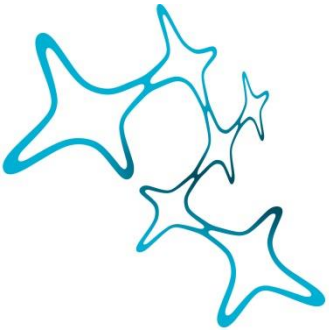
Using Genetics to Explore Novel Risk Factors and Drug Targets for Cerebrovascular Disease

Marios K. Georgakis



Graduate School of Systemic Neurosciences

LMU Munich

Dissertation der
Graduate School of Systemic Neurosciences der
Ludwig-Maximilians-Universität München

December 6th, 2019

Supervisor - 1st Reviewer

Prof. Dr. med. Martin Dichgans

Director, Institute for Stroke and Dementia Research

University Hospital, LMU Munich

2nd Reviewer

Prof. Dr. Annette Peters

Director, Institute of Epidemiology

Helmholtz Zentrum München

Date of defense: Tuesday, April 21st 2020

To my parents and my brother

Στους γονείς μου και τον αδερφό μου

TABLE OF CONTENTS

ABSTRACT

Cerebrovascular disease is a major cause of mortality and disability worldwide. Treatment options for stroke, the acute clinical manifestation of cerebrovascular disease, remain restricted and the development of new therapeutic strategies is hampered by the difficulties in identifying neuroprotective approaches. Thus, current efforts are focused on prevention. The development of effective preventive strategies requires enriching our knowledge regarding stroke etiology. Stroke is a highly heterogeneous disease. While multiple risk factors have been found for stroke as a whole, there is only limited evidence about specific risk factors for etiologically-defined stroke subtypes. Evidence regarding risk factors for stroke subtypes is mainly derived from observational studies, which could be biased because of confounding and reverse causation.

In this thesis, I aimed to explore potential risk factors and drug targets for stroke, stroke subtypes, and manifestations of cerebral small vessel disease (SVD) by using Mendelian randomization (MR). MR is a novel approach for exploring associations based on the use of data from genome-wide association studies (GWAS). By using genetic variants as proxies for a trait of interest, MR overcomes key limitations of observational studies and allows for investigation of causal effects on outcomes. By restricting the selection of variants to the vicinity of genes encoding candidate drug targets, MR further enables the prediction of the effects and side-effects of pharmacological interventions.

On the basis of experimental and clinical evidence suggesting a key role of inflammation in atherosclerosis, my collaborators and I explored the associations of inflammatory mediators with the risk of stroke and stroke subtypes. First, using GWAS data on the circulating levels of 41 cytokines and growth factors (n=8,293), we explored the association of their genetic determinants with stroke and stroke subtypes (n=521,612). We found genetic predisposition to higher levels of monocyte-chemoattractant protein-1 (MCP-1), a chemokine attracting monocytes to the sites of inflammation, to be associated with a higher risk of ischemic stroke, and particularly large artery and cardioembolic stroke. We further found similar associations for the atherosclerotic phenotypes of coronary artery disease and myocardial infarction. Second, to validate these findings, we explored the association between MCP-1 levels and risk of incident stroke in a meta-analysis of 6 population-based cohort studies (n=17,180, mean age 57 years, 51%

women). Over a follow-up of 16 years, we found higher MCP-1 levels to be associated with a higher risk of any stroke and ischemic stroke, but not hemorrhagic stroke. Third, we constructed a genetic score reflecting the activity of interleukin-6 (IL-6) signaling and explored associations with ischemic stroke. We found genetically downregulated IL-6 signaling to be associated with lower risk of ischemic stroke, particularly large artery and small vessel stroke. Interestingly, in observational data, IL-6 and MCP-1 levels were both associated with ischemic stroke risk, independently of each other, thus indicating that targeting the two pathways might offer complementary benefits in reducing stroke risk.

We further explored the effects of genetic predisposition to traditional vascular risk factors on different stroke subtypes and SVD. Using GWAS data (n=757,601), We identified genetic proxies for the effects of common first-line antihypertensive medications that showed associations with stroke and coronary artery disease that were comparable to those derived from clinical trials. In a phenome-wide association study, using these genetic proxies, we were able to detect and validate with observational data a previously unreported adverse effect of calcium channel blockers on risk of diverticulosis. We then used these genetic proxies to explore associations with stroke subtypes and cerebral SVD. Aside from the expected associations between genetically predicted blood pressure and all major stroke subtypes, we found the proxies for calcium-channel blockers to show particularly strong associations with small vessel stroke and the related radiological SVD phenotype of white matter hyperintensities (WMH). When exploring blood lipids, genetic predisposition to higher levels of high-density lipoprotein cholesterol (HDL-C) was associated with lower risk of small vessel stroke risk and WMH. This effect was primarily driven by cholesterol concentration in medium-sized HDL particles. Genetic proxies for CETP inhibitors, an HDL-C-raising drug class, showed associations with lower risk of ischemic SVD manifestations (small vessel stroke, WMH), but also with higher risk of intracerebral hemorrhage, a hemorrhagic SVD manifestation.

In conclusion, using genetic data from humans, this thesis identified novel risk factors and drug targets for stroke subtypes and cerebral SVD. Specifically, I provide evidence supporting MCP-1 and IL-6 signaling as promising novel strategies for lowering the risk of ischemic stroke. Already approved strategies like lowering blood pressure with calcium channel blockade and increasing HDL-C by CETP inhibition might offer benefits for preventing the ischemic manifestations of cerebral SVD.

LIST OF ABBREVIATIONS

AF	atrial fibrillation
CCL2	CC chemokine ligand-2
CCR2	CC chemokine receptor-2
CETP	cholesteryl-ester transfer protein
CNS	central nervous system
CRP	C-reactive protein
CT	computed tomography
DBP	diastolic blood pressure
GWAS	genome-wide association study
HDL-C	high-density lipoprotein cholesterol
ICH	intracerebral hemorrhage
IL-1β	interleukin-1β
IL-6	interleukin-6
IL-6R	interleukin-6 receptor
LDL-C	low-density lipoprotein cholesterol
MR	Mendelian randomization
MCP-1	monocyte chemoattractant protein-1
MRI	magnetic resonance imaging
RCT	randomized controlled trial
PheWAS	phenome-wide association study
SBP	systolic blood pressure
SVD	small vessel disease
TG	triglycerides
TOAST	trial of ORG 10172 in acute stroke treatment
WMH	white matter hyperintensities

INTRODUCTION

Cerebrovascular disease refers to a variety of disorders that affect the brain vessels or blood supply to the brain. Stroke, the most extreme clinical manifestation of cerebrovascular disease, is a focal neurological injury due to a vascular cause, which includes infarction of the central nervous system (CNS), defined as cell death in the brain, the spinal cord or the retina based on either pathological and imaging evidence of ischemic injury in a defined vascular distribution or clinical evidence of symptoms of acute neurological dysfunction persisting for ≥24 hours or until death (Sacco *et al.*, 2013). Stroke may occur as a result of either CNS ischemia (ischemic stroke) or CNS hemorrhage (hemorrhagic stroke) (Hankey, 2017). Despite the undisputable progresses in our understanding of the etiology, the pathophysiology, and the consequences of stroke, it remains a devastating disease with a huge burden for the patients, their caregivers, the public health systems (Feigin *et al.*, 2016b). Given the limited options and difficulties associated with stroke treatment and rehabilitation, the attention has shifted towards stroke prevention (Feigin *et al.*, 2016a). Current evidence suggests that stroke is a disease with a very high potential for prevention (Tikk *et al.*, 2014). Although the major risk factors for stroke, like hypertension, diabetes mellitus, hypercholesterolemia, and smoking are in common with those for other vascular disorders, little is known about the associations of these risk factors with the highly heterogeneous etiological subtypes of stroke. Taking into account the heterogeneity of the underlying etiology might further be important in identifying novel risk factors and drug targets for specific stroke subtypes, thus moving towards more targeted personalized approaches.

Epidemiology of stroke

Stroke is a major public health issue. It is estimated that more than 80 million people live with stroke worldwide, 15 million people suffer a stroke, and 6 million people die because of stroke every year (Feigin *et al.*, 2017; Thrift *et al.*, 2017; G. B. D. Stroke Collaborators, 2019). Stroke accounts for more than 5% of disability-adjusted life-years worldwide, comprising along with ischemic heart disease, the most common cause of disability worldwide (G. B. D. DALYs Hale Collaborators, 2017). Furthermore, stroke accounts for more than 10% of all deaths worldwide being the second most common cause of death after ischemic heart disease (G. B. D. Causes of Death Collaborators, 2017). Besides, functional disability and death, stroke has also been recognized as a major contributor to cognitive decline and dementia (Pendlebury *et al.*, 2019), as well as to depression (Towfighi *et al.*, 2017).

The lifetime risk of stroke from the age of 25 onwards is around 25% and is similar between men and women. Yet, there is significant regional and between-country heterogeneity, with the highest lifetime risk being noted in East Asia (mainly due to a very high rate in China), Central Europe, and Eastern Europe (G. B. D. Lifetime Risk of Stroke Collaborators *et al.*, 2018). Stroke is a disease of the elderly with a steep increase in incidence rates with increasing ages (Benjamin *et al.*, 2017). Men have a higher incidence of stroke at ages lower than 75 years, but this trend is reversed at around 75 years of age, with women having a higher incidence thereafter (Leening *et al.*, 2014; Benjamin *et al.*, 2017). In a background of increasing life expectancy worldwide (G. B. D. Mortality Collaborators, 2017), the lifetime risk of stroke has increased over the last decades (G. B. D. Lifetime Risk of Stroke Collaborators *et al.*, 2018).

The incidence of stroke is decreasing in high-income countries (Koton *et al.*, 2014; Vangen-Lonne *et al.*, 2017), but is increasing in low-income countries (Feigin *et al.*, 2014). Although stroke mortality and disability rates have decreased worldwide since 1990, its global burden in terms of absolute numbers of people who died from stroke, survived a stroke, remained disabled from stroke, and affected by stroke remains very high and has increased dramatically (1.4- to 1.8-fold increases) (Feigin *et al.*, 2017). These numbers call for preventive actions, especially given the strong evidence suggesting that substantial prevention of stroke is feasible (Tikk *et al.*, 2014). There are well-known

modifiable risk factors for stroke, which account for more than 90% of the attributable risk of stroke across both sexes, all age groups, and ethnicities (O'Donnell *et al.*, 2016).

Stroke subtypes and underlying etiology

Stroke is a heterogeneous disease. At a primary level, stroke may be classified as ischemic or hemorrhagic, depending on whether the underlying cause is an occlusion or a rupture of a blood vessel, respectively (Hankey, 2017). Yet, even the two major subtypes have different underlying etiologies.

According to the Trial of Organization 10172 in Acute Stroke Treatment (TOAST) classification system, which is the most commonly used tool for stroke classification in clinical practice, ischemic stroke may be classified to five major etiological subtypes: large artery atherosclerosis, cardioembolism, small vessel disease (SVD), stroke of other determined etiology (e.g. arterial dissection), and stroke of undetermined etiology (Adams *et al.*, 1993). Determination of stroke etiology is important in clinical practice as it then guides secondary prevention of recurrent stroke (Kernan *et al.*, 2014). Based on the TOAST criteria, epidemiological studies suggest that approximately 15% of ischemic strokes may be defined as large artery atherosclerotic strokes, 30% as cardioembolic strokes, 25% as small vessel strokes, and <5% as strokes of other determined etiologies (Petty *et al.*, 1999; Kolominsky-Rabas *et al.*, 2001; Schneider *et al.*, 2004). In around 25-30% of patients with stroke, the etiology cannot be sufficiently determined (Petty *et al.*, 1999; Kolominsky-Rabas *et al.*, 2001; Schneider *et al.*, 2004).

Large artery atherosclerotic stroke is caused by atherosclerotic lesions in the large extracranial (common and internal carotids, vertebral arteries) or intracranial arteries (circle of Willis and proximal branches). Atherosclerotic plaques in extracranial and intracranial arteries may cause a stroke by either reducing blood flow beyond obstructive lesions or by serving as sources of intra-arterial emboli. Thrombi are often superimposed upon the atherosclerotic plaques. Clinically, the determination of large artery atherosclerotic stroke requires a stenosis of >50% or occlusion of the relevant artery, as determined by vascular imaging (Adams *et al.*, 1993; Adams and Biller, 2015). Vascular imaging initially included arteriography and carotid duplex or transcranial Doppler

ultrasonography, and has now further expanded to magnetic resonance arteriography or computed tomography (CT) angiography. The patients usually manifest neurological findings consistent with infarction of the cerebral cortex or both deep and cortical structures, the brain stem, or cerebellum, accompanied by imaging findings of infarction of the respective areas. In the majority of cases, there is also evidence for risk factors for accelerated atherosclerosis or symptomatic atherosclerotic disease in other vascular beds (e.g. coronary and peripheral arterial disease) (Adams *et al.*, 1993; Adams and Biller, 2015).

Cardioembolic stroke is caused by emboli arising from the heart, which are transmitted with blood flow to the brain, thus leading to occlusion of cerebral arteries and infarction of the supplied territories (Adams *et al.*, 1993; Adams and Biller, 2015; Kamel and Healey, 2017). Cardioembolic strokes may arise from a known cardiac source or may be of a possible cardiac or ascending aortic source based on transthoracic or transesophageal echocardiographic findings (Adams *et al.*, 1993; Adams and Biller, 2015). Cardioembolism leads to strokes of higher severity, as compared to other stroke etiologies (Lin *et al.*, 1996). The main cardiac source of emboli to the brain is atrial fibrillation (AF) (Kamel and Healey, 2017), which may be diagnosed by electrographic investigation and cardiac rhythm monitoring. Other causes of cardioembolism might include valvular disease, patent foramen ovale, recent myocardial infarction, forms of cardiomyopathy, infective endocarditis, atrial septal aneurysm, and atheroma of the ascending aorta or the proximal aortic arch (Adams *et al.*, 1993; Adams and Biller, 2015).

Small vessel stroke refers to small (0.2 to 15 mm in diameter) noncortical infarcts caused by occlusion of a penetrating branch of a large cerebral artery (Adams *et al.*, 1993; Adams and Biller, 2015). These branches arise from the large arteries of the circle of Willis, stem of the middle cerebral artery, and the basilar artery. Most lacunes occur in the basal ganglia, subcortical white matter, and pons (Pantoni, 2010). These arteries are usually affected by either lipohyalinosis and fibrinoid degeneration (usually secondary to hypertension) or microatheroma at their origin or in the parent large artery (Fisher, 1968, 1978, 1979; Pantoni, 2010). Clinically, these patients usually have evidence of arterial hypertension or diabetes mellitus, which are recognized risk factors for SVD (Jackson and Sudlow, 2005). Imaging requirements further include demonstration of a small (<1.5 cm) deep infarction restricted to the basal ganglia, internal capsule, thalamus, or brain stem (Adams *et al.*, 1993; Adams and Biller, 2015). In addition, vascular imaging should not

demonstrate findings consistent with large artery atherosclerosis in the clinically relevant vessel (Adams *et al.*, 1993; Adams and Biller, 2015).

Hemorrhagic stroke may be subdivided into intracerebral (ICH) and subarachnoid hemorrhages, which relate to bleeding directly into the brain parenchyma or the surrounding subarachnoid space, respectively (Cordonnier *et al.*, 2018). The primary causes of ICH include pathologies of the small arteries, which might include lipohyalinosis or cerebral amyloid angiopathy (Cordonnier *et al.*, 2018). Interestingly, there is an anatomical distinction between ICHs caused by lipohyalinosis versus those causes by cerebral amyloid angiopathy. Most commonly, isolated lobar hemorrhages are caused by cerebral amyloid angiopathy (Rodrigues *et al.*, 2018), whereas hemorrhages in deeper brain structures are primarily caused by lipohyalinosis, usually as a consequence of hypertension (Jackson and Sudlow, 2006; Martini *et al.*, 2012). Clinically, the neurologic symptoms related to ICH may not begin abruptly and are not at maximal intensity at onset, but they usually increase gradually over minutes or a few hours. Headache, vomiting, and a decreased level of consciousness develop if the hematoma becomes large enough to increase intracranial pressure or cause shifts in intracranial contents (Cordonnier *et al.*, 2018). ICH causes damage to the brain parenchyma as it enlarges. The pressure created by blood and surrounding edema is life-threatening; ICHs have very high mortality and morbidity (van Asch *et al.*, 2010; Cordonnier *et al.*, 2018).

The two major causes of subarachnoid hemorrhage are rupture of arterial aneurysms that are situated at the base of the brain and bleeding from vascular malformations that lie near the pial surface (Lawton and Vates, 2017; Muehlschlegel, 2018). Symptoms of subarachnoid hemorrhage begin abruptly in contrast to the more gradual onset of ICH (Edlow *et al.*, 2008; Lawton and Vates, 2017) with instantly severe and widespread headache comprising the most common presenting symptom (Linn *et al.*, 1994). Vomiting occurs soon after onset (Edlow *et al.*, 2008). The prognosis of the disease is poor with the pre-hospital and 30-day fatality rates being at 15% and 35%, respectively (Nieuwkamp *et al.*, 2009; Lovelock *et al.*, 2010).

Cerebral small vessel disease

The term cerebral SVD describes the pathological processes affecting the perforating arterioles, capillaries, and venules of the brain (Pantoni, 2010; Wardlaw *et al.*, 2013a). Cerebral SVD is strongly associated with aging and its manifestations are at least to some extent present in almost all elderly individuals (de Leeuw *et al.*, 2001). SVD is the major vascular contributor to dementia (Debette *et al.*, 2018), accounts for about 25% of ischemic strokes (Sudlow and Warlow, 1997) and for almost all cases of intracerebral hemorrhage (Qureshi *et al.*, 2001; Qureshi *et al.*, 2009), and is an independent predictor of mortality (Debette *et al.*, 2018). SVD has further been associated with physical and psychological sequalae in the elderly, including gait (de Laat *et al.*, 2010), functional (Inzitari *et al.*, 2009), mood (van Agtmaal *et al.*, 2017), and urinary disturbances (Poggesi *et al.*, 2008).

As conventional imaging techniques do not allow direct assessment of cerebral small vessels *in vivo*, SVD is mainly defined by its neuroimaging manifestations, according to standard criteria (Wardlaw *et al.*, 2013b). The typical lesions that are associated with SVD include lacunes and recent small subcortical infarcts, alterations in white matter, which are observed as hyperintensities in T2 sequences in magnetic resonance imaging (MRI), cerebral microbleeds and intracerebral hemorrhages, and enlarged perivascular spaces (Wardlaw *et al.*, 2013b). More sensitive imaging methods further show pathological changes in the otherwise normal-appearing parenchyma, especially in the white matter, which are believed to arise as a result of demyelination or increased interstitial fluid (Baykara *et al.*, 2016; Munoz Maniega *et al.*, 2017).

The most common pathological alterations associated with cerebral SVD include arteriolosclerosis and cerebral amyloid angiopathy (Pantoni, 2010). Arteriolosclerosis, otherwise commonly called “hypertensive arteriopathy”, describes a pathological process related to degenerative alterations of the vessel walls, which is common in small perforating end arterioles of the deep grey nuclei and deep white matter of the brain, but is also a systemic disorder (Pantoni, 2010). Arteriolosclerotic microvascular lesions outside the brain are characteristically found in the microcirculation of the kidneys and the retina. They are strongly associated with aging and with vascular risk factors, particularly hypertension and diabetes. This type of SVD is characterized by loss of

smooth muscle cells from the tunica media, deposits of fibro-hyaline material (lipohyalinosis), narrowing of the lumen, and thickening of the vessel wall (Fisher, 1968, 1978, 1979; Lammie, 2002; Pantoni, 2010). Microaneurysms and microatheromas in the parent vessels often co-exist with arteriolosclerosis (Fisher, 1968, 1978, 1979; Pantoni, 2010).

Cerebral amyloid angiopathy is the second most common type of SVD and is characterized by the accumulation of the amyloid β (Aβ) protein in the media and adventitia of small-to-medium-sized arteries and arterioles predominantly located in the leptomeningeal space and the cortex (Vinters and Gilbert, 1983; Banerjee *et al.*, 2017). The accumulation of Aβ makes the vessel fragile and vulnerable to rupture and is thus associated with microbleeds and intracerebral hemorrhages (Vinters and Gilbert, 1983; Banerjee *et al.*, 2017). Although the extent to which it might contribute to ischemic manifestations of SVD remains uncertain, the presence of microbleeds due to CAA might influence treatment decisions in patients with ischemic stroke, such as the use of antithrombotic and anticoagulant agents (Wang *et al.*, 2014; Wilson *et al.*, 2018). A definite diagnosis of CAA requires neuropathological evidence of presence of Aβ in the vessel wall. Yet, the development of standard clinical and diagnostic criteria with high specificity for the diagnosis of CAA, have enabled the investigation of the disease with neuroimaging studies (Knudsen *et al.*, 2001; Linn *et al.*, 2010).

Despite its major impact on public health, the etiology of SVD remains largely unknown, thus hampering the development of treatments that could reduce the burden of the disease (Bath and Wardlaw, 2015; Wardlaw *et al.*, 2019). Several mechanisms have been proposed as playing a key role in the pathogenesis of the disease. These include endothelial dysfunction leading to blood-brain barrier dysfunction and impaired vasodilation (Wardlaw *et al.*; Rajani *et al.*, 2018; Nation *et al.*, 2019), vessel stiffening causing dysfunctional blood flow and interstitial fluid drainage (Shi *et al.*, 2016; Mestre *et al.*, 2018; Shi *et al.*, 2018), as well as inflammation (Shoamanesh *et al.*, 2015).

Genetic risk factors for stroke and cerebral small vessel disease

Genetic risk factors contribute to cerebrovascular disease. Having a first-degree relative with stroke increases the risk of stroke by 30% (Flossmann *et al.*, 2004), whereas monozygotic twin are twice more likely to be concordant for stroke as compared to dizygotic twins (de Faire *et al.*, 1975; Flossmann *et al.*, 2004). The overall heritability of ischemic stroke is estimated to 38%, but varies across subtypes (from 16% for small vessel stroke to >40% for large artery stroke) (Bevan *et al.*, 2012) and heritability for ICH is estimated to 29% (Devan *et al.*, 2013), based on genome-wide associations study (GWAS) data. The heritability of subclinical radiological markers of cerebral SVD (WMH, enlarged perivascular spaces), is estimated to be even >50% (Duperron *et al.*, 2018). Hereditary factors that increase the risk of cerebrovascular disease could either comprise rare single mutations with high penetration that lead to mendelian forms of disease or common genetic variants that increase the risk modestly.

Rare single mutations may cause familial disorders with stroke as the primary manifestation. Advances in sequencing technology have facilitated the discovery of such single-gene disorders associated with stroke, and especially SVD, which can manifest with either stroke or cognitive decline and other manifestations (Haffner *et al.*, 2016). Typical examples in this category involve cerebral autosomal dominant arteriopathy with subcortical infarcts and leukoencephalopathy (CADASIL), cerebral autosomal recessive arteriopathy with subcortical infarcts and leukoencephalopathy (CARASIL), or pontine autosomal dominant microangiopathy with leukoencephalopathy (PADMAL) (Dichgans *et al.*, 2019). CADASIL is the most common hereditary cause of stroke and cognitive decline, and has gained great interest as the model disease to study the more common sporadic form of SVD (Chabriat *et al.*, 2009; Dichgans *et al.*, 2019). It is caused by mutations in *NOTCH3* and leads to thickening of the arteriolar walls, accumulation of an amyloid granular osmiophilic material in the arterial walls, and prominent degeneration of vascular smooth muscle cells (Joutel *et al.*, 1996; Chabriat *et al.*, 2009; Dichgans *et al.*, 2019). CADASIL is considered a disease model for arteriolosclerosis. Similarly, hereditary forms of cerebral amyloid angiopathy have been described, but they are very rare in the general population (Biffi and Greenberg, 2011). Single mutations could also lead to familial disorders with more complex phenotypes with cerebrovascular disease being one of several manifestations. Typical examples in this category include Marfan's syndrome

and the vascular Ehler's Danlos syndrome, which are both characteristically associated with arterial dissection (Dichgans *et al.*, 2019).

Common genetic variants (typically single nucleotide polymorphisms with allele frequencies >0.5-1%) might be associated with small increases in the risk of stroke. These genetic variants are identified through GWASs, which compare the frequency of all common variants in the genome between individuals having suffered a stroke and stroke-free controls. The largest to-date GWAS for stroke using data from the MEGASTROKE Consortium, was a meta-analysis of 67,162 cases and 454,450 controls and identified 32 loci associated with either any stroke, ischemic stroke, or one of the major ischemic stroke subtypes (Malik *et al.*, 2018a). A subsequent meta-analysis of the MEGASTROKE data with the UK Biobank (72,147 stroke patients and 823,869 controls) further increased the number of significant loci to 35 (Malik *et al.*, 2018b). Some of the identified variants were specific for etiological stroke subtype: 6 loci were specific for large artery stroke, 4 for cardioembolic stroke, and 2 for small vessel stroke (Malik *et al.*, 2018a). The majority of the identified variants were common (minor allele frequency >5%), showed modest increases in risk of stroke (OR<1.30) and were located in non-coding regions (Malik *et al.*, 2018a; Dichgans *et al.*, 2019). Some genetic variants can confer risk of stroke by influencing the risk of causal stroke risk factors. Indeed, almost half of the identified loci in the abovementioned GWAS meta-analyses have been associated in previous studies with high blood pressure, blood lipid levels, carotid plaque, and atrial fibrillation (AF) (Malik *et al.*, 2018a). Similarly, trans-ethnic and European-based GWAS meta-analyses for white matter hyperintensities (WMH) have identified 9 loci associated at a genome-wide significance threshold (Verhaaren *et al.*, 2015; Traylor *et al.*, 2019). Interestingly, a genetic risk score for WMH volume constructed based on these studies has been found to be also associated with the risk of small vessel stroke, thus indicating common genetic architecture of the different SVD manifestations (Traylor *et al.*, 2016).

Risk factors for stroke and cerebral small vessel disease

Although the high heterogeneity in the etiology of cerebrovascular disease complicates the exploration of risk factors, multiple modifiable and non-modifiable risk factors have been identified. For stroke, an international case-control study (INTERSTROKE) recently showed that a set of 10 risk factors including hypertension, smoking, waist-to-hip ratio, diet, physical activity, alcohol intake, psychosocial stress, cardiac disease (e.g. atrial fibrillation), and the ratio of apolipoprotein B to A1 could explain up to 90% of stroke population-attributable risk (O'Donnell *et al.*, 2010). All of these risk factors were associated with risk of ischemic stroke, whereas hypertension, smoking, waist-to-hip ratio, diet, and alcohol intake were also significantly associated with the risk of hemorrhagic stroke (O'Donnell *et al.*, 2010).

Hypertension is the leading risk factor for both ischemic and hemorrhagic stroke estimated to account for ~50% of the population attributed risk of stroke worldwide (Feigin *et al.*, 2016b; O'Donnell *et al.*, 2016; Forouzanfar *et al.*, 2017). Both systolic (SBP) and diastolic blood pressure (DBP) are linearly associated with stroke incidence and mortality in both sexes and across all age groups (Lewington *et al.*, 2002; Lacey *et al.*, 2018). Furthermore, hypertension is considered the primary risk factors for cerebral SVD and particularly arteriolosclerosis (Wardlaw *et al.*, 2019). Antihypertensive treatment remains one of the primary targets for lowering the global burden of cerebrovascular disease (Blood Pressure Lowering Treatment Trialists, 2014; Meschia *et al.*, 2014; Ettehad *et al.*, 2016; Xie *et al.*, 2016; Brunstrom and Carlberg, 2018). Interestingly, clinical trials show that BP lowering with different antihypertensive drug classes might differentially influence the risk of stroke. Specifically, calcium channel blockers were shown to be superior for primary prevention of stroke, as compared to beta blockers (Rothwell *et al.*, 2010; Webb *et al.*, 2010; Ettehad *et al.*, 2016). While hypertension is an established risk factor for both ischemic and hemorrhagic stroke (O'Donnell *et al.*, 2016; Lacey *et al.*, 2018), the effects of BP and different antihypertensive drug classes on etiologically defined stroke subtypes (Adams *et al.*, 1993), in particular, large artery stroke, cardioembolic stroke, small vessel stroke, deep and lobar intracerebral hemorrhage (ICH), remain largely unknown. Only a few observational studies have examined whether BP is differentially associated with stroke subtypes (Schulz and Rothwell, 2003; Ohira *et*

al., 2006; Zia *et al.*, 2007; Li *et al.*, 2015), but data from BP lowering trials on stroke subtypes are missing (Ettehad *et al.*, 2016).

Blood lipids are a well-established risk factor for large artery atherosclerosis (Collins *et al.*, 2016). Lipid-modifying therapies have shown benefits in reducing risk of both coronary artery disease and large artery stroke (Cholesterol Treatment Trialists Collaboration *et al.*, 2010; Chou *et al.*, 2016). Current guidelines for secondary stroke prevention recommend treatment with statins after ischemic stroke or transient ischemic attack (European Stroke Organisation Executive Committee and E. S. O. Writing Committee, 2008; Kernan *et al.*, 2014; Intercollegiate Stroke Working Party, 2016; Stroke Foundation, 2017) referring to large-scale clinical trials data and meta-analyses (Amarenco *et al.*, 2006; Amarenco and Labreuche, 2009; Manktelow and Potter, 2009). However, most trials provided no sub-analyses for ischemic stroke subtypes. Thus, the role of blood lipids in forms of cerebrovascular disease other than large artery stroke remains largely elusive. The J-STARS trial, the only study providing sub-analyses, found statins to reduce recurrence of large artery stroke but not small vessel stroke (Hosomi *et al.*, 2015). Results from the SPARCL trial suggest that statins may increase the risk of ICH in patients with stroke or transient ischemic attack (Amarenco *et al.*, 2006), especially in patients with SVS as an entry event (Goldstein *et al.*, 2008).

AF is an arrhythmia with very high prevalence among the elderly population (>10% among those aged >80 years) (Chugh *et al.*, 2014). AF is associated with a predisposition towards atrial thrombi formation and is considered a major source of cardioembolism, thus increasing the risk for cardioembolic stroke. Observational cohort studies have found AF to increase the risk of stroke by 3- to 5-fold (Wolf *et al.*, 1991). The number of patients with AF may double and the number of AF-related strokes may triple in the next few decades based on projections from high-income countries, such as the United States (Go *et al.*, 2001). Current guidelines for stroke classification and prevention of recurrent stroke clearly highlight the need to identify AF among patients with stroke, and treating them with anticoagulation depending on the cumulative risk of stroke among every individual patient (Kernan *et al.*, 2014).

Diabetes mellitus is another major risk factor for cerebrovascular disease. Prospective cohort studies have consistently shown presence of diabetes mellitus to be associated with 1.5- to 2-fold increases in risk of ischemic and hemorrhagic stroke (Emerging Risk

Factors *et al.*, 2010). Fasting glucose levels in non-diabetic individuals, pre-diabetes, and duration of the period living with diabetes are also strong independent risk factors for stroke and cerebral SVD (Abbott *et al.*, 1987; Sui *et al.*, 2011; Banerjee *et al.*, 2012; Lee *et al.*, 2012). Furthermore, intensive glycemic control in patients with type II diabetes mellitus has been shown to decrease stroke risk and stroke mortality (Gaede *et al.*, 2008).

A number of other behavioral risk factors have further been associated with the risk of stroke in a similar way that they have been associated with other forms of vascular disease. These include overweight and obesity (Strazzullo *et al.*, 2010), alcohol intake (Millwood *et al.*, 2019b), smoking (Larsson *et al.*, 2019; Pan *et al.*, 2019), physical inactivity and sedentary behavior (Kivimaki *et al.*, 2019), and specific nutritional choices (Psaltopoulou *et al.*, 2013; Iacoviello *et al.*, 2018). Although the associations of all these factors with risk of stroke are considered well-established, again there is only limited evidence regarding associations with specific stroke etiologies.

Inflammation as a novel risk factor for cerebrovascular disease

Extensive experimental evidence suggests atherosclerosis, one of the major underlying etiologies of ischemic stroke, to be a primarily chronic inflammatory disorder (Hansson, 2005; Libby *et al.*, 2011). Both innate and adaptive immune mechanisms have been found to participate in the initiation and propagation of atherosclerosis (Hansson, 2005; Libby *et al.*, 2011). Furthermore, inflammation has pro-thrombotic effects and is even involved in the pathogenesis of major risk factors for stroke such as atrial fibrillation (Kamel and Iadecola, 2012). Epidemiological studies consistently show biomarkers of inflammation to be associated with the risk of atherosclerotic events independently of traditional vascular risk factors (Kaptoge *et al.*, 2010; Kaptoge *et al.*, 2014). Despite the aggressive control of blood pressure, lipids, and glucose levels, there is still a high prevalence of both coronary artery disease events and stroke. As in these individuals, inflammatory biomarkers, and particularly circulating levels of high-sensitivity C-reactive protein (hsCRP), have been associated with the risk of vascular events, a new concept of residual inflammatory risk has been developed (Ridker, 2017; Bohula *et al.*, 2018). This concept is further supported by clinical data. Statin treatment has been shown in several trials to reduce markers of inflammation in a dose-response manner and these decreases are

proportional to the final vascular benefit, especially among patients with high hsCRP levels at the time of treatment initiation (Ridker *et al.*, 2008; Bohula *et al.*, 2018).

In the last years, two large randomized clinical trials specifically targeted vascular inflammation in patients with established cardiovascular disease and provided novel insights into the role of inflammatory mechanisms as potential drug targets for reducing vascular risk. In the CANTOS trial, treatment with an anti-IL-1β (interleukin-1β) monoclonal antibody reduced the levels of IL-6 (interleukin-6) and hsCRP leading to a reduction in the combined primary endpoint of nonfatal myocardial infarction, nonfatal stroke or cardiovascular death independent of low-density lipoprotein cholesterol (LDL-C) levels in patients with a history of myocardial infarction and hsCRP levels ≥2 mg/l (Ridker *et al.*, 2017). On the contrary, treatment with low-dose methotrexate in the CIRT trial neither reduced cardiovascular event rates nor the levels of IL-1β, IL-6, and hsCRP in patients with stable coronary artery disease (Ridker *et al.*, 2019a).

These discordant results from the CANTOS (Ridker *et al.*, 2017; Aday and Ridker, 2019; Ridker, 2019; Ridker *et al.*, 2019a) and CIRT (Ridker *et al.*, 2019a) randomized controlled trials emphasize the importance of targeting specific mediators and pathways for lowering vascular risk (Ridker *et al.*, 2017; Aday and Ridker, 2019; Ridker, 2019; Ridker *et al.*, 2019a). IL-6, a key regulator of the inflammatory cascade, acts by binding to either its membrane-bound or soluble receptor (IL-6R, IL-6 receptor) and induces proinflammatory downstream effects including increases in the circulating levels of CRP (Scheller *et al.*, 2011; Ridker, 2016). IL-6 has been implicated in the pathogenesis of several inflammatory diseases (Scott, 2017; Stone *et al.*, 2017; Danese *et al.*, 2019) and downregulation of its signaling cascade has been proposed as a potential strategy for lowering cardiovascular risk (Ridker, 2016, 2019). IL-6 levels have consistently been associated with risk of coronary artery disease in cohort studies (Ridker *et al.*, 2000; Kaptoge *et al.*, 2014). Mendelian randomization (MR) studies further showed that a variant in the gene encoding IL-6R with effects resembling pharmacological IL-6R inhibition is associated with a lower risk of coronary artery disease (Il R. Genetics Consortium Emerging Risk Factors Collaboration *et al.*, 2012; Interleukin-6 Receptor Mendelian Randomisation Analysis Consortium *et al.*, 2012). Finally, secondary analyses from CANTOS demonstrated that the magnitude of the therapeutic benefit of IL-1β targeting was associated with the reduction of circulating IL-6 levels (Ridker *et al.*, 2018a; Ridker, 2019) and that even after IL-1β inhibition, the residual cardiovascular risk was

proportional to the post-treatment IL-6 levels (Ridker *et al.*, 2019b). These results from CANTOS provide indirect clinical evidence that interfering with IL-6 signaling might lower cardiovascular risk and suggest that an approach directly targeting IL-6 signaling could offer additional benefit for cardiovascular prevention beyond IL-1β inhibition.

However, the IL-1β/IL-6/CRP axis is only one of the inflammatory pathways that are involved in the pathogenesis of atherosclerosis. Identification and exploration of different pathways, which might include other inflammatory mediators with more specific roles in the initiation and progression of atherosclerosis are considered crucial for the development of successful anti-inflammatory strategies. The complex system of chemokines is one such pathway (Noels *et al.*, 2019). Chemokines are part of the innate immunity and act in an orchestrated and complex way by attracting specific inflammatory cells in the sites of inflammation. They contribute to accumulation of inflammatory cells in the atherosclerotic plaque, expansion of the plaque, plaque destabilization, and thrombosis (Noels *et al.*, 2019). Although animal studies in experimental models of atherosclerosis have long suggested the chemokine system as a potential target for lowering vascular risk, there is only scarce evidence from humans regarding the potential efficacy of such an approach (Noels *et al.*, 2019). Monocyte-chemoattractant protein-1 (MCP-1) or CC chemokine ligand-2 (CCL2), the prototypical CC chemokine has attracted major attention, due to its key role in recruiting monocytes to the atherosclerotic plaque (Nelken *et al.*, 1991; Lutgens *et al.*, 2005; Lin *et al.*, 2014). Evidence from animal models of atherosclerosis suggests that knocking out either MCP-1 or its receptor CCR2 (CC chemokine receptor-2) could lead to attenuation of the atherosclerotic phenotype (Boring *et al.*, 1998; Gu *et al.*, 1998; Combadiere *et al.*, 2008; Liehn *et al.*, 2010; Bot *et al.*, 2017). Yet, data from humans regarding the role of MCP-1 in atherosclerosis remain scarce.

Mendelian Randomization

The gold-standard approach for inferring causality in medical research is a randomized controlled trial (RCT). However, the majority of the known risk factors for cerebrovascular disease have been identified through epidemiological observational studies, typically population-based longitudinal cohort studies that might be biased because of confounding and reverse causation (Fewell *et al.*, 2007). Yet, conducting RCTs is very expensive and time-consuming and RCTs have further been found to have very low success rates. The main reason for RCT failures is lack of efficacy of the respective drug target (Harrison, 2016). This is assumed to be partly the result of a low-quality evidence basis that supports conducting an RCT (Khakoo *et al.*, 2019). Particularly, consistently biased results from large-scale observational studies and the inability to translate findings from animal models to humans are believed to be the main reasons for these high failure rates (Smith *et al.*, 2007). Hence, it is of crucial importance to prioritize interventions to be tested in RCTs based on unbiased evidence in order to maximize the probability of success.

MR is a methodology developed over the last three decades that overcomes the inherent limitations of observational studies (confounding and reverse causation) while using observational data and thus enables exploration of causal associations without the need of conducting an RCT (Zheng *et al.*, 2017; Bandres-Ciga *et al.*, 2019). It is a type of instrumental variable analysis that instead of measuring a risk factor directly, uses information from genetics to create an instrument of genetic predisposition to the risk factor under study (Zheng *et al.*, 2017). Then, using this genetic instrument, MR explores its effect on outcomes of interest (Zheng *et al.*, 2017). By grouping individuals in the population according to the presence or not of specific genetic variants that modify a risk factor allows researchers exploring if a biomarker is causally related to a disease (Holmes *et al.*, 2017). This inference is based on the fundamental characteristics of the genome: first, because of the random allocation of alleles during meiotic segregation, genetic variants used as instrument variables should be free from confounding effects; second, owing to the non-modifiable nature of the germline genome, bias due to reverse causation can be excluded (Holmes *et al.*, 2017).

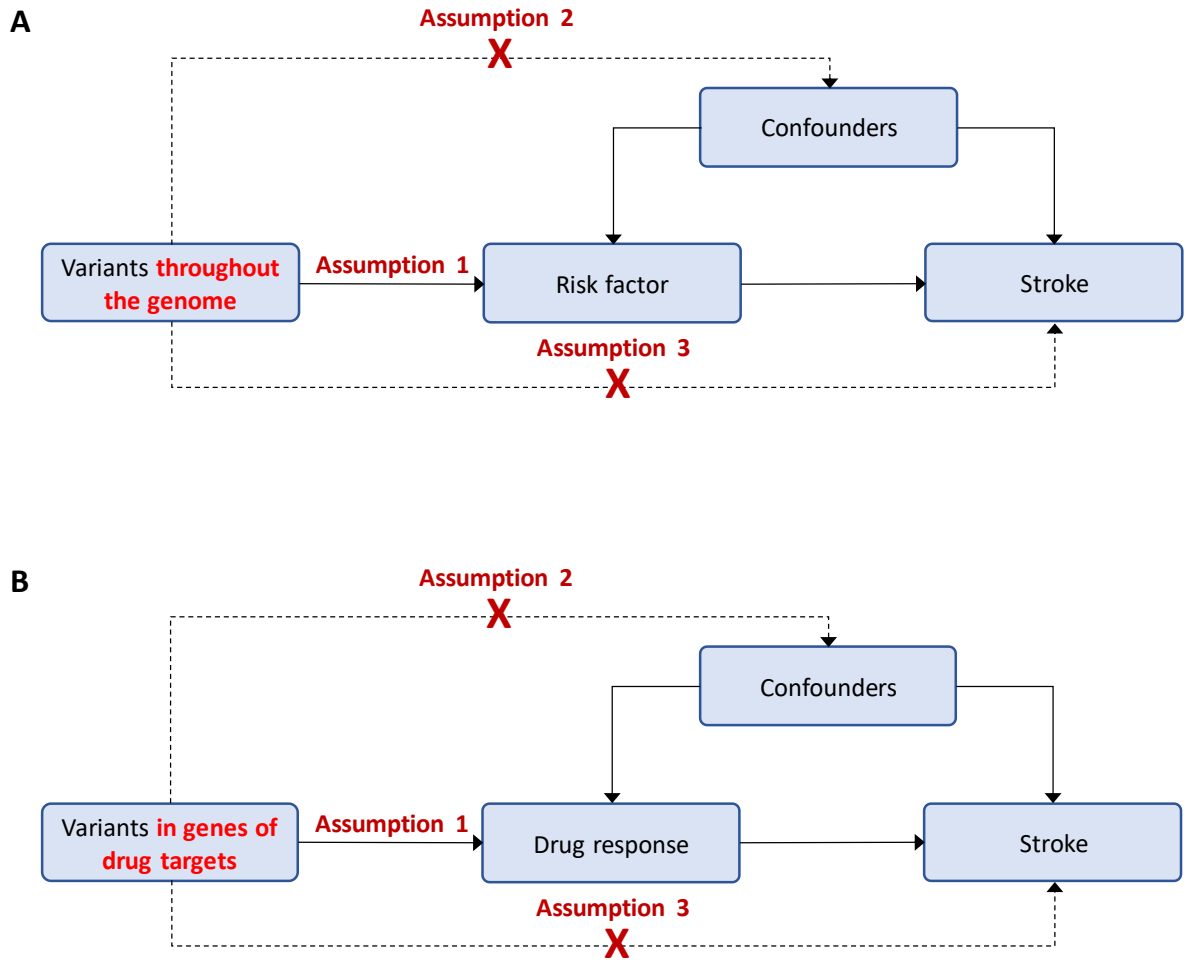


Figure 1. Exploration of associations of (A) potential risk factors and (B) drug target effects with risk of stroke using Mendelian randomization (MR). Schematic representations of the principles and assumptions of MR analyses. (A) By using genetic variants throughout the genome that associate with a risk factor we can generate causal association estimates with a disease outcome (e.g. stroke). (B) When restricting the selection of variants to those located in the vicinity of the gene encoding a protein drug target and associated with a downstream drug target effect, we can estimate the effects that a drug intervention could have on the studied disease outcome. In both occasions the assumptions are the same: the genetic variants (instruments) must be associated with the exposure (assumption 1); the variants must not be associated with confounders (assumption 2); the variants must influence the outcome only through the risk factor under study (assumption 3).

In the last decade, the expansion of GWASs to a broad range of phenotypes, the wide availability of summary data from these studies, and the development of statistical methods to utilize these data to perform MR analyses, have led to an explosion in the MR field (Bandres-Ciga *et al.*, 2019). MR may be used to explore the global effects of a risk factor on a disease, but might also offer insights about the efficacy of specific drug targets.

In the first case, MR uses genetic variants throughout the genome, which have been found in a GWAS to independently influence a risk factor, as instruments to proxy the effects of the risk factor (Burgess and Davey Smith, 2019). In the second approach, the selection of instruments may be restricted to the locus of a known or promising novel drug target (Mokry *et al.*, 2015). By then using genetic variants in this locus associated with the risk factor under study, MR may be used to study the effects of a specific target-exerted modification in the risk factor (Ference, 2018; Gill *et al.*, 2019). This approach has consistently been found to compute association estimates with disease endpoints that are comparable to those derived from RCTs (Ference, 2018; Roberts, 2018).

MR has been proven especially effective in establishing causal relationships and prioritizing drug targets for clinical study in the field of cardiovascular medicine. For example, in the absence of data from clinical trials, MR analyses using genetic variants that influence CRP circulating levels in the gene encoding CRP, showed that there is no evidence for a causal effect of CRP on risk of stroke, coronary artery disease, or other atherosclerotic phenotypes (Zacho *et al.*, 2008; Elliott *et al.*, 2009; Prins *et al.*, 2016; Ligthart *et al.*, 2018). Furthermore, MR provided evidence for a lack of a protective effect of low alcohol consumption on risk of stroke and coronary artery disease (Holmes *et al.*, 2014; Millwood *et al.*, 2019a). Regarding drug targets, in a series of studies using MR analyses, it was shown that lipid-lowering drug targets primarily exert their efficacy against coronary artery disease by influencing the levels of apolipoprotein B and LDL-C (Ference *et al.*, 2015; Ference *et al.*, 2017a; Ference *et al.*, 2017b; Ference *et al.*, 2018; Ference *et al.*, 2019).

However, as every methodological design, MR is based on specific assumptions and could be prone to bias, if these assumptions are violated. The primary assumptions are the following: (1) the genetic variants used as instruments must be associated with the risk factor under study; (2) these variants should affect the risk of the disease under study only through the explored risk factor and not through other independent mechanisms; (3) these variants should not be associated with known or unknown confounders that are associated both with the risk factor and the disease under study (Holmes *et al.*, 2017; Zheng *et al.*, 2017; Burgess and Davey Smith, 2019). While the first assumption relates to the quality and strength of the genetic instruments and might be relatively easy to assess with GWAS data, the second and third assumption might be violated if the variants used as instruments affect other phenotypes beyond the risk factor under study. Specifically,

pleiotropic genetic variants, i.e. variants influencing several phenotypes, might violate the latest two assumptions and if used as genetic instruments in an MR study might bias the results. The use of multiple genetic variants as instruments and the development of more advanced statistical approaches allow for the assessment of the probability of horizontal pleiotropy and the adjustment of the results (Holmes *et al.*, 2017; Zheng *et al.*, 2017; Burgess and Davey Smith, 2019). Furthermore, the use of alternative MR approaches with different underlying assumptions enables the exploration of the robustness of the findings (Burgess and Davey Smith, 2019). Studies using MR should therefore explore potential violations of the assumptions of the method and these requirements should be kept in mind when interpreting any MR results.

AIMS OF THE THESIS

While multiple risk factors have long been identified for stroke, the highly heterogeneous nature of the disease makes it extremely challenging to identify specific risk factors for the major etiological stroke subtypes. In light of largely variable mechanisms between stroke subtypes, there might further exist differences in the effects of approved drugs or treatments under development, which could have relevance for therapeutic decisions in specific patient subgroups.

The large number of events and the depth of phenotypic information available through GWASs permits exploration of outcomes for which there are no adequate data from observational studies or RCTs, such as stroke subtypes. Therefore, the overarching goal of the current PhD thesis was to leverage large-scale genetic data and apply MR analyses to (i) detect novel risk factors and promising drug targets for cerebrovascular disease and (ii) investigate the effects of known risk factors and drug targets for stroke as a whole on etiological stroke subtypes and cerebral SVD,

The following specific aims were addressed by the six individual studies included in the current PhD thesis:

(1) First, we used data from a large-scale GWAS investigating genetic determinants for the circulating levels of 41 cytokines and growth factors and implemented two-sample MR analyses to: (i) explore associations between genetic predisposition to higher or lower circulating cytokine levels with risk of any stroke; (ii) evaluate specific associations with ischemic stroke and its major etiologic subtypes (large artery stroke, cardioembolic stroke, and small vessel stroke), as well as with intracerebral hemorrhage; (iii) examine associations with etiologically related cardiovascular outcomes including coronary artery disease (CAD), myocardial infarction (MI), and atrial fibrillation (AF).

(2) Second, motivated by the results from this study, we performed a meta-analysis of 6 population-based cohorts encompassing 17,180 stroke-free individuals with long-term follow-up and aimed to: (i) determine the association between circulating MCP-1 levels and risk of incident stroke, (ii) explore associations of MCP-1 levels with risk of major

stroke subtypes (incident ischemic and hemorrhagic stroke); (iii) assess whether any association with stroke risk is independent of the IL-6 and CRP levels.

(3) Third, we identified and validated genetic proxies for downregulated IL-6 signaling on the basis of effects on upstream regulators and downstream effectors of the pathway aiming to: (i) explore associations of genetic predisposition to downregulated IL-6 signaling with risk of ischemic stroke and coronary artery disease; (ii) examine associations with major etiological subtypes of ischemic stroke (large artery, cardioembolic, and small vessel stroke); (iii) examine associations with a broad range of other cardiovascular phenotypes.

(4) Fourth, we set out to: (i) identify genetic variants within genes corresponding to the targets of common antihypertensive agents for hypertension that proxy the effects of these treatments; (ii) validate these variants by exploring their effects on coronary artery disease and stroke risk in MR and comparing them with those derived from RCTs; (iii) offer insights towards their adverse effect profiles and repurposing potential by undertaking a phenome-wide association study (PheWAS).

(5) Fifth, we used large-scale genetic data for blood pressure and the abovementioned genetic proxies for antihypertensive treatments with the aims to: (i) examine the effects of genetically determined SBP and DBP on the risk of etiological stroke subtypes; (ii) explore the effects of genetic proxies for different antihypertensive drug classes on etiological stroke subtypes; (iii) examine associations of these genetic proxies with the radiological phenotype of WMH, a manifestation of cerebral SVD etiologically related to small vessel stroke and ICH.

(6) Finally, we leveraged data from large GWASs on blood lipid levels, as well as on ischemic (small vessel stroke, WMH volume) and hemorrhagic (ICH) manifestations of cerebral SVD aiming to: (i) examine the effects of genetic determinants of levels of HDL-C, LDL-C, and triglycerides (TG) on SVD manifestations; (ii) explore associations between genetic determinants of lipoprotein particle fractions with these phenotypes; (iii) determine the effects of genetic predisposition to HDL-C-raising, LDL-C-lowering, and TG-lowering through variants in genes encoding lipid-modifying drug targets on SVD.

LIST OF PUBLICATIONS IN THE THESIS

(1) **Georgakis MK,** Gill D, Rannikmäe K, Traylor M, Anderson CD, Lee JM, Kamatani Y, Hopewell JC, Worrall BB, Bernhagen J, Sudlow CLM, Malik R, Dichgans M. Genetically Determined Levels of Circulating Cytokines and Risk of Stroke. ***Circulation***. 2019 Jan 8;139(2):256-268.
doi: 10.1161/CIRCULATIONAHA.118.035905. [PMID: 30586705]
Impact factor: 23.1

(2) **Georgakis MK,** Malik R, Björkbacka H, Pana TA, Demissie S, Ayers C, Elhadad MA, Fornage M, Beiser AS, Benjamin EJ, Boekholdt MS, Engström G, Herder C, Hoogeveen RC, Koenig W, Melander O, Orho-Melander M, Schiopu A, Söderholm M, Wareham N, Ballantyne CM, Peters A, Seshadri S, Myint PK, Nilsson J, de Lemos JA, Dichgans M. Circulating Monocyte Chemoattractant Protein-1 and Risk of Stroke: Meta-Analysis of Population-Based Studies Involving 17 180 Individuals. ***Circ Res***. 2019 Sep 27;125(8):773-782.
doi: 10.1161/CIRCRESAHA.119.315380. Epub 2019 Sep 3. [PMID: 31476962]
Impact factor: 15.9

(3) **Georgakis MK,** Malik R, Gill D, Franceschini N, Sudlow CLM, INVENT Consortium, CHARGE Inflammation Working Group, Dichgans M. Interleukin-6 signaling effects on ischemic stroke and other cardiovascular outcomes: a Mendelian Randomization study. [Preprint in MedRxiv – Under review].
doi: https://doi.org/10.1101/19007682.

(4) Gill D*, **Georgakis MK*,** Koskeridis F, Jiang L, Feng Q, Wei WQ, Theodoratou E, Elliott P, Denny JC, Malik R, Evangelou E, Dehghan A, Dichgans M†, Tzoulaki I†. Use of Genetic Variants Related to Antihypertensive Drugs to Inform on Efficacy and Side Effects. ***Circulation***. 2019 Jul 23;140(4):270-279.
doi: 10.1161/CIRCULATIONAHA.118.038814. Epub 2019 Jun 25. [PMID: 31234639], *,† equally contributed
Impact factor: 23.1

(5) **Georgakis MK*,** Gill D*, Webb AJS, Evangelou E, Elliott P, Sudlow CLM, Dehghan A, Malik R, Tzoulaki I†, Dichgans D†. Genetically determined blood pressure, antihypertensive drug classes, and risk of stroke subtypes. [In minor revision] ***Neurology.*** *,† equally contributed

Impact factor: 8.7

(6) **Georgakis MK,** Malik R, Anderson CD, Parhofer KG, Hopewell JC, Dichgans M. Genetically determined blood lipids and cerebral small vessel disease: role of HDL cholesterol. [Accepted] ***Brain***

Impact factor: 11.8

MANUSCRIPT I: Genetically Determined Levels of Circulating Cytokines and Risk of Stroke: Role of Monocyte Chemoattractant Protein-1

Georgakis MK, Gill D, Rannikmäe K, Traylor M, Anderson CD, Lee JM, Kamatani Y, Hopewell JC, Worrall BB, Bernhagen J, Sudlow CLM, Malik R*, Dichgans M*. Genetically Determined Levels of Circulating Cytokines and Risk of Stroke. ***Circulation***. 2019 Jan 8;139(2):256-268. * equally contributed

Author contributions: MKG, RM, and MD conceptualized and designed the study. MKG and RM performed the statistical analyses. All authors interpreted the results. MKG and MD drafted the manuscript. All authors critically revised the manuscript for intellectual content. All authors approved the submitted version and are accountable for the integrity of the work.

Circulation

ORIGINAL RESEARCH ARTICLE

Genetically Determined Levels of Circulating Cytokines and Risk of Stroke

Role of Monocyte Chemoattractant Protein-1

Marios K. Georgakis, MD
Dipender Gill, MD
Kristiina Rannikmäe, MD
Matthew Traylor, PhD
Christopher D. Anderson, MD
MEGASTROKE consortium of the International Stroke Genetics Consortium (ISGC)
Jin-Moo Lee, MD, PhD
Yoichiro Kamatani, MD, PhD
Jemma C. Hopewell, PhD
Bradford B. Worrall, MD
Jürgen Bernhagen, PhD
Cathie L. M. Sudlow, DPhil
Rainer Malik, PhD*
Martin Dichgans, MD*

BACKGROUND: Cytokines and growth factors have been implicated in the initiation and propagation of vascular disease. Observational studies have shown associations of their circulating levels with stroke. Our objective was to explore whether genetically determined circulating levels of cytokines and growth factors are associated with stroke and its etiologic subtypes by conducting a 2-sample Mendelian randomization (MR) study.

METHODS: Genetic instruments for 41 cytokines and growth factors were obtained from a genome-wide association study of 8293 healthy adults. Their associations with stroke and stroke subtypes were evaluated in the MEGASTROKE genome-wide association study data set (67 162 cases; 454 450 controls) applying inverse variance–weighted meta-analysis, weighted-median analysis, Mendelian randomization–Egger regression, and multivariable Mendelian randomization. The UK Biobank cohort was used as an independent validation sample (4985 cases; 364 434 controls). Genetic instruments for monocyte chemoattractant protein-1 (MCP-1/CCL2) were further tested for association with etiologically related vascular traits by using publicly available genome-wide association study data.

RESULTS: Genetic predisposition to higher MCP-1 levels was associated with higher risk of any stroke (odds ratio [OR] per 1 SD increase, 1.06; 95% CI, 1.02–1.09; *P*=0.0009), any ischemic stroke (OR, 1.06; 95% CI, 1.02–1.10; *P*=0.002), large-artery stroke (OR, 1.19; 95% CI, 1.09–1.30; *P*=0.0002), and cardioembolic stroke (OR, 1.14; 95% CI, 1.06–1.23; *P*=0.0004), but not with small-vessel stroke or intracerebral hemorrhage. The results were stable in sensitivity analyses and remained significant after adjustment for cardiovascular risk factors. Analyses in the UK Biobank showed similar associations for available phenotypes (any stroke: OR, 1.08; 95% CI, 0.99–1.17; *P*=0.09; any ischemic stroke: OR, 1.07; 95% CI, 0.97–1.18; *P*=0.17). Genetically determined higher MCP-1 levels were further associated with coronary artery disease (OR, 1.04; 95% CI, 1.00–1.08; *P*=0.04) and myocardial infarction (OR, 1.05; 95% CI, 1.01–1.09; *P*=0.02), but not with atrial fibrillation. A meta-analysis of observational studies showed higher circulating MCP-1 levels in patients with stroke in comparison with controls.

CONCLUSIONS: Genetic predisposition to elevated circulating levels of MCP-1 is associated with higher risk of stroke, in particular with large-artery stroke and cardioembolic stroke. Whether targeting MCP-1 or its receptors can lower stroke incidence requires further study.

*Drs Malik and Dichgans jointly supervised this work.

Key Words: atherosclerosis ■ chemokine CCL2 ■ cytokines ■ human genetics ■ inflammation ■ Mendelian randomization analysis ■ stroke

Sources of Funding, see page 265

https://www.ahajournals.org/journal/circ

Clinical Perspective

What Is New?

- Genetic predisposition to higher circulating levels of monocyte chemoattractant protein-1 was associated with higher risk of stroke.
- Associations were also found for etiologic stroke subtypes, specifically large-artery stroke and cardioembolic stroke.
- Genetically determined levels monocyte chemoattractant protein-1 also associated with higher risk of the related phenotypes of coronary artery disease and myocardial infarction.

What Are the Clinical Implications?

- Additional work is needed to determine whether targeting monocyte chemoattractant protein-1 or its downstream effectors is a meaningful strategy for lowering stroke risk.

Stroke is the leading cause of long-term disability and the second most common cause of death worldwide,[1,2] with a growing burden on global health.[3] Inflammatory mechanisms have been implicated in stroke and etiologic stroke subtypes,[4–6] and specifically demonstrated for large-artery atherosclerotic stroke.[4,5] Cytokines and growth factors regulate the inflammatory response[4] and thus may serve as targets for cardiovascular disease prevention.[7] Indeed, the CANTOS trial (Canakinumab Anti-Inflammatory Thrombosis Outcome Study) recently demonstrated the potential of targeting specific inflammatory cytokines in reducing vascular end points.[8]

Few studies have investigated associations between the circulating levels of inflammatory cytokines and risk of stroke. Levels of interleukin (IL)–1β and IL-6 were found to be associated with incident and recurrent ischemic stroke.[4] However, these associations derived from observational studies preclude conclusions about causal relationships because of possible confounding and reverse causation.[9] Also, associations with etiologic stroke subtypes were not investigated in depth.[4] Hence, the potential causative role of individual cytokines in determining stroke risk remains elusive. Developing meaningful strategies for stroke prevention will require defining these relationships.[10]

Mendelian randomization (MR) aims to overcome the limitations of conventional epidemiological studies with respect to confounding and reverse causation. By using genetic variants as instrumental variables for a trait, MR enables an investigation of associations independent of the conventional biases accompanying observational studies.[11] A recent genome-wide association study (GWAS) in 8293 healthy subjects of Finnish ancestry identified multiple common genetic variants that influence circulating levels of 41 cytokines and growth factors (referred to hereafter as cytokines for simplicity),[12] thus providing comprehensive data on genetic determinants of circulating inflammatory biomarkers.[12]

Here, by leveraging data from this recent GWAS on cytokines[12] and the largest GWAS meta-analysis on stroke and stroke subtypes to date,[13] we implemented a 2-sample MR study to (1) explore the associations between genetic predisposition to higher or lower circulating cytokine levels with risk of any stroke; (2) evaluate specific associations with ischemic stroke and its major etiologic subtypes (large-artery stroke, cardioembolic stroke, and small-vessel stroke), and with intracerebral hemorrhage, as well; (3) validate these findings in UK Biobank as an independent cohort; (4) compare the MR associations with estimates of association derived from meta-analyses of observational studies; and (5) examine the association with etiologically related cardiovascular outcomes including coronary artery disease (CAD), myocardial infarction (MI), and atrial fibrillation (AF).

METHODS

Access to Publicly Available Data

The analyses for this study were based on publicly available summary statistics from GWAS consortia. The web links for downloading the data are provided in Table I in the online-only Data Supplement along with descriptive characteristics of the consortia. The retrieved summary data for the current analysis and the code script are available on reasonable request to the corresponding author. Because all analyses have been based on publicly available summary statistics and not individual-level data, no ethical approval from an institutional review board was required.

Study Design and Data Sources

The overall design of this study is displayed in Figure 1. Table I in the online-only Data Supplement summarizes our data sources for this MR study. The genetic instruments were taken from publicly available summary statistics.[12] For each of the 41 cytokines (full list provided in Table II in the online-only Data Supplement) we selected single-nucleotide polymorphisms (SNPs) associated with their circulating levels at a significance threshold of a false discovery rate <5%.[14] To avoid bias by selection of false-positive instruments, we performed additional analyses using a genome-wide threshold of significance ($P<5\times10^{-8}$). After extracting the summary statistics for significant SNPs, we pruned all SNPs in linkage disequilibrium ($r^2<0.1$ in the European 1000 Genomes Project reference panel), retaining SNPs with the lowest P value as an independent instrument. We identified 698 SNPs not in linkage disequilibrium to be significantly associated with circulating cytokine levels; 615 of them were also available in the MEGASTROKE data set. To avoid the use of pleiotropic instruments, we excluded 126 SNPs that were associated with levels of >1 cytokine,[15] leaving 489 SNPs as the final instruments. These instruments related to the circulating levels of

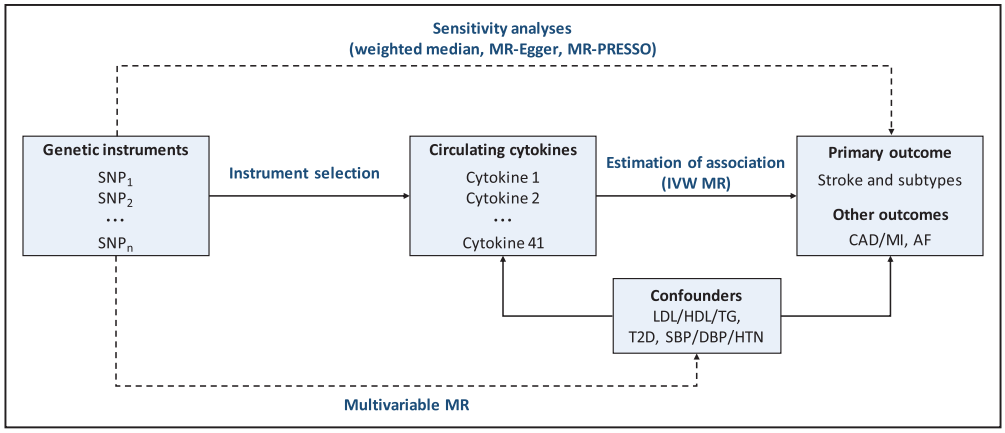


Figure 1. Schematic representation of the study design.
Methods used to test for associations and for violations of the Mendelian randomization assumptions (dashed lines). AF indicates atrial fibrillation; CAD, coronary artery disease; DBP, diastolic blood pressure; HDL, high-density lipoprotein; HTN, hypertension; IVW, inverse variance–weighted; LDL, low-density lipoprotein; MI, myocardial infarction; MR, Mendelian randomization; MR-PRESSO, Mendelian Randomization Pleiotropy Residual Sum and Outlier; SBP, systolic blood pressure; SNP, single-nucleotide polymorphism; T2D, type 2 diabetes mellitus; and TG, triglyceride.

23 cytokines, whereas for 18 cytokines, no SNPs associated with their circulating levels at a significance level of false discovery rate <5% could be identified.

The primary outcomes for this study were any stroke, any ischemic stroke, etiologic ischemic stroke subtypes defined by TOAST criteria (Trial of Org 10172 in Acute Stroke Treatment) (large-artery stroke, cardioembolic stroke, and small-vessel stroke),[16] and intracerebral hemorrhage. We extracted estimates for the associations of the selected instruments with any stroke, any ischemic stroke and its subtypes from the MEGASTROKE multiancestry GWAS data set (67 162 cases; 454 450 controls).[13] Sensitivity analyses restricted to individuals of European ancestry (40 528 cases; 445 396 controls) were conducted, to minimize ancestral mismatch with the Finnish population used for the discovery GWAS on cytokines.[12] For intracerebral hemorrhage, we extracted data from publicly available summary statistics of a GWAS meta-analysis on 1545 cases and 1481 controls of European ancestry.[17]

We computed *F* statistics to quantify the strength of the selected instruments[18] and performed power calculations.[19] The *F* statistic for the 489 instrument SNPs ranged from 17 to 789 (Table III in the online-only Data Supplement), well above the threshold of *F*>10 typically recommended for MR analyses.[20] Based on the sample size of MEGASTROKE, there was >80% power to detect significant associations with any stroke and any ischemic stroke for 18 of 23 cytokines at an effect size (odds ratio [OR]) of 1.10. Power was lower for the remaining 5 cytokines and for subanalyses for ischemic stroke subtypes and intracerebral hemorrhage (Table III in the online-only Data Supplement).

For validation of significant associations in MEGASTROKE, we used the UK Biobank data set as detailed in the Methods in the online-only Data Supplement. We included cases of prevalent and incident stroke. Cases with an unconfirmed self-reported diagnosis of stroke were excluded from the analysis. The final sample size consisted of 369 419 individuals, including 4985 patients with any stroke and 3628 patients with any ischemic stroke. No data were available on ischemic stroke subtypes.

Cytokines that were significantly associated with stroke were subsequently explored for an association with etiologically related vascular outcomes. Publicly available summary statistics were extracted from the CARDIoGRAMplusC4D (Coronary Artery Disease Genome wide Replication and Meta-analysis [CARDIoGRAM] plus The Coronary Artery Disease [C4D] Genetics) consortium for CAD and MI (60 801 CAD and 43 676 MI cases; 123 504 controls),[21] and the AFGen (Atrial Fibrillation Genetics) consortium for AF (17 931 cases; 115 142 controls).[22]

Statistical Analysis

After extraction of data and harmonization of the effect alleles across GWASs, we computed individual MR estimates and standard errors from the SNP-cytokine and SNP-outcome associations using the Wald estimator and the Delta method that weight all estimates based on the magnitude of the SNP-cytokine association.[23] The MR association between each cytokine and stroke was estimated after pooling individual SNP MR estimates using fixed-effects inverse variance–weighted (IVW) meta-analysis.[23] Statistical significance for the MR associations with stroke was set at a *P* value corrected for multiple comparisons (based on the number of cytokines) using the Bonferroni method. We further report on results corrected for both the number of cytokines and the number of examined phenotypes. A *P*<0.05, but above the Bonferroni-corrected threshold, was considered as suggestive for association. The IVW MR approach assumes that instruments affect the outcome only through the exposure under consideration, and not by some alternative pathway.[23] Any violation of this assumption would represent horizontal pleiotropy of the instrument and could introduce bias to the MR estimate. In the absence of any such horizontal pleiotropy, there would not be any expected heterogeneity in the MR estimates obtained from different instruments. As such, heterogeneity markers (I^2>25% or Cochran *Q*-derived *P*<0.05) from the IVW MR were used as indicators of possible horizontal pleiotropy.[24]

For cytokines showing either significant or suggestive associations or significant heterogeneity in the primary IVW MR analysis, we conducted additional sensitivity analyses that vary

in their underlying assumptions regarding the presence of pleiotropic genetic variants that may be associated with the outcome independently of the exposure. In particular, we used MR-Egger regression, which requires that the strengths of the instruments are independent of their direct associations with the outcome,[25] and the weighted median method, which requires that at least half of the information for the MR analysis comes from valid instruments.[26] We used the intercept obtained from the MR-Egger regression as a measure of directional pleiotropy (P<0.05 was considered significant),[25] and also tested for outlier SNPs using MR-Pleiotropy Residual Sum and Outlier.[27]

To generate MR estimates unaffected by the presence of pleiotropic pathways acting through cardiovascular risk factors, we performed regression-based multivariable MR with summary genetic association estimates[28] that adjusted for the genetic association of instruments with circulating lipid levels (low-density lipoprotein cholesterol, high-density lipoprotein cholesterol, triglycerides), type 2 diabetes mellitus, and blood pressure measurements (systolic and diastolic blood pressure, hypertension). Genetic association estimates for these phenotypes were extracted from the GLGC (Global Lipids Genetic Consortium),[29] the DIAGRAM consortium (Diabetes Genetics Replication and Meta-Analysis),[30] and the UK Biobank,[31] published by the Neale laboratory, respectively.

Instrument SNPs for cytokines showing significant associations with stroke were mapped to the nearest gene by using the GRCh37/hg19 reference genome. We used the STRING database (Search Tool for the Retrieval of Interacting Genes)[32] to look for protein-protein interactions between gene products and the cytokines and identified interacting subnetworks. As a sensitivity analysis, and to gain further insight into the biological processes involved in the examined associations, we performed IVW MR analyses with SNPs restricted to the specific subnetworks.

The GWAS used to select cytokine instruments included no replication, and its estimates of association were further adjusted for body mass index, besides age and sex.[12] As a sensitivity analysis for bias that may be introduced by this body mass index adjustment,[33] we also calculated an unweighted allele score for any cytokines demonstrating a significant association in our main IVW MR analysis.[34] Such an unweighted allele score may offer evidence of a causal effect of the exposure on the outcome without suffering from bias in the genetic association estimates for the exposure, although this is at the cost of not being able to estimate the magnitude of any such effect.[34] Statistical analyses were conducted in Stata 13.1 (StataCorp).

Meta-Analysis of Observational Studies

For the cytokines that showed significant associations with stroke in MR, we performed a meta-analysis of observational studies. We searched Medline until December 10, 2017 (search strategy is available in the Methods in the online-only Data Supplement), for case-control studies comparing the circulating cytokine levels between patients with stroke and controls, and cohort studies exploring the association of baseline levels with incident or recurrent stroke. We extracted relevant data and applied random-effects meta-analyses for hazard ratios (cohort studies) or standardized mean differences (case-control studies). We evaluated heterogeneity with the I^2 and the Cochran Q.

RESULTS

Genetically Determined Circulating Levels of Cytokines and Risk of Stroke

The primary results of the MR analyses for the 23 cytokines are presented in Figure 2. Following Bonferroni correction for testing multiple cytokines (P<0.05/23=2.2×10^{-3}), the only cytokine showing sta-

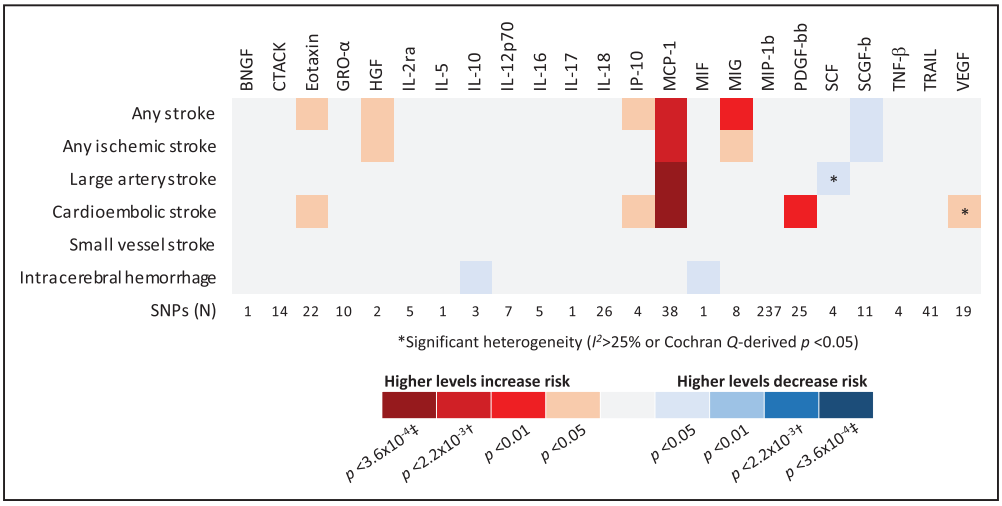


Figure 2. Mendelian randomization associations of circulating cytokine and growth factor levels with stroke and stroke subtypes.
Shown are the results derived from the fixed-effects inverse variance weighted meta-analysis. BNGF indicates beta nerve growth factor; CTACK, cutaneous T-cell-attracting chemokine; GRO-α, growth-regulated oncogene alpha; HGF, hepatocyte growth factor; IL, interleukin; IP-10, interferon gamma-induced protein 10 MCP-1, monocyte chemoattractant protein-1; MIF, macrophage migration inhibitory factor; MIG, monokine induced by gamma interferon; MIP-1b, macrophage inflammatory protein 1 beta; PDGF-bb, platelet-derived growth factor-bb; SCF, stem cell factor; SCGF-b, stem cell growth factor beta; SNP, single-nucleotide polymorphism; TNF, tumor necrosis factor; TRAIL, TNF-related apoptosis-inducing ligand; and VEGF, vascular endothelial growth factor. *Significant heterogeneity (I^2>25% or Cochran Q-derived P<0.05). †Bonferroni-corrected threshold for number of tested cytokines. ‡Bonferroni-corrected threshold for number of cytokines and number of phenotypes.

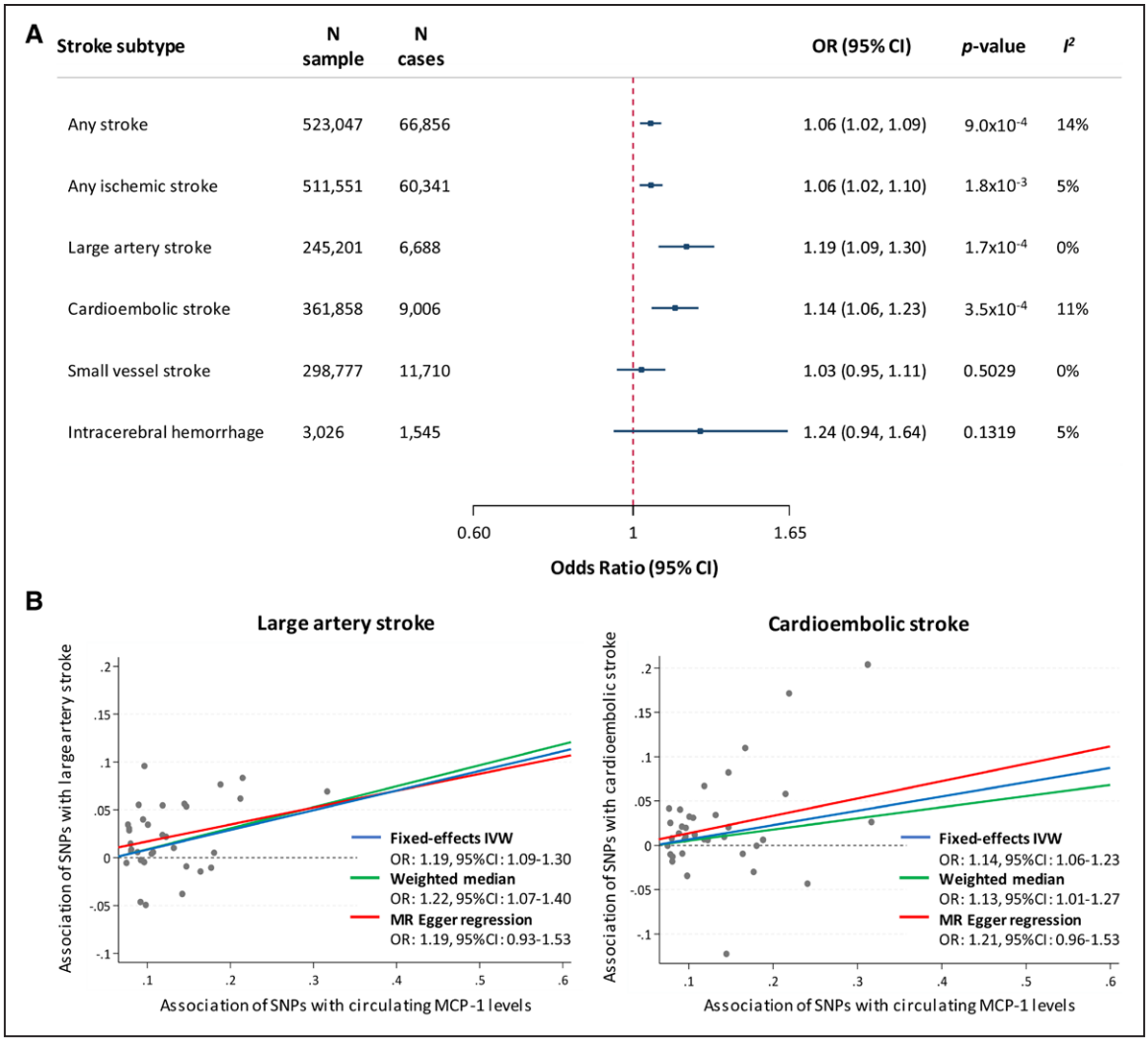


Figure 3. Mendelian randomization analysis for circulating MCP-1 levels and risk of stroke.
A, MR-derived associations between genetically determined circulating MCP-1 levels (1 SD increase) and risk of any stroke and stroke subtypes. **B**, Associations between genetically determined circulating MCP-1 levels and risk of large-artery (**left**) and cardioembolic (**right**) stroke based on different MR methods. I^2 refers to heterogeneity in the Mendelian randomization analysis (inverse variance–weighted method). IVW indicates inverse variance–weighted; MCP-1, monocyte chemoattractant protein-1; MR, Mendelian randomization; OR, odds ratio; and SNP, single-nucleotide polymorphism.

tistically significant associations with stroke was the CC chemokine monocyte chemoattractant protein-1 (MCP-1/CCL2). As depicted in Figure 3A and Figure I in the online-only Data Supplement, genetically determined higher circulating MCP-1 levels (1 SD increase) were associated with 6% higher odds for both any stroke (OR, 1.06; 95% CI, 1.02–1.09; $P=9\times10^{-4}$; 523 047 individuals; 66 856 cases) and any ischemic stroke (OR, 1.06; 95% CI, 1.02–1.10; $P=1.8\times10^{-3}$; 511 551 individuals; 60 341 cases) in MR analyses. Corresponding analyses for ischemic stroke subtypes revealed significant associations for large-artery stroke (OR, 1.19; 95% CI, 1.09–1.30; $P=1.7\times10^{-4}$; 245 201 individuals; 6688 cases) and cardioembolic stroke (OR, 1.14; 95% CI, 1.06–1.23; $P=3.5\times10^{-4}$; 361 858 individuals; 9006 cases), but not for small-vessel stroke (OR, 1.03; 95% CI, 0.95–1.11; $P=0.50$; 298 777 individuals; 11 710 cases). In addition, we found no significant association of genetically determined MCP-1 levels with intracerebral hemorrhage (OR, 1.24; 95% CI, 0.94–1.64; $P=0.13$), although this might be related to the lower sample size (3026 individuals; 1545 cases). It is important to note that the results for large-artery stroke and cardioembolic stroke remained significant when further correcting for both the number of examined cytokines and the number of examined phenotypes ($P<0.05/138=3.6\times10^{-4}$; Figure 2). Subanalyses restricted to lobar (OR, 1.25; 95% CI, 0.88–1.79; $P=0.22$; 2145 individuals; 664 cases) and nonlobar intracerebral hemorrhage (OR, 1.03; 95% CI, 0.72–1.49; $P=0.16$; 2362 individuals; 881 cases) also showed no significant associations with genetically determined MCP-1 levels. The individual SNPs associated with MCP-1 levels explained 14.7% of the variance of MCP-1 levels (Table III in the online-only Data Supplement) and are presented in Table IV in the online-only Data Supplement.

There was no evidence for heterogeneity in any of the MCP-1 associations as measured by I^2 and Cochran Q (Figure 3A), and no outlier SNPs were detected with the MR-Pleiotropy Residual Sum and Outlier method. Also, there was no indication for directional pleiotropy effects as assessed by the MR-Egger intercept (any stroke, P=0.41; any ischemic stroke, P=0.39; large-artery stroke, P=0.98; cardioembolic stroke, P=0.67; small-vessel stroke, P=0.70; intracerebral hemorrhage, P=0.94). The weighted median estimator and the MR-Egger regression analysis provided estimates of the same magnitude as the fixed-effects IVW meta-analysis for large-artery stroke (OR, 1.22; 95% CI, 1.07–1.40; $P=2\times10^{-3}$ and OR, 1.19; 95% CI, 0.93–1.53; P=0.13, respectively) and cardioembolic stroke (OR, 1.13; 95% CI, 1.01–1.27; P=0.04 and OR, 1.21; 95% CI, 0.96–1.53; P=0.09, respectively, Figure 3B), although with wider confidence intervals as would be expected given the lower statistical power of these approaches.[25,26] Use of an unweighted allele score for the MCP-1 instrument SNPs also showed statistically significant associations with risk of large-artery ($P=1.5\times10^{-4}$) and cardioembolic stroke ($P=2.8\times10^{-4}$). The significant association between MCP-1 and outcomes was retained both when restricting the analysis to individuals of European ancestry (Figure II in the online-only Data Supplement), and when applying the more conservative threshold of $P<5\times10^{-8}$ for instrument selection (Figure III in the online-only Data Supplement).

To explore whether the MR association between genetically determined MCP-1 levels and stroke was attributable through pleiotropic pathways relating to cardiovascular risk factors, we conducted multivariable MR analysis adjusting for circulating lipid levels, type 2 diabetes mellitus, and blood pressure. The results remained stable regardless of the model (unadjusted, single, or fully adjusted model), thus supporting an independent association between MCP-1 levels and stroke and stroke subtypes (Table).

None of the genetic instruments for MCP-1 was within or close to the *MCP1* gene. Assessing genes closest to the instruments for MCP-1, we noted that several of them encoded proteins that show a biological relationship with MCP-1, eg, CCR2, the main receptor for MCP-1 (Table IV in the online-only Data Supplement). To minimize the risk of using nonspecific instruments that might exert pleiotropic effects, we performed an additional sensitivity analysis focusing on instruments in the vicinity of these genes. Using the STRING database, we found the chemokine receptors CCR2, CCR1, CCR3, and CCR9, the chemokine-binding protein CCBP2, and the receptor of the complement C5a (C5aR1) to integrate into a subnetwork of established interactions with MCP-1 (Figure IVA in the online-only Data Supplement). Restricting the MR analysis to the respective SNPs resulted in significant estimates of association for large-artery (OR per 1 SD increase in MCP-1 levels, 1.25; 95% CI, 1.08–1.45; $P=2\times10^{-3}$) and cardioembolic stroke (OR, 1.21; 95% CI, 1.07–1.37; $P=3\times10^{-3}$), and intracerebral hemorrhage, as well (OR, 2.19; 95% CI, 1.30–3.69; $P=3\times10^{-3}$) (Figure IVB in the online-only Data Supplement).

Several other cytokines not reaching the Bonferroni-corrected threshold showed suggestive (P<0.05) associations with risk of stroke in MR analyses: genetic predisposition to higher levels of eotaxin, interferon gamma-induced protein 10, monokine induced by gamma interferon, platelet-derived growth factor-bb, and vas-

Table. Multivariable Mendelian Randomization Associations Between Circulating MCP-1 Levels and Risk of Stroke and Its Subtypes Adjusting for Cardiovascular Risk Factors

Model	Any Stroke	Any Ischemic Stroke	Large-Artery Stroke	Cardioembolic Stroke	Small-Vessel Stroke	Intracerebral Hemorrhage
No. in sample	523 047	511 551	245 201	361 858	298 777	3026
N. of cases	66 856	60 341	6688	9006	11 710	1545
Unadjusted model	1.06 (1.02–1.09)	1.06 (1.02–1.10)	1.19 (1.09–1.30)	1.14 (1.06–1.23)	1.03 (0.95–1.11)	1.24 (0.94–1.64)
Adjusted for T2D	1.07 (1.03–1.11)	1.07 (1.03–1.11)	1.22 (1.12–1.33)	1.17 (1.08–1.27)	1.03 (0.97–1.10)	1.06 (0.94–1.20)
Adjusted for LDL-C	1.06 (1.02–1.10)	1.06 (1.02–1.11)	1.20 (1.10–1.31)	1.16 (1.06–1.24)	1.03 (0.98–1.09)	1.26 (0.93–1.71)
Adjusted for HDL-C	1.07 (1.03–1.11)	1.07 (1.02–1.11)	1.21 (1.11–1.33)	1.15 (1.06–1.25)	1.04 (0.97–1.10)	1.27 (0.94–1.72)
Adjusted for TG	1.06 (1.02–1.10)	1.06 (1.02–1.10)	1.19 (1.09–1.30)	1.16 (1.06–1.26)	1.03 (0.97–1.10)	1.28 (0.94–1.73)
Adjusted for SBP	1.08 (1.04–1.12)	1.09 (1.05–1.14)	1.23 (1.12–1.35)	1.20 (1.10–1.32)	1.03 (0.96–1.11)	1.81 (1.13–1.90)
Adjusted for DBP	1.08 (1.04–1.13)	1.09 (1.05–1.14)	1.22 (1.11–1.34)	1.20 (1.10–1.32)	1.04 (0.96–1.11)	1.53 (0.89–2.65)
Adjusted for HTN	1.07 (1.03–1.11)	1.07 (1.03–1.11)	1.19 (1.09–1.29)	1.18 (1.08–1.29)	1.03 (0.95–1.11)	1.03 (0.93–1.14)
Fully adjusted model (T2D, LDL-C*, SBP†)	1.08 (1.03–1.12)	1.09 (1.04–1.13)	1.23 (1.11–1.35)	1.20 (1.10–1.32)	1.04 (0.97–1.12)	1.06 (0.92–1.21)

The results are presented as odds ratios (95% CIs) for the effect of 1 SD increase in MCP-1 levels. DBP indicates diastolic blood pressure; HDL-C, high-density lipoprotein cholesterol; HTN, hypertension; LDL-C, low-density lipoprotein cholesterol; MCP-1, monocyte chemoattractant protein-1; SBP, systolic blood pressure; T2D, type 2 diabetes mellitus; and TG, triglyceride.

*Restricted to LDL-C to avoid collinearity with HDL-C and TG levels.

†Restricted to SBP to avoid collinearity with DBP and HTN.

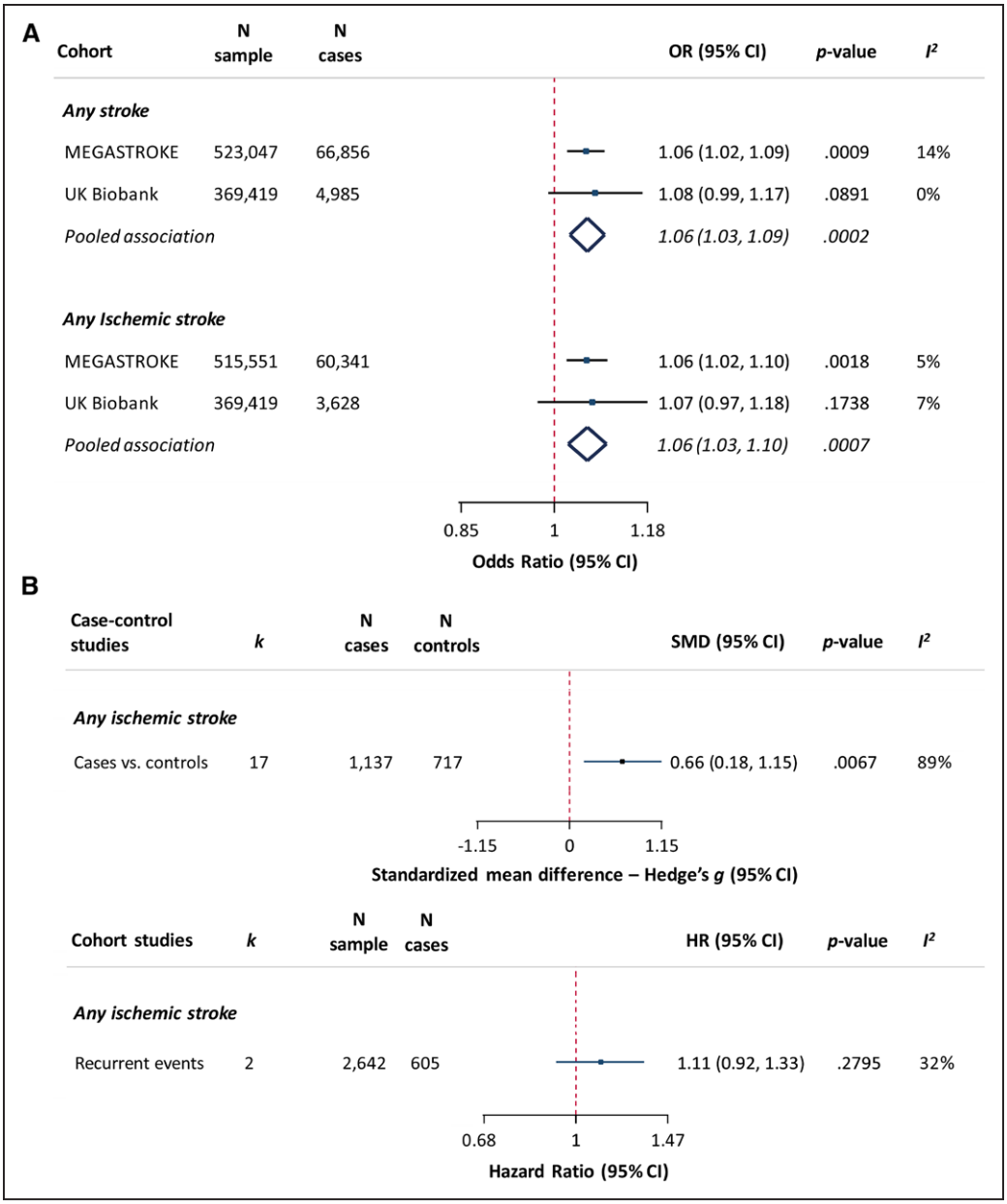


Figure 4. Associations between circulating MCP-1 levels and risk of stroke in Mendelian randomization and in observational studies.
A, MR-derived associations between genetically determined circulating MCP-1 levels (1 SD increase) and risk of any stroke and any ischemic stroke in MEGASTROKE, in UK Biobank, and a meta-analysis of both samples. **B**, Meta-analysis–derived associations between circulating MCP-1 levels (1 SD increase) and risk of ischemic stroke in case-control and cohort studies. *k* refers to number of included studies. I^2 in Figure 4A refers to heterogeneity in the MR analysis (inverse variance weighted method), and, in Figure 4B, to heterogeneity in the random-effects meta-analyses of observational studies. HR indicates hazard ratio; MCP-1, monocyte chemoattractant protein-1; MR, Mendelian randomization; OR, odds ratio; and SMD, standardized mean difference.

cular endothelial growth factor were associated with an higher risk of stroke, whereas predisposition to higher levels of stem cell factor and stem cell growth factor beta were associated with lower risk of stroke (Figure 2).

Genetically Determined Circulating Levels of MCP-1 and Risk of Stroke in UK Biobank

We next explored the MR association between genetically determined MCP-1 levels and risk of any stroke and risk of any ischemic stroke in the independent UK Biobank sample and meta-analyzed the MEGASTROKE and UK Biobank data (Figure 4A and Figure V in the online-only Data Supplement). Estimates of association in UK Biobank were similar to MEGASTROKE for any stroke (OR per 1 SD increase, 1.08; 95% CI, 0.99–1.17; *P*=0.09; 369419 individuals, 4985 cases) and any ischemic stroke (OR, 1.07; 95% CI, 0.97–1.18; *P*=0.17; 369419, 3628 cases), but did not reach statistical significance. Genetically elevated circulating MCP-1 levels were significantly associated with both any stroke (OR,

1.06; 95% CI, 1.03–1.09; $P=2\times10^{-4}$) and any ischemic stroke (OR, 1.06; 95% CI, 1.03–1.10; $P=7\times10^{-4}$) in the meta-analysis of MEGASTROKE and UK Biobank.

Circulating Levels of MCP-1 and Risk of Stroke: Meta-Analysis of Observational Studies

Next, we compared the MR estimates with those derived from a meta-analysis of observational studies. Our search yielded 17 case-control studies of patients with ischemic stroke and controls, 2 cohort studies on patients with a history of stroke or cardiovascular disease exploring the risk of recurrent ischemic stroke, and 1 case-cohort study of incident ischemic stroke in a community population (Tables V and VI in the online-only Data Supplement and Figure VI in the online-only Data Supplement). Patients with any ischemic stroke were found to have significantly higher MCP-1 levels than controls in the case-control studies (Hedges' *g*: 0.66; 95% CI, 0.18–1.15 [corresponding to a medium to strong effect size[35]]; 1137 cases, 717 controls; heterogeneity: I^2=89%, P<0.001; Figure 4B and Figure VIIA in the online-only Data Supplement). Studies on recurrent stroke (2642 individuals, 605 events) yielded a hazard ratio of 1.11 (95% CI, 0.92–1.33) for 1 SD increase in MCP-1 levels (heterogeneity: I^2=32%, P=0.28; Figure 4B and Figure VIIB in the online-only Data Supplement), whereas the single study examining incident ischemic stroke (95 cases, 190 controls) reported a hazard ratio of 0.99 (95% CI, 0.68–1.45).

Genetically Determined Circulating Levels of MCP-1 and Etiologically Related Vascular Outcomes

Figure 5 depicts the MR association between genetically determined MCP-1 levels and risk of CAD, MI, and AF. Genetic predisposition to higher MCP-1 levels was associated with CAD (OR per 1 SD increase, 1.04; 95% CI, 1.00–1.08; P=0.04; 184 305 individuals, 60 801 cases) and MI (OR, 1.05; 95% CI, 1.01–1.09; P=0.02; 167 180 individuals, 43 676 cases). Given the association of MCP-1 with cardioembolic stroke, we further explored the relationship between genetically determined MCP-1 levels and risk of AF in MR analysis, but found no association (OR, 0.96; 95% CI, 0.91–1.01; P=0.09).

DISCUSSION

Exploring 41 cytokines in a 2-sample MR approach involving the largest GWAS data sets available, we found that genetic predisposition to higher levels of MCP-1/CCL2 is associated with higher risk of any stroke, any ischemic stroke, large-artery stroke, and cardioembolic stroke. The results were stable in alternative MR methods and sensitivity analyses and remained significant after adjustment for cardiovascular risk factors. Moreover, effect sizes for any stroke and any ischemic stroke were similar in the UK Biobank. We further found associations between genetic predisposition to higher MCP-1 levels and higher risk of CAD and MI as etiologically related outcomes. Collectively, our findings suggest that lifelong elevated circulating MCP-1 levels increase the risk of stroke.

The directionality of the MR association between genetically determined levels of MCP-1 and risk of large-artery stroke is consistent with experimental data showing a key role for this chemokine in atherogenesis and atheroprogression. Acting mainly through its receptor CCR2, MCP-1 is the prototypical CC family chemokine that is upregulated by chronic inflammatory conditions and attracts monocytes to the subendothelial space of the atherogenic arterial wall.[36] Mice lacking MCP-1[37] or CCR2[38] are less susceptible to atherosclerosis, and anti–MCP-1 gene therapy,[39] MCP-1 competitors,[40] and CCR2

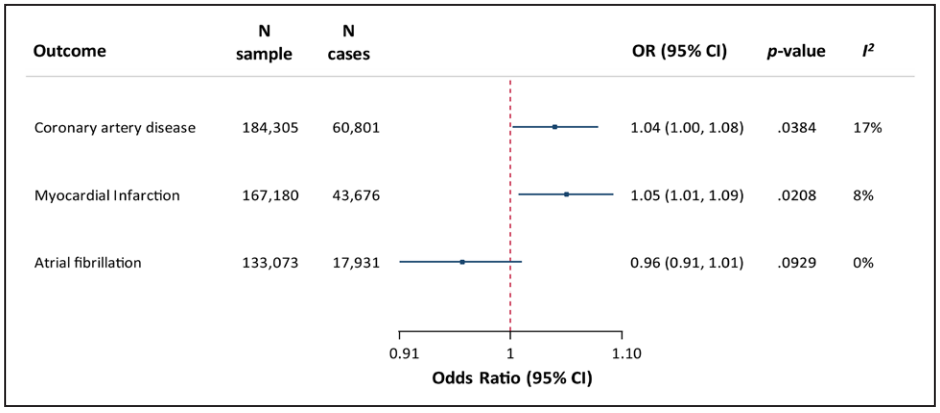


Figure 5. Mendelian randomization (MR) analysis for genetically determined circulating MCP-1 levels and etiologically related vascular outcomes. MR-derived associations between genetically determined circulating MCP-1 levels (1 SD increase) and risk of coronary artery disease, myocardial infarction, and atrial fibrillation. I^2 refers to heterogeneity in the MR analysis (inverse variance weighted method). MCP-1 indicates monocyte chemoattractant protein-1; and OR, odds ratio.

antagonists[41] reduce plaque size and inhibit plaque progression and destabilization in experimental atherosclerosis. Conversely, overexpression of MCP-1 leads to inflammation, accumulation of lipids, and smooth muscle cell proliferation in atherosclerotic plaques.[42]

We further found an MR association between genetic predisposition to higher MCP-1 levels and risk of cardioembolic stroke. Genetic predisposition to higher MCP-1 levels is associated with higher risk of CAD and MI, which could promote the formation of left ventricular thrombus from myocardial damage, thus resulting in cardioembolic stroke. Furthermore, MCP-1 has been reported to promote myocardial fibrosis,[43] an established risk factor for AF.[44] However, we found no association between the genetic instruments for MCP-1 and AF risk. Other investigators have found an association between circulating MCP-1 levels and the presence of atrial thrombi in patients with AF.[45] Hence, it might be that MCP-1 increases the risk of cardioembolic stroke by promoting thrombus formation in patients with established AF. Alternative explanations for the association between circulating MCP-1 levels and cardioembolic stroke might include less frequent causes of cardioembolism, such as valvular disease and the misclassification of patients with multiple competing stroke etiologies including atherosclerosis.

In contrast, our analysis provides no evidence for an association of genetically determined MCP-1 levels with small-vessel stroke even though the sample size was larger than for other stroke subtypes. In fact, we found none of the cytokines to be associated with small-vessel stroke (all $P>0.05$, Figure 2). Overall, these observations agree with the notion that inflammatory processes are less important in small-vessel disease than in large-artery atherosclerosis, although this has so far not been systematically examined.

The lack of a signal with intracerebral hemorrhage, and, in particular, deep intracerebral hemorrhage, which, like small-vessel stroke, is attributed to small-vessel disease,[17] is in line with this result. However, this analysis was based on a rather small sample size. Also, following restriction of the analysis to SNPs in the vicinity of genes interacting with MCP-1, we identified a significant association between genetically determined MCP-1 levels and intracerebral hemorrhage. This difference in results might relate to the exclusion of nonspecific instruments in the sensitivity analyses and should be explored further in larger samples.

Our meta-analysis of case-control studies revealed higher circulating MCP-1 levels in patients with ischemic stroke than in healthy controls. Our systematic search identified only 3 prospective cohort studies, one on incident[46] and 2 on recurrent stroke events,[47,48] none of which showed significant results. However, these studies had small sample sizes and a low number of events. Also, ischemic stroke subtypes were not considered, thus precluding meaningful comparisons with our MR results. It is interesting to note that observational cohort studies on CAD found higher MCP-1 levels to be associated with a higher risk of incident[49] and recurrent[50] events, consistent with the observed association with atherosclerotic stroke. Serial measurements of MCP-1 in large population-based cohorts with data on ischemic stroke subtypes would offer further insights into the relationship between MCP-1 and the risk of stroke.

Targeting specific inflammatory cytokines might reduce vascular risk. The recent multicenter CANTOS trial showed that canakinumab, a monoclonal antibody against IL-1β, decreases the rate of recurrent cardiovascular events, including nonfatal MI, nonfatal stroke, and cardiovascular mortality, among patients with MI and elevated circulating C-reactive protein levels.[8] Unfortunately, the original cytokine GWAS did not identify any genetic instruments for IL-1β circulating levels,[12] thus precluding a comparison of the MR results with the results of the CANTOS trial.[8] The MCP-1/CCR2 pathway was targeted in a small phase II clinical trial in patients with risk factors for atherosclerosis and elevated circulating C-reactive protein levels. MLN1202, a humanized monoclonal antibody against CCR2, reduced C-reactive protein levels after 4 and 12 weeks.[51] However, the effects on clinical end points were not assessed[51] and would need to be determined in a larger trial.

This study has several methodological strengths. We used the most recent and comprehensive data set for cytokine levels and the largest available GWAS data set for stroke and stroke subtypes. Results were confirmed through sensitivity analyses for pleiotropy, including alternative MR methods, in subanalyses on a biologically plausible protein-protein interaction network, and in analyses on etiologically related outcomes (CAD and MI).

Our study also has limitations. First, none of the SNPs used as instruments for MCP-1 were located in the vicinity of the *MCP1* gene, thus precluding analyses restricted to SNPs within this locus. Consequently, although we found no statistical evidence for pleiotropy, we cannot preclude nonspecific effects of the MCP-1 trans-acting instruments. Second, our instrument selection was based on a single-discovery GWAS that adjusted for body mass index. Although the association remained consistent when using an unweighted allele score, we cannot exclude that the body mass index adjustment led to collider bias during instrument selection. Third, we could not obtain reliable genetic instruments for 18 cytokines, and several analyses for ischemic stroke subtypes were underpowered. Thus, we might have missed associations for several cytokines that have previously been implicated in vascular disease such as IL-1β, tumor necrosis factor-α, and IL-

6. Targeted studies incorporating further GWAS data on individual cytokines might reveal additional associations not captured by our approach. Fourth, genetic instruments were selected using a false discovery rate–based approach, which might have weakened the instruments. However, the *F* statistics were high, and the results were in line with those derived when selecting instruments based on the genome-wide threshold ($P<5\times10^{-8}$). Finally, the UK Biobank analysis was rather underpowered and did not include stroke subtypes. Yet the consistency of both the direction and magnitude of the associations between genetically determined MCP-1 and risk of any stroke and any ischemic stroke supports our results.

In conclusion, this study demonstrates that lifelong elevated circulating MCP-1 levels are associated with higher risk of stroke and, in particular, with the large-artery and the cardioembolic subtypes. Future studies should explore in more depth whether targeting MCP-1 or its downstream effectors could be a meaningful strategy to reduce stroke risk.

ARTICLE INFORMATION

Received May 11, 2018; accepted August 17, 2018.

The online-only Data Supplement is available with this article at https://www.ahajournals.org/doi/suppl/10.1161/CIRCULATIONAHA.118.035905

Correspondence

Martin Dichgans, MD, Institute for Stroke and Dementia Research, University Hospital of Ludwig-Maximilians-University, Feodor-Lynen-Str 17, 81377 Munich, Germany. Email martin.dichgans@med.uni-muenchen.de

Affiliations

Institute for Stroke and Dementia Research, University Hospital of Ludwig-Maximilians-University, Munich, Germany (M.K.G., J.B., R.M., M.D.). Graduate School for Systemic Neurosciences, Ludwig-Maximilians-University, Munich, Germany (M.K.G.). Department of Biostatistics and Epidemiology, School of Public Health, Imperial College London, UK (D.G., C.L.M.S.). Centre for Clinical Brain Sciences, University of Edinburgh, UK (K.R.). Stroke Research Group, Department of Clinical Neurosciences, University of Cambridge, UK (M.T.). Center for Genomic Medicine (C.D.A.), Division of Neurocritical Care and Emergency Neurology, Department of Neurology (C.D.A.), Massachusetts General Hospital, Boston. Program in Medical and Population Genetics, Broad Institute, Cambridge, MA (C.D.A.). Department of Neurology, Radiology, and Biomedical Engineering, Washington University School of Medicine, St Louis, MO (J.-M.L.). Laboratory for Statistical Analysis, RIKEN Center for Integrative Medical Sciences, Yokohama, Japan (Y.K.). Clinical Trial Service Unit and Epidemiological Studies Unit, Nuffield Department of Population Health, University of Oxford, UK (J.C.H.). Departments of Neurology and Public Health Sciences, University of Virginia School of Medicine, Charlottesville (B.B.W.). Munich Cluster for Systems Neurology (SyNergy), Munich, Germany (J.B., M.D.). Institute for Genetics and Molecular Medicine, University of Edinburgh, UK (C.L.M.S.). German Centre for Neurodegenerative Diseases, Munich (M.D.).

Acknowledgments

This research has been conducted using the UK Biobank Resource (UK Biobank application 2532, "UK Biobank stroke study: developing an in-depth understanding of the determinants of stroke and its subtypes"). We thank the following consortia for making data publicly available: CARDIoGRAMplusC4D Consortium, AFGen Consortium, GLGC Consortium, and DIAGRAM Consortium.

Sources of Funding

This project has received funding from the European Union's Horizon 2020 research and innovation program under grant agreements No. 666881, "Small vessel diseases in a mechanistic perspective: Targets for Intervention" (SVDs@target; to Dr Dichgans) and No. 667375, "Common mechanisms and pathways in stroke and Alzheimer's disease" (CoSTREAM; to Dr Dichgans); the German Research Foundation as part of the "Munich Cluster for Systems Neurology" (SyNergy; EXC 1010; to Drs Dichgans and Bernhagen) and the Collaborative Research Center 1123 (B3; to Dr Dichgans and A3 to Dr Bernhagen); the Corona Foundation (to Dr Dichgans); the Foundation Leducq (Transatlantic Network of Excellence on the Pathogenesis of Small Vessel Disease of the Brain; to Dr Dichgans); the e:Med program ("Systems medicine of myocardial infarction and stroke"; e:AtheroSysMed; to Dr Dichgans); and the European Union's Seventh Framework Programme (FP7/2007–2103) project "Exploitation of Genomic Variants Affecting Coronary Artery Disease and Stroke Risk for Therapeutic Intervention" (CVgenes@target; grant agreement number Health-F2-2013–601456; to Dr Dichgans). Dr Gill is funded by the Wellcome Trust. Dr Hopewell is supported by a fellowship from the British Heart Foundation (FS/14/55/30806).

Disclosures

None.

APPENDIX

The full investigator list of the MEGASTROKE consortium of the International Stroke Genetics Consortium (ISGC) is at http://megastroke.org/authors.html..

REFERENCES

1. GBD 2015 DALYs and Hale Collaborators. Global, regional, and national disability-adjusted life-years (DALYs) for 315 diseases and injuries and healthy life expectancy (HALE), 1990–2015: a systematic analysis for the Global Burden of Disease Study 2015. *Lancet*. 2016;388:1603–1658.
2. GBD 2015 Mortality and Causes of Death Collaborators. Global, regional, and national life expectancy, all-cause mortality, and cause-specific mortality for 249 causes of death, 1980–2015: a systematic analysis for the Global Burden of Disease Study 2015. *Lancet*. 2016;388:1459–1544.
3. Feigin VL, Norrving B, Mensah GA. Global burden of stroke. *Circ Res*. 2017;120:439–448. doi: 10.1161/CIRCRESAHA.116.308413
4. Esenwa CC, Elkind MS. Inflammatory risk factors, biomarkers and associated therapy in ischaemic stroke. *Nat Rev Neurol*. 2016;12:594–604. doi: 10.1038/nrneurol.2016.125
5. Libby P, Ridker PM, Hansson GK. Progress and challenges in translating the biology of atherosclerosis. *Nature*. 2011;473:317–325. doi: 10.1038/nature10146
6. Rouhl RP, Damoiseaux JG, Lodder J, Theunissen RO, Knottnerus IL, Staals J, Henskens LH, Kroon AA, de Leeuw PW, Tervaert JW, van Oostenbrugge RJ. Vascular inflammation in cerebral small vessel disease. *Neurobiol Aging*. 2012;33:1800–1806. doi: 10.1016/j.neurobiolaging.2011.04.008
7. Ridker PM. Targeting inflammatory pathways for the treatment of cardiovascular disease. *Eur Heart J*. 2014;35:540–543. doi: 10.1093/eurheartj/eht398
8. Ridker PM, Everett BM, Thuren T, MacFadyen JG, Chang WH, Ballantyne C, Fonseca F, Nicolau J, Koenig W, Anker SD, Kastelein JJP, Cornel JH, Pais P, Pella D, Genest J, Cifkova R, Lorenzatti A, Forster T, Kobalava Z, Vida-Simiti L, Flather M, Shimokawa H, Ogawa H, Dellborg M, Rossi PRF, Troquay RPT, Libby P, Glynn RJ; CANTOS Trial Group. Antiinflammatory therapy with canakinumab for atherosclerotic disease. *N Engl J Med*. 2017;377:1119–1131. doi: 10.1056/NEJMoa1707914
9. Smith GD, Ebrahim S. 'Mendelian randomization': can genetic epidemiology contribute to understanding environmental determinants of disease? *Int J Epidemiol*. 2003;32:1–22.
10. Kelly PJ, Murphy S, Coveney S, Purroy F, Lemmens R, Tsivgoulis G, Price C. Anti-inflammatory approaches to ischaemic stroke prevention. *J Neurol Neurosurg Psychiatry*. 2018;89:211–218. doi: 10.1136/jnnp-2016-314817
11. Holmes MV, Ala-Korpela M, Smith GD. Mendelian randomization in cardiometabolic disease: challenges in evaluating causality. *Nat Rev Cardiol*. 2017;14:577–590. doi: 10.1038/nrcardio.2017.78
12. Ahola-Olli AV, Würtz P, Havulinna AS, Aalto K, Pitkänen N, Lehtimäki T, Kähönen M, Lyytikäinen LP, Raitoharju E, Seppälä I, Sarin AP, Ripatti S, Palotie A, Perola M, Viikari JS, Jalkanen S, Maksimow M, Salomaa V, Salmi

M, Kettunen J, Raitakari OT. Genome-wide association study identifies 27 loci influencing concentrations of circulating cytokines and growth factors. *Am J Hum Genet*. 2017;100:40–50. doi: 10.1016/j.ajhg.2016.11.007

13. Malik R, Chauhan G, Traylor M, Sargurupremraj M, Okada Y, Mishra A, Rutten-Jacobs L, Giese AK, van der Laan SW, Gretarsdottir S, Anderson CD, Chong M, Adams HHH, Ago T, Almgren P, Amouyel P, Ay H, Bartz TM, Benavente OR, Bevan S, Boncoraglio GB, Brown RD Jr, Butterworth AS, Carrera C, Carty CL, Chasman DI, Chen WM, Cole JW, Correa A, Cotlarciuc I, Cruchaga C, Danesh J, de Bakker PIW, DeStefano AL, den Hoed M, Duan Q, Engelter ST, Falcone GJ, Gottesman RF, Grewal RP, Gudnason V, Gustafsson S, Haessler J, Harris TB, Hassan A, Havulinna AS, Heckbert SR, Holliday EG, Howard G, Hsu FC, Hyacinth HI, Ikram MA, Ingelsson E, Irvin MR, Jian X, Jiménez-Conde J, Johnson JA, Jukema JW, Kanai M, Keene KL, Kissela BM, Kleindorfer DO, Kooperberg C, Kubo M, Lange LA, Langefeld CD, Langenberg C, Launer LJ, Lee JM, Lemmens R, Leys D, Lewis CM, Lin WY, Lindgren AG, Lorentzen E, Magnusson PK, Maguire J, Manichaikul A, McArdle PF, Meschia JF, Mitchell BD, Mosley TH, Nalls MA, Ninomiya T, O'Donnell MJ, Psaty BM, Pulit SL, Rannikmäe K, Reiner AP, Rexrode KM, Rice K, Rich SS, Ridker PM, Rost NS, Rothwell PM, Rotter JI, Rundek T, Sacco RL, Sakaue S, Sale MM, Salomaa V, Sapkota BR, Schmidt R, Schmidt CO, Schminke U, Sharma P, Slowik A, Sudlow CLM, Tanislav C, Tatlisumak T, Taylor KD, Thijs VNS, Thorleifsson G, Thorsteinsdottir U, Tiedt S, Trompet S, Tzourio C, van Duijn CM, Walters M, Wareham NJ, Wassertheil-Smoller S, Wilson JG, Wiggins KL, Yang Q, Yusuf S, Bis JC, Pastinen T, Ruusalepp A, Schadt EE, Koplev S, Björkegren JLM, Codoni V, Civelek M, Smith NL, Trégouët DA, Christophersen IE, Roselli C, Lubitz SA, Ellinor PT, Tai ES, Kooner JS, Kato N, He J, van der Harst P, Elliott P, Chambers JC, Takeuchi F, Johnson AD, Sanghera DK, Melander O, Jern C, Strbian D, Fernandez-Cadenas I, Longstreth WT Jr, Rolfs A, Hata J, Woo D, Rosand J, Pare G, Hopewell JC, Saleheen D, Stefansson K, Worrall BB, Kittner SJ, Seshadri S, Fornage M, Markus HS, Howson JMM, Kamatani Y, Debette S, Dichgans M, Malik R, Chauhan G, Traylor M, Sargurupremraj M, Okada Y, Mishra A, Rutten-Jacobs L, Giese AK, van der Laan SW, Gretarsdottir S, Anderson CD, Chong M, Adams HHH, Ago T, Almgren P, Amouyel P, Ay H, Bartz TM, Benavente OR, Bevan S, Boncoraglio GB, Brown RD Jr, Butterworth AS, Carrera C, Carty CL, Chasman DI, Chen WM, Cole JW, Correa A, Cotlarciuc I, Cruchaga C, Danesh J, de Bakker PIW, DeStefano AL, Hoed MD, Duan Q, Engelter ST, Falcone GJ, Gottesman RF, Grewal RP, Gudnason V, Gustafsson S, Haessler J, Harris TB, Hassan A, Havulinna AS, Heckbert SR, Holliday EG, Howard G, Hsu FC, Hyacinth HI, Ikram MA, Ingelsson E, Irvin MR, Jian X, Jiménez-Conde J, Johnson JA, Jukema JW, Kanai M, Keene KL, Kissela BM, Kleindorfer DO, Kooperberg C, Kubo M, Lange LA, Langefeld CD, Langenberg C, Launer LJ, Lee JM, Lemmens R, Leys D, Lewis CM, Lin WY, Lindgren AG, Lorentzen E, Magnusson PK, Maguire J, Manichaikul A, McArdle PF, Meschia JF, Mitchell BD, Mosley TH, Nalls MA, Ninomiya T, O'Donnell MJ, Psaty BM, Pulit SL, Rannikmäe K, Reiner AP, Rexrode KM, Rice K, Rich SS, Ridker PM, Rost NS, Rothwell PM, Rotter JI, Rundek T, Sacco RL, Sakaue S, Sale MM, Salomaa V, Sapkota BR, Schmidt R, Schmidt CO, Schminke U, Sharma P, Slowik A, Sudlow CLM, Tanislav C, Tatlisumak T, Taylor KD, Thijs VNS, Thorleifsson G, Thorsteinsdottir U, Tiedt S, Trompet S, Tzourio C, van Duijn CM, Walters M, Wareham NJ, Wassertheil-Smoller S, Wilson JG, Wiggins KL, Yang Q, Yusuf S, Amin N, Aparicio HS, Arnett DK, Attia J, Beiser AS, Berr C, Buring JE, Bustamante M, Caso V, Cheng YC, Choi SH, Chowhan A, Cullell N, Dartigues JF, Delavaran H, Delgado P, Dörr M, Engström G, Ford I, Gurpreet WS, Hamsten A, Heitsch L, Hozawa A, Ibanez L, Ilinca A, Ingelsson M, Iwasaki M, Jackson RD, Jood K, Jousilahti P, Kaffashian S, Kalra L, Kamouchi M, Kitazono T, Kjartansson O, Kloss M, Koudstaal PJ, Krupinski J, Labovitz DL, Laurie CC, Levi CR, Li L, Lind L, Lindgren CM, Lioutas V, Liu YM, Lopez OL, Makoto H, Martinez-Majander N, Matsuda K, Minegishi N, Montaner J, Morris AP, Muiño E, Müller-Nurasyid M, Norrving B, Ogishima S, Parati EA, Peddareddygari LR, Pedersen NL, Pera J, Perola M, Pezzini A, Pileggi S, Rabionet R, Riba-Llena I, Ribasés M, Romero JR, Roquer J, Rudd AG, Sarin AP, Sarju R, Sarnowski C, Sasaki M, Satizabal CL, Satoh M, Sattar N, Sawada N, Sibolt G, Sigurdsson Á, Smith A, Sobue K, Soriano-Tárraga C, Stanne T, Stine OC, Stott DJ, Strauch K, Takai T, Tanaka H, Tanno K, Teumer A, Tomppo L, Torres-Aguila NP, Touze E, Tsugane S, Uitterlinden AG, Valdimarsson EM, van der Lee SJ, Völzke H, Wakai K, Weir D, Williams SR, Wolfe CDA, Wong Q, Xu H, Yamaji T, Sanghera DK, Melander O, Jern C, Strbian D, Fernandez-Cadenas I, Longstreth WT Jr, Rolfs A, Hata J, Woo D, Rosand J, Pare G, Hopewell JC, Saleheen D, Stefansson K, Worrall BB, Kittner SJ, Seshadri S, Fornage M, Markus HS, Howson JMM, Kamatani Y, Debette S, Dichgans M; AFGen Consortium; Cohorts for Heart and Aging Research in Genomic Epidemiology (CHARGE) Consortium; International Genomics of Blood Pressure (iGEN-BP) Consortium; INVENT Consortium; STARNET; BioBank Japan Cooperative Hospital Group; COMPASS Consortium; EPIC-CVD Consortium; EPIC-InterAct Consortium; International Stroke Genetics Consortium (ISGC); METASTROKE Consortium; Neurology Working Group of the CHARGE Consortium; NINDS Stroke Genetics Network (SiGN); UK Young Lacunar DNA Study; MEGASTROKE Consortium; MEGASTROKE Consortium:. Multiancestry genome-wide association study of 520,000 subjects identifies 32 loci associated with stroke and stroke subtypes. *Nat Genet*. 2018;50:524–537. doi: 10.1038/s41588-018-0058-3
14. Nelson CP, Goel A, Butterworth AS, Kanoni S, Webb TR, Marouli E, Zeng L, Ntalla I, Lai FY, Hopewell JC, Giannakopoulou O, Jiang T, Hamby SE, Di Angelantonio E, Assimes TL, Bottinger EP, Chambers JC, Clarke R, Palmer CNA, Cubbon RM, Ellinor P, Ermel R, Evangelou E, Franks PW, Grace C, Gu D, Hingorani AD, Howson JMM, Ingelsson E, Kastrati A, Kessler T, Kyriakou T, Lehtimäki T, Lu X, Lu Y, März W, McPherson R, Metspalu A, Pujades-Rodriguez M, Ruusalepp A, Schadt EE, Schmidt AF, Sweeting MJ, Zalloua PA, AlGhalayini K, Keavney BD, Kooner JS, Loos RJF, Patel RS, Rutter MK, Tomaszewski M, Tzoulaki I, Zeggini E, Erdmann J, Dedoussis G, Björkegren JLM, Schunkert H, Farrall M, Danesh J, Samani NJ, Watkins H, Deloukas P; EPIC-CVD Consortium; CARDIoGRAMplusC4D; UK Biobank CardioMetabolic Consortium CHD working group. Association analyses based on false discovery rate implicate new loci for coronary artery disease. *Nat Genet*. 2017;49:1385–1391. doi: 10.1038/ng.3913
15. Holmes MV, Asselbergs FW, Palmer TM, Drenos F, Lanktree MB, Nelson CP, Dale CE, Padmanabhan S, Finan C, Swerdlow DI, Tragante V, van Iperen EP, Sivapalaratnam S, Shah S, Elbers CC, Shah T, Engmann J, Giambartolomei C, White J, Zabaneh D, Sofat R, McLachlan S, Doevendans PA, Balmforth AJ, Hall AS, North KE, Almoguera B, Hoogeveen RC, Cushman M, Fornage M, Patel SR, Redline S, Siscovick DS, Tsai MY, Karczewski KJ, Hofker MH, Verschuren WM, Bots ML, van der Schouw YT, Melander O, Dominiczak AF, Morris R, Ben-Shlomo Y, Price J, Kumari M, Baumert J, Peters A, Thorand B, Koenig W, Gaunt TR, Humphries SE, Clarke R, Watkins H, Farrall M, Wilson JG, Rich SS, de Bakker PI, Lange LA, Davey Smith G, Reiner AP, Talmud PJ, Kivimäki M, Lawlor DA, Dudbridge F, Samani NJ, Keating BJ, Hingorani AD, Casas JP; UCLEB consortium. Mendelian randomization of blood lipids for coronary heart disease. *Eur Heart J*. 2015;36:539–550. doi: 10.1093/eurheartj/eht571
16. Adams HP Jr, Bendixen BH, Kappelle LJ, Biller J, Love BB, Gordon DL, Marsh EE 3rd. Classification of subtype of acute ischemic stroke. Definitions for use in a multicenter clinical trial. TOAST. Trial of Org 10172 in Acute Stroke Treatment. *Stroke*. 1993;24:35–41.
17. Woo D, Falcone GJ, Devan WJ, Brown WM, Biffi A, Howard TD, Anderson CD, Brouwers HB, Valant V, Battey TW, Radmanesh F, Raffeld MR, Baedorf-Kassis S, Deka R, Woo JG, Martin LJ, Haverbusch M, Moomaw CJ, Sun G, Broderick JP, Flaherty ML, Martini SR, Kleindorfer DO, Kissela B, Comeau ME, Jagiella JM, Schmidt H, Freudenberger P, Pichler A, Enzinger C, Hansen BM, Norrving B, Jimenez-Conde J, Giralt-Steinhauer E, Elosua R, Cuadrado-Godia E, Soriano C, Roquer J, Kraft P, Ayres AM, Schwab K, McCauley JL, Pera J, Urbanik A, Rost NS, Goldstein JN, Viswanathan A, Stögerer EM, Tirschwell DL, Selim M, Brown DL, Silliman SL, Worrall BB, Meschia JF, Kidwell CS, Montaner J, Fernandez-Cadenas I, Delgado P, Malik R, Dichgans M, Greenberg SM, Rothwell PM, Lindgren A, Slowik A, Schmidt R, Langefeld CD, Rosand J; International Stroke Genetics Consortium. Meta-analysis of genome-wide association studies identifies 1q22 as a susceptibility locus for intracerebral hemorrhage. *Am J Hum Genet*. 2014;94:511–521. doi: 10.1016/j.ajhg.2014.02.012
18. Pierce BL, Ahsan H, Vanderweele TJ. Power and instrument strength requirements for Mendelian randomization studies using multiple genetic variants. *Int J Epidemiol*. 2011;40:740–752. doi: 10.1093/ije/dyq151
19. Burgess S. Sample size and power calculations in Mendelian randomization with a single instrumental variable and a binary outcome. *Int J Epidemiol*. 2014;43:922–929. doi: 10.1093/ije/dyu005
20. Palmer TM, Lawlor DA, Harbord RM, Sheehan NA, Tobias JH, Timpson NJ, Davey Smith G, Sterne JA. Using multiple genetic variants as instrumental variables for modifiable risk factors. *Stat Methods Med Res*. 2012;21:223–242. doi: 10.1177/0962280210394459
21. Nikpay M, Goel A, Won HH, Hall LM, Willenborg C, Kanoni S, Saleheen D, Kyriakou T, Nelson CP, Hopewell JC, Webb TR, Zeng L, Dehghan A, Alver M, Armasu SM, Auro K, Bjonnes A, Chasman DI, Chen S, Ford I, Franceschini N, Gieger C, Grace C, Gustafsson S, Huang J, Hwang SJ, Kim YK, Kleber ME, Lau KW, Lu X, Lu Y, Lyytikäinen LP, Mihailov E, Morrison AC, Pervjakova N, Qu L, Rose LM, Salfati E, Saxena R, Scholz M, Smith AV, Tikkanen E, Uitterlinden A, Yang X, Zhang W, Zhao W, de Andrade M, de Vries PS, van Zuydam NR, Anand SS, Bertram L, Beutner F, Dedous-

sis G, Frossard P, Gauguier D, Goodall AH, Gottesman O, Haber M, Han BG, Huang J, Jalilzadeh S, Kessler T, König IR, Lannfelt L, Lieb W, Lind L, Lindgren CM, Lokki ML, Magnusson PK, Mallick NH, Mehra N, Meitinger T, Memon FU, Morris AP, Nieminen MS, Pedersen NL, Peters A, Rallidis LS, Rasheed A, Samuel M, Shah SH, Sinisalo J, Stirrups KE, Trompet S, Wang L, Zaman KS, Ardissino D, Boerwinkle E, Borecki IB, Bottinger EP, Buring JE, Chambers JC, Collins R, Cupples LA, Danesh J, Demuth I, Elosua R, Epstein SE, Esko T, Feitosa MF, Franco OH, Franzosi MG, Granger CB, Gu D, Gudnason V, Hall AS, Hamsten A, Harris TB, Hazen SL, Hengstenberg C, Hofman A, Ingelsson E, Iribarren C, Jukema JW, Karhunen PJ, Kim BJ, Kooner JS, Kullo IJ, Lehtimäki T, Loos RJF, Melander O, Metspalu A, März W, Palmer CN, Perola M, Quertermous T, Rader DJ, Ridker PM, Ripatti S, Roberts R, Salomaa V, Sanghera DK, Schwartz SM, Seedorf U, Stewart AF, Stott DJ, Thiery J, Zalloua PA, O'Donnell CJ, Reilly MP, Assimes TL, Thompson JR, Erdmann J, Clarke R, Watkins H, Kathiresan S, McPherson R, Deloukas P, Schunkert H, Samani NJ, Farrall M. A comprehensive 1,000 Genomes-based genome-wide association meta-analysis of coronary artery disease. *Nat Genet*. 2015;47:1121–1130. doi: 10.1038/ng.3396

22. Christophersen IE, Rienstra M, Roselli C, Yin X, Geelhoed B, Barnard J, Lin H, Arking DE, Smith AV, Albert CM, Chaffin M, Tucker NR, Li M, Klarin D, Bihlmeyer NA, Low SK, Weeke PE, Müller-Nurasyid M, Smith JG, Brody JA, Niemeijer MN, Dörr M, Trompet S, Huffman J, Gustafsson S, Schurmann C, Kleber ME, Lyytikäinen LP, Seppälä I, Malik R, Horimoto ARVR, Perez M, Sinisalo J, Aeschbacher S, Thériault S, Yao J, Radmanesh F, Weiss S, Teumer A, Choi SH, Weng LC, Clauss S, Deo R, Rader DJ, Shah SH, Sun A, Hopewell JC, Debette S, Chauhan G, Yang Q, Worrall BB, Paré G, Kamatani Y, Hagemeijer YP, Verweij N, Siland JE, Kubo M, Smith JD, Van Wagoner DR, Bis JC, Perz S, Psaty BM, Ridker PM, Magnani JW, Harris TB, Launer LJ, Shoemaker MB, Padmanabhan S, Haessler J, Bartz TM, Waldenberger M, Lichtner P, Arendt M, Krieger JE, Kähönen M, Risch L, Mansur AJ, Peters A, Smith BH, Lind L, Scott SA, Lu Y, Bottinger EB, Hernesniemi J, Lindgren CM, Wong JA, Huang J, Eskola M, Morris AP, Ford I, Reiner AP, Delgado G, Chen LY, Chen YI, Sandhu RK, Li M, Boerwinkle E, Eisele L, Lannfelt L, Rost N, Anderson CD, Taylor KD, Campbell A, Magnusson PK, Porteous D, Hocking LJ, Vlachopoulou E, Pedersen NL, Nikus K, Orho-Melander M, Hamsten A, Heeringa J, Denny JC, Kriebel J, Darbar D, Newton-Cheh C, Shaffer C, Macfarlane PW, Heilmann-Heimbach S, Almgren P, Huang PL, Sotoodehnia N, Soliman EZ, Uitterlinden AG, Hofman A, Franco OH, Völker U, Jöckel KH, Sinner MF, Lin HJ, Guo X, Dichgans M, Ingelsson E, Kooperberg C, Melander O, Loos RJF, Laurikka J, Conen D, Rosand J, van der Harst P, Lokki ML, Kathiresan S, Pereira A, Jukema JW, Hayward C, Rotter JI, März W, Lehtimäki T, Stricker BH, Chung MK, Felix SB, Gudnason V, Alonso A, Roden DM, Kääb S, Chasman DI, Heckbert SR, Benjamin EJ, Tanaka T, Lunetta KL, Lubitz SA, Ellinor PT; METASTROKE Consortium of the ISGC; Neurology Working Group of the CHARGE Consortium; AFGen Consortium. Large-scale analyses of common and rare variants identify 12 new loci associated with atrial fibrillation. *Nat Genet*. 2017;49:946–952. doi: 10.1038/ng.3843
23. Burgess S, Butterworth A, Thompson SG. Mendelian randomization analysis with multiple genetic variants using summarized data. *Genet Epidemiol*. 2013;37:658–665. doi: 10.1002/gepi.21758
24. Greco M FD, Minelli C, Sheehan NA, Thompson JR. Detecting pleiotropy in Mendelian randomisation studies with summary data and a continuous outcome. *Stat Med*. 2015;34:2926–2940. doi: 10.1002/sim.6522
25. Bowden J, Davey Smith G, Burgess S. Mendelian randomization with invalid instruments: effect estimation and bias detection through Egger regression. *Int J Epidemiol*. 2015;44:512–525. doi: 10.1093/ije/dyv080
26. Bowden J, Davey Smith G, Haycock PC, Burgess S. Consistent estimation in mendelian randomization with some invalid instruments using a weighted median estimator. *Genet Epidemiol*. 2016;40:304–314. doi: 10.1002/gepi.21965
27. Verbanck M, Chen CY, Neale B, Do R. Detection of widespread horizontal pleiotropy in causal relationships inferred from Mendelian randomization between complex traits and diseases. *Nat Genet*. 2018;50:693–698. doi: 10.1038/s41588-018-0099-7
28. Burgess S, Thompson SG. Multivariable Mendelian randomization: the use of pleiotropic genetic variants to estimate causal effects. *Am J Epidemiol*. 2015;181:251–260. doi: 10.1093/aje/kwu283
29. Willer CJ, Schmidt EM, Sengupta S, Peloso GM, Gustafsson S, Kanoni S, Ganna A, Chen J, Buchkovich ML, Mora S, Beckmann JS, Bragg-Gresham JL, Chang HY, Demirkan A, Den Hertog HM, Do R, Donnelly LA, Ehret GB, Esko T, Feitosa MF, Ferreira T, Fischer K, Fontanillas P, Fraser RM, Freitag DF, Gurdasani D, Heikkilä K, Hyppönen E, Isaacs A, Jackson AU, Johansson Å, Johnson T, Kaakinen M, Kettunen J, Kleber ME, Li X, Luan J, Lyytikäinen LP, Magnusson PKE, Mangino M, Mihailov E, Montasser ME, Müller-Nurasyid M, Nolte IM, O'Connell JR, Palmer CD, Perola M, Petersen AK, Sanna S, Saxena R, Service SK, Shah S, Shungin D, Sidore C, Song C, Strawbridge RJ, Surakka I, Tanaka T, Teslovich TM, Thorleifsson G, Van den Herik EG, Voight BF, Volcik KA, Waite LL, Wong A, Wu Y, Zhang W, Absher D, Asiki G, Barroso I, Been LF, Bolton JL, Bonnycastle LL, Brambilla P, Burnett MS, Cesana G, Dimitriou M, Doney ASF, Döring A, Elliott P, Epstein SE, Ingi Eyjolfsson G, Gigante B, Goodarzi MO, Grallert H, Gravito ML, Groves CJ, Hallmans G, Hartikainen AL, Hayward C, Hernandez D, Hicks AA, Holm H, Hung YJ, Illig T, Jones MR, Kaleebu P, Kastelein JJP, Khaw KT, Kim E, Klopp N, Komulainen P, Kumari M, Langenberg C, Lehtimäki T, Lin SY, Lindström J, Loos RJF, Mach F, McArdle WL, Meisinger C, Mitchell BD, Müller G, Nagaraja R, Narisu N, Nieminen TVM, Nsubuga RN, Olafsson I, Ong KK, Palotie A, Papamarkou T, Pomilla C, Pouta A, Rader DJ, Reilly MP, Ridker PM, Rivadeneira F, Rudan I, Ruokonen A, Samani N, Scharnagl H, Seeley J, Silander K, Stančáková A, Stirrups K, Swift AJ, Tiret L, Uitterlinden AG, van Pelt LJ, Vedantam S, Wainwright N, Wijmenga C, Wild SH, Willemsen G, Wilsgaard T, Wilson JF, Young EH, Zhao JH, Adair LS, Arveiler D, Assimes TL, Bandinelli S, Bennett F, Bochud M, Boehm BO, Boomsma DI, Borecki IB, Bornstein SR, Bovet P, Burnier M, Campbell H, Chakravarti A, Chambers JC, Chen YI, Collins FS, Cooper RS, Danesh J, Dedoussis G, de Faire U, Feranil AB, Ferrières J, Ferrucci L, Freimer NB, Gieger C, Groop LC, Gudnason V, Gyllensten U, Hamsten A, Harris TB, Hingorani A, Hirschhorn JN, Hofman A, Hovingh GK, Hsiung CA, Humphries SE, Hunt SC, Hveem K, Iribarren C, Järvelin MR, Jula A, Kähönen M, Kaprio J, Kesäniemi A, Kivimaki M, Kooner JS, Koudstaal PJ, Krauss RM, Kuh D, Kuusisto J, Kyvik KO, Laakso M, Lakka TA, Lind L, Lindgren CM, Martin NG, März W, McCarthy MI, McKenzie CA, Meneton P, Metspalu A, Moilanen L, Morris AD, Munroe PB, Njølstad I, Pedersen NL, Power C, Pramstaller PP, Price JF, Psaty BM, Quertermous T, Rauramaa R, Saleheen D, Salomaa V, Sanghera DK, Saramies J, Schwarz PEH, Sheu WH, Shuldiner AR, Siegbahn A, Spector TD, Stefansson K, Strachan DP, Tayo BO, Tremoli E, Tuomilehto J, Uusitupa M, van Duijn CM, Vollenweider P, Wallentin L, Wareham NJ, Whitfield JB, Wolffenbuttel BHR, Ordovas JM, Boerwinkle E, Palmer CNA, Thorsteinsdottir U, Chasman DI, Rotter JI, Franks PW, Ripatti S, Cupples LA, Sandhu MS, Rich SS, Boehnke M, Deloukas P, Kathiresan S, Mohlke KL, Ingelsson E, Abecasis GR; Global Lipids Genetics Consortium. Discovery and refinement of loci associated with lipid levels. *Nat Genet*. 2013;45:1274–1283. doi: 10.1038/ng.2797
30. Morris AP, Voight BF, Teslovich TM, Ferreira T, Segrè AV, Steinthorsdottir V, Strawbridge RJ, Khan H, Grallert H, Mahajan A, Prokopenko I, Kang HM, Dina C, Esko T, Fraser RM, Kanoni S, Kumar A, Lagou V, Langenberg C, Luan J, Lindgren CM, Müller-Nurasyid M, Pechlivanis S, Rayner NW, Scott LJ, Wiltshire S, Yengo L, Kinnunen L, Rossin EJ, Raychaudhuri S, Johnson AD, Dimas AS, Loos RJ, Vedantam S, Chen H, Florez JC, Fox C, Liu CT, Rybin D, Couper DJ, Kao WH, Li M, Cornelis MC, Kraft P, Sun Q, van Dam RM, Stringham HM, Chines PS, Fischer K, Fontanillas P, Holmen OL, Hunt SE, Jackson AU, Kong A, Lawrence R, Meyer J, Perry JR, Platou CG, Potter S, Rehnberg E, Robertson N, Sivapalaratnam S, Stančáková A, Stirrups K, Thorleifsson G, Tikkanen E, Wood AR, Almgren P, Atalay M, Benediktsson R, Bonnycastle LL, Burtt N, Carey J, Charpentier G, Crenshaw AT, Doney AS, Dorkhan M, Edkins S, Emilsson V, Eury E, Forsen T, Gertow K, Gigante B, Grant GB, Groves CJ, Guiducci C, Herder C, Hreidarsson AB, Hui J, James A, Jonsson A, Rathmann W, Klopp N, Kravic J, Krjutškov K, Langford C, Leander K, Lindholm E, Lobbens S, Männistö S, Mirza G, Mühleisen TW, Musk B, Parkin M, Rallidis L, Saramies J, Sennblad B, Shah S, Sigurðsson G, Silveira A, Steinbach G, Thorand B, Trakalo J, Veglia F, Wennauer R, Winckler W, Zabaneh D, Campbell H, van Duijn C, Uitterlinden AG, Hofman A, Sijbrands E, Abecasis GR, Owen KR, Zeggini E, Trip MD, Forouhi NG, Syvänen AC, Eriksson JG, Peltonen L, Nöthen MM, Balkau B, Palmer CN, Lyssenko V, Tuomi T, Isomaa B, Hunter DJ, Qi L, Shuldiner AR, Roden M, Barroso I, Wilsgaard T, Beilby J, Hovingh K, Price JF, Wilson JF, Rauramaa R, Lakka TA, Lind L, Dedoussis G, Njølstad I, Pedersen NL, Khaw KT, Wareham NJ, Keinanen-Kiukaanniemi SM, Saaristo TE, Korpi-Hyövälti E, Saltevo J, Laakso M, Kuusisto J, Metspalu A, Collins FS, Mohlke KL, Bergman RN, Tuomilehto J, Boehm BO, Gieger C, Hveem K, Cauchi S, Froguel P, Baldassarre D, Tremoli E, Humphries SE, Saleheen D, Danesh J, Ingelsson E, Ripatti S, Salomaa V, Erbel R, Jöckel KH, Moebus S, Peters A, Illig T, de Faire U, Hamsten A, Morris AD, Donnelly PJ, Frayling TM, Hattersley AT, Boerwinkle E, Melander O, Kathiresan S, Nilsson PM, Deloukas P, Thorsteinsdottir U, Groop LC, Stefansson K, Hu F, Pankow JS, Dupuis J, Meigs JB, Altshuler D, Boehnke M, McCarthy MI; Wellcome Trust Case Control Consortium; Meta-Analyses of Glucose and Insulin-related traits Consortium (MAGIC) Investigators; Genetic Investigation of ANthropometric Traits (GIANT) Consortium; Asian Genetic Epidemiology

Network–Type 2 Diabetes (AGEN-T2D) Consortium; South Asian Type 2 Diabetes (SAT2D) Consortium; DIAbetes Genetics Replication And Meta-analysis (DIAGRAM) Consortium. Large-scale association analysis provides insights into the genetic architecture and pathophysiology of type 2 diabetes. *Nat Genet*. 2012;44:981–990. doi: 10.1038/ng.2383
31. biobank[uk]. http://www.nealelab.is/uk-biobank. Accessed December 14, 2018.
32. Szklarczyk D, Morris JH, Cook H, Kuhn M, Wyder S, Simonovic M, Santos A, Doncheva NT, Roth A, Bork P, Jensen LJ, von Mering C. The STRING database in 2017: quality-controlled protein-protein association networks, made broadly accessible. *Nucleic Acids Res*. 2017;45(D1):D362–D368. doi: 10.1093/nar/gkw937
33. Ioannidis JP, Ntzani EE, Trikalinos TA, Contopoulos-Ioannidis DG. Replication validity of genetic association studies. *Nat Genet*. 2001;29:306–309. doi: 10.1038/ng749
34. Charoen P, Nitsch D, Engmann J, Shah T, White J, Zabaneh D, Jefferis B, Wannamethee G, Whincup P, Mulick Cassidy A, Gaunt T, Day I, McLachlan S, Price J, Kumari M, Kivimaki M, Brunner E, Langenberg C, Ben-Shlomo Y, Hingorani A, Whittaker J, Pablo Casas J, Dudbridge F; UCLEB Consortium. Mendelian randomisation study of the influence of eGFR on coronary heart disease. *Sci Rep*. 2016;6:28514. doi: 10.1038/srep28514
35. Cohen J. *Statistical Power Analysis for the Behavioral Sciences*. 2nd ed. Hillsdale, NJ: Erlbaum; 1988.
36. Lin J, Kakkar V, Lu X. Impact of MCP-1 in atherosclerosis. *Curr Pharm Des*. 2014;20:4580–4588.
37. Gu L, Okada Y, Clinton SK, Gerard C, Sukhova GK, Libby P, Rollins BJ. Absence of monocyte chemoattractant protein-1 reduces atherosclerosis in low density lipoprotein receptor-deficient mice. *Mol Cell*. 1998;2:275–281.
38. Boring L, Gosling J, Cleary M, Charo IF. Decreased lesion formation in CCR2-/- mice reveals a role for chemokines in the initiation of atherosclerosis. *Nature*. 1998;394:894–897. doi: 10.1038/29788
39. Inoue S, Egashira K, Ni W, Kitamoto S, Usui M, Otani K, Ishibashi M, Hiasa K, Nishida K, Takeshita A. Anti-monocyte chemoattractant protein-1 gene therapy limits progression and destabilization of established atherosclerosis in apolipoprotein E-knockout mice. *Circulation*. 2002;106:2700–2706.
40. Liehn EA, Piccinini AM, Koenen RR, Soehnlein O, Adage T, Fatu R, Curaj A, Popescu A, Zernecke A, Kungl AJ, Weber C. A new monocyte chemotactic protein-1/chemokine CC motif ligand-2 competitor limiting neointima formation and myocardial ischemia/reperfusion injury in mice. *J Am Coll Cardiol*. 2010;56:1847–1857. doi: 10.1016/j.jacc.2010.04.066
41. Bot I, Ortiz Zacarías NV, de Witte WE, de Vries H, van Santbrink PJ, van der Velden D, Kröner MJ, van der Berg DJ, Stamos D, de Lange EC, Kuiper J, IJzerman AP, Heitman LH. A novel CCR2 antagonist inhibits atherogenesis in apoE deficient mice by achieving high receptor occupancy. *Sci Rep*. 2017;7:52. doi: 10.1038/s41598-017-00104-z
42. Aiello RJ, Bourassa PA, Lindsey S, Weng W, Natoli E, Rollins BJ, Milos PM. Monocyte chemoattractant protein-1 accelerates atherosclerosis in apolipoprotein E-deficient mice. *Arterioscler Thromb Vasc Biol*. 1999;19:1518–1525.
43. Biernacka A, Frangogiannis NG. Aging and cardiac fibrosis. *Aging Dis*. 2011;2:158–173.
44. Dzeshka MS, Lip GY, Snezhitskiy V, Shantsila E. Cardiac fibrosis in patients with atrial fibrillation: mechanisms and clinical implications. *J Am Coll Cardiol*. 2015;66:943–959. doi: 10.1016/j.jacc.2015.06.1313
45. Hammwöhner M, Ittenson A, Dierkes J, Bukowska A, Klein HU, Lendeckel U, Goette A. Platelet expression of CD40/CD40 ligand and its relation to inflammatory markers and adhesion molecules in patients with atrial fibrillation. *Exp Biol Med (Maywood)*. 2007;232:581–589.
46. Canouï-Poitrine F, Luc G, Mallat Z, Machez E, Bingham A, Ferrieres J, Ruidavets JB, Montaye M, Yarnell J, Haas B, Arveiler D, Morange P, Kee F, Evans A, Amouyel P, Ducimetiere P, Empana JP; PRIME Study Group. Systemic chemokine levels, coronary heart disease, and ischemic stroke events: the PRIME study. *Neurology*. 2011;77:1165–1173. doi: 10.1212/WNL.0b013e31822dc7c8
47. Ganz P, Amarenco P, Goldstein LB, Sillesen H, Bao W, Preston GM, Welch KMA; SPARCL Steering Committee. Association of osteopontin, neopterin, and myeloperoxidase with stroke risk in patients with prior stroke or transient ischemic attacks: results of an analysis of 13 biomarkers from the Stroke Prevention by Aggressive Reduction in Cholesterol Levels Trial. *Stroke*. 2017;48:3223–3231. doi: 10.1161/STROKEAHA.117.017965
48. Haim M, Tanne D, Boyko V, Reshef T, Goldbourt U, Battler A, Mekori YA, Behar S. Monocyte chemoattractant protein-1 and recurrent cardiovascular events in patients with stable coronary heart disease. *Clin Cardiol*. 2005;28:31–35.
49. Hoogeveen RC, Morrison A, Boerwinkle E, Miles JS, Rhodes CE, Sharrett AR, Ballantyne CM. Plasma MCP-1 level and risk for peripheral arterial disease and incident coronary heart disease: Atherosclerosis Risk in Communities study. *Atherosclerosis*. 2005;183:301–307. doi: 10.1016/j.atherosclerosis.2005.03.007
50. de Lemos JA, Morrow DA, Sabatine MS, Murphy SA, Gibson CM, Antman EM, McCabe CH, Cannon CP, Braunwald E. Association between plasma levels of monocyte chemoattractant protein-1 and long-term clinical outcomes in patients with acute coronary syndromes. *Circulation*. 2003;107:690–695.
51. Gilbert J, Lekstrom-Himes J, Donaldson D, Lee Y, Hu M, Xu J, Wyant T, Davidson M; MLN1202 Study Group. Effect of CC chemokine receptor 2 CCR2 blockade on serum C-reactive protein in individuals at atherosclerotic risk and with a single nucleotide polymorphism of the monocyte chemoattractant protein-1 promoter region. *Am J Cardiol*. 2011;107:906–911. doi: 10.1016/j.amjcard.2010.11.005

MANUSCRIPT II: Circulating Monocyte Chemoattractant Protein-1 and Risk of Stroke: Meta-Analysis of Population-Based Studies Involving 17 180 Individuals

Georgakis MK, Malik R, Björkbacka H, Pana TA, Demissie S, Ayers C, Elhadad MA, Fornage M, Beiser AS, Benjamin EJ, Boekholdt SM, Engström G, Herder C, Hoogeveen RC, Koenig W, Melander O, Orho-Melander M, Schiopu A, Söderholm M, Wareham N, Ballantyne CM, Peters A, Seshadri S, Myint PK, Nilsson J, de Lemos JA, Dichgans M. Circulating Monocyte Chemoattractant Protein-1 and Risk of Stroke: Meta-Analysis of Population-Based Studies Involving 17 180 Individuals. ***Circ Res***. 2019 Sep 27;125(8):773-782.

Author contributions: MKG and MD conceptualized and designed the study. MKG performed the systematic review and the meta-analyses of the pooled data that were received from the individual studies. HB, TAP, SD, CA, MAE, ASB, EJB, SMB, GE, CH, RCH, WK, OM, MOM, AS, MS, NW, CMB, AP, SS, PKM, JN, and JAdL performed the statistical analyses of the individual studies and provided the summary data. All authors interpreted the results. MKG and MD drafted the manuscript. All authors critically revised the manuscript for intellectual content. All authors approved the submitted version and are accountable for the integrity of the work.

Circulation Research

ORIGINAL RESEARCH

Circulating Monocyte Chemoattractant Protein-1 and Risk of Stroke

Meta-Analysis of Population-Based Studies Involving 17 180 Individuals

Marios K. Georgakis, Rainer Malik, Harry Björkbacka, Tiberiu Alexandru Pana, Serkalem Demissie, Colby Ayers, Mohamed A. Elhadad, Myriam Fornage, Alexa S. Beiser, Emelia J. Benjamin, Matthijs S. Boekholdt, Gunnar Engström, Christian Herder, Ron C. Hoogeveen, Wolfgang Koenig, Olle Melander, Marju Orho-Melander, Alexandru Schiopu, Martin Söderholm, Nick Wareham, Christie M. Ballantyne, Annette Peters, Sudha Seshadri, Phyo K. Myint, Jan Nilsson, James A. de Lemos, Martin Dichgans

RATIONALE: Proinflammatory cytokines have been identified as potential targets for lowering vascular risk. Experimental evidence and Mendelian randomization suggest a role of MCP-1 (monocyte chemoattractant protein-1) in atherosclerosis and stroke. However, data from large-scale observational studies are lacking.

OBJECTIVE: To determine whether circulating levels of MCP-1 are associated with risk of incident stroke in the general population.

METHODS AND RESULTS: We used previously unpublished data on 17 180 stroke-free individuals (mean age, 56.7±8.1 years; 48.8% men) from 6 population-based prospective cohort studies and explored associations between baseline circulating MCP-1 levels and risk of any stroke, ischemic stroke, and hemorrhagic stroke during a mean follow-up interval of 16.3 years (280 522 person-years at risk; 1435 incident stroke events). We applied Cox proportional-hazards models and pooled hazard ratios (HRs) using random-effects meta-analyses. After adjustments for age, sex, race, and vascular risk factors, higher MCP-1 levels were associated with increased risk of any stroke (HR per 1-SD increment in ln-transformed MCP-1, 1.07; 95% CI, 1.01–1.14). Focusing on stroke subtypes, we found a significant association between baseline MCP-1 levels and higher risk of ischemic stroke (HR, 1.11 [1.02–1.21]) but not hemorrhagic stroke (HR, 1.02 [0.82–1.29]). The results followed a dose-response pattern with a higher risk of ischemic stroke among individuals in the upper quartiles of MCP-1 levels as compared with the first quartile (HRs, second quartile: 1.19 [1.00–1.42]; third quartile: 1.35 [1.14–1.59]; fourth quartile: 1.38 [1.07–1.77]). There was no indication for heterogeneity across studies, and in a subsample of 4 studies (12 516 individuals), the risk estimates were stable after additional adjustments for circulating levels of IL (interleukin)-6 and high-sensitivity CRP (C-reactive protein).

CONCLUSIONS: Higher circulating levels of MCP-1 are associated with increased long-term risk of stroke. Our findings along with genetic and experimental evidence suggest that MCP-1 signaling might represent a therapeutic target to lower stroke risk.

VISUAL OVERVIEW: An online visual overview is available for this article.

Key Words: atherosclerosis ■ cerebrovascular disorders ■ chemokine CCL2 ■ inflammation ■ stroke

In This Issue, see p 725 | Meet the First Author, see p 726

Stroke is the leading cause of adult disability and the second most common cause of death worldwide.[1,2] Inflammatory mechanisms contribute to the pathogenesis of stroke, most notably to large artery atherosclerotic stroke,[3,4] but the specific proinflammatory factors mediating stroke risk are largely elusive. Discordant results from the CANTOS (Canakinumab Anti-Inflammatory Thrombosis Outcomes Study)[5–8] and CIRT (Cardiovascular

Correspondence to: Martin Dichgans, MD, Institute for Stroke and Dementia Research, University Hospital of Ludwig-Maximilians-University, Feodor-Lynen-Straße 17, 81377 Munich, Germany. Email martin.dichgans@med.uni-muenchen.de

The online-only Data Supplement is available with this article at https://www.ahajournals.org/doi/suppl/10.1161/CIRCRESAHA.119.315380.

For Sources of Funding and Disclosures, see page 781.

Circulation Research is available at www.ahajournals.org/journal/res

Novelty and Significance

What Is Known?

- Inflammatory mechanisms contribute to the pathogenesis of vascular disease, and inflammatory cytokines have been identified as potential therapeutic targets for lowering vascular risk.
- Using genetic data, we recently showed in Mendelian randomization that lifetime higher MCP-1 (monocyte chemoattractant protein-1) levels are associated with a higher risk of ischemic stroke.
- Preclinical studies in animal models of experimental atherosclerosis further suggest a critical role of MCP-1 in the initiation and propagation of atherosclerosis.

What New Information Does This Article Contribute?

- We performed a meta-analysis of 6 population-based cohort studies involving 17 000 stroke-free individuals who were followed up for 16 years.
- After adjustment for traditional vascular risk factors, higher baseline MCP-1 levels were associated with a higher risk of any stroke and ischemic stroke but not hemorrhagic stroke over follow-up.
- On top of experimental and genetic data, our findings provide additional evidence supporting MCP-1 signaling as a promising target for lowering stroke risk.

In view of recent findings suggesting the efficacy of anti-inflammatory approaches in lowering vascular risk, there is a need for identification of specific inflammatory mediators that show promise as potential therapeutic targets. Experimental and genetic evidence suggests MCP-1—a chemokine involved in monocyte recruitment—to play a critical role in atherosclerosis and stroke. Here, we aimed to amplify this concept by exploring in a meta-analysis of 6 previously unpublished cohort studies whether MCP-1 levels are associated with risk of stroke. Following up 17 000 stroke-free individuals for a mean of 16 years, we found baseline MCP-1 levels to be associated with a higher risk of any stroke, independently of traditional vascular risk factors. Across stroke subtypes, there was a significant association of MCP-1 levels with the risk of ischemic stroke but not hemorrhagic stroke. Adjustments for IL-6 (interleukin-6) and CRP (C-reactive protein) levels did not attenuate these associations, thus indicating that MCP-1 signaling might contribute to stroke risk independently of the well-established IL-6–CRP axis. Along with genetic and experimental data, our findings provide triangulation of evidence suggesting MCP-1 as a causal risk factor for stroke and MCP-1 signaling as a potential therapeutic target.

Nonstandard Abbreviations and Acronyms

ARIC	Atherosclerosis Risk in Communities
CAD	coronary artery disease
CANTOS	Canakinumab Anti-Inflammatory Thrombosis Outcomes Study
CCL2	CC-chemokine ligand 2
CRP	C-reactive protein
DHS	Dallas Heart Study
eGFR	estimated glomerular filtration rate
EPIC-Norfolk	Norfolk Arm of the European Prospective Investigation of Cancer
FHS	Framingham Heart Study
HbA1c	glycosylated hemoglobin type A1C
HDL	high-density lipoprotein
HR	hazard ratio
hsCRP	high-sensitivity C-reactive protein
IL	interleukin
KORA	Kooperative Gesundheitsforschung in der Region Augsburg
LDL	low-density lipoprotein
MCP-1	monocyte chemoattractant protein-1
MDCS	Malmö Diet and Cancer Study
MONICA	Monitoring of Trends and Determinants in Cardiovascular Disease Communities

Inflammation Reduction Trial)[6] randomized controlled trials emphasize the importance of targeting specific mediators and pathways for lowering vascular risk.[5–8] Treatment with an anti–IL (interleukin)-1β monoclonal antibody reduced the levels of IL-6 and high-sensitivity CRP [C-reactive protein] (hsCRP) leading to a reduction in the combined primary end point of nonfatal myocardial infarction, nonfatal stroke, or cardiovascular death independent of LDL (low-density lipoprotein) cholesterol levels,[5] whereas treatment with low-dose methotrexate neither reduced cardiovascular event rates nor the levels of IL-1β, IL-6, and hsCRP.[6]

In a Mendelian randomization study on circulating levels of 41 cytokines and growth factors, we recently found genetic predisposition to higher levels of the CC-chemokine MCP-1 (monocyte chemoattractant protein-1; also known as CCL2 [CC-chemokine ligand 2]) to be associated with increased risk of stroke, ischemic stroke, coronary artery disease (CAD), and myocardial infarction.[9] MCP-1 recruits monocytes to the subendothelial space of the atherogenic arterial wall,[10–12] and studies in experimental models of atherosclerosis suggest that targeting MCP-1 or its receptor CCR2 (C-C chemokine receptor type 2) limits plaque size, plaque progression, and plaque destabilization.[13–17] These findings define the MCP-1/CCR2 axis as a potential additional target for reducing residual inflammatory risk in vascular disease. However, data on MCP-1 and vascular risk in humans remain scarce.

Among patients with acute coronary syndromes in the OPUS-TIMI 16 (Orbofiban in Patients With Unstable Coronary Syndromes by the Thrombolysis in Myocardial Infarction Study Group)[18] and A-to-Z trial,[19] high circulating MCP-1 levels were associated with a significantly increased risk of death or myocardial infarction during follow-up, independently of baseline variables including hsCRP levels. In population-based studies, higher MCP-1 levels were associated with subclinical atherosclerosis and incident CAD during follow-up.[20,21] In contrast, the relationship between circulating MCP-1 levels and incident stroke remains unknown as does the relationship between MCP-1, IL-6, and CRP in mediating vascular risk.

Here, leveraging data from 6 population-based prospective cohort studies encompassing 17 180 stroke-free individuals with long-term follow-up, we set out to (1) determine the association between circulating MCP-1 levels at baseline and risk of incident stroke, (2) explore associations of MCP-1 levels with risk of major stroke subtypes (incident ischemic and hemorrhagic stroke), and (3) assess whether any association with stroke risk is independent of the IL-6/CRP axis by adjusting for the circulating levels of IL-6 and hsCRP.

METHODS

This study is based on summary statistics produced by the studies included in the systematic review. The main individual-study results are provided in the Online Data Supplement. All summary data that support the findings of this study are further available from the corresponding author on reasonable request. For accessing individual-level data of the included studies the readers should contact the authors representing the respective studies and follow the required processes.

Systematic Review

We systematically searched PubMed from inception through March 15, 2019, for population-based prospective cohort studies exploring associations between circulating MCP-1 levels and the risk of incident vascular outcomes including CAD, myocardial infarction, fatal or nonfatal stroke, and peripheral artery disease. The reference lists of the identified studies were further hand searched. The detailed search strategy is available in the Online Appendix. We subsequently contacted the corresponding authors of the selected studies inquiring about their interest to contribute data for the current meta-analysis examining the association between circulating MCP-1 levels and risk of incident stroke. Investigators of the following 6 studies agreed to participate, and the following studies were thus included in the current meta-analysis: the ARIC study (Atherosclerosis Risk in Communities),[20] DHS (Dallas Heart Study),[21] the EPIC-Norfolk study (Norfolk Arm of the European Prospective Investigation of Cancer),[22] the Offspring Cohort of FHS (Framingham Heart Study),[23] the MONICA (Monitoring of Trends and Determinants in Cardiovascular Disease) subcohort of the KORA study (Kooperative Gesundheitsforschung in der Region Augsburg),[24] and the cardiovascular subcohort of MDCS (Malmö Diet and Cancer Study).[25] With the exception of the FHS Offspring study, which had previously published part of the data included in this analysis (96 versus 172 incident events),[23] none of the studies previously published data on the association between circulating MCP-1 levels and risk of incident stroke. The flowchart describing the study selection is depicted in Online Figure I.

Study Populations, MCP-1 Level Measurements, and Assessment of Stroke Outcomes

The study design, population characteristics, methods used for quantifying circulating MCP-1 levels, stroke outcome definitions, and assessments in individual cohorts are detailed in Online Table I. In brief, all studies were population-based prospective cohorts, and participants included in the current analyses were selected from these cohorts based on the availability of MCP-1 measurements at baseline. Circulating MCP-1 levels were measured in serum or plasma samples drawn during the baseline assessments. Because incident stroke was the primary outcome of the current study, all participants with a history of stroke at baseline assessments (prevalent cases) were excluded from subsequent analyses. Stroke occurrence was assessed at follow-up visits during mean intervals of 11 to 23 years based on self-reported information and validation from medical records of the participants. In addition to information on any stroke, all studies further provided information on the major stroke subtypes (ischemic versus hemorrhagic stroke).

Quality Assessment

Study quality was assessed using the cohort subscale of the Newcastle-Ottawa scale.[26] The criteria for awarding quality points were the following: a general population sample (representativeness of exposed cohort), selection of patients for inclusion independently of MCP-1 levels (selection of the nonexposed cohort), measurement of MCP-1 levels in the serum or plasma based on a validated assay (ascertainment of exposure), exclusion of patients with prevalent stroke at baseline (outcome not present at the start of study), adjustments for age and sex, as well as for conventional vascular risk factors (comparability items), assessment of stroke outcomes blindly to MCP-1 levels with validation based on medical records (assessment of outcome), a follow-up interval >5 years (follow-up duration), and a completion of follow-up rate of >90% (adequacy of follow-up cohorts).

Statistical Analysis

A predefined analysis protocol was circulated to investigators of each of the cohort studies requesting summary results for meta-analysis. MCP-1 levels were ln-transformed in all studies for normalization. We did not consider absolute MCP-1 values because of marked differences in mean MCP-1 level values between studies, probably related to different assays used for MCP-1 quantification (Table). We first examined descriptive associations between MCP-1 levels and conventional vascular risk factors. We pooled study-specific Z scores reflecting differences of MCP-1 levels from the overall mean of each study with random-effects models across the risk factor categories and statistically examined associations using meta-regression.

To examine associations between baseline MCP-1 levels and incident stroke, Cox proportional-hazards models were fit in each study. MCP-1 levels were included in the models as either a continuous variable (1-SD increment in ln-transformed MCP-1 levels) or categorized in 4 quartiles (first quartile as reference category) to also assess for potential

Table. Descriptive Baseline Characteristics of the 6 Included Population-Based Prospective Cohort Studies

Cohort	ARIC	DHS	EPIC-Norfolk	FHS Offspring	MONICA/KORA	MDCS-CV
Geographic setting (baseline assessment)	The United States (1986–1989)	The United States (2000–2002)	United Kingdom (1993–1997)	The United States (1998–2001)	Germany (1984–2002)	Sweden (1991–1994)
Individuals included in the analysis, n	1234	2931	3182	3069	2055	4709
Follow-up, y	23.0 [13.2–27.8]	11.0 (1.7)	16.8 (6.4)	13.8 (3.7)	15.7 (6.4)	19.5 (4.9)
Incident stroke events, n	153	64	503	172	116	427
Incident ischemic stroke events, n	141	42	458	141	99	352
Incident hemorrhagic stroke events, n	12	9	76	22	17	69
Fatal stroke events, n	10	6	132	26	22	30
Age, y	56.9 (5.3)	44.0 (10.0)	65.3 (7.8)	61.6 (9.4)	52.4 (10.3)	57.5 (4.9)
Male sex, n (%)	738 (59.8)	1254 (42.8)	2009 (63.1)	1421 (46.3)	1093 (53.2)	1873 (39.8)
Hypertension, n (%)	417 (33.9)	944 (32.7)	2029 (63.8)	1378 (44.9)	877 (42.7)	2958 (62.8)
SBP, mm Hg	125 (20)	124 (19)	141 (18)	127 (19)	133 (19)	141 (19)
DBP, mm Hg	74 (12)	78 (10)	85 (11)	74 (10)	82 (11)	87 (9)
Diabetes mellitus, n (%)	156 (12.6)	296 (10.1)	623 (19.6)	379 (12.3)	103 (5.0)	183 (3.9)
Hypercholesterolemia, n (%)	760 (61.6)	377 (12.9)	414 (13.0)	1615 (52.6)	1251 (57.4)	2918 (62.8)
LDL cholesterol levels, mg/dL	142.8 (39.9)	107.4 (35.3)	160.1 (39.4)	119.9 (32.7)	148.5 (2.4)	161.3 (37.9)
HDL cholesterol levels, mg/dL	49.6 (16.5)	50.0 (14.6)	51.8 (15.1)	53.9 (16.7)	56.0 (17.0)	53.8 (14.3)
BMI, kg/m^2	27.4 (5.1)	29.7 (7.0)	26.6 (3.6)	28.1 (5.3)	27.2 (4.1)	25.6 (3.9)
Smoking status, n (%)						
Never smokers	461 (37.3)	1639 (55.9)	1201 (10.3)	1077 (35.1)	947 (46.1)	1916 (40.1)
Ex-smokers	397 (32.2)	496 (16.9)	1652 (51.9)	1604 (52.3)	591 (28.8)	1777 (37.8)
Current smokers	376 (30.5)	796 (27.2)	329 (37.7)	388 (12.6)	517 (25.1)	1010 (21.5)
eGFR, mL/min per 1.73 m^2	100.0 (16.6)	99.5 (23.7)	74.5 (24.9)	83.3 (16.5)	87.9 (17.4)	76.9 (15.3)
Coronary artery disease, n (%)	68 (5.5)	79 (2.7)	0 (0)	265 (8.6)	46 (2.2)	78 (1.7)
Atrial fibrillation, n (%)	1 (0.1)	35 (1.2)	NA	119 (3.9)	NA	34 (0.7)
Heart failure, n (%)	53 (4.3)	83 (2.8)	0 (0)	31 (1.0)	119 (5.7)	2 (0.04)
hsCRP levels, mg/L	2.4 [1.3–5.3]	2.8 [1.2–6.8]	2.0 [1.0–3.8]	2.2 [1.0–5.1]	1.4 [0.7–3.3]	1.3 [0.7-2.7]
Sample used for MCP-1 assessment	Plasma	Plasma	Serum	Serum	Serum	Plasma
MCP-1 levels, pg/mL	398.9 [348.4–467.1]	166.5 [122.9–224.4]	51.5 [38.8–68.1]	313.4 [253.9–382.3]	298.0 [127.6–323.8]	2.52 [2.22–2.82]*

The numbers correspond to n (%) for categorical variables and to mean (SD) or median [25th–75th percentile] for continuous variables. ARIC indicates Atherosclerosis Risk in Communities Study; BMI, body mass index; DBP, diastolic blood pressure; DHS, Dallas Heart Study; eGFR, estimated glomerular filtration rate; EPIC-Norfolk, European Prospective Investigation of Cancer, Norfolk; FHS Offspring, Framingham Heart Study–Offspring Cohort; HDL, high-density lipoprotein; hsCRP, high-sensitivity C-reactive protein; KORA, Kooperative Gesundheitsforschung in der Region Augsburg; LDL, low-density lipoprotein; MCP-1, monocyte chemoattractant protein-1; MDCS-CV, Malmö Diet and Cancer Study–Cardiovascular Subcohort; MONICA, Monitoring of Trends and Determinants in Cardiovascular Disease; NA, not available; PEA, proximity extension assay; and SBP, systolic blood pressure.

*The used assay in MDCS did not provide MCP-1 measurements as absolute values but as relative expression levels obtained by PEA.

nonlinear associations. We applied 3 models with different levels of adjustment: model 1 was adjusted for age, sex, and race; model 2 was additionally adjusted for conventional vascular risk factors (hypertension, diabetes mellitus, hypercholesterolemia, body mass index, smoking [current versus noncurrent], estimated glomerular filtration rate [eGFR], CAD, atrial fibrillation, and heart failure); and model 3 was further adjusted for circulating hsCRP levels on top of these variables. Model 2 was predefined as our main model for analyses. In these models, we defined hypertension as a history of physician-diagnosed hypertension, systolic blood pressure ≥140 mm Hg, diastolic blood pressure ≥90 mm Hg, or use of ≥1 antihypertensive medications.[27] We defined diabetes mellitus as a history of physician-diagnosed diabetes mellitus, HbA1c (glycosylated hemoglobin type A1C) ≥6.5%, fasting glucose ≥126 mg/dL, random glucose levels ≥200 mg/dL, or use of glucose-lowering medications.[28] Hypercholesterolemia was defined as LDL cholesterol levels ≥130 mg/dL, total cholesterol levels ≥200 mg/dL (if LDL cholesterol was not available) or use of lipid-lowering drugs,[29] and chronic kidney disease as eGFR <60 mL/min per 1.73 m^2.[30] In an alternative model (alternative model 2), we directly adjusted for the components of these definitions instead of the binary variables: thus, instead of hypertension, diabetes mellitus, hypercholesterolemia, and chronic kidney disease, we included systolic blood pressure (as a continuous variable), use of antihypertensive medications, fasting glucose levels (as continuous), use of glucose-lowering medications, LDL cholesterol levels (as continuous), administration of lipid-lowering medications, and eGFR (as continuous).

The purpose of the main models was to explore MCP-1 as a potentially causal risk factor for stroke and not to evaluate the predictive values of its levels. In subsequent models, we aimed to explore whether the association between MCP-1 levels and risk of stroke is independent of the IL-6/CRP pathway that was recently shown to provide an efficient drug target for reducing vascular risk.[31] To indirectly examine this, we applied additional adjustments for circulating IL-6 and hsCRP levels. In one model, we included IL-6 on top of age, sex, race, and vascular risk factors, and in a subsequent model, we included both IL-6 and hsCRP levels. We did this because CRP is a downstream effector of IL-6 but also comprises a more general marker of inflammation, and thus the alternative adjustments provide different levels of information regarding the involved inflammatory pathways. Data for IL-6 circulating levels were not available in ARIC and the EPIC-Norfolk. Thus, these cohorts were not included in these analyses.

Analyses were conducted separately for any stroke, ischemic stroke, and hemorrhagic stroke. DHS was excluded from the analysis for hemorrhagic stroke, where MCP-1 was examined in quartiles, because of the low numbers of incident events across the quartile categories of MCP-1 levels. The hazard ratios (HRs) and the 95% CIs derived from each study were pooled with random-effects (DerSimonian-Laird) meta-analyses to allow for heterogeneity across studies related to the different baseline characteristics and the different methods of MCP-1 assessment. Heterogeneity across studies was assessed with the I^2 and the Cochran Q statistic (I^2 >50% and P<0.10 were considered statistically significant).

To examine whether the pooled risk estimates were driven by any individual study, we also applied sensitivity analyses by pooling the risk estimates across studies after excluding one study at a time. To explore potential interactions between MCP-1 levels and known cardiovascular risk factors, we performed meta-regression analyses examining how the prevalence of cardiovascular risk factors or the mean or median values of biomarkers were associated with the risk estimates for stroke in each study. We further performed subgroup analyses by sex, presence of hypertension, presence of diabetes mellitus, and body mass index levels (<30 versus ≥30 kg/m^2). Differences in the effect sizes across the subgroup categories were examined by assessing heterogeneity (I^2 >50% and P<0.10 were considered statistically significant). Finally, we performed separate analyses for fatal and nonfatal stroke (fatal stroke defined as death occurring within 30 days after the stroke event).

Statistical significance was set at a 2-sided P<0.05 for the main analysis for any stroke. For the subsequent analysis for stroke subtypes, we corrected for multiple comparisons based on the Bonferroni method (P<0.05/2 stroke subtypes, 0.025). Finally, we corrected for multiple comparisons in the descriptive analyses exploring the correlations between MCP-1 levels and baseline variables (threshold for statistical significance at P<0.05/12 variables, 0.004). All analyses were conducted with SAS (v9.4) and Stata (v13.0).

RESULTS

Following a systematic review and contact with the lead investigators, 6 population-based prospective cohort studies contributed previously unpublished data for this meta-analysis. All studies scored high in quality as they fulfilled the full set of Newcastle-Ottawa scale criteria (Online Table II). The baseline characteristics of each study are presented in the Table. In total, 17 180 individuals (mean age, 56.7±8.1 years; 48.8% men), who were stroke-free at baseline, were followed for a mean interval of 16.3 years (range of mean follow-up, 11–23 years) with 280 522 person-years at risk. A total of 1435 incident stroke cases were diagnosed during follow-up, which were classified as ischemic in 1233 cases and as hemorrhagic in 205 cases. Two hundred twenty-six (15.7%) incident stroke events were fatal. Median MCP-1 levels differed between studies possibly reflecting differences in the methods used for MCP-1 quantification (Online Table I). Figure 1 displays associations of standardized MCP-1 levels with conventional vascular risk factors in the pooled sample. We found the following baseline factors to be associated with higher circulating MCP-1 levels: older age, male sex, higher systolic blood pressure, presence of diabetes mellitus, higher LDL cholesterol levels, higher HDL (high-density lipoprotein) cholesterol levels, higher body mass index, current smoking, lower eGFR, history of CAD, higher hsCRP levels, and higher IL-6 levels.

In the pooled analysis, we found higher MCP-1 levels at baseline to be associated with an increased risk of any stroke both in a model adjusted for age, sex, and race (model 1: HR per 1-SD increment in ln-transformed MCP-1, 1.10; 95% CI, 1.01–1.19; P=0.02) and in the main model further adjusted for vascular risk factors (model 2: HR, 1.07; 95% CI, 1.01–1.14; P=0.03; Figure 2; Online Table III). In analyses comparing MCP-1 quartiles, we found the association between MCP-1 levels and risk of stroke to follow a dose-response pattern with a higher risk among individuals in the upper quartiles of circulating MCP-1 levels as compared with the first quartile (HRs from model 2: second quartile, 1.16 [95% CI, 0.99–1.36; P=0.07]; third quartile, 1.31 [95% CI, 1.12–1.53; P=0.001]; fourth quartile, 1.33 [95% CI, 1.05–1.68; P=0.008]). The results were further stable in a model additionally adjusting for circulating hsCRP levels (model 3 in Figure 2 and Online Table III).

We next examined the associations of circulating MCP-1 levels at baseline with stroke subtypes (Figure 3; Online Tables IV and V) and found significant associations of higher MCP-1 levels at baseline with the risk of ischemic stroke (HR per 1-SD increment in ln-MCP-1 from model 2, 1.11; 95% CI, 1.02–1.21; P=0.009) but not with hemorrhagic stroke (model: HR, 1.02; 95% CI, 0.82–1.29; P=0.83). MCP-1 levels in the second, third, and fourth quartiles, as compared to the first, were associated with a higher risk for ischemic stroke after adjusting for age, sex, race, and vascular risk factors (HRs from model 2: second quartile, 1.19 [95% CI, 1.00–1.42; P=0.05]; third quartile, 1.35 [95% CI, 1.14–1.59; P<0.001]; fourth quartile, 1.38 [95% CI, 1.07–1.77; P=0.008]). The results

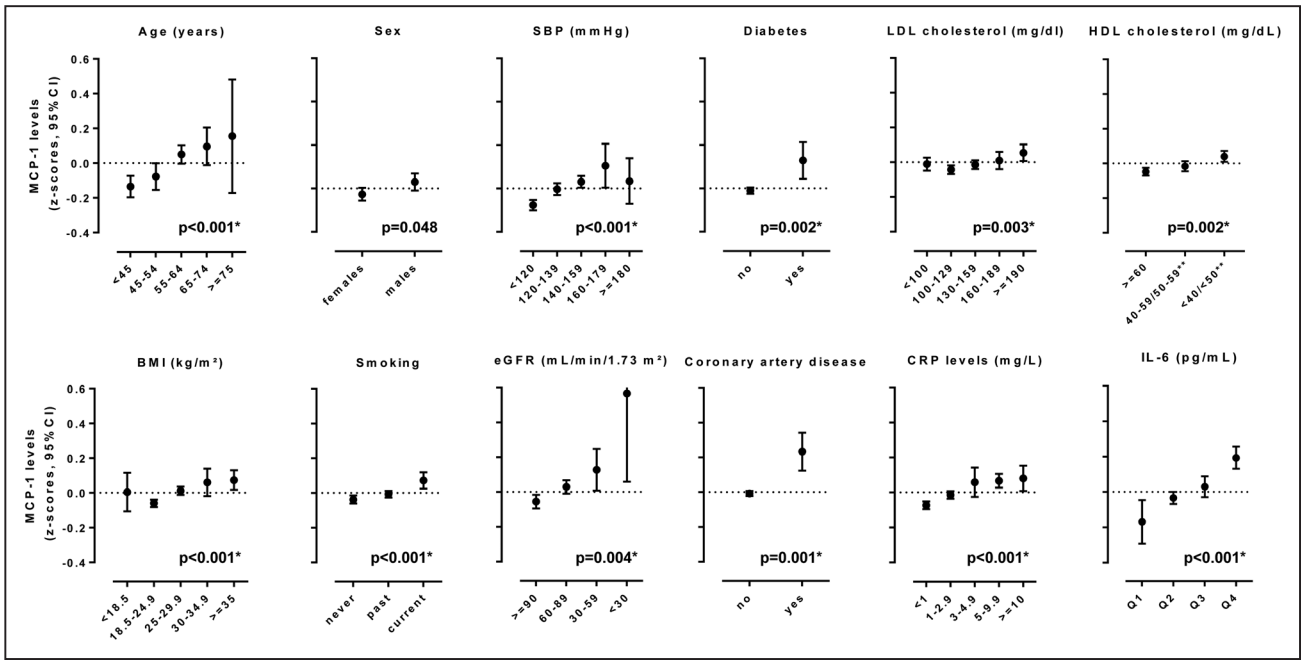


Figure 1. Cross-sectional associations between baseline circulating MCP-1 (monocyte chemoattractant protein-1) levels, demographic factors, conventional vascular risk factors, and inflammatory biomarkers.
Shown are the results from the pooled sample consisting of 6 population-based studies. *Z* score for circulating MCP-1 levels correspond to differences from the mean value of each study. *P* values are derived from meta-regression. BMI indicates body mass index; eGFR, estimated glomerular filtration rate; HDL, high-density lipoprotein; hsCRP, high-sensitivity C-reactive protein; IL, interleukin; LDL, low-density lipoprotein; and SBP, systolic blood pressure. *Statistically significant results (after correction for multiple comparisons, statistical significance was set at $P<0.05/12=0.004$). **<40 and 40 to 59 mg/dL for men, <50 and 50 to 59 mg/dL for women.

were highly consistent in the model additionally adjusting for circulating hsCRP levels on top of the vascular risk factors (model 3 in Figure 3 and Online Table IV).

Study-specific risk estimates are depicted in Online Figures II through IV. There was no evidence of heterogeneity in any of the analyses (I^2 <50% and Cochran Q-derived $P>0.10$), except for moderate heterogeneity in the analysis of the upper fourth MCP-1 quartile for any stroke and ischemic stroke (I^2=49.8%, P=0.08 and I^2=46.1%, P=0.10, respectively). The results were similar for both fatal and nonfatal stroke (I^2=0% for between-subgroup comparisons), although the CIs

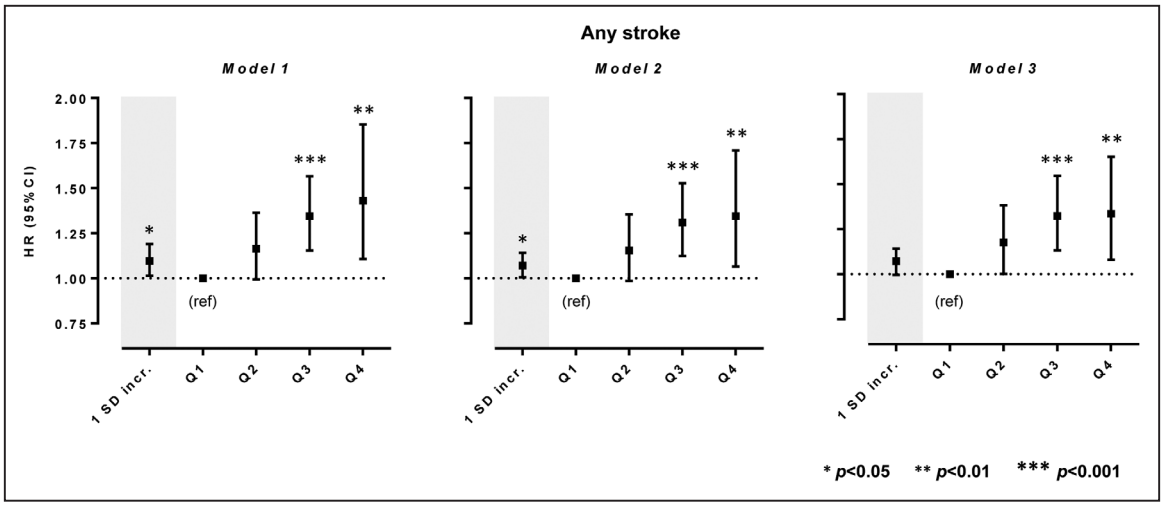


Figure 2. Associations between baseline circulating MCP-1 (monocyte chemoattractant protein-1) levels and risk of any stroke.
Shown are the results from random-effects meta-analyses of the pooled sample consisting of 6 population-based studies. Model 1 is adjusted for age, sex, and race. Model 2 is adjusted for age, sex, race, and vascular risk factors including body mass index (1 kg/m² increment [incr.]), smoking (current vs noncurrent), estimated glomerular filtration rate (1 mL/min per 1.73 m² incr.), history of coronary artery disease, diabetes mellitus, hypercholesterolemia, hypertension, atrial fibrillation, and heart failure at baseline. Model 3 is additionally adjusted for circulating high-sensitivity CRP (C-reactive protein) levels. Analyses for 1-SD incr. correspond to ln-transformed MCP-1 levels. HR indicates hazard ratio.

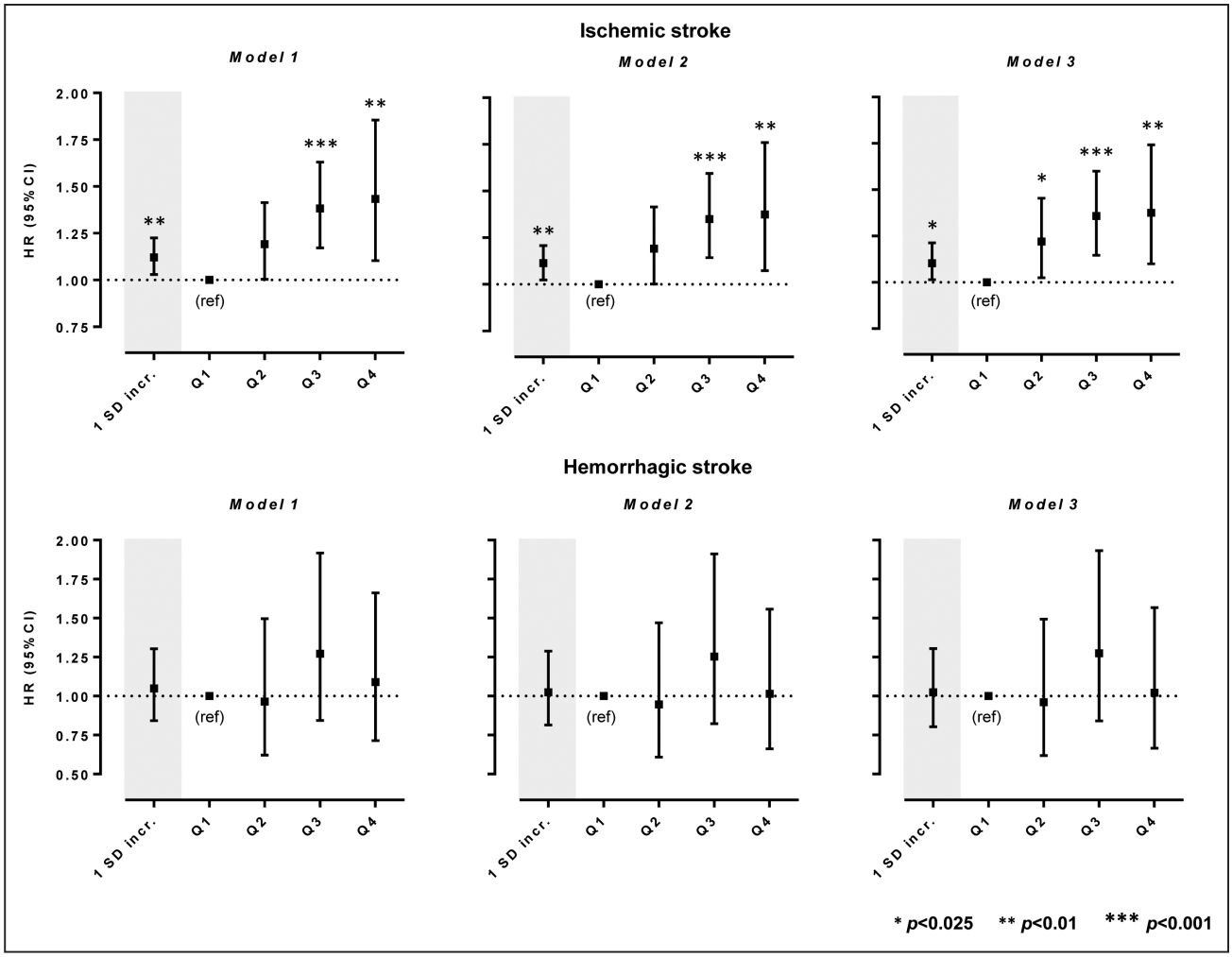


Figure 3. Associations between baseline circulating MCP-1 levels and risk of ischemic and hemorrhagic stroke. Shown are the results from random-effects meta-analyses of the pooled sample consisting of 6 population-based studies for (**A**) ischemic and (**B**) hemorrhagic stroke. Model 1 is adjusted for age, sex, and race. Model 2 is adjusted for age, sex, race, and vascular risk factors including body mass index (1 kg/m^2 increment [incr.]), smoking (current vs noncurrent), estimated glomerular filtration rate (1 mL/min per 1.73 m^2 incr.), history of coronary artery disease, diabetes mellitus, hypercholesterolemia, hypertension, atrial fibrillation, and heart failure at baseline. Model 3 is additionally adjusted for circulating high-sensitivity CRP (C-reactive protein) levels. Analyses for 1-SD incr. correspond to ln-transformed MCP-1 levels. HR indicates hazard ratio. *Statistical significance threshold was set at $P<0.05/2=0.025$ after correction for multiple comparisons (2 stroke subtypes).

for fatal stroke were wider probably because of lower statistical power (Online Figure V). The association estimates remained consistent in alternative models directly adjusting for the crude components of vascular risk factors (systolic blood pressure, fasting glucose levels, LDL cholesterol, and eGFR) and use of antihypertensive, glucose-lowering, or lipid-lowering medications (alternative model 2; Online Tables III through V). Furthermore, the results remained stable in sensitivity analyses omitting one study per time (leave-one-out analysis) showing that the results were not driven by any individual study (Online Figures VI through VIII). Meta-regression analyses showed that none of the examined study population characteristics nor the sample source (serum versus plasma) modified the associations of MCP-1 with the risk of any stroke, ischemic stroke, or hemorrhagic stroke (Online Table VI). Finally, in subgroup analyses stratifying for sex, hypertension, diabetes mellitus, and body mass index (≥30 versus <30 kg/m^2), there was no indication for heterogeneity in the risk estimates for any stroke, ischemic stroke, and hemorrhagic stroke between subgroups (I^2=0%; Online Figure IX).

As a last step, we performed analyses with additional adjustments for IL-6 and hsCRP levels in 4 studies (12516 individuals; 758 incident stroke events) with available data. Adjustment for IL-6 levels showed that the risk estimates between MCP-1 levels and risk of stroke and stroke subtypes remained stable, although with wider CIs than the main analysis, as would be expected given the smaller sample sizes (Online Table VII). Similarly, simultaneous adjustments for both IL-6 and hsCRP did not alter the risk estimates between MCP-1 and risk of stroke or stroke subtypes, even though both variables

were associated with the risk of any stroke and ischemic stroke (Online Table VII).

DISCUSSION

Pooling data from 6 population-based cohort studies involving 17 180 stroke-free individuals, we found higher circulating levels of MCP-1 at baseline to be associated with a higher long-term risk of stroke after accounting for age, sex, race, and vascular risk factors. In analyses for stroke subtypes, MCP-1 levels were specifically associated with the risk of ischemic stroke but not with hemorrhagic stroke. These associations followed a dose-response pattern, and risk estimates were stable after additional adjustments for serum levels of IL-6 or hsCRP.

Our results, which were obtained in studies with long-term follow-up, confirm and extend our recent Mendelian randomization finding of a higher stroke risk among individuals with genetic predisposition to higher lifetime MCP-1 levels.[9] The results were remarkably consistent between the 2 approaches: with Mendelian randomization, the odds ratio for stroke was 1.06 per SD increment in genetically determined MCP-1 levels, which is almost identical to the HR for incident stroke observed in the current meta-analysis of observational studies. In accord with the Mendelian randomization results, higher MCP-1 levels were further associated with a higher risk of incident ischemic stroke, but not hemorrhagic stroke, which is consistent with the established role of MCP-1 in experimental atherosclerosis. The magnitude of association of MCP-1 with incident ischemic stroke was modest suggesting that MCP-1 measurement is not likely to be of value as a risk marker for stroke although this would need to be formally examined. Of note, however, risk estimates compare well with those for lipoprotein (a),[32,33] which is established as a causal risk factor for atherosclerosis currently under investigation in clinical trials.[34,35] When viewed together with the genetic[9] and experimental data,[13–17] our findings provide triangulation of evidence regarding a role of MCP-1 as a causal risk factor for stroke.

Only limited human data exist supporting vascular benefits by reducing inflammation. Secondary analyses from the CANTOS trial showed that the reductions in vascular event rates after IL-1β inhibition were restricted to individuals with a substantial decrease in IL-6 or hsCRP levels.[31,36] Importantly, the risk estimates for stroke by MCP-1 levels in our study remained stable after additional adjustments for the baseline levels of IL-6, hsCRP, and both IL-6 and hsCRP. This observation provides indirect evidence suggesting that elevated levels of MCP-1 might influence risk of stroke independently of the IL-1β/IL-6/CRP axis. Thus, targeting the MCP-1/CCR2 pathway might serve as an alternative anti-inflammatory strategy with independent and complementary effects in reducing vascular event rates on top of current approaches.

Deficiency of either MCP-1[15,17] or its receptor CCR2[16] decreases plaque burden and limits lipid deposition and macrophage infiltration in experimental models of atherosclerosis. Similar effects are observed with pharmacological treatment using MCP-1 competitors[13] or CCR2 antagonists.[14,37–39] In contrast, overexpression of MCP-1 promotes oxidized lipid accumulation, macrophage infiltration, and smooth muscle cell proliferation, thus accelerating atherosclerosis.[40] To our knowledge, there has been only one small phase II randomized controlled trial in the context of atherosclerosis in humans that targeted the MCP-1/CCR2 axis. Among 108 patients with cardiovascular risk factors and hsCRP levels >3 mg/L, those treated with a single intravenous infusion of MLN1202—a humanized monoclonal antibody against CCR2—exhibited significant reductions in hsCRP levels after 4 weeks and continuing through 12 weeks after dosing.[41] However, this study did not assess clinical outcomes, which would need to be examined in a larger trial.[41]

Our study has several strengths. The pooled analysis was based on a large sample size of >17 000 individuals from 6 previously unpublished population-based prospective studies with long follow-up intervals and a large number of incident events, thus providing sufficient statistical power to identify robust associations. The included studies fulfilled all of the criteria of quality assessment, which minimized the risk of several sources of bias. We further applied extensive adjustments for demographic and vascular risk factors thus accounting for confounding and enabling the identification of independent associations between MCP-1 levels and risk of stroke. Finally, in 4 of the cohorts, we had available data on IL-6 and hsCRP measurements, which allowed examining the associations between MCP-1 and stroke after adjusting for these biomarkers.

Our study also has limitations. First, the different assays used by individual studies to quantify circulating MCP-1 levels and the different sample sources (plasma versus serum) resulted in substantial variations in MCP-1 levels between studies. Although our analyses standardized MCP-1 levels across studies, it was not possible to explore associations between absolute MCP-1 values and risk of stroke. Second, studies differed in terms of demographic characteristics and prevalence of vascular risk factors. While we found no evidence of substantial heterogeneity between studies, there was moderate heterogeneity in the analyses for the highest quartiles of MCP-1, which could possibly be explained by the differences in baseline MCP-1 levels and in vascular risk profiles between studies. Third, we could not explore associations between MCP-1 levels and risk of ischemic stroke subtypes (large artery, cardioembolic, and small vessel stroke) because information on deeper phenotyping was not available for the majority of studies. Fourth, our analyses were based on predominantly

European ancestry individuals and do thus not necessarily apply to other ethnic groups. Fifth, we cannot exclude residual confounding. Finally, based on our a priori determined approach and power calculations, we corrected for multiple comparisons within each level of analysis but not across all analyses. Although this would not be expected to have any impact on the findings, future studies with even larger sample sizes would be useful in replicating our results

In conclusion, this meta-analysis demonstrates that higher circulating levels of MCP-1 among stroke-free individuals are associated with increased long-term risk of ischemic stroke. The results extend and corroborate experimental and genetic evidence suggesting a key role of MCP-1 in atherosclerosis and stroke. Additional work is needed to examine whether interventions aimed at interfering with MCP-1 signaling would lower stroke risk.

ARTICLE INFORMATION

Received May 14, 2019; revision received August 30, 2019; accepted September 1, 2019.

Affiliations

From the Institute for Stroke and Dementia Research, University Hospital (M.K.G., R.M., M.D.) and Graduate School for Systemic Neurosciences (M.K.G.), Ludwig-Maximilians-University, Munich; Clinical Sciences, Malmö, Lund University, Sweden (H.B., G.E., O.M., M.O.-M., A.S., M.S., J.N.); Institute of Applied Health Sciences, School of Medicine, Medical Sciences and Nutrition, University of Aberdeen, Scotland (T.A.P., P.K.M.); Biostatistics, Boston University School of Public Health, MA (S.D., A.S.B.); National Heart, Lung, and Blood Institute's and Boston University's Framingham Heart Study, MA (S.D., A.S.B., E.J.B., S.S.); Cardiology, University of Texas Southwestern Medical Center, Dallas (C.A., J.A.d.L.); Research Unit of Molecular Epidemiology (M.A.E.) and Institute of Epidemiology (M.A.E., A.P.), Helmholtz Zentrum München, German Research Center for Environmental Health, Neuherberg; German Research Center for Cardiovascular Disease, Partner site Munich Heart Alliance (M.A.E., W.K.); Brown Foundation Institute of Molecular Medicine, McGovern Medical School and Human Genetics Center, School of Public Health, University of Texas Health Science Center, Houston (M.F.); Department of Neurology, Boston University School of Medicine, MA (A.S.B.); Medicine, Boston University School of Medicine, MA (E.J.B., S.S.); Epidemiology, Boston University School of Public Health, MA (E.J.B.); Amsterdam UMC, University of Amsterdam, Cardiology, the Netherlands (M.S.B.); Institute for Clinical Diabetology, German Diabetes Center, Leibniz Center for Diabetes Research at Heinrich Heine University Düsseldorf (C.H.); German Center for Diabetes Research, Partner Düsseldorf (C.H.); Endocrinology and Diabetology, Medical Faculty, Heinrich Heine University Düsseldorf, Germany (C.H.); Medicine, Baylor College of Medicine, Houston, TX (R.C.H., C.M.B.); Deutsches Herzzentrum München, Technische Universität München, Germany (W.K.); Institute of Epidemiology and Biostatistics, University of Ulm, Germany (W.K.); Cardiology, Skåne University Hospital, Malmö, Sweden (A.S.); MRC Epidemiology Unit, University of Cambridge, United Kingdom (N.W.); Glenn Biggs Institute for Alzheimer's and Neurodegenerative Diseases, University of Texas Health Sciences Center, San Antonio (S.S.); Munich Cluster for Systems Neurology, Germany (M.D.); and German Centre for Neurodegenerative Diseases, Munich, Germany (M.D.).

Acknowledgments

We thank the staff and participants of all the included studies for their important contributions.

Sources of Funding

M.K. Georgakis is funded by scholarships from the German Academic Exchange Service and Onassis Foundation. The ARIC study (Atherosclerosis Risk in Communities Study) has been funded in whole or in part with Federal funds from the National Heart, Lung, and Blood Institute, National Institutes of Health (NIH), Department of Health and Human Services, under contract No. HHSN2682017000011, HHSN2682017000021, HHSN2682017000031, HHSN2682017000051, and HHSN2682017000041. The DHS (Dallas Heart Study) was funded by a grant from the Donald W. Reynolds Foundation. The EPIC-Norfolk study (Norfolk Arm of the European Prospective Investigation of Cancer) is funded by grants from the Medical Research Council UK (G9502233 and G0401527) and Cancer Research UK (C864/A8257 and C864/A2883). FHS (Framingham Heart Study) is supported by the National Heart, Lung and Blood Institute Framingham Heart Study (contract No. N01-HC-25195, HHSN268201500001I, and 75N92019D00031), received funding by grants from the National Institute of Aging (R01s AG054076, AG049607, AG059421, U01-AG049505, AG058589, and AG052409) and the National Institute of Neurological Disorders and Stroke (R01 NS017950 and UH2 NS100605), as well as grants for the MCP-1 (monocyte chemoattractant protein-1) measurements by NIH (1RO1 HL64753, R01 HL076784, and 1 R01 AG028321). The KORA study (Kooperative Gesundheitsforschung in der Region Augsburg) was initiated and financed by the Helmholtz Zentrum München–German Research Center for Environmental Health, which is funded by the German Federal Ministry of Education and Research and by the State of Bavaria. Furthermore, KORA research was supported within the Munich Center of Health Sciences (MC-Health), Ludwig-Maximilians-Universität, as part of LMUinnovativ. The MDCS-CV study (Malmö Diet and Cancer Study–Cardiovascular Subcohort) has been supported with funding from the Swedish Research Council, Swedish Heart and Lung Foundations, and the Swedish Foundation for Strategic Research. This project has received funding from the European Union's Horizon 2020 research and innovation programme (No. 666881), SVDs@target (to M. Dichgans) and No. 667375, CoSTREAM (Common Mechanisms and Pathways in Stroke and Alzheimer's Disease; to M. Dichgans); the DFG (Deutsche Forschungsgemeinschaft) as part of the Munich Cluster for Systems Neurology (EXC 2145 SyNergy—ID 390857198) and the CRC 1123 (B3, to M. Dichgans); the Corona Foundation (to M. Dichgans); the Fondation Leducq (Transatlantic Network of Excellence on the Pathogenesis of Small Vessel Disease of the Brain, to M. Dichgans); the e:Med program (e:AtheroSysMed, to M. Dichgans); and the FP7/2007-2103 European Union project CVgenes@target (grant agreement No. Health-F2-2013-601456, to M. Dichgans).

The funders had no role in study design, data collection, analysis, decision to publish, or preparation of the manuscript.

Disclosures

J.A. de Lemos reports research grants from Abbott Diagnostics and Roche Diagnostics, consulting from Ortho Clinical Diagnostics and Jannsen, honoraria for steering committee membership from Amgen, and DSMB (Data and Safety Monitoring Board) membership from Novo Nordisc and Regeneron. W. Koenig reports personal fees for consulting from AstraZeneca, Novartis, DalCor, Kowa, Amgen, and Sanofi and grants and nonfinancial support from Roche Diagnostics, Beckmann, Singulex, and Abbott. C. Ayers reports statistical consulting fees from the National Institutes of Health. R.C. Hoogeveen reports research grants and personal fees for consulting from Denka Seiken. The other authors report no conflicts.

REFERENCES

1. DALYs GBD, HALE Collaborators. Global, regional, and national disability-adjusted life-years (dalys) for 315 diseases and injuries and healthy life expectancy (hale), 1990-2015: a systematic analysis for the global burden of disease study 2015. *Lancet*. 2016;388:1603–1658.
2. Mortality GBD, Causes of Death Collaborators. Global, regional, and national life expectancy, all-cause mortality, and cause-specific mortality for 249 causes of death, 1980-2015: a systematic analysis for the global burden of disease study 2015. *Lancet*. 2016;388:1459–1544.
3. Esenwa CC, Elkind MS. Inflammatory risk factors, biomarkers and associated therapy in ischaemic stroke. *Nat Rev Neurol*. 2016;12:594–604. doi: 10.1038/nrneurol.2016.125
4. Libby P, Ridker PM, Hansson GK; Leducq Transatlantic Network on Atherothrombosis. Inflammation in atherosclerosis: from pathophysiology to practice. *J Am Coll Cardiol*. 2009;54:2129–2138. doi: 10.1016/j.jacc.2009.09.009
5. Ridker PM, Everett BM, Thuren T, MacFadyen JG, Chang WH, Ballantyne C, Fonseca F, Nicolau J, Koenig W, Anker SD, et al; CANTOS Trial Group. Antiinflammatory therapy with wanakinumab for atherosclerotic disease. *N Engl J Med*. 2017;377:1119–1131. doi: 10.1056/NEJMoa1707914
6. Ridker PM, Everett BM, Pradhan A, MacFadyen JG, Solomon DH, Zaharris E, Mam V, Hasan A, Rosenberg Y, Iturriaga E, et al; CIRT Investigators. Low-dose methotrexate for the prevention of atherosclerotic events. *N Engl J Med*. 2019;380:752–762. doi: 10.1056/NEJMoa1809798
7. Ridker PM. Anticytokine agents: targeting interleukin signaling pathways for the treatment of atherothrombosis. *Circ Res*. 2019;124:437–450. doi: 10.1161/CIRCRESAHA.118.313129

8. Aday AW, Ridker PM. Targeting residual inflammatory risk: a shifting paradigm for atherosclerotic disease. *Front Cardiovasc Med*. 2019;6:16. doi: 10.3389/fcvm.2019.00016
9. Georgakis MK, Gill D, Rannikmäe K, Traylor M, Anderson CD, Lee JM, Kamatani Y, Hopewell JC, Worrall BB, Bernhagen J, et al. Genetically determined levels of circulating cytokines and risk of stroke. *Circulation*. 2019;139:256–268. doi: 10.1161/CIRCULATIONAHA.118.035905
10. Lin J, Kakkar V, Lu X. Impact of MCP-1 in atherosclerosis. *Curr Pharm Des*. 2014;20:4580–4588. doi: 10.2174/1381612820666140522115801
11. Nelken NA, Coughlin SR, Gordon D, Wilcox JN. Monocyte chemoattractant protein-1 in human atheromatous plaques. *J Clin Invest*. 1991;88:1121–1127. doi: 10.1172/JCI115411
12. Lutgens E, Faber B, Schapira K, Evelo CT, van Haaften R, Heeneman S, Cleutjens KB, Bijnens AP, Beckers L, Porter JG, et al. Gene profiling in atherosclerosis reveals a key role for small inducible cytokines: validation using a novel monocyte chemoattractant protein monoclonal antibody. *Circulation*. 2005;111:3443–3452. doi: 10.1161/CIRCULATIONAHA.104.510073
13. Liehn EA, Piccinini AM, Koenen RR, Soehnlein O, Adage T, Fatu R, Curaj A, Popescu A, Zernecke A, Kungl AJ, et al. A new monocyte chemotactic protein-1/chemokine CC motif ligand-2 competitor limiting neointima formation and myocardial ischemia/reperfusion injury in mice. *J Am Coll Cardiol*. 2010;56:1847–1857. doi: 10.1016/j.jacc.2010.04.066
14. Bot I, Ortiz Zacarías NV, de Witte WE, de Vries H, van Santbrink PJ, van der Velden D, Kröner MJ, van der Berg DJ, Stamos D, de Lange EC, et al. A novel CCR2 antagonist inhibits atherogenesis in apoE deficient mice by achieving high receptor occupancy. *Sci Rep*. 2017;7:52. doi: 10.1038/s41598-017-00104-z
15. Gu L, Okada Y, Clinton SK, Gerard C, Sukhova GK, Libby P, Rollins BJ. Absence of monocyte chemoattractant protein-1 reduces atherosclerosis in low density lipoprotein receptor-deficient mice. *Mol Cell*. 1998;2:275–281.
16. Boring L, Gosling J, Cleary M, Charo IF. Decreased lesion formation in CCR2-/- mice reveals a role for chemokines in the initiation of atherosclerosis. *Nature*. 1998;394:894–897. doi: 10.1038/29788
17. Combadière C, Potteaux S, Rodero M, Simon T, Pezard A, Esposito B, Merval R, Proudfoot A, Tedgui A, Mallat Z. Combined inhibition of CCL2, CX3CR1, and CCR5 abrogates Ly6C(hi) and Ly6C(lo) monocytosis and almost abolishes atherosclerosis in hypercholesterolemic mice. *Circulation*. 2008;117:1649–1657. doi: 10.1161/CIRCULATIONAHA.107.745091
18. de Lemos JA, Morrow DA, Sabatine MS, Murphy SA, Gibson CM, Antman EM, McCabe CH, Cannon CP, Braunwald E. Association between plasma levels of monocyte chemoattractant protein-1 and long-term clinical outcomes in patients with acute coronary syndromes. *Circulation*. 2003;107:690–695. doi: 10.1161/01.cir.0000049742.68848.99
19. de Lemos JA, Morrow DA, Blazing MA, Jarolim P, Wiviott SD, Sabatine MS, Califf RM, Braunwald E. Serial measurement of monocyte chemoattractant protein-1 after acute coronary syndromes: results from the A to Z trial. *J Am Coll Cardiol*. 2007;50:2117–2124. doi: 10.1016/j.jacc.2007.06.057
20. Hoogeveen RC, Morrison A, Boerwinkle E, Miles JS, Rhodes CE, Sharrett AR, Ballantyne CM. Plasma MCP-1 level and risk for peripheral arterial disease and incident coronary heart disease: Atherosclerosis Risk in Communities study. *Atherosclerosis*. 2005;183:301–307. doi: 10.1016/j.atherosclerosis.2005.03.007
21. Deo R, Khera A, McGuire DK, Murphy SA, Meo Neto Jde P, Morrow DA, de Lemos JA. Association among plasma levels of monocyte chemoattractant protein-1, traditional cardiovascular risk factors, and subclinical atherosclerosis. *J Am Coll Cardiol*. 2004;44:1812–1818. doi: 10.1016/j.jacc.2004.07.047
22. van Wijk DF, van Leuven SI, Sandhu MS, Tanck MW, Hutten BA, Wareham NJ, Kastelein JJ, Stroes ES, Khaw KT, Boekholdt SM. Chemokine ligand 2 genetic variants, serum monocyte chemoattractant protein-1 levels, and the risk of coronary artery disease. *Arterioscler Thromb Vasc Biol*. 2010;30:1460–1466. doi: 10.1161/ATVBAHA.110.205526
23. Shoamanesh A, Preis SR, Beiser AS, Kase CS, Wolf PA, Vasan RS, Benjamin EJ, Seshadri S, Romero JR. Circulating biomarkers and incident ischemic stroke in the framingham offspring Study. *Neurology*. 2016;87:1206–1211. doi: 10.1212/WNL.0000000000003115
24. Herder C, Baumert J, Thorand B, Martin S, Löwel H, Kolb H, Koenig W. Chemokines and incident coronary heart disease: results from the MONICA/KORA augsburg case-cohort study, 1984-2002. *Arterioscler Thromb Vasc Biol*. 2006;26:2147–2152. doi: 10.1161/01.ATV.0000235691.84430.86
25. Schiopu A, Bengtsson E, Goncalves I, Nilsson J, Fredrikson GN, Bjorkbacka H. Associations between macrophage colony-stimulating factor and monocyte chemotactic protein 1 in plasma and first-time coronary events: a nested case-control study. *J Am Heart Assoc*. 2016;5:e002851.
26. Wells GA, Shea B, O'Connell D, Peterson J, Welch V, Losos M. The newcastle-ottawa scale (NOS) for assessing the quality of nonrandomized studies in meta-analyses [Online]. Available at: http://www.ohri.ca/programs/clinical_epidemiology/oxford.asp. Accessed September 13, 2019.
27. Williams B, Mancia G, Spiering W, Agabiti Rosei E, Azizi M, Burnier M, Clement DL, Coca A, de Simone G, Dominiczak A, et al; ESC Scientific Document Group. 2018 ESC/ESH guidelines for the management of arterial hypertension. *Eur Heart J*. 2018;39:3021–3104. doi: 10.1093/eurheartj/ehy339
28. American Diabetes Association. 2. Classification and diagnosis of diabetes: standards of medical care in diabetes-2019. *Diabetes Care*. 2019;42:S13–S28.
29. Jellinger PS, Handelsman Y, Rosenblit PD, Bloomgarden ZT, Fonseca VA, Garber AJ, Grunberger G, Guerin CK, Bell DSH, Mechanick JI, et al. American Association of clinical endocrinologists And American College of endocrinology guidelines for management of dyslipidemia and prevention of cardiovascular disease. *Endocr Pract*. 2017;23:1–87. doi: 10.4158/EP171764.APPGL
30. Stevens PE, Levin A, Kidney disease: improving global outcomes chronic kidney disease guideline development work Group M. Evaluation and management of chronic kidney disease: synopsis of the kidney disease: improving global outcomes 2012 clinical practice guideline. *Ann Intern Med*. 2013;158:825–830
31. Ridker PM, Libby P, MacFadyen JG, Thuren T, Ballantyne C, Fonseca F, Koenig W, Shimokawa H, Everett BM, Glynn RJ. Modulation of the interleukin-6 signalling pathway and incidence rates of atherosclerotic events and all-cause mortality: analyses from the Canakinumab Anti-Inflammatory Thrombosis Outcomes Study (CANTOS). *Eur Heart J*. 2018;39:3499–3507. doi: 10.1093/eurheartj/ehy310
32. Erqou S, Kaptoge S, Perry PL, Di Angelantonio E, Thompson A, White IR, Marcovina SM, Collins R, Thompson SG, Danesh J; Emerging Risk Factors Collaboration. Lipoprotein(a) concentration and the risk of coronary heart disease, stroke, and nonvascular mortality. *JAMA*. 2009;302:412–423. doi: 10.1001/jama.2009.1063
33. Willeit P, Ridker PM, Nestel PJ, Simes J, Tonkin AM, Pedersen TR, Schwartz GG, Olsson AG, Colhoun HM, Kronenberg F, et al. Baseline and on-statin treatment lipoprotein(a) levels for prediction of cardiovascular events: individual patient-data meta-analysis of statin outcome trials. *Lancet*. 2018;392:1311–1320. doi: 10.1016/S0140-6736(18)31652-0
34. Nordestgaard BG, Langsted A. Lipoprotein (a) as a cause of cardiovascular disease: insights from epidemiology, genetics, and biology. *J Lipid Res*. 2016;57:1953–1975. doi: 10.1194/jlr.R071233
35. Khera AV, Kathiresan S. Genetics of coronary artery disease: discovery, biology and clinical translation. *Nat Rev Genet*. 2017;18:331–344. doi: 10.1038/nrg.2016.160
36. Ridker PM, MacFadyen JG, Everett BM, Libby P, Thuren T, Glynn RJ; CANTOS Trial Group. Relationship of C-reactive protein reduction to cardiovascular event reduction following treatment with canakinumab: a secondary analysis from the CANTOS randomised controlled trial. *Lancet*. 2018;391:319–328. doi: 10.1016/S0140-6736(17)32814-3
37. Winter C, Silvestre-Roig C, Ortega-Gomez A, Lemnitzer P, Poelman H, Schumski A, Winter J, Drechsler M, de Jong R, Immler R, et al. Chrono-pharmacological targeting of the CCL2-CCR2 axis ameliorates atherosclerosis. *Cell Metab*. 2018;28:175.e5–182.e5. doi: 10.1016/j.cmet.2018.05.002
38. Okamoto M, Fuchigami M, Suzuki T, Watanabe N. A novel C-C chemokine receptor 2 antagonist prevents progression of albuminuria and atherosclerosis in mouse models. *Biol Pharm Bull*. 2012;35:2069–2074. doi: 10.1248/bpb.b12-00528
39. Yamashita T, Kawashima S, Ozaki M, Namiki M, Inoue N, Hirata K, Yokoyama M. Propagermanium reduces atherosclerosis in apolipoprotein E knockout mice via inhibition of macrophage infiltration. *Arterioscler Thromb Vasc Biol*. 2002;22:969–974. doi: 10.1161/01.atv.0000019051.88366.9c
40. Aiello RJ, Bourassa PA, Lindsey S, Weng W, Natoli E, Rollins BJ, Milos PM. Monocyte chemoattractant protein-1 accelerates atherosclerosis in apolipoprotein E-deficient mice. *Arterioscler Thromb Vasc Biol*. 1999;19:1518–1525. doi: 10.1161/01.atv.19.6.1518
41. Gilbert J, Lekstrom-Himes J, Donaldson D, Lee Y, Hu M, Xu J, Wyant T, Davidson M; MLN1202 Study Group. Effect of CC chemokine receptor 2 CCR2 blockade on serum C-reactive protein in individuals at atherosclerotic risk and with a single nucleotide polymorphism of the monocyte chemoattractant protein-1 promoter region. *Am J Cardiol*. 2011;107:906–911. doi: 10.1016/j.amjcard.2010.11.005

MANUSCRIPT III: Interleukin-6 signaling effects on ischemic stroke and other cardiovascular outcomes: a Mendelian Randomization study

Georgakis MK, Malik R, Gill D, Franceschini N, Sudlow CLM, INVENT Consortium, CHARGE Inflammation Working Group, Dichgans M. Interleukin-6 signaling effects on ischemic stroke and other cardiovascular outcomes: a Mendelian Randomization study. [Preprint in ***MedRxiv***, Accepted in ***Circ Gen Prec Med***]

Author contributions: MKG conceptualized the study. MKG, RM, and MD designed the study. MKG performed the statistical analyses. All authors interpreted the results. MKG and MD drafted the manuscript. All authors critically revised the manuscript for intellectual content. All authors approved the submitted version and are accountable for the integrity of the work.

Interleukin-6 signaling effects on ischemic stroke and other cardiovascular outcomes: a Mendelian Randomization study

Running title: *Georgakis et al.; IL-6 signaling and cardiovascular disease*

Marios K. Georgakis, MD, MSc[1, 2], Rainer Malik, PhD[1], Dipender Gill, MD[3], Nora Franceschini, MD, MPH[4], Cathie L. M. Sudlow, DPhil[3, 5], INVENT Consortium, CHARGE Inflammation Working Group, Martin Dichgans, MD[1, 6, 7]

[1]Institute for Stroke and Dementia Research (ISD), University Hospital, Ludwig-Maximilians-University LMU, Munich, Germany; [2]Graduate School for Systemic Neurosciences (GSN), Ludwig-Maximilians-University LMU, Munich, Germany; [3]Department of Epidemiology and Biostatistics, School of Public Health, Imperial College London, London, United Kingdom; [4]Department of Epidemiology, UNC Gillings Global School of Public Health, Chapel Hill, NC, USA; [5]Institute for Genetics and Molecular Medicine, University of Edinburgh, UK; [6]Munich Cluster for Systems Neurology (SyNergy), Munich, Germany; [7]German Centre for Neurodegenerative Diseases (DZNE), Munich, Germany

Address for correspondence:
Martin Dichgans, MD
Institute for Stroke and Dementia Research,
University Hospital of Ludwig-Maximilians-University (LMU)
Feodor-Lynen-Str. 17
81377 Munich, Germany
Phone: +49-89-4400-46018
Fax: +49-89-4400-46040
e-mail: martin.dichgans@med.uni-muenchen.de

Word count: 12,617 (title page, abstract, text, references, tables, figure legends)

ABSTRACT

Background: Studies in humans and experimental models highlight a role of interleukin-6 (IL-6) in cardiovascular disease. Indirect evidence suggests that inhibition of IL-6 signaling could lower risk of coronary artery disease. However, whether such an approach would be effective for ischemic stroke and other cardiovascular outcomes remains unknown.

Methods: In a genome-wide association study (GWAS) of 204,402 European individuals, we identified genetic proxies for downregulated IL-6 signaling as genetic variants in the IL-6 receptor (*IL6R*) locus that were associated with lower C-reactive protein (CRP) levels, a downstream effector of IL-6 signaling. We then applied two-sample Mendelian randomization (MR) to explore associations with ischemic stroke and its major subtypes (large artery stroke, cardioembolic stroke, small vessel stroke) in the MEGASTROKE dataset (34,217 cases and 404,630 controls), with coronary artery disease in the CARDIoGRAMplusC4D dataset (60,801 cases and 123,504 control), and with other cardiovascular outcomes in the UK Biobank (up to 321,406 individuals) and in phenotype-specific GWAS datasets. All effect estimates were scaled to the CRP-decreasing effects of tocilizumab, a monoclonal antibody targeting IL-6R.

Results: We identified 7 genetic variants as proxies for downregulated IL-6 signaling, which showed effects on upstream regulators (IL-6 and soluble IL-6R levels) and downstream effectors (CRP and fibrinogen levels) of the pathway that were consistent with pharmacological blockade of IL-6R. In MR, proxies for downregulated IL-6 signaling were associated with lower risk of ischemic stroke (Odds Ratio [OR]: 0.89, 95%CI: 0.82-0.97) and coronary artery disease (OR: 0.84, 95%CI: 0.77-0.90). Focusing on ischemic stroke subtypes, we found significant associations with risk of large artery (OR: 0.76, 95%CI: 0.62-0.93) and small vessel stroke (OR: 0.71, 95%CI: 0.59-0.86), but not cardioembolic stroke (OR: 0.95, 95%CI: 0.74-1.22). Proxies for IL-6 signaling inhibition were further associated with a lower risk of myocardial infarction, aortic aneurysm, atrial fibrillation and carotid plaque.

Conclusions: We provide evidence for a causal effect of IL-6 signaling on ischemic stroke, particularly large artery and small vessel stroke, and a range of other cardiovascular outcomes. IL-6R blockade might represent a valid therapeutic target for lowering cardiovascular risk and should thus be investigated in clinical trials.

Key Words: Interleukin-6; inflammation; cytokines; Mendelian randomization; genetics, human; atherosclerosis; stroke; coronary artery disease; cardiovascular disease.

CLINICAL PERSPECTIVE

What is new?

- We identified genetic proxies for downregulated IL-6 signaling that had effects on upstream and downstream regulators of the IL-6 signaling pathway consistent with those of pharmacological IL-6R blockade
- Genetically downregulated IL-6 signaling was associated with a lower risk of ischemic stroke, and in particular large artery and small vessel stroke
- Similar associations were obtained for a broad range of other cardiovascular outcomes

What are the clinical implications?

- Inhibition of IL-6 signaling is a promising therapeutic target for lowering risk of stroke and other cardiovascular outcomes and should be further investigated in clinical trials

INTRODUCTION

Stroke is the leading cause of adult disability and the second most common cause of mortality worldwide[1, 2] with an increasing burden on global health.[3, 4] Inflammation is involved in the pathogenesis of ischemic stroke, as has specifically been demonstrated for large artery atherosclerotic stroke.[5, 6] Cytokines regulate inflammatory responses[5] and could thus serve as targets for cardiovascular disease prevention.[7] In the recent Canakinumab Anti-Inflammatory Thrombosis Outcomes Study (CANTOS), treatment with an interleukin-1β (IL-1β) antagonist reduced cardiovascular event rates in patients with a history of myocardial infarction.[8] However, whether interfering with other cytokines would likewise offer benefit remains largely unknown. Also, there are few data on stroke and other cardiovascular outcomes beyond coronary artery disease.[9-11]

Interleukin-6 (IL-6), a key regulator of the inflammatory cascade, acts by binding to either its membrane-bound or soluble receptor (IL-6R) and induces proinflammatory downstream effects including increases in the levels of C-reactive protein (CRP).[12, 13] IL-6 has been implicated in the pathogenesis of multiple inflammatory diseases and inhibitors of IL-6R are used for the treatment of rheumatoid arthritis,[14] inflammatory bowel disease,[15] and other autoimmune disorders.[16] Downregulation of IL-6 signaling has further been proposed as a potential strategy for lowering cardiovascular risk.[11, 13] IL-6 levels have consistently been associated with risk of coronary artery disease in cohort studies.[17, 18] Mendelian randomization (MR) studies further showed that a variant in the gene encoding IL-6R with effects resembling pharmacological IL-6R inhibition is associated with a lower risk of coronary artery disease.[19, 20] Finally, secondary analyses from CANTOS demonstrated that the magnitude of the therapeutic benefit of IL-1β targeting was associated with the reduction of circulating IL-6 levels[11, 21] and that even after IL-1β inhibition, the residual cardiovascular risk was proportional to the post-treatment IL-6 levels.[22] These results provide indirect clinical evidence that interfering with IL-6 signaling

might lower cardiovascular risk and suggest that an approach directly targeting IL-6 signaling could offer additional benefit for cardiovascular prevention beyond IL-1β inhibition.

The effects of IL-6 signaling on risk of ischemic stroke remain largely unknown. While population-based cohort studies have found that circulating IL-6 levels are associated with a higher risk of ischemic stroke,[23, 24] these associations preclude conclusions about causal relationships because of possible confounding and reverse causation bias.[25] Also, there are no data on etiological stroke subtypes and other cardiovascular outcomes beyond coronary artery disease. Developing meaningful strategies for the prevention of ischemic stroke and cardiovascular disease in general would require defining these relationships.[26]

By using genetic variants as proxies for a trait of interest, MR overcomes key limitations of observational studies such as confounding and reverse causation and allows for investigation of causal effects on outcomes.[27, 28] MR further allows for prediction of the effects of pharmacological interventions by using variants located close to genes encoding candidate drug targets.[29, 30] Hence, MR has become a powerful strategy to prioritize interventions for exploration in clinical trials.[28]

Here, leveraging data from large genome-wide association studies (GWASs)[31-33] and applying MR analyses, we aimed to: (i) identify genetic proxies for downregulated IL-6 signaling on the basis of their effects on CRP levels, a well-established IL-6 signaling downstream effector,[13, 20, 34] (ii) validate their utility by comparing the consistency of their effects on upstream regulators and downstream effectors of the IL-6 signaling pathway with the effects of pharmacological IL-6R inhibition, as derived from clinical trials, (iii) explore associations of genetic predisposition to downregulated IL-6 signaling with the risk of ischemic stroke and coronary artery disease, (iv) examine associations with major etiological subtypes of ischemic stroke (large artery, cardioembolic, and small vessel stroke), and (v) examine associations with a broad range of other cardiovascular phenotypes. To derive clinically meaningful effect sizes that would be

comparable to those derived from potential future clinical trials, we weighted our instruments based on the CRP-decreasing effects of tocilizumab, a monoclonal antibody targeting IL-6R.

METHODS

Selection of genetic proxies for IL-6 signaling and validation of the instruments

The data sources for this study are described in **Table 1**. To identify instruments for genetic predisposition to downregulated IL-6 signaling, we selected variants within or near the *IL6R* gene, which encodes the receptor of IL-6. Specifically, we selected single-nucleotide polymorphisms (SNPs) in the *IL6R* gene or a region of 300 kB upstream or downstream from the *IL6R* gene (GRCh37/hg19 coordinates: chr1:154,077,669-154,741,926; **Supplementary Figure 1**) that were associated with circulating CRP levels. We selected and weighted genetic instruments for genetic predisposition to IL-6 signaling on the basis of their associations with CRP levels, because elevated CRP levels are a well-described downstream effect of IL-6 signaling (**Figure 1**).[13, 20, 34] Genetic association estimates with circulating CRP levels were obtained from a GWAS of 204,402 individuals of European ancestry drawn from the Cohorts for Heart and Aging Research in Genomic Epidemiology (CHARGE) Inflammation Working Group.[31] We selected variants that were associated with circulating CRP levels at genome-wide significance ($p<5x10^{-8}$) and clumped these variants to a linkage disequilibrium (LD) threshold of $r^2 < 0.1$ according to the European reference panel of the 1000 Genomes project.[35] We estimated the variance in CRP levels explained by each of the SNPs by calculating the R^2,[36] and the strength of the instruments by calculating the F-statistic.[37]

In sensitivity analyses, we restricted our selection of instruments to SNPs within the *IL6R* gene (GRCh37/hg19 coordinates: chr1:154,377,669-154,441,926), to avoid potential pleiotropic effects through genes neighboring *IL6R* and increase confidence in the effects of the instruments through IL-6 signaling. As the instruments used in the current setting were not identified based

on established biological effects, but solely on the basis of their statistical associations with CRP levels, in an additional sensitivity analysis, we restricted our genetic instrument to a single SNP (rs2228145) within the *IL6R* gene with well-established biological effects leading to a downregulation of the IL-6 signaling.[20, 34, 38, 39]

To disentangle the effects of IL-6 signaling from the respective effects of CRP, we selected SNPs associated with CRP levels at genome-wide significance ($p<5x10^{-8}$) throughout the genome and clumped them to $r^2 < 0.1$. We then performed MR analyses using all these SNPs as instruments, and performed 10,000 permutations for each outcome using 7 randomly selected SNPs (the same number as those used as instruments for IL-6 signaling). We further performed MR analyses using SNPs at the *CRP* locus as instruments (within a region of 300 kB upstream or downstream to the *CRP* gene; GRCh37/hg19 coordinates: chr1: 159,382,079- 159,984,379).

To validate the instruments, we explored their associations with circulating levels of IL-6 and soluble IL-6R, which have previously been reported to increase as a result of both pharmacological inhibition and genetic downregulation of IL-6 signaling.[20] We further explored association with fibrinogen levels, which is a downstream effector of IL-6 signaling and decreases after its blockade.[20] The effects of genetic variants on IL-6 levels were obtained from a GWAS of 8,293 healthy individuals of Finnish ancestry.[40] For soluble IL-6R levels, we used the summary statistics from the INTERVAL study exploring the human plasma proteome,[41] as made publicly available through the PhenoScanner database.[42] For fibrinogen levels, we used GWAS data from the CHARGE Inflammation Working Group on 120,246 European individuals.[43]

Outcomes

The primary outcomes for this study were ischemic stroke and coronary artery disease. Genetic association estimates for ischemic stroke and coronary artery disease were derived from the MEGASTROKE[32] and CARDIoGRAMplusC4D[44] consortia, respectively. Specifically, for

ischemic stroke we used the European sub-dataset of MEGASTROKE (34,217 cases and 404,630 controls) to avoid population stratification with the CRP GWAS dataset, which also included solely individuals of European ancestry.[32] The CARDIoGRAMplusC4D refers to a GWAS of 60,801 cases with coronary artery disease and 123,504 controls, primarily (77%) of European ancestry.[44] Definitions for major ischemic stroke subtypes in MEGASTROKE followed the Trial of Org 10172 in Acute Stroke Treatment (TOAST) criteria with the following samples for analysis: large artery stroke (4,373 cases), cardioembolic stroke (7,193 cases), and small vessel stroke (5,386 cases; 404,630 controls for all subtypes).[45] We further extended our analyses to other cardiovascular outcomes including myocardial infarction, aortic aneurysm, carotid artery plaque, peripheral artery disease, heart failure, atrial fibrillation, venous thromboembolism, deep vein thrombosis, and pulmonary embolism. The data sources and the sample sizes for these studies are presented in **Table 1**. For aortic aneurysm, heart failure, peripheral artery disease, deep vein thrombosis, and pulmonary embolism we used data from the UK Biobank, as described in **Supplementary Methods**.

Mendelian Randomization analyses

After extracting the association estimates between the variants and the outcomes and harmonizing the direction of estimates by effect alleles, we computed MR estimates for each instrument with the Wald estimator and standard errors with the Delta method.[46] We then pooled individual MR estimates using fixed-effects inverse-variance weighted (IVW) meta-analyses.[47] To provide clinically relevant results, all effect estimates were scaled to the CRP-decreasing effect of tocilizumab (8 mg/kg), between 4 and 24 weeks after administration (a decrease of CRP levels by 67%), as determined by a meta-analysis of 4 clinical trials.[20] For the main IVW analyses, we performed power calculations and estimated the minimum and maximum effects that we had 80% statistical power to detect.[48]

The IVW method was our primary MR analysis approach. Although the selection of instruments on a specific gene reduces the possibility of invalid variants,[49] the derived estimates might still be biased in case of directional pleiotropy. Hence, we further applied sensitivity MR analyses that are more robust to the inclusion of pleiotropic variants: the weighted median estimator, the contamination mixture method, and the MR Pleiotropy Residual Sum and Outlier (MR-PRESSO). The weighted median estimator provides consistent estimates as long as at least half of the variants used in the MR analysis are valid.[50] The contamination mixture method constructs a likelihood function of the individual estimates and under the assumption that the estimates of the valid instruments would follow a distribution centered around the causal effect and any invalid instruments would follow a distribution around zero, it calculates MR estimates that would maximize this likelihood.[51] The contamination method assumes that only some of the genetic variants used are valid instruments and it has been found to perform better than other methods under the presence of invalid instruments.[52] Finally, we applied MR-PRESSO, which regresses the SNP-outcome estimates against the SNP-exposure estimates to test, using residual errors, whether there are outlier SNPs. Outliers are detected by sequentially removing all genetic variants from the analyses and comparing the residual sum of squares as a global heterogeneity measure (p-value for detecting outliers <0.05).[53] MR-PRESSO then removes the identified outliers and provides outlier-corrected MR estimates.[53] MR-PRESSO, is outlier-robust, but still relies on the assumption that at least half of the variants are valid instruments.[53]

For the primary analyses (associations between downregulated IL-6 signaling and risk of ischemic stroke or coronary artery disease), we set a statistical significance threshold at a two-sided *p*-value of < 0.05. For ischemic stroke subtypes and for other cardiovascular outcomes, we corrected for multiple comparisons with the Bonferroni method. Thus, the statistical significance thresholds were set at $p<0.05/3=0.017$ for the 3 ischemic stroke subtypes, and at $p<0.05/9=0.0055$ for the 9 cardiovascular outcomes. Associations not reaching these thresholds, but showing *p*-values <0.05

were considered suggestive. All analyses were performed in R (v3.5.0; The R Foundation for Statistical Computing).

RESULTS

Identification and validation of genetic variants as proxies of downregulated IL-6 signaling

Using our pre-defined selection criteria, we identified 7 SNPs to serve as instruments for downregulated IL-6 signaling (**Table 2)**. Three of these instruments were situated within the *IL6R* gene (**Supplementary Figure 1**). The F-statistics of the 7 SNPs ranged from 81 to 764 indicating a low probability of weak instrument bias.[37] Power calculations indicated that these instruments provide adequate statistical power (>80%) to detect ORs at the magnitude of 0.90 or lower for ischemic stroke and coronary artery disease regarding the effect of genetically downregulated IL-6 signaling (scaled to the CRP-decreasing effect of tocilizumab) (**Table S1**). We were further sufficiently powered (>80%) to detect ORs at the magnitude of 0.80 or lower for ischemic stroke subtypes.

To validate the 7 instruments, we explored associations of genetically downregulated IL-6 signaling with circulating IL-6, soluble IL-6R, and fibrinogen levels. In accordance with randomized clinical trials testing the effects of tocilizumab versus placebo (8 mg/kg),[20] genetically downregulated IL-6 signaling was associated with higher circulating IL-6 and soluble IL-6R levels and with lower circulating concentration of fibrinogen with the strongest effects seen for soluble IL-6R levels (**Figure 2**).

Genetically downregulated IL-6 signaling, ischemic stroke and coronary artery disease

We next explored associations between genetically downregulated IL-6 signaling (scaled to the CRP-decreasing effect of tocilizumab) with the risk of ischemic stroke and coronary artery disease (**Figure 3**). In the primary IVW analysis, downregulated IL-6 signaling was associated with a lower risk of both ischemic stroke (OR: 0.89, 95%CI: 0.82-0.97, $p=3x10^{-3}$) and coronary artery disease (OR: 0.84, 95%CI: 0.77-0.90, $p=7x10^{-6}$). The alternative MR approaches (weighted median, contamination mixture, MR-PRESSO) all showed consistent association estimates (**Figure S2**).

In sensitivity analyses restricted to the 3 instruments within the *IL6R* gene, we likewise found genetically downregulated IL-6 signaling to be associated with a lower risk of ischemic stroke and coronary artery disease (**Figure 3** and **Figure S2**). Further restricting the analysis to a single SNP (rs2228145) with a well-described effect proxying the effects of pharmacological IL-6 signaling inhibition,[20] we found presence of the allele linked to downregulated IL-6 signaling to be associated with lower risk of both ischemic stroke (OR: 0.88, 95%CI: 0.79-0.99, $p=0.033$) and coronary artery disease (OR: 0.75, 95%CI: 0.67-0.85, $p=2x10^{-7}$).

To disentangle the effect of downregulated IL-6 signaling from the effect of CRP, we next performed MR analyses to explore associations between SNPs associated with CRP, and risk of ischemic stroke and coronary artery disease. These analyses showed no associations between genetically determined CRP levels and risk of either ischemic stroke or coronary artery disease independent of whether we used all variants reaching genome-wide significance ($p<5x10^{-8}$) for association with CRP (187 SNPs), or whether we restricted the analyses to significant SNPs at the *CRP* locus (24 SNPs) (**Figure S3**). We further performed 10,000 permutations of MR analyses randomly selecting 7 out of the 187 SNPs associated with CRP. The effects of the 7 SNPs selected as instruments for downregulated IL-6 signaling on ischemic stroke and coronary artery disease were located at the 4th and 1st lowest percentiles of the respective distributions,

corresponding to *p*-values of 0.04 and 0.01, respectively (**Figure 3C** and **Figure S4**), thus indicating that the effects of IL-6 signaling are independent of the effects of CRP itself.

Genetically downregulated IL-6 signaling and ischemic stroke subtypes

Focusing on etiological stroke subtypes (**Figure 4**), we found genetic downregulation of IL-6 signaling to be associated with a lower risk of large artery stroke (OR: 0.76, 95%CI: 0.62-0.93, $p=8x10^{-3}$) and small vessel stroke (OR: 0.71, 95%CI: 0.59-0.86, $p=3x10^{-4}$), but not cardioembolic stroke (OR: 0.95, 95%CI: 0.74-1.22, $p=0.667$). The results were stable in all MR sensitivity analyses, including when restricting the analyses to the instruments within the *IL6R* gene (**Figure S5**). We further found no associations between genetically determined CRP levels, as determined by SNPs throughout the genome or SNPs at the *CRP* locus, and any of the ischemic stroke subtypes (**Figure S6**). Similarly, in permutations of analyses including 7 randomly allocated SNPs throughout the genome, the effects of the SNPs proxying the downregulated IL-6 signaling on large artery and small vessel stroke, were at the 3rd and 0.1th percentiles (corresponding to *p*-values of 0.03 and 0.001), respectively, thus supporting that the observed effects were again independent of CRP (**Figure S7**).

Genetically downregulated IL-6 signaling and other cardiovascular outcomes

In a last step, we expanded the analyses to other cardiovascular outcomes (**Figure 5**). Genetic predisposition to downregulated IL-6 signaling was associated with lower risks of myocardial infarction (OR: 0.88, 95%CI: 0.81-0.96, $p=3x10^{-3}$) and aortic aneurysm (OR: 0.51, 95%CI: 0.37-0.68, $p=1x10^{-5}$). We further found suggestive associations ($p<0.05$) with atrial fibrillation (OR: 0.83, 95%CI: 0.71-0.96, $p=0.013$) and carotid plaque (OR: 0.87, 95%CI: 0.77-0.99, $p=0.041$). In contrast, we found no significant associations with peripheral artery disease (OR: 0.91, 95%CI:

0.74-1.11, *p*=0.349), heart failure (OR: 0.90, 95%CI: 0.79-1.04, *p*=0.156), venous thromboembolism (OR: 0.98, 95%CI: 0.81-1.15, *p*=0.809), deep vein thrombosis (OR: 1.15, 95%CI: 0.94-1.40, *p*=0.183), and pulmonary embolism (OR: 0.92, 95%CI: 0.78-1.10, *p*=0.373).

DISCUSSION

Leveraging large-scale genetic data from multiple sources we identified variants serving as proxies for a genetic predisposition to downregulated IL-6 signaling and validated them using clinical trial data on pharmacological IL-6R inhibition. The identified proxies showed significant associations with a lower risk of both ischemic stroke and coronary artery disease. Among ischemic stroke subtypes, genetic predisposition to downregulated IL-6 signaling was associated with lower risks of large artery and small vessel stroke, but not cardioembolic stroke. Proxies for IL-6 signaling inhibition further showed significant associations with myocardial infarction and aortic aneurysm, and suggestive associations with atrial fibrillation and carotid plaque.

The MR association between genetically downregulated IL-6 signaling and lower risk of large artery stroke extends previous clinical,[17, 18, 21] genetic,[19, 20] and experimental[54, 55] data demonstrating a key role of IL-6 signaling in atherosclerosis. By binding to IL-6R, IL-6 promotes downstream effects that include induction of macrophage recruitment[56] and arterial smooth muscle cell proliferation,[55, 57] and have been linked with plaque initiation,[58] plaque destabilization,[54] microvascular flow dysfunction,[59] and adverse outcomes in the setting of acute ischemia.[60] Moreover, pharmacological inhibition of IL-6R has been shown to attenuate atherosclerotic lesions in an experimental model of atherosclerosis.[61] Our finding of an effect of genetic predisposition to downregulated IL-6 signaling on multiple atherosclerotic phenotypes (large artery stroke, coronary artery disease, myocardial infarction, aortic aneurysm, atrial

fibrillation, carotid plaque) provides further support that IL-6 signaling is critically implicated in atherogenesis and atheroprogression and might represent a valid therapeutic target.

Notably, we found genetically downregulated IL-6 signaling to be further associated with small vessel stroke. There is only limited evidence regarding a role of inflammation in general and of IL-6 signaling in particular in cerebral small vessel disease.[62] In a small prospective study of 123 patients with manifestations of cerebral small vessel disease, IL-6 circulating levels were associated with a higher risk of incident lacunes, a marker of small vessel disease on brain magnetic resonance imaging.[63] However, cross-sectional analyses from larger population-based studies showed inconsistent findings for lacunes, silent brain infarcts and other manifestations of small vessel disease.[64-69] While the specific mechanisms underlying our MR results remain unknown, our findings suggest that inhibition of IL-6 signaling aside from being a candidate treatment for atherosclerosis might also lower the risk of small vessel stroke.

Our results strongly support the candidacy of IL-6 signaling as a target for vascular prevention over and beyond previous data. The CANTOS trial targeted IL-1β rather than IL-6R and thus provided only indirect evidence for a benefit of interfering with IL-6 signaling.[11, 21] Interestingly, the study further showed that part of the residual vascular risk after IL-1β inhibition could be explained by IL-6 levels, thus providing evidence that direct IL-6 signaling inhibition might represent a more effective strategy.[22] Also, CANTOS was based on a population of individuals with coronary artery disease and explored a combined vascular endpoint rather than offering information on individual cardiovascular outcomes. With respect to stroke, there was a 7% reduction in incident stroke events in the IL-1β arm, which however did not reach statistical significance, possibly because of insufficient power.[8] Our MR results provide evidence for directionally consistent effects of IL-6 signaling in multiple cardiovascular outcomes. Thus, our findings offer a solid basis for future clinical trials exploring the benefit of pharmacological IL-6R inhibition for the range of phenotypes examined here.

Interestingly, we found a particularly strong effect of genetically downregulated IL-6 signaling on aortic aneurysm. A role of IL-6 signaling in the pathogenesis of aortic aneurysm has been previously demonstrated by genetic studies.[38, 70, 71] IL-6 signaling might contribute to the formation of aortic aneurysms through mechanisms aside from atherosclerosis, thus explaining the large effect. For instance, IL-6 signaling is a key pathway in the pathogenesis of large vessel vasculitides,[72] which are strongly associated with the formation of aortic aneurysms.[16, 73, 74]

Our analysis provides no evidence for an association of genetically downregulated IL-6 signaling with cardioembolic stroke. In conjunction with the lack of significant MR associations with thrombotic phenotypes (venous thromboembolism, deep vein thrombosis, pulmonary embolism), our results do not support a role of IL-6 signaling in promoting coagulation and thrombosis. Yet, in accord with previous observational studies,[75-77] we found IL-6 signaling to show a suggestive association with atrial fibrillation, the primary cause of cardioembolism and a common complication of coronary artery disease.[78, 79] Given the relatively small magnitude of this association, any effect of IL-6 signaling on risk of cardioembolic stroke through atrial fibrillation would be expected to be small.

Our study has several strengths. Utilizing the most recent genetic data on CRP levels, ischemic stroke, and other cardiovascular phenotypes, we were sufficiently powered to show significant associations between genetically downregulated IL-6 signaling and multiple outcomes of interest. Using CRP levels, as a proxy for downstream IL-6 signaling enabled us to scale the derived association estimates to the respective effects of tocilizumab, as recorded in previous clinical trials, thus providing clinically meaningful estimates that might be comparable to those obtained from future trials. We further validated the effects of the selected proxies on upstream regulators (IL-6 and soluble IL-6R) and downstream effectors (fibrinogen) of IL-6 signaling, which were consistent with the effects observed with pharmacological inhibition of IL-6R. Finally, we could disentangle the effect of IL-6 signaling from the direct effect of CRP by

determining the effects of CRP levels on risk of the examined outcomes and performing permutations for the effects of randomly selected CRP-decreasing variants.

Our study also has limitations. First, to proxy IL-6 signaling we used CRP levels, which are a downstream effect of the classical membrane-bound IL-6R-mediated signaling in hepatocytes.[80] However, IL-6 also acts on other tissues not expressing the membrane-bound IL-6R, by binding to its soluble form, which is known as trans-signaling.[80] Thus, our results may be interpreted as an effect of downstream regulation of classical IL-6 signaling but not IL-6 trans-signaling. Second, by design, our MR study assessed the effects of lifetime downregulated IL-6 signaling, which might differ from a shorter pharmacological inhibition. Third, there might be unknown pleiotropic effects of the genetic proxies used as instruments in the current study that might bias the associations. Of note, however, the results were remarkably consistent in sensitivity MR methods that are more robust to the inclusion of pleiotropic variants. Finally, our results were mainly based on individuals of European origin, and might thus not apply to other ethnic groups.

In conclusion, this study provides evidence for a causal effect of IL-6 signaling on ischemic stroke, particularly large artery and small vessel stroke, as well as a range of cardiovascular phenotypes. IL-6R blockade might represent a valid therapeutic target for lowering cardiovascular risk and should thus be further investigated in clinical trials.

Acknowledgements: We thank the following consortia for making data publicly available: MEGASTROKE Consortium, CARDIoGRAMplusC4D Consortium, AFGen Consortium, the YFS/FINRISK studies, and the INTERVAL study. This research has been conducted using the UK Biobank Resource (UK Biobank application 2532, "UK Biobank stroke study: developing an in-depth understanding of the determinants of stroke and its subtypes").

Funding: M. Georgakis was funded by scholarships from the Onassis Foundation and the German Academic Exchange Service (DAAD). D. Gill is funded by the Wellcome Trust. This project has received funding from the European Union's Horizon 2020 research and innovation programme (No 666881), SVDs@target (to M. Dichgans) and No 667375, CoSTREAM (to M. Dichgans); the DFG as part of the Munich Cluster for Systems Neurology (EXC 2145 SyNergy – ID 390857198) and the CRC 1123 (B3) (to M. Dichgans); the Corona Foundation (to M. Dichgans); the Fondation Leducq (Transatlantic Network of Excellence on Pathogenesis of Small Vessel Disease of the Brain, to M. Dichgans); the e:Med program (e:AtheroSysMed, to M. Dichgans) and the FP7/2007-2103 European Union project CVgenes@target (grant agreement number Health-F2-2013-601456, to M. Dichgans).

Disclosures: No conflicts of interest to disclose.

Affiliations: From Institute for Stroke and Dementia Research (ISD), University Hospital of Ludwig-Maximilians-University (LMU), Munich, Germany (M.G., R.M., M.D.); Graduate School for Systemic Neurosciences (GSN), Ludwig-Maximilians-University LMU, Munich, Germany (M.G.); Department of Biostatistics and Epidemiology, School of Public Health, Imperial College London, London, UK (D.G., C.S.); Department of Epidemiology, UNC Gillings Global School of Public Health, Chapel Hill, NC, USA (N.F.); Institute for Genetics and Molecular Medicine, University of Edinburgh, UK (C.S.); Munich Cluster for Systems Neurology (SyNergy), Munich, Germany (M.D.); German Centre for Neurodegenerative Diseases (DZNE), Munich, Germany (M.D.).

REFERENCES

1. DALYs GBD and Collaborators H. Global, regional, and national disability-adjusted life-years (DALYs) for 315 diseases and injuries and healthy life expectancy (HALE), 1990-2015: a systematic analysis for the Global Burden of Disease Study 2015. *Lancet*. 2016;388:1603-1658.
2. Mortality GBD and Causes of Death C. Global, regional, and national life expectancy, all-cause mortality, and cause-specific mortality for 249 causes of death, 1980-2015: a systematic analysis for the Global Burden of Disease Study 2015. *Lancet*. 2016;388:1459-1544.
3. Collaborators GBDLRoS, Feigin VL, Nguyen G, Cercy K, Johnson CO, Alam T, Parmar PG, Abajobir AA, Abate KH, Abd-Allah F, Abejie AN, Abyu GY, Ademi Z, Agarwal G, Ahmed MB, Akinyemi RO, Al-Raddadi R, Aminde LN, Amlie-Lefond C, Ansari H, Asayesh H, Asgedom SW, Atey TM, Ayele HT, Banach M, Banerjee A, Barac A, Barker-Collo SL, Barnighausen T, Barregard L, Basu S, Bedi N, Behzadifar M, Bejot Y, Bennett DA, Bensenor IM, Berhe DF, Boneya DJ, Brainin M, Campos-Nonato IR, Caso V, Castaneda-Orjuela CA, Rivas JC, Catala-Lopez F, Christensen H, Criqui MH, Damasceno A, Dandona L, Dandona R, Davletov K, de Courten B, deVeber G, Dokova K, Edessa D, Endres M, Faraon EJA, Farvid MS, Fischer F, Foreman K, Forouzanfar MH, Gall SL, Gebrehiwot TT, Geleijnse JM, Gillum RF, Giroud M, Goulart AC, Gupta R, Gupta R, Hachinski V, Hamadeh RR, Hankey GJ, Hareri HA, Havmoeller R, Hay SI, Hegazy MI, Hibstu DT, James SL, Jeemon P, John D, Jonas JB, Jozwiak J, Kalani R, Kandel A, Kasaeian A, Kengne AP, Khader YS, Khan AR, Khang YH, Khubchandani J, Kim D, Kim YJ, Kivimaki M, Kokubo Y, Kolte D, Kopec JA, Kosen S, Kravchenko M, Krishnamurthi R, Kumar GA, Lafranconi A, Lavados PM, Legesse Y, Li Y, Liang X, Lo WD, Lorkowski S, Lotufo PA, Loy CT, Mackay MT, Abd El Razek HM, Mahdavi M, Majeed A, Malekzadeh R, Malta DC, Mamun AA, Mantovani LG, Martins SCO, Mate KK, Mazidi M, Mehata S, Meier T, Melaku YA, Mendoza W, Mensah GA, Meretoja A, Mezgebe HB, Miazgowski T, Miller TR, Ibrahim NM, Mohammed S, Mokdad AH, Moosazadeh M, Moran AE, Musa KI, Negoi RI, Nguyen M, Nguyen QL, Nguyen TH, Tran TT, Nguyen TT, Anggraini Ningrum DN, Norrving B, Noubiap JJ, O'Donnell MJ, Olagunju AT, Onuma OK, Owolabi MO, Parsaeian M, Patton GC, Piradov M, Pletcher MA, Pourmalek F, Prakash V, Qorbani M, Rahman M, Rahman MA, Rai RK, Ranta A, Rawaf D, Rawaf S, Renzaho AM, Robinson SR, Sahathevan R, Sahebkar A, Salomon JA, Santalucia P, Santos IS, Sartorius B, Schutte AE, Sepanlou SG, Shafieesabet A, Shaikh MA, Shamsizadeh M, Sheth KN, Sisay M, Shin MJ, Shiue I, Silva DAS, Sobngwi E, Soljak M, Sorensen RJD, Sposato LA, Stranges S, Suliankatchi RA, Tabares-Seisdedos R, Tanne D, Nguyen CT, Thakur JS, Thrift AG, Tirschwell DL, Topor-Madry R, Tran BX, Nguyen LT, Truelsen T, Tsilimparis N, Tyrovolas S, Ukwaja KN, Uthman OA, Varakin Y, Vasankari T, Venketasubramanian N, Vlassov VV, Wang W, Werdecker A, Wolfe CDA, Xu G, Yano Y, Yonemoto N, Yu C, Zaidi Z, El Sayed Zaki M, Zhou M, Ziaeian B, Zipkin B, Vos T, Naghavi M, Murray CJL and Roth GA. Global, Regional, and Country-Specific Lifetime Risks of Stroke, 1990 and 2016. *N Engl J Med*. 2018;379:2429-2437.
4. Feigin VL, Norrving B and Mensah GA. Global Burden of Stroke. *Circ Res*. 2017;120:439-448.
5. Esenwa CC and Elkind MS. Inflammatory risk factors, biomarkers and associated therapy in ischaemic stroke. *Nat Rev Neurol*. 2016;12:594-604.
6. Libby P, Ridker PM, Hansson GK and Leducq Transatlantic Network on Atherothrombosis. Inflammation in atherosclerosis: from pathophysiology to practice. *J Am Coll Cardiol*. 2009;54:2129-38.
7. Ridker PM. Targeting inflammatory pathways for the treatment of cardiovascular disease. *Eur Heart J*. 2014;35:540-3.
8. Ridker PM, Everett BM, Thuren T, MacFadyen JG, Chang WH, Ballantyne C, Fonseca F, Nicolau J, Koenig W, Anker SD, Kastelein JJP, Cornel JH, Pais P, Pella D, Genest J, Cifkova R, Lorenzatti A, Forster T, Kobalava Z, Vida-Simiti L, Flather M, Shimokawa H, Ogawa H, Dellborg M, Rossi PRF, Troquay RPT, Libby P, Glynn RJ and Group CT. Antiinflammatory Therapy with Canakinumab for Atherosclerotic Disease. *N Engl J Med*. 2017;377:1119-1131.
9. Georgakis MK, Gill D, Rannikmae K, Traylor M, Anderson CD, Lee JM, Kamatani Y, Hopewell JC, Worrall BB, Bernhagen J, Sudlow CLM, Malik R and Dichgans M. Genetically Determined Levels of Circulating Cytokines and Risk of Stroke. *Circulation*. 2019;139:256-268.
10. Georgakis MK, Malik R, Bjorkbacka H, Pana TA, Demissie S, Ayers C, Elhadad MA, Fornage M, Beiser AS, Benjamin EJ, Boekholdt MS, Engstrom G, Herder C, Hoogeveen RC, Koenig W, Melander O, Orho-Melander M, Schiopu A, Soderholm M, Wareham N, Ballantyne CM, Peters A, Seshadri S, Myint PK, Nilsson J, de Lemos JA and Dichgans M. Circulating Monocyte Chemoattractant Protein-1 and Risk of Stroke: A Meta-Analysis of Population-Based Studies Involving 17,180 Individuals. *Circ Res*. 2019.
11. Ridker PM. Anticytokine Agents: Targeting Interleukin Signaling Pathways for the Treatment of Atherothrombosis. *Circ Res*. 2019;124:437-450.
12. Scheller J, Chalaris A, Schmidt-Arras D and Rose-John S. The pro- and anti-inflammatory properties of the cytokine interleukin-6. *Biochim Biophys Acta*. 2011;1813:878-88.
13. Ridker PM. From C-Reactive Protein to Interleukin-6 to Interleukin-1: Moving Upstream To Identify Novel Targets for Atheroprotection. *Circ Res*. 2016;118:145-56.

14. Scott LJ. Tocilizumab: A Review in Rheumatoid Arthritis. *Drugs*. 2017;77:1865-1879.
15. Danese S, Vermeire S, Hellstern P, Panaccione R, Rogler G, Fraser G, Kohn A, Desreumaux P, Leong RW, Comer GM, Cataldi F, Banerjee A, Maguire MK, Li C, Rath N, Beebe J and Schreiber S. Randomised trial and open-label extension study of an anti-interleukin-6 antibody in Crohn's disease (ANDANTE I and II). *Gut*. 2019;68:40-48.
16. Stone JH, Tuckwell K, Dimonaco S, Klearman M, Aringer M, Blockmans D, Brouwer E, Cid MC, Dasgupta B, Rech J, Salvarani C, Schett G, Schulze-Koops H, Spiera R, Unizony SH and Collinson N. Trial of Tocilizumab in Giant-Cell Arteritis. *N Engl J Med*. 2017;377:317-328.
17. Kaptoge S, Seshasai SR, Gao P, Freitag DF, Butterworth AS, Borglykke A, Di Angelantonio E, Gudnason V, Rumley A, Lowe GD, Jorgensen T and Danesh J. Inflammatory cytokines and risk of coronary heart disease: new prospective study and updated meta-analysis. *Eur Heart J*. 2014;35:578-89.
18. Ridker PM, Rifai N, Stampfer MJ and Hennekens CH. Plasma concentration of interleukin-6 and the risk of future myocardial infarction among apparently healthy men. *Circulation*. 2000;101:1767-72.
19. Collaboration IRGCERF, Sarwar N, Butterworth AS, Freitag DF, Gregson J, Willeit P, Gorman DN, Gao P, Saleheen D, Rendon A, Nelson CP, Braund PS, Hall AS, Chasman DI, Tybjaerg-Hansen A, Chambers JC, Benjamin EJ, Franks PW, Clarke R, Wilde AA, Trip MD, Steri M, Witteman JC, Qi L, van der Schoot CE, de Faire U, Erdmann J, Stringham HM, Koenig W, Rader DJ, Melzer D, Reich D, Psaty BM, Kleber ME, Panagiotakos DB, Willeit J, Wennberg P, Woodward M, Adamovic S, Rimm EB, Meade TW, Gillum RF, Shaffer JA, Hofman A, Onat A, Sundstrom J, Wassertheil-Smoller S, Mellstrom D, Gallacher J, Cushman M, Tracy RP, Kauhanen J, Karlsson M, Salonen JT, Wilhelmsen L, Amouyel P, Cantin B, Best LG, Ben-Shlomo Y, Manson JE, Davey-Smith G, de Bakker PI, O'Donnell CJ, Wilson JF, Wilson AG, Assimes TL, Jansson JO, Ohlsson C, Tivesten A, Ljunggren O, Reilly MP, Hamsten A, Ingelsson E, Cambien F, Hung J, Thomas GN, Boehnke M, Schunkert H, Asselbergs FW, Kastelein JJ, Gudnason V, Salomaa V, Harris TB, Kooner JS, Allin KH, Nordestgaard BG, Hopewell JC, Goodall AH, Ridker PM, Holm H, Watkins H, Ouwehand WH, Samani NJ, Kaptoge S, Di Angelantonio E, Harari O and Danesh J. Interleukin-6 receptor pathways in coronary heart disease: a collaborative meta-analysis of 82 studies. *Lancet*. 2012;379:1205-13.
20. Interleukin-6 Receptor Mendelian Randomisation Analysis C, Swerdlow DI, Holmes MV, Kuchenbaecker KB, Engmann JE, Shah T, Sofat R, Guo Y, Chung C, Peasey A, Pfister R, Mooijaart SP, Ireland HA, Leusink M, Langenberg C, Li KW, Palmen J, Howard P, Cooper JA, Drenos F, Hardy J, Nalls MA, Li YR, Lowe G, Stewart M, Bielinski SJ, Peto J, Timpson NJ, Gallacher J, Dunlop M, Houlston R, Tomlinson I, Tzoulaki I, Luan J, Boer JM, Forouhi NG, Onland-Moret NC, van der Schouw YT, Schnabel RB, Hubacek JA, Kubinova R, Baceviciene M, Tamosiunas A, Pajak A, Topor-Madry R, Malyutina S, Baldassarre D, Sennblad B, Tremoli E, de Faire U, Ferrucci L, Bandenelli S, Tanaka T, Meschia JF, Singleton A, Navis G, Mateo Leach I, Bakker SJ, Gansevoort RT, Ford I, Epstein SE, Burnett MS, Devaney JM, Jukema JW, Westendorp RG, Jan de Borst G, van der Graaf Y, de Jong PA, Mailand-van der Zee AH, Klungel OH, de Boer A, Doevendans PA, Stephens JW, Eaton CB, Robinson JG, Manson JE, Fowkes FG, Frayling TM, Price JF, Whincup PH, Morris RW, Lawlor DA, Smith GD, Ben-Shlomo Y, Redline S, Lange LA, Kumari M, Wareham NJ, Verschuren WM, Benjamin EJ, Whittaker JC, Hamsten A, Dudbridge F, Delaney JA, Wong A, Kuh D, Hardy R, Castillo BA, Connolly JJ, van der Harst P, Brunner EJ, Marmot MG, Wassel CL, Humphries SE, Talmud PJ, Kivimaki M, Asselbergs FW, Voevoda M, Bobak M, Pikhart H, Wilson JG, Hakonarson H, Reiner AP, Keating BJ, Sattar N, Hingorani AD and Casas JP. The interleukin-6 receptor as a target for prevention of coronary heart disease: a mendelian randomisation analysis. *Lancet*. 2012;379:1214-24.
21. Ridker PM, Libby P, MacFadyen JG, Thuren T, Ballantyne C, Fonseca F, Koenig W, Shimokawa H, Everett BM and Glynn RJ. Modulation of the interleukin-6 signalling pathway and incidence rates of atherosclerotic events and all-cause mortality: analyses from the Canakinumab Anti-Inflammatory Thrombosis Outcomes Study (CANTOS). *Eur Heart J*. 2018;39:3499-3507.
22. Ridker PM, MacFadyen JG, Thuren T and Libby P. Residual inflammatory risk associated with interleukin-18 and interleukin-6 after successful interleukin-1beta inhibition with canakinumab: further rationale for the development of targeted anti-cytokine therapies for the treatment of atherothrombosis. *Eur Heart J*. 2019.
23. Cainzos-Achirica M, Enjuanes C, Greenland P, McEvoy JW, Cushman M, Dardari Z, Nasir K, Budoff MJ, Al-Mallah MH, Yeboah J, Miedema MD, Blumenthal RS, Comin-Colet J and Blaha MJ. The prognostic value of interleukin 6 in multiple chronic diseases and all-cause death: The Multi-Ethnic Study of Atherosclerosis (MESA). *Atherosclerosis*. 2018;278:217-225.
24. Jenny NS, Callas PW, Judd SE, McClure LA, Kissela B, Zakai NA and Cushman M. Inflammatory cytokines and ischemic stroke risk: The REGARDS cohort. *Neurology*. 2019;92:e2375-e2384.
25. Smith GD and Ebrahim S. 'Mendelian randomization': can genetic epidemiology contribute to understanding environmental determinants of disease? *Int J Epidemiol*. 2003;32:1-22.
26. Kelly PJ, Murphy S, Coveney S, Purroy F, Lemmens R, Tsivgoulis G and Price C. Anti-inflammatory approaches to ischaemic stroke prevention. *J Neurol Neurosurg Psychiatry*. 2018;89:211-218.
27. Holmes MV, Ala-Korpela M and Smith GD. Mendelian randomization in cardiometabolic disease: challenges in evaluating causality. *Nat Rev Cardiol*. 2017;14:577-590.
28. Roberts R. Mendelian Randomization Studies Promise to Shorten the Journey to FDA Approval. *JACC Basic Transl Sci*. 2018;3:690-703.

29. Walker VM, Davey Smith G, Davies NM and Martin RM. Mendelian randomization: a novel approach for the prediction of adverse drug events and drug repurposing opportunities. *Int J Epidemiol*. 2017;46:2078-2089.
30. Gill D, Georgakis MK, Koskeridis F, Jiang L, Feng Q, Wei WQ, Theodoratou E, Elliott P, Denny JC, Malik R, Evangelou E, Dehghan A, Dichgans M and Tzoulaki I. Use of Genetic Variants Related to Antihypertensive Drugs to Inform on Efficacy and Side Effects. *Circulation*. 2019;140:270-279.
31. Ligthart S, Vaez A, Vosa U, Stathopoulou MG, de Vries PS, Prins BP, Van der Most PJ, Tanaka T, Naderi E, Rose LM, Wu Y, Karlsson R, Barbalic M, Lin H, Pool R, Zhu G, Mace A, Sidore C, Trompet S, Mangino M, Sabater-Lleal M, Kemp JP, Abbasi A, Kacprowski T, Verweij N, Smith AV, Huang T, Marzi C, Feitosa MF, Lohman KK, Kleber ME, Milaneschi Y, Mueller C, Huq M, Vlachopoulou E, Lyytikainen LP, Oldmeadow C, Deelen J, Perola M, Zhao JH, Feenstra B, LifeLines Cohort S, Amini M, Group CIW, Lahti J, Schraut KE, Fornage M, Suktitipat B, Chen WM, Li X, Nutile T, Malerba G, Luan J, Bak T, Schork N, Del Greco MF, Thiering E, Mahajan A, Marioni RE, Mihailov E, Eriksson J, Ozel AB, Zhang W, Nethander M, Cheng YC, Aslibekyan S, Ang W, Gandin I, Yengo L, Portas L, Kooperberg C, Hofer E, Rajan KB, Schurmann C, den Hollander W, Ahluwalia TS, Zhao J, Draisma HHM, Ford I, Timpson N, Teumer A, Huang H, Wahl S, Liu Y, Huang J, Uh HW, Geller F, Joshi PK, Yanek LR, Trabetti E, Lehne B, Vozzi D, Verbanck M, Biino G, Saba Y, Meulenbelt I, O'Connell JR, Laakso M, Giulianini F, Magnusson PKE, Ballantyne CM, Hottenga JJ, Montgomery GW, Rivadineira F, Rueedi R, Steri M, Herzig KH, Stott DJ, Menni C, Franberg M, St Pourcain B, Felix SB, Pers TH, Bakker SJL, Kraft P, Peters A, Vaidya D, Delgado G, Smit JH, Grossmann V, Sinisalo J, Seppala I, Williams SR, Holliday EG, Moed M, Langenberg C, Raikkonen K, Ding J, Campbell H, Sale MM, Chen YI, James AL, Ruggiero D, Soranzo N, Hartman CA, Smith EN, Berenson GS, Fuchsberger C, Hernandez D, Tiesler CMT, Giedraitis V, Liewald D, Fischer K, Mellstrom D, Larsson A, Wang Y, Scott WR, Lorentzon M, Beilby J, Ryan KA, Pennell CE, Vuckovic D, Balkau B, Concas MP, Schmidt R, Mendes de Leon CF, Bottinger EP, Kloppenburg M, Paternoster L, Boehnke M, Musk AW, Willemsen G, Evans DM, Madden PAF, Kahonen M, Kutalik Z, Zoledziewska M, Karhunen V, Kritchevsky SB, Sattar N, Lachance G, Clarke R, Harris TB, Raitakari OT, Attia JR, van Heemst D, Kajantie E, Sorice R, Gambaro G, Scott RA, Hicks AA, Ferrucci L, Standl M, Lindgren CM, Starr JM, Karlsson M, Lind L, Li JZ, Chambers JC, Mori TA, de Geus E, Heath AC, Martin NG, Auvinen J, Buckley BM, de Craen AJM, Waldenberger M, Strauch K, Meitinger T, Scott RJ, McEvoy M, Beekman M, Bombieri C, Ridker PM, Mohlke KL, Pedersen NL, Morrison AC, Boomsma DI, Whitfield JB, Strachan DP, Hofman A, Vollenweider P, Cucca F, Jarvelin MR, Jukema JW, Spector TD, Hamsten A, Zeller T, Uitterlinden AG, Nauck M, Gudnason V, Qi L, Grallert H, Borecki IB, Rotter JI, Marz W, Wild PS, Lokki ML, Boyle M, Salomaa V, Melbye M, Eriksson JG, Wilson JF, Penninx B, Becker DM, Worrall BB, Gibson G, Krauss RM, Ciullo M, Zaza G, Wareham NJ, Oldehinkel AJ, Palmer LJ, Murray SS, Pramstaller PP, Bandinelli S, Heinrich J, Ingelsson E, Deary IJ, Magi R, Vandenput L, van der Harst P, Desch KC, Kooner JS, Ohlsson C, Hayward C, Lehtimaki T, Shuldiner AR, Arnett DK, Beilin LJ, Robino A, Froguel P, Pirastu M, Jess T, Koenig W, Loos RJF, Evans DA, Schmidt H, Smith GD, Slagboom PE, Eiriksdottir G, Morris AP, Psaty BM, Tracy RP, Nolte IM, Boerwinkle E, Visvikis-Siest S, Reiner AP, Gross M, Bis JC, Franke L, Franco OH, Benjamin EJ, Chasman DI, Dupuis J, Snieder H, Dehghan A and Alizadeh BZ. Genome Analyses of >200,000 Individuals Identify 58 Loci for Chronic Inflammation and Highlight Pathways that Link Inflammation and Complex Disorders. *Am J Hum Genet*. 2018;103:691-706.
32. Malik R, Chauhan G, Traylor M, Sargurupremraj M, Okada Y, Mishra A, Rutten-Jacobs L, Giese AK, van der Laan SW, Gretarsdottir S, Anderson CD, Chong M, Adams HHH, Ago T, Almgren P, Amouyel P, Ay H, Bartz TM, Benavente OR, Bevan S, Boncoraglio GB, Brown RD, Jr., Butterworth AS, Carrera C, Carty CL, Chasman DI, Chen WM, Cole JW, Correa A, Cotlarciuc I, Cruchaga C, Danesh J, de Bakker PIW, DeStefano AL, den Hoed M, Duan Q, Engelter ST, Falcone GJ, Gottesman RF, Grewal RP, Gudnason V, Gustafsson S, Haessler J, Harris TB, Hassan A, Havulinna AS, Heckbert SR, Holliday EG, Howard G, Hsu FC, Hyacinth HI, Ikram MA, Ingelsson E, Irvin MR, Jian X, Jimenez-Conde J, Johnson JA, Jukema JW, Kanai M, Keene KL, Kissela BM, Kleindorfer DO, Kooperberg C, Kubo M, Lange LA, Langefeld CD, Langenberg C, Launer LJ, Lee JM, Lemmens R, Leys D, Lewis CM, Lin WY, Lindgren AG, Lorentzen E, Magnusson PK, Maguire J, Manichaikul A, McArdle PF, Meschia JF, Mitchell BD, Mosley TH, Nalls MA, Ninomiya T, O'Donnell MJ, Psaty BM, Pulit SL, Rannikmae K, Reiner AP, Rexrode KM, Rice K, Rich SS, Ridker PM, Rost NS, Rothwell PM, Rotter JI, Rundek T, Sacco RL, Sakaue S, Sale MM, Salomaa V, Sapkota BR, Schmidt R, Schmidt CO, Schminke U, Sharma P, Slowik A, Sudlow CLM, Tanislav C, Tatlisumak T, Taylor KD, Thijs VNS, Thorleifsson G, Thorsteinsdottir U, Tiedt S, Trompet S, Tzourio C, van Duijn CM, Walters M, Wareham NJ, Wassertheil-Smoller S, Wilson JG, Wiggins KL, Yang Q, Yusuf S, Bis JC, Pastinen T, Ruusalepp A, Schadt EE, Koplev S, Bjorkegren JLM, Codoni V, Civelek M, Smith NL, Tregouet DA, Christophersen IE, Roselli C, Lubitz SA, Ellinor PT, Tai ES, Kooner JS, Kato N, He J, van der Harst P, Elliott P, Chambers JC, Takeuchi F, Johnson AD, Sanghera DK, Melander O, Jern C, Strbian D, Fernandez-Cadenas I, Longstreth WT, Jr., Rolfs A, Hata J, Woo D, Rosand J, Pare G, Hopewell JC, Saleheen D, Stefansson K, Worrall BB, Kittner SJ, Seshadri S, Fornage M, Markus HS, Howson JMM, Kamatani Y, Debette S, Dichgans M, Malik R, Chauhan G, Traylor M, Sargurupremraj M, Okada Y, Mishra A, Rutten-Jacobs L, Giese AK, van der Laan SW, Gretarsdottir S, Anderson CD, Chong M, Adams HHH, Ago T, Almgren P, Amouyel P, Ay H, Bartz TM, Benavente OR, Bevan S, Boncoraglio GB, Brown RD, Jr., Butterworth AS, Carrera C, Carty CL, Chasman DI, Chen WM, Cole JW, Correa A, Cotlarciuc I, Cruchaga C, Danesh J, de Bakker PIW, DeStefano AL, Hoed MD, Duan Q, Engelter ST, Falcone GJ, Gottesman RF, Grewal RP, Gudnason V, Gustafsson S, Haessler J, Harris TB,

Hassan A, Havulinna AS, Heckbert SR, Holliday EG, Howard G, Hsu FC, Hyacinth HI, Ikram MA, Ingelsson E, Irvin MR, Jian X, Jimenez-Conde J, Johnson JA, Jukema JW, Kanai M, Keene KL, Kissela BM, Kleindorfer DO, Kooperberg C, Kubo M, Lange LA, Langefeld CD, Langenberg C, Launer LJ, Lee JM, Lemmens R, Leys D, Lewis CM, Lin WY, Lindgren AG, Lorentzen E, Magnusson PK, Maguire J, Manichaikul A, McArdle PF, Meschia JF, Mitchell BD, Mosley TH, Nalls MA, Ninomiya T, O'Donnell MJ, Psaty BM, Pulit SL, Rannikmae K, Reiner AP, Rexrode KM, Rice K, Rich SS, Ridker PM, Rost NS, Rothwell PM, Rotter JI, Rundek T, Sacco RL, Sakaue S, Sale MM, Salomaa V, Sapkota BR, Schmidt R, Schmidt CO, Schminke U, Sharma P, Slowik A, Sudlow CLM, Tanislav C, Tatlisumak T, Taylor KD, Thijs VNS, Thorleifsson G, Thorsteinsdottir U, Tiedt S, Trompet S, Tzourio C, van Duijn CM, Walters M, Wareham NJ, Wassertheil-Smoller S, Wilson JG, Wiggins KL, Yang Q, Yusuf S, Amin N, Aparicio HS, Arnett DK, Attia J, Beiser AS, Berr C, Buring JE, Bustamante M, Caso V, Cheng YC, Choi SH, Chowhan A, Cullell N, Dartigues JF, Delavaran H, Delgado P, Dorr M, Engstrom G, Ford I, Gurpreet WS, Hamsten A, Heitsch L, Hozawa A, Ibanez L, Ilinca A, Ingelsson M, Iwasaki M, Jackson RD, Jood K, Jousilahti P, Kaffashian S, Kalra L, Kamouchi M, Kitazono T, Kjartansson O, Kloss M, Koudstaal PJ, Krupinski J, Labovitz DL, Laurie CC, Levi CR, Li L, Lind L, Lindgren CM, Lioutas V, Liu YM, Lopez OL, Makoto H, Martinez-Majander N, Matsuda K, Minegishi N, Montaner J, Morris AP, Muino E, Muller-Nurasyid M, Norrving B, Ogishima S, Parati EA, Peddareddygari LR, Pedersen NL, Pera J, Perola M, Pezzini A, Pileggi S, Rabionet R, Riba-Llena I, Ribases M, Romero JR, Roquer J, Rudd AG, Sarin AP, Sarju R, Sarnowski C, Sasaki M, Satizabal CL, Satoh M, Sattar N, Sawada N, Sibolt G, Sigurdsson A, Smith A, Sobue K, Soriano-Tarraga C, Stanne T, Stine OC, Stott DJ, Strauch K, Takai T, Tanaka H, Tanno K, Teumer A, Tomppo L, Torres-Aguila NP, Touze E, Tsugane S, Uitterlinden AG, Valdimarsson EM, van der Lee SJ, Volzke H, Wakai K, Weir D, Williams SR, Wolfe CDA, Wong Q, Xu H, Yamaji T, Sanghera DK, Melander O, Jern C, Strbian D, Fernandez-Cadenas I, Longstreth WT, Jr., Rolfs A, Hata J, Woo D, Rosand J, Pare G, Hopewell JC, Saleheen D, Stefansson K, Worrall BB, Kittner SJ, Seshadri S, Fornage M, Markus HS, Howson JMM, Kamatani Y, Debette S, Dichgans M, Consortium AF, Cohorts for H, Aging Research in Genomic Epidemiology C, International Genomics of Blood Pressure C, Consortium I, Starnet, BioBank Japan Cooperative Hospital G, Consortium C, Consortium E-C, Consortium EP-I, International Stroke Genetics C, Consortium M, Neurology Working Group of the CC, Network NSG, Study UKYLD, Consortium M and Consortium M. Multiancestry genome-wide association study of 520,000 subjects identifies 32 loci associated with stroke and stroke subtypes. *Nat Genet*. 2018;50:524-537.

33. Malik R, Rannikmae K, Traylor M, Georgakis MK, Sargurupremraj M, Markus HS, Hopewell JC, Debette S, Sudlow CLM, Dichgans M, consortium M and the International Stroke Genetics C. Genome-wide meta-analysis identifies 3 novel loci associated with stroke. *Ann Neurol*. 2018;84:934-939.

34. Parisinos CA, Serghiou S, Katsoulis M, George MJ, Patel RS, Hemingway H and Hingorani AD. Variation in Interleukin 6 Receptor Gene Associates With Risk of Crohn's Disease and Ulcerative Colitis. *Gastroenterology*. 2018;155:303-306 e2.

35. Genomes Project C, Auton A, Brooks LD, Durbin RM, Garrison EP, Kang HM, Korbel JO, Marchini JL, McCarthy S, McVean GA and Abecasis GR. A global reference for human genetic variation. *Nature*. 2015;526:68-74.

36. Shim H, Chasman DI, Smith JD, Mora S, Ridker PM, Nickerson DA, Krauss RM and Stephens M. A multivariate genome-wide association analysis of 10 LDL subfractions, and their response to statin treatment, in 1868 Caucasians. *PLoS One*. 2015;10:e0120758.

37. Palmer TM, Lawlor DA, Harbord RM, Sheehan NA, Tobias JH, Timpson NJ, Davey Smith G and Sterne JA. Using multiple genetic variants as instrumental variables for modifiable risk factors. *Stat Methods Med Res*. 2012;21:223-42.

38. Cai T, Zhang Y, Ho YL, Link N, Sun J, Huang J, Cai TA, Damrauer S, Ahuja Y, Honerlaw J, Huang J, Costa L, Schubert P, Hong C, Gagnon D, Sun YV, Gaziano JM, Wilson P, Cho K, Tsao P, O'Donnell CJ, Liao KP and Program VAMV. Association of Interleukin 6 Receptor Variant With Cardiovascular Disease Effects of Interleukin 6 Receptor Blocking Therapy: A Phenome-Wide Association Study. *JAMA Cardiol*. 2018;3:849-857.

39. Ferreira RC, Freitag DF, Cutler AJ, Howson JM, Rainbow DB, Smyth DJ, Kaptoge S, Clarke P, Boreham C, Coulson RM, Pekalski ML, Chen WM, Onengut-Gumuscu S, Rich SS, Butterworth AS, Malarstig A, Danesh J and Todd JA. Functional IL6R 358Ala allele impairs classical IL-6 receptor signaling and influences risk of diverse inflammatory diseases. *PLoS Genet*. 2013;9:e1003444.

40. Ahola-Olli AV, Wurtz P, Havulinna AS, Aalto K, Pitkanen N, Lehtimaki T, Kahonen M, Lyytikainen LP, Raitoharju E, Seppala I, Sarin AP, Ripatti S, Palotie A, Perola M, Viikari JS, Jalkanen S, Maksimow M, Salomaa V, Salmi M, Kettunen J and Raitakari OT. Genome-wide Association Study Identifies 27 Loci Influencing Concentrations of Circulating Cytokines and Growth Factors. *Am J Hum Genet*. 2017;100:40-50.

41. Sun BB, Maranville JC, Peters JE, Stacey D, Staley JR, Blackshaw J, Burgess S, Jiang T, Paige E, Surendran P, Oliver-Williams C, Kamat MA, Prins BP, Wilcox SK, Zimmerman ES, Chi A, Bansal N, Spain SL, Wood AM, Morrell NW, Bradley JR, Janjic N, Roberts DJ, Ouwehand WH, Todd JA, Soranzo N, Suhre K, Paul DS, Fox CS, Plenge RM, Danesh J, Runz H and Butterworth AS. Genomic atlas of the human plasma proteome. *Nature*. 2018;558:73-79.

42. Kamat MA, Blackshaw JA, Young R, Surendran P, Burgess S, Danesh J, Butterworth AS and Staley JR. PhenoScanner V2: an expanded tool for searching human genotype-phenotype associations. *Bioinformatics*. 2019.

43. de Vries PS, Chasman DI, Sabater-Lleal M, Chen MH, Huffman JE, Steri M, Tang W, Teumer A, Marioni RE, Grossmann V, Hottenga JJ, Trompet S, Muller-Nurasyid M, Zhao JH, Brody JA, Kleber ME, Guo X, Wang JJ, Auer PL, Attia JR, Yanek LR, Ahluwalia TS, Lahti J, Venturini C, Tanaka T, Bielak LF, Joshi PK, Rocanin-Arjo A, Kolcic I, Navarro P, Rose LM, Oldmeadow C, Riess H, Mazur J, Basu S, Goel A, Yang Q, Ghanbari M, Willemsen G, Rumley A, Fiorillo E, de Craen AJ, Grotevendt A, Scott R, Taylor KD, Delgado GE, Yao J, Kifley A, Kooperberg C, Qayyum R, Lopez LM, Berentzen TL, Raikkonen K, Mangino M, Bandinelli S, Peyser PA, Wild S, Tregouet DA, Wright AF, Marten J, Zemunik T, Morrison AC, Sennblad B, Tofler G, de Maat MP, de Geus EJ, Lowe GD, Zoledziewska M, Sattar N, Binder H, Volker U, Waldenberger M, Khaw KT, McKnight B, Huang J, Jenny NS, Holliday EG, Qi L, McEvoy MG, Becker DM, Starr JM, Sarin AP, Hysi PG, Hernandez DG, Jhun MA, Campbell H, Hamsten A, Rivadeneira F, McArdle WL, Slagboom PE, Zeller T, Koenig W, Psaty BM, Haritunians T, Liu J, Palotie A, Uitterlinden AG, Stott DJ, Hofman A, Franco OH, Polasek O, Rudan I, Morange PE, Wilson JF, Kardia SL, Ferrucci L, Spector TD, Eriksson JG, Hansen T, Deary IJ, Becker LC, Scott RJ, Mitchell P, Marz W, Wareham NJ, Peters A, Greinacher A, Wild PS, Jukema JW, Boomsma DI, Hayward C, Cucca F, Tracy R, Watkins H, Reiner AP, Folsom AR, Ridker PM, O'Donnell CJ, Smith NL, Strachan DP and Dehghan A. A meta-analysis of 120 246 individuals identifies 18 new loci for fibrinogen concentration. *Hum Mol Genet*. 2016;25:358-70.
44. Nikpay M, Goel A, Won HH, Hall LM, Willenborg C, Kanoni S, Saleheen D, Kyriakou T, Nelson CP, Hopewell JC, Webb TR, Zeng L, Dehghan A, Alver M, Armasu SM, Auro K, Bjonnes A, Chasman DI, Chen S, Ford I, Franceschini N, Gieger C, Grace C, Gustafsson S, Huang J, Hwang SJ, Kim YK, Kleber ME, Lau KW, Lu X, Lu Y, Lyytikainen LP, Mihailov E, Morrison AC, Pervjakova N, Qu L, Rose LM, Salfati E, Saxena R, Scholz M, Smith AV, Tikkanen E, Uitterlinden A, Yang X, Zhang W, Zhao W, de Andrade M, de Vries PS, van Zuydam NR, Anand SS, Bertram L, Beutner F, Dedoussis G, Frossard P, Gauguier D, Goodall AH, Gottesman O, Haber M, Han BG, Huang J, Jalilzadeh S, Kessler T, Konig IR, Lannfelt L, Lieb W, Lind L, Lindgren CM, Lokki ML, Magnusson PK, Mallick NH, Mehra N, Meitinger T, Memon FU, Morris AP, Nieminen MS, Pedersen NL, Peters A, Rallidis LS, Rasheed A, Samuel M, Shah SH, Sinisalo J, Stirrups KE, Trompet S, Wang L, Zaman KS, Ardissino D, Boerwinkle E, Borecki IB, Bottinger EP, Buring JE, Chambers JC, Collins R, Cupples LA, Danesh J, Demuth I, Elosua R, Epstein SE, Esko T, Feitosa MF, Franco OH, Franzosi MG, Granger CB, Gu D, Gudnason V, Hall AS, Hamsten A, Harris TB, Hazen SL, Hengstenberg C, Hofman A, Ingelsson E, Iribarren C, Jukema JW, Karhunen PJ, Kim BJ, Kooner JS, Kullo IJ, Lehtimaki T, Loos RJF, Melander O, Metspalu A, Marz W, Palmer CN, Perola M, Quertermous T, Rader DJ, Ridker PM, Ripatti S, Roberts R, Salomaa V, Sanghera DK, Schwartz SM, Seedorf U, Stewart AF, Stott DJ, Thiery J, Zalloua PA, O'Donnell CJ, Reilly MP, Assimes TL, Thompson JR, Erdmann J, Clarke R, Watkins H, Kathiresan S, McPherson R, Deloukas P, Schunkert H, Samani NJ and Farrall M. A comprehensive 1,000 Genomes-based genome-wide association meta-analysis of coronary artery disease. *Nat Genet*. 2015;47:1121-1130.
45. Adams HP, Jr., Bendixen BH, Kappelle LJ, Biller J, Love BB, Gordon DL and Marsh EE, 3rd. Classification of subtype of acute ischemic stroke. Definitions for use in a multicenter clinical trial. TOAST. Trial of Org 10172 in Acute Stroke Treatment. *Stroke*. 1993;24:35-41.
46. Lawlor DA, Harbord RM, Sterne JA, Timpson N and Davey Smith G. Mendelian randomization: using genes as instruments for making causal inferences in epidemiology. *Stat Med*. 2008;27:1133-63.
47. Burgess S, Butterworth A and Thompson SG. Mendelian randomization analysis with multiple genetic variants using summarized data. *Genet Epidemiol*. 2013;37:658-65.
48. Brion MJ, Shakhbazov K and Visscher PM. Calculating statistical power in Mendelian randomization studies. *Int J Epidemiol*. 2013;42:1497-501.
49. Swerdlow DI, Kuchenbaecker KB, Shah S, Sofat R, Holmes MV, White J, Mindell JS, Kivimaki M, Brunner EJ, Whittaker JC, Casas JP and Hingorani AD. Selecting instruments for Mendelian randomization in the wake of genome-wide association studies. *Int J Epidemiol*. 2016;45:1600-1616.
50. Hartwig FP, Davey Smith G and Bowden J. Robust inference in summary data Mendelian randomization via the zero modal pleiotropy assumption. *Int J Epidemiol*. 2017;46:1985-1998.
51. Burgess S, Foley CN, Allara E, Staley JR and Howson JM. A robust and efficient method for Mendelian randomization with hundreds of genetic variants: unravelling mechanisms linking HDL-cholesterol and coronary heart disease. *bioRxiv (Pre-print)*. 2019.
52. Slob EAW and Burgess S. A Comparison Of Robust Mendelian Randomization Methods Using Summary Data. *bioRxiv (Pre-print)*. 2019.
53. Verbanck M, Chen CY, Neale B and Do R. Detection of widespread horizontal pleiotropy in causal relationships inferred from Mendelian randomization between complex traits and diseases. *Nat Genet*. 2018;50:693-698.
54. Huber SA, Sakkinen P, Conze D, Hardin N and Tracy R. Interleukin-6 exacerbates early atherosclerosis in mice. *Arterioscler Thromb Vasc Biol*. 1999;19:2364-7.
55. Ikeda U, Ikeda M, Oohara T, Oguchi A, Kamitani T, Tsuruya Y and Kano S. Interleukin 6 stimulates growth of vascular smooth muscle cells in a PDGF-dependent manner. *Am J Physiol*. 1991;260:H1713-7.
56. Jenkins BJ, Grail D, Inglese M, Quilici C, Bozinovski S, Wong P and Ernst M. Imbalanced gp130-dependent signaling in macrophages alters macrophage colony-stimulating factor responsiveness via regulation of c-fms expression. *Mol Cell Biol*. 2004;24:1453-63.

57. Wang D, Liu Z, Li Q, Karpurapu M, Kundumani-Sridharan V, Cao H, Dronadula N, Rizvi F, Bajpai AK, Zhang C, Muller-Newen G, Harris KW and Rao GN. An essential role for gp130 in neointima formation following arterial injury. *Circ Res*. 2007;100:807-16.
58. Schieffer B, Selle T, Hilfiker A, Hilfiker-Kleiner D, Grote K, Tietge UJ, Trautwein C, Luchtefeld M, Schmittkamp C, Heeneman S, Daemen MJ and Drexler H. Impact of interleukin-6 on plaque development and morphology in experimental atherosclerosis. *Circulation*. 2004;110:3493-500.
59. Guo F, Dong M, Ren F, Zhang C, Li J, Tao Z, Yang J and Li G. Association between local interleukin-6 levels and slow flow/microvascular dysfunction. *J Thromb Thrombolysis*. 2014;37:475-82.
60. Lindmark E, Diderholm E, Wallentin L and Siegbahn A. Relationship between interleukin 6 and mortality in patients with unstable coronary artery disease: effects of an early invasive or noninvasive strategy. *JAMA*. 2001;286:2107-13.
61. Akita K, Isoda K, Sato-Okabayashi Y, Kadoguchi T, Kitamura K, Ohtomo F, Shimada K and Daida H. An Interleukin-6 Receptor Antibody Suppresses Atherosclerosis in Atherogenic Mice. *Front Cardiovasc Med*. 2017;4:84.
62. Low A, Mak E, Rowe JB, Markus HS and O'Brien JT. Inflammation and cerebral small vessel disease: A systematic review. *Ageing Res Rev*. 2019;53:100916.
63. Staszewski J, Piusinska-Macoch R, Brodacki B, Skrobowska E and Stepien A. IL-6, PF-4, sCD40 L, and homocysteine are associated with the radiological progression of cerebral small-vessel disease: a 2-year follow-up study. *Clin Interv Aging*. 2018;13:1135-1141.
64. Shoamanesh A, Preis SR, Beiser AS, Vasan RS, Benjamin EJ, Kase CS, Wolf PA, DeCarli C, Romero JR and Seshadri S. Inflammatory biomarkers, cerebral microbleeds, and small vessel disease: Framingham Heart Study. *Neurology*. 2015;84:825-32.
65. Satizabal CL, Zhu YC, Mazoyer B, Dufouil C and Tzourio C. Circulating IL-6 and CRP are associated with MRI findings in the elderly: the 3C-Dijon Study. *Neurology*. 2012;78:720-7.
66. Hoshi T, Kitagawa K, Yamagami H, Furukado S, Hougaku H and Hori M. Relations of serum high-sensitivity C-reactive protein and interleukin-6 levels with silent brain infarction. *Stroke*. 2005;36:768-72.
67. Yoshida M, Tomitori H, Machi Y, Katagiri D, Ueda S, Horiguchi K, Kobayashi E, Saeki N, Nishimura K, Ishii I, Kashiwagi K and Igarashi K. Acrolein, IL-6 and CRP as markers of silent brain infarction. *Atherosclerosis*. 2009;203:557-62.
68. Fornage M, Chiang YA, O'Meara ES, Psaty BM, Reiner AP, Siscovick DS, Tracy RP and Longstreth WT, Jr. Biomarkers of Inflammation and MRI-Defined Small Vessel Disease of the Brain: The Cardiovascular Health Study. *Stroke*. 2008;39:1952-9.
69. Baune BT, Ponath G, Rothermundt M, Roesler A and Berger K. Association between cytokines and cerebral MRI changes in the aging brain. *J Geriatr Psychiatry Neurol*. 2009;22:23-34.
70. Jones GT, Tromp G, Kuivaniemi H, Gretarsdottir S, Baas AF, Giusti B, Strauss E, Van't Hof FN, Webb TR, Erdman R, Ritchie MD, Elmore JR, Verma A, Pendergrass S, Kullo IJ, Ye Z, Peissig PL, Gottesman O, Verma SS, Malinowski J, Rasmussen-Torvik LJ, Borthwick KM, Smelser DT, Crosslin DR, de Andrade M, Ryer EJ, McCarty CA, Bottinger EP, Pacheco JA, Crawford DC, Carrell DS, Gerhard GS, Franklin DP, Carey DJ, Phillips VL, Williams MJ, Wei W, Blair R, Hill AA, Vasudevan TM, Lewis DR, Thomson IA, Krysa J, Hill GB, Roake J, Merriman TR, Oszkinis G, Galora S, Saracini C, Abbate R, Pulli R, Pratesi C, Saratzis A, Verissimo AR, Bumpstead S, Badger SA, Clough RE, Cockerill G, Hafez H, Scott DJ, Futers TS, Romaine SP, Bridge K, Griffin KJ, Bailey MA, Smith A, Thompson MM, van Bockxmeer FM, Matthiasson SE, Thorleifsson G, Thorsteinsdottir U, Blankensteijn JD, Teijink JA, Wijmenga C, de Graaf J, Kiemeney LA, Lindholt JS, Hughes A, Bradley DT, Stirrups K, Golledge J, Norman PE, Powell JT, Humphries SE, Hamby SE, Goodall AH, Nelson CP, Sakalihasan N, Courtois A, Ferrell RE, Eriksson P, Folkersen L, Franco-Cereceda A, Eicher JD, Johnson AD, Betsholtz C, Ruusalepp A, Franzen O, Schadt EE, Bjorkegren JL, Lipovich L, Drolet AM, Verhoeven EL, Zeebregts CJ, Geelkerken RH, van Sambeek MR, van Sterkenburg SM, de Vries JP, Stefansson K, Thompson JR, de Bakker PI, Deloukas P, Sayers RD, Harrison SC, van Rij AM, Samani NJ and Bown MJ. Meta-Analysis of Genome-Wide Association Studies for Abdominal Aortic Aneurysm Identifies Four New Disease-Specific Risk Loci. *Circ Res*. 2017;120:341-353.
71. Harrison SC, Smith AJ, Jones GT, Swerdlow DI, Rampuri R, Bown MJ, Aneurysm C, Folkersen L, Baas AF, de Borst GJ, Blankensteijn JD, Price JF, van der Graaf Y, McLachlan S, Agu O, Hofman A, Uitterlinden AG, Franco-Cereceda A, Ruigrok YM, van't Hof FN, Powell JT, van Rij AM, Casas JP, Eriksson P, Holmes MV, Asselbergs FW, Hingorani AD and Humphries SE. Interleukin-6 receptor pathways in abdominal aortic aneurysm. *Eur Heart J*. 2013;34:3707-16.
72. Terrades-Garcia N and Cid MC. Pathogenesis of giant-cell arteritis: how targeted therapies are influencing our understanding of the mechanisms involved. *Rheumatology (Oxford)*. 2018;57:ii51-ii62.
73. Evans JM, Bowles CA, Bjornsson J, Mullany CJ and Hunder GG. Thoracic aortic aneurysm and rupture in giant cell arteritis. A descriptive study of 41 cases. *Arthritis Rheum*. 1994;37:1539-47.
74. Villiger PM, Adler S, Kuchen S, Wermelinger F, Dan D, Fiege V, Butikofer L, Seitz M and Reichenbach S. Tocilizumab for induction and maintenance of remission in giant cell arteritis: a phase 2, randomised, double-blind, placebo-controlled trial. *Lancet*. 2016;387:1921-7.

75. Marcus GM, Whooley MA, Glidden DV, Pawlikowska L, Zaroff JG and Olgin JE. Interleukin-6 and atrial fibrillation in patients with coronary artery disease: data from the Heart and Soul Study. *Am Heart J*. 2008;155:303-9.
76. Amdur RL, Mukherjee M, Go A, Barrows IR, Ramezani A, Shoji J, Reilly MP, Gnanaraj J, Deo R, Roas S, Keane M, Master S, Teal V, Soliman EZ, Yang P, Feldman H, Kusek JW, Tracy CM, Raj DS and Investigators CS. Interleukin-6 Is a Risk Factor for Atrial Fibrillation in Chronic Kidney Disease: Findings from the CRIC Study. *PLoS One*. 2016;11:e0148189.
77. Li J, Solus J, Chen Q, Rho YH, Milne G, Stein CM and Darbar D. Role of inflammation and oxidative stress in atrial fibrillation. *Heart Rhythm*. 2010;7:438-44.
78. Ntalla I, Kanoni S, Zeng L, Giannakopoulou O, Danesh J, Watkins H, Samani NJ, Deloukas P, Schunkert H and Group UKBCCCW. Genetic Risk Score for Coronary Disease Identifies Predispositions to Cardiovascular and Noncardiovascular Diseases. *J Am Coll Cardiol*. 2019;73:2932-2942.
79. Schmitt J, Duray G, Gersh BJ and Hohnloser SH. Atrial fibrillation in acute myocardial infarction: a systematic review of the incidence, clinical features and prognostic implications. *Eur Heart J*. 2009;30:1038-45.
80. Lissilaa R, Buatois V, Magistrelli G, Williams AS, Jones GW, Herren S, Shang L, Malinge P, Guilhot F, Chatel L, Hatterer E, Jones SA, Kosco-Vilbois MH and Ferlin WG. Although IL-6 trans-signaling is sufficient to drive local immune responses, classical IL-6 signaling is obligate for the induction of T cell-mediated autoimmunity. *J Immunol*. 2010;185:5512-21.
81. Sudlow C, Gallacher J, Allen N, Beral V, Burton P, Danesh J, Downey P, Elliott P, Green J, Landray M, Liu B, Matthews P, Ong G, Pell J, Silman A, Young A, Sprosen T, Peakman T and Collins R. UK biobank: an open access resource for identifying the causes of a wide range of complex diseases of middle and old age. *PLoS Med*. 2015;12:e1001779.
82. Franceschini N, Giambartolomei C, de Vries PS, Finan C, Bis JC, Huntley RP, Lovering RC, Tajuddin SM, Winkler TW, Graff M, Kavousi M, Dale C, Smith AV, Hofer E, van Leeuwen EM, Nolte IM, Lu L, Scholz M, Sargurupremraj M, Pitkanen N, Franzen O, Joshi PK, Noordam R, Marioni RE, Hwang SJ, Musani SK, Schminke U, Palmas W, Isaacs A, Correa A, Zonderman AB, Hofman A, Teumer A, Cox AJ, Uitterlinden AG, Wong A, Smit AJ, Newman AB, Britton A, Ruusalepp A, Sennblad B, Hedblad B, Pasaniuc B, Penninx BW, Langefeld CD, Wassel CL, Tzourio C, Fava C, Baldassarre D, O'Leary DH, Teupser D, Kuh D, Tremoli E, Mannarino E, Grossi E, Boerwinkle E, Schadt EE, Ingelsson E, Veglia F, Rivadeneira F, Beutner F, Chauhan G, Heiss G, Snieder H, Campbell H, Volzke H, Markus HS, Deary IJ, Jukema JW, de Graaf J, Price J, Pott J, Hopewell JC, Liang J, Thiery J, Engmann J, Gertow K, Rice K, Taylor KD, Dhana K, Kiemeney L, Lind L, Raffield LM, Launer LJ, Holdt LM, Dorr M, Dichgans M, Traylor M, Sitzer M, Kumari M, Kivimaki M, Nalls MA, Melander O, Raitakari O, Franco OH, Rueda-Ochoa OL, Roussos P, Whincup PH, Amouyel P, Giral P, Anugu P, Wong Q, Malik R, Rauramaa R, Burkhardt R, Hardy R, Schmidt R, de Mutsert R, Morris RW, Strawbridge RJ, Wannamethee SG, Hagg S, Shah S, McLachlan S, Trompet S, Seshadri S, Kurl S, Heckbert SR, Ring S, Harris TB, Lehtimaki T, Galesloot TE, Shah T, de Faire U, Plagnol V, Rosamond WD, Post W, Zhu X, Zhang X, Guo X, Saba Y, Consortium M, Dehghan A, Seldenrijk A, Morrison AC, Hamsten A, Psaty BM, van Duijn CM, Lawlor DA, Mook-Kanamori DO, Bowden DW, Schmidt H, Wilson JF, Wilson JG, Rotter JI, Wardlaw JM, Deanfield J, Halcox J, Lyytikainen LP, Loeffler M, Evans MK, Debette S, Humphries SE, Volker U, Gudnason V, Hingorani AD, Bjorkegren JLM, Casas JP and O'Donnell CJ. GWAS and colocalization analyses implicate carotid intima-media thickness and carotid plaque loci in cardiovascular outcomes. *Nat Commun*. 2018;9:5141.
83. Christophersen IE, Rienstra M, Roselli C, Yin X, Geelhoed B, Barnard J, Lin H, Arking DE, Smith AV, Albert CM, Chaffin M, Tucker NR, Li M, Klarin D, Bihlmeyer NA, Low SK, Weeke PE, Muller-Nurasyid M, Smith JG, Brody JA, Niemeijer MN, Dorr M, Trompet S, Huffman J, Gustafsson S, Schurmann C, Kleber ME, Lyytikainen LP, Seppala I, Malik R, Horimoto A, Perez M, Sinisalo J, Aeschbacher S, Theriault S, Yao J, Radmanesh F, Weiss S, Teumer A, Choi SH, Weng LC, Clauss S, Deo R, Rader DJ, Shah SH, Sun A, Hopewell JC, Debette S, Chauhan G, Yang Q, Worrall BB, Pare G, Kamatani Y, Hagemeijer YP, Verweij N, Siland JE, Kubo M, Smith JD, Van Wagoner DR, Bis JC, Perz S, Psaty BM, Ridker PM, Magnani JW, Harris TB, Launer LJ, Shoemaker MB, Padmanabhan S, Haessler J, Bartz TM, Waldenberger M, Lichtner P, Arendt M, Krieger JE, Kahonen M, Risch L, Mansur AJ, Peters A, Smith BH, Lind L, Scott SA, Lu Y, Bottinger EB, Hernesniemi J, Lindgren CM, Wong JA, Huang J, Eskola M, Morris AP, Ford I, Reiner AP, Delgado G, Chen LY, Chen YI, Sandhu RK, Li M, Boerwinkle E, Eisele L, Lannfelt L, Rost N, Anderson CD, Taylor KD, Campbell A, Magnusson PK, Porteous D, Hocking LJ, Vlachopoulou E, Pedersen NL, Nikus K, Orho-Melander M, Hamsten A, Heeringa J, Denny JC, Kriebel J, Darbar D, Newton-Cheh C, Shaffer C, Macfarlane PW, Heilmann-Heimbach S, Almgren P, Huang PL, Sotoodehnia N, Soliman EZ, Uitterlinden AG, Hofman A, Franco OH, Volker U, Jockel KH, Sinner MF, Lin HJ, Guo X, ISGC MCot, Neurology Working Group of the CC, Dichgans M, Ingelsson E, Kooperberg C, Melander O, Loos RJF, Laurikka J, Conen D, Rosand J, van der Harst P, Lokki ML, Kathiresan S, Pereira A, Jukema JW, Hayward C, Rotter JI, Marz W, Lehtimaki T, Stricker BH, Chung MK, Felix SB, Gudnason V, Alonso A, Roden DM, Kaab S, Chasman DI, Heckbert SR, Benjamin EJ, Tanaka T, Lunetta KL, Lubitz SA, Ellinor PT and Consortium AF. Large-scale analyses of common and rare variants identify 12 new loci associated with atrial fibrillation. *Nat Genet*. 2017;49:946-952.

84. Germain M, Chasman DI, de Haan H, Tang W, Lindstrom S, Weng LC, de Andrade M, de Visser MC, Wiggins KL, Suchon P, Saut N, Smadja DM, Le Gal G, van Hylckama Vlieg A, Di Narzo A, Hao K, Nelson CP, Rocanin-Arjo A, Folkersen L, Monajemi R, Rose LM, Brody JA, Slagboom E, Aissi D, Gagnon F, Deleuze JF, Deloukas P, Tzourio C, Dartigues JF, Berr C, Taylor KD, Civelek M, Eriksson P, Cardiogenics C, Psaty BM, Houwing-Duitermaat J, Goodall AH, Cambien F, Kraft P, Amouyel P, Samani NJ, Basu S, Ridker PM, Rosendaal FR, Kabrhel C, Folsom AR, Heit J, Reitsma PH, Tregouet DA, Smith NL and Morange PE. Meta-analysis of 65,734 individuals identifies TSPAN15 and SLC44A2 as two susceptibility loci for venous thromboembolism. *Am J Hum Genet*. 2015;96:532-42.

Table 1. Data sources that were used in the analyses for the current study.

Phenotype	Source	N (Total or Cases/Controls	Imputation reference panel	Ancestry	Adjustments
CRP levels	CHARGE Inflammation Working Group[31]	204,402	HapMap	European	age, sex, population structure
IL-6 levels	YFS/FINRISK studies[40]	8,293	1000 Genomes Phase 1	Finnish	age, sex, BMI, population structure
sIL-6R levels	INTERVAL study[41]	3,301	1000 Genomes Phase 3	European	age, sex, duration between blood draw and processing, population structure
Fibrinogen levels	CHARGE Inflammation Working Group[43]	120,246	1000 Genomes Phase 1	European	age, sex, population structure
Ischemic stroke	MEGASTROKE Consortium[32]	34,217/406,630	1000 Genomes Phase 1	European	age, sex, population structure
Large artery stroke	MEGASTROKE Consortium[32]	4,373/406,111	1000 Genomes Phase 1	European	age, sex, population structure
Cardioembolic stroke	MEGASTROKE Consortium[32]	7,193/406,111	1000 Genomes Phase 1	European	age, sex, population structure
Small vessel stroke	MEGASTROKE Consortium[32]	5,386/406,111	1000 Genomes Phase 1	European	age, sex, population structure
Coronary artery disease	CARDIoGRAMplusC4D Consortium[44]	60,801/123,504	HapMap	European and Asian	age, sex, population structure
Myocardial infarction	CARDIoGRAMplusC4D Consortium[44]	43,676/123,504	HapMap	European and Asian	age, sex, population structure
Aortic aneurysm	UK Biobank[81]	1,817/314,325	HRC + UK10K	White British	age, sex, population structure, genotyping platform array
Carotid plaque	CHARGE Consortium[82]	21,540/26894	1000 Genomes Phase 1	European	age, sex, population structure
Peripheral artery disease	UK Biobank[81]	3,992/313,725	HRC + UK10K	White British	age, sex, population structure, genotyping platform array
Heart failure	UK Biobank[81]	8,970/312,436	HRC + UK10K	White British	age, sex, population structure, genotyping platform array
Atrial fibrillation	AFGen Consortium[83]	18,398/111,433	1000 Genomes Phase 1	European and Asian	age, sex, population structure
Venous thromboembolism	INVENT Consortium[84]	7,507/52,632	1000 Genomes Phase 1	European	age, sex, population structure
Deep vein thrombosis	UK Biobank[81]	4,135/302,337	HRC + UK10K	White British	age, sex, population structure, genotyping platform array
Pulmonary embolism	UK Biobank[81]	5,400/302,186	HRC + UK10K	White British	age, sex, population structure, genotyping platform array

Table 2. Single nucleotide polymorphisms (SNP) used in the current analyses for proxying the effects of IL-6 signaling. The betas, standard errors, and p-values refer to associations of these SNPs with CRP levels.

SNP	Effect allele	Non-effect allele	Chromosome	MAF	Position (GRCh37/hg19)	Beta †	Standard Error	P-value	R^2 ‡	F §
rs73026617	t	c	1	0.097	154,369,981	0.0474	0.0068	3.16E-12	3.94E-04	80.5
rs12083537*	a	g	1	0.193	154,381,103	0.0643	0.0053	7.14E-34	1.29E-03	263.6
rs4556348*	t	c	1	0.148	154,394,296	0.0541	0.0067	6.77E-16	7.38E-04	151.0
rs2228145*	a	c	1	0.360	154,426,970	0.0899	0.0042	1.21E-101	3.72E-03	764.1
rs11264224	a	c	1	0.193	154,568,086	0.0465	0.0057	3.41E-16	6.74E-04	137.8
rs12059682	t	c	1	0.196	154,579,585	-0.0441	0.0049	2.26E-19	6.13E-04	125.4
rs34693607	c	g	1	0.184	154,661,369	0.0368	0.0057	1.07E-10	4.07E-04	83.2

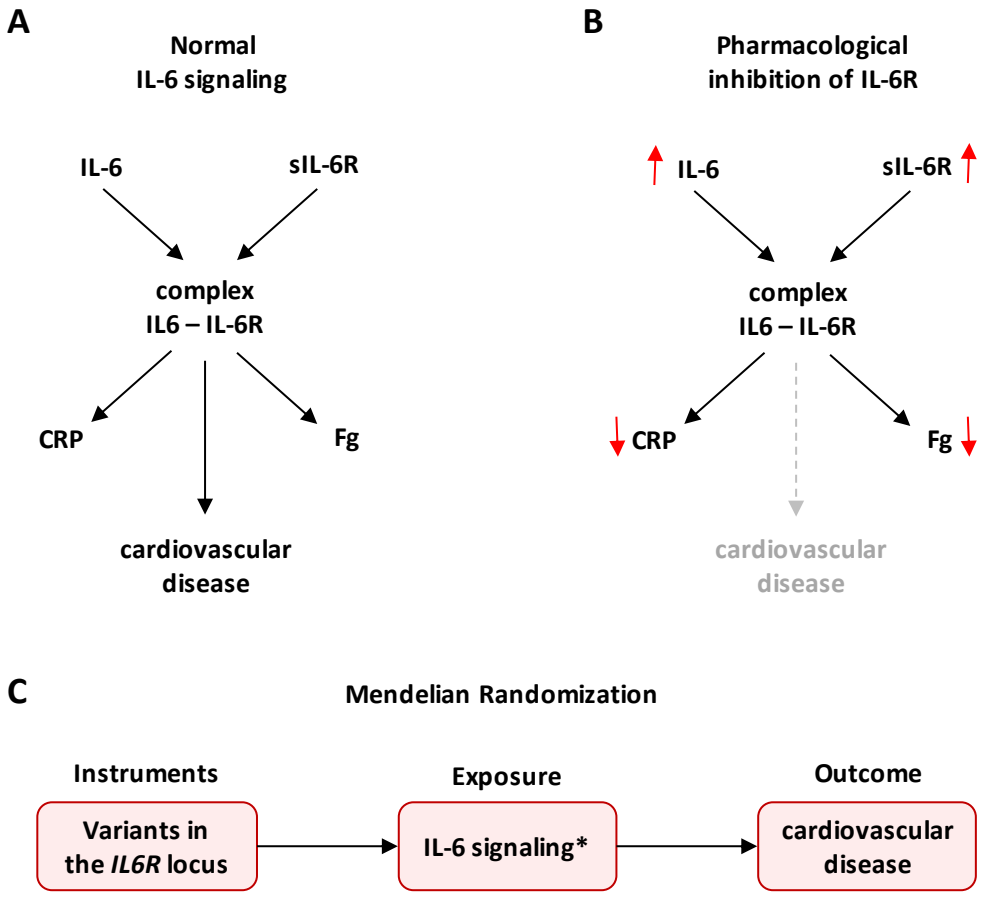
* Variants located within the *IL6R* gene.

† Beta coefficients correspond to 1-unit changes in the natural-log-transformed CRP (mg/L) per copy increment in effect allele.

‡ $R^2 = 2 \times MAF \times (1 - MAF) \times beta^2$, where MAF is the minimum allele frequency and beta is the effect estimate of the SNP on CRP levels.[36]

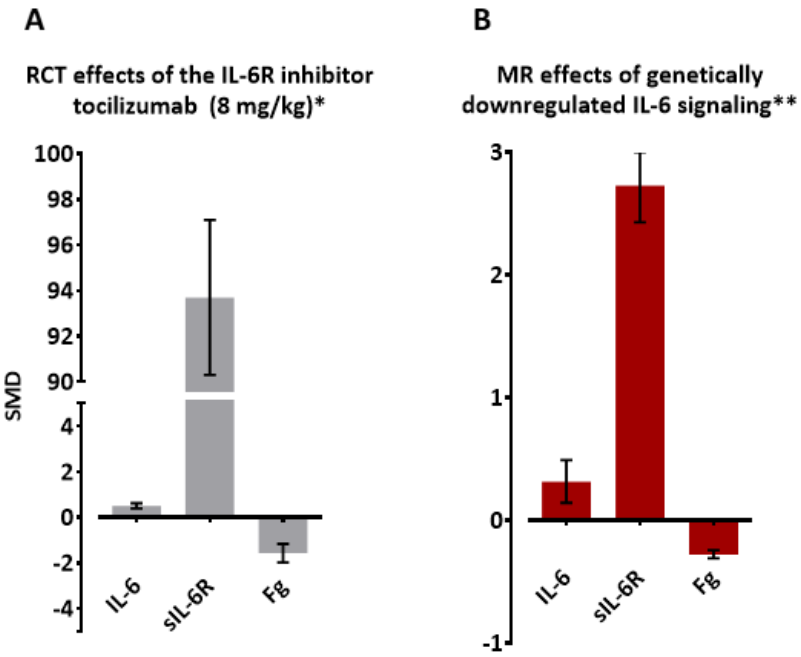
§ $F = \frac{R^2 \times (N-2)}{1-R^2}$ where R^2 is the variance of CRP explained by the specific SNP (as explained above) and N the number of individuals in the GWAS analysis.[37]

Figure 1. Conceptual framework and design of the current Mendelian Randomization approach. (**A**) Shown is a simplified scheme of IL-6 signaling, which is induced by binding of IL-6 to the soluble or the membrane-bound form of its receptor (IL-6R). IL-6 signaling results in increased C-reactive protein (CRP) and Fibrinogen (Fg) levels and is associated with a higher risk of cardiovascular disease. (**B**) Pharmacological inhibition of IL-6R leads to increases in the levels of upstream regulators (IL-6 and sIL-6R), and decreases in the levels of downstream effectors (CRP and Fg) of the IL-6 signaling pathway, but its effects on cardiovascular disease remain unknown. (**C**) In the current MR approach, we selected genetic variants within the *IL6R* locus, which significantly associated with lower CRP levels, as instruments (proxies) for a downregulated IL-6 signaling, and explored their effects on ischemic stroke, coronary artery disease and other cardiovascular disease phenotypes.



* IL-6 signaling was determined by the effects of the instruments on CRP levels. The instruments were further validated by exploring their effects on other upstream regulators (IL-6, sIL-6-R) and downstream effectors (Fg) of IL-6 signaling.
sIL-6R, soluble IL-6 receptor.

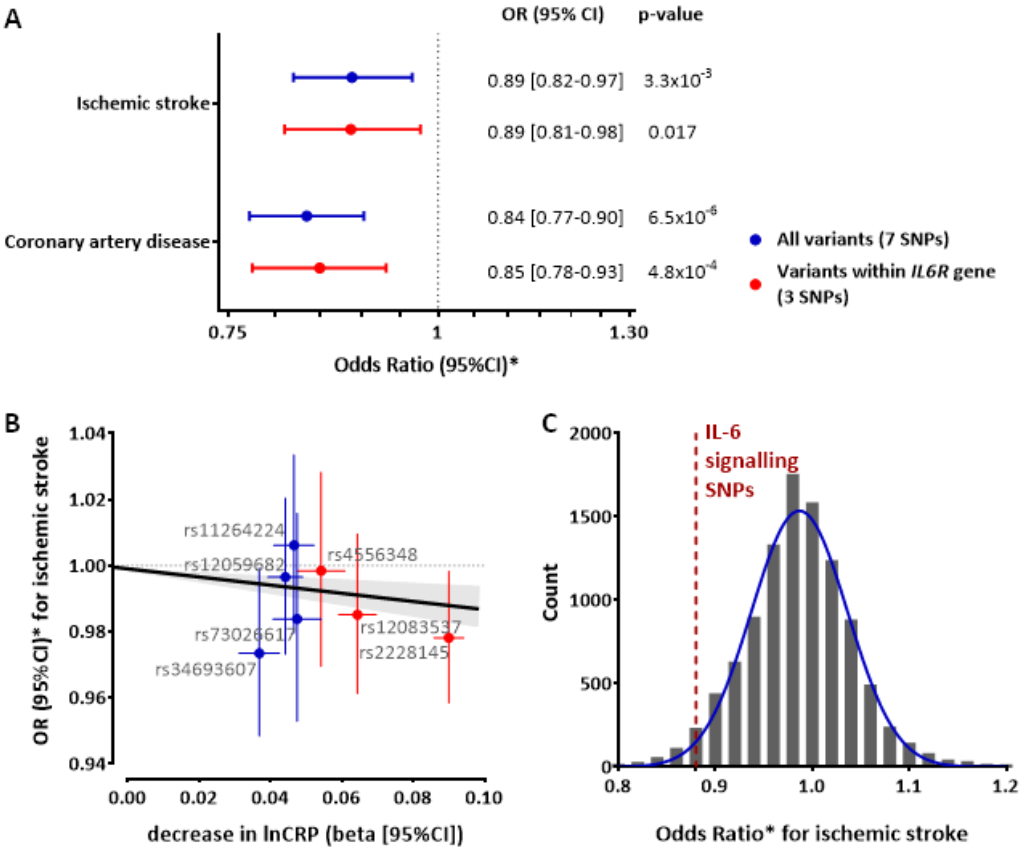
Figure 2. Effects of pharmacological inhibition of IL-6R and of genetic downregulation of IL-6 signaling on circulating levels of IL-6, soluble IL-6R (sIL-6R), and fibrinogen (Fg). (**A**) Effects of pharmacological inhibition of IL-6R on IL-6, sIL-6R, and Fg levels by administration of tocilizumab (8 mg/kg), as compared to placebo in a meta-analysis of 4 randomized clinical trials (RCT). Effects represent the standardized mean differences (SMD) in IL-6, sIL-6R, and Fg levels between 8 and 24 weeks after administration of tocilizumab (8 mg/kg), as compared to placebo. (**B**) Effects of genetic downregulation of IL-6 signaling on IL-6, sIL-6R, and Fg levels as determined by Mendelian Randomization (MR) analyses. Effects represent SMDs in IL-6, sIL-6R, and Fg levels.



* The SMDs for RCTs are derived from a meta-analysis of 4 studies.[20]

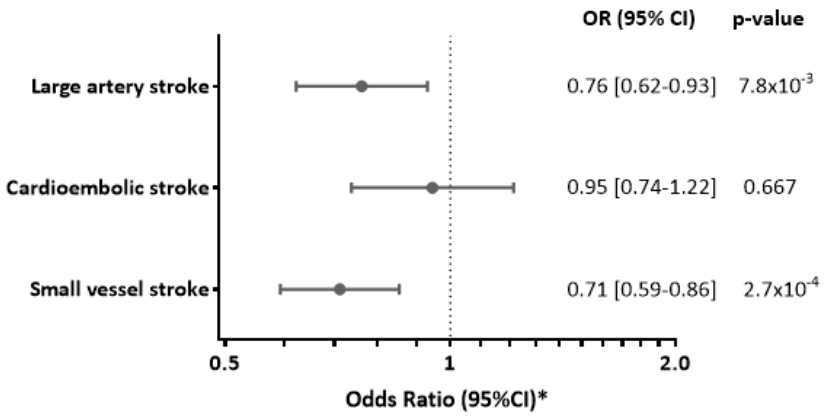
** The SMDs for the MR analyses are scaled to the CRP-decreasing effects of tocilizumab (8 mg/kg).

Figure 3. Mendelian Randomization associations of genetically downregulated IL-6 signaling with ischemic stroke. (**A**) Genetically downregulated IL-6 signaling in association with ischemic stroke and coronary artery disease as derived from IVW MR analyses either using the full set of genetic instruments (7 SNPs), or the restricted set of instruments (3 SNPs located within the *IL6R* gene). (**B**) SNP-specific effects regarding the associations of genetically downregulated IL-6 signaling with ischemic stroke and results derived from the IVW MR analysis. (**C**) Distributions of the effects of 7 randomly selected CRP-decreasing SNPs on risk of ischemic stroke and the position of the IL-6 signaling downregulating effect (7 SNPs included in our analyses).



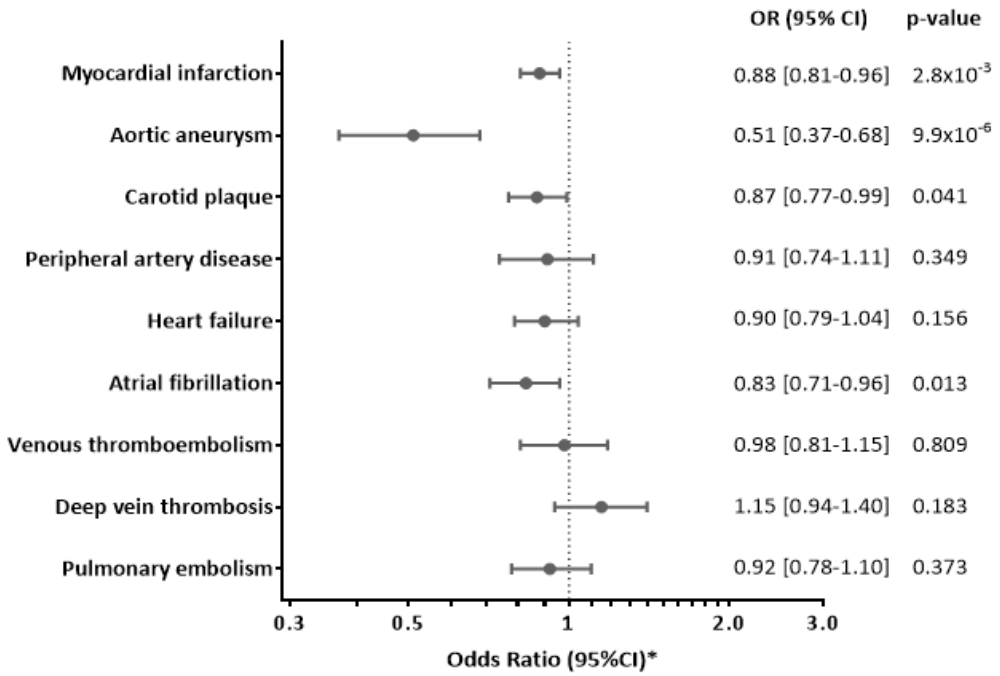
* Odds Ratios for genetically downregulated IL-6 signaling are scaled to the CRP-decreasing effects of tocilizumab (8 mg/kg).

Figure 4. Mendelian Randomization associations of genetically downregulated IL-6 signaling with ischemic stroke etiological subtypes. The effects represent Odds Ratios (OR) derived from inverse-variance-weighted MR analyses.



* Odds Ratios for genetically downregulated IL-6 signaling are scaled to the CRP-decreasing effects of tocilizumab (8 mg/kg).

Figure 5. Mendelian Randomization associations of genetically downregulated IL-6 signaling with other cardiovascular outcomes. The effects represent Odds Ratios (OR) derived from inverse-variance-weighted MR analyses.



* Odds Ratios for genetically downregulated IL-6 signaling are scaled to the CRP-decreasing effects of tocilizumab (8 mg/kg).

MANUSCRIPT IV: Use of Genetic Variants Related to Antihypertensive Drugs to Inform on Efficacy and Side Effects

Gill D*, **Georgakis MK*,** Koskeridis F, Jiang L, Feng Q, Wei WQ, Theodoratou E, Elliott P, Denny JC, Malik R, Evangelou E, Dehghan A, Dichgans M†, Tzoulaki I†. Use of Genetic Variants Related to Antihypertensive Drugs to Inform on Efficacy and Side Effects. ***Circulation***. 2019 Jul 23;140(4):270-279. *,† equally contributed

Authors contributions: DG, IT, MKG and MD conceptualized and designed the study. DG, MKG, FK and LJ collectively had full access to the data and performed the analysis. All authors interpreted the results. DG and IT drafted the manuscript. All authors critically revised the manuscript for intellectual content. All authors approved the submitted version and are accountable for the integrity of the work.

Circulation

ORIGINAL RESEARCH ARTICLE

Use of Genetic Variants Related to Antihypertensive Drugs to Inform on Efficacy and Side Effects

BACKGROUND: Drug effects can be investigated through natural variation in the genes for their protein targets. The present study aimed to use this approach to explore the potential side effects and repurposing potential of antihypertensive drugs, which are among the most commonly used medications worldwide.

METHODS: Genetic proxies for the effect of antihypertensive drug classes were identified as variants in the genes for the corresponding targets that associated with systolic blood pressure at genome-wide significance. Mendelian randomization estimates for drug effects on coronary heart disease and stroke risk were compared with randomized, controlled trial results. A phenome-wide association study in the UK Biobank was performed to identify potential side effects and repurposing opportunities, with findings investigated in the Vanderbilt University biobank (BioVU) and in observational analysis of the UK Biobank.

RESULTS: Suitable genetic proxies for angiotensin-converting enzyme inhibitors, β-blockers, and calcium channel blockers (CCBs) were identified. Mendelian randomization estimates for their effect on coronary heart disease and stroke risk, respectively, were comparable to results from randomized, controlled trials against placebo. A phenome-wide association study in the UK Biobank identified an association of the CCB standardized genetic risk score with increased risk of diverticulosis (odds ratio, 1.02 per standard deviation increase; 95% CI, 1.01–1.04), with a consistent estimate found in BioVU (odds ratio, 1.01; 95% CI, 1.00–1.02). Cox regression analysis of drug use in the UK Biobank suggested that this association was specific to nondihydropyridine CCBs (hazard ratio 1.49 considering thiazide diuretic agents as a comparator; 95% CI, 1.04–2.14) but not dihydropyridine CCBs (hazard ratio, 1.04; 95% CI, 0.83–1.32).

CONCLUSIONS: Genetic variants can be used to explore the efficacy and side effects of antihypertensive medications. The identified potential effect of nondihydropyridine CCBs on diverticulosis risk could have clinical implications and warrants further investigation.

Dipender Gill, MD*
Marios K. Georgakis, MD*
Fotios Koskeridis, MSc
Lan Jiang, MS
Qiping Feng, PhD
Wei-Qi Wei, MD, PhD
Evropi Theodoratou, PhD
Paul Elliott, FMedSci
Joshua C. Denny, MD, MS
Rainer Malik, PhD
Evangelos Evangelou, PhD
Abbas Dehghan, MD, PhD
Martin Dichgans, MD†
Ioanna Tzoulaki, PhD†

*Drs Gill and Georgakis contributed equally.

†Drs Dichgans and Tzoulaki contributed equally.

Key Words: antihypertensive drugs ■ Mendelian randomization analysis

Sources of Funding, see page 278

https://www.ahajournals.org/journal/circ

Clinical Perspective

What Is New?

- This work identifies genetic variants that serve as proxies for the effect of angiotensin-converting enzyme inhibitor, β-blocker, and calcium channel blocker antihypertensive drugs.
- Mendelian randomization using the genetic proxies for each respective drug class provides estimates consistent with those of randomized, controlled trials against placebo for effects on risk of coronary heart disease and stroke.
- Phenome-wide association study identifies diverticulosis as a previously unreported possible side effect of calcium channel blockers, with observational analysis further supporting an association between nondihydropyridine calcium channel blocker use and increased risk of diverticulosis.

What Are the Clinical Implications?

- Any increase in the risk of diverticulosis related to use of nondihydropyridine calcium channel blockers could have notable consequences and warrants further study.
- No other potential side effects of angiotensin-converting enzyme inhibitors, β-blockers, or calcium channel blockers were identified.

In 2015, the 874 million adults worldwide estimated to have a systolic blood pressure (SBP) of ≥140 mm Hg accounted for 106 deaths per 100 000 and loss of 143 million disability-adjusted life-years,[1] making hypertension a leading cause of mortality and morbidity. Blood pressure lowering through lifestyle modification or pharmacological treatment can significantly decrease cardiovascular risk, with every 10 mm Hg reduction estimated to decrease risk of all-cause mortality by 13%.[2]

The pharmacological treatment of hypertension is founded on strong evidence, underpinned by a large number of outcome-based randomized, controlled trials (RCTs) that have identified several drug classes to be effective for lowering blood pressure.[3] However, RCTs based on clinical outcomes have limitations[4]; they are largely restricted to older or high-risk patients and have a relatively short duration of follow-up, rarely beyond 5 years.[5] Therefore, recommendations for treatment are often based on extrapolation of the available evidence, with known side effects frequently limited to relatively common outcomes captured in RCTs.[6] At the same time, particular drug treatments for hypertension may have beneficial effects beyond their blood pressure–lowering properties,[6] thus offering potential for repurposing. However, observational research used to study such opportunities suffers from well-characterized biases, including confounding by indication.[7]

With the growing availability of genome-wide association study (GWAS) meta-analyses, it is becoming increasingly feasible to study drug effects by investigating genetic variants in the genes of their protein targets, as has previously been applied to lipid-lowering drugs.[7] In this study, human genetic variants within genes corresponding to the targets of common pharmacological agents for hypertension were first identified to serve as a proxy for the effects of these treatments. Second, the validity of this approach for studying the effects of these drugs was investigated by exploring consistency in mendelian randomization (MR) estimates for their effect on coronary heart disease (CHD) and stroke risk with corresponding RCT findings. Finally, to offer insight into their adverse effect profiles and repurposing potential, phenome-wide association study (PheWAS) analyses were undertaken with replication in an external dataset, as well as further investigation in observational analysis of drug use.

METHODS

All supporting data are available within the article, the online-only Data Supplement, and the web links provided. UK Biobank data were accessed through application 236. Relevant ethical approval and participant consent were already obtained in all studies that contributed data to this work. Statistical analysis was undertaken with R version 3.4.1 (The R Foundation for Statistical Computing) and Stata 14.2 (StataCorp LP).

Genetic Variant Selection

Common antihypertensive drugs were selected for study on the basis of recent consensus guidelines[6]: angiotensin-converting enzyme (ACE) inhibitors, angiotensin receptor blockers, β-blockers (BB), calcium channel blockers (CCB) and thiazide diuretic agents. Genes encoding the targets of these drugs related to effects on blood pressure were identified using the DrugBank database,[8] with promoter and enhancer regions identified using the GeneHancer database in the GeneCards online platform (version 4.7).[9] Genetic association estimates for SBP were obtained from a GWAS meta-analysis of 757 601 individuals with European ancestry drawn from the UK Biobank and the International Consortium of Blood Pressure GWAS meta-analysis,[10] where correction was made for antihypertensive medication use by adding 15 mm Hg to the SBP of participants receiving medication, with further adjustment for body mass index.[10] In sensitivity analyses, a GWAS of SBP on ≈337 000 white British individuals in the UK Biobank was also used, without correction for medication use or adjustment for body mass index .[11] Genetic variants to serve as proxies (ie, instruments) for the effect of lower SBP through antihypertensive drug targets were selected as single-nucleotide polymorphisms (SNPs) in corresponding genes, promoter regions, or enhancers that were associated with SBP at genome-wide significance ($P<5\times10^{-8}$) and clumped to a linkage disequilibrium (LD) threshold of $r^2<0.1$ using the 1000G European reference panel. This approach does not distinguish between selection of loss-of-function variants or those related to gene expression. The R^2 and F statistics

were used to estimate the variance in SBP explained and the strength of each SNP, respectively.[12]

Statistical Analysis

Mendelian Randomization

MR uses randomly allocated genetic variants related to an exposure of interest to study the effect of that exposure on a given outcome. In this study, the exposure of interest was SBP lowering through a particular antihypertensive drug class. All antihypertensive drug classes for which SNPs were identified as proxies using the larger SBP GWAS were taken forward to MR analysis investigating their effect on CHD and stroke risk. Genetic association estimates for CHD were obtained from the CARDIoGRAMplusC4D (Coronary Artery Disease Genome-wide Replication and Meta-analysis [CARDIOGRAM] plus the Coronary Artery Disease [C4D] Genetics) Consortium's 1000 Genomes–based transethnic meta-analysis of 60 801 case subjects and 123 504 control subjects.[13] Estimates for stroke risk were obtained from the MEGASTROKE Consortium's transethnic meta-analysis of 67 162 cases of any stroke and 454 450 control subjects.[14] Details for the MR analyses are provided in the online-only Data Supplement Methods. To allow comparison with RCT results, all MR estimates were scaled to the SBP-lowering effect of their respective drug class as measured in these RCTs.[3]

Investigation of Genetic Pleiotropy Unrelated to Drug Effect

The MR estimates can be biased if any of the genetic variants used affect the outcome under consideration through a pleiotropic pathway that is independent of the drug effect for which they serve as proxies. The PhenoScanner curated database of publicly available SNP-phenotype associations (accessed on March 30, 2018) was used to explore whether any of the selected SNPs or proxies with LD r^2>0.8 (using a 1000G reference panel) were also associated at genome-wide significance (P<5×10^{-8}) with traits that may potentially be exerting such pleiotropy,[15] and any such SNPs were excluded in sensitivity analyses. PhenoScanner includes SNP-phenotype associations identified in analysis of UK Biobank data.[11] Statistical evaluations of pleiotropy were also incorporated where multiple genetic variants were available to serve as proxies for the drug effect[16] and are detailed in the Methods in the online-only Data Supplement.

Phenome-Wide Association Study

The UK Biobank, a prospective study comprising approximately half a million middle-aged individuals,[17] served as the cohort for the PheWAS investigating drug side effects and repurposing opportunities. The participants provided self-reported information, with blood samples collected for biochemical tests and genotyping and physical measurements performed as described previously.[17] Individuals were linked retrospectively and prospectively to the National Health Service's Hospital Episode Statistics database.

PheWAS was restricted to participants of self-reported European descent, with random exclusion of 1 participant from each pair of relatives based on a kinship coefficient >0.0884. For antihypertensive drugs for which genetic variants were identified to serve as proxies, PLINK was used to construct a genetic risk score (GRS) for each individual, weighted for the SBP-lowering effect of each participating SNP,[18] and standardized to have a mean of 0 and an SD of 1 across all individuals. The 9th and 10th revisions of the *International Classification of Diseases* were used to define cases based on inpatient Hospital Episode Statistics data. The phecode grouping system was used to align diagnoses used in clinical practice with genomic analysis.[19] A series of case-control groups were generated for each phecode, with control subjects identified as individuals with no record of the respective outcome and its related phecodes.[19] Analysis was performed with logistic regression after adjustment for age, sex, and first 4 genetic principal components. Only outcomes that had a minimum of 200 cases were considered, to maintain sufficient statistical power to identify associations with common variants.[20] A 5% threshold with the false-discovery rate method was used in ascertaining the statistical significance of associations, to correct for multiple testing of correlated phenotypes. As for the MR analysis, sensitivity analyses were performed using genetic association estimates derived from the SBP GWAS that did not correct for medication use or adjust for body mass index, and after the exclusion of any SNPs with potentially pleiotropic associations at genome-wide significance that were identified with PhenoScanner.[15]

PheWAS associations for noncardiovascular conditions were investigated for relation to SBP more generally using a permutation-based approach that repeated association analyses 1000 times, with the standardized GRS created on each instance using a matched number of randomly sampled SBP-related SNPs from throughout the genome (ie, associated with SBP at genome-wide significance and clumped to LD r^2<0.001; Table I in the online-only Data Supplement). Compared with the investigation of antihypertensive drug targets, a more stringent LD threshold was used, because variants for SBP were selected from throughout the genome rather than any particular locus. The proportion of such permutation analyses that have a consistent direction of effect and P value lower than in the main PheWAS analysis would serve as an adjusted P value of the null hypothesis. Further study of any PheWAS associations significant at a false-discovery rate threshold of 5% for noncardiovascular conditions was also undertaken in the Vanderbilt University Biobank (BioVU), for which genetic data on ≈50 000 individuals are linked to a deidentified electronic health record system.[21] Similar to the main PheWAS, a standardized GRS was constructed, and logistic regression with the outcome was performed after adjustment for age, sex, and first 3 principal components. The analysis was restricted to individuals identified as white, with control subjects based on the same exclusions as the main PheWAS. Results between the UK Biobank and BioVU analysis were pooled by use of a fixed-effects meta-analysis model.

Observational Analysis of Drug Use

PheWAS associations significant at a 5% false-discovery rate for noncardiovascular conditions related to any antihypertensive class were further explored in observational analysis of drug use among individuals in the UK Biobank. This additionally allowed for investigation of the dihydropyridine

and nondihydropyridine CCB subclasses, which was not possible when using genetic proxies because of overlap in the genes for their corresponding protein targets. Cox regression analysis was used to compare time to first incident outcome between individuals orally taking different antihypertensive drug classes at baseline. Individuals who died during the follow-up period before a relevant diagnosis were censored. The categories of antihypertensive drug treatment considered were ACE inhibitors alone, angiotensin receptor blockers alone, BBs alone, dihydropyridine CCBs alone, nondihydropyridine CCBs alone, thiazide diuretic agents alone, a combination of medications from any 2 antihypertensive classes, and a combination of medications from 3 or more antihypertensive classes. In a separate model, individuals who were taking any subclass of CCBs were pooled into a single category. Adjustment was made for age, sex, body mass index, Townsend Deprivation Index, smoking status, previous cancer diagnosis, number of noncancer diagnoses, and number of previous surgical operations. Individuals with a diagnosis of the condition under consideration before recruitment were excluded.

RESULTS

Genetic Variant Selection

The genes and enhancer and promoter regions corresponding to the targets of each antihypertensive drug class are shown in Table II in the online-only Data Supplement. There was 1 gene identified for each drug target for ACE inhibitors (*ACE*), angiotensin receptor blockers (*AGTR1*), BBs (*ADRB1*), and thiazide diuretic agents (*SLC12A3*), and 11 genes for CCBs (*CACNA1D*, *CACNA1F*, *CACNA2D1*, *CACNA2D2*, *CACNA1S*, *CACNB1*, *CACNB2*, *CACNB3*, *CACNB4*, *CACNG1*, and *CACNA1C*) encoding the different calcium channel subunits related to effects on blood pressure. The *CACNA1F* gene is located on the X chromosome, and SNPs corresponding to this region were not available. Using the predefined selection criteria, there was 1 SNP identified for ACE inhibitors, 6 for BBs, and 24 for CCBs (Tables III through V in the online-only Data Supplement). The larger number of SNPs and correspondingly greater proportion of variation in SBP explained for CCBs was related to the availability of more genes from which to identify variants. The F statistic for SNPs ranged from 54 to 534 (Tables III through V in the online-only Data Supplement), consistent with a low risk of weak instrument bias.[12]

Mendelian Randomization

To allow comparison with RCT meta-analysis effect estimates, MR results for each drug class were scaled to their respective SBP-lowering effect in these studies. Thus, for ACE inhibitors, MR estimates are given per 21.14 mm Hg decrease, for BBs per 9.51 mm Hg decrease, and for CCBs per 8.90 mm Hg decrease.[3] MR analysis using the single genetic variant identified for ACE inhibitors showed a protective effect on stroke (relative risk [RR], 0.21; 95% CI, 0.06–0.72; P=0.01) but not CHD risk (RR, 0.67; 95% CI, 0.16–2.56; P=0.58). The main MR analysis using the 6 variants for BBs identified a protective effect on CHD risk (RR, 0.62; 95% CI, 0.47–0.81; $P=4\times10^{-4}$) but not stroke risk (RR, 0.91; 95% CI, 0.73–1.14; P=0.41). For CCBs, the main MR analysis using the 24 SNPs identified a protective effect on both CHD risk (RR, 0.73; 95% CI, 0.64–0.84; $P=6\times10^{-6}$) and stroke risk (RR, 0.75; 95% CI, 0.66–0.84; $P=1\times10^{-6}$). Similar results for all drug classes were obtained when the incidence of CHD and stroke was considered to be 1%, 5%, and 10% (Table VI in the online-only Data Supplement). The MR estimates had overlapping 95% CIs to those from RCTs of these drugs versus placebo[3] (Figure 1). Individual MR estimates for each BB and CCB SNP are given in Figures I through IV in the online-only Data Supplement. Consistent MR results were found in sensitivity analyses, as detailed in the online-only Data Supplement (Results, Tables VII through IX, and Figures V through VIII).

Phenome-Wide Association Study

After quality control and mapping of *International Classification of Diseases, 9th Revision* and *10th Revision*, to phecodes, data for 424 439 individuals across 909 distinct phenotypes were available for PheWAS analysis. Details of the number of phenotypes and cases per disease category are provided in the Table, with the number of cases and controls for each outcome in Tables X through XVI in the online-only Data Supplement. Using the ACE inhibitor, BB, and CCB standardized GRS, the respective PheWAS analyses revealed associations with hypertension and related cardiovascular disease (Figures 2–4 and Tables X through XII in the online-only Data Supplement). CCBs additionally showed an association with higher risk of diverticulosis (odds ratio per SD increase in standardized GRS, 1.02; 95% CI, 1.01–1.04, $P=2\times10^{-4}$). Similar results were obtained in PheWAS sensitivity analyses (Tables XIII through XVI in the online-only Data Supplement). Random sampling of 24 SBP SNPs from throughout the genome (Table I in the online-only Data Supplement) to create standardized GRSs and measurement of associations with diverticulosis risk in permutation analyses (N=1000) showed effect estimates centered close to the null (mean odds ratio per SD increase in standardized GRS, 1.00; 95% CI, 0.98–1.02, P=0.79; Figure IX in the online-only Data Supplement). Of the 1000 permutation analyses, only 10 had a consistent direction of effect and P value lower than that observed for the association of the standardized CCB GRS with diverticulosis in PheWAS, thus generating an adjusted P value=0.01.

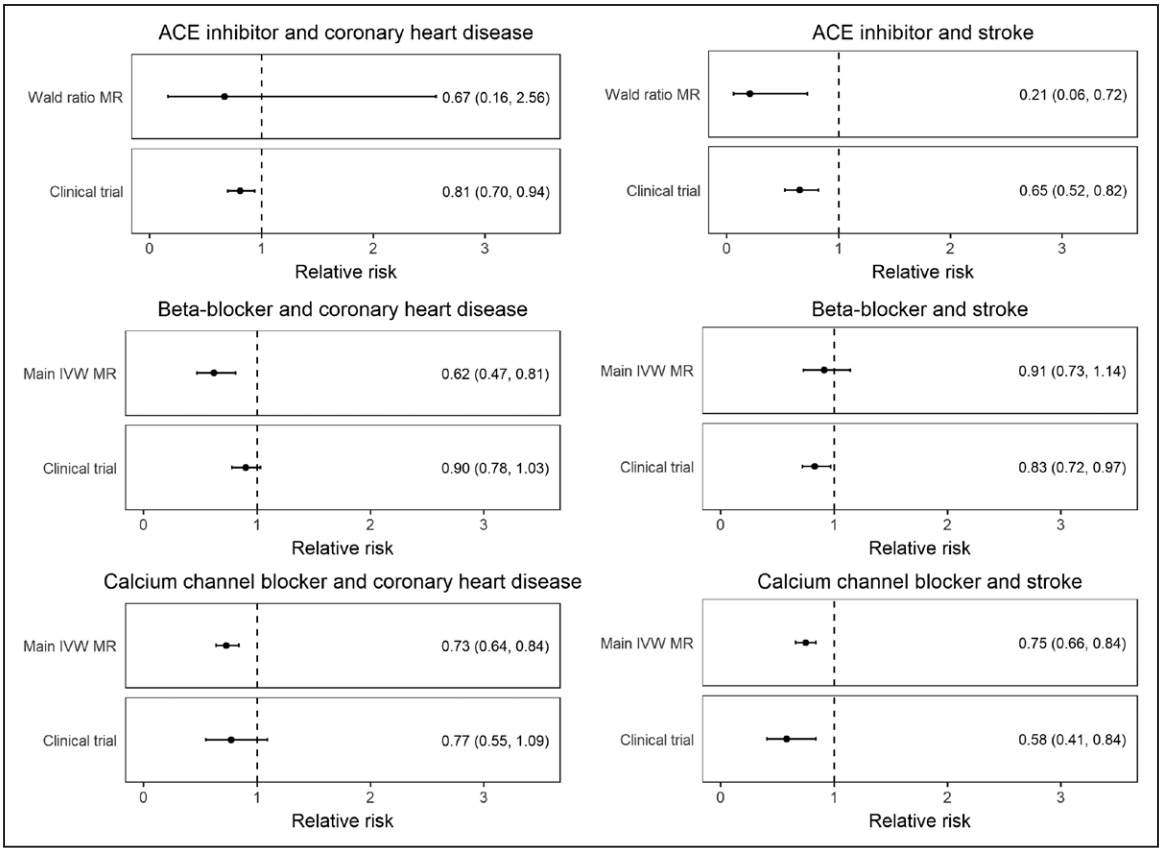


Figure 1. MR estimates for the effect of genetically lower systolic blood pressure through the ACE inhibitor, β-blocker, and calcium channel blocker variants, respectively, on risk of coronary heart disease and stroke, compared with randomized, controlled trial meta-analysis results.[3]
ACE indicates angiotensin-converting enzyme; IVW, inverse variance weighted; and MR, Mendelian randomization.

Data for 45 517 individuals were available in BioVU to further investigate novel PheWAS findings for traits unrelated to hypertension. General cohort characteristics for the considered populations from the UK Biobank and BioVU are detailed in Table XVII in the online-only Data Supplement. The prevalence of diverticulosis in BioVU was 12%, comparable to the 10% observed in the UK Biobank. In BioVU, the CCB standardized GRS association with diverticulosis had an odds ratio per SD increase of 1.01 (95% CI, 1.00–1.02; P=0.17). The meta-analyses of UK Biobank and BioVU estimates had an odds ratio of 1.02 (95% CI, 1.01–1.03; P=3×10^{-4}; Figure 5).

Observational Analysis of Drug Use

For the observational analysis of antihypertensive drug use in the UK Biobank, there were 1408 incident diverticulosis diagnoses up to February 13, 2016, in the 54 612 individuals taking any of the considered antihypertensive drug classes at recruitment (March 13, 2006, to October 1, 2010), with a mean follow-up of 2538 days. In adjusted Cox regression (with use of thiazide diuretic antihypertensive medications alone as the reference category), there was no evidence for an association between use of any CCB and risk of diverticulosis (hazard ratio, 1.10; 95% CI, 0.88–1.35; P=0.43). Considering CCB subclasses, there was evidence for an association with risk of diverticulosis for nondihydropyridine CCB use (hazard ratio, 1.49; 95% CI, 1.03–2.14; P=0.03) but not dihydropyridine CCB use (hazard ratio, 1.01; 95% CI, 0.80–1.28; P=0.91) or any other antihypertensive drug class (Table XVIII in the online-only Data Supplement).

DISCUSSION

This work leveraged large-scale GWAS data from >750 000 individuals and generated genetic proxies for the effect of ACE inhibitors, BBs, and CCBs, 3 of the most commonly used medications worldwide. The MR estimates for risk of CHD and stroke were comparable to those observed in RCTs against placebo, which supports the validity of the approach. PheWAS on 909 outcomes corroborated the known efficacy of these agents in preventing hypertension and related vascular diseases, thus further supporting the robustness of the genetic variants used.

The PheWAS investigation also revealed an increased risk of diverticulosis associated with the standardized

Table. Number of Phenotypes and Cases per Disease Category in the UK Biobank Phenome-Wide Association Study Analysis

Disease Category	Phenotypes, n	Cases, n			
		Minimum	Median	Mean	Maximum
Circulatory system	98	202	1048	6308	133749
Congenital anomalies	19	211	442	557	1823
Dermatologic	43	218	799	4765	82669
Digestive	116	228	1455	4817	79488
Endocrine/metabolic	49	208	773	4076	45303
Genitourinary	106	203	1376	4153	103829
Hematopoietic	22	201	569	2690	12759
Infectious diseases	25	219	1012	2237	10752
Injuries and poisonings	59	222	536	1513	16683
Mental disorders	36	202	710	3280	29405
Musculoskeletal	57	213	925	4164	53823
Neoplasms	82	215	1124	4261	90826
Neurological	44	204	567	2286	40703
Pregnancy complications	17	208	1113	1854	9534
Respiratory	56	200	1124	3837	62168
Sense organs	64	210	774	2443	39998
Symptoms	16	304	2341	7036	42311

GRS for CCBs. No significant association with diverticulosis risk was identified when SBP SNPs were explored more generally, which makes effects through systemic SBP lowering unlikely to account for this. A consistent association between the standardized CCB GRS and diverticulosis risk was found in BioVU, which contained fewer cases and had a correspondingly wider CI that crossed the null. The finding was further supported by observa-

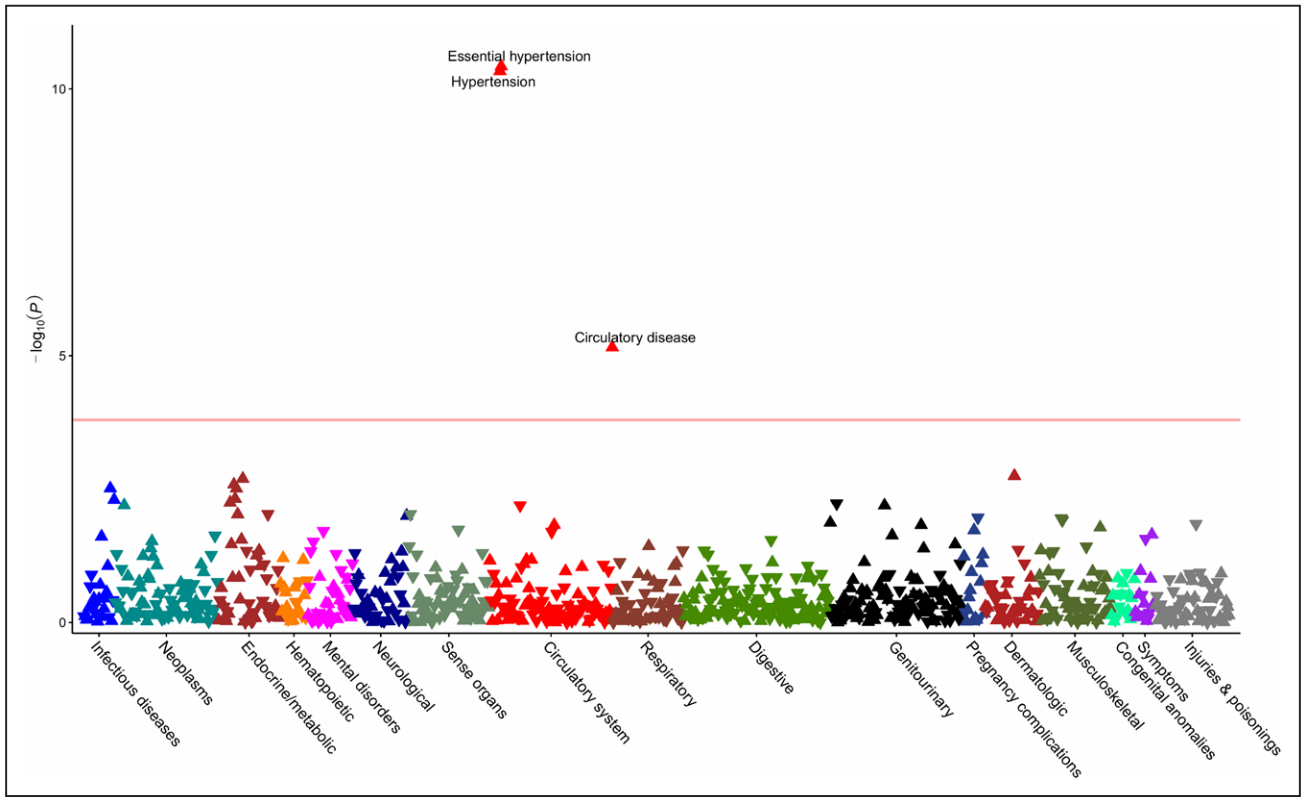


Figure 2. Phenome-wide association study of the standardized genetic risk score for angiotensin-converting enzyme inhibitors.
The horizontal line depicts the 5% false-discovery rate threshold.

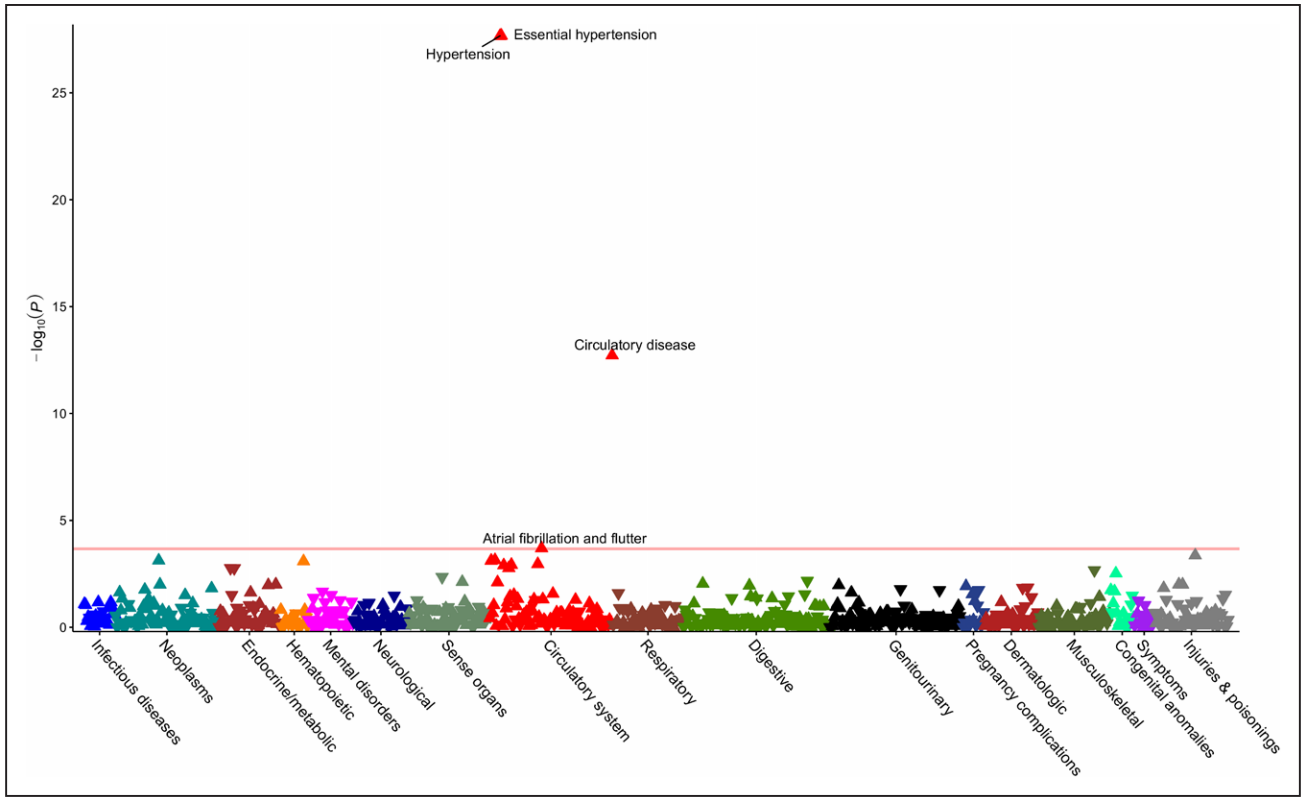


Figure 3. Phenome-wide association study of the standardized genetic risk score for β-blockers.
The horizontal line depicts the 5% false-discovery rate threshold.

tional analysis suggesting that nondihydropyridine CCB treatment at baseline in the UK Biobank was associated with increased risk of diverticulosis. Dihydropyridine and nondihydropyridine CCBs have different pharmacological effects, and it also follows that their side effect profiles vary.[22] In terms of a possible mechanism, constipation is an established side effect of nondihydropyridine CCBs, related to their role in reducing bowel contractility,[23] and it may be through a similar process that the risk of diverticulosis is increased. Alternatively, there may be specific effects on the vasa recta vessels that penetrate the muscle layer of the colon, thus giving rise to weak points where diverticulae consequently form.[24] Complications related to diverticulosis are a common reason for hospital admission[25] and have a rising incidence.[26] Given that more than one-tenth of the world's adults have hypertension, and CCBs are recommended as a first-line pharmacological agent, with nondihydropyridine drugs in particular recommended for individuals with concurrent atrial fibrillation,[1,6] the clinical implications of these findings merit consideration. For example, individuals with or at increased risk of developing diverticulosis might benefit from alternative pharmacological treatments for hypertension. The genetic proxies for ACE inhibitors, BBs, and CCBs did not show detrimental associations with any of the other traits examined in PheWAS. Although absence of evidence is not evidence of absence, this does provide some assurance that long-term pharmacological inhibition of these drug targets is generally safe, with other side effects that require hospitalization being smaller or rarer.

A major strength of our work is that it uses genetic variants to investigate the effect of antihypertensive drugs using existing data obtained from large-scale studies, thus avoiding the time and resource constraints associated with such study through RCTs[4] and overcoming the limitations of potential confounding and reverse causation from use of standard observational methods.[7] A range of sensitivity analyses supported the robustness of this approach, with PheWAS allowing rapid investigation of hundreds of clinically relevant traits across the phenome. Additionally, observational analysis allowed for consideration of CCB subclasses and further replication of novel findings.

Concerning the limitations of the study, the MR and PheWAS results estimate the cumulative effect of lifelong exposure to genetic variants, rather than the consequence of a clinical intervention. Furthermore, there may be unknown pleiotropic effects of the genetic variants that bias the association estimates.[16] Although less stringent criteria for selecting instruments (such as a more relaxed *P* value threshold for association with SBP, or a more lenient LD criterion for clumping) might have increased the number of variants available, this could also have reduced the sensitivity and specificity of the analysis because of the introduction of weak

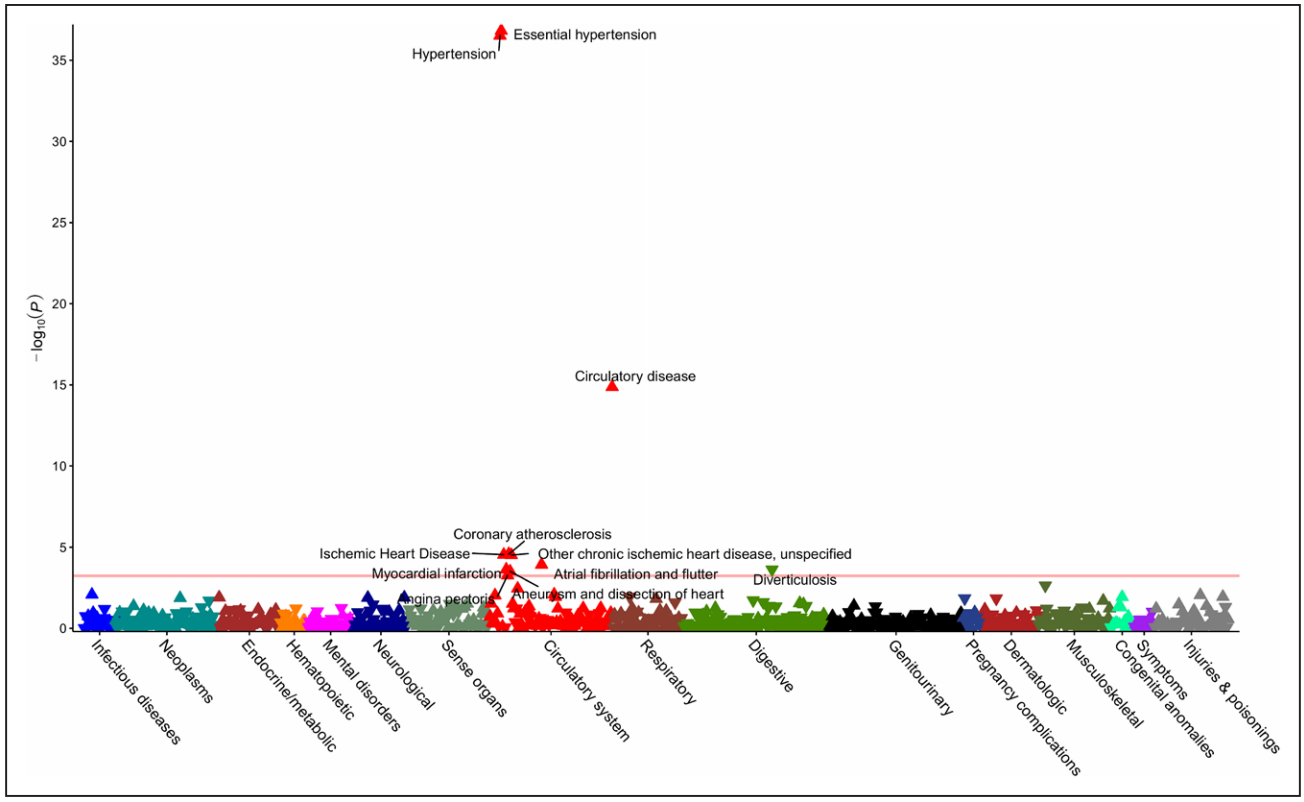


Figure 4. Phenome-wide association study of the standardized genetic risk score for calcium channel blockers.
The horizontal line depicts the 5% false-discovery rate threshold.

instrument bias and invalid instruments, respectively. Similarly, information on gene expression was not incorporated in this work, and although such an approach could offer an additional strategy for identifying genetic variants that serve as proxies for drug effects,[7] this would be restricted to the cells or tissues in which gene expression was measured, limiting applicability for exploration of general side effects or repurposing opportunities. Although the PheWAS analysis was performed to explore clinically relevant outcomes identified using harmonized Hospital Episode Statistics data in UK Biobank participants, there is also the potential to extend this approach to other cohorts and summary-level genetic data.[15] Finally, although the observational analysis of drug use in the UK Biobank did support an association between nondihydropyridine CCB use and risk of diverticulosis, it is not clear whether this finding may in part relate to ascertainment bias or residual confounding. Diverticulosis can be incidental in asymptomatic individuals, and as such, increased interaction with healthcare services could lead to a greater chance of diagnosis.

In conclusion, this work has identified genetic variants that serve as proxies for the effect of the ACE in-

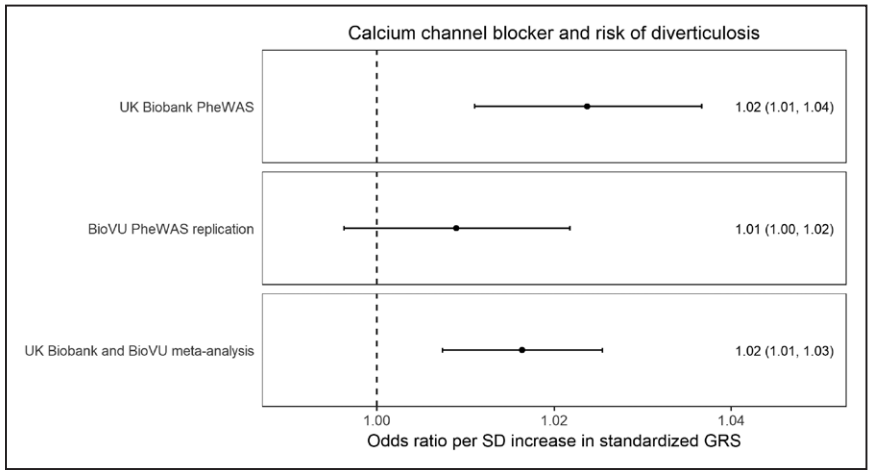


Figure 5. Estimates for genetic association between calcium channel blockers and diverticulosis risk derived from PheWAS analyses in the UK Biobank and BioVU, respectively, and their fixed-effects pooled estimate.
BioVU indicates Vanderbilt University Biobank; GRS, genetic risk score; and PheWAS, phenome-wide association study.

hibitor, BB, and CCB classes of antihypertensive medication. In MR and PheWAS, our instrumental variable approaches corroborated the established associations of these agents with a range of traits related to hypertension. Additionally, this study identified an apparent, previously unreported detrimental effect of nondihydropyridine CCBs on risk of diverticulosis, a finding that requires further replication before it should alter clinical practice. No other potential side effects of any drug class were identified to suggest a lack of long-term safety. This study demonstrates that the use of genetic variants offers a powerful complement to existing RCT and observational approaches for investigating the efficacy, side effects, and repurposing potential of antihypertensive agents.

ARTICLE INFORMATION

Received November 9, 2018; accepted May 2, 2019.

The online-only Data Supplement, podcast, and transcript are available with this article at https://www.ahajournals.org/doi/suppl/10.1161/circulationaha.118.038814.

Correspondence

Dipender Gill, MD, Department of Biostatistics and Epidemiology, School of Public Health, St Mary's Hospital, Norfolk Place, Imperial College London, United Kingdom, W2 1PG. Email dipender.gill@imperial.ac.uk

Affiliations

Department of Epidemiology and Biostatistics, School of Public Health, Imperial College London, United Kingdom (D.G., P.E., E.E., A.D., I.T.). Institute for Stroke and Dementia Research, University Hospital (M.K.G., R.M., M.D.) and Graduate School for Systemic Neurosciences (M.K.G.), Ludwig-Maximilians-Universität LMU, Munich, Germany. Department of Hygiene and Epidemiology, University of Ioannina Medical School, Greece (F.K., E.E., I.T.). Division of Clinical Pharmacology, Department of Medicine (L.J., Q.F.) and Department of Biomedical Informatics (W.-Q.W., J.C.D.), Vanderbilt University Medical Center, Nashville, TN. Centre for Global Health Research, Usher Institute of Population Health Sciences and Informatics, University of Edinburgh, United Kingdom (E.T.). Medical Research Council-Public Health England Centre for Environment, School of Public Health, Imperial College London, United Kingdom (P.E., A.D., I.T.). Imperial Biomedical Research Centre, Imperial College London and Imperial College NHS Healthcare Trust, UK (P.E.). UK Dementia Research Institute at Imperial College London, UK (P.E., A.D., I.T.). Health Data Research UK-London (P.E.). Munich Cluster for Systems Neurology (SyNergy), Germany (M.D.). German Center for Neurodegenerative Diseases (DZNE, Munich), Germany (M.D.).

Acknowledgments

D.G., M.K.G., M.D., and I.T. designed the study. D.G., M.K.G., F.K., and L.J. collectively had full access to the data and performed the analysis. All authors interpreted the results. D.G. and I.T. drafted the manuscript. All authors critically revised the manuscript for intellectual content. All authors approved the submitted version and are accountable for the integrity of the work.

Sources of Funding

This work was funded by the Wellcome 4i Clinical PhD Program at Imperial College London (to D.G.); scholarships from the Deutscher Akademischer Austauschdienst and the Onassis Foundation (to M.K.G.); National Institute of Health grants R01 HL133786, R01 GM120523 and R01 LM010685 (to Q.F., W.-Q.W., and J.C.D.); Cancer Research UK grants C31250/A22804 (to E.T.); the Medical Research Council and Public Health England (grant MR/L01341X/1) (to P.E., as part of the Medical Research Council-Public Health England Centre for Environment and Health); the National Institute for Health Research Imperial Biomedical Research Centre in collaboration with Imperial College Healthcare National Health Service Trust (to P.E.); the UK Medical Research Council, Alzheimer's Society and Alzheimer's Research UK (to P.E., as part of the UK Dementia Research Institute); a consortium led by the UK Medical Research Council (to P.E., as associate director of Health Data Research UK); the European Union Horizon 2020 research and innovation programme "Small vessel diseases in a mechanistic perspective: Targets for Intervention" (SVDs@target, No 666881; to M.D.); the German Research Foundation (DFG) as part of the "Munich Cluster for Systems Neurology" (SyNergy, EXC 1010) and the Collaborative Research Center 1123 (B3) (to M.D.).

Disclosures

None.

REFERENCES

1. Forouzanfar MH, Liu P, Roth GA, Ng M, Biryukov S, Marczak L, Alexander L, Estep K, Hassen Abate K, Akinyemiju TF, et al. Global burden of hypertension and systolic blood pressure of at least 110 to 115 mm Hg, 1990-2015. *JAMA.* 2017;317:165–182. doi: 10.1001/jama.2016.19043
2. Ettehad D, Emdin CA, Kiran A, Anderson SG, Callender T, Emberson J, Chalmers J, Rodgers A, Rahimi K. Blood pressure lowering for prevention of cardiovascular disease and death: a systematic review and meta-analysis. *Lancet.* 2016;387:957–967. doi: 10.1016/S0140-6736(15)01225-8
3. Wright JM, Musini VM, Gill R. First-line drugs for hypertension. *Cochrane Database Syst Rev.* 2018;4:CD001841. doi: 10.1002/14651858.CD001841.pub3
4. Frieden TR. Evidence for health decision making - beyond randomized, controlled trials. *N Engl J Med.* 2017;377:465–475. doi: 10.1056/NEJMra1614394
5. Tsang R, Colley L, Lynd LD. Inadequate statistical power to detect clinically significant differences in adverse event rates in randomized controlled trials. *J Clin Epidemiol.* 2009;62:609–616. doi: 10.1016/j.jclinepi.2008.08.005
6. Williams B, Mancia G, Spiering W, Agabiti Rosei E, Azizi M, Burnier M, Clement DL, Coca A, de Simone G, Dominiczak A, et al; Authors/Task Force Members. 2018 ESC/ESH guidelines for the management of arterial hypertension: the Task Force for the management of arterial hypertension of the European Society of Cardiology and the European Society of Hypertension. *J Hypertens.* 2018;36:1953–2041. doi: 10.1097/HJH.0000000000001940
7. Walker VM, Davey Smith G, Davies NM, Martin RM. Mendelian randomization: a novel approach for the prediction of adverse drug events and drug repurposing opportunities. *Int J Epidemiol.* 2017;46:2078–2089. doi: 10.1093/ije/dyx207
8. Wishart DS, Knox C, Guo AC, Shrivastava S, Hassanali M, Stothard P, Chang Z, Woolsey J. DrugBank: a comprehensive resource for in silico drug discovery and exploration. *Nucleic Acids Res.* 2006;34(database issue):D668–D672. doi: 10.1093/nar/gkj067
9. Fishilevich S, Nudel R, Rappaport N, Hadar R, Plaschkes I, Iny Stein T, Rosen N, Kohn A, Twik M, Safran M, et al. GeneHancer: genome-wide integration of enhancers and target genes in GeneCards. *Database.* 2017;2017:bax028. doi: 10.1093/database/bax028
10. Evangelou E, Warren HR, Mosen-Ansorena D, Mifsud B, Pazoki R, Gao H, Ntritsos G, Dimou N, Cabrera CP, Karaman I, et al; Million Veteran Program. Genetic analysis of over 1 million people identifies 535 new loci associated with blood pressure traits. *Nat Genet.* 2018;50:1412–1425. doi: 10.1038/s41588-018-0205-x
11. Churchhouse C, Neale B. Rapid GWAS of thousands of phenotypes for 337,000 samples in the UK Biobank. Neale Lab web site. http://www.nealelab.is/blog/2017/7/19/rapid-gwas-of-thousands-of-phenotypes-for-337000-samples-in-the-uk-biobank. Published July 19, 2017. Accessed March 30, 2018.
12. Palmer TM, Lawlor DA, Harbord RM, Sheehan NA, Tobias JH, Timpson NJ, Davey Smith G, Sterne JA. Using multiple genetic variants as instrumental variables for modifiable risk factors. *Stat Methods Med Res.* 2012;21:223–242. doi: 10.1177/0962280210394459
13. Nikpay M, Goel A, Won HH, Hall LM, Willenborg C, Kanoni S, Saleheen D, Kyriakou T, Nelson CP, Hopewell JC, et al. A comprehensive 1,000 Genomes-based genome-wide association meta-analysis of coronary artery disease. *Nat Genet.* 2015;47:1121–1130. doi: 10.1038/ng.3396
14. Malik R, Chauhan G, Traylor M, Sargurupremraj M, Okada Y, Mishra A, Rutten-Jacobs L, Giese AK, van der Laan SW, Gretarsdottir S, et al; AFGen Consortium; Cohorts for Heart and Aging Research in Genomic Epidemiology (CHARGE) Consortium; International Genomics of Blood Pressure (iGEN-BP) Consortium; INVENT Consortium; STARNET; BioBank Japan

Cooperative Hospital Group; COMPASS Consortium; EPIC-CVD Consortium; EPIC-InterAct Consortium; International Stroke Genetics Consortium (ISGC); METASTROKE Consortium; Neurology Working Group of the CHARGE Consortium; NINDS Stroke Genetics Network (SiGN); UK Young Lacunar DNA Study; MEGASTROKE Consortium; MEGASTROKE Consortium:. Multiancestry genome-wide association study of 520,000 subjects identifies 32 loci associated with stroke and stroke subtypes. *Nat Genet.* 2018;50:524–537. doi: 10.1038/s41588-018-0058-3

15. Staley JR, Blackshaw J, Kamat MA, Ellis S, Surendran P, Sun BB, Paul DS, Freitag D, Burgess S, Danesh J, et al. PhenoScanner: a database of human genotype-phenotype associations. *Bioinformatics.* 2016;32:3207–3209. doi: 10.1093/bioinformatics/btw373
16. Burgess S, Bowden J, Fall T, Ingelsson E, Thompson SG. Sensitivity analyses for robust causal inference from Mendelian randomization analyses with multiple genetic variants. *Epidemiology.* 2017;28:30–42. doi: 10.1097/EDE.0000000000000559
17. Sudlow C, Gallacher J, Allen N, Beral V, Burton P, Danesh J, Downey P, Elliott P, Green J, Landray M, et al. UK Biobank: an open access resource for identifying the causes of a wide range of complex diseases of middle and old age. *PLoS Med.* 2015;12:e1001779. doi: 10.1371/journal.pmed.1001779
18. Purcell S, Neale B, Todd-Brown K, Thomas L, Ferreira MA, Bender D, Maller J, Sklar P, de Bakker PI, Daly MJ, et al. PLINK: a tool set for whole-genome association and population-based linkage analyses. *Am J Hum Genet.* 2007;81:559–575. doi: 10.1086/519795
19. Li X, Meng X, Spiliopoulou A, Timofeeva M, Wei WQ, Gifford A, Shen X, He Y, Varley T, McKeigue P, et al. MR-PheWAS: exploring the causal effect of SUA level on multiple disease outcomes by using genetic instruments in UK Biobank. *Ann Rheum Dis.* 2018;77:1039–1047. doi: 10.1136/annrheumdis-2017-212534
20. Verma A, Bradford Y, Dudek S, Lucas AM, Verma SS, Pendergrass SA, Ritchie MD. A simulation study investigating power estimates in phenome-wide association studies. *BMC Bioinformatics.* 2018;19:120. doi: 10.1186/s12859-018-2135-0
21. Denny JC, Bastarache L, Ritchie MD, Carroll RJ, Zink R, Mosley JD, Field JR, Pulley JM, Ramirez AH, Bowton E, et al. Systematic comparison of phenome-wide association study of electronic medical record data and genome-wide association study data. *Nat Biotechnol.* 2013;31:1102–1110. doi: 10.1038/nbt.2749
22. Frishman WH. Calcium channel blockers: differences between subclasses. *Am J Cardiovasc Drugs.* 2007;7(suppl 1):17–23. doi: 10.2165/00129784-200707001-00003
23. Morris CR, Harvey IM, Stebbings WS, Speakman CT, Kennedy HJ, Hart AR. Do calcium channel blockers and antimuscarinics protect against perforated colonic diverticular disease? A case control study. *Gut.* 2003;52:1734–1737. doi: 10.1136/gut.52.12.1734
24. Brian West A. The pathology of diverticulosis: classical concepts and mucosal changes in diverticula. *J Clin Gastroenterol.* 2006;40 Suppl 3:S126–S131. doi: 10.1097/01.mcg.0000225508.90417.07
25. Everhart JE, Ruhl CE. Burden of digestive diseases in the United States part II: lower gastrointestinal diseases. *Gastroenterology.* 2009;136:741–754. doi: 10.1053/j.gastro.2009.01.015
26. Etzioni DA, Mack TM, Beart RW Jr, Kaiser AM. Diverticulitis in the United States: 1998-2005: changing patterns of disease and treatment. *Ann Surg.* 2009;249:210–217. doi: 10.1097/SLA.0b013e3181952888

MANUSCRIPT V: Genetically determined blood pressure, antihypertensive drug classes, and risk of stroke subtypes

Georgakis MK*, Gill D*, Webb AJS, Evangelou E, Elliott P, Sudlow CLM, Dehghan A, Malik R, Tzoulaki I†, Dichgans D†. Genetically determined blood pressure, antihypertensive drug classes, and risk of stroke subtypes. [Accepted in ***Neurology***]. *,† equally contributed

Author contributions: MKG, DG, RM, IT, and MD conceptualized and designed the study. MKG and DG performed the statistical analysis. All authors interpreted the results. MKG and MD drafted the manuscript. All authors critically revised the manuscript for intellectual content. All authors approved the submitted version and are accountable for the integrity of the work.

Genetically determined blood pressure, antihypertensive drug classes and risk of stroke subtypes

Marios K. Georgakis MD [1,2] *, Dipender Gill MD [3] *, Alastair J. S. Webb DPhil [4], Evangelos Evangelou PhD [3,5], Paul Elliott PhD [6,7,8], Cathie L. M. Sudlow DPhil [3,9], Abbas Dehghan MD PhD [3,6], Rainer Malik PhD [1], Ioanna Tzoulaki PhD [3,5,10] †, Martin Dichgans MD [1,11,12] †,#

[1] Institute for Stroke and Dementia Research (ISD), University Hospital, Ludwig-Maximilians-Universität LMU, Munich, Germany.
[2] Graduate School for Systemic Neurosciences, Ludwig-Maximilians-Universität LMU, Munich, Germany
[3] Department of Biostatistics and Epidemiology, School of Public Health, Imperial College London, London, United Kingdom.
[4] Centre for Prevention of Stroke and Dementia, Department of Clinical Neurosciences, University of Oxford, Oxford, United Kingdom.
[5] Department of Hygiene and Epidemiology, University of Ioannina Medical School, Ioannina, Greece.
[6] UK Dementia Research Institute, Imperial College London, London, United Kingdom.
[7] National Institute for Health Research Imperial College Biomedical Research Centre, London, United Kingdom.
[8] Health Data Research-UK London at Imperial College London, London, United Kingdom.
[9] Institute for Genetics and Molecular Medicine, University of Edinburgh, Edinburgh, United Kingdom.
[10] MRC-PHE Centre for Environment, School of Public Health, Imperial College London, London, United Kingdom.
[11] Munich Cluster for Systems Neurology (SyNergy), Munich, Germany.
[12] German Centre for Neurodegenerative Diseases (DZNE), Munich, Germany.

* these authors contributed equally to the manuscript
† these authors contributed equally to the manuscript

Corresponding Author:

Martin Dichgans, MD
Institute for Stroke and Dementia Research, University Hospital
Ludwig-Maximilians-University (LMU)
Feodor-Lynen-Str. 17, 81377 Munich, Germany
Phone: +49-89-4400-46018; Fax: +49-89-4400-46040
E-mail: martin.dichgans@med.uni-muenchen.de

Statistical analyses were conducted by Marios K. Georgakis, MD, Institute for Stroke and Dementia Research, University Hospital, Ludwig-Maximilians-University (LMU) and Dipender Gill, MD, Department of Biostatistics and Epidemiology, School of Public Health, Imperial College.

Supplemental Data: Online Supplement includes more detailed methods (eMethods) and 6 Tables (e-1 to e-6). Data available from dryad, doi: https://doi.org/10.5061/dryad.dfn2z34wj.

Word count:

Title character count: 96
Word count abstract: 244
Word count article: 3,006
Number of references: 53
Number of tables: 1

Number of figures: 4

Search terms: [2] All Cerebrovascular disease/Stroke, [7] Intracerebral hemorrhage, [91] All Genetics, [59] Risk factors in epidemiology, Genetically determined blood pressure, Mendelian randomization

Study funding: M. Georgakis is funded by scholarships from the German Academic Exchange Service (DAAD) and Onassis Foundation. D. Gill is funded by the Wellcome Trust. P. Elliott acknowledges support from the Medical Research Council and Public Health England (MR/L01341X/1) for the MRC-PHE Centre for Environment and Health. P. Elliott is supported by the UK Dementia Research Institute which receives its funding from UK DRI Ltd funded by the UK Medical Research Council, Alzheimer's Society and Alzheimer's Research UK; and the National Institute of Health Research Imperial College Biomedical Research Centre. P. Elliott is associate director of the Health Data Research UK London funded by a consortium led by the UK Medical Research Council. This project has received funding from the European Union's Horizon 2020 research and innovation programme (No 666881), SVDs@target (to M. Dichgans) and No 667375, CoSTREAM (to M. Dichgans); the DFG as part of the Munich Cluster for Systems Neurology (EXC 1010 SyNergy) and the CRC 1123 (B3) (to M. Dichgans); the Corona Foundation (to M. Dichgans); the Fondation Leducq (Transatlantic Network of Excellence on the Pathogenesis of Small Vessel Disease of the Brain)(to M. Dichgans); the e:Med program (e:AtheroSysMed) (to M. Dichgans) and the FP7/2007-2103 European Union project CVgenes@target (grant agreement number Health-F2-2013-601456) (to M. Dichgans).

Role of the funder/sponsor: The funding organizations had no role in the design and conduct of the study; collection, management, analysis, and interpretation of the data; preparation, review, or approval of the manuscript; and decision to submit the manuscript for publication.

Acknowledgements: This research has been conducted using the UK Biobank Resource (UK Biobank application 2532). We acknowledge the contributions by the MEGASTROKE Consortium, Woo et al. for the ICH GWAS meta-analysis, the ICBP Consortium, the CHARGE Consortium, the AFGen Consortium, and the Neale lab for performing GWAS analyses in the UK Biobank data. MEGASTROKE has received funding from the sources detailed at http://www.megastroke.org/acknowledgments.html. All MEGASTROKE authors and their affiliations are available at http://www.megastroke.org/authors.html.

Author Disclosures:

Dr. Georgakis reports no disclosures.
Dr. Gill reports no disclosures.
Dr. Webb reports no disclosures.
Dr. Evangelou reports no disclosures.
Dr. Elliott reports no disclosures.
Dr. Sudlow reports no disclosures.
Dr. Dehghan reports no disclosures.
Dr. Malik reports no disclosures.
Dr. Tzoulaki reports no disclosures.
Dr. Dichgans reports no disclosures.

ABSTRACT

Objective: We employed Mendelian Randomization to explore whether the effects of blood pressure (BP) and BP lowering through different antihypertensive drug classes on stroke risk vary by stroke etiology.

Methods: We selected genetic variants associated with systolic and diastolic BP and BP-lowering variants in genes encoding antihypertensive drug targets from a GWAS on 757,601 individuals. Applying two-sample Mendelian randomization, we examined associations with any stroke (67,162 cases; 454,450 controls), ischemic stroke and its subtypes (large artery, cardioembolic, small vessel stroke), intracerebral hemorrhage (ICH, deep and lobar), and the related small vessel disease phenotype of WMH.

Results: Genetic predisposition to higher systolic and diastolic BP was associated with higher risk of any stroke, ischemic stroke, and ICH. We found associations between genetically determined BP and all ischemic stroke subtypes with a higher risk of large artery and small vessel stroke compared to cardioembolic stroke, as well as associations with deep, but not lobar ICH. Genetic proxies for calcium channel blockers, but not beta blockers, were associated with lower risk of any stroke and ischemic stroke. Proxies for CCBs showed particularly strong associations with small vessel stroke and the related radiological phenotype of WMH.

Conclusions: This study supports a causal role of hypertension in all major stroke subtypes except lobar ICH. We find differences in the effects of BP and BP lowering through antihypertensive drug classes between stroke subtypes and identify calcium channel blockade as a promising strategy for preventing manifestations of cerebral small vessel disease.

INTRODUCTION

Stroke ranks among the leading causes of death and disability worldwide.[1,2] High blood pressure (BP) is the major risk factor for both ischemic and hemorrhagic stroke, accounting for ~50% of the population attributable risk worldwide.[3-6] BP lowering reduces stroke risk with known differences between antihypertensive drug classes.[7,8] Randomized-controlled trials (RCTs) found calcium channel blockers (CCBs) to be superior to other drug classes, and specifically beta-blockers (BB), in lowering stroke risk.[7,9,10] However, it remains unknown whether the effects of BP or BP lowering through specific drug classes vary between stroke etiologies. In light of largely variable mechanisms between large artery stroke (LAS), cardioembolic stroke (CES), small vessel stroke (SVS), and deep and lobar intracerebral hemorrhage (ICH),[11,12] differences seem possible and might have relevance for therapeutic decisions.

Mendelian Randomization (MR) uses genetic variants as proxies for traits of interest and is by design less prone to confounding and reverse causation than observational studies.[13] As such, MR has been proven valuable in exploring causality and in predicting the effects of interventions,[13-17] as we recently showed for the effects of antihypertensive drugs on vascular outcomes.[18] The large samples in genome-wide association studies (GWAS) further permit exploration of outcomes for which there are no adequate data from RCTs, as is the case for BP lowering and stroke subtypes. Here, leveraging genetic data on BP[19] and stroke[20] we employed MR to examine the effects of genetically determined BP and genetic proxies for antihypertensive drug classes on stroke subtypes, as well as on white matter hyperintensities (WMH), a radiological manifestation of small vessel disease (SVD).

METHODS

This study was conducted in accordance with the Guidelines for strengthening the reporting of Mendelian randomization studies (STROBE-MR).[21]

Genetic instrument selection

Data sources are detailed in **Table 1**. All data were derived from studies that had already obtained ethical review board approvals. We used summary statistics from the discovery GWAS meta-analysis of the International Consortium for Blood Pressure (ICBP) and the UK Biobank (UKB), based on 757,601 individuals of European ancestry.[19] In the pooled sample, mean systolic (SBP) and diastolic BP (DBP) were 138.4 (SD: 21.5) and 82.8 (SD: 11.4) mmHg, respectively. As genetic instruments for SBP and DBP, we selected single-nucleotide polymorphisms (SNPs) associated with SBP or DBP at genome-wide significance level ($p<5x10^{-8}$) and clumped for linkage disequilibrium (LD) to $r^2<0.001$ based on the European 1000 Genomes panel. We estimated the proportion of variance in SBP and DBP explained by each instrument[22] and calculated F-statistics to measure instrument strength (data available from dryad, **Tables e-1** and **e-2**, doi: https://doi.org/10.5061/dryad.dfn2z34wj).[23]

We further selected genetic variants as proxies for the SBP-lowering effects of common antihypertensive drug classes (**Figure 1)**. According to our previously described strategy,[18] we identified the genes encoding pharmacological targets related to BP-lowering for common antihypertensive drug classes in DrugBank[24] and screened the genomic regions corresponding to these genes and their regulatory regions (promoters and enhancers).[25] For the main analyses, we selected SNPs associated with SBP at genome-wide significance ($p<5x10^{-8}$) that were at moderate to low LD ($r^2<0.4$) according to previously described approaches,[26-28] with sensitivity

analyses using a more stringent LD threshold (r^2<0.1) (data available from dryad, **Table e-3**, doi: https://doi.org/10.5061/dryad.dfn2z34wj). The genes and the specific genomic regions screened for identification of genetic proxies for each antihypertensive drug class are detailed in **Table e-4 (**data available from dryad, doi: https://doi.org/10.5061/dryad.dfn2z34wj).

Primary outcomes and etiologically related phenotypes

The primary outcomes for our analyses were any stroke, ischemic stroke and its TOAST-defined subtypes (LAS, CES, SVS),[29] ICH and its location-specific subtypes, i.e. lobar (originating at cerebral cortex or cortical-subcortical junction) and deep (originating at thalamus, internal capsule, basal ganglia, deep periventricular white matter, cerebellum, or brainstem).[30] Genetic association estimates for any stroke, ischemic stroke and its subtypes were obtained from the MEGASTROKE multi-ethnic GWAS meta-analysis of 67,162 cases (60,341 ischemic stroke, 6,688 LAS, 9,006 CES, 11,710 SVS) and 454,450 controls.[20,31] For ICH, we used the summary statistics from the ISGC meta-analysis by Woo *et al.* including 1,545 cases (664 lobar, 881 deep) and 1,481 controls.[30] In addition, we performed MR analyses for the radiological phenotype of WMH volume, a manifestation of cerebral SVD etiologically related to SVS and ICH. We performed a genome-wide association study (GWAS) analysis for total volume of WMH, derived from T1 and T2-FLAIR images in the UK Biobank data following a previously described approach,[32] as detailed in **eMethods (**data available from dryad, doi: https://doi.org/10.5061/dryad.dfn2z34wj).

Statistical analysis

For SBP and DBP, we calculated individual MR estimates and standard errors from the SNP-exposure and SNP-outcome associations using the Wald estimator and the Delta method;

second-order weights were used.[33] The MR associations for SBP and DBP with the primary outcomes were estimated by pooling individual MR estimates using fixed-effects inverse-variance weighted (IVW) meta-analyses.[33] All MR associations between SBP, DBP, and stroke were scaled to 10 mmHg increment in SBP and 5 mmHg in DBP.

For the antihypertensive drug classes, including instruments at moderate to low LD ($r^2<0.4$), we applied generalized linear regression analyses weighted for the correlation between the instruments, as previously described.[26] This relatively lenient LD correlation threshold allows for an increase in proportion of variance explained and thus in statistical power.[26,27] In sensitivity analyses we restricted our instrument selection to a lower LD correlation threshold ($r^2<0.1$) and applied fixed-effects IVW. All MR associations between antihypertensive drug classes and stroke were scaled to 10 mmHg decrease in SBP.

MR analyses might be biased due to pleiotropic instruments. As measures of pleiotropy, we assessed heterogeneity across MR estimates with I^2 and the Cochran's Q test ($I^2>50\%$ and $p<0.05$ were considered statistically significant)[34] and the intercept obtained from MR-Egger regression ($p<0.05$ considered statistically significant).[35] We further used alternative methods (weighted-median estimator,[36] MR-Egger,[35] weighted-modal estimator[37]) with relaxing assumptions regarding pleiotropic variants. The weighted median estimator requires that at least half of the information for the MR analysis comes from valid instruments.[36] MR-Egger regression requires that the strengths of potential pleiotropic instruments are independent of their direct associations with the outcome.[35] The weighted modal estimator provides correct estimates under the assumption that a plurality of genetic variants are valid instruments.[37] We further tested for the presence of pleiotropic outlier variants using the Mendelian randomization pleiotropy residual sum and outlier (MR- PRESSO) test[38] and in sensitivity IVW MR analyses excluded these variants.

The genetic association estimates used in the analyses for BP were corrected for antihypertensive medication use and were adjusted for body mass index,[19] thus introducing potential bias due to medication non-compliance or collider effects, respectively. Thus, we performed sensitivity analyses using unadjusted estimates for BP from a UK Biobank GWAS (317,756 individuals).[39] To minimize ancestral mismatch with the European population used in the BP GWAS, in sensitivity analyses we further restricted our MR analyses for stroke to the MEGASTROKE European subset.

Statistical significance for all analyses was set at a two-sided p-value <0.05. To examine whether BP differentially associated with stroke subtypes or whether there were differential effects of antihypertensive drugs on stroke risk, we compared the derived ORs by computing z-score for the differences of their natural logarithms. All statistical analyses were undertaken in R (v3.5.0; The R Foundation for Statistical Computing) using the MendelianRandomization, TwoSampleMR, and the MRPRESSO packages.

Data availability statement

This study was based on summary statistics. The GWAS data from the ICBP and UKB meta-analysis are publicly available through the GRASP repository of the National Heart, Lung, and Blood Institute (https://grasp.nhlbi.nih.gov/FullResults.aspx). The data from the GWAS studies for stroke and ICH are publicly available and may be accessed through the MEGASTROKE (http://www.megastroke.org/download.html) and the ISGC (http://cerebrovascularportal.org/informational/downloads) websites, respectively. Data from the UK Biobank GWAS for WMH volume may be accessed through an application to the UK Biobank. The summary data for the genetic instruments used for the purposes of the current

study are available in the Online Supplement (data available from dryad, **Tables e-1** to **e-3**, doi: https://doi.org/10.5061/dryad.dfn2z34wj).

RESULTS

Genetically determined BP and risk of stroke subtypes

We first examined the relationship between genetically determined BP and the risk of stroke and stroke subtypes. We identified 462 genetic variants associated with SBP and 460 variants associated with DBP. F-statistic was >10 for all variants indicating low risk of weak instrument bias (data available from dryad, **Tables e-1** and **e-2**, doi: https://doi.org/10.5061/dryad.dfn2z34wj). MR analyses showed statistically significant associations of both SBP and DBP with risk of any stroke, ischemic stroke and all of its major subtypes (LAS, CES, SVS), ICH, and deep ICH, but not lobar ICH (**Figure 2**). The effects of genetically determined BP were larger for LAS and SVS compared to CES (p for LAS-CES comparisons of ORs=$2x10^{-8}$ for SBP and 0.004 for DBP; p for SVS-CES comparisons of ORs =0.001 for SBP and $9x10^{-4}$ for DBP), and for deep compared to lobar ICH (p for comparisons of ORs =0.016 for SBP and 0.009 for DBP), as depicted in **Figure 2**.

The effect estimates remained stable in the weighted median, MR-Egger, and weighted-modal analyses, analyses excluding outliers detected with MR-PRESSO, European-restricted analyses, and analyses based on unadjusted BP estimates (data available from dryad, **Table e-5**, doi: https://doi.org/10.5061/dryad.dfn2z34wj). Tests for heterogeneity and the MR-Egger intercepts were not significant in any of the analyses (I^2<50% and p>0.05, respectively) providing no evidence for pleiotropy.

Genetic proxies for antihypertensive drugs and risk of stroke subtypes

Next, we selected BP-lowering variants in genes encoding drug targets as proxies for the effects of antihypertensive drug classes, as detailed in **Figure 1** and as has been previously described,[18] and examined their effects on stroke in MR analyses. We identified 8 proxies (variants) for BBs and 60 proxies for CCBs (data available from dryad, **Table e-3**, doi: https://doi.org/10.5061/dryad.dfn2z34wj). We further identified a single proxy for ACE inhibitors, which we did not consider in the following analyses given the lack of power. A 10-mmHg reduction in SBP through variants in genes encoding targets of CCBs, but not BBs, was associated with a significantly lower risk of any stroke and ischemic stroke (**Figure 3**). In analyses for ischemic stroke subtypes, we found a 10-mmHg reduction in SBP through CCB variants to be associated with significantly lower risks of LAS, CES, and SVS. The effect for SVS was stronger than that for both LAS (p for comparison of ORs=0.002) and CES (p for comparison of ORs=$6x10^{-4}$) (**Figure 3**). BB variants were not associated with any of the ischemic stroke subtypes. We found no significant associations for any of the drug classes for ICH and its subtypes, which is probably related to limited power (data available from dryad, **Table e-6**, doi: https://doi.org/10.5061/dryad.dfn2z34wj).

Sensitivity analyses for BBs and CCBs restricted to the set of variants with a more stringent LD threshold (r^2<0.1) showed consistent association estimates with the primary analyses for all of the examined phenotypes (data available from dryad, **Table e-6**, doi: https://doi.org/10.5061/dryad.dfn2z34wj). For CCBs, we found no evidence for pleiotropy (heterogeneity: I^2<50%; p of MR-Egger intercepts>0.05). There was heterogeneity in the associations of BBs with any stroke (I^2=59%), ischemic stroke (I^2=67%), and SVS (I^2=66%), which was however attenuated following exclusion of 2 outlier SNPs in MR-PRESSO (I^2=0%, following exclusion of outlier SNPs), while the association estimates remained stable (data available from dryad, **Table e-6**, doi: https://doi.org/10.5061/dryad.dfn2z34wj). The results

remained consistent across the alternative MR methods (data available from dryad, **Table e-6**, doi: https://doi.org/10.5061/dryad.dfn2z34wj).

Genetically determined BP and WMH volume

To gain additional insight in the relationship between genetically determined BP and cerebral SVD, we next calculated MR estimates for the associations of BP with WMH volume. We found genetically elevated SBP and DBP to be significantly associated with higher WMH volume (**Figure 4A**). Examining the effects of genetic proxies for antihypertensive drug classes (**Figure 4B**), we found significant associations of CCBs with lower WMH volume (β=-0.491, 95%CI=-0.591 to -0.391, $p=3.5x10^{-7}$), whereas proxies for BBs were not associated with WMH volume. The results were consistent across sensitivity analyses (data available from dryad, **Table e-5**, doi: https://doi.org/10.5061/dryad.dfn2z34wj).

DISCUSSION

We investigated the relationship between the leading modifiable risk factor for stroke and etiologically defined stroke subtypes by leveraging large-scale genetic data. We found genetic predisposition to higher BP to be associated with greater risk of any stroke, ischemic stroke, each of its main subtypes, and deep but not lobar ICH. Risk was higher for LAS and SVS compared to CES. Using genetic proxies for different antihypertensive drug classes we found BP-lowering through CCBs, but not BBs to be associated with lower risk of stroke and ischemic stroke. CCB variants were associated with a lower risk of all major ischemic stroke subtypes showing particularly strong effects on SVS and the related phenotype of WMH.

Our study provides evidence for a causal effect of higher BP on LAS, CES, and SVS, thus demonstrating a broad involvement of BP in the pathogenesis of ischemic stroke. Of note, however, we found the effects on stroke risk to vary depending on stroke mechanisms.

Specifically, risk was more pronounced for LAS and SVS than for CES and was restricted to deep ICH. Unlike deep ICH, lobar ICH is often related to cerebral amyloid angiopathy and the absence of an association signal between BP and lobar ICH is consistent with observational data.[40,41] As demonstrated by our drug target analyses, the effects of specific antihypertensive drug classes also differed according to stroke subtype. Collectively, these data emphasize the need to consider stroke etiologies when studying the effects of BP on stroke risk in observational and interventional studies.

Among the major findings is a benefit of BP lowering through genetic proxies for CCBs over BBs for SVS and the related phenotype of WMH. In contrast, we found no disparity in effects between genetic proxies for CCBs and BBs for LAS and CES. This suggests that CCBs may be particularly effective in preventing manifestations of cerebral small vessel disease. The mechanisms underlying this observation are currently unknown but may include direct effects of CCBs on cerebral microvessels or systemic effects for instance from the established influence of CCBs on BP variability.[9,10,42]

Patients with cerebral small vessel disease mark a population at increased risk for stroke, dementia and death.[43] Small vessel disease manifestations are highly prevalent in the ageing population with figures reaching up to 90% in patients aged 65 years and above.[44] Yet, there have been no informative trials on specific antihypertensive agents for the prevention of SVS, WMH or other manifestations of small vessel disease.[45-47] Our MR results suggest that BP lowering with CCBs should be tested in clinical trials for prevention of SVS and other outcomes related to small vessel disease.

The consistency of our results for stroke obtained from genetic proxies for different drug classes with those from previous RCTs[7,9,10] is worth noting and lends confidence to our findings on etiological stroke subtypes for which no data from RCTs exist. The disparity in treatment effects between CCBs and BBs on stroke risk has been related to the opposite actions of these drugs on

BP variability; CCBs decrease whereas BBs increase BP variability.[9,10] However, whether the effects of BP variability on stroke risk vary by stroke etiology is unresolved and deserves further investigation.

Our study has several methodological strengths. We used large datasets offering sufficient statistical power for most analyses and applied multiple methods to exclude pleiotropic effects and other biases. We also examined phenotypes etiologically related to stroke subtypes and performed mediation analyses that allowed inferences on mechanistic aspects regarding the association of BP with stroke. Finally, we used genetic proxies for antihypertensive drug classes that have been previously validated and have shown comparable effects to those derived from RCTs.[18]

Our study also has limitations. First, MR examines the lifetime effects of genetically determined BP, which might differ from the effect of a clinical intervention for BP lowering. Second, based on our selection criteria we identified only a single genetic proxy for ACE inhibitors that did not offer sufficient statistical power to perform meaningful analyses. Future studies encompassing larger GWAS datasets for BP might identify such variants and might thus offer deeper insights into differential effects between different classes of BP-lowering agents including ACE inhibitors, angiotensin-receptor blockers, and thiazide diuretics on stroke and stroke subtypes. Third, by design, we could not examine non-linear associations between BP and stroke risk.[48] However, current evidence suggests that the association of mid-life SBP and DBP with stroke seems to follow a linear pattern.[49] Fourth, our results apply stroke incidence and not stroke recurrence. While we found high BP to not be associated with risk of lobar ICH, hypertension has been shown in observational studies to increase the risk for both deep and lobar ICH recurrence,[50] which could not be examined in the context of the current study. Fifth, the small sample size for the ICH GWAS did not offer sufficient power to examine the effects of antihypertensive drug classes on any, lobar, and deep ICH. Sixth, our GWAS data for BP were

restricted to individuals of European ancestry which could limit generalizability of our findings to this population. This might specifically apply for ICH [30] given the evidence from observational studies for differential associations of BP with lobar ICH depending on ethnicity.[51] Furthermore, there is evidence for differential responses to antihypertensive drug classes by ethnicity, which could not be examined in the current study.[52] The availability of large-scale GWAS data from more diverse populations with higher representation of non-European ethnicities will enable future MR studies to explore potential ethnic disparities in more detail. Finally, it was not possible to disentangle the effects of dihydropyridine and non-dihydropyridine CCBs with MR, because the differences in the subunits of the voltage-gated calcium channels that are the targets of these drug subclasses in the vessels and the heart, respectively, are encoded by the same genes but are the result of alternative splicing.[53]

In conclusion, we provide evidence for a causal association of higher BP with risk of any stroke and all stroke subtypes except lobar ICH, with a higher risk of large artery and small vessel stroke compared to cardioembolic stroke. Our findings support CCBs, but not BBs, to lower ischemic stroke risk. Genetic proxies for the effects of CCBs showed particularly strong associations with SVS and WMH, highlighting calcium channel blockade as a promising strategy for the prevention of cerebral small vessel disease.

Appendix 1. Authors.

Name	Location	Role	Contribution
Marios K. Georgakis, MD	LMU Munich, Germany	Author	Concept and design; data acquisition, analysis, and interpretation of data; statistical analysis; drafting of the manuscript; critical revision of the manuscript for intellectual content
Dipender Gill MD	Imperial College London, UK	Author	Concept and design; data acquisition, analysis, and interpretation of data; statistical analysis; critical revision of the manuscript for intellectual content
Alastair J. S. Webb DPhil	University of Oxford, UK	Author	Data acquisition, analysis, and interpretation of data; critical revision of the manuscript for intellectual content
Evangelos Evangelou PhD	University of Ioannina, Greece	Author	Data acquisition, analysis, and interpretation of data; critical revision of the manuscript for intellectual content
Paul Elliott PhD	Imperial College London, UK	Author	Data acquisition, analysis, and interpretation of data; critical revision of the manuscript for intellectual content
Cathie L. M. Sudlow DPhil	University of Edinburgh, UK	Author	Data acquisition, analysis, and interpretation of data; critical revision of the manuscript for intellectual content
Abbas Dehghan MD	Imperial College London, UK	Author	Data acquisition, analysis, and interpretation of data; critical revision of the manuscript for intellectual content
Rainer Malik PhD	LMU Munich, Germany	Author	Concept and design; data acquisition, analysis, and interpretation of data; statistical analysis; critical revision of the manuscript for intellectual content
Ioanna Tzoulaki PhD	Imperial College London, UK	Author	Concept and design; data acquisition, analysis, and interpretation of data; statistical analysis; critical revision of the manuscript for intellectual content
Martin Dichgans, MD	LMU Munich, Germany	Author	Concept and design; data acquisition, analysis, and interpretation of data; drafting of the manuscript; critical revision of the manuscript for intellectual content

REFERENCES

1. DALYs GBD, Collaborators H. Global, regional, and national disability-adjusted life-years (DALYs) for 315 diseases and injuries and healthy life expectancy (HALE), 1990-2015: a systematic analysis for the Global Burden of Disease Study 2015. Lancet 2016;388:1603-1658.

2. Mortality GBD, Causes of Death C. Global, regional, and national life expectancy, all-cause mortality, and cause-specific mortality for 249 causes of death, 1980-2015: a systematic analysis for the Global Burden of Disease Study 2015. Lancet 2016;388:1459-1544.

3. O'Donnell MJ, Chin SL, Rangarajan S, et al. Global and regional effects of potentially modifiable risk factors associated with acute stroke in 32 countries (INTERSTROKE): a case-control study. Lancet 2016;388:761-775.

4. Feigin VL, Roth GA, Naghavi M, et al. Global burden of stroke and risk factors in 188 countries, during 1990-2013: a systematic analysis for the Global Burden of Disease Study 2013. Lancet Neurol 2016;15:913-924.

5. Lewington S, Clarke R, Qizilbash N, Peto R, Collins R, Prospective Studies C. Age-specific relevance of usual blood pressure to vascular mortality: a meta-analysis of individual data for one million adults in 61 prospective studies. Lancet 2002;360:1903-1913.

6. Lacey B, Lewington S, Clarke R, et al. Age-specific association between blood pressure and vascular and non-vascular chronic diseases in 0.5 million adults in China: a prospective cohort study. Lancet Glob Health 2018;6:e641-e649.

7. Ettehad D, Emdin CA, Kiran A, et al. Blood pressure lowering for prevention of cardiovascular disease and death: a systematic review and meta-analysis. Lancet 2016;387:957-967.

8. Xie X, Atkins E, Lv J, et al. Effects of intensive blood pressure lowering on cardiovascular and renal outcomes: updated systematic review and meta-analysis. Lancet 2016;387:435-443.

9. Rothwell PM, Howard SC, Dolan E, et al. Effects of beta blockers and calcium-channel blockers on within-individual variability in blood pressure and risk of stroke. Lancet Neurol 2010;9:469-480.

10. Webb AJ, Fischer U, Mehta Z, Rothwell PM. Effects of antihypertensive-drug class on interindividual variation in blood pressure and risk of stroke: a systematic review and meta-analysis. Lancet 2010;375:906-915.

11. Zia E, Hedblad B, Pessah-Rasmussen H, Berglund G, Janzon L, Engstrom G. Blood pressure in relation to the incidence of cerebral infarction and intracerebral hemorrhage. Hypertensive hemorrhage: debated nomenclature is still relevant. Stroke 2007;38:2681-2685.

12. Ohira T, Shahar E, Chambless LE, Rosamond WD, Mosley TH, Jr., Folsom AR. Risk factors for ischemic stroke subtypes: the Atherosclerosis Risk in Communities study. Stroke 2006;37:2493-2498.

13. Holmes MV, Ala-Korpela M, Smith GD. Mendelian randomization in cardiometabolic disease: challenges in evaluating causality. Nature reviews Cardiology 2017;14:577-590.

14. Roberts R. Mendelian Randomization Studies Promise to Shorten the Journey to FDA Approval. JACC Basic Transl Sci 2018;3:690-703.

15. Georgakis MK, Gill D, Rannikmae K, et al. Genetically Determined Levels of Circulating Cytokines and Risk of Stroke. Circulation 2019;139:256-268.

16. Ference BA, Robinson JG, Brook RD, et al. Variation in PCSK9 and HMGCR and Risk of Cardiovascular Disease and Diabetes. N Engl J Med 2016;375:2144-2153.

17. Ference BA, Kastelein JJP, Ginsberg HN, et al. Association of Genetic Variants Related to CETP Inhibitors and Statins With Lipoprotein Levels and Cardiovascular Risk. JAMA 2017;318:947-956.

18. Gill D, Georgakis MK, Koskeridis F, et al. Use of Genetic Variants Related to Antihypertensive Drugs to Inform on Efficacy and Side Effects. Circulation 2019;140:270-279.

19. Evangelou E, Warren HR, Mosen-Ansorena D, et al. Genetic analysis of over 1 million people identifies 535 new loci associated with blood pressure traits. Nat Genet 2018;50:1412-1425.

20. Malik R, Chauhan G, Traylor M, et al. Multiancestry genome-wide association study of 520,000 subjects identifies 32 loci associated with stroke and stroke subtypes. Nat Genet 2018;50:524-537.

21. Davey Smith G, Davies NM, Dimou N, et al. STROBE-MR: Guidelines for strengthening the reporting of Mendelian randomization studies. PeerJ Preprints 2019;7:e27857v27851. Available from: https://peerj.com/preprints/27857/#aff-1. [Accessed on Nov 18, 2019].

22. Shim H, Chasman DI, Smith JD, et al. A multivariate genome-wide association analysis of 10 LDL subfractions, and their response to statin treatment, in 1868 Caucasians. PLoS One 2015;10:e0120758.

23. Palmer TM, Lawlor DA, Harbord RM, et al. Using multiple genetic variants as instrumental variables for modifiable risk factors. Stat Methods Med Res 2012;21:223-242.

24. Wishart DS, Feunang YD, Guo AC, et al. DrugBank 5.0: a major update to the DrugBank database for 2018. Nucleic Acids Res 2018;46:D1074-D1082.

25. Fishilevich S, Nudel R, Rappaport N, et al. GeneHancer: genome-wide integration of enhancers and target genes in GeneCards. Database (Oxford) 2017;2017.

26. Burgess S, Dudbridge F, Thompson SG. Combining information on multiple instrumental variables in Mendelian randomization: comparison of allele score and summarized data methods. Stat Med 2016;35:1880-1906.

27. Burgess S, Ference BA, Staley JR, et al. Association of LPA Variants With Risk of Coronary Disease and the Implications for Lipoprotein(a)-Lowering Therapies: A Mendelian Randomization Analysis. JAMA Cardiol 2018;3:619-627.

28. Nowak C, Arnlov J. A Mendelian randomization study of the effects of blood lipids on breast cancer risk. Nat Commun 2018;9:3957.

29. Adams HP, Jr., Bendixen BH, Kappelle LJ, et al. Classification of subtype of acute ischemic stroke. Definitions for use in a multicenter clinical trial. TOAST. Trial of Org 10172 in Acute Stroke Treatment. Stroke 1993;24:35-41.

30. Woo D, Falcone GJ, Devan WJ, et al. Meta-analysis of genome-wide association studies identifies 1q22 as a susceptibility locus for intracerebral hemorrhage. Am J Hum Genet 2014;94:511-521.

31. Malik R, Rannikmae K, Traylor M, et al. Genome-wide meta-analysis identifies 3 novel loci associated with stroke. Ann Neurol 2018;84:934-939.

32. Rutten-Jacobs LCA, Tozer DJ, Duering M, et al. Genetic Study of White Matter Integrity in UK Biobank (N=8448) and the Overlap With Stroke, Depression, and Dementia. Stroke 2018;49:1340-1347.

33. Burgess S, Butterworth A, Thompson SG. Mendelian randomization analysis with multiple genetic variants using summarized data. Genet Epidemiol 2013;37:658-665.

34. Del Greco M F, Minelli C, Sheehan NA, Thompson JR. Detecting pleiotropy in Mendelian randomisation studies with summary data and a continuous outcome. Stat Med 2015;34:2926-2940.

35. Bowden J, Davey Smith G, Burgess S. Mendelian randomization with invalid instruments: effect estimation and bias detection through Egger regression. International journal of epidemiology 2015;44:512-525.

36. Bowden J, Davey Smith G, Haycock PC, Burgess S. Consistent estimation in Mendelian randomization with some invalid instruments using a weighted median estimator. Genet Epidemiol 2016;40:304-314.

37. Hartwig FP, Davey Smith G, Bowden J. Robust inference in summary data Mendelian randomization via the zero modal pleiotropy assumption. International journal of epidemiology 2017;46:1985-1998.

38. Verbanck M, Chen CY, Neale B, Do R. Detection of widespread horizontal pleiotropy in causal relationships inferred from Mendelian randomization between complex traits and diseases. Nat Genet 2018;50:693-698.

39. Flossmann E, Schulz UG, Rothwell PM. Systematic review of methods and results of studies of the genetic epidemiology of ischemic stroke. Stroke 2004;35:212-227.

40. Jackson CA, Sudlow CL. Is hypertension a more frequent risk factor for deep than for lobar supratentorial intracerebral haemorrhage? J Neurol Neurosurg Psychiatry 2006;77:1244-1252.

41. Martini SR, Flaherty ML, Brown WM, et al. Risk factors for intracerebral hemorrhage differ according to hemorrhage location. Neurology 2012;79:2275-2282.

42. Yamaguchi Y, Wada M, Sato H, et al. Impact of ambulatory blood pressure variability on cerebral small vessel disease progression and cognitive decline in community-based elderly Japanese. Am J Hypertens 2014;27:1257-1267.

43. Debette S, Schilling S, Duperron MG, Larsson SC, Markus HS. Clinical Significance of Magnetic Resonance Imaging Markers of Vascular Brain Injury: A Systematic Review and Meta-analysis. JAMA Neurol 2018.

44. de Leeuw FE, de Groot JC, Achten E, et al. Prevalence of cerebral white matter lesions in elderly people: a population based magnetic resonance imaging study. The Rotterdam Scan Study. J Neurol Neurosurg Psychiatry 2001;70:9-14.

45. Group SPSS, Benavente OR, Coffey CS, et al. Blood-pressure targets in patients with recent lacunar stroke: the SPS3 randomised trial. Lancet 2013;382:507-515.

46. Croall ID, Tozer DJ, Moynihan B, et al. Effect of Standard vs Intensive Blood Pressure Control on Cerebral Blood Flow in Small Vessel Disease: The PRESERVE Randomized Clinical Trial. JAMA Neurol 2018;75:720-727.

47. van Middelaar T, Argillander TE, Schreuder F, Deinum J, Richard E, Klijn CJM. Effect of Antihypertensive Medication on Cerebral Small Vessel Disease: A Systematic Review and Meta-Analysis. Stroke 2018;49:1531-1533.

48. Kannel WB, Wolf PA, McGee DL, Dawber TR, McNamara P, Castelli WP. Systolic blood pressure, arterial rigidity, and risk of stroke. The Framingham study. JAMA 1981;245:1225-1229.

49. Bangalore S, Messerli FH, Wun CC, et al. J-curve revisited: An analysis of blood pressure and cardiovascular events in the Treating to New Targets (TNT) Trial. Eur Heart J 2010;31:2897-2908.

50. Biffi A, Anderson CD, Battey TW, et al. Association Between Blood Pressure Control and Risk of Recurrent Intracerebral Hemorrhage. JAMA 2015;314:904-912.

51. Sawyer RP, Sekar P, Osborne J, et al. Racial/ethnic variation of APOE alleles for lobar intracerebral hemorrhage. Neurology 2018;91:e410-e420.

52. Preston RA, Materson BJ, Reda DJ, et al. Age-race subgroup compared with renin profile as predictors of blood pressure response to antihypertensive therapy. Department of Veterans Affairs Cooperative Study Group on Antihypertensive Agents. JAMA 1998;280:1168-1172.

53. Hu Z, Liang MC, Soong TW. Alternative Splicing of L-type CaV1.2 Calcium Channels: Implications in Cardiovascular Diseases. Genes (Basel) 2017;8.

Figure 1. Selection strategy for genetic variants used as proxies for antihypertensive drug classes. Shown are the steps for genetic instrument selection and the respective criteria and resources.

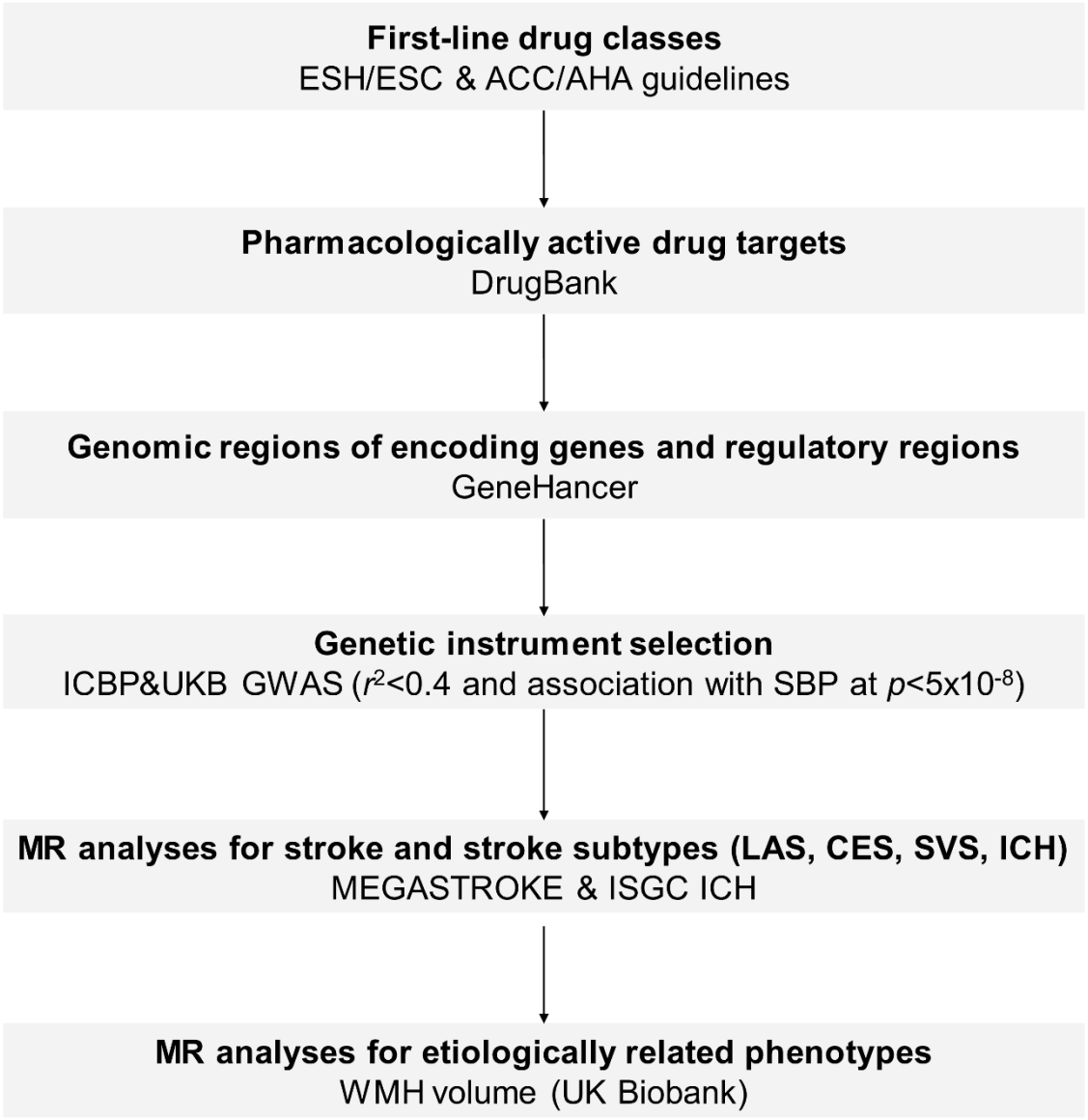


Figure 2. Mendelian randomization associations between genetically determined blood pressure and risk of stroke and stroke subtypes.

Shown are the results from the fixed-effects IVW analysis.

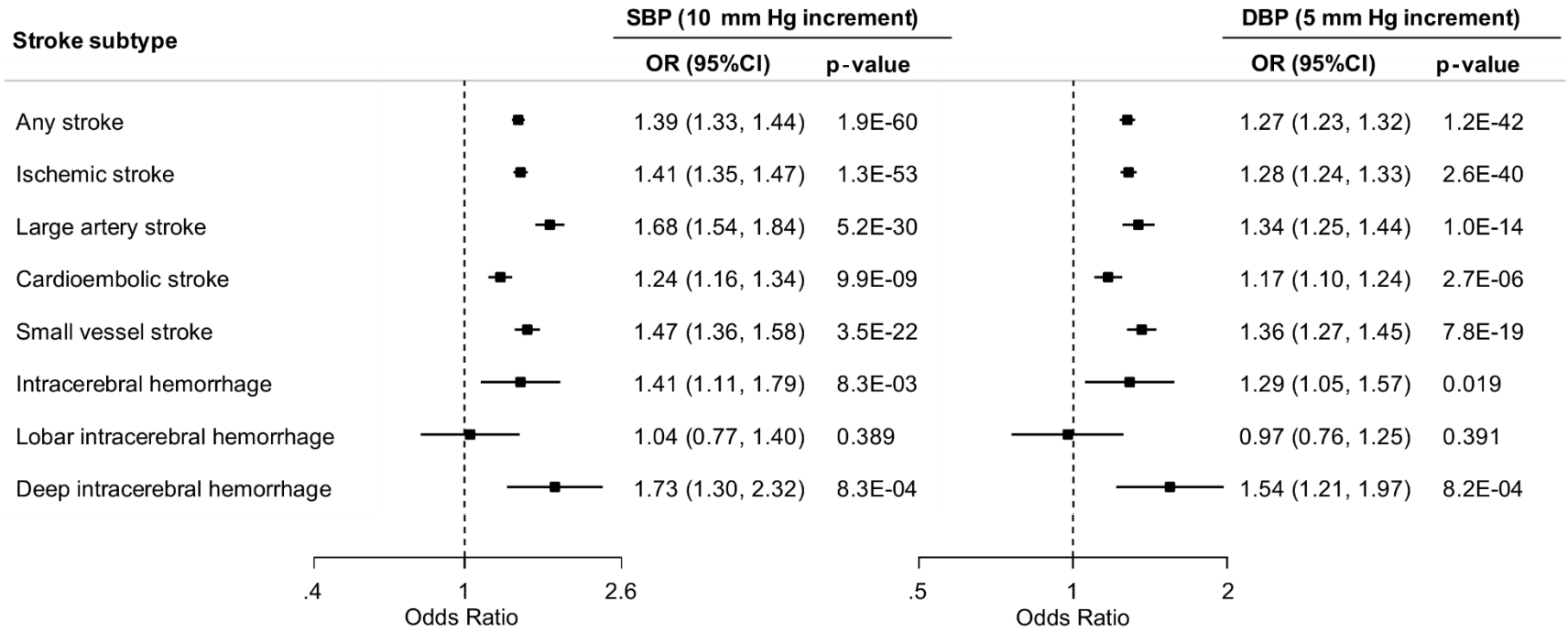


Figure 3. Mendelian randomization associations between genetic proxies for antihypertensive drug classes and risk of stroke and stroke subtypes. Shown are the results from the MR analysis adjusting for correlation between variants.

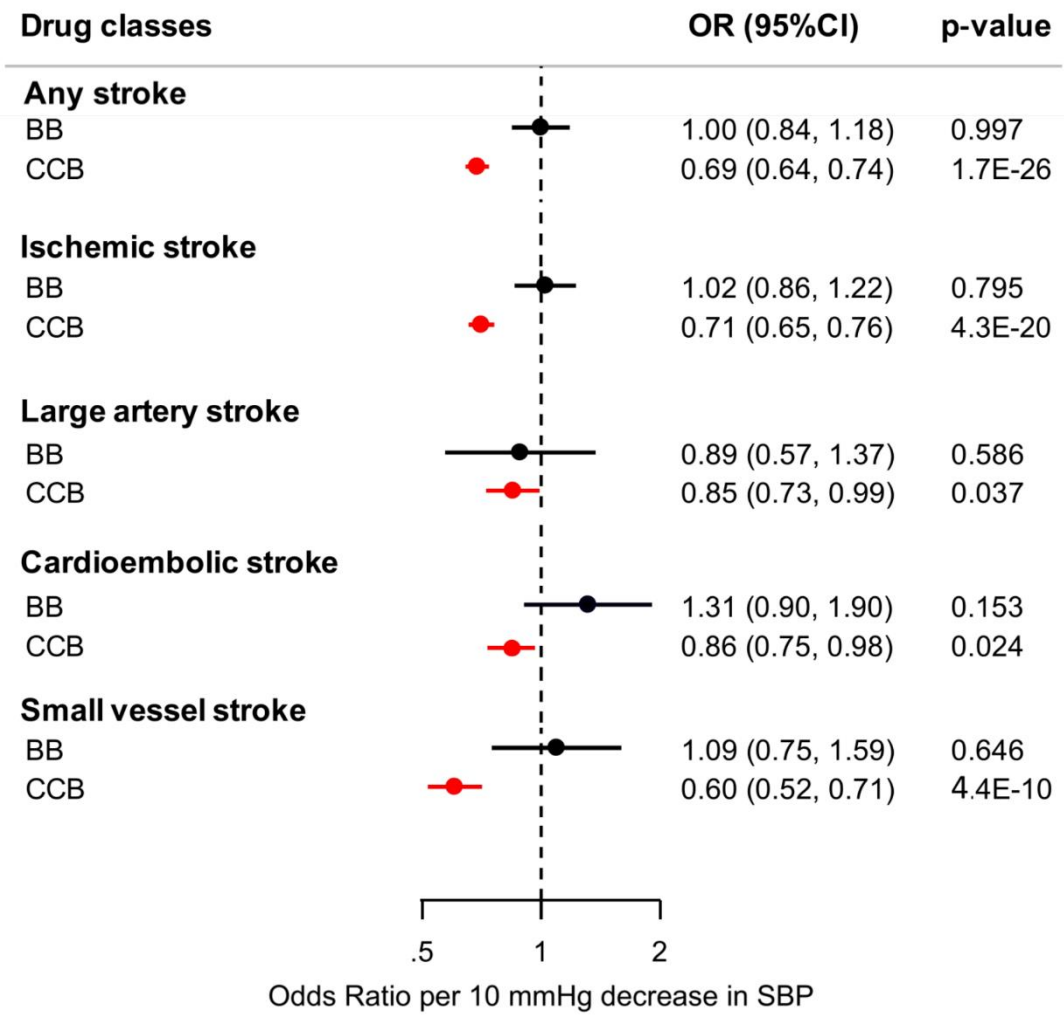


Figure 4. Mendelian randomization associations of (A) genetically determined blood pressure and (B) genetic proxies for antihypertensive drug classes with WMH volume. (A) Shown are the results from the fixed-effects IVW analysis. (B) Shown are the results from the MR analysis adjusting for correlation between variants.

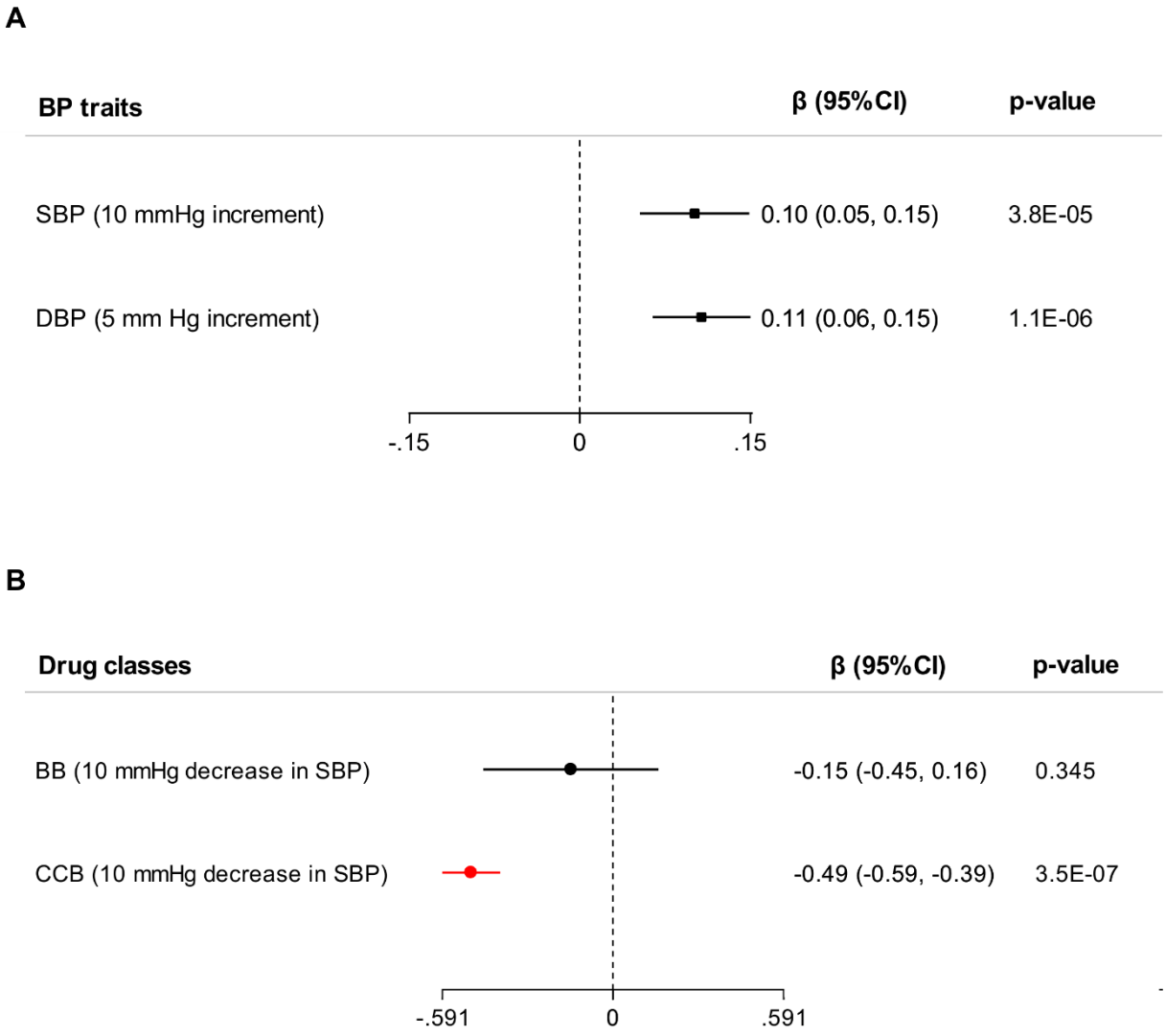


Table 1. Descriptive characteristics of the genome-wide association study (GWAS) meta-analyses that were included in this Mendelian randomization study.

Study stage	GWAS	Phenotype	Sample size	Ancestry	Adjustments[a]
Instrument selection	ICBP & UK Biobank[19]	SBP, DBP	757,601 individuals	European	age, sex, BMI
Use of instruments for sensitivity analysis	UK Biobank (Neale lab analysis)[39]	SBP, DBP	317,756 individuals	European	none
Primary outcome	MEGASTROKE[20]	Any stroke, IS and subtypes (LAS, CES, SVS)	67,162cases/ 454,450 controls	Multi-ancestry/ European	age, sex
Primary outcome	ISGC ICH GWAS[30]	ICH and subtypes (lobar, deep ICH)	1,545 cases/ 1,481 controls	European	age, sex
Etiologically related outcome	UK Biobank	WMH volume	10,597 individuals	European	age, sex

[a] All GWAS studies have further adjusted for principal components.

BMI, body mass index; CES, cardioembolic stroke; DBP, diastolic blood pressure; ICBP, International Consortium for Blood Pressure; ICH, intracerebral hemorrhage; LAS, large artery stroke; SBP, systolic blood pressure; SVS, small vessel stroke; WMH, white matter hyperintensities.

MANUSCRIPT VI: Genetically determined blood lipids and cerebral small vessel disease: role of HDL cholesterol

Georgakis MK, Malik R, Anderson CD, Parhofer KG, Hopewell JC, Dichgans M. Genetically determined blood lipids and cerebral small vessel disease: role of HDL cholesterol. **Brain.** 2020 Feb 1;143(2):597-610.

Authors contributions: MKG, RM, and MD conceptualized the study. MKG, RM, CDA, JCH, and MD designed the study. MKG and RM performed the statistical analysis. All authors interpreted the results. MKG and MD drafted the manuscript. All authors critically revised the manuscript for intellectual content. All authors approved the submitted version and are accountable for the integrity of the work.

doi:10.1093/brain/awz413 BRAIN 2020: 0; 1–14 | 1

BRAIN
A JOURNAL OF NEUROLOGY

Genetic determinants of blood lipids and cerebral small vessel disease: role of high-density lipoprotein cholesterol

Marios K. Georgakis,[1,2] Rainer Malik,[1] Christopher D. Anderson,[3,4,5,6] Klaus G. Parhofer,[7] Jemma C. Hopewell[8] and Martin Dichgans[1,9,10]

Blood lipids are causally involved in the pathogenesis of atherosclerosis, but their role in cerebral small vessel disease remains largely elusive. Here, we explored associations of genetic determinants of blood lipid levels, lipoprotein particle components, and targets for lipid-modifying drugs with small vessel disease phenotypes. We selected genetic instruments for blood levels of high-density lipoprotein cholesterol (HDL-C), low-density lipoprotein cholesterol (LDL-C), and triglycerides, for cholesterol and triglycerides components of size-defined lipoprotein particles, and for lipid-modifying drug targets based on published genome-wide association studies (up to 617 303 individuals). Applying two-sample Mendelian randomization approaches we investigated associations with ischaemic and haemorrhagic manifestations of small vessel disease [small vessel stroke: 11 710 cases, 287 067 controls; white matter hyperintensities (WMH): 10 597 individuals; intracerebral haemorrhage: 1545 cases, 1481 controls]. We applied the inverse-variance weighted method and multivariable Mendelian randomization as our main analytical approaches. Genetic predisposition to higher HDL-C levels was associated with lower risk of small vessel stroke [odds ratio (OR) per standard deviation = 0.85, 95% confidence interval (CI) = 0.78–0.92] and lower WMH volume (β = –0.07, 95% CI = –0.12 to –0.02), which in multivariable Mendelian randomization remained stable after adjustments for LDL-C and triglycerides. In analyses of lipoprotein particle components by size, we found these effects to be specific for cholesterol concentration in medium-sized high-density lipoprotein, and not large or extra-large high-density lipoprotein particles. Association estimates for intracerebral haemorrhage were negatively correlated with those for small vessel stroke and WMH volume across all lipid traits and lipoprotein particle components. HDL-C raising genetic variants in the gene locus of the target of CETP inhibitors were associated with lower risk of small vessel stroke (OR: 0.82, 95% CI = 0.75–0.89) and lower WMH volume (β = –0.08, 95% CI = –0.13 to –0.02), but a higher risk of intracerebral haemorrhage (OR: 1.64, 95% CI = 1.26–2.13). Genetic predisposition to higher HDL-C, specifically to cholesterol in medium-sized high-density lipoprotein particles, is associated with both a lower risk of small vessel stroke and lower WMH volume. These analyses indicate that HDL-C raising strategies could be considered for the prevention of ischaemic small vessel disease but the net benefit of such an approach would need to be tested in a randomized controlled trial.

1 Institute for Stroke and Dementia Research (ISD), University Hospital of Ludwig-Maximilians-University (LMU), Munich, Germany
2 Graduate School for Systemic Neurosciences (GSN), Ludwig-Maximilians-University (LMU), Munich, Germany
3 Center for Genomic Medicine, Massachusetts General Hospital, Boston, MA, USA
4 Division of Neurocritical Care and Emergency Neurology, Department of Neurology, Massachusetts General Hospital, Boston, MA, USA
5 Program in Medical and Population Genetics, Broad Institute, Cambridge, MA, USA
6 Henry and Allison McCance Center for Brain Health, Department of Neurology, Massachusetts General Hospital, Boston, MA, USA
7 Department of Internal Medicine IV, University Hospital of Ludwig-Maximilians-University (LMU), Munich, Germany

Received June 07, 2019. Revised October 26, 2019. Accepted November 19, 2019

8 Clinical Trial Service Unit and Epidemiological Studies Unit, Nuffield Department of Population Health, University of Oxford, Oxford, UK
9 Munich Cluster for Systems Neurology (SyNergy), Munich, Germany
10 German Centre for Neurodegenerative Diseases (DZNE), Munich, Germany

Correspondence to: Martin Dichgans, MD
Institute for Stroke and Dementia Research
University Hospital of Ludwig-Maximilians-University (LMU)
Feodor-Lynen-Str. 17, 81377 Munich, Germany
E-mail: martin.dichgans@med.uni-muenchen.de

Keywords: lipids; small vessel disease; Mendelian randomization; high-density lipoprotein; lacunar stroke

Abbreviations: GLGC = Global Lipids Genetics Consortium; GWAS = genome-wide association study; HDL-C = high-density lipoprotein cholesterol; ICH = intracerebral haemorrhage; LDL-C = low-density lipoprotein cholesterol; SVD = small vessel disease; SVS = small vessel stroke; WMH = white matter hyperintensities

Introduction

Cerebral small vessel disease (SVD) accounts for ~20% of all ischaemic strokes (Sudlow and Warlow, 1997) and most cases of intracerebral haemorrhage (ICH) (Qureshi *et al.*, 2001, 2009). SVD is the leading cause of vascular dementia (O'Brien and Thomas, 2015; Iadecola *et al.*, 2019) and an independent predictor of mortality (Debette *et al.*, 2019; Georgakis *et al.*, 2019). Manifestations of SVD on MRI are highly prevalent in the ageing population with figures reaching 90% for white matter hyperintensities (WMH) in patients aged 65 years and above (de Leeuw *et al.*, 2001; Pantoni, 2010; Wardlaw *et al.*, 2019). However, the mechanisms underlying SVD are poorly understood, thus impeding the development of effective strategies for prevention.

Blood lipids are a well-established risk factor for large artery atherosclerosis (Collins *et al.*, 2016) and lipid-modifying therapies have shown benefits in reducing risk of both coronary artery disease and stroke [Cholesterol Treatment Trialists' (CTT) Collaboration *et al.*, 2010; Chou *et al.*, 2016]. Yet, their role in SVD remains largely elusive. Current guidelines for secondary stroke prevention recommend treatment with statins after ischaemic stroke or transient ischaemic attack [European Stroke Organisation (ESO) Executive Committee and ESO Writing Committee, 2008; Kernan *et al.*, 2014; Intercollegiate Stroke Working Party, 2016; Stroke Foundation, 2017) referring to clinical trials data and meta-analyses (Amarenco *et al.*, 2006; Amarenco and Labreuche, 2009; Manktelow and Potter, 2009). However, most trials provided no sub-analyses for ischaemic stroke subtypes. The J-STARS trial, the only study providing sub-analyses, found statins to reduce recurrence of large artery stroke but not small vessel stroke (SVS) (Hosomi *et al.*, 2015). Results from the SPARCL trial suggest that statins may increase the risk of ICH in patients with stroke or transient ischaemic attack (Amarenco *et al.*, 2006), especially in patients with SVS as an entry event (Goldstein *et al.*, 2008).

Mendelian randomization makes use of genetic variants that are associated with an exposure or risk factor as instruments, and investigates their associations with disease outcomes thus overcoming some of the key limitations of observational studies such as confounding and reverse causation (Hopewell and Clarke, 2016; Holmes *et al.*, 2017). Hence, Mendelian randomization analyses can assess the causal relevance of a risk factor for disease and facilitate prioritization of interventions to be tested in clinical trials (Holmes *et al.*, 2017; O'Donnell and Sabatine, 2018) as has specifically been demonstrated for lipid-modifying drugs (Khera and Kathiresan, 2017; Ference *et al.*, 2018). In fact, there are several examples where Mendelian randomization studies have predicted the success or failure of clinical trials (Ference *et al.*, 2015, 2016, 2017*b*, 2019*b*; Gill *et al.*, 2019; Ray *et al.*, 2019). The availability of large scale genome-wide association studies (GWAS) for an expanding range of phenotypes and the development of two-sample Mendelian randomization approaches enable the exploration of associations for which there is a paucity of evidence from clinical trials, as is the case for lipids and cerebral SVD.

Here, we leveraged data from the largest GWAS currently available on blood lipid levels (617 303 individuals) (Willer *et al.*, 2013; Klarin *et al.*, 2018) and on both ischaemic (SVS, WMH volume) and haemorrhagic (ICH) manifestations of cerebral SVD (Woo *et al.*, 2014; Malik *et al.*, 2018*a*; Rutten-Jacobs *et al.*, 2018) with the aim to: (i) examine the effects of genetic determinants of blood levels of high-density lipoprotein cholesterol (HDL-C), low-density lipoprotein cholesterol (LDL-C), and triglycerides on SVD manifestations; (ii) explore associations between genetic determinants of size-defined lipoprotein particle fractions with these phenotypes; and (iii) determine the effects of genetic predisposition to HDL-C raising, LDL-C lowering, and triglyceride lowering through variants in genes encoding targets of lipid-modifying drugs on SVD manifestations.

Materials and methods

This study follows the guidelines for strengthening the reporting of Mendelian randomization studies (STROBE-MR)

(Davey Smith *et al.*, 2019). We applied two-sample Mendelian randomization analyses, which allow selection of genetic variants as instruments for a risk factor (blood lipid traits) in one sample and explore associations of theses variants with outcomes (manifestations of SVD) in another sample (Davey Smith and Hemani, 2014; Burgess *et al.*, 2015). By overcoming the requirement for assessing the exposure and outcome in the same dataset, this approach enables the exploration of associations in publicly available summary statistics from large GWASs with a corresponding increase in power. Also, two-sample Mendelian randomization is less prone to the winner's curse bias than one-sample Mendelian randomization (Davey Smith and Hemani, 2014; Taylor *et al.*, 2014).

Study design and data sources

The data sources used for this study are detailed in Supplementary Table 1. In Mendelian randomization analyses, we examined associations of blood lipid levels, size-defined lipoprotein particle fractions, and lipid-modifying drug targets, with ischaemic and haemorrhagic SVD phenotypes. We selected genetic instruments from the GWAS summary statistics of the Million Veteran Program (MVP) (Klarin *et al.*, 2018), the Global Lipids Genetics Consortium (GLGC) (Willer *et al.*, 2013), and from a GWAS on nuclear magnetic resonance (NMR) measured circulating metabolites (Kettunen *et al.*, 2016). We then examined associations of the selected instruments with SVS in the GWAS summary statistics of the MEGASTROKE Consortium (Malik *et al.*, 2018*a*), with WMH volume in a GWAS analysis that we undertook in the UK Biobank neuroimaging dataset (Alfaro-Almagro *et al.*, 2018), and with ICH in the International Stroke Genetics Consortium (ISGC) GWAS meta-analysis (Woo *et al.*, 2014).

Genetic instrument selection

Blood lipid levels

We selected genetic instruments for the blood levels of HDL-C, LDL-C, and triglycerides, based on the results of the GWAS multi-ethnic meta-analysis of the MVP and the GLGC samples (617 303 individuals) (Klarin *et al.*, 2018). Specifically, we used independent genetic variants that reached genome-wide level of significance ($P < 5 \times 10^{-8}$) for their associations with HDL-C, LDL-C and triglycerides, in the conditional GWAS meta-analyses as instruments. We identified 312 instruments for HDL-C, 219 for LDL-C, and 253 for triglycerides (Supplementary Table 2). In our primary analyses, we weighted the instruments based on the joint regression coefficients from the conditional GWAS meta-analysis of MVP and GLGC. As the GLGC further excluded participants on lipid-lowering treatment (Willer *et al.*, 2013), to exclude sources of biases related to treatment-mediated effects on blood lipids in the MVP dataset, we performed sensitivity analyses weighting the instruments using the GLGC effect sizes only. Both MVP and GLGC were imputed to the 1000 Genomes Project (Phase 3 and Phase 1, respectively) (1000 Genomes Project Consortium *et al.*, 2012) and included adjustments for age, age^2, sex, and population structure.

In a secondary approach, we restricted our selection of instruments to HDL-C-, LDL-C-, and triglyceride-specific variants. In particular, we used the GLGC dataset (188 577 individuals), for which we had access to the full GWAS summary statistics (Willer *et al.*, 2013), and identified those independent genetic variants associated with HDL-C, LDL-C, or triglycerides at genome-wide significance ($P < 5 \times 10^{-8}$), but showed associations of $P > 0.01$ with the other two traits. We found 19 HLD-C-specific, 25 LDL-C-specific, and four triglyceride-specific variants (Supplementary Table 3) and performed sensitivity analyses using them as instruments.

Size-defined lipoprotein particle fractions

We then selected genetic instruments for cholesterol and triglyceride concentrations in size-defined lipoprotein particles available from a GWAS for NMR-measured circulating metabolites on 24 925 European individuals (Kettunen *et al.*, 2016). The GWAS analyses were imputed to the 1000 Genomes Project (Phase 1) and adjusted for age, sex, time from last meal, and population structure (Kettunen *et al.*, 2016). Based on summary statistics for each trait, we extracted variants after clumping for linkage disequilibrium (LD) at $r^2 < 0.1$ that reached genome-wide significance ($P < 5 \times 10^{-8}$). The identified instruments for each metabolite are available in Supplementary Table 4.

Variants in genes encoding known lipid-modifying drug targets

Next, we selected variants clumped for linkage disequilibrium at $r^2 < 0.1$ within a region of 100 kb upstream or downstream from genes encoding known drug targets that were associated with the respective lipid trait at a genome-wide significant level ($P < 5 \times 10^{-8}$) in the GLGC dataset (Willer *et al.*, 2013). Specifically, we searched for genetic variants in the *CETP* locus (encoding the target of CETP inhibitors) associated with HDL-C levels; variants in the loci of *HMGCR* (target of statins), *NPC1L1* (target of ezetimibe), *PCSK9* (target of PCSK9 inhibitors), *ABCG5* and *ABCG8* (targets of bile acid resins), and *LDLR* (therapeutic target of the LDL receptor) associated with LDL-C levels; and variants in the *PPARA* locus (target for fibrates) associated with triglyceride levels, in accordance with similar approaches applied by other studies (Ference *et al.*, 2012, 2015, 2017*b*, 2019*a*; Anderson *et al.*, 2016; Harrison *et al.*, 2018; Nowak and Arnlov, 2018). We identified 24 HDL-C raising variants in *CETP*, and for LDL-C lowering targets, four variants in *HMGCR*, three in *NPC1L1*, 11 in *PCSK9*, six in *ABCG5/G8*, and eight in *LDLR* (Supplementary Table 5). No triglyceride-lowering variants were identified in the *PPARA* locus based on our selection criteria for instruments.

For each genetic instrument, we estimated the proportion of variance explained for the respective phenotype and measured instrument strength with *F*-statistics (Supplementary Tables 2–5). *F* was >10 for all selected instruments, indicating a low probability for weak instrument bias (Palmer *et al.*, 2012). Furthermore, we performed power calculations (Burgess, 2014) to identify the range of association estimates that we had >80% power (1 − β) to detect at $\alpha = 0.05$ (Supplementary Table 6).

Associations with outcomes

The outcomes examined in this study were ischaemic and haemorrhagic manifestations of SVD including SVS, WMH

volume, and ICH. Genetic association estimates for SVS—defined according to the TOAST (Trial of Org 10172 in Acute Stroke Treatment) criteria (Adams *et al.*, 1993)—were obtained from the MEGASTROKE multi-ethnic GWAS meta-analysis (Malik *et al.*, 2018*a*, *b*) on 11 710 cases and 287 067 controls. For WMH volume, we performed a GWAS analysis in the UK Biobank Imaging dataset (10 597 individuals of White-British ancestry), based on the measurements of WMH volume in T_1 and T_2 FLAIR MRI sequences, as previously described, following adjustments for age, sex, and the first 10 principal components (Rutten-Jacobs *et al.*, 2018). We further examined ICH, as well as ICH subtypes defined according to haemorrhage location (deep and lobar). We used summary statistics from the ISGC GWAS meta-analysis including 1545 cases of spontaneous ICH defined by acute neurological onset and compatible neuroimaging showing intraparenchymal haemorrhage (664 lobar, 881 deep) and 1481 controls of European ancestry (Woo *et al.*, 2014).

Statistical analysis

Main analyses

We applied two-sample Mendelian randomization analyses based on association estimates derived from the abovementioned sources. Following extraction of the association estimates between the instruments and the outcomes and harmonization of the direction of estimates by effect alleles, we computed Mendelian randomization estimates for each instrument with the Wald estimator and standard errors with the Delta method. All Mendelian randomization estimates were scaled to 1 – SD (standard deviation) increment in the lipid levels or the lipoprotein particle fractions. We then pooled individual Mendelian randomization estimates using random-effects inverse-variance weighted (IVW) meta-analyses. IVW is the most widely used main method for Mendelian randomization analysis because it provides robust causal estimates under absence of directional pleiotropy (Burgess *et al.*, 2013).

Given the correlation between HDL-C, LDL-C, and triglyceride levels, and between cholesterol and triglyceride concentrations in specific size-defined lipoprotein particles, we further performed multivariable Mendelian randomization to disentangle their independent associations with SVD phenotypes (Burgess and Thompson, 2015). For HDL-C, LDL-C, and triglyceride blood levels, we used the respective instruments and adjusted for their effects on the other two traits from the GLGC dataset. For cholesterol concentration in HDL particles, we combined all unique variants associated with either total HDL-C levels or size-defined HDL cholesterol concentration and adjusted for their effects on blood LDL-C and triglyceride levels. Similarly, for cholesterol concentration in LDL and larger particles, we combined all variants associated with either total LDL-C levels or size-defined LDL and larger particle cholesterol concentrations and adjusted for their effects on HDL-C and triglyceride levels. Finally, we combined instruments for either total circulating triglyceride levels or for particle-specific triglyceride concentrations and adjusted for their effects on HDL-C and LDL-C.

For all analyses, we corrected for multiple comparisons with the false discovery rate (FDR) approach and set statistical significance at a q-value < 0.05. Associations not reaching this threshold, but showing a $P < 0.05$, were considered suggestive of an association.

Assessment of pleiotropy and sensitivity analyses

The IVW method was our primary Mendelian randomization analysis approach, but the derived estimates might be biased in case of directional pleiotropy. As a measure of pleiotropy, we assessed heterogeneity across the Mendelian randomization estimates for each instrument in the IVW Mendelian randomization analyses with the Cochran's Q statistic (Bowden *et al.*, 2018). Under presence of nominal heterogeneity (P from Cochran's $Q < 0.10$) we further applied alternative Mendelian randomization methods, which are more robust to the use of pleiotropic instruments. These were the weighted median estimator and the Mendelian randomization (MR)-Egger regression. The weighted median estimator allows the use of invalid instruments under the assumption that at least half of the instruments used in the Mendelian randomization analysis are valid (Hartwig *et al.*, 2017). The MR-Egger regression allows for the estimation of an intercept term, which can be used as an indicator of unbalanced directional pleiotropy (Bowden *et al.*, 2015). MR-Egger provides less precise estimates and relies on the assumption that the strengths of potential pleiotropic instruments are independent of their direct associations with the outcome (Bowden *et al.*, 2015). The intercept obtained from MR-Egger regression was used as a measure of directional pleiotropy ($P < 0.05$ indicated statistical significance) (Bowden *et al.*, 2015).

In case of evidence of directional pleiotropy (as assessed by both the Cochran's Q statistic and the intercept in the MR-Egger regression) and inconsistent results between the different approaches, we further applied the generalized summary data-based Mendelian randomization (GSMR) approach. This method uses all variants reaching genome-wide significance as instruments by accounting for linkage disequilibrium correlation between them and further identifies and eliminates outliers that exert apparent pleiotropic effects on both the risk factor and the outcome using the HEIDI-outlier method (Zhu *et al.*, 2018). GSMR further provides a measure of remaining global heterogeneity following exclusion of outliers that also takes into account the low linkage disequilibrium across the used instruments.

All analyses were performed in R (v3.5.0; The R Foundation for Statistical Computing) using the MendelianRandomization and the gsmr packages.

Data availability

The data used for the current study are publicly available and may also become available from the corresponding author on reasonable request.

Results

Genetic determinants of blood lipid levels and ischaemic small vessel disease

The primary results of the IVW Mendelian randomization analyses for the associations between genetic determinants of blood lipid levels and SVS and WMH volume are presented in Fig. 1. Genetic predisposition to elevated HDL-C

levels were associated with both a lower risk of SVS [odds ratio (OR): 0.85, 95% CI: 0.78–0.92, $P = 5 \times 10^{-4}$] and lower WMH volume (β: –0.07, 95% CI: –0.12 to –0.02, $P = 0.004$). We further found genetic predisposition to higher triglyceride levels to be associated with higher risk of SVS and a suggestive association between genetic predisposition to higher LDL-C levels and SVS risk. In multivariable Mendelian randomization, the associations between genetic determinants of HDL-C levels and SVS and WMH volume remained stable and statistically significant (Fig. 1). In contrast, the association between genetic determinants of triglyceride levels and SVS was attenuated when adjusting for HDL-C and LDL-C.

The Mendelian randomization results were stable when weighting the genetic instruments for the three lipid traits based on their association estimates in the GLGC dataset, which excluded individuals on lipid-modifying treatment (Supplementary Figs 1 and 2). In Mendelian randomization analyses restricted to the instruments specifically associated with HDL-C, LDL-C, or triglycerides, the association estimates of genetic determinants of HDL-C for both risk of SVS (OR: 0.78, 95% CI: 0.62–0.98) and WMH volume (β: –0.27, 95% CI: –0.45 to –0.08) were even stronger (Supplementary Fig. 3). GSMR-HEIDI, which identifies and excludes pleiotropic outlier variants, also showed significant associations between genetic predisposition to higher HDL-C and both lower SVS risk and lower WMH volume (Supplementary Figs 1 and 2).

Genetic determinants of size-defined lipoprotein particle fractions and ischaemic small vessel disease

To obtain a deeper understanding of the observed associations, we next selected genetic instruments for cholesterol and triglyceride concentrations in size-defined lipoprotein particles and examined their associations with SVS and WMH volume (Fig. 2 and Supplementary Table 7). We found genetic predisposition to higher cholesterol concentration in the medium-sized, but not in the large- or extra-large sized HDL particles, to be associated with both lower SVS risk (OR: 0.84, 95% CI: 0.73–0.96, $P = 0.007$) and lower WMH volume (β: –0.09, 95% CI: –0.16 to –0.02, $P = 0.009$). There was no heterogeneity and the associations remained significant when adjusting for the effects of the instruments on circulating LDL-C and triglyceride levels (Fig. 2 and Supplementary Table 8).

Because of evidence for heterogeneity (Cochran's Q $P < 0.10$) and inconsistent results for the associations of genetic determinants of total HDL-C with SVS risk and WMH volume across sensitivity analyses (weighted median and MR-Egger) (Fig. 3 and Supplementary Figs 1 and 2), we next restricted the set of instruments for total HDL-C to those associated with medium-sized HDL-C ($P < 5 \times 10^{-8}$). These analyses revealed stronger association estimates between genetic predisposition to higher HDL-C and both lower risk of SVS (OR: 0.69, 95% CI: 0.56–0.84, $P = 4 \times 10^{-4}$) and lower WMH volume (β: –0.23, 95% CI: –0.35 to –0.10, $P = 2 \times 10^{-4}$) (Fig. 3). Moreover, the estimates were highly consistent in alternative Mendelian randomization approaches with no evidence for heterogeneity, thus suggesting that heterogeneity in the overall analyses was driven by non-medium sized HDL-C increasing variants.

To explore whether the observed associations were specific to genetic predisposition to higher cholesterol concentration in the medium-sized HDL, we next expanded our analyses to other components of the HDL particles (Supplementary Fig. 4). In this *post hoc* analysis, we found similar association estimates between genetic determinants of the concentration of any of the medium-sized HDL particle components (total cholesterol, cholesterol-esters, free cholesterol, total lipids, and phospholipids) and SVS risk as well as WMH volume suggesting that the associations are driven by the medium-sized HDL particles as a whole.

Regarding other lipoprotein particle components, we further found genetic predisposition to higher concentration of triglycerides in the small-sized HDL particles to be associated with higher risk of SVS (Fig. 2 and Supplementary Tables 7–9).

Genetic variants in loci of lipid-modifying drug targets and ischaemic small vessel disease

We next selected genetic variants in genes encoding known HDL-C-raising or LDL-C-lowering drug targets and examined their associations with ischaemic SVD phenotypes. HDL-C-raising variants in the *CETP* locus were associated with lower risk of SVS (OR: 0.82, 95% CI = 0.75–0.89, $P = 9 \times 10^{-6}$) and lower WMH volume (β = –0.08, 95% CI = –0.13 to –0.02, $P = 0.008$) (Figs 4, 5A and B). While there was heterogeneity in the association between *CETP* variants and SVS ($P = 0.03$), the results remained significant in the weighted median and MR-Egger approaches (Supplementary Table 10). As previous analyses from the REVEAL trial (HPS3/TIMI55–REVEAL Collaborative Group *et al.*, 2017) had shown the beneficial effects of cholesteryl-ester transfer protein (CETP) inhibitors on vascular disease to be mainly driven by their LDL-C lowering and not their HDL-C raising capacity, we further explored the associations between genetic predisposition to LDL-C lowering through *CETP* variants and ischaemic SVD manifestations. While genetic predisposition to LDL-C lowering was associated with lower risk of SVS and lower WMH volume in univariable IVW Mendelian randomization analyses, these effects were entirely reversed after adjusting for the HDL-C raising effects of the variants in multivariable Mendelian randomization (Supplementary Table 11). Analyses for genetic variants in LDL-C lowering drug target loci showed no statistically significant results (Fig. 4).

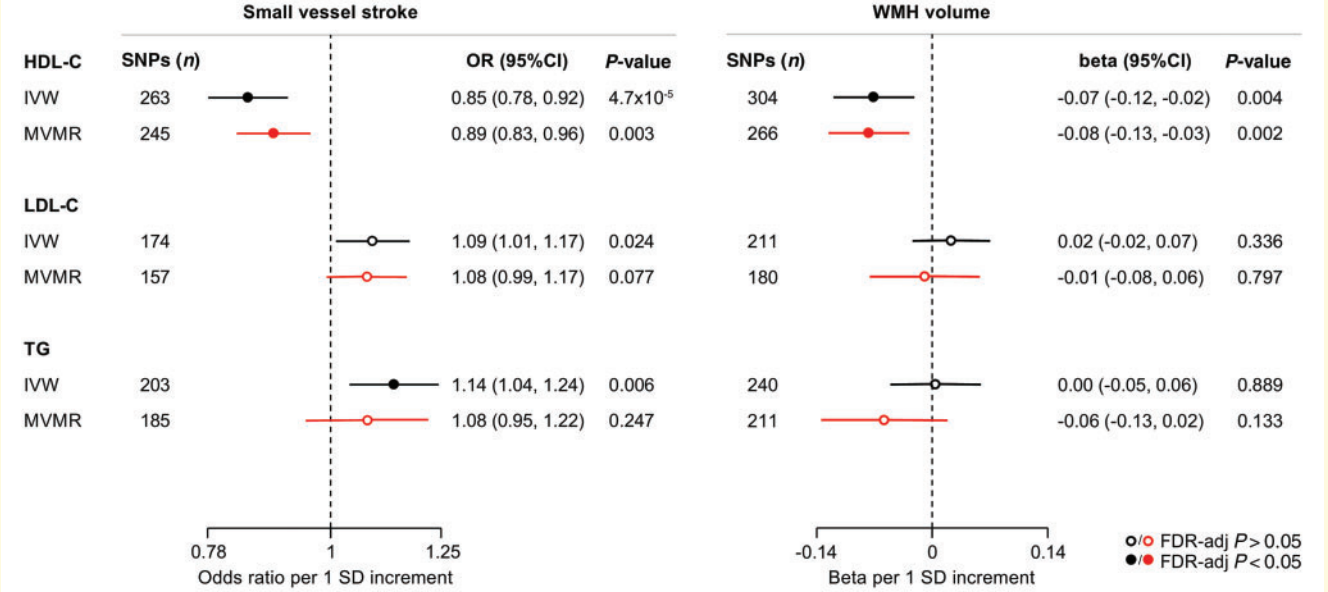


Figure 1 Mendelian randomization associations of genetic determinants of blood lipid levels (HDL-C, LDL-C, triglycerides) with risk of small vessel stroke and WMH volume. Shown are the results derived from random-effects IVW (inverse-variance weighted) Mendelian randomization and multivariable Mendelian randomization (MVMR) analyses adjusting for the effects of the genetic variants on all the three blood lipid traits. SD = standard deviation; TG = triglycerides.

Genetic associations of lipid traits with intracerebral haemorrhage

IVW-Mendelian randomization analyses showed no significant associations of genetic determinants of HDL-C, LDL-C, and triglycerides with risk of ICH (Fig. 6). When examining lipoprotein particle fractions, we found associations of the opposite direction, as compared to both SVS and WMH volume (Fig. 7). However, confidence intervals were wide, likely due to lack of statistical power (Supplementary Fig. 5 and Supplementary Tables 6 and 12). Across drug target loci (Supplementary Fig. 6) we found HDL-C raising variants in the *CETP* locus to be associated with a higher risk of ICH (OR: 1.64, 95% CI: 1.26–2.13, $P = 2.6 \times 10^{-4}$) (Fig. 5C). This effect was significant for both deep (OR: 2.01, 95% CI: 1.27–3.18, $P = 0.003$) and lobar ICH (OR: 1.78, 95% CI: 1.06–2.89, $P = 0.028$) (Supplementary Fig. 7).

Discussion

The main findings from this study can be summarized as follows: (i) we found significant associations between genetic predisposition to higher HDL-C levels and both lower risk of SVS and lower WMH volume; (ii) associations were specific for cholesterol concentrations in the medium and not large or extra-large sized HDL particles; (iii) exploring genetic variants at loci for targets of lipid-modifying drugs, we found HDL-C raising variants in *CETP* to be associated with a lower SVS risk and lower WMH volume; and (iv) we found these HDL-C raising variants in *CETP* to be associated with a higher risk of ICH, with consistent results for both lobar and deep ICH.

Our Mendelian randomization results provide evidence for a protective role of HDL-C on ischaemic SVD. This agrees with findings from two small observational studies. In the Women's Healthy Ageing Project, midlife HDL-C levels among 135 females were inversely associated with WMH volume after 20 years, independently of other vascular risk factors (Aljondi *et al.*, 2018). Similarly, in a cross-sectional study of 817 participants aged $\geqslant 50$ years, higher HDL-C levels were associated with lower volumes of both deep and periventricular WMH after adjusting for vascular risk factors (Yin *et al.*, 2018). The mechanisms underlying the observed inverse association between HDL-C levels and ischaemic SVD are unknown but may involve protective effects on the vascular endothelium (Sorrentino *et al.*, 2010; Prosser *et al.*, 2012; Tran-Dinh *et al.*, 2013; Monette *et al.*, 2016). Endothelial cells, including those of the brain microvasculature (Lapergue *et al.*, 2010; Fung *et al.*, 2017), express receptors, which upon HDL binding, induce intracellular signalling eventually leading to vasodilatory (Yuhanna *et al.*, 2001; Spieker *et al.*, 2002; Nofer *et al.*, 2004), anti-inflammatory (Cockerill *et al.*, 1995; Nicholls *et al.*, 2005; Murphy *et al.*, 2008), antioxidative (Garner *et al.*, 1998; Lee *et al.*, 2005; Terasaka *et al.*, 2007), and anti-thrombotic effects (Viswambharan *et al.*, 2004; Calkin *et al.*, 2009).

Our findings contrast with Mendelian randomization analyses on atherosclerotic phenotypes supporting no association of genetic determinants of HDL-C levels with

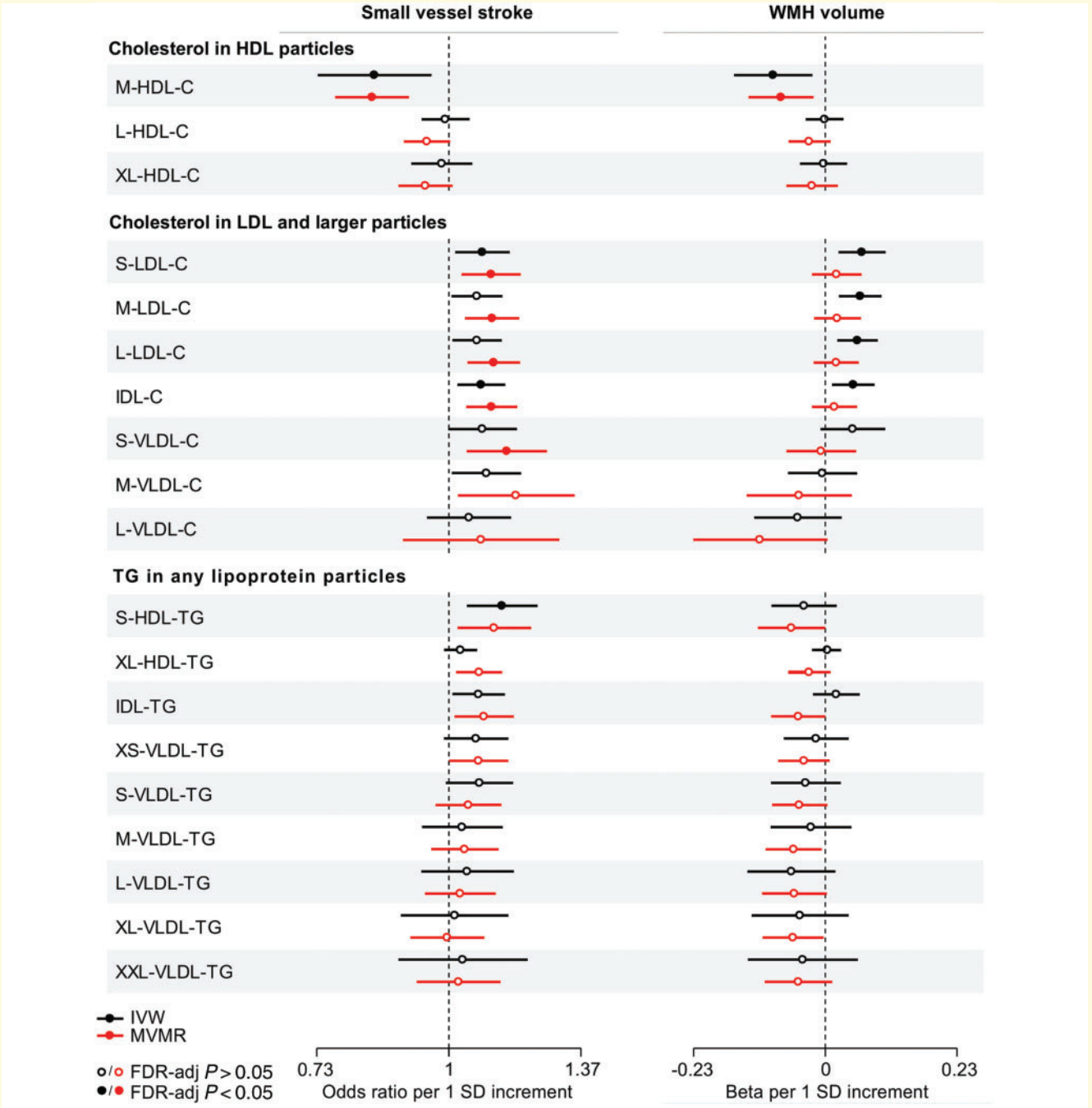


Figure 2 Mendelian randomization associations of genetic determinants of cholesterol (C) and triglyceride (TG) concentrations in size-defined lipoprotein particles with risk of small vessel stroke and WMH volume. Shown are the results derived from random-effects inverse-variance weighted (IVW) Mendelian randomization and multivariable Mendelian randomization (MVMR) analyses. MVMR for cholesterol in HDL particles adjusted for LDL-C and triglycerides; for cholesterol in LDL and larger particles adjusted for HDL-C and triglycerides; and for triglycerides in any particles adjusted for HDL-C and LDL-C. IDL = intermediate density lipoprotein; L = large; M = medium; S = small; VLDL = very low density lipoprotein; XL = extra-large.

coronary artery disease (Voight *et al.*, 2012; Holmes *et al.*, 2015; White *et al.*, 2016) and large artery stroke (Hindy *et al.*, 2018) thus suggesting differential effects of HDL-C on cerebral SVD and large artery atherosclerosis. A disparity in the effect of lipid levels between small and large vessel pathologies has also been reported for LDL-C: previous Mendelian randomization studies found strong effects of genetic predisposition to higher LDL-C on the risk of coronary artery disease, large artery stroke, and peripheral artery disease (Holmes *et al.*, 2015; Hindy *et al.*, 2018; Valdes-Marquez *et al.*, 2019; Emanuelsson *et al.*, 2019), but no effect on risk of retinopathy and neuropathy,

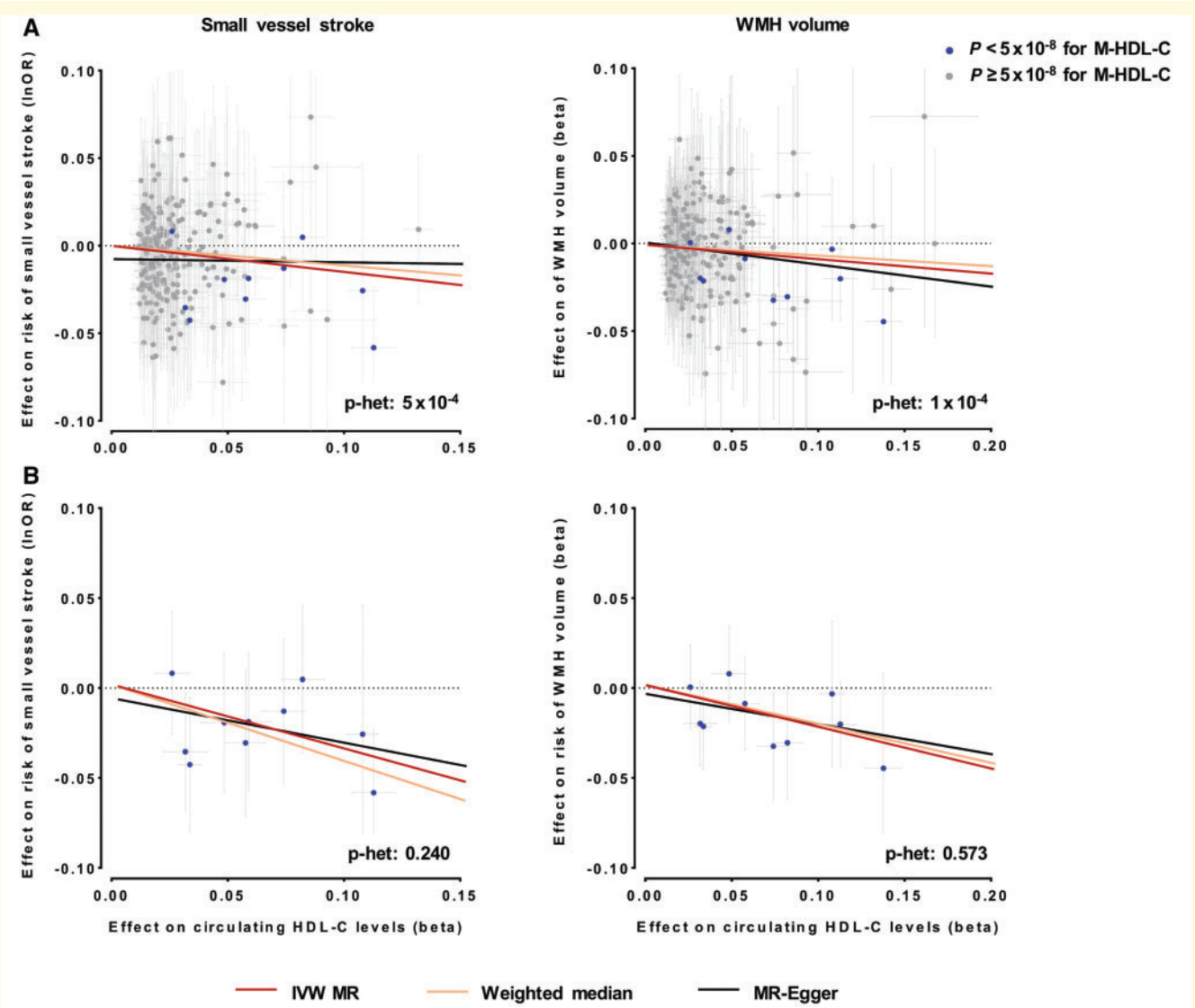


Figure 3 Mendelian randomization (MR) associations of genetic determinants of HDL-C with risk of small vessel stroke and WMH volume. Shown are the results from random-effects inverse-variance weighted (IVW), weighted median and MR-Egger analyses when (**A**) using the full set of genetic instruments and (**B**) restricting the analyses to instruments also associated with cholesterol concentration in medium-sized HDL.

which are typically related to small vessel pathology (Emanuelsson *et al.*, 2019). Future studies should explore potentially distinct mechanisms through which blood lipids influence the risk of small versus large vessel disease.

Analysing size-defined lipoprotein particle subfractions we found that the protective effects of HDL-C on ischaemic SVD are specific for medium-sized, and not larger HDL particles. In additional analyses, this effect seemed to be not specific to a particular component of the HDL particles but rather uniform across the different components, thus suggesting that medium-sized HDL particles as a whole could underlie this observation. HDL comprises a heterogeneous pool of lipoprotein particles (Kontush and Chapman, 2010) and the few observational studies that have performed analyses stratified by particle size indeed found differential effects on vascular outcomes (Martin *et al.*, 2015; Wurtz *et al.*, 2015; Joshi *et al.*, 2016; Holmes *et al.*, 2018). There are technical challenges related to different methods of HDL subfractioning (Superko *et al.*, 2012), making it challenging to compare our results with those from previous studies. Still, our results agree with the general notion that the favourable effects observed for HDL are predominantly exerted by the smaller and denser HDL particles (Yu *et al.*, 2003; Williams, 2012; Martin *et al.*, 2014). Of note, previous Mendelian randomization studies on blood lipids that showed no significant associations between HDL-C levels and atherosclerotic phenotypes did not consider particle subfractions (Holmes *et al.*, 2015; White *et al.*, 2016; Hindy *et al.*, 2018). Conceivably, disregarding subfractions might result in

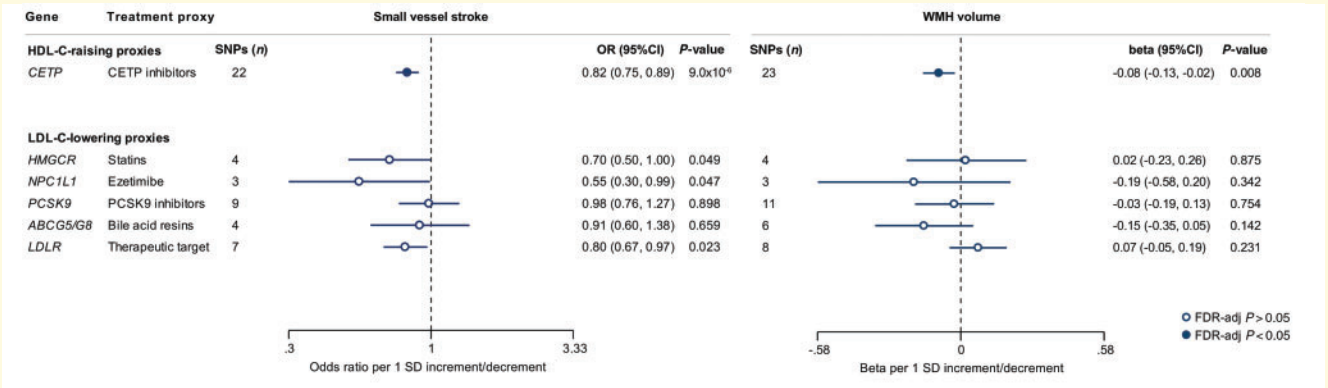


Figure 4 Mendelian randomization associations of HDL-C-raising and LDL-C-lowering genetic variants in the loci of known lipid-modifying drug targets with risk of SVS and WMH volume. Shown are the results derived from random-effects inverse-variance weighted (IVW) Mendelian randomization analyses. The results are scaled per 1 SD increment in circulating HDL-C levels (HDL-C-raising drug targets) and per 1 SD increment in circulating LDL-C levels (LDL-C-lowering drug targets).

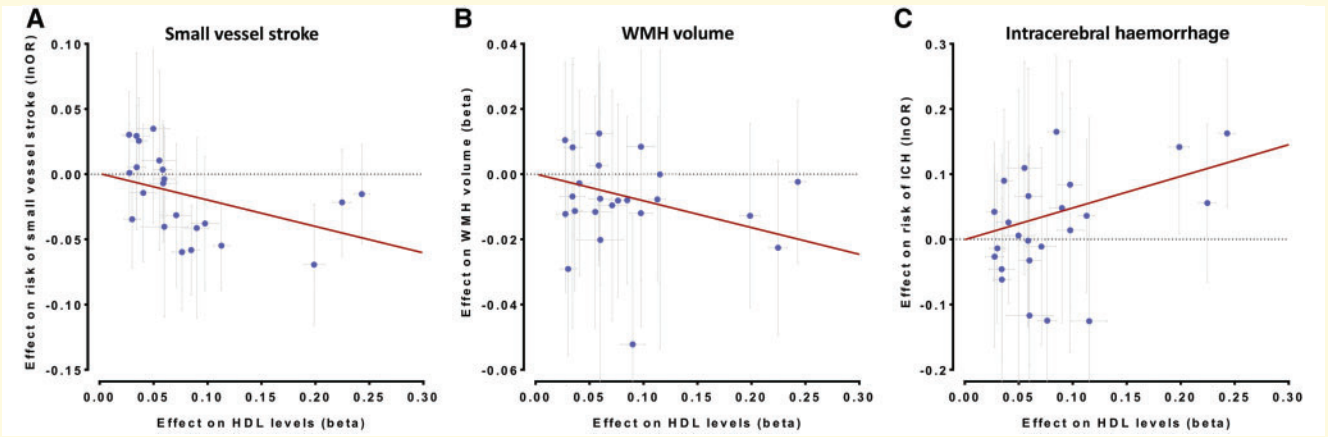


Figure 5 Mendelian randomization associations between HDL-C raising genetic variants in the *CETP* locus and (A) risk of SVS, (B) WMH volume, and (C) risk of ICH. Shown are the results from the random-effects inverse-variance weighted (IVW) Mendelian randomization approach. The results are scaled per 1 SD increment in circulating HDL-C levels.

masking causal effects of potential biological relevance. As such, our findings highlight the importance of sub-analyses stratifying by lipoprotein particle size, but the complexity of the potential underlying mechanisms necessitates further study of our observations.

Importantly, we found HDL-C raising genetic variants in the *CETP* locus to also associate with lower SVS risk and WMH volume. Pharmacological CETP inhibition leads to an increase in the circulating pool of HDL particles (Armitage *et al.*, 2019). While initial randomized trials investigating CETP inhibitors on top of statins found no benefit of CETP inhibition on vascular risk (Barter *et al.*, 2007; Schwartz *et al.*, 2012; Lincoff *et al.*, 2017), the most recent REVEAL trial showed a reduced risk for major coronary events (HPS3/TIMI55–REVEAL Collaborative Group *et al.*, 2017). In light of the relatively small effect (relative risk reduction in REVEAL: 9%) it seems unlikely that CETP inhibitors will achieve approval for prevention of cardiovascular disease (Hegele, 2017; Badimon, 2018). However, none of these trials explicitly reported effects on risk of SVS or other SVD manifestations. Our Mendelian randomization results suggest that *post hoc* analyses should consider stratifying for stroke subtypes, and that HDL-C raising approaches might show promise as a strategy for lowering the burden of ischaemic SVD.

The exact mechanism by which CETP inhibition might reduce risk of SVS and WMH volume is poorly understood. In the REVEAL trial, the reduction in vascular risk by CETP inhibition was mediated by a reduction in LDL-C rather than an increase in HDL-C (HPS3/TIMI55–REVEAL Collaborative Group *et al.*, 2017). In our analyses, most of the HDL-C raising *CETP* variants also showed strong associations with lower LDL-C levels. Yet, in multivariable Mendelian randomization analyses adjusting for the effects of the variants on both HDL-C and LDL-C, we found only the effects of genetic predisposition to higher HDL-C

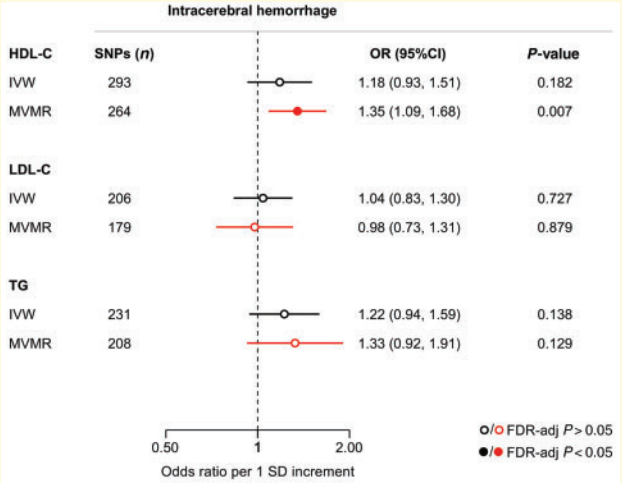


Figure 6 Mendelian randomization associations of genetic determinants of blood lipid levels (HDL-C, LDL-C, triglycerides) with risk of intracerebral haemorrhage. Shown are the results derived from random-effects inverse-variance weighted (IVW) Mendelian randomization and multivariable Mendelian randomization (MVMR) analyses. MVMR for cholesterol in HDL particles adjusted for LDL-C and triglycerides; for cholesterol in LDL and larger particles adjusted for HDL-C and triglycerides; and for triglycerides in any particles adjusted for HDL-C and LDL-C. TG = triglycerides.

through these *CETP* variants to remain consistent in terms of magnitude and directionality. Thus, although we were not sufficiently powered to entirely disentangle the effects of the two traits, our results suggest that in contrast with the REVEAL trial results, the effects of *CETP* variants on SVD manifestations might be primarily exerted by HDL-C raising. Administration of CETP inhibitors increases HDL particle size (Brousseau *et al.*, 2004) and genetic predisposition to higher CETP concentration is associated with increased concentrations of medium- and large-sized, but not smaller HDL particles (Blauw *et al.*, 2019). However, whether the expected effects of CETP inhibition on SVS and WMH volume are mediated through increases in the pool of specific HDL subparticles would need to be explored in future studies.

Previous observational and genetic studies found high HDL-C and low LDL-C levels to be associated with a higher risk of ICH (Wang *et al.*, 2013; Anderson *et al.*, 2016; Sun *et al.*, 2019). Also, clinical trials have shown that LDL-C lowering with statins might increase risk for ICH (Amarenco *et al.*, 2006; Goldstein *et al.*, 2008). We found HDL-C raising variants in the *CETP* locus to be associated with a higher risk of both deep and lobar ICH, which relate to different vascular pathologies. Specifically, deep ICH has been associated with hypertensive SVD, whereas lobar ICH is typically related to cerebral amyloid angiopathy (Martini *et al.*, 2012). While speculative, low serum LDL-C and high HDL-C levels may be associated with a fragile vascular endothelium, eventually leading to vessel permeability and a higher susceptibility to rupture (Konishi *et al.*, 1993).

The main analytical approaches used in the current study, IVW and multivariable Mendelian randomization, are sensitive to directional pleiotropy (Lawlor *et al.*, 2008; Burgess and Thompson, 2015). Specifically, if the single nucleotide polymorphisms used as genetic instruments for blood lipid levels associate with manifestations of SVD through pathways independent of blood lipid levels, the results could be biased. To ameliorate this risk, we performed a series of sensitivity analyses, which are based on statistical models that are more robust to pleiotropy, are focused on a subset of genetic instruments that are more specifically associated with the blood lipid traits under study, or excluded outlier single nucleotide polymorphisms with out of average effects on SVD manifestations, which are more likely to exert pleiotropic effects. Importantly, our results for an association between genetic determinants of HDL-C levels with risk of SVS and WMH volume were robust across these sensitivity analyses, thus supporting the results of the main analyses.

Our study has several strengths. The use of large genetic datasets enabled us to explore associations with a range of phenotypes, covering key manifestations of cerebral SVD. Also, the use of GWAS data for NMR-derived measurements enabled analyses stratified for lipoprotein particle subfractions. We further performed multiple tests for the detection of unbalanced pleiotropy and used multiple sensitivity analyses including advanced approaches such as GSMR-HEIDI. These analyses showed consistent results, thus minimizing the possibility of bias in the Mendelian randomization analyses. Finally, we explored the effects of HDL-C raising or LDL-C lowering genetic variants in genes encoding known lipid-modifying drug targets; this approach has previously been validated with the Mendelian randomization effects being comparable to those derived from randomized controlled trials.

Our study also has limitations. First, Mendelian randomization examines the lifetime effect of genetic determinants of blood lipid levels, which might differ from the effects of clinical lipid-modifying interventions. Second, we were not sufficiently powered to identify significant associations for ICH, and especially for ICH subtypes. Similarly, the non-significant, but still suggestive associations between LDL-C levels and SVS risk should be tested in larger datasets offering greater statistical power. Third, we had no access to the full summary statistics from the meta-analysis of the MVP and the GLGC studies. Hence, some analyses were restricted to the smaller GLGC dataset. Fourth, we are not aware of any sufficiently powered GWAS on cerebral microbleeds that would more accurately capture the spectrum of haemorrhagic SVD pathology than the currently used phenotype of ICH. While SVD is an important cause of ICH as a severe clinical manifestation, SVD more frequently manifests with subclinical cerebral microbleeds. Future GWAS on cerebral microbleeds will facilitate Mendelian randomization analyses on the relationship with

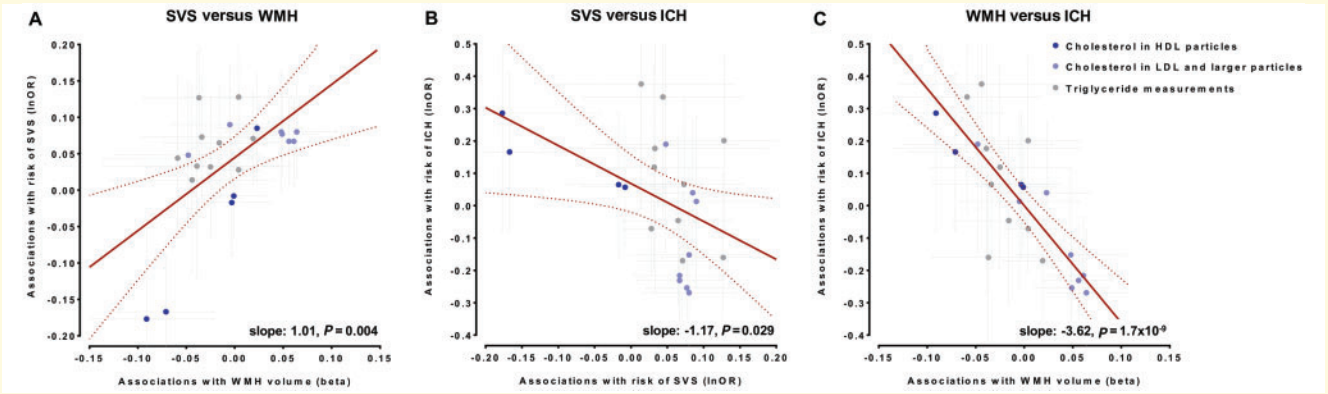


Figure 7 Comparisons of association estimates for genetic determinants of lipid traits (blood lipid levels and concentrations of lipoprotein particle components) between SVS, WMH volume, and ICH. Comparisons of the Mendelian randomization association estimates between genetic determinants of lipid traits and risk of SVS with the Mendelian randomization association estimates for WMH volume and risk of ICH. Shown are the meta-regression slopes for the comparisons of these association estimates for: (**A**) risk of SVS and WMH volume, (**B**) risk of SVS and risk of ICH, and (**C**) WMH volume and risk of ICH. Estimates are scaled per 1 SD increment.

blood lipids. Finally, we could not identify valid triglyceride-lowering variants in the locus of the target for fibrates. Hence, we could not explore their associations with SVD phenotypes. Future studies leveraging even larger GWAS datasets on blood lipid levels might identify genetic instruments for the full range of lipid-modifying drug classes.

In conclusion, our results suggest causal associations between higher HDL-C levels and both a lower risk of SVS and lower WMH volume, which were driven by cholesterol concentrations in medium-sized, and not larger HDL particles. HDL-C raising strategies might be of benefit for the prevention of ischaemic SVD. Considering the predicted increase in risk of ICH, the net benefit of such an approach would need to be tested in a randomized controlled trial.

Acknowledgements

This research has been conducted using the UK Biobank Resource (UK Biobank application 2532). We acknowledge the contributions by the MEGASTROKE Consortium, the ISGC, the GLGC, the MVP, as well as by Kettunen *et al.* (2016) for performing the original GWASs and for making summary statistics data publicly available.

Funding

M.K.G. has been supported by scholarships from the Onassis Foundation and the German Academic Exchange Service (DAAD). C.D.A. received grants from the National Institutes of Health and the National Institute of Neurological Disorders and Stroke (Grant numbers: R01NS103924, K23NS086873). This project has received funding from the European Union's Horizon 2020 research and innovation programme (No 666881), Small vessel diseases in a mechanistic perspective: Targets for Intervention (to M.D.) and No 667375, Common mechanisms and pathways in stroke and Alzheimer's disease (to M.D.); the German Research Foundation (Deutsche Forschungsgemeinschaft) as part of the Munich Cluster for Systems Neurology (EXC 2145 SyNergy ID 390857198) and the Collaborative Research Center 1123 (B3) (to M.D.); the Corona Foundation (to M.D.); the Fondation Leducq (Transatlantic Network of Excellence on the Pathogenesis of Small Vessel Disease of the Brain) (to M.D.); the e: Med program (Systems medicine of myocardial infarction and stroke) (to M.D.) and the FP7/2007-2103 European Union project Exploitation of Genomic Variants Affecting Coronary Artery Disease and Stroke Risk for Therapeutic Intervention (grant agreement number Health-F2-2013-601456) (to M.D.).

Competing interests

C.D.A. receives sponsored research support from the National Institutes of Health of the United States, the American Heart Association, Massachusetts General Hospital, and Bayer AG, and has consulted for ApoPharma, Inc. J.C.H. receives personal fellowship support from the British Heart Foundation [FS/14/55/30806]. J.C.H. works in the Clinical Trial Service Unit & Epidemiological Studies Unit of the Nuffield Department of Population Health at the University of Oxford, which has received research grants from Abbott, AstraZeneca, Bayer, Boehringer Ingelheim, GlaxoSmithKline, The Medicines Company, Merck, Mylan, Novartis, Pfizer, Roche, Schering, and Solvay, which are governed by University of Oxford contracts that protect their independence. In line with the Clinical Trial Service Unit & Epidemiological Studies Unit staff policy, J.C.H. does not take any personal payments directly or indirectly from industry (with reimbursement sought only for the costs of travel and accommodation to attend scientific meetings)

for clinical trial involvement. M.K.G., R.M., K.G.P., and M.D. have no competing interests to declare.

Supplementary material

Supplementary material is available at *Brain* online.

References

1000 Genomes Project Consortium, Abecasis GR, Auton A, Brooks LD, DePristo MA, Durbin RM, et al. An integrated map of genetic variation from 1,092 human genomes. Nature 2012; 491: 56–65.

Adams HP Jr, Bendixen BH, Kappelle LJ, Biller J, Love BB, Gordon DL, et al. Classification of subtype of acute ischemic stroke. Definitions for use in a multicenter clinical trial. TOAST. Trial of Org 10172 in acute stroke treatment. Stroke 1993; 24: 35–41.

Alfaro-Almagro F, Jenkinson M, Bangerter NK, Andersson JLR, Griffanti L, Douaud G, et al. Image processing and quality control for the first 10,000 brain imaging datasets from UK Biobank. Neuroimage 2018; 166: 400–24.

Aljondi R, Szoeke C, Steward C, Gorelik A, Desmond P. The effect of midlife cardiovascular risk factors on white matter hyperintensity volume and cognition two decades later in normal ageing women. Brain Imaging Behav 2018. Advance Access published on September 26, 2018. doi: 10.1007/s11682-018-9970-5.

Amarenco P, Bogousslavsky J, Callahan A 3rd, Goldstein LB, Hennerici M, Rudolph AE, et al. High-dose atorvastatin after stroke or transient ischemic attack. N Engl J Med 2006; 355: 549–59.

Amarenco P, Labreuche J. Lipid management in the prevention of stroke: review and updated meta-analysis of statins for stroke prevention. Lancet Neurol 2009; 8: 453–63.

Anderson CD, Falcone GJ, Phuah CL, Radmanesh F, Brouwers HB, Battey TW, et al. Genetic variants in CETP increase risk of intracerebral hemorrhage. Ann Neurol 2016; 80: 730–40.

Armitage J, Holmes MV, Preiss D. Cholesteryl ester transfer protein inhibition for preventing cardiovascular events: jACC review topic of the week. J Am Coll Cardiol 2019; 73: 477–87.

Badimon L. CETP inhibition and HDL: what is the trial REVEALing? Cardiovasc Res 2018; 114: e15–16.

Barter PJ, Caulfield M, Eriksson M, Grundy SM, Kastelein JJ, Komajda M, et al. Effects of torcetrapib in patients at high risk for coronary events. N Engl J Med 2007; 357: 2109–22.

Blauw LL, Noordam R, Soidinsalo S, Blauw CA, Li-Gao R, de Mutsert R, et al. Mendelian randomization reveals unexpected effects of CETP on the lipoprotein profile. Eur J Hum Genet 2019; 27: 422–31.

Bowden J, Davey Smith G, Burgess S. Mendelian randomization with invalid instruments: effect estimation and bias detection through Egger regression. Int J Epidemiol 2015; 44: 512–25.

Bowden J, Hemani G, Davey Smith G. Invited commentary: detecting individual and global horizontal pleiotropy in mendelian randomization-a job for the humble heterogeneity statistic? Am J Epidemiol 2018; 187: 2681–5.

Brousseau ME, Schaefer EJ, Wolfe ML, Bloedon LT, Digenio AG, Clark RW, et al. Effects of an inhibitor of cholesteryl ester transfer protein on HDL cholesterol. N Engl J Med 2004; 350: 1505–15.

Burgess S. Sample size and power calculations in Mendelian randomization with a single instrumental variable and a binary outcome. Int J Epidemiol 2014; 43: 922–9.

Burgess S, Butterworth A, Thompson SG. Mendelian randomization analysis with multiple genetic variants using summarized data. Genet Epidemiol 2013; 37: 658–65.

Burgess S, Scott RA, Timpson NJ, Davey Smith G, Thompson SG, Consortium E-I. Using published data in Mendelian randomization: a blueprint for efficient identification of causal risk factors. Eur J Epidemiol 2015; 30: 543–52.

Burgess S, Thompson SG. Multivariable Mendelian randomization: the use of pleiotropic genetic variants to estimate causal effects. Am J Epidemiol 2015; 181: 251–60.

Calkin AC, Drew BG, Ono A, Duffy SJ, Gordon MV, Schoenwaelder SM, et al. Reconstituted high-density lipoprotein attenuates platelet function in individuals with type 2 diabetes mellitus by promoting cholesterol efflux. Circulation 2009; 120: 2095–104.

Cholesterol Treatment Trialists' (CTT) Collaboration; Baigent C, Blackwell L, Emberson J, Holland LE, Reith C, et al. Efficacy and safety of more intensive lowering of LDL cholesterol: a meta-analysis of data from 170,000 participants in 26 randomised trials. Lancet 2010; 376: 1670–81.

Chou R, Dana T, Blazina I, Daeges M, Jeanne TL. Statins for prevention of cardiovascular disease in adults: evidence report and systematic review for the US Preventive Services Task Force. JAMA 2016; 316: 2008–24.

Cockerill GW, Rye KA, Gamble JR, Vadas MA, Barter PJ. High-density lipoproteins inhibit cytokine-induced expression of endothelial cell adhesion molecules. Arterioscler Thromb Vasc Biol 1995; 15: 1987–94.

Collins R, Reith C, Emberson J, Armitage J, Baigent C, Blackwell L, et al. Interpretation of the evidence for the efficacy and safety of statin therapy. Lancet 2016; 388: 2532–61.

Davey Smith G, Davies NM, Dimou N, Egger M, Gallo V, Golub R, et al. STROBE-MR: guidelines for strengthening the reporting of Mendelian randomization studies. PeerJ Preprints 2019; 7: e27857v1.

Davey Smith G, Hemani G. Mendelian randomization: genetic anchors for causal inference in epidemiological studies. Hum Mol Genet 2014; 23: R89–98.

de Leeuw FE, de Groot JC, Achten E, Oudkerk M, Ramos LM, Heijboer R, et al. Prevalence of cerebral white matter lesions in elderly people: a population based magnetic resonance imaging study. The Rotterdam Scan Study. J Neurol Neurosurg Psychiatry 2001; 70: 9–14.

Debette S, Schilling S, Duperron MG, Larsson SC, Markus HS. Clinical significance of magnetic resonance imaging markers of vascular brain injury: a systematic review and meta-analysis. JAMA Neurol 2019; 76: 81–94.

Emanuelsson F, Nordestgaard BG, Tybjaerg-Hansen A, Benn M. Impact of LDL cholesterol on microvascular versus macrovascular disease: a Mendelian randomization study. J Am Coll Cardiol 2019; 74: 1465–76.

European Stroke Organisation (ESO) Executive Committee; ESO Writing Committee. Guidelines for management of ischaemic stroke and transient ischaemic attack 2008. Cerebrovasc Dis 2008; 25: 457–507.

Ference BA, Ginsberg HN, Graham I, Ray KK, Packard CJ, Bruckert E, et al. Low-density lipoproteins cause atherosclerotic cardiovascular disease. 1. Evidence from genetic, epidemiologic, and clinical studies. A consensus statement from the European Atherosclerosis Society Consensus Panel. Eur Heart J 2017a; 38: 2459–72.

Ference BA, Graham I, Tokgozoglu L, Catapano AL. Impact of lipids on cardiovascular health: jACC health promotion series. J Am Coll Cardiol 2018; 72: 1141–56.

Ference BA, Kastelein JJP, Ginsberg HN, Chapman MJ, Nicholls SJ, Ray KK, et al. Association of genetic variants related to CETP inhibitors and statins with lipoprotein levels and cardiovascular risk. JAMA 2017b; 318: 947–56.

Ference BA, Kastelein JJP, Ray KK, Ginsberg HN, Chapman MJ, Packard CJ, et al. Association of triglyceride-lowering LPL variants and LDL-C-lowering LDLR variants with risk of coronary heart disease. JAMA 2019a; 321: 364–73.

Ference BA, Majeed F, Penumetcha R, Flack JM, Brook RD. Effect of naturally random allocation to lower low-density lipoprotein cholesterol on the risk of coronary heart disease mediated by polymorphisms in NPC1L1, HMGCR, or both: a 2 x 2 factorial Mendelian randomization study. J Am Coll Cardiol 2015; 65: 1552–61.

Ference BA, Ray KK, Catapano AL, Ference TB, Burgess S, Neff DR, et al. Mendelian randomization study of ACLY and cardiovascular disease. N Engl J Med 2019b; 380: 1033–42.

Ference BA, Robinson JG, Brook RD, Catapano AL, Chapman MJ, Neff DR, et al. Variation in PCSK9 and HMGCR and risk of cardiovascular disease and diabetes. N Engl J Med 2016; 375: 2144–53.

Ference BA, Yoo W, Alesh I, Mahajan N, Mirowska KK, Mewada A, et al. Effect of long-term exposure to lower low-density lipoprotein cholesterol beginning early in life on the risk of coronary heart disease: a Mendelian randomization analysis. J Am Coll Cardiol 2012; 60: 2631–9.

Fung KY, Wang C, Nyegaard S, Heit B, Fairn GD, Lee WL. SR-BI mediated transcytosis of HDL in brain microvascular endothelial cells is independent of caveolin, clathrin, and PDZK1. Front Physiol 2017; 8: 841.

Garner B, Waldeck AR, Witting PK, Rye KA, Stocker R. Oxidation of high density lipoproteins. II. Evidence for direct reduction of lipid hydroperoxides by methionine residues of apolipoproteins AI and AII. J Biol Chem 1998; 273: 6088–95.

Georgakis MK, Duering M, Wardlaw JM, Dichgans M. WMH and long-term outcomes in ischemic stroke: a systematic review and meta-analysis. Neurology 2019; 92: e1298–308.

Gill D, Georgakis MK, Koskeridis F, Feng Q, Wei W-Q, Theodoratou E, et al. Use of genetic variants related to antihypertensive drugs to inform on efficacy and side effects. Circulation 2019; 140: 270–9.

Goldstein LB, Amarenco P, Szarek M, Callahan A 3rd, Hennerici M, Sillesen H, et al. Hemorrhagic stroke in the stroke prevention by aggressive reduction in cholesterol levels study. Neurology 2008; 70: 2364–70.

Harrison SC, Holmes MV, Burgess S, Asselbergs FW, Jones GT, Baas AF, et al. Genetic association of lipids and lipid drug targets with abdominal aortic aneurysm: a meta-analysis. JAMA Cardiol 2018; 3: 26–33.

Hartwig FP, Davey Smith G, Bowden J. Robust inference in summary data Mendelian randomization via the zero modal pleiotropy assumption. Int J Epidemiol 2017; 46: 1985–98.

Hegele RA. CETP inhibitors-a new inning? N Engl J Med 2017; 377: 1284–5.

Hindy G, Engstrom G, Larsson SC, Traylor M, Markus HS, Melander O, et al. Role of blood lipids in the development of ischemic stroke and its subtypes: a Mendelian randomization study. Stroke 2018; 49: 820–7.

Holmes MV, Ala-Korpela M, Smith GD. Mendelian randomization in cardiometabolic disease: challenges in evaluating causality. Nat Rev Cardiol 2017; 14: 577–90.

Holmes MV, Asselbergs FW, Palmer TM, Drenos F, Lanktree MB, Nelson CP, et al. Mendelian randomization of blood lipids for coronary heart disease. Eur Heart J 2015; 36: 539–50.

Holmes MV, Millwood IY, Kartsonaki C, Hill MR, Bennett DA, Boxall R, et al. Lipids, lipoproteins, and metabolites and risk of myocardial infarction and stroke. J Am Coll Cardiol 2018; 71: 620–32.

Hopewell JC, Clarke R. Emerging risk factors for stroke: what have we learned from mendelian randomization studies? J Cereb Circ 2016; 47: 1673–8.

Hosomi N, Nagai Y, Kohriyama T, Ohtsuki T, Aoki S, Nezu T, et al. The Japan Statin treatment against recurrent stroke (J-STARS): a multicenter, randomized, open-label, parallel-group study. EBioMedicine 2015; 2: 1071–8.

HPS3/TIMI55–REVEAL Collaborative Group, Bowman L, Hopewell JC, Chen F, Wallendszus K, Stevens W, et al. Effects of anacetrapib in patients with atherosclerotic vascular disease. N Engl J Med 2017; 377: 1217–27.

Iadecola C, Duering M, Hachinski V, Joutel A, Pendlebury ST, Schneider J, et al. Vascular cognitive impairment and dementia: JACC scientific expert panel. J Am Coll Cardiol 2019; 73: 3326–44.

Intercollegiate Stroke Working Party. National clinical guideline for stroke. 5th edn. London: Royal College of Physicians; 2016.

Joshi PH, Toth PP, Lirette ST, Griswold ME, Massaro JM, Martin SS, et al. Association of high-density lipoprotein subclasses and incident coronary heart disease: the Jackson Heart and Framingham Offspring Cohort Studies. Eur J Prev Cardiol 2016; 23: 41–9.

Kernan WN, Ovbiagele B, Black HR, Bravata DM, Chimowitz MI, Ezekowitz MD, et al. Guidelines for the prevention of stroke in patients with stroke and transient ischemic attack: a guideline for healthcare professionals from the American Heart Association/American Stroke Association. Stroke 2014; 45: 2160–236.

Kettunen J, Demirkan A, Wurtz P, Draisma HH, Haller T, Rawal R, et al. Genome-wide study for circulating metabolites identifies 62 loci and reveals novel systemic effects of LPA. Nat Commun 2016; 7: 11122.

Khera AV, Kathiresan S. Genetics of coronary artery disease: discovery, biology and clinical translation. Nat Rev Genet 2017; 18: 331–44.

Klarin D, Damrauer SM, Cho K, Sun YV, Teslovich TM, Honerlaw J, et al. Genetics of blood lipids among ~300,000 multi-ethnic participants of the Million Veteran Program. Nat Genet 2018; 50: 1514–23.

Konishi M, Iso H, Komachi Y, Iida M, Shimamoto T, Jacobs DR Jr, et al. Associations of serum total cholesterol, different types of stroke, and stenosis distribution of cerebral arteries. The Akita Pathology Study. Stroke 1993; 24: 954–64.

Kontush A, Chapman MJ. Antiatherogenic function of HDL particle subpopulations: focus on antioxidative activities. Curr Opin Lipidol 2010; 21: 312–8.

Lapergue B, Moreno JA, Dang BQ, Coutard M, Delbosc S, Raphaeli G, et al. Protective effect of high-density lipoprotein-based therapy in a model of embolic stroke. Stroke 2010; 41: 1536–42.

Lawlor DA, Harbord RM, Sterne JA, Timpson N, Davey Smith G. Mendelian randomization: using genes as instruments for making causal inferences in epidemiology. Stat Med 2008; 27: 1133–63.

Lee CM, Chien CT, Chang PY, Hsieh MY, Jui HY, Liau CS, et al. High-density lipoprotein antagonizes oxidized low-density lipoprotein by suppressing oxygen free-radical formation and preserving nitric oxide bioactivity. Atherosclerosis 2005; 183: 251–8.

Lincoff AM, Nicholls SJ, Riesmeyer JS, Barter PJ, Brewer HB, Fox KAA, et al. Evacetrapib and cardiovascular outcomes in high-risk vascular disease. N Engl J Med 2017; 376: 1933–42.

Malik R, Chauhan G, Traylor M, Sargurupremraj M, Okada Y, Mishra A, et al. Multiancestry genome-wide association study of 520,000 subjects identifies 32 loci associated with stroke and stroke subtypes. Nat Genet 2018a; 50: 524–37.

Malik R, Rannikmae K, Traylor M, Georgakis MK, Sargurupremraj M, Markus HS, et al. Genome-wide meta-analysis identifies 3 novel loci associated with stroke. Ann Neurol 2018b; 84: 934–9.

Manktelow BN, Potter JF. Interventions in the management of serum lipids for preventing stroke recurrence. Cochrane Database Syst Rev 2009; 3: CD002091.

Martin SS, Jones SR, Toth PP. High-density lipoprotein subfractions: current views and clinical practice applications. Trends Endocrinol Metab 2014; 25: 329–36.

Martin SS, Khokhar AA, May HT, Kulkarni KR, Blaha MJ, Joshi PH, et al. HDL cholesterol subclasses, myocardial infarction, and mortality in secondary prevention: the Lipoprotein Investigators Collaborative. Eur Heart J 2015; 36: 22–30.

Martini SR, Flaherty ML, Brown WM, Haverbusch M, Comeau ME, Sauerbeck LR, et al. Risk factors for intracerebral hemorrhage differ according to hemorrhage location. Neurology 2012; 79: 2275–82.

Monette JS, Hutchins PM, Ronsein GE, Wimberger J, Irwin AD, Tang C, et al. Patients with coronary endothelial dysfunction have impaired cholesterol efflux capacity and reduced HDL particle concentration. Circ Res 2016; 119: 83–90.

Murphy AJ, Woollard KJ, Hoang A, Mukhamedova N, Stirzaker RA, McCormick SP, et al. High-density lipoprotein reduces the human monocyte inflammatory response. Arterioscler Thromb Vasc Biol 2008; 28: 2071–7.

Nicholls SJ, Dusting GJ, Cutri B, Bao S, Drummond GR, Rye KA, et al. Reconstituted high-density lipoproteins inhibit the acute pro-oxidant and proinflammatory vascular changes induced by a periarterial collar in normocholesterolemic rabbits. Circulation 2005; 111: 1543–50.

Nofer JR, van der Giet M, Tolle M, Wolinska I, von Wnuck Lipinski K, Baba HA, et al. HDL induces NO-dependent vasorelaxation via the lysophospholipid receptor S1P3. J Clin Invest 2004; 113: 569–81.

Nowak C, Arnlov J. A Mendelian randomization study of the effects of blood lipids on breast cancer risk. Nat Commun 2018; 9: 3957.

O'Brien JT, Thomas A. Vascular dementia. Lancet 2015; 386: 1698–706.

O'Donnell CJ, Sabatine MS. Opportunities and challenges in Mendelian randomization studies to guide trial design. JAMA Cardiol 2018; 3: 967.

Palmer TM, Lawlor DA, Harbord RM, Sheehan NA, Tobias JH, Timpson NJ, et al. Using multiple genetic variants as instrumental variables for modifiable risk factors. Stat Methods Med Res 2012; 21: 223–42.

Pantoni L. Cerebral small vessel disease: from pathogenesis and clinical characteristics to therapeutic challenges. Lancet Neurol 2010; 9: 689–701.

Prosser HC, Ng MK, Bursill CA. The role of cholesterol efflux in mechanisms of endothelial protection by HDL. Curr Opin Lipidol 2012; 23: 182–9.

Qureshi AI, Mendelow AD, Hanley DF. Intracerebral haemorrhage. Lancet 2009; 373: 1632–44.

Qureshi AI, Tuhrim S, Broderick JP, Batjer HH, Hondo H, Hanley DF. Spontaneous intracerebral hemorrhage. N Engl J Med 2001; 344: 1450–60.

Ray KK, Bays HE, Catapano AL, Lalwani ND, Bloedon LT, Sterling LR, et al. Safety and efficacy of bempedoic acid to reduce LDL cholesterol. N Engl J Med 2019; 380: 1022–32.

Rutten-Jacobs LCA, Tozer DJ, Duering M, Malik R, Dichgans M, Markus HS, et al. Genetic study of white matter integrity in UK biobank (N=8448) and the overlap with stroke, depression, and dementia. Stroke 2018; 49: 1340–7.

Schwartz GG, Olsson AG, Abt M, Ballantyne CM, Barter PJ, Brumm J, et al. Effects of dalcetrapib in patients with a recent acute coronary syndrome. N Engl J Med 2012; 367: 2089–99.

Sorrentino SA, Besler C, Rohrer L, Meyer M, Heinrich K, Bahlmann FH, et al. Endothelial-vasoprotective effects of high-density lipoprotein are impaired in patients with type 2 diabetes mellitus but are improved after extended-release niacin therapy. Circulation 2010; 121: 110–22.

Spieker LE, Sudano I, Hurlimann D, Lerch PG, Lang MG, Binggeli C, et al. High-density lipoprotein restores endothelial function in hypercholesterolemic men. Circulation 2002; 105: 1399–402.

Stroke Foundation. Clinical guidelines for stroke management. Melbourne: Stroke Foundation; 2017.

Sudlow CL, Warlow CP. Comparable studies of the incidence of stroke and its pathological types: results from an International Collaboration. International Stroke Incidence Collaboration. Stroke 1997; 28: 491–9.

Sun L, Clarke R, Bennett D, Guo Y, Walters RG, Hill M, et al. Causal associations of blood lipids with risk of ischemic stroke and intracerebral hemorrhage in Chinese adults. Nat Med 2019; 25: 569–74.

Superko HR, Pendyala L, Williams PT, Momary KM, King SB 3rd, Garrett BC. High-density lipoprotein subclasses and their relationship to cardiovascular disease. J Clin Lipidol 2012; 6: 496–523.

Taylor AE, Davies NM, Ware JJ, VanderWeele T, Smith GD, Munafo MR. Mendelian randomization in health research: using appropriate genetic variants and avoiding biased estimates. Econ Hum Biol 2014; 13: 99–106.

Terasaka N, Wang N, Yvan-Charvet L, Tall AR. High-density lipoprotein protects macrophages from oxidized low-density lipoprotein-induced apoptosis by promoting efflux of 7-ketocholesterol via ABCG1. Proc Natl Acad Sci U S A 2007; 104: 15093–8.

Tran-Dinh A, Diallo D, Delbosc S, Varela-Perez LM, Dang QB, Lapergue B, et al. HDL and endothelial protection. Br J Pharmacol 2013; 169: 493–511.

Valdes-Marquez E, Parish S, Clarke R, Stari T, Worrall BB, Hopewell JC. Relative effects of LDL-C on ischemic stroke and coronary disease: a Mendelian randomization study. Neurology 2019; 92: e1176–87.

Viswambharan H, Ming XF, Zhu S, Hubsch A, Lerch P, Vergeres G, et al. Reconstituted high-density lipoprotein inhibits thrombin-induced endothelial tissue factor expression through inhibition of RhoA and stimulation of phosphatidylinositol 3-kinase but not Akt/endothelial nitric oxide synthase. Circ Res 2004; 94: 918–25.

Voight BF, Peloso GM, Orho-Melander M, Frikke-Schmidt R, Barbalic M, Jensen MK, et al. Plasma HDL cholesterol and risk of myocardial infarction: a mendelian randomisation study. Lancet 2012; 380: 572–80.

Wang X, Dong Y, Qi X, Huang C, Hou L. Cholesterol levels and risk of hemorrhagic stroke: a systematic review and meta-analysis. Stroke 2013; 44: 1833–9.

Wardlaw JM, Smith C, Dichgans M. Small vessel disease: mechanisms and clinical implications. Lancet Neurol 2019; 18: 684–96.

White J, Swerdlow DI, Preiss D, Fairhurst-Hunter Z, Keating BJ, Asselbergs FW, et al. Association of lipid fractions with risks for coronary artery disease and diabetes. JAMA Cardiol 2016; 1: 692–9.

Willer CJ, Schmidt EM, Sengupta S, Peloso GM, Gustafsson S, Kanoni S, et al. Discovery and refinement of loci associated with lipid levels. Nat Genet 2013; 45: 1274–83.

Williams PT. Low high-density lipoprotein 3 reduces the odds of men surviving to age 85 during 53-year follow-up. J Am Geriatr Soc 2012; 60: 430–6.

Woo D, Falcone GJ, Devan WJ, Brown WM, Biffi A, Howard TD, et al. Meta-analysis of genome-wide association studies identifies 1q22 as a susceptibility locus for intracerebral hemorrhage. Am J Hum Genet 2014; 94: 511–21.

Wurtz P, Havulinna AS, Soininen P, Tynkkynen T, Prieto-Merino D, Tillin T, et al. Metabolite profiling and cardiovascular event risk: a prospective study of 3 population-based cohorts. Circulation 2015; 131: 774–85.

Yin ZG, Wang QS, Yu K, Wang WW, Lin H, Yang ZH. Sex differences in associations between blood lipids and cerebral small vessel disease. Nutr Metab Cardiovasc Dis 2018; 28: 28–34.

Yu S, Yarnell JW, Sweetnam P, Bolton CH. High density lipoprotein subfractions and the risk of coronary heart disease: 9-years follow-up in the Caerphilly Study. Atherosclerosis 2003; 166: 331–8.

Yuhanna IS, Zhu Y, Cox BE, Hahner LD, Osborne-Lawrence S, Lu P, et al. High-density lipoprotein binding to scavenger receptor-BI activates endothelial nitric oxide synthase. Nat Med 2001; 7: 853–7.

Zhu Z, Zheng Z, Zhang F, Wu Y, Trzaskowski M, Maier R, et al. Causal associations between risk factors and common diseases inferred from GWAS summary data. Nat Commun 2018; 9: 224.

DISCUSSION

Summary of the findings

In this thesis, I used large-scale genetic data to identify novel risk factors and drug targets for cerebrovascular disease. Applying Mendelian randomization, this thesis provides evidence that genetic predisposition to higher MCP-1 levels and to upregulated IL-6 signaling are associated with higher risk of ischemic stroke and other cardiovascular phenotypes. These results for MCP-1 were further replicated in a meta-analysis of observational population-based cohort studies, where MCP-1 levels among stroke-free individuals were associated with a higher risk of incident ischemic stroke over a 16-year follow-up period. Interestingly, circulating MCP-1 and IL-6 levels were both associated with ischemic stroke risk, independently of each other, thus indicating that targeting the two pathways might offer complementary benefits in reducing stroke risk. These results highlight the potential of targeting inflammatory mechanisms for the treatment of atherosclerosis.

By leveraging GWAS data I further examined the associations of genetic predisposition to well-established risk factors for stroke with manifestations of cerebral SVD. Specifically, genetic predisposition to high blood pressure was found to be associated with a higher risk of small vessel stroke, deep ICH, and the radiological phenotype of WMH volume. My collaborators and I further showed that genetically determined higher HDL cholesterol levels, and particularly cholesterol levels in medium-sized HDL particles, are associated with a lower risk of ischemic manifestations of cerebral SVD (small vessel stroke, WMH volume). By focusing on genetic variants in genes encoding drug targets, we were able to identify genetic proxies for the effects of common blood pressure-lowering and lipid-modifying drugs drug classes. Interestingly, we found BP-lowering variants at loci for targets of calcium channel blockers and HDL-C-raising variants at loci for targets of CETP (cholesterol-ester transfer protein) inhibitors to be associated with significantly lower risk of small vessel stroke and lower WMH volume. These findings demonstrate that the use of genetics might offer a powerful approach for investigating the efficacy and repurposing potential of commonly used pharmacological agents for cerebral SVD.

Cytokines as drug targets for cerebrovascular disease

Although inflammation has long been identified as a key contributor to atherosclerosis, it was only recetnly that the results of a large-scale clinical trial provided evidence for the efficacy of anti-inflammatory approaches for lowering vascular risk. Yet, the discrepancy in the results of the CANTOS trial testing a monoclonal antibody against IL-1β (Ridker *et al.*, 2017; Aday and Ridker, 2019; Ridker, 2019; Ridker *et al.*, 2019a) and CIRT (Ridker *et al.*, 2019a), which tested low-dose methotrexate, highlight the importance of targeting specific inflammatory cytokines and pathways for lowering vascular risk (Ridker *et al.*, 2017; Aday and Ridker, 2019; Ridker, 2019; Ridker *et al.*, 2019a).

In the MR analyses, we systematically explored the associations between genetic predisposition to circulating levels of 41 cytokines with the risk of stroke, aiming to identify specific mediators with the highest potential to be tested as drug targets for lowering stroke risk. Across these cytokines, genetic predisposition to higher lifetime MCP-1 levels came up as showing the strongest association with the risk of stroke, and specifically large artery stroke. We then confirmed and extended these results in a meta-analysis of 6 population-based cohort studies with long-term follow-up involving 17,180 stroke-free individuals. Again, high MCP-1 levels in midlife were associated with a higher risk of incident ischemic stroke over follow-up independently of conventional vascular risk factors. The results were remarkably consistent between the two approaches: with MR the odds ratio for stroke was 1.06 per SD increment in genetically determined MCP-1 levels, which is almost identical to the hazard ratio for incident stroke observed in the current meta-analysis of observational studies.

Besides the evidence from genetic and observational studies provided in the current thesis, experimental studies in animal models of atherosclerosis further suggest MCP-1 as a key molecule in atherogenesis and atheroprogression. By binding to its receptor CCR2, MCP-1 is the prototypical CC family chemokine that is upregulated by chronic inflammatory conditions and attracts monocytes to the subendothelial space of the atherogenic arterial wall (Lin *et al.*, 2014). Mice lacking MCP-1 (Gu *et al.*, 1998; Combadiere *et al.*, 2008) or CCR2 (Boring *et al.*, 1998) are less susceptible to atherosclerosis and anti-MCP-1 gene therapy (Inoue *et al.*, 2002), MCP-1 inhibitors (Grassia *et al.*, 2009), MCP-1 competitors (Liehn *et al.*, 2010), and CCR2 antagonists

(Yamashita *et al.*, 2002; Okamoto *et al.*, 2012; Bot *et al.*, 2017; Winter *et al.*, 2018) reduce plaque size and inhibit plaque progression and destabilization in experimental atherosclerosis. In contrast, overexpression of MCP-1 promotes oxidized lipid accumulation, macrophage infiltration, and smooth muscle cell proliferation, thus accelerating atherosclerosis (Aiello *et al.*, 1999) .

When viewed together with the existing experimental data (Boring *et al.*, 1998; Gu *et al.*, 1998; Combadiere *et al.*, 2008; Liehn *et al.*, 2010; Bot *et al.*, 2017), the data presented here from two different approaches in humans (MR and population-based cohort studies) provide triangulation of evidence regarding a role of MCP-1 as a causal risk factor for ischemic stroke. This thesis thus provides strong evidence for the candidacy of the MCP-1/CCR2 as a target for lowering risk of ischemic stroke. The MCP-1/CCR2 pathway has to our knowledge only been targeted in a small phase II clinical trial in 108 patients with risk factors for atherosclerosis and elevated circulating CRP levels. MLN1202, a humanized monoclonal antibody against CCR2 reduced CRP levels after 4 and 12 weeks (Gilbert *et al.*, 2011). However, effects on clinical endpoints were not assessed (Gilbert *et al.*, 2011) and would need to be determined in a larger trial.

In addition to MCP-1, we identified variants at the locus of *IL6R* that could be used as proxies for genetically downregulated IL-6 signaling. Exploring the associations of these variants with ischemic stroke and its subtypes, we found significant reductions in the risk of large artery and small vessel stroke. The MR association between genetically downregulated IL-6 signaling and lower risk of large artery stroke extends previous clinical (Ridker *et al.*, 2000; Kaptoge *et al.*, 2014; Ridker *et al.*, 2018a), genetic (Il R. Genetics Consortium Emerging Risk Factors Collaboration *et al.*, 2012; Interleukin-6 Receptor Mendelian Randomisation Analysis Consortium *et al.*, 2012), and experimental (Ikeda *et al.*, 1991; Huber *et al.*, 1999) data demonstrating a key role of IL-6 signaling in atherosclerosis. Moreover, pharmacological inhibition of IL-6R has been shown to attenuate atherosclerotic lesions in an experimental model of atherosclerosis (Akita *et al.*, 2017). Our finding of an effect of genetic predisposition to downregulated IL-6 signaling on multiple atherosclerotic phenotypes (large artery stroke, coronary artery disease, myocardial infarction, aortic aneurysm, atrial fibrillation, carotid plaque) provides further support that IL-6 signaling is critically implicated in atherogenesis and atheroprogression and might represent a valid therapeutic target.

Notably, genetically downregulated IL-6 signaling was further associated with small vessel stroke. There is only limited evidence regarding a role of inflammation in general and of IL-6 signaling in particular in cerebral SVD (Low *et al.*, 2019). In a small prospective study of 123 patients with manifestations of cerebral SVD, IL-6 circulating levels were associated with a higher risk of incident lacunes, a marker of SVD on brain magnetic resonance imaging (Staszewski *et al.*, 2018). However, cross-sectional analyses from larger population-based studies showed inconsistent findings for lacunes, silent brain infarcts and other SVD manifestations (Hoshi *et al.*, 2005; Fornage *et al.*, 2008; Baune *et al.*, 2009; Yoshida *et al.*, 2009; Satizabal *et al.*, 2012; Shoamanesh *et al.*, 2015). While the specific mechanisms underlying our MR results remain unknown, our findings suggest that inhibition of IL-6 signaling aside from being a candidate treatment for atherosclerosis might also lower the risk of small vessel stroke.

The CANTOS trial targeted IL-1β rather than IL-6R and thus provided only indirect evidence for a benefit of interfering with IL-6 signaling (Ridker *et al.*, 2018a; Ridker, 2019). Interestingly, the study further showed that part of the residual vascular risk after IL-1β inhibition could be explained by IL-6 levels, thus providing evidence that direct IL-6 signaling inhibition might represent a more effective strategy (Ridker *et al.*, 2019b). Also, CANTOS was based on a population of individuals with coronary artery disease and explored a combined vascular endpoint rather than offering information on individual cardiovascular outcomes. With respect to stroke, there was a 7% reduction in incident stroke events in the IL-1β arm, which however did not reach statistical significance, possibly because of insufficient power (Ridker *et al.*, 2017). Our MR results provide evidence for directionally consistent effects of IL-6 signaling in multiple cardiovascular outcomes. Thus, our findings offer a solid basis for future clinical trials exploring the benefit of pharmacological IL-6R inhibition for the range of phenotypes examined here.

Secondary analyses from the CANTOS trial showed that the reductions in vascular event rates after IL-1β inhibition were restricted to individuals with a substantial decrease in IL-6 or hsCRP levels (Ridker *et al.*, 2018a; Ridker *et al.*, 2018b). Intriguingly, the risk estimates for stroke by MCP-1 levels in our meta-analysis of observational studies remained stable after additional adjustments for the baseline levels of IL-6, hsCRP, and both IL-6 and hsCRP. This observation provides indirect evidence suggesting that elevated levels of MCP-1 might influence risk of stroke independently of the IL-1β/IL-6/CRP axis. Thus, targeting the MCP-1/CCR2 pathway might serve as an alternative anti-

inflammatory strategy with independent and complementary effects in reducing vascular event rates on top of current approaches.

Identifying novel drug targets for cerebral small vessel disease

Manifestations of cerebral SVD are highly prevalent in the ageing population with figures reaching up to 90% in patients aged 65 years and above (de Leeuw *et al.*, 2001) and patients with these manifestations mark a population at increased risk for stroke, dementia and death (Debette *et al.*, 2018; Georgakis *et al.*, 2019). However, the risk factors for cerebral SVD remain elusive and to date there have been no informative trials exploring the efficacy of specific interventions for the prevention of outcomes related to SVD (Group *et al.*, 2013; Croall *et al.*, 2018; van Middelaar *et al.*, 2018). With the MR studies presented here, I explored the associations of blood pressure and blood lipid levels, two major risk factors for large vessel disease, with manifestations of cerebral SVD. I further aimed to explore if genetic variants at loci of the targets of common antihypertensive and lipid-modifying drugs associate with the risk of cerebral SVD.

As expected, we found genetic predisposition to higher blood pressure to associate with a higher risk of small vessel stroke, WMH volume, and deep ICH. Unlike deep ICH, lobar ICH is often related to cerebral amyloid angiopathy and the absence of an association signal between BP and lobar ICH is consistent with observational data (Jackson and Sudlow, 2006; Martini *et al.*, 2012). Most interestingly, we also found a benefit of BP lowering through genetic proxies for CCBs over BBs for SVS and the related phenotype of WMH. In contrast, we found no disparity in effects between genetic proxies for CCBs and BBs for other stroke subtypes. This might suggest that CCBs may be particularly effective in preventing manifestations of cerebral SVD. The mechanisms underlying this observation are currently unknown, but might be related to the established influence of CCBs on BP variability (Rothwell *et al.*, 2010; Webb *et al.*, 2010; Yamaguchi *et al.*, 2014).

Our MR analyses for blood lipids further provide evidence for a protective role of HDL-C on ischemic SVD (small vessel stroke and WMH volume), which is in agreement with findings from small observational studies (Aljondi *et al.*, 2018; Yin *et al.*, 2018). The mechanisms underlying this observation may involve protective effects of HDL particles on the vascular endothelium (Sorrentino *et al.*, 2010; Prosser *et al.*, 2012; Tran-Dinh *et al.*,

2013; Monette *et al.*, 2016). Endothelial cells of the brain microvasculature (Lapergue *et al.*, 2010; Fung *et al.*, 2017), express receptors which upon HDL binding induce intracellular signaling eventually leading to vasodilatory (Yuhanna *et al.*, 2001; Spieker *et al.*, 2002; Nofer *et al.*, 2004), anti-inflammatory (Cockerill *et al.*, 1995; Nicholls *et al.*, 2005; Murphy *et al.*, 2008), anti-oxidative(Garner *et al.*, 1998; Lee *et al.*, 2005; Terasaka *et al.*, 2007), and anti-thrombotic effects (Viswambharan *et al.*, 2004; Calkin *et al.*, 2009). The observed effect was specific for cholesterol concentrations in medium-sized HDL particles, which agrees with the general notion that the favorable effects observed for HDL are predominantly exerted by the smaller and denser HDL particles (Yu *et al.*, 2003; Williams, 2012; Martin *et al.*, 2014). The elucidation of the mechanisms by which HDL-C and particularly medium-sized HDL particles influence the risk of cerebral SVD might offer novel insights into the mechanisms of cerebral SVD.

HDL-C raising genetic variants in the *CETP* locus were also associated with lower SVS risk and WMH volume. Pharmacological CETP inhibition leads to an increase in the circulating pool of HDL particles (Armitage *et al.*, 2019). While a number of randomized trials have investigated the efficacy of CETP inhibition on top of statins for reducing vascular risk (Barter *et al.*, 2007; Schwartz *et al.*, 2012; HPS3/TIMI55–REVEAL Collaborative Group *et al.*, 2017; Lincoff *et al.*, 2017), none of them explicitly reported effects on risk of small vessel stroke or other SVD manifestations. On the basis of our MR results, *post hoc* analyses should consider stratifying for stroke subtypes. In the REVEAL trial, the reduction in vascular risk by CETP inhibition was mediated by a reduction in LDL-C rather than an increase in HDL-C (HPS3/TIMI55–REVEAL Collaborative Group *et al.*, 2017). Yet, our multivariable MR results suggest, that in contrast with the REVEAL trial, the effects of *CETP* variants on SVD manifestations might be primarily exerted by HDL-C raising and not LDL-C lowering.

Our results suggest opposite effects of the blood lipid traits on risk of ischemic SVD phenotypes (small vessel stroke, WMH volume), as compared with ICH. This agrees with previous observational, genetic, and clinical studies that have found high HDL-C and low LDL-C levels to be associated with a higher risk of ICH (Amarenco *et al.*, 2006; Goldstein *et al.*, 2008; Wang *et al.*, 2013; Anderson *et al.*, 2016; Sun *et al.*, 2019). Here, HDL-C raising variants in the *CETP* locus were also associated with a higher risk of both deep and lobar ICH. Thus, HDL-C raising strategies might decrease the risk for small vessel stroke and

WMH volume, but such effects should be counterbalanced to potential increases in the risk of ICH.

Collectively, with the MR analyses presented in the current thesis, I provide evidence that BP-lowering and HDL-raising approaches might be effective strategies for decreasing the risk of ischemic SVD manifestations. More specifically, our findings support that BP-lowering through calcium channel blockade and HDL-C-raising through CETP inhibition might be promising approaches in preventing ischemic manifestations of cerebral SVD, worth exploring in future clinical trials.

Methodological considerations

The validity of MR analyses is based on specific assumptions. First, the variants used as instruments need to represent valid genetic determinants of the risk factor under study and be strongly associated with it. In our MR for cytokine levels, instrument selection was based on a single discovery GWAS that adjusted for BMI. While the associations remained consistent when using unweighted allele scores, it cannot be excluded that the BMI adjustment led to collider bias during instrument selection. The genetic instruments used in all other MR analyses were selected from GWAS meta-analyses including both discovery and replication samples. In all occasions, we were very strict with our statistical significance thresholds for instrument selection to preclude both selection of invalid instruments and introduction of weak instrument bias. Second, the genetic variants used as instruments must exert any of their effects on the clinical outcomes only through the risk factors under study and not through alternative pathways. The use of horizontally pleiotropic variants may violate this assumption. To decrease the possibility of pleiotropy we either (1) focused our analyses on variants in specific loci closely related to the risk factor under study (e.g. IL6R for IL-6 signaling or genes encoding the drug targets when studying drug effects), or (2) performed sensitivity analyses that control for this type of bias (e.g. MR-Egger, weighted median approach, MR-PRESSO). Yet, for MCP-1, it was not possible to identify genetic variants located in the vicinity of the *CCL2* gene thus precluding analyses restricted to SNPs within this locus. Consequently, while no statistical evidence for pleiotropy was found, nonspecific effects of the MCP-1 *trans*-acting instruments cannot be entirely excluded.

MR analyses exploring drug effects estimate the cumulative effects of lifelong exposure to genetic variants, which might differ from those of a clinical intervention. Still, the estimates of the associations between the identified genetic instruments for antihypertensive drug classes and vascular endpoints were comparable to those from clinical trials. Similarly, using CRP levels as a proxy for downstream IL-6 signaling enabled the scaling of the respective association estimates to the effects of tocilizumab, as determined from previous trials, thus providing clinically meaningful estimates. Indeed, when exploring the effects of the selected proxies on upstream regulators (IL-6 and soluble IL-6R) and downstream effectors (fibrinogen) of IL-6 signaling for validation, we found consistent estimates with the effects observed with pharmacological inhibition of IL-6R.

With regards to the meta-analysis of observational studies, the different assays used by individual studies to quantify circulating MCP-1 levels and the different sample sources (plasma vs. serum) resulted in substantial variations in MCP-1 levels. Although our analyses standardized MCP-1 levels across studies, it was not possible to explore associations between absolute MCP-1 values and risk of stroke. Finally, I should note that most of the datasets analysed in the current thesis were based on individuals of primarily European origin, and might thus not apply to other ethnic groups.

CONCLUSIONS AND FUTURE DIRECTIONS

Cerebrovascular disease remains a major cause of mortality and disability worldwide. In the context of lack of specific neuroprotective treatments, current efforts are focused on prevention of its clinical consequences. This requires a deep understanding of underlying pathophysiological mechanisms and the identification of modifiable risk factors that could be targeted in the context of preventive strategies. However, the etiological heterogeneity of cerebrovascular disease and the inherent limitations of observational studies to explore its causes hamper progress regarding identification of risk factors and drug targets. With this thesis I aimed to address this issue by using large-scale genetic data and the approach of Mendelian randomization that enables exploration of causal inference in a more robust framework.

I provide support for a key role of inflammatory mechanisms in ischemic stroke and for the potential of anti-inflamatory approaches for lowering ischemic stroke risk. Based on genetic studies and population-based cohorts, our findings suggest circulating MCP-1 levels as a novel risk factor for ischemic stroke. Similarly, I provide evidence for a key role of IL-6 signaling in ischemic stroke and other cardiovascular phenotypes. These results extend and corroborate previous experimental and clinical evidence supporting the MCP-1 and IL-6 signaling pathways to play a causal role in the progression of atherosclerosis and the pathogenesis of stroke. Future clinical trials should explore whether taregting MCP-1 or IL-6R signaling could represent valid therapeutic targets for lowering ischemic stroke risk.

I further explored in large-scale genetic data how blood pressure and blood lipid levels associate with manifestations of cerebral SVD. I provide evidence for causal associations of genetically determined higher BP with all major manifestations of cerebral SVD (small vessel stroke, WMH, deep ICH), except for lobar ICH. Our findings further support that genetically determined lower HDL-C levels are associated with higher risk of small vessel stroke and higher WMH volume. Genetic proxies for calcium channel blockers and CETP inhibitors showed strong associations with small vessel stroke and WMH. Thus, calcium channel blockade and CETP inhibition might comprise promising strategy for the prevention of ischemic manifestations of cerebral SVD and its clinical sequalae and would need to be explored in future clinical trials.

REFERENCES

Abbott RD, Donahue RP, MacMahon SW, Reed DM, Yano K. Diabetes and the risk of stroke. The Honolulu Heart Program. JAMA 1987; 257(7): 949-52.

Adams HP, Jr., Bendixen BH, Kappelle LJ, Biller J, Love BB, Gordon DL*, et al.* Classification of subtype of acute ischemic stroke. Definitions for use in a multicenter clinical trial. TOAST. Trial of Org 10172 in Acute Stroke Treatment. Stroke 1993; 24(1): 35-41.

Adams HP, Jr., Biller J. Classification of subtypes of ischemic stroke: history of the trial of org 10172 in acute stroke treatment classification. Stroke 2015; 46(5): e114-7.

Aday AW, Ridker PM. Targeting Residual Inflammatory Risk: A Shifting Paradigm for Atherosclerotic Disease. Front Cardiovasc Med 2019; 6: 16.

Aiello RJ, Bourassa PA, Lindsey S, Weng W, Natoli E, Rollins BJ*, et al.* Monocyte chemoattractant protein-1 accelerates atherosclerosis in apolipoprotein E-deficient mice. Arteriosclerosis, thrombosis, and vascular biology 1999; 19(6): 1518-25.

Akita K, Isoda K, Sato-Okabayashi Y, Kadoguchi T, Kitamura K, Ohtomo F*, et al.* An Interleukin-6 Receptor Antibody Suppresses Atherosclerosis in Atherogenic Mice. Front Cardiovasc Med 2017; 4: 84.

Aljondi R, Szoeke C, Steward C, Gorelik A, Desmond P. The effect of midlife cardiovascular risk factors on white matter hyperintensity volume and cognition two decades later in normal ageing women. Brain Imaging Behav 2018.

Amarenco P, Bogousslavsky J, Callahan A, 3rd, Goldstein LB, Hennerici M, Rudolph AE*, et al.* High-dose atorvastatin after stroke or transient ischemic attack. N Engl J Med 2006; 355(6): 549-59.

Amarenco P, Labreuche J. Lipid management in the prevention of stroke: review and updated meta-analysis of statins for stroke prevention. Lancet Neurol 2009; 8(5): 453-63.

Anderson CD, Falcone GJ, Phuah CL, Radmanesh F, Brouwers HB, Battey TW*, et al.* Genetic variants in CETP increase risk of intracerebral hemorrhage. Ann Neurol 2016; 80(5): 730-40.

Armitage J, Holmes MV, Preiss D. Cholesteryl Ester Transfer Protein Inhibition for Preventing Cardiovascular Events: JACC Review Topic of the Week. J Am Coll Cardiol 2019; 73(4): 477-87.

Bandres-Ciga S, Noyce AJ, Traynor BJ. Mendelian Randomization-A Journey From Obscurity to Center Stage With a Few Potholes Along the Way. JAMA Neurol 2019.

Banerjee C, Moon YP, Paik MC, Rundek T, Mora-McLaughlin C, Vieira JR*, et al.* Duration of diabetes and risk of ischemic stroke: the Northern Manhattan Study. Stroke 2012; 43(5): 1212-7.

Banerjee G, Carare R, Cordonnier C, Greenberg SM, Schneider JA, Smith EE*, et al.* The increasing impact of cerebral amyloid angiopathy: essential new insights for clinical practice. J Neurol Neurosurg Psychiatry 2017; 88(11): 982-94.

Barter PJ, Caulfield M, Eriksson M, Grundy SM, Kastelein JJ, Komajda M*, et al.* Effects of torcetrapib in patients at high risk for coronary events. N Engl J Med 2007; 357(21): 2109-22.

Bath PM, Wardlaw JM. Pharmacological treatment and prevention of cerebral small vessel disease: a review of potential interventions. Int J Stroke 2015; 10(4): 469-78.

Baune BT, Ponath G, Rothermundt M, Roesler A, Berger K. Association between cytokines and cerebral MRI changes in the aging brain. J Geriatr Psychiatry Neurol 2009; 22(1): 23-34.

Baykara E, Gesierich B, Adam R, Tuladhar AM, Biesbroek JM, Koek HL*, et al.* A Novel Imaging Marker for Small Vessel Disease Based on Skeletonization of White Matter Tracts and Diffusion Histograms. Ann Neurol 2016; 80(4): 581-92.

Benjamin EJ, Blaha MJ, Chiuve SE, Cushman M, Das SR, Deo R*, et al.* Heart Disease and Stroke Statistics-2017 Update: A Report From the American Heart Association. Circulation 2017; 135(10): e146-e603.

Bevan S, Traylor M, Adib-Samii P, Malik R, Paul NL, Jackson C*, et al.* Genetic heritability of ischemic stroke and the contribution of previously reported candidate gene and genomewide associations. Stroke 2012; 43(12): 3161-7.

Biffi A, Greenberg SM. Cerebral amyloid angiopathy: a systematic review. J Clin Neurol 2011; 7(1): 1-9.

Blood Pressure Lowering Treatment Trialists C. Blood pressure-lowering treatment based on cardiovascular risk: a meta-analysis of individual patient data. Lancet 2014; 384(9943): 591-8.

Bohula EA, Giugliano RP, Leiter LA, Verma S, Park JG, Sever PS*, et al.* Inflammatory and Cholesterol Risk in the FOURIER Trial. Circulation 2018; 138(2): 131-40.

Boring L, Gosling J, Cleary M, Charo IF. Decreased lesion formation in CCR2-/- mice reveals a role for chemokines in the initiation of atherosclerosis. Nature 1998; 394(6696): 894-7.

Bot I, Ortiz Zacarias NV, de Witte WE, de Vries H, van Santbrink PJ, van der Velden D*, et al.* A novel CCR2 antagonist inhibits atherogenesis in apoE deficient mice by achieving high receptor occupancy. Sci Rep 2017; 7(1): 52.

Brunstrom M, Carlberg B. Association of Blood Pressure Lowering With Mortality and Cardiovascular Disease Across Blood Pressure Levels: A Systematic Review and Meta-analysis. JAMA Intern Med 2018; 178(1): 28-36.

Burgess S, Davey Smith G. How humans can contribute to Mendelian randomization analyses. International journal of epidemiology 2019; 48(3): 661-4.

Calkin AC, Drew BG, Ono A, Duffy SJ, Gordon MV, Schoenwaelder SM*, et al.* Reconstituted high-density lipoprotein attenuates platelet function in individuals with type 2 diabetes mellitus by promoting cholesterol efflux. Circulation 2009; 120(21): 2095-104.

Chabriat H, Joutel A, Dichgans M, Tournier-Lasserve E, Bousser MG. Cadasil. Lancet Neurol 2009; 8(7): 643-53.

Cholesterol Treatment Trialists Collaboration, Baigent C, Blackwell L, Emberson J, Holland LE, Reith C*, et al.* Efficacy and safety of more intensive lowering of LDL cholesterol: a meta-analysis of data from 170,000 participants in 26 randomised trials. Lancet 2010; 376(9753): 1670-81.

Chou R, Dana T, Blazina I, Daeges M, Jeanne TL. Statins for Prevention of Cardiovascular Disease in Adults: Evidence Report and Systematic Review for the US Preventive Services Task Force. JAMA 2016; 316(19): 2008-24.

Chugh SS, Havmoeller R, Narayanan K, Singh D, Rienstra M, Benjamin EJ*, et al.* Worldwide epidemiology of atrial fibrillation: a Global Burden of Disease 2010 Study. Circulation 2014; 129(8): 837-47.

Cockerill GW, Rye KA, Gamble JR, Vadas MA, Barter PJ. High-density lipoproteins inhibit cytokine-induced expression of endothelial cell adhesion molecules. Arteriosclerosis, thrombosis, and vascular biology 1995; 15(11): 1987-94.

Collins R, Reith C, Emberson J, Armitage J, Baigent C, Blackwell L*, et al.* Interpretation of the evidence for the efficacy and safety of statin therapy. Lancet 2016; 388(10059): 2532-61.

Combadiere C, Potteaux S, Rodero M, Simon T, Pezard A, Esposito B*, et al.* Combined inhibition of CCL2, CX3CR1, and CCR5 abrogates Ly6C(hi) and Ly6C(lo) monocytosis and almost abolishes atherosclerosis in hypercholesterolemic mice. Circulation 2008; 117(13): 1649-57.

Cordonnier C, Demchuk A, Ziai W, Anderson CS. Intracerebral haemorrhage: current approaches to acute management. Lancet 2018; 392(10154): 1257-68.

Croall ID, Tozer DJ, Moynihan B, Khan U, O'Brien JT, Morris RG*, et al.* Effect of Standard vs Intensive Blood Pressure Control on Cerebral Blood Flow in Small Vessel Disease: The PRESERVE Randomized Clinical Trial. JAMA Neurol 2018; 75(6): 720-7.

Danese S, Vermeire S, Hellstern P, Panaccione R, Rogler G, Fraser G*, et al.* Randomised trial and open-label extension study of an anti-interleukin-6 antibody in Crohn's disease (ANDANTE I and II). Gut 2019; 68(1): 40-8.

de Faire U, Friberg L, Lundman T. Concordance for mortality with special reference to ischaemic heart disease and cerebrovascular disease. A study on the Swedish Twin Registry. Prev Med 1975; 4(4): 509-17.

de Laat KF, van Norden AG, Gons RA, van Oudheusden LJ, van Uden IW, Bloem BR*, et al.* Gait in elderly with cerebral small vessel disease. Stroke 2010; 41(8): 1652-8.

de Leeuw FE, de Groot JC, Achten E, Oudkerk M, Ramos LM, Heijboer R*, et al.* Prevalence of cerebral white matter lesions in elderly people: a population based magnetic resonance imaging study. The Rotterdam Scan Study. J Neurol Neurosurg Psychiatry 2001; 70(1): 9-14.

Debette S, Schilling S, Duperron MG, Larsson SC, Markus HS. Clinical Significance of Magnetic Resonance Imaging Markers of Vascular Brain Injury: A Systematic Review and Meta-analysis. JAMA Neurol 2018.

Devan WJ, Falcone GJ, Anderson CD, Jagiella JM, Schmidt H, Hansen BM*, et al.* Heritability estimates identify a substantial genetic contribution to risk and outcome of intracerebral hemorrhage. Stroke 2013; 44(6): 1578-83.

Dichgans M, Pulit SL, Rosand J. Stroke genetics: discovery, biology, and clinical applications. Lancet Neurol 2019; 18(6): 587-99.

Duperron MG, Tzourio C, Sargurupremraj M, Mazoyer B, Soumare A, Schilling S*, et al.* Burden of Dilated Perivascular Spaces, an Emerging Marker of Cerebral Small Vessel Disease, Is Highly Heritable. Stroke 2018; 49(2): 282-7.

Edlow JA, Malek AM, Ogilvy CS. Aneurysmal subarachnoid hemorrhage: update for emergency physicians. J Emerg Med 2008; 34(3): 237-51.

Elliott P, Chambers JC, Zhang W, Clarke R, Hopewell JC, Peden JF*, et al.* Genetic Loci associated with C-reactive protein levels and risk of coronary heart disease. Jama 2009; 302(1): 37-48.

Emerging Risk Factors C, Sarwar N, Gao P, Seshasai SR, Gobin R, Kaptoge S*, et al.* Diabetes mellitus, fasting blood glucose concentration, and risk of vascular disease: a collaborative meta-analysis of 102 prospective studies. Lancet 2010; 375(9733): 2215-22.

Ettehad D, Emdin CA, Kiran A, Anderson SG, Callender T, Emberson J*, et al.* Blood pressure lowering for prevention of cardiovascular disease and death: a systematic review and meta-analysis. Lancet 2016; 387(10022): 957-67.

European Stroke Organisation Executive Committee, E. S. O. Writing Committee. Guidelines for management of ischaemic stroke and transient ischaemic attack 2008. Cerebrovasc Dis 2008; 25(5): 457-507.

Feigin VL, Forouzanfar MH, Krishnamurthi R, Mensah GA, Connor M, Bennett DA*, et al.* Global and regional burden of stroke during 1990-2010: findings from the Global Burden of Disease Study 2010. Lancet 2014; 383(9913): 245-54.

Feigin VL, Norrving B, George MG, Foltz JL, Roth GA, Mensah GA. Prevention of stroke: a strategic global imperative. Nat Rev Neurol 2016a; 12(9): 501-12.

Feigin VL, Norrving B, Mensah GA. Global Burden of Stroke. Circ Res 2017; 120(3): 439-48.

Feigin VL, Roth GA, Naghavi M, Parmar P, Krishnamurthi R, Chugh S*, et al.* Global burden of stroke and risk factors in 188 countries, during 1990-2013: a systematic analysis for the Global Burden of Disease Study 2013. Lancet Neurol 2016b; 15(9): 913-24.

Ference BA. How to use Mendelian randomization to anticipate the results of randomized trials. Eur Heart J 2018; 39(5): 360-2.

Ference BA, Ginsberg HN, Graham I, Ray KK, Packard CJ, Bruckert E*, et al.* Low-density lipoproteins cause atherosclerotic cardiovascular disease. 1. Evidence from genetic, epidemiologic, and clinical studies. A consensus statement from the European Atherosclerosis Society Consensus Panel. Eur Heart J 2017a; 38(32): 2459-72.

Ference BA, Graham I, Tokgozoglu L, Catapano AL. Impact of Lipids on Cardiovascular Health: JACC Health Promotion Series. J Am Coll Cardiol 2018; 72(10): 1141-56.

Ference BA, Kastelein JJP, Ginsberg HN, Chapman MJ, Nicholls SJ, Ray KK*, et al.* Association of Genetic Variants Related to CETP Inhibitors and Statins With Lipoprotein Levels and Cardiovascular Risk. JAMA 2017b; 318(10): 947-56.

Ference BA, Majeed F, Penumetcha R, Flack JM, Brook RD. Effect of naturally random allocation to lower low-density lipoprotein cholesterol on the risk of coronary heart disease mediated by polymorphisms in NPC1L1, HMGCR, or both: a 2 x 2 factorial Mendelian randomization study. J Am Coll Cardiol 2015; 65(15): 1552-61.

Ference BA, Ray KK, Catapano AL, Ference TB, Burgess S, Neff DR*, et al.* Mendelian Randomization Study of ACLY and Cardiovascular Disease. N Engl J Med 2019; 380(11): 1033-42.

Fewell Z, Davey Smith G, Sterne JA. The impact of residual and unmeasured confounding in epidemiologic studies: a simulation study. American journal of epidemiology 2007; 166(6): 646-55.

Fisher CM. The arterial lesions underlying lacunes. Acta Neuropathol 1968; 12(1): 1-15.

Fisher CM. Thalamic pure sensory stroke: a pathologic study. Neurology 1978; 28(11): 1141-4.

Fisher CM. Capsular infarcts: the underlying vascular lesions. Arch Neurol 1979; 36(2): 65-73.

Flossmann E, Schulz UG, Rothwell PM. Systematic review of methods and results of studies of the genetic epidemiology of ischemic stroke. Stroke 2004; 35(1): 212-27.

Fornage M, Chiang YA, O'Meara ES, Psaty BM, Reiner AP, Siscovick DS*, et al.* Biomarkers of Inflammation and MRI-Defined Small Vessel Disease of the Brain: The Cardiovascular Health Study. Stroke 2008; 39(7): 1952-9.

Forouzanfar MH, Liu P, Roth GA, Ng M, Biryukov S, Marczak L*, et al.* Global Burden of Hypertension and Systolic Blood Pressure of at Least 110 to 115 mm Hg, 1990-2015. JAMA 2017; 317(2): 165-82.

Fung KY, Wang C, Nyegaard S, Heit B, Fairn GD, Lee WL. SR-BI Mediated Transcytosis of HDL in Brain Microvascular Endothelial Cells Is Independent of Caveolin, Clathrin, and PDZK1. Front Physiol 2017; 8: 841.

G. B. D. Causes of Death Collaborators. Global, regional, and national age-sex specific mortality for 264 causes of death, 1980-2016: a systematic analysis for the Global Burden of Disease Study 2016. Lancet 2017; 390(10100): 1151-210.

G. B. D. DALYs Hale Collaborators. Global, regional, and national disability-adjusted life-years (DALYs) for 333 diseases and injuries and healthy life expectancy (HALE) for 195 countries and territories, 1990-2016: a systematic analysis for the Global Burden of Disease Study 2016. Lancet 2017; 390(10100): 1260-344.

G. B. D. Lifetime Risk of Stroke Collaborators, Feigin VL, Nguyen G, Cercy K, Johnson CO, Alam T*, et al.* Global, Regional, and Country-Specific Lifetime Risks of Stroke, 1990 and 2016. N Engl J Med 2018; 379(25): 2429-37.

G. B. D. Mortality Collaborators. Global, regional, and national under-5 mortality, adult mortality, age-specific mortality, and life expectancy, 1970-2016: a systematic analysis for the Global Burden of Disease Study 2016. Lancet 2017; 390(10100): 1084-150.

G. B. D. Stroke Collaborators. Global, regional, and national burden of stroke, 1990-2016: a systematic analysis for the Global Burden of Disease Study 2016. Lancet Neurol 2019; 18(5): 439-58.

Gaede P, Lund-Andersen H, Parving HH, Pedersen O. Effect of a multifactorial intervention on mortality in type 2 diabetes. N Engl J Med 2008; 358(6): 580-91.

Garner B, Waldeck AR, Witting PK, Rye KA, Stocker R. Oxidation of high density lipoproteins. II. Evidence for direct reduction of lipid hydroperoxides by methionine residues of apolipoproteins AI and AII. J Biol Chem 1998; 273(11): 6088-95.

Georgakis MK, Duering M, Wardlaw JM, Dichgans M. WMH and long-term outcomes in ischemic stroke: A systematic review and meta-analysis. Neurology 2019; 92(12): e1298-e308.

Gilbert J, Lekstrom-Himes J, Donaldson D, Lee Y, Hu M, Xu J*, et al.* Effect of CC chemokine receptor 2 CCR2 blockade on serum C-reactive protein in individuals at atherosclerotic risk and with a single nucleotide polymorphism of the monocyte chemoattractant protein-1 promoter region. Am J Cardiol 2011; 107(6): 906-11.

Gill D, Georgakis MK, Koskeridis F, Jiang L, Feng Q, Wei WQ*, et al.* Use of Genetic Variants Related to Antihypertensive Drugs to Inform on Efficacy and Side Effects. Circulation 2019; 140(4): 270-9.

Go AS, Hylek EM, Phillips KA, Chang Y, Henault LE, Selby JV*, et al.* Prevalence of diagnosed atrial fibrillation in adults: national implications for rhythm management and stroke prevention: the AnTicoagulation and Risk Factors in Atrial Fibrillation (ATRIA) Study. JAMA 2001; 285(18): 2370-5.

Goldstein LB, Amarenco P, Szarek M, Callahan A, 3rd, Hennerici M, Sillesen H*, et al.* Hemorrhagic stroke in the Stroke Prevention by Aggressive Reduction in Cholesterol Levels study. Neurology 2008; 70(24 Pt 2): 2364-70.

Grassia G, Maddaluno M, Guglielmotti A, Mangano G, Biondi G, Maffia P*, et al.* The anti-inflammatory agent bindarit inhibits neointima formation in both rats and hyperlipidaemic mice. Cardiovasc Res 2009; 84(3): 485-93.

Group SPSS, Benavente OR, Coffey CS, Conwit R, Hart RG, McClure LA*, et al.* Blood-pressure targets in patients with recent lacunar stroke: the SPS3 randomised trial. Lancet 2013; 382(9891): 507-15.

Gu L, Okada Y, Clinton SK, Gerard C, Sukhova GK, Libby P*, et al.* Absence of monocyte chemoattractant protein-1 reduces atherosclerosis in low density lipoprotein receptor-deficient mice. Mol Cell 1998; 2(2): 275-81.

Haffner C, Malik R, Dichgans M. Genetic factors in cerebral small vessel disease and their impact on stroke and dementia. J Cereb Blood Flow Metab 2016; 36(1): 158-71.

Hankey GJ. Stroke. Lancet 2017; 389(10069): 641-54.

Hansson GK. Inflammation, atherosclerosis, and coronary artery disease. N Engl J Med 2005; 352(16): 1685-95.

Harrison RK. Phase II and phase III failures: 2013-2015. Nature reviews Drug discovery 2016; 15(12): 817-8.

Holmes MV, Ala-Korpela M, Smith GD. Mendelian randomization in cardiometabolic disease: challenges in evaluating causality. Nature reviews Cardiology 2017; 14(10): 577-90.

Holmes MV, Dale CE, Zuccolo L, Silverwood RJ, Guo Y, Ye Z*, et al.* Association between alcohol and cardiovascular disease: Mendelian randomisation analysis based on individual participant data. Bmj 2014; 349: g4164.

Hoshi T, Kitagawa K, Yamagami H, Furukado S, Hougaku H, Hori M. Relations of serum high-sensitivity C-reactive protein and interleukin-6 levels with silent brain infarction. Stroke 2005; 36(4): 768-72.

Hosomi N, Nagai Y, Kohriyama T, Ohtsuki T, Aoki S, Nezu T*, et al.* The Japan Statin Treatment Against Recurrent Stroke (J-STARS): A Multicenter, Randomized, Open-label, Parallel-group Study. EBioMedicine 2015; 2(9): 1071-8.

HPS3/TIMI55–REVEAL Collaborative Group, Bowman L, Hopewell JC, Chen F, Wallendszus K, Stevens W*, et al.* Effects of Anacetrapib in Patients with Atherosclerotic Vascular Disease. N Engl J Med 2017; 377(13): 1217-27.

Huber SA, Sakkinen P, Conze D, Hardin N, Tracy R. Interleukin-6 exacerbates early atherosclerosis in mice. Arteriosclerosis, thrombosis, and vascular biology 1999; 19(10): 2364-7.

Iacoviello L, Bonaccio M, Cairella G, Catani MV, Costanzo S, D'Elia L*, et al.* Diet and primary prevention of stroke: Systematic review and dietary recommendations by the ad hoc Working Group of the Italian Society of Human Nutrition. Nutr Metab Cardiovasc Dis 2018; 28(4): 309-34.

Ikeda U, Ikeda M, Oohara T, Oguchi A, Kamitani T, Tsuruya Y*, et al.* Interleukin 6 stimulates growth of vascular smooth muscle cells in a PDGF-dependent manner. Am J Physiol 1991; 260(5 Pt 2): H1713-7.

Il R. Genetics Consortium Emerging Risk Factors Collaboration, Sarwar N, Butterworth AS, Freitag DF, Gregson J, Willeit P*, et al.* Interleukin-6 receptor pathways in coronary heart disease: a collaborative meta-analysis of 82 studies. Lancet 2012; 379(9822): 1205-13.

Inoue S, Egashira K, Ni W, Kitamoto S, Usui M, Otani K*, et al.* Anti-monocyte chemoattractant protein-1 gene therapy limits progression and destabilization of established atherosclerosis in apolipoprotein E-knockout mice. Circulation 2002; 106(21): 2700-6.

Intercollegiate Stroke Working Party. National clinical guideline for stroke. 5th Ed. ed. London: Royal College of Physicians; 2016.

Interleukin-6 Receptor Mendelian Randomisation Analysis Consortium, Swerdlow DI, Holmes MV, Kuchenbaecker KB, Engmann JE, Shah T*, et al.* The interleukin-6 receptor as a target for prevention of coronary heart disease: a mendelian randomisation analysis. Lancet 2012; 379(9822): 1214-24.

Inzitari D, Pracucci G, Poggesi A, Carlucci G, Barkhof F, Chabriat H*, et al.* Changes in white matter as determinant of global functional decline in older independent outpatients: three year follow-up of LADIS (leukoaraiosis and disability) study cohort. BMJ 2009; 339: b2477.

Jackson C, Sudlow C. Are lacunar strokes really different? A systematic review of differences in risk factor profiles between lacunar and nonlacunar infarcts. Stroke 2005; 36(4): 891-901.

Jackson CA, Sudlow CL. Is hypertension a more frequent risk factor for deep than for lobar supratentorial intracerebral haemorrhage? J Neurol Neurosurg Psychiatry 2006; 77(11): 1244-52.

Joutel A, Corpechot C, Ducros A, Vahedi K, Chabriat H, Mouton P*, et al.* Notch3 mutations in CADASIL, a hereditary adult-onset condition causing stroke and dementia. Nature 1996; 383(6602): 707-10.

Kamel H, Healey JS. Cardioembolic Stroke. Circ Res 2017; 120(3): 514-26.

Kamel H, Iadecola C. Brain-immune interactions and ischemic stroke: clinical implications. Arch Neurol 2012; 69(5): 576-81.

Kaptoge S, Di Angelantonio E, Lowe G, Pepys MB, Thompson SG, Collins R*, et al.* C-reactive protein concentration and risk of coronary heart disease, stroke, and mortality: an individual participant meta-analysis. Lancet 2010; 375(9709): 132-40.

Kaptoge S, Seshasai SR, Gao P, Freitag DF, Butterworth AS, Borglykke A*, et al.* Inflammatory cytokines and risk of coronary heart disease: new prospective study and updated meta-analysis. Eur Heart J 2014; 35(9): 578-89.

Kernan WN, Ovbiagele B, Black HR, Bravata DM, Chimowitz MI, Ezekowitz MD*, et al.* Guidelines for the prevention of stroke in patients with stroke and transient ischemic attack: a guideline for healthcare professionals from the American Heart Association/American Stroke Association. Stroke 2014; 45(7): 2160-236.

Khakoo AY, Yurgin NR, Eisenberg PR, Fonarow GC. Overcoming Barriers to Development of Novel Therapies for Cardiovascular Disease: Insights From the Oncology Drug Development Experience. JACC Basic to translational science 2019; 4(2): 269-74.

Kivimaki M, Singh-Manoux A, Pentti J, Sabia S, Nyberg ST, Alfredsson L*, et al.* Physical inactivity, cardiometabolic disease, and risk of dementia: an individual-participant meta-analysis. BMJ 2019; 365: l1495.

Knudsen KA, Rosand J, Karluk D, Greenberg SM. Clinical diagnosis of cerebral amyloid angiopathy: validation of the Boston criteria. Neurology 2001; 56(4): 537-9.

Kolominsky-Rabas PL, Weber M, Gefeller O, Neundoerfer B, Heuschmann PU. Epidemiology of ischemic stroke subtypes according to TOAST criteria: incidence, recurrence, and long-term survival in ischemic stroke subtypes: a population-based study. Stroke 2001; 32(12): 2735-40.

Koton S, Schneider AL, Rosamond WD, Shahar E, Sang Y, Gottesman RF*, et al.* Stroke incidence and mortality trends in US communities, 1987 to 2011. JAMA 2014; 312(3): 259-68.

Lacey B, Lewington S, Clarke R, Kong XL, Chen Y, Guo Y*, et al.* Age-specific association between blood pressure and vascular and non-vascular chronic diseases in 0.5 million adults in China: a prospective cohort study. Lancet Glob Health 2018; 6(6): e641-e9.

Lammie GA. Hypertensive cerebral small vessel disease and stroke. Brain Pathol 2002; 12(3): 358-70.

Lapergue B, Moreno JA, Dang BQ, Coutard M, Delbosc S, Raphaeli G*, et al.* Protective effect of high-density lipoprotein-based therapy in a model of embolic stroke. Stroke 2010; 41(7): 1536-42.

Larsson SC, Burgess S, Michaelsson K. Smoking and stroke: A mendelian randomization study. Ann Neurol 2019; 86(3): 468-71.

Lawton MT, Vates GE. Subarachnoid Hemorrhage. N Engl J Med 2017; 377(3): 257-66.

Lee CM, Chien CT, Chang PY, Hsieh MY, Jui HY, Liau CS*, et al.* High-density lipoprotein antagonizes oxidized low-density lipoprotein by suppressing oxygen free-radical formation and preserving nitric oxide bioactivity. Atherosclerosis 2005; 183(2): 251-8.

Lee M, Saver JL, Hong KS, Song S, Chang KH, Ovbiagele B. Effect of pre-diabetes on future risk of stroke: meta-analysis. BMJ 2012; 344: e3564.

Leening MJ, Ferket BS, Steyerberg EW, Kavousi M, Deckers JW, Nieboer D*, et al.* Sex differences in lifetime risk and first manifestation of cardiovascular disease: prospective population based cohort study. BMJ 2014; 349: g5992.

Lewington S, Clarke R, Qizilbash N, Peto R, Collins R, Prospective Studies C. Age-specific relevance of usual blood pressure to vascular mortality: a meta-analysis of individual data for one million adults in 61 prospective studies. Lancet 2002; 360(9349): 1903-13.

Li L, Yiin GS, Geraghty OC, Schulz UG, Kuker W, Mehta Z*, et al.* Incidence, outcome, risk factors, and long-term prognosis of cryptogenic transient ischaemic attack and ischaemic stroke: a population-based study. Lancet Neurol 2015; 14(9): 903-13.

Libby P, Ridker PM, Hansson GK. Progress and challenges in translating the biology of atherosclerosis. Nature 2011; 473(7347): 317-25.

Liehn EA, Piccinini AM, Koenen RR, Soehnlein O, Adage T, Fatu R*, et al.* A new monocyte chemotactic protein-1/chemokine CC motif ligand-2 competitor limiting neointima formation and myocardial ischemia/reperfusion injury in mice. J Am Coll Cardiol 2010; 56(22): 1847-57.

Ligthart S, Vaez A, Vosa U, Stathopoulou MG, de Vries PS, Prins BP*, et al.* Genome Analyses of >200,000 Individuals Identify 58 Loci for Chronic Inflammation and Highlight Pathways that Link Inflammation and Complex Disorders. Am J Hum Genet 2018; 103(5): 691-706.

Lin HJ, Wolf PA, Kelly-Hayes M, Beiser AS, Kase CS, Benjamin EJ*, et al.* Stroke severity in atrial fibrillation. The Framingham Study. Stroke 1996; 27(10): 1760-4.

Lin J, Kakkar V, Lu X. Impact of MCP-1 in atherosclerosis. Curr Pharm Des 2014; 20(28): 4580-8.

Lincoff AM, Nicholls SJ, Riesmeyer JS, Barter PJ, Brewer HB, Fox KAA*, et al.* Evacetrapib and Cardiovascular Outcomes in High-Risk Vascular Disease. N Engl J Med 2017; 376(20): 1933-42.

Linn FH, Wijdicks EF, van der Graaf Y, Weerdesteyn-van Vliet FA, Bartelds AI, van Gijn J. Prospective study of sentinel headache in aneurysmal subarachnoid haemorrhage. Lancet 1994; 344(8922): 590-3.

Linn J, Halpin A, Demaerel P, Ruhland J, Giese AD, Dichgans M*, et al.* Prevalence of superficial siderosis in patients with cerebral amyloid angiopathy. Neurology 2010; 74(17): 1346-50.

Lovelock CE, Rinkel GJ, Rothwell PM. Time trends in outcome of subarachnoid hemorrhage: Population-based study and systematic review. Neurology 2010; 74(19): 1494-501.

Low A, Mak E, Rowe JB, Markus HS, O'Brien JT. Inflammation and cerebral small vessel disease: A systematic review. Ageing Res Rev 2019; 53: 100916.

Lutgens E, Faber B, Schapira K, Evelo CT, van Haaften R, Heeneman S*, et al.* Gene profiling in atherosclerosis reveals a key role for small inducible cytokines: validation using a novel monocyte chemoattractant protein monoclonal antibody. Circulation 2005; 111(25): 3443-52.

Malik R, Chauhan G, Traylor M, Sargurupremraj M, Okada Y, Mishra A*, et al.* Multiancestry genome-wide association study of 520,000 subjects identifies 32 loci associated with stroke and stroke subtypes. Nat Genet 2018a; 50(4): 524-37.

Malik R, Rannikmae K, Traylor M, Georgakis MK, Sargurupremraj M, Markus HS*, et al.* Genome-wide meta-analysis identifies 3 novel loci associated with stroke. Ann Neurol 2018b; 84(6): 934-9.

Manktelow BN, Potter JF. Interventions in the management of serum lipids for preventing stroke recurrence. Cochrane Database Syst Rev 2009(3): CD002091.

Martin SS, Jones SR, Toth PP. High-density lipoprotein subfractions: current views and clinical practice applications. Trends Endocrinol Metab 2014; 25(7): 329-36.

Martini SR, Flaherty ML, Brown WM, Haverbusch M, Comeau ME, Sauerbeck LR*, et al.* Risk factors for intracerebral hemorrhage differ according to hemorrhage location. Neurology 2012; 79(23): 2275-82.

Meschia JF, Bushnell C, Boden-Albala B, Braun LT, Bravata DM, Chaturvedi S*, et al.* Guidelines for the primary prevention of stroke: a statement for healthcare professionals from the American Heart Association/American Stroke Association. Stroke 2014; 45(12): 3754-832.

Mestre H, Tithof J, Du T, Song W, Peng W, Sweeney AM*, et al.* Flow of cerebrospinal fluid is driven by arterial pulsations and is reduced in hypertension. Nat Commun 2018; 9(1): 4878.

Millwood IY, Walters RG, Mei XW, Guo Y, Yang L, Bian Z*, et al.* Conventional and genetic evidence on alcohol and vascular disease aetiology: a prospective study of 500 000 men and women in China. Lancet 2019a; 393(10183): 1831-42.

Millwood IY, Walters RG, Mei XW, Guo Y, Yang L, Bian Z*, et al.* Conventional and genetic evidence on alcohol and vascular disease aetiology: a prospective study of 500 000 men and women in China. Lancet 2019b; 393(10183): 1831-42.

Mokry LE, Ahmad O, Forgetta V, Thanassoulis G, Richards JB. Mendelian randomisation applied to drug development in cardiovascular disease: a review. Journal of medical genetics 2015; 52(2): 71-9.

Monette JS, Hutchins PM, Ronsein GE, Wimberger J, Irwin AD, Tang C*, et al.* Patients With Coronary Endothelial Dysfunction Have Impaired Cholesterol Efflux Capacity and Reduced HDL Particle Concentration. Circ Res 2016; 119(1): 83-90.

Muehlschlegel S. Subarachnoid Hemorrhage. Continuum (Minneap Minn) 2018; 24(6): 1623-57.

Munoz Maniega S, Chappell FM, Valdes Hernandez MC, Armitage PA, Makin SD, Heye AK*, et al.* Integrity of normal-appearing white matter: Influence of age, visible lesion burden and hypertension in patients with small-vessel disease. J Cereb Blood Flow Metab 2017; 37(2): 644-56.

Murphy AJ, Woollard KJ, Hoang A, Mukhamedova N, Stirzaker RA, McCormick SP*, et al.* High-density lipoprotein reduces the human monocyte inflammatory response. Arteriosclerosis, thrombosis, and vascular biology 2008; 28(11): 2071-7.

Nation DA, Sweeney MD, Montagne A, Sagare AP, D'Orazio LM, Pachicano M*, et al.* Blood-brain barrier breakdown is an early biomarker of human cognitive dysfunction. Nat Med 2019; 25(2): 270-6.

Nelken NA, Coughlin SR, Gordon D, Wilcox JN. Monocyte chemoattractant protein-1 in human atheromatous plaques. J Clin Invest 1991; 88(4): 1121-7.

Nicholls SJ, Dusting GJ, Cutri B, Bao S, Drummond GR, Rye KA*, et al.* Reconstituted high-density lipoproteins inhibit the acute pro-oxidant and proinflammatory vascular changes induced by a periarterial collar in normocholesterolemic rabbits. Circulation 2005; 111(12): 1543-50.

Nieuwkamp DJ, Setz LE, Algra A, Linn FH, de Rooij NK, Rinkel GJ. Changes in case fatality of aneurysmal subarachnoid haemorrhage over time, according to age, sex, and region: a meta-analysis. Lancet Neurol 2009; 8(7): 635-42.

Noels H, Weber C, Koenen RR. Chemokines as Therapeutic Targets in Cardiovascular Disease. Arteriosclerosis, thrombosis, and vascular biology 2019; 39(4): 583-92.

Nofer JR, van der Giet M, Tolle M, Wolinska I, von Wnuck Lipinski K, Baba HA*, et al.* HDL induces NO-dependent vasorelaxation via the lysophospholipid receptor S1P3. J Clin Invest 2004; 113(4): 569-81.

O'Donnell MJ, Chin SL, Rangarajan S, Xavier D, Liu L, Zhang H*, et al.* Global and regional effects of potentially modifiable risk factors associated with acute stroke in 32 countries (INTERSTROKE): a case-control study. Lancet 2016; 388(10046): 761-75.

O'Donnell MJ, Xavier D, Liu L, Zhang H, Chin SL, Rao-Melacini P*, et al.* Risk factors for ischaemic and intracerebral haemorrhagic stroke in 22 countries (the INTERSTROKE study): a case-control study. Lancet 2010; 376(9735): 112-23.

Ohira T, Shahar E, Chambless LE, Rosamond WD, Mosley TH, Jr., Folsom AR. Risk factors for ischemic stroke subtypes: the Atherosclerosis Risk in Communities study. Stroke 2006; 37(10): 2493-8.

Okamoto M, Fuchigami M, Suzuki T, Watanabe N. A novel C-C chemokine receptor 2 antagonist prevents progression of albuminuria and atherosclerosis in mouse models. Biol Pharm Bull 2012; 35(11): 2069-74.

Pan B, Jin X, Jun L, Qiu S, Zheng Q, Pan M. The relationship between smoking and stroke: A meta-analysis. Medicine (Baltimore) 2019; 98(12): e14872.

Pantoni L. Cerebral small vessel disease: from pathogenesis and clinical characteristics to therapeutic challenges. Lancet Neurol 2010; 9(7): 689-701.

Pendlebury ST, Rothwell PM, Oxford Vascular S. Incidence and prevalence of dementia associated with transient ischaemic attack and stroke: analysis of the population-based Oxford Vascular Study. Lancet Neurol 2019; 18(3): 248-58.

Petty GW, Brown RD, Jr., Whisnant JP, Sicks JD, O'Fallon WM, Wiebers DO. Ischemic stroke subtypes: a population-based study of incidence and risk factors. Stroke 1999; 30(12): 2513-6.

Poggesi A, Pracucci G, Chabriat H, Erkinjuntti T, Fazekas F, Verdelho A*, et al.* Urinary complaints in nondisabled elderly people with age-related white matter changes: the Leukoaraiosis And DISability (LADIS) Study. J Am Geriatr Soc 2008; 56(9): 1638-43.

Prins BP, Abbasi A, Wong A, Vaez A, Nolte I, Franceschini N*, et al.* Investigating the Causal Relationship of C-Reactive Protein with 32 Complex Somatic and Psychiatric Outcomes: A Large-Scale Cross-Consortium Mendelian Randomization Study. PLoS medicine 2016; 13(6): e1001976.

Prosser HC, Ng MK, Bursill CA. The role of cholesterol efflux in mechanisms of endothelial protection by HDL. Curr Opin Lipidol 2012; 23(3): 182-9.

Psaltopoulou T, Sergentanis TN, Panagiotakos DB, Sergentanis IN, Kosti R, Scarmeas N. Mediterranean diet, stroke, cognitive impairment, and depression: A meta-analysis. Ann Neurol 2013; 74(4): 580-91.

Qureshi AI, Mendelow AD, Hanley DF. Intracerebral haemorrhage. Lancet 2009; 373(9675): 1632-44.

Qureshi AI, Tuhrim S, Broderick JP, Batjer HH, Hondo H, Hanley DF. Spontaneous intracerebral hemorrhage. N Engl J Med 2001; 344(19): 1450-60.

Rajani RM, Quick S, Ruigrok SR, Graham D, Harris SE, Verhaaren BFJ, *et al.* Reversal of endothelial dysfunction reduces white matter vulnerability in cerebral small vessel disease in rats. Sci Transl Med 2018; 10(448).

Ridker PM. From C-Reactive Protein to Interleukin-6 to Interleukin-1: Moving Upstream To Identify Novel Targets for Atheroprotection. Circ Res 2016; 118(1): 145-56.

Ridker PM. How Common Is Residual Inflammatory Risk? Circ Res 2017; 120(4): 617-9.

Ridker PM. Anticytokine Agents: Targeting Interleukin Signaling Pathways for the Treatment of Atherothrombosis. Circ Res 2019; 124(3): 437-50.

Ridker PM, Danielson E, Fonseca FA, Genest J, Gotto AM, Jr., Kastelein JJ, *et al.* Rosuvastatin to prevent vascular events in men and women with elevated C-reactive protein. N Engl J Med 2008; 359(21): 2195-207.

Ridker PM, Everett BM, Pradhan A, MacFadyen JG, Solomon DH, Zaharris E, *et al.* Low-Dose Methotrexate for the Prevention of Atherosclerotic Events. N Engl J Med 2019a; 380(8): 752-62.

Ridker PM, Everett BM, Thuren T, MacFadyen JG, Chang WH, Ballantyne C, *et al.* Antiinflammatory Therapy with Canakinumab for Atherosclerotic Disease. N Engl J Med 2017; 377(12): 1119-31.

Ridker PM, Libby P, MacFadyen JG, Thuren T, Ballantyne C, Fonseca F, *et al.* Modulation of the interleukin-6 signalling pathway and incidence rates of atherosclerotic events and all-cause mortality: analyses from the Canakinumab Anti-Inflammatory Thrombosis Outcomes Study (CANTOS). Eur Heart J 2018a; 39(38): 3499-507.

Ridker PM, MacFadyen JG, Everett BM, Libby P, Thuren T, Glynn RJ, *et al.* Relationship of C-reactive protein reduction to cardiovascular event reduction following treatment with canakinumab: a secondary analysis from the CANTOS randomised controlled trial. Lancet 2018b; 391(10118): 319-28.

Ridker PM, MacFadyen JG, Thuren T, Libby P. Residual inflammatory risk associated with interleukin-18 and interleukin-6 after successful interleukin-1beta inhibition with canakinumab: further rationale for the development of targeted anti-cytokine therapies for the treatment of atherothrombosis. Eur Heart J 2019b.

Ridker PM, Rifai N, Stampfer MJ, Hennekens CH. Plasma concentration of interleukin-6 and the risk of future myocardial infarction among apparently healthy men. Circulation 2000; 101(15): 1767-72.

Roberts R. Mendelian Randomization Studies Promise to Shorten the Journey to FDA Approval. JACC Basic Transl Sci 2018; 3(5): 690-703.

Rodrigues MA, Samarasekera N, Lerpiniere C, Humphreys C, McCarron MO, White PM, *et al.* The Edinburgh CT and genetic diagnostic criteria for lobar intracerebral haemorrhage associated with cerebral amyloid angiopathy: model development and diagnostic test accuracy study. Lancet Neurol 2018; 17(3): 232-40.

Rothwell PM, Howard SC, Dolan E, O'Brien E, Dobson JE, Dahlof B, *et al.* Effects of beta blockers and calcium-channel blockers on within-individual variability in blood pressure and risk of stroke. Lancet Neurol 2010; 9(5): 469-80.

Sacco RL, Kasner SE, Broderick JP, Caplan LR, Connors JJ, Culebras A*, et al.* An updated definition of stroke for the 21st century: a statement for healthcare professionals from the American Heart Association/American Stroke Association. Stroke 2013; 44(7): 2064-89.

Satizabal CL, Zhu YC, Mazoyer B, Dufouil C, Tzourio C. Circulating IL-6 and CRP are associated with MRI findings in the elderly: the 3C-Dijon Study. Neurology 2012; 78(10): 720-7.

Scheller J, Chalaris A, Schmidt-Arras D, Rose-John S. The pro- and anti-inflammatory properties of the cytokine interleukin-6. Biochim Biophys Acta 2011; 1813(5): 878-88.

Schneider AT, Kissela B, Woo D, Kleindorfer D, Alwell K, Miller R*, et al.* Ischemic stroke subtypes: a population-based study of incidence rates among blacks and whites. Stroke 2004; 35(7): 1552-6.

Schulz UG, Rothwell PM. Differences in vascular risk factors between etiological subtypes of ischemic stroke: importance of population-based studies. Stroke 2003; 34(8): 2050-9.

Schwartz GG, Olsson AG, Abt M, Ballantyne CM, Barter PJ, Brumm J*, et al.* Effects of dalcetrapib in patients with a recent acute coronary syndrome. N Engl J Med 2012; 367(22): 2089-99.

Scott LJ. Tocilizumab: A Review in Rheumatoid Arthritis. Drugs 2017; 77(17): 1865-79.

Shi Y, Thrippleton MJ, Blair GW, Dickie DA, Marshall I, Hamilton I*, et al.* Small vessel disease is associated with altered cerebrovascular pulsatility but not resting cerebral blood flow. J Cereb Blood Flow Metab 2018: 271678X18803956.

Shi Y, Thrippleton MJ, Makin SD, Marshall I, Geerlings MI, de Craen AJM*, et al.* Cerebral blood flow in small vessel disease: A systematic review and meta-analysis. J Cereb Blood Flow Metab 2016; 36(10): 1653-67.

Shoamanesh A, Preis SR, Beiser AS, Vasan RS, Benjamin EJ, Kase CS*, et al.* Inflammatory biomarkers, cerebral microbleeds, and small vessel disease: Framingham Heart Study. Neurology 2015; 84(8): 825-32.

Smith GD, Lawlor DA, Harbord R, Timpson N, Day I, Ebrahim S. Clustered environments and randomized genes: a fundamental distinction between conventional and genetic epidemiology. PLoS medicine 2007; 4(12): e352.

Sorrentino SA, Besler C, Rohrer L, Meyer M, Heinrich K, Bahlmann FH*, et al.* Endothelial-vasoprotective effects of high-density lipoprotein are impaired in patients with type 2 diabetes mellitus but are improved after extended-release niacin therapy. Circulation 2010; 121(1): 110-22.

Spieker LE, Sudano I, Hurlimann D, Lerch PG, Lang MG, Binggeli C*, et al.* High-density lipoprotein restores endothelial function in hypercholesterolemic men. Circulation 2002; 105(12): 1399-402.

Staszewski J, Piusinska-Macoch R, Brodacki B, Skrobowska E, Stepien A. IL-6, PF-4, sCD40 L, and homocysteine are associated with the radiological progression of cerebral small-vessel disease: a 2-year follow-up study. Clin Interv Aging 2018; 13: 1135-41.

Stone JH, Tuckwell K, Dimonaco S, Klearman M, Aringer M, Blockmans D*, et al.* Trial of Tocilizumab in Giant-Cell Arteritis. N Engl J Med 2017; 377(4): 317-28.

Strazzullo P, D'Elia L, Cairella G, Garbagnati F, Cappuccio FP, Scalfi L. Excess body weight and incidence of stroke: meta-analysis of prospective studies with 2 million participants. Stroke 2010; 41(5): e418-26.

Stroke Foundation. Clinical Guidelines for Stroke Management Melbourne, Australia; 2017.

Sudlow CL, Warlow CP. Comparable studies of the incidence of stroke and its pathological types: results from an international collaboration. International Stroke Incidence Collaboration. Stroke 1997; 28(3): 491-9.

Sui X, Lavie CJ, Hooker SP, Lee DC, Colabianchi N, Lee CD*, et al.* A prospective study of fasting plasma glucose and risk of stroke in asymptomatic men. Mayo Clin Proc 2011; 86(11): 1042-9.

Sun L, Clarke R, Bennett D, Guo Y, Walters RG, Hill M*, et al.* Causal associations of blood lipids with risk of ischemic stroke and intracerebral hemorrhage in Chinese adults. Nat Med 2019; 25(4): 569-74.

Terasaka N, Wang N, Yvan-Charvet L, Tall AR. High-density lipoprotein protects macrophages from oxidized low-density lipoprotein-induced apoptosis by promoting efflux of 7-ketocholesterol via ABCG1. Proc Natl Acad Sci U S A 2007; 104(38): 15093-8.

Thrift AG, Thayabaranathan T, Howard G, Howard VJ, Rothwell PM, Feigin VL*, et al.* Global stroke statistics. Int J Stroke 2017; 12(1): 13-32.

Tikk K, Sookthai D, Monni S, Gross ML, Lichy C, Kloss M*, et al.* Primary preventive potential for stroke by avoidance of major lifestyle risk factors: the European Prospective Investigation into Cancer and Nutrition-Heidelberg cohort. Stroke 2014; 45(7): 2041-6.

Towfighi A, Ovbiagele B, El Husseini N, Hackett ML, Jorge RE, Kissela BM*, et al.* Poststroke Depression: A Scientific Statement for Healthcare Professionals From the American Heart Association/American Stroke Association. Stroke 2017; 48(2): e30-e43.

Tran-Dinh A, Diallo D, Delbosc S, Varela-Perez LM, Dang QB, Lapergue B*, et al.* HDL and endothelial protection. Br J Pharmacol 2013; 169(3): 493-511.

Traylor M, Rutten-Jacobs LC, Thijs V, Holliday EG, Levi C, Bevan S*, et al.* Genetic Associations With White Matter Hyperintensities Confer Risk of Lacunar Stroke. Stroke 2016; 47(5): 1174-9.

Traylor M, Tozer DJ, Croall ID, Lisiecka-Ford DM, Olorunda AO, Boncoraglio G*, et al.* Genetic variation in PLEKHG1 is associated with white matter hyperintensities (n = 11,226). Neurology 2019; 92(8): e749-e57.

van Agtmaal MJM, Houben A, Pouwer F, Stehouwer CDA, Schram MT. Association of Microvascular Dysfunction With Late-Life Depression: A Systematic Review and Meta-analysis. JAMA Psychiatry 2017; 74(7): 729-39.

van Asch CJ, Luitse MJ, Rinkel GJ, van der Tweel I, Algra A, Klijn CJ. Incidence, case fatality, and functional outcome of intracerebral haemorrhage over time, according to age, sex, and ethnic origin: a systematic review and meta-analysis. Lancet Neurol 2010; 9(2): 167-76.

van Middelaar T, Argillander TE, Schreuder F, Deinum J, Richard E, Klijn CJM. Effect of Antihypertensive Medication on Cerebral Small Vessel Disease: A Systematic Review and Meta-Analysis. Stroke 2018; 49(6): 1531-3.

Vangen-Lonne AM, Wilsgaard T, Johnsen SH, Lochen ML, Njolstad I, Mathiesen EB. Declining Incidence of Ischemic Stroke: What Is the Impact of Changing Risk Factors? The Tromso Study 1995 to 2012. Stroke 2017; 48(3): 544-50.

Verhaaren BF, Debette S, Bis JC, Smith JA, Ikram MK, Adams HH*, et al.* Multiethnic genome-wide association study of cerebral white matter hyperintensities on MRI. Circ Cardiovasc Genet 2015; 8(2): 398-409.

Vinters HV, Gilbert JJ. Cerebral amyloid angiopathy: incidence and complications in the aging brain. II. The distribution of amyloid vascular changes. Stroke 1983; 14(6): 924-8.

Viswambharan H, Ming XF, Zhu S, Hubsch A, Lerch P, Vergeres G*, et al.* Reconstituted high-density lipoprotein inhibits thrombin-induced endothelial tissue factor expression through inhibition of RhoA and stimulation of phosphatidylinositol 3-kinase but not Akt/endothelial nitric oxide synthase. Circ Res 2004; 94(7): 918-25.

Wang X, Dong Y, Qi X, Huang C, Hou L. Cholesterol levels and risk of hemorrhagic stroke: a systematic review and meta-analysis. Stroke 2013; 44(7): 1833-9.

Wang Z, Soo YO, Mok VC. Cerebral microbleeds: is antithrombotic therapy safe to administer? Stroke 2014; 45(9): 2811-7.

Wardlaw JM, Makin SJ, Valdés Hernández MC, Armitage PA, Heye AK, Chappell FM*, et al.* Blood-brain barrier failure as a core mechanism in cerebral small vessel disease and dementia: evidence from a cohort study: Alzheimers Dement. 2017 Jun;13(6):634-43. doi: 10.1016/j.jalz.2016.09.006.

Wardlaw JM, Smith C, Dichgans M. Mechanisms of sporadic cerebral small vessel disease: insights from neuroimaging. Lancet Neurol 2013a; 12(5): 483-97.

Wardlaw JM, Smith C, Dichgans M. Small vessel disease: mechanisms and clinical implications. Lancet Neurol 2019; 18(7): 684-96.

Wardlaw JM, Smith EE, Biessels GJ, Cordonnier C, Fazekas F, Frayne R*, et al.* Neuroimaging standards for research into small vessel disease and its contribution to ageing and neurodegeneration. Lancet Neurol 2013b; 12(8): 822-38.

Webb AJ, Fischer U, Mehta Z, Rothwell PM. Effects of antihypertensive-drug class on interindividual variation in blood pressure and risk of stroke: a systematic review and meta-analysis. Lancet 2010; 375(9718): 906-15.

Williams PT. Low high-density lipoprotein 3 reduces the odds of men surviving to age 85 during 53-year follow-up. J Am Geriatr Soc 2012; 60(3): 430-6.

Wilson D, Ambler G, Shakeshaft C, Brown MM, Charidimou A, Al-Shahi Salman R*, et al.* Cerebral microbleeds and intracranial haemorrhage risk in patients anticoagulated for atrial fibrillation after acute ischaemic stroke or transient ischaemic attack (CROMIS-2): a multicentre observational cohort study. Lancet Neurol 2018; 17(6): 539-47.

Winter C, Silvestre-Roig C, Ortega-Gomez A, Lemnitzer P, Poelman H, Schumski A*, et al.* Chrono-pharmacological Targeting of the CCL2-CCR2 Axis Ameliorates Atherosclerosis. Cell Metab 2018; 28(1): 175-82 e5.

Wolf PA, Abbott RD, Kannel WB. Atrial fibrillation as an independent risk factor for stroke: the Framingham Study. Stroke 1991; 22(8): 983-8.

Xie X, Atkins E, Lv J, Bennett A, Neal B, Ninomiya T*, et al.* Effects of intensive blood pressure lowering on cardiovascular and renal outcomes: updated systematic review and meta-analysis. Lancet 2016; 387(10017): 435-43.

Yamaguchi Y, Wada M, Sato H, Nagasawa H, Koyama S, Takahashi Y*, et al.* Impact of ambulatory blood pressure variability on cerebral small vessel disease progression and cognitive decline in community-based elderly Japanese. Am J Hypertens 2014; 27(10): 1257-67.

Yamashita T, Kawashima S, Ozaki M, Namiki M, Inoue N, Hirata K*, et al.* Propagermanium reduces atherosclerosis in apolipoprotein E knockout mice via inhibition of macrophage infiltration. Arteriosclerosis, thrombosis, and vascular biology 2002; 22(6): 969-74.

Yin ZG, Wang QS, Yu K, Wang WW, Lin H, Yang ZH. Sex differences in associations between blood lipids and cerebral small vessel disease. Nutr Metab Cardiovasc Dis 2018; 28(1): 28-34.

Yoshida M, Tomitori H, Machi Y, Katagiri D, Ueda S, Horiguchi K*, et al.* Acrolein, IL-6 and CRP as markers of silent brain infarction. Atherosclerosis 2009; 203(2): 557-62.

Yu S, Yarnell JW, Sweetnam P, Bolton CH. High density lipoprotein subfractions and the risk of coronary heart disease: 9-years follow-up in the Caerphilly Study. Atherosclerosis 2003; 166(2): 331-8.

Yuhanna IS, Zhu Y, Cox BE, Hahner LD, Osborne-Lawrence S, Lu P*, et al.* High-density lipoprotein binding to scavenger receptor-BI activates endothelial nitric oxide synthase. Nat Med 2001; 7(7): 853-7.

Zacho J, Tybjaerg-Hansen A, Jensen JS, Grande P, Sillesen H, Nordestgaard BG. Genetically elevated C-reactive protein and ischemic vascular disease. N Engl J Med 2008; 359(18): 1897-908.

Zheng J, Baird D, Borges MC, Bowden J, Hemani G, Haycock P*, et al.* Recent Developments in Mendelian Randomization Studies. Current epidemiology reports 2017; 4(4): 330-45.

Zia E, Hedblad B, Pessah-Rasmussen H, Berglund G, Janzon L, Engstrom G. Blood pressure in relation to the incidence of cerebral infarction and intracerebral hemorrhage. Hypertensive hemorrhage: debated nomenclature is still relevant. Stroke 2007; 38(10): 2681-5.

ACKNOWLEDGEMENTS

Towards the end of this journey, I would like to thank a number of people who contributed with their skills and energy to the completion of the current PhD thesis. First of all, I would like to express my gratitude to my supervisor, Martin Dichgans, for believing in me and offering me the opportunity to work on these projects. In addition to providing me an excellent and inspiring working environment, Martin acted as my mentor during these years and also worked hard with me towards the completion of the projects included in this thesis. I would also like to deeply thank Rainer Malik, the closest of my collaborators during this period. Rainer overviewed my work on a daily basis, introduced me to the magnificent world of large-scale -omics analyses, supported me for the statistical analyses of the studies, and was always there to solve emerging problems and discuss new ideas with me. I learned more than I would have expected from both of them.

I would further like to thank the members of my thesis advisory committee (TAC), Stefan Lichtenhaler, Annette Peters, and Marco Duering for providing me with their expert inputs during and between the TAC meetings. During my stay at the Institute for Stroke and Dementia Research I had the opportunity to also collaborate with a number of people for projects related or not to this PhD thesis. These people include Jürgen Bernhagen, Frank Wollenweber, Vera Zietemann, Anna Kopczak, Steffen Tiedt, Elisabeth Völkl, and Rupert Aigner. In one way or another, all of them contributed towards the completion of my projects and I would like to express them my gratitude.

Moreover, I would like to acknowledge the key contributions of external collaborators. Dipender Gill from Imperial College London provided me with excellent advice regarding the technical aspects of Mendelian randomization and was always willing to discuss with me new ideas, methodological challenges, and new perspectives about the technical aspects of my work. For the purposes of some of the projects, I also had the opportunity to collaborate with world-leading experts in specific fields related to cerebrovascular disease: specifically, I am really grateful for the key inputs provided by Alastair Webb, Chris Anderson, and Jemma Hopewell. I would like to thank the principal investigators and the analysts of the individual studies that agreed to contribute data to the meta-analysis about MCP-1 and stroke. They include: Harry Björkbacka and Jan Nilsson from the MDCS-CV study, Tiberiu Alexandru Pana and Phyo Myint from the EPIC-Norfolk Study,

Serkalem Demissie and Sudha Seshadri from the Framingham Heart Study, Colby Ayers and James de Lemos from the Dallas Heart Study, Mohamed Elhadad and Annette Peters from MONICA/KORA, and finally Myriam Fornage and Christie Ballantyne from ARIC.

I am further grateful to all the GWAS Consortia for making publicly their summary data publicly available. Specifically, we used data from MEGASTROKE, CardioGramPLUSC4D, FINRISK, CHARGE, INVENT, GLGC, ICBP, AFGen, and the UK Biobank. The current thesis is a proof for the benefits of data sharing and open science, as it would not have been possible to accomplish without data from these Consortia.

Furthermore, I would like to express my deepest gratitude to all the members of the Faculty and the Administration Boards of the Graduate School of Systemic Neurosciences. They provided all of us with free access to all the resources available from LMU Munich (courses, lectures, labs, supervision) and tremendous physical, but also financial support to complete our PhD projects. Their dedication towards a multidisciplinary approach for studying neurosciences from its most basic level to clinical applications and neurophilosophy will remain a great source of inspiration for the future.

Last but not least, I would like to thank the Onassis Public Benefit Foundation and the German Academic Exchange Service (Deutscher Akademischer Austauschdienst, DAAD) for supporting me financially during my PhD studies.

APPENDIX – SUPPLEMENTARY MATERIAL

Supplementary Material of the included studies, as published in the original papers or as submitted in the latest version of the respective manuscripts.

MANUSCRIPT I: Genetically Determined Levels of Circulating Cytokines and Risk of Stroke: Role of Monocyte Chemoattractant Protein-1

MANUSCRIPT II: Circulating Monocyte Chemoattractant Protein-1 and Risk of Stroke: Meta-Analysis of Population-Based Studies Involving 17 180 Individuals

MANUSCRIPT III: Interleukin-6 signaling effects on ischemic stroke and other cardiovascular outcomes: a Mendelian Randomization study

MANUSCRIPT IV: Use of Genetic Variants Related to Antihypertensive Drugs to Inform on Efficacy and Side Effects

MANUSCRIPT V: Genetically determined blood pressure, antihypertensive drug classes, and risk of stroke subtypes

MANUSCRIPT VI: Genetically determined blood lipids and cerebral small vessel disease: role of HDL cholesterol

SUPPLEMENTAL MATERIAL

Genetically Determined Circulating Levels of Cytokines and Risk of Stroke: Role of Monocyte Chemoattractant Protein-1

Running title: *Georgakis et al.; MCP-1 levels and stroke: Mendelian randomization*

Marios K. Georgakis, MD[1, 2], Dipender Gill, MD[3], Kristiina Rannikmäe, MD[4], Matthew Traylor, PhD[5], Christopher D. Anderson, MD[6, 7, 8], MEGASTROKE consortium of the International Stroke Genetics Consortium (ISGC), Jin-Moo Lee, MD, PhD[9], Yoichiro Kamatani, MD, PhD[10], Jemma C. Hopewell, PhD[11], Bradford B. Worrall, MD[12], Jürgen Bernhagen, PhD[1, 13], Cathie L. M. Sudlow, DPhil[3, 14], Rainer Malik, PhD[1, *], Martin Dichgans, MD[1, 13, 15, *]

[1]Institute for Stroke and Dementia Research (ISD), University Hospital of Ludwig-Maximilians-University (LMU), Munich, Germany; [2]Graduate School for Systemic Neurosciences (GSN), Ludwig-Maximilians-University (LMU), Munich, Germany; [3]Department of Biostatistics and Epidemiology, School of Public Health, Imperial College London, London, UK; [4]Centre for Clinical Brain Sciences, The University of Edinburgh, Edinburgh, UK; [5]Stroke Research Group, Department of Clinical Neurosciences, University of Cambridge, Cambridge, UK; [6]Center for Genomic Medicine, Massachusetts General Hospital (MGH), Boston, MA, USA; [7]Division of Neurocritical Care and Emergency Neurology, Department of Neurology, MGH, Boston, MA, USA; [8]Program in Medical and Population Genetics, Broad Institute, Cambridge, MA, USA; [9]Department of Neurology, Radiology, and Biomedical Engineering, Washington University School of Medicine, St. Louis, MO, USA; [10]Laboratory for Statistical Analysis, RIKEN Center for Integrative Medical Sciences, Yokohama, Japan; [11]Clinical Trial Service Unit and Epidemiological Studies Unit, Nuffield Department of Population Health, University of Oxford, Oxford, UK; [12]Departments of Neurology and Public Health Sciences, University of Virginia School of Medicine, Charlottesville, VA, USA; [13]Munich Cluster for Systems Neurology (SyNergy), Munich, Germany; [14]Institute for Genetics and Molecular Medicine, University of Edinburgh, UK; [15]German Centre for Neurodegenerative Diseases (DZNE), Munich, Germany

***** jointly supervised this work

Supplemental Methods

Analysis of the UK Biobank data

We used the June 2017 release of the imputed genetic data from UK Biobank (downloaded on July 13, 2017). Details on the design of the arrays, sample processing and quality control have been previously described (1). In brief, two closely related arrays from Affymetrix, the UK BiLEVE Axiom array (9.9% of individuals) and the UK Biobank Axiom array were used to genotype approximately 805,426 markers with good genome-wide coverage. Phasing was performed using SHAPEIT3 and imputation to a merged HRC reference panel (39,131,578 autosomal SNPs) and UK10K & 1000 Genomes Phase 3 panel was carried out using the IMPUTE4 package. Imputed genotypes were available for 488,369 individuals (1). From the resulting dataset, we excluded individuals of self-reported ancestry other than White-British, related individuals (pi-hat >0.1875), individuals with high levels heterozygosity and missingness (>5%), and individuals whose reported sex was inconsistent with sex inferred from the genetic data. In addition, only SNPs imputed from the HRC panel were included in this analysis. Stroke in UK Biobank was based on self-reported medical history, and linkage to hospitalization and mortality data. We used the stroke variables provided by UK Biobank that have been created using algorithmic definitions. Details of the stroke algorithm have been described previously and are available on the UK Biobank website ("Definitions of stroke and main stroke pathological types for UK Biobank phase 1 outcomes adjudication", Version 1, http://www.ukbiobank.ac.uk). Individuals with stroke based on self-report only were excluded from the analysis.

Search strategy for meta-analysis

Medline was searched via PubMed using the following combination of search terms:

(CCL2 OR MCP1 OR CCL-2 OR MCP-1 OR "monocyte chemoattractant protein 1" OR "small inducible cytokine A2" OR "chemokine (C-C motif) ligand 2" OR "C-C motif ligand 2") AND

(stroke OR cerebrovascular OR (coronary AND artery AND disease) OR (ischemic AND heart AND disease) OR (myocardial AND infarction))

1. Bycroft C, Freeman C, Petkova D, et al. Genome-wide genetic data on ~500,000 UK Biobank participants. BioRxiv [serial online] 2017.

Supplemental Table 1. Descriptive characteristics of the genome-wide association studies (GWAS) that were included in the Mendelian randomization study.

GWAS	Phenotype	Sample size	Ancestry	Adjustments [a]	Use in this MR study	URL for data download
FINRISK/ CRYFS	Circulating levels of 41 cytokines and growth factors	8,293 individuals	Finnish	age, sex, BMI	Exposure variable in all analyses (selection of instruments)	http://computationalmedicine.fi/data#Cytokine_GWAS
MEGASTROKE	Any stroke, any ischemic stroke and subtypes (LAS, CES, SVS)	67,162 cases/ 454,450 controls	Multi-ancestry	age, sex	Primary outcome in discovery analysis	http://megastroke.org/download.html
Woo et al, 2014	ICH	1,545 cases/ 1,481 controls	European	age, sex	Primary outcome in discovery analysis	http://cerebrovascularportal.org/informational/downloads
UK Biobank	Any stroke, any ischemic stroke	4,985 cases/ 364,434 controls	European	age, sex, genotyping platform array	Primary outcome in validation analysis	http://www.ukbiobank.ac.uk/ (after official data request)
CARDIoGRAMplus C4D	CAD/MI	60,801 cases/ 123,504 controls	European	age, sex	Etiologically related vascular outcome	http://www.cardiogramplusc4d.org/data-downloads/
AFGen	AF	17,931 cases/ 115,142 controls	Multi-ancestry	age, sex	Etiologically related vascular outcome	http://www.broadcvdi.org/informational/data (search for variants of interest)
GLGC	LDL-C, HDL-C, TGL	188,578 individuals	Multi-ancestry	age, sex	Confounder in multivariable MR	http://lipidgenetics.org/
DIAGRAM	T2D	34,840 cases/ 114,981 controls	European/ Pakistani	age, sex	Confounder in multivariable MR	http://diagram-consortium.org/downloads.html
UK Biobank (Neale lab analysis)	SBP, DBP, hypertension	317,754 individuals	European	sex	Confounder in multivariable MR	http://www.nealelab.is/data/

[a] All GWAS studies have further adjusted for principal components.

GWAS names: AGFen, Atrial Fibrillation Genetics; CARDIoGRAMplusC4D, Coronary ARtery DIsease Genome-wide Replication and Meta-analysis (CARDIoGRAM) plus The Coronary Artery Disease (C4D) Genetics; CRYFS, Cardiovascular Risk in Young Finns Study; GLGC, global lipids genetics consortium; DIAGRAM, DIAbetes Genetics Replication And Meta-analysis.

Phenotypes: AF, atrial fibrillation; CAD, coronary artery disease; CES, cardioembolic stroke; DBP, diastolic blood pressure; HDL-C, high-density lipoprotein cholesterol; ICH, intracerebral hemorrhage; LAS, large artery stroke; LDL-C, low-density lipoprotein cholesterol; MI, myocardial infarction; SBP, systolic blood pressure; SVS, small vessel stroke; T2D, type 2 diabetes; TGL, triglycerides

Supplemental Table 2. Cytokines and growth factors examined by the original GWAS study and number of genetic instruments (SNPs) identified from our approach.

Cytokines/ Growth factors	SNPs (N)
BNGF	1
CTACK	14
Eotaxin	22
FGF-basic	0
G-CSF	0
GRO-a	10
HGF	2
IFN-γ	0
IL-1ra	0
IL-1β	0
IL-2	0
IL-2ra	5
IL-4	0
IL-5	1
IL-6	0
IL-7	0
IL-8	0
IL-9	0
IL-10	3
IL-12p70	7
IL-13	0
IL-16	5
IL-17	1
IL-18	26
IP-10	4
MCP-1	38
MCP-3	0
M-CSF	0
MIF	1
MIG	8
MIP-1a	0
MIP-1b	237
PDGF-bb	25
RANTES	0
SCF	4
SCGF-b	11
SDF-1a	0
TNF-α	0
TNF-β	4
TRAIL	41
VEGF	19

Supplemental Table 3. Characteristics of the genetic instruments selected for the circulating levels of cytokines and growth factors. Variance explained by the selected instruments, power of the instruments assessed by the F-statistic and power calculations for the Mendelian randomization study based on the sample sizes of the multi-ancestry MEGASTROKE dataset.

Cytokines/ Growth factors	SNPs (N)	Variance explained (R^2) [a]	*F*-statistic median [range] [b]	Odds Ratio for α <0.05 and [1-β] >0.8					
				Any stroke	Any ischemic stroke	Large artery stroke	Cardio-embolic stroke	Small vessel stroke	Intra-cerebral hemorrhage
BNGF	1	0.012	37	1.107	1.112	1.318	1.275	1.242	2.477
CTACK	14	0.161	27 [24-142]	1.029	1.031	1.087	1.075	1.066	1.288
Eotaxin	22	0.112	28 [22-94]	1.037	1.035	1.104	1.089	1.079	1.354
GRO-a	10	0.162	28 [22-183]	1.029	1.031	1.086	1.074	1.066	1.287
HGF	2	0.017	49 [41-58]	1.090	1.094	1.268	1.229	1.204	2.155
IL-2ra	5	0.143	41 [24-168]	1.031	1.033	1.093	1.079	1.070	1.308
IL-5	1	0.012	38	1.107	1.112	1.318	1.275	1.242	2.477
IL-10	3	0.012	26 [25-37]	1.107	1.112	1.318	1.275	1.242	2.477
IL-12p70	7	0.025	25 [23-35]	1.074	1.077	1.220	1.190	1.168	1.890
IL-16	5	0.123	30 [25-131]	1.034	1.035	1.099	1.085	1.076	1.336
IL-17	1	0.006	39	1.153	1.160	1.450	1.390	1.346	3.508
IL-18	26	0.352	25 [21-96]	1.020	1.021	1.059	1.051	1.045	1.187
IP-10	4	0.036	27 [26-31]	1.062	1.065	1.185	1.158	1.140	1.703
MCP-1	38	0.147	25 [21-92]	1.031	1.031	1.091	1.078	1.069	1.303
MIF	1	0.012	39	1.107	1.112	1.318	1.275	1.242	2.477
MIG	8	0.077	27 [24-42]	1.042	1.044	1.126	1.108	1.095	1.441
MIP-1b	237	1.000[c]	25 [17-789]	1.012	1.013	1.035	1.030	1.027	1.107
PDGF-bb	25	0.118	29 [23-102]	1.034	1.036	1.101	1.087	1.077	1.344
SCF	4	0.023	31 [27-48]	1.077	1.081	1.230	1.197	1.175	1.940
SCGF-b	11	0.174	35 [24-98]	1.028	1.030	1.083	1.072	1.064	1.276
TNF-b	4	0.174	108 [35-123]	1.028	1.030	1.083	1.072	1.064	1.276
TRAIL	41	0.454	35 [21-370]	1.018	1.018	1.052	1.045	1.040	1.163
VEGF	19	0.094	25 [21-63]	1.038	1.040	1.113	1.098	1.086	1.392

Shown are the Odds Ratios per 1 SD increase in circulating levels of the cytokine/growth factor, for which there is power (β) ≥80% to detect an existed association at a type I error of α <0.05.

[a] $R^2 = (beta\ x\ \sqrt{2\ x\ MAF(1-MAF)})^2$, where MAF is the minimum allele frequency and beta is the effect of the SNP on the respective cytokine levels (Park *et al* 2010, Nat. Genet. 42, 570–575). Total variance was calculated in an additive model assuming no interaction between the individual SNPs.

[b] $F = beta^2 / SE^2$, where beta is the effect estimate of the SNP and SE its standard error on the respective cytokine levels (Li & Martin 2002, Comput Stat Data Anal, 40, 21-26).

[c] Due to many variants in low linkage disequilibrium (r^2<0.1), variance estimates and power calculations for MIP-1b are unreliable.

Supplemental Table 4. SNPs that were used as instruments for the circulating levels of MCP-1.

SNP	Gene	Chr	Position	Effect allele	Other allele	Effect allele frequency	Beta[a]	SE	p-value
rs56212190	HIVEP3	1	42168539	c	t	0.95	-0.181	0.0373	9.85E-07
rs145155829	KDM4A	1	44165646	c	t	0.96	0.2153	0.0463	3.72E-06
rs10888395	CTSK	1	150762171	c	t	0.63	0.0814	0.0163	5.98E-07
rs7519506	MNDA	1	158859138	c	t	0.68	0.0987	0.0191	2.44E-07
rs12727764	IFI16	1	158982477	g	t	0.83	-0.0971	0.0213	5.02E-06
rs2281300	CADM3	1	159156285	c	t	0.77	0.0889	0.017	1.71E-07
rs115936758	LOC100131825	1	159170343	c	t	0.94	0.1775	0.0349	5.74E-07
rs35333710	CADM3	1	159172854	g	a	0.9	-0.1476	0.0268	3.71E-08
rs12047264	APCS	1	159535626	g	a	0.19	0.0929	0.0191	1.87E-06
rs7527322	APCS	1	159579533	g	a	0.79	0.0805	0.0171	1.54E-06
rs12073356	LOC148696	1	208007848	g	a	0.93	0.1426	0.0311	4.17E-06
rs111995966	LIMS1	2	109174969	g	t	0.03	-0.1452	0.031	2.53E-06
rs11926788	SEC22C	3	42623498	g	c	0.04	0.1887	0.0363	2.06E-07
rs56300632	NKTR	3	42685911	g	a	0.4	-0.0772	0.0165	3.04E-06
rs4682860	CCBP2	3	42863804	g	a	0.61	-0.0811	0.0157	2.31E-07
rs116425179	LARS2	3	45598703	g	a	0.07	0.1476	0.0255	5.68E-09
rs75265958	SACM1L	3	45758020	g	t	0.05	-0.1231	0.0271	5.16E-06
rs1386930	LZTFL1	3	45884003	c	t	0.54	-0.0903	0.0157	7.86E-09
rs75826707	CCR9	3	45908859	g	a	0.97	-0.1676	0.0369	5.51E-06
rs3774641	CCR9	3	45937833	g	t	0.81	-0.1324	0.0191	5.04E-12
rs2036297	CCR1	3	46172903	g	a	0.63	-0.119	0.016	1.09E-13
rs41338844	CCR3	3	46272951	g	a	0.98	-0.2195	0.0462	3.22E-06
rs138591554	CCR3	3	46289206	t	a	0.1	0.3171	0.0331	7.94E-22
rs112313229	CCR2	3	46364860	g	a	0.96	0.1646	0.0313	1.43E-07
rs62242985	CCR2	3	46385638	g	a	0.4	0.1013	0.0164	5.54E-10
rs35060576	CCRL2	3	46434525	g	a	0.58	-0.0955	0.0162	3.79E-09
rs11720094	LRRC2	3	46559911	g	c	0.53	0.1058	0.0157	1.54E-11
rs141676607	LOC100132146	3	46653244	c	t	0.98	-0.3128	0.064	9.48E-07
rs142043796	LOC100132146	3	46679831	g	c	0.98	0.2414	0.0487	8.30E-07
rs34190208	ALS2CL	3	46736217	c	t	0.75	-0.1076	0.022	9.91E-07
rs78629618	PRSS42	3	46880130	c	t	0.12	-0.1188	0.0259	4.23E-06
rs11710798	PFKFB4	3	48570686	c	a	0.11	-0.0924	0.0197	2.87E-06
rs2712431	RPN1	3	128316890	c	a	0.32	0.0787	0.0172	4.76E-06
rs7019112	FLJ35282	9	23028336	g	t	0.21	0.2126	0.0471	7.12E-06
rs10744620	EFCAB4B	12	3739094	c	t	0.62	-0.0788	0.0161	9.91E-07
rs10145849	SEL1L	14	82941991	g	a	0.6	0.0755	0.0162	3.41E-06
rs7197349	WWOX	16	78687219	g	a	0.13	-0.0968	0.0206	2.62E-06
rs146522229	C5AR1	19	47798480	c	t	0.98	0.5976	0.1177	3.56E-07
rs191688264	SF3A1	22	30745361	c	a	0.99	0.4834	0.0991	1.10E-06

[a] Beta refers to 1 standard deviation increase in circulating MCP-1 levels.
Chr, chromosome; SE, standard error; SNP, single-nucleotide polymorphism.

Supplemental Table 5. Characteristics of the observational case-control studies that were included in the meta-analysis of circulating MCP-1 levels and risk of stroke.

Study	Place (study period)	Definition of cases	Definition of controls	N cases	N controls	MCP-1 assessment	MCP-1 (pg/dl, mean)	Age (y, mean)	Males (%)	Diabetes mellitus (%)	Hypertension (%)	BMI (kg/m^2, mean)	Dyslipidemia (%)	Adjustment/ Matching factors
Losy et al, 2001	Poznan, Poland	First-ever ischemic stroke	Individuals with tension headache	23	15	ELISA	269	72.2	NR	17	52	NR	NR	Age, gender
Sánchez-Moreno et al, 2004	Boston, USA	Ischemic stroke	Outpatients without neurological disorders	15	24	ELISA	133	64	NR	NR	NR	NR	NR	-
Arakelyan et al, 2005	Yerevan, Armenia	Ischemic stroke	Individuals without history of stroke and myocardial infarction	40	40	ELISA	312	59.8	70	4	25	NR	NR	-
Zaremba et al, 2006	Poznan, Poland	First-ever ischemic stroke	Individuals without stroke	27	20	ELISA	182	65.5	40	15	34	NR	NR	Age, gender
Davi et al, 2009	Palermo, Italy (2002-2006)	Ischemic stroke and diabetes	Healthy controls	90	45	ELISA	185	72	57	67	69	29.6	31	Age, gender
Kuriyama et al, 2009	Kyoto, Japan (2004-2006)	First-time or recurrent ischemic stroke	Individuals with normal MRI	100	90	Multiplex assay	139	66.1	52	29	50	NR	38	Age
Chen et al, 2012	Hannover, Germany (2007-2009)	Ischemic stroke	Individuals without cerebrovascular or cardiovascular disease	58	32	ELISA	226	71.6	51	11	63	25.7	46	-
Khurana et al, 2013	Chandigarh, India	Ischemic stroke with carotid plaque	Individuals without stroke or carotid plaque	57	15	ELISA	4.25	59.3	50	29	73	NR	5	-
Gao et al, 2014	Meta-analysis of 8 Asian studies	Ischemic stroke	Health controls	716	425	ELISA	NR	62.6	59	NR	NR	NR	NR	-
Grosse et al, 2016	Hannover, Germany (2011-2012)	Cardioembolic stroke	Individuals without cardiovascular disease	11	11	ELISA	165	69.5	82	NR	NR	26.3	18	Age, gender

ELISA, Enzyme-linked Immunosorbent Assay; NR, not reported.

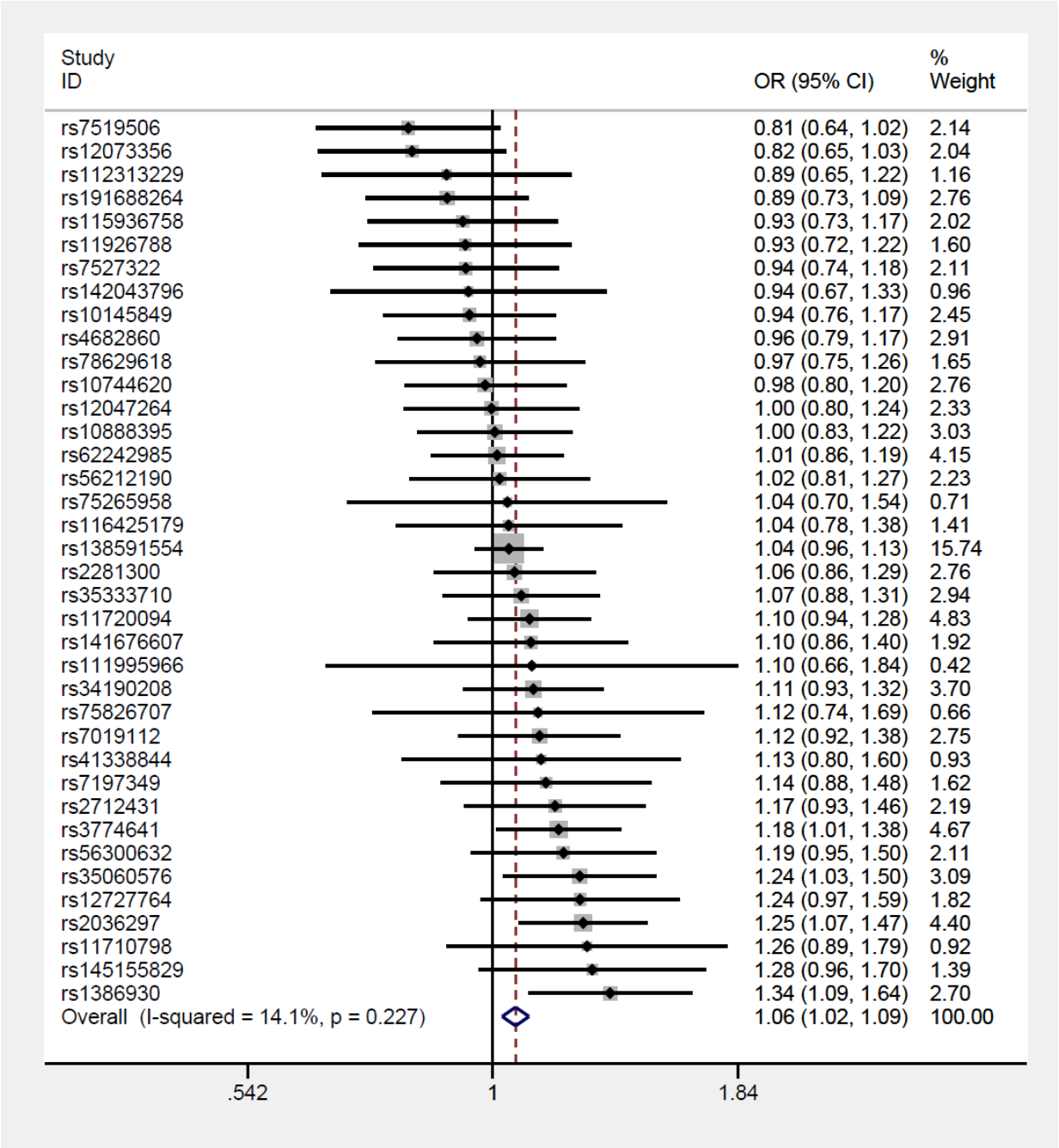
Supplemental Table 6. Characteristics of the observational cohort studies that were included in the meta-analysis of circulating MCP-1 levels and risk of stroke.

Study	Place (study period)	Population	Outcome	Follow-up (y, mean)	N population	N events	MCP-1 assessment	MCP-1 (pg/dl, mean)	Age (y, mean)	Males (%)	Diabetes mellitus (%)	Hypertension (%)	BMI (kg/m^2, mean)	Dyslipidemia (%)	Adjustment/ Matching factors
Haim et al, 2005 (BIP)	Israel	Patients with myocardial infarction in the last 5 years (part of a trial for bezafibrate)	Ischemic stroke	6.2	466	123	ELISA	319	61.1	94	11.5	34.5	26.7	NR	age, sex, smoking, circulating lipoprotein concentrations at baseline, diabetes mellitus, hypertension, and history of myocardial infarction, fibrinogen, soluble ICAM-1
Canoui-Poitrine et al, 2011 (PRIME)	Belfast, Northern Ireland & Lille, Strasbourg, Toulouse (France)	General male population	Ischemic stroke	10	285	95	Multiplex bioassay	82	55.5	100	4.9	47.7	26.7	NR	age, sex, hypertension, smoking, diabetes, body mass index, high-density lipoprotein and total cholesterol, triglycerides, high sensitivity C-reactive protein, and fibrinogen
Ganz et al, 2017 (SPARCL)	Multicenter trial	Individuals with an ischemic stroke in the previous 6-12 months (part of a trial for atorvastatin)	Ischemic and hemorrhagic stroke	5	2176	482	ELISA	116	62.9	61.4	17.2	62.5	27.4	NR	age, sex, race, treatment group, entry event (stroke or transient ischemic attack), time since entry event, geographic region, smoking, diabetes, systolic blood pressure, hypertension treatment, high-density lipoprotein cholesterol, and apolipoprotein A1

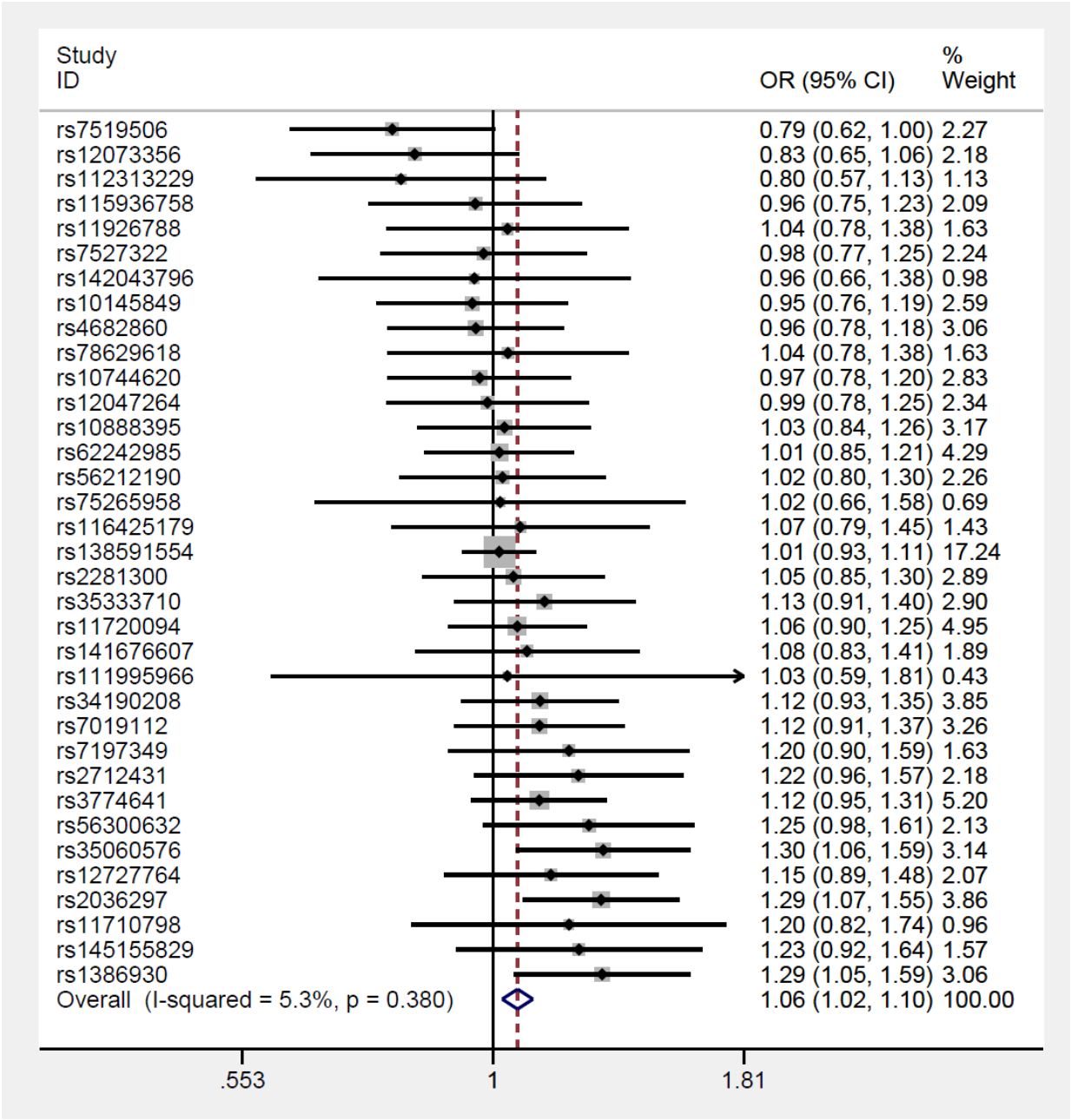
BIP, Bezafibrate Infarction Prevention; ELISA, Enzyme-linked Immunosorbent Assay; NR, not reported; PRIME, étude Prospective sur l'Infarctus du Myocarde; SPARCL, Stroke Prevention by Aggressive Reduction of Cholesterol Levels.

Supplemental Figure 1. Forest plots of SNP-specific Mendelian randomization associations between circulating MCP-1 levels with the odds of (A) any stroke, (B) any ischemic stroke, (C) large artery stroke, (D) cardioembolic stroke, (E) small-vessel stroke, and (F) intracerebral hemorrhage. The results are expressed as Odds Ratios (OR) with their 95% Confidence Intervals (95%CI) for the effect of 1 SD increase in circulating MCP-1 levels and are based on the MEGASTROKE data.

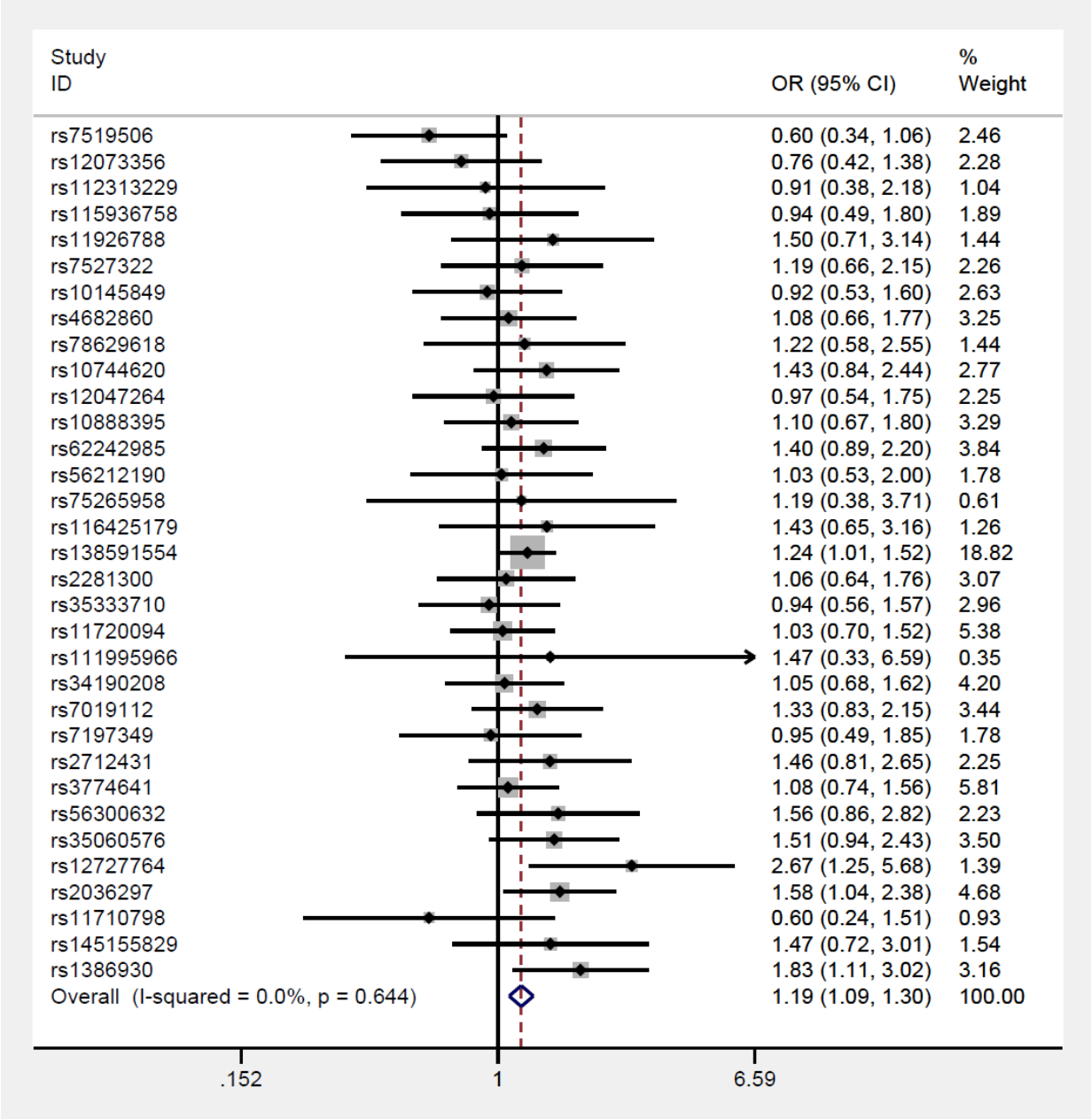
A



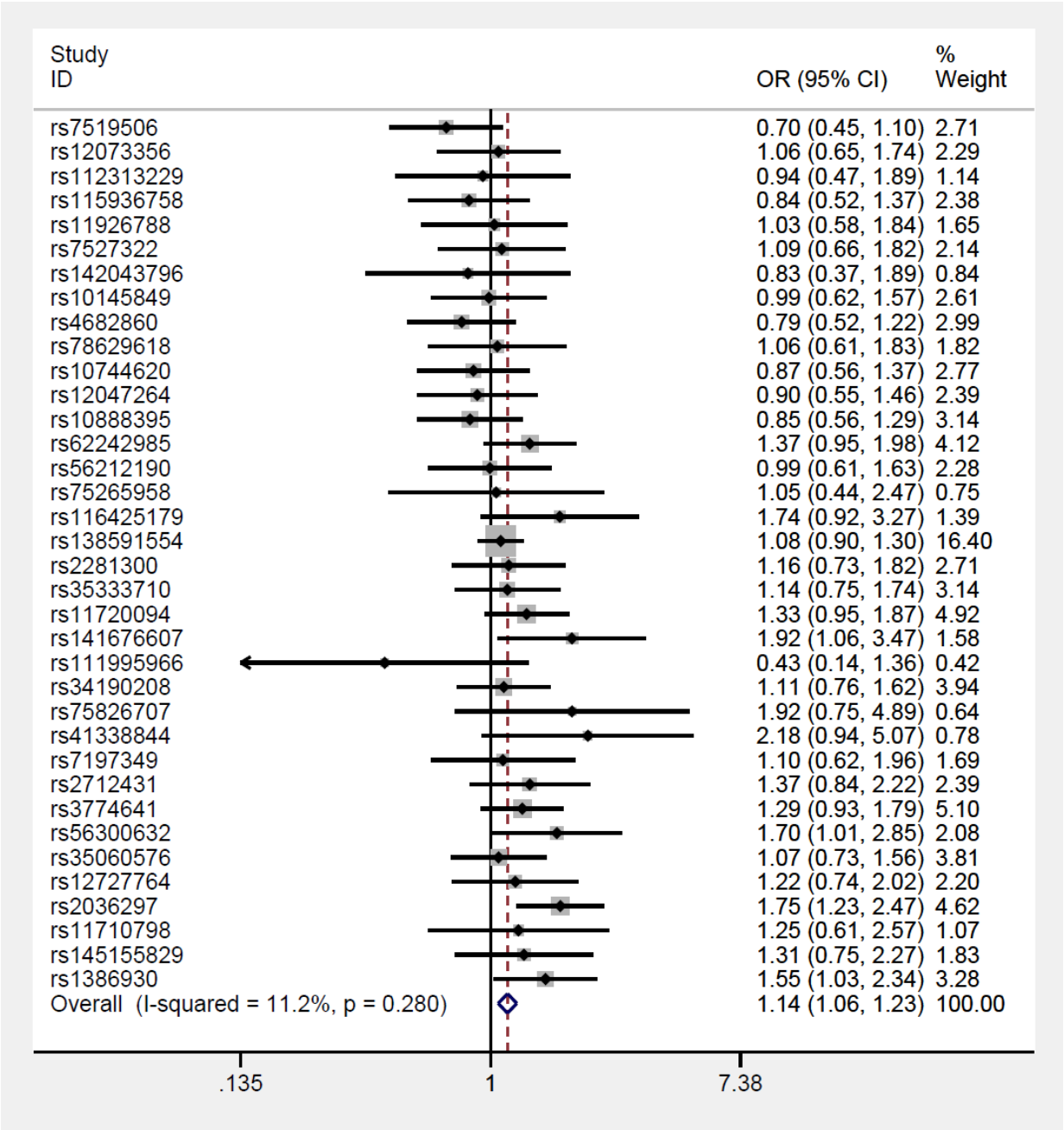
B


Study ID
OR (95% CI)
% Weight
rs7519506 0.79 (0.62, 1.00) 2.27
rs12073356 0.83 (0.65, 1.06) 2.18
rs112313229 0.80 (0.57, 1.13) 1.13
rs115936758 0.96 (0.75, 1.23) 2.09
rs11926788 1.04 (0.78, 1.38) 1.63
rs7527322 0.98 (0.77, 1.25) 2.24
rs142043796 0.96 (0.66, 1.38) 0.98
rs10145849 0.95 (0.76, 1.19) 2.59
rs4682860 0.96 (0.78, 1.18) 3.06
rs78629618 1.04 (0.78, 1.38) 1.63
rs10744620 0.97 (0.78, 1.20) 2.83
rs12047264 0.99 (0.78, 1.25) 2.34
rs10888395 1.03 (0.84, 1.26) 3.17
rs62242985 1.01 (0.85, 1.21) 4.29
rs56212190 1.02 (0.80, 1.30) 2.26
rs75265958 1.02 (0.66, 1.58) 0.69
rs116425179 1.07 (0.79, 1.45) 1.43
rs138591554 1.01 (0.93, 1.11) 17.24
rs2281300 1.05 (0.85, 1.30) 2.89
rs35333710 1.13 (0.91, 1.40) 2.90
rs11720094 1.06 (0.90, 1.25) 4.95
rs141676607 1.08 (0.83, 1.41) 1.89
rs111995966 1.03 (0.59, 1.81) 0.43
rs34190208 1.12 (0.93, 1.35) 3.85
rs7019112 1.12 (0.91, 1.37) 3.26
rs7197349 1.20 (0.90, 1.59) 1.63
rs2712431 1.22 (0.96, 1.57) 2.18
rs3774641 1.12 (0.95, 1.31) 5.20
rs56300632 1.25 (0.98, 1.61) 2.13
rs35060576 1.30 (1.06, 1.59) 3.14
rs12727764 1.15 (0.89, 1.48) 2.07
rs2036297 1.29 (1.07, 1.55) 3.86
rs11710798 1.20 (0.82, 1.74) 0.96
rs145155829 1.23 (0.92, 1.64) 1.57
rs1386930 1.29 (1.05, 1.59) 3.06
Overall (I-squared = 5.3%, p = 0.380) 1.06 (1.02, 1.10) 100.00
.553
1
1.81

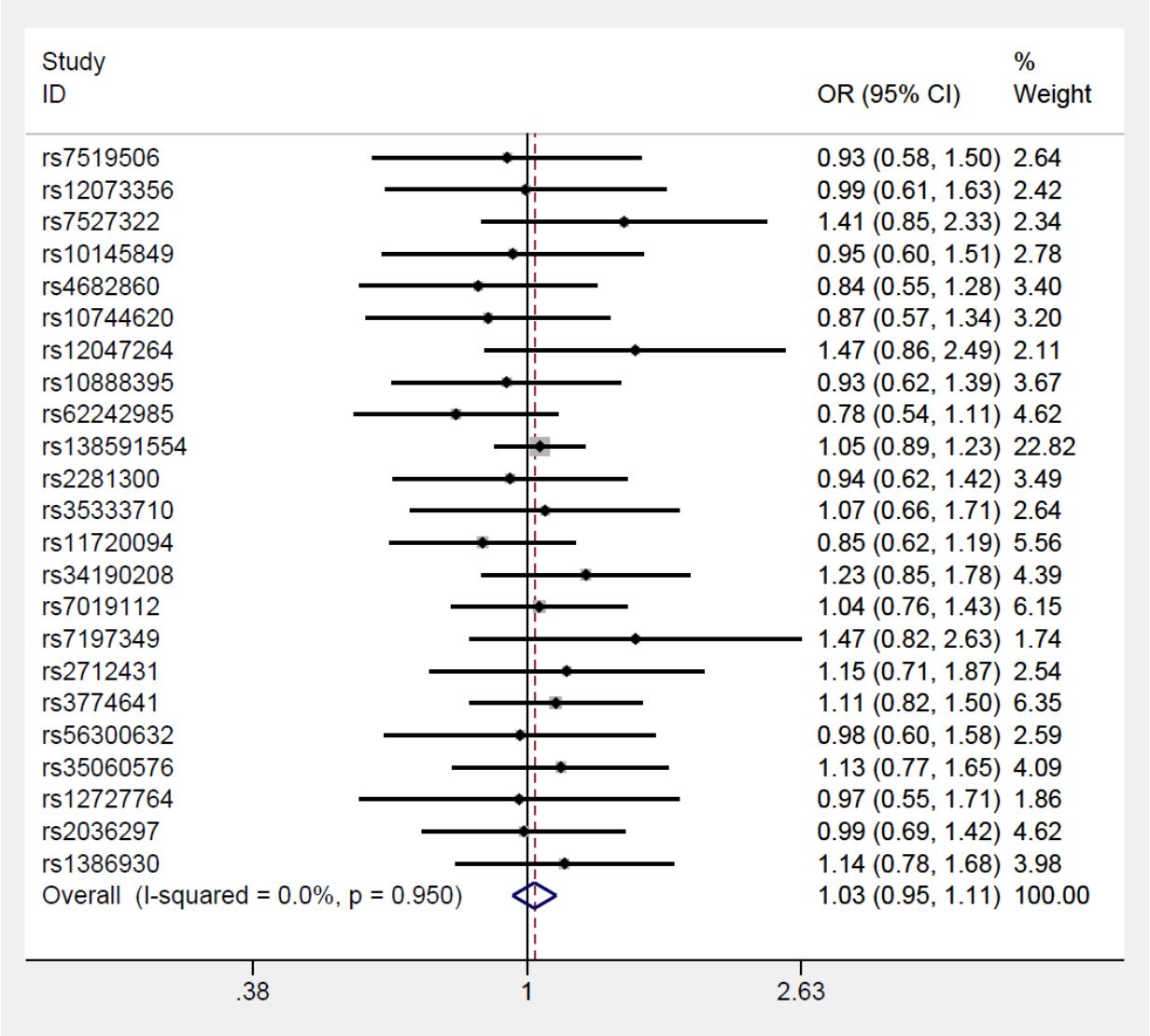
C


Study ID
OR (95% CI)
% Weight
rs7519506 0.60 (0.34, 1.06) 2.46
rs12073356 0.76 (0.42, 1.38) 2.28
rs112313229 0.91 (0.38, 2.18) 1.04
rs115936758 0.94 (0.49, 1.80) 1.89
rs11926788 1.50 (0.71, 3.14) 1.44
rs7527322 1.19 (0.66, 2.15) 2.26
rs10145849 0.92 (0.53, 1.60) 2.63
rs4682860 1.08 (0.66, 1.77) 3.25
rs78629618 1.22 (0.58, 2.55) 1.44
rs10744620 1.43 (0.84, 2.44) 2.77
rs12047264 0.97 (0.54, 1.75) 2.25
rs10888395 1.10 (0.67, 1.80) 3.29
rs62242985 1.40 (0.89, 2.20) 3.84
rs56212190 1.03 (0.53, 2.00) 1.78
rs75265958 1.19 (0.38, 3.71) 0.61
rs116425179 1.43 (0.65, 3.16) 1.26
rs138591554 1.24 (1.01, 1.52) 18.82
rs2281300 1.06 (0.64, 1.76) 3.07
rs35333710 0.94 (0.56, 1.57) 2.96
rs11720094 1.03 (0.70, 1.52) 5.38
rs111995966 1.47 (0.33, 6.59) 0.35
rs34190208 1.05 (0.68, 1.62) 4.20
rs7019112 1.33 (0.83, 2.15) 3.44
rs7197349 0.95 (0.49, 1.85) 1.78
rs2712431 1.46 (0.81, 2.65) 2.25
rs3774641 1.08 (0.74, 1.56) 5.81
rs56300632 1.56 (0.86, 2.82) 2.23
rs35060576 1.51 (0.94, 2.43) 3.50
rs12727764 2.67 (1.25, 5.68) 1.39
rs2036297 1.58 (1.04, 2.38) 4.68
rs11710798 0.60 (0.24, 1.51) 0.93
rs145155829 1.47 (0.72, 3.01) 1.54
rs1386930 1.83 (1.11, 3.02) 3.16
Overall (I-squared = 0.0%, p = 0.644) 1.19 (1.09, 1.30) 100.00
.152
1
6.59

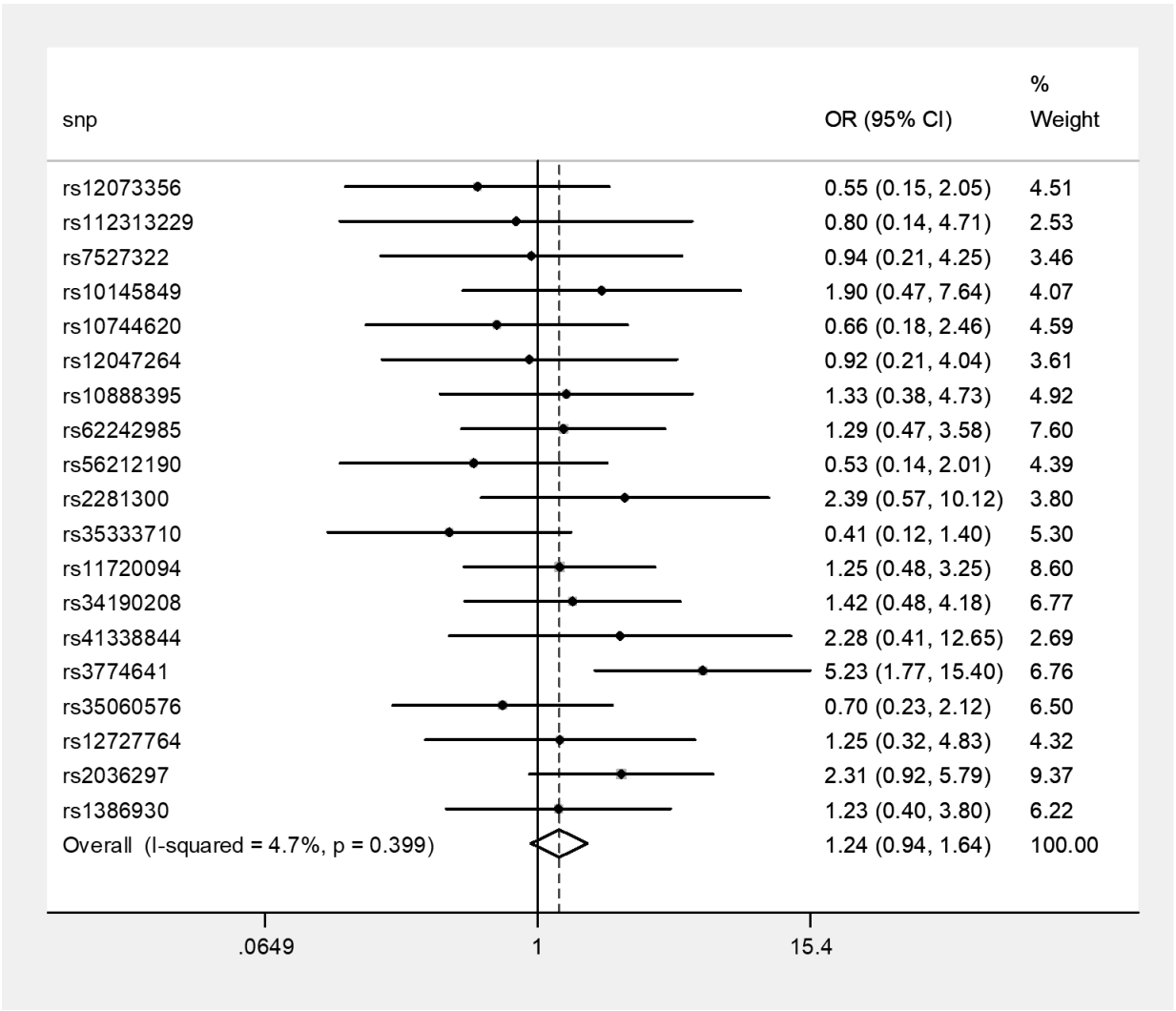
D


Study ID
OR (95% CI)
% Weight
rs7519506 0.70 (0.45, 1.10) 2.71
rs12073356 1.06 (0.65, 1.74) 2.29
rs112313229 0.94 (0.47, 1.89) 1.14
rs115936758 0.84 (0.52, 1.37) 2.38
rs11926788 1.03 (0.58, 1.84) 1.65
rs7527322 1.09 (0.66, 1.82) 2.14
rs142043796 0.83 (0.37, 1.89) 0.84
rs10145849 0.99 (0.62, 1.57) 2.61
rs4682860 0.79 (0.52, 1.22) 2.99
rs78629618 1.06 (0.61, 1.83) 1.82
rs10744620 0.87 (0.56, 1.37) 2.77
rs12047264 0.90 (0.55, 1.46) 2.39
rs10888395 0.85 (0.56, 1.29) 3.14
rs62242985 1.37 (0.95, 1.98) 4.12
rs56212190 0.99 (0.61, 1.63) 2.28
rs75265958 1.05 (0.44, 2.47) 0.75
rs116425179 1.74 (0.92, 3.27) 1.39
rs138591554 1.08 (0.90, 1.30) 16.40
rs2281300 1.16 (0.73, 1.82) 2.71
rs35333710 1.14 (0.75, 1.74) 3.14
rs11720094 1.33 (0.95, 1.87) 4.92
rs141676607 1.92 (1.06, 3.47) 1.58
rs111995966 0.43 (0.14, 1.36) 0.42
rs34190208 1.11 (0.76, 1.62) 3.94
rs75826707 1.92 (0.75, 4.89) 0.64
rs41338844 2.18 (0.94, 5.07) 0.78
rs7197349 1.10 (0.62, 1.96) 1.69
rs2712431 1.37 (0.84, 2.22) 2.39
rs3774641 1.29 (0.93, 1.79) 5.10
rs56300632 1.70 (1.01, 2.85) 2.08
rs35060576 1.07 (0.73, 1.56) 3.81
rs12727764 1.22 (0.74, 2.02) 2.20
rs2036297 1.75 (1.23, 2.47) 4.62
rs11710798 1.25 (0.61, 2.57) 1.07
rs145155829 1.31 (0.75, 2.27) 1.83
rs1386930 1.55 (1.03, 2.34) 3.28
Overall (I-squared = 11.2%, p = 0.280) 1.14 (1.06, 1.23) 100.00
.135
1
7.38

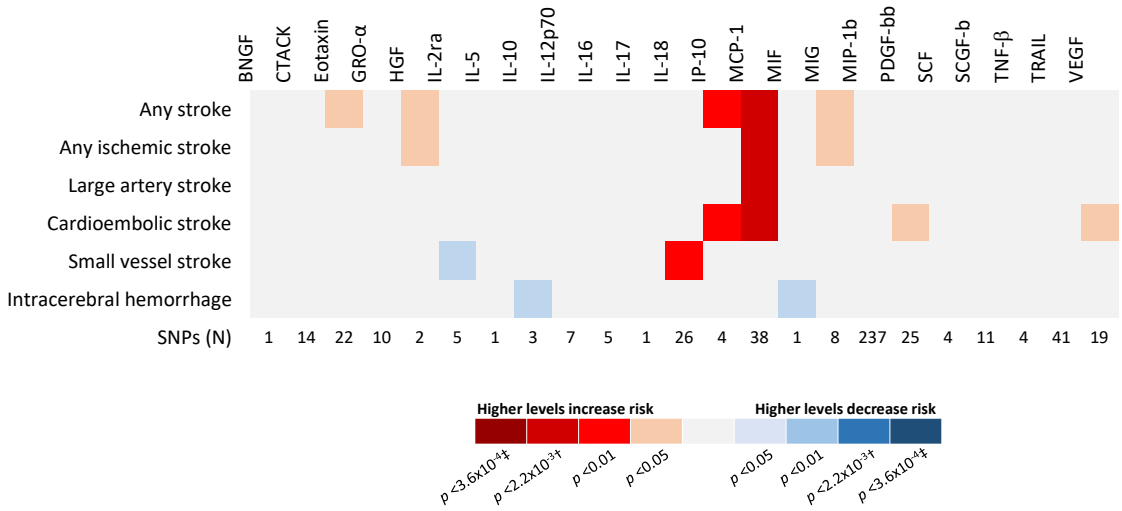
E


Study ID
OR (95% CI)
% Weight
rs7519506 0.93 (0.58, 1.50) 2.64
rs12073356 0.99 (0.61, 1.63) 2.42
rs7527322 1.41 (0.85, 2.33) 2.34
rs10145849 0.95 (0.60, 1.51) 2.78
rs4682860 0.84 (0.55, 1.28) 3.40
rs10744620 0.87 (0.57, 1.34) 3.20
rs12047264 1.47 (0.86, 2.49) 2.11
rs10888395 0.93 (0.62, 1.39) 3.67
rs62242985 0.78 (0.54, 1.11) 4.62
rs138591554 1.05 (0.89, 1.23) 22.82
rs2281300 0.94 (0.62, 1.42) 3.49
rs35333710 1.07 (0.66, 1.71) 2.64
rs11720094 0.85 (0.62, 1.19) 5.56
rs34190208 1.23 (0.85, 1.78) 4.39
rs7019112 1.04 (0.76, 1.43) 6.15
rs7197349 1.47 (0.82, 2.63) 1.74
rs2712431 1.15 (0.71, 1.87) 2.54
rs3774641 1.11 (0.82, 1.50) 6.35
rs56300632 0.98 (0.60, 1.58) 2.59
rs35060576 1.13 (0.77, 1.65) 4.09
rs12727764 0.97 (0.55, 1.71) 1.86
rs2036297 0.99 (0.69, 1.42) 4.62
rs1386930 1.14 (0.78, 1.68) 3.98
Overall (I-squared = 0.0%, p = 0.950) 1.03 (0.95, 1.11) 100.00
.38
1
2.63

F

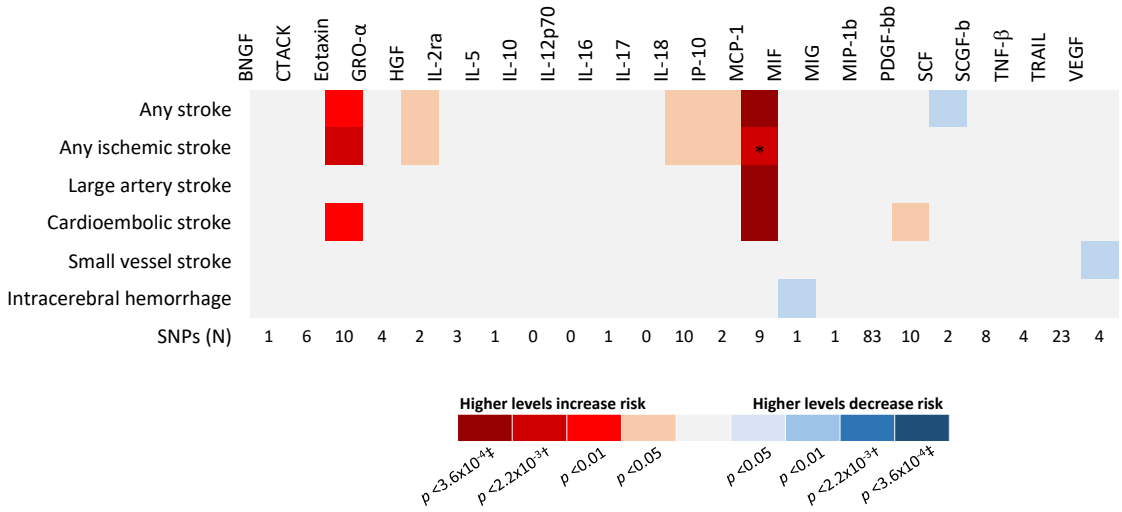

snp
OR (95% CI)
% Weight
rs12073356 0.55 (0.15, 2.05) 4.51
rs112313229 0.80 (0.14, 4.71) 2.53
rs7527322 0.94 (0.21, 4.25) 3.46
rs10145849 1.90 (0.47, 7.64) 4.07
rs10744620 0.66 (0.18, 2.46) 4.59
rs12047264 0.92 (0.21, 4.04) 3.61
rs10888395 1.33 (0.38, 4.73) 4.92
rs62242985 1.29 (0.47, 3.58) 7.60
rs56212190 0.53 (0.14, 2.01) 4.39
rs2281300 2.39 (0.57, 10.12) 3.80
rs35333710 0.41 (0.12, 1.40) 5.30
rs11720094 1.25 (0.48, 3.25) 8.60
rs34190208 1.42 (0.48, 4.18) 6.77
rs41338844 2.28 (0.41, 12.65) 2.69
rs3774641 5.23 (1.77, 15.40) 6.76
rs35060576 0.70 (0.23, 2.12) 6.50
rs12727764 1.25 (0.32, 4.83) 4.32
rs2036297 2.31 (0.92, 5.79) 9.37
rs1386930 1.23 (0.40, 3.80) 6.22
Overall (I-squared = 4.7%, p = 0.399) 1.24 (0.94, 1.64) 100.00
.0649
1
15.4

Supplemental Figure 2. Mendelian randomization associations of the circulating levels of cytokines and growth factors in with any stroke and stroke subtypes restricted to individuals of European ancestry in the MEGASTROKE data. The results are derived from the fixed-effects inverse-variance weighted (IVW) method.



* Significant heterogeneity (I^2>25% or Cochran *Q*-derived *p* <0.05)
† Bonferroni-corrected threshold for number of cytokines
‡ Bonferroni-corrected threshold for number of cytokines and number of phenotypes

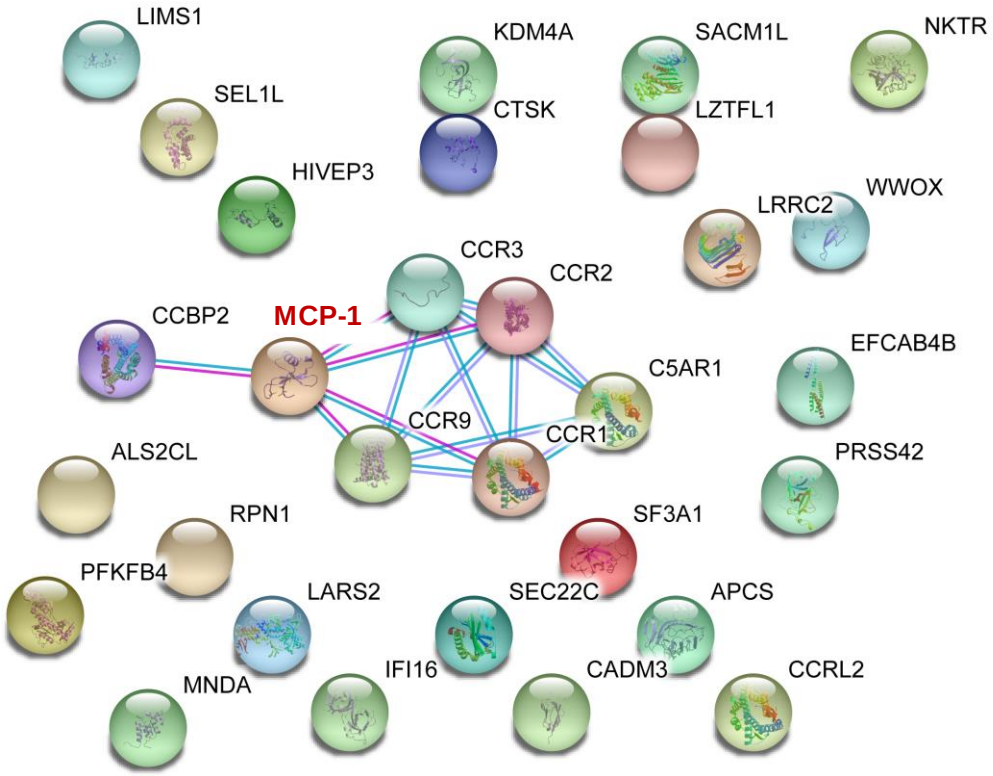
Supplemental Figure 3. Mendelian randomization associations of the circulating levels of cytokines and growth factors with any stroke and stroke subtypes, using a GWAS threshold of $p < 5 \times 10^{-8}$ for the selection of genetic instruments in the MEGASTROKE data. The results are derived from the fixed-effects inverse-variance weighted (IVW) method.



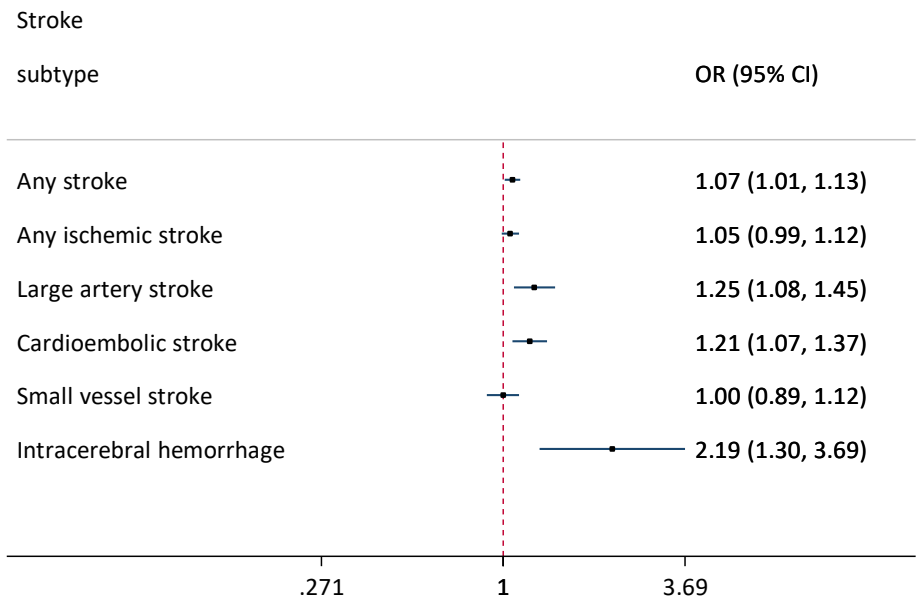
* Significant heterogeneity (I^2>25% or Cochran *Q*-derived p <0.05)
† Bonferroni-corrected threshold for number of cytokines
‡ Bonferroni-corrected threshold for number of cytokines and number of phenotypes

Supplemental Figure 4. (A) Protein-protein interactions between MCP-1 and proteins encoded by genes in the vicinity of the genetic instruments for MCP-1. (B) Results from the Mendelian randomization analysis restricted to the respective genetic instruments at CCR1, CCR2, CCR3, CCR9, C5AR1, and CCBP2.

A



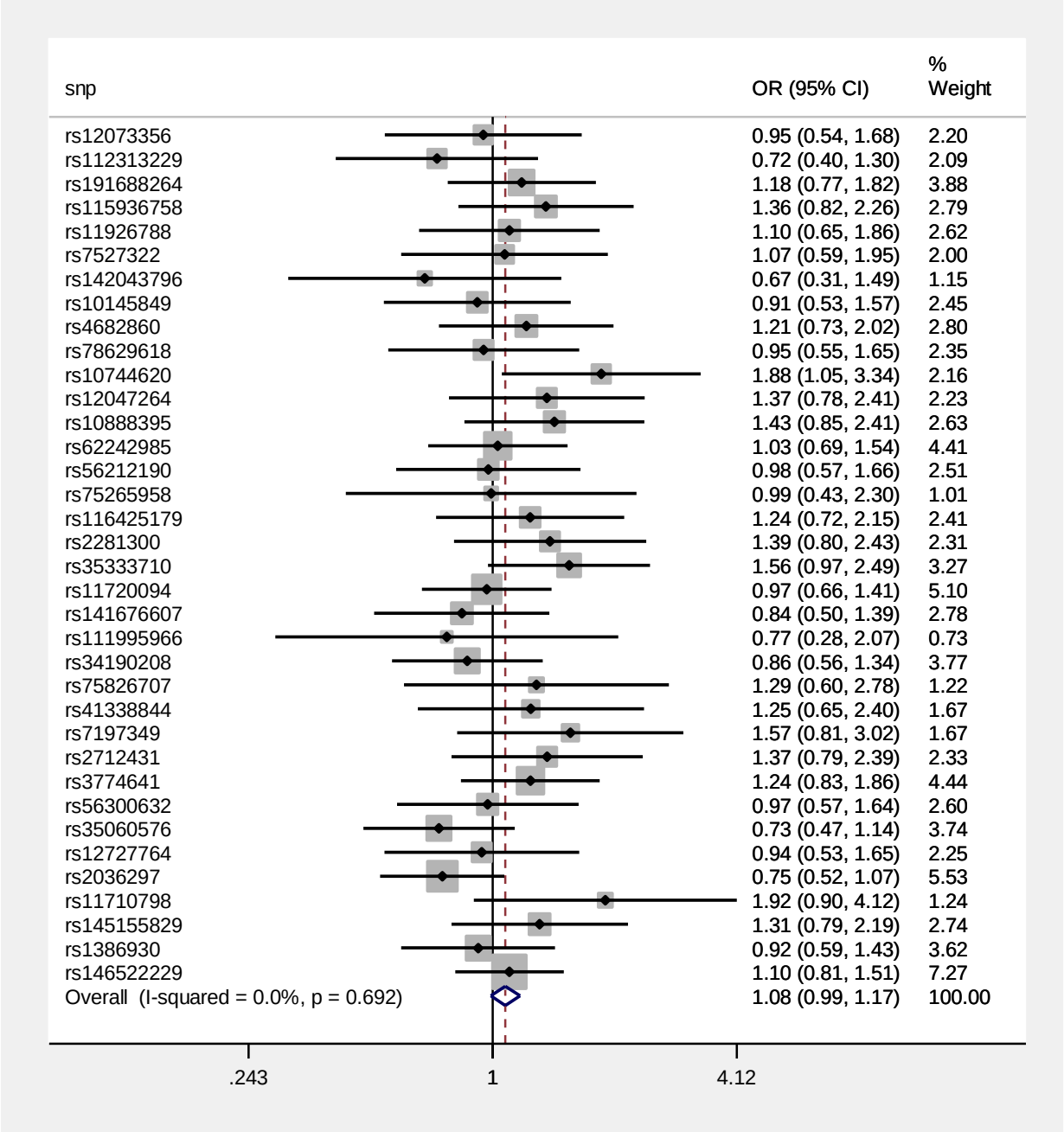
B



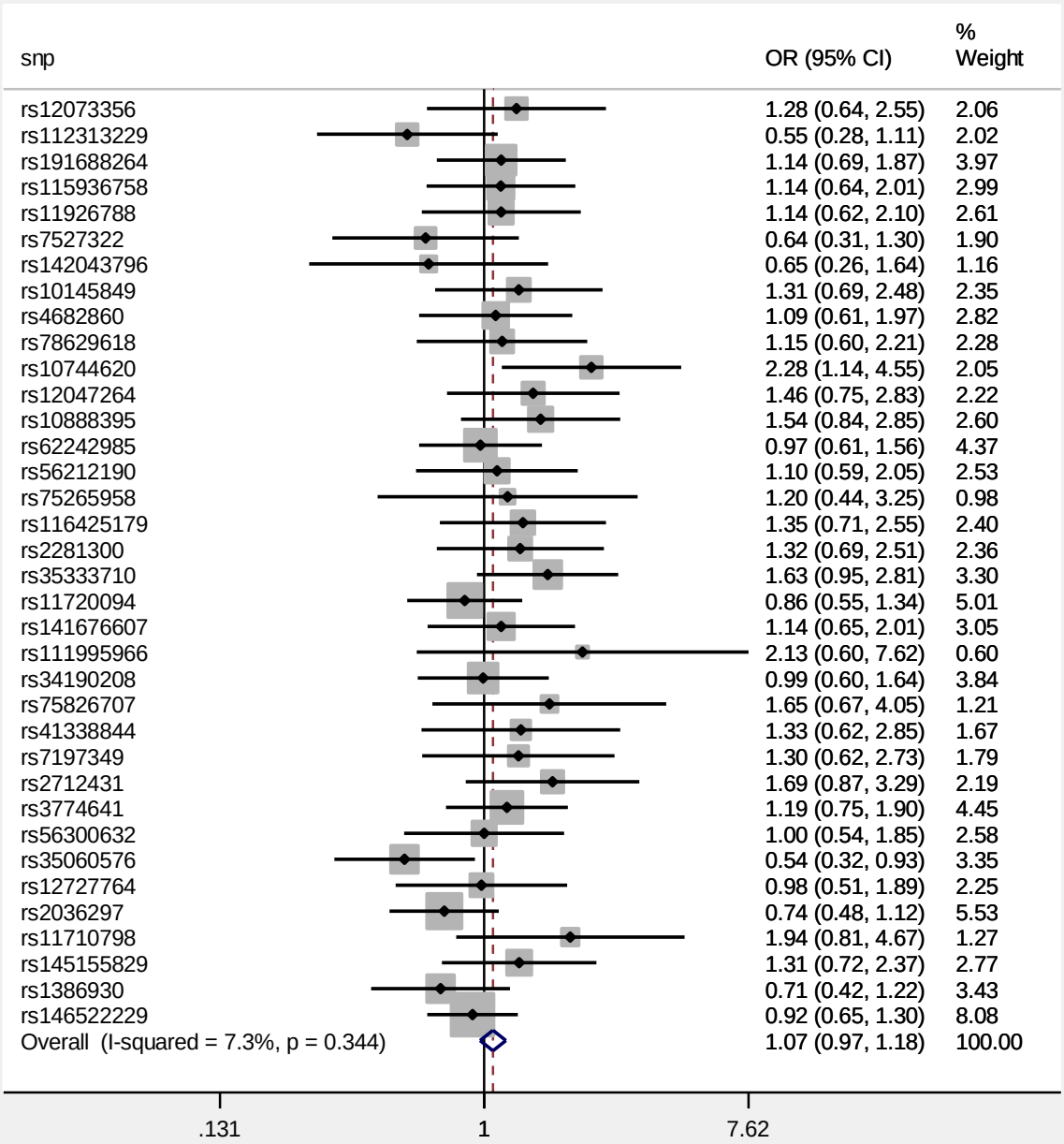
The protein-protein interaction network has been produced by the STRING database. The Mendelian randomization has been conducted within the MEGASTROKE database and has been based on the fixed-effects IVW meta-analysis method.

Supplemental Figure 5. Forest plots of SNP-specific Mendelian randomization associations between circulating MCP-1 levels with the odds of (A) any stroke, and (B) any ischemic stroke in the UK Biobank. The results are expressed as Odds Ratios (OR) with their 95% Confidence Intervals (95%CI) for the effect of 1 SD increase in circulating MCP-1 levels.

A

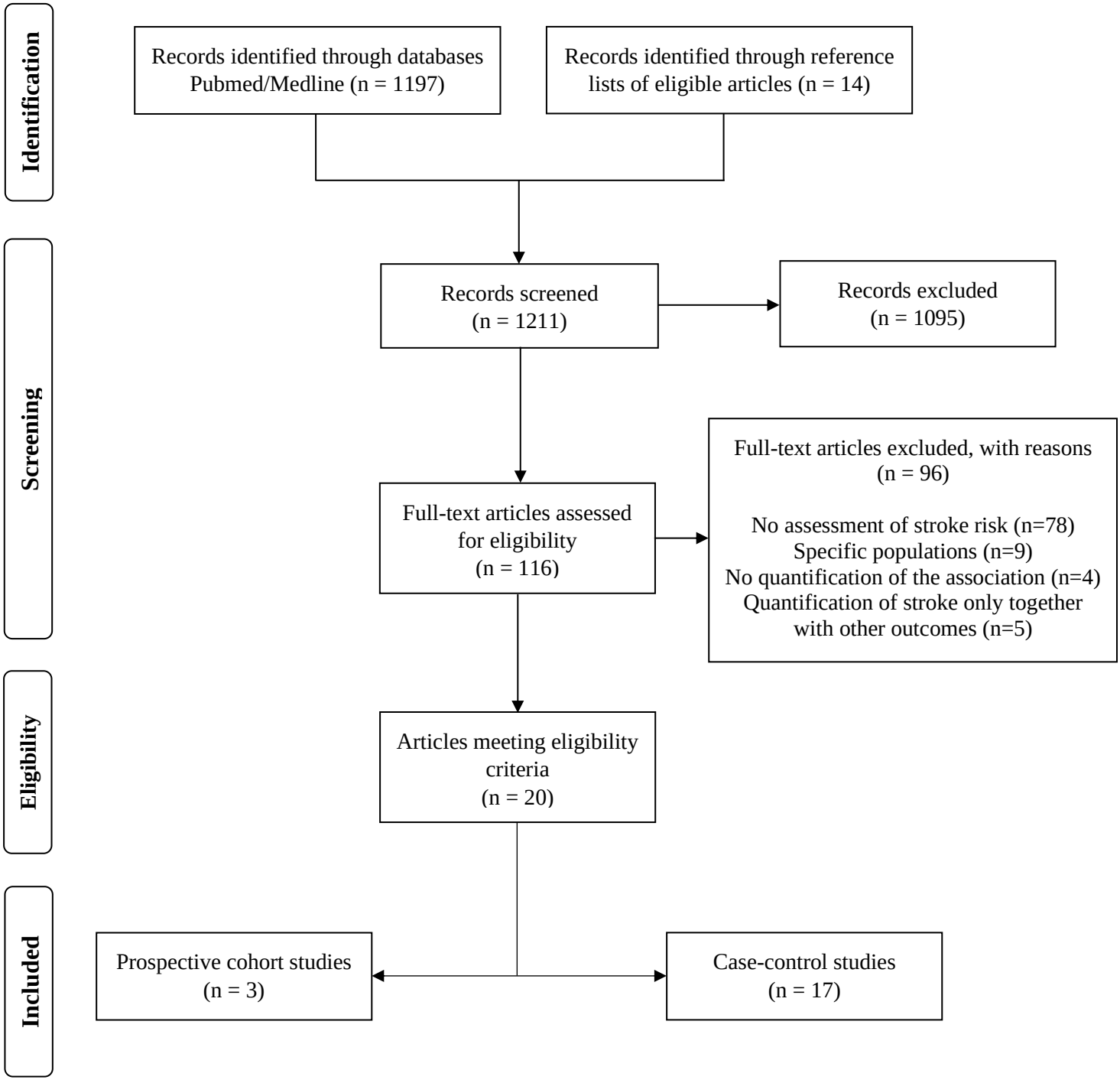


B


snp
OR (95% CI)
% Weight
rs12073356 1.28 (0.64, 2.55) 2.06
rs112313229 0.55 (0.28, 1.11) 2.02
rs191688264 1.14 (0.69, 1.87) 3.97
rs115936758 1.14 (0.64, 2.01) 2.99
rs11926788 1.14 (0.62, 2.10) 2.61
rs7527322 0.64 (0.31, 1.30) 1.90
rs142043796 0.65 (0.26, 1.64) 1.16
rs10145849 1.31 (0.69, 2.48) 2.35
rs4682860 1.09 (0.61, 1.97) 2.82
rs78629618 1.15 (0.60, 2.21) 2.28
rs10744620 2.28 (1.14, 4.55) 2.05
rs12047264 1.46 (0.75, 2.83) 2.22
rs10888395 1.54 (0.84, 2.85) 2.60
rs62242985 0.97 (0.61, 1.56) 4.37
rs56212190 1.10 (0.59, 2.05) 2.53
rs75265958 1.20 (0.44, 3.25) 0.98
rs116425179 1.35 (0.71, 2.55) 2.40
rs2281300 1.32 (0.69, 2.51) 2.36
rs35333710 1.63 (0.95, 2.81) 3.30
rs11720094 0.86 (0.55, 1.34) 5.01
rs141676607 1.14 (0.65, 2.01) 3.05
rs111995966 2.13 (0.60, 7.62) 0.60
rs34190208 0.99 (0.60, 1.64) 3.84
rs75826707 1.65 (0.67, 4.05) 1.21
rs41338844 1.33 (0.62, 2.85) 1.67
rs7197349 1.30 (0.62, 2.73) 1.79
rs2712431 1.69 (0.87, 3.29) 2.19
rs3774641 1.19 (0.75, 1.90) 4.45
rs56300632 1.00 (0.54, 1.85) 2.58
rs35060576 0.54 (0.32, 0.93) 3.35
rs12727764 0.98 (0.51, 1.89) 2.25
rs2036297 0.74 (0.48, 1.12) 5.53
rs11710798 1.94 (0.81, 4.67) 1.27
rs145155829 1.31 (0.72, 2.37) 2.77
rs1386930 0.71 (0.42, 1.22) 3.43
rs146522229 0.92 (0.65, 1.30) 8.08
Overall (I-squared = 7.3%, p = 0.344) 1.07 (0.97, 1.18) 100.00
.131
1
7.62

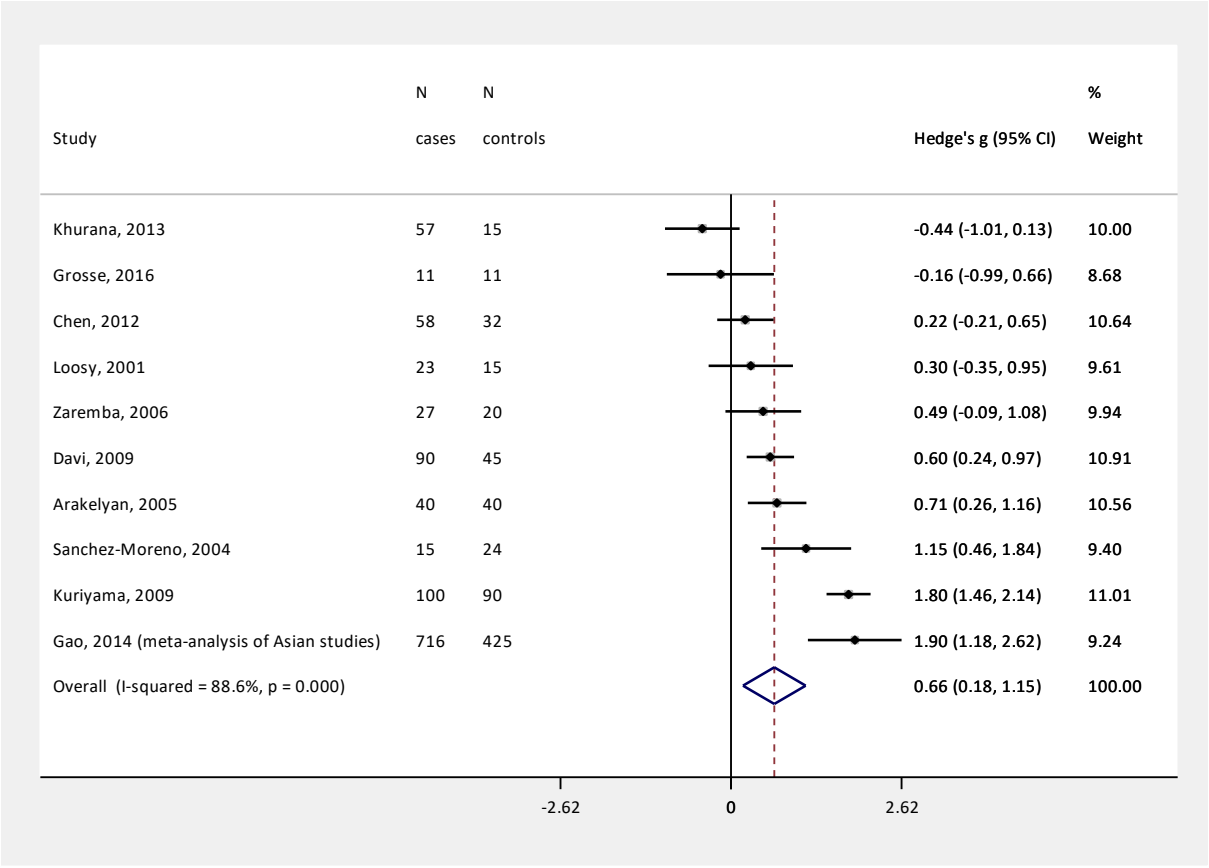
Supplemental Figure 6. Flowchart of the study selection process in the meta-analysis of MCP-1 levels with stroke.

A

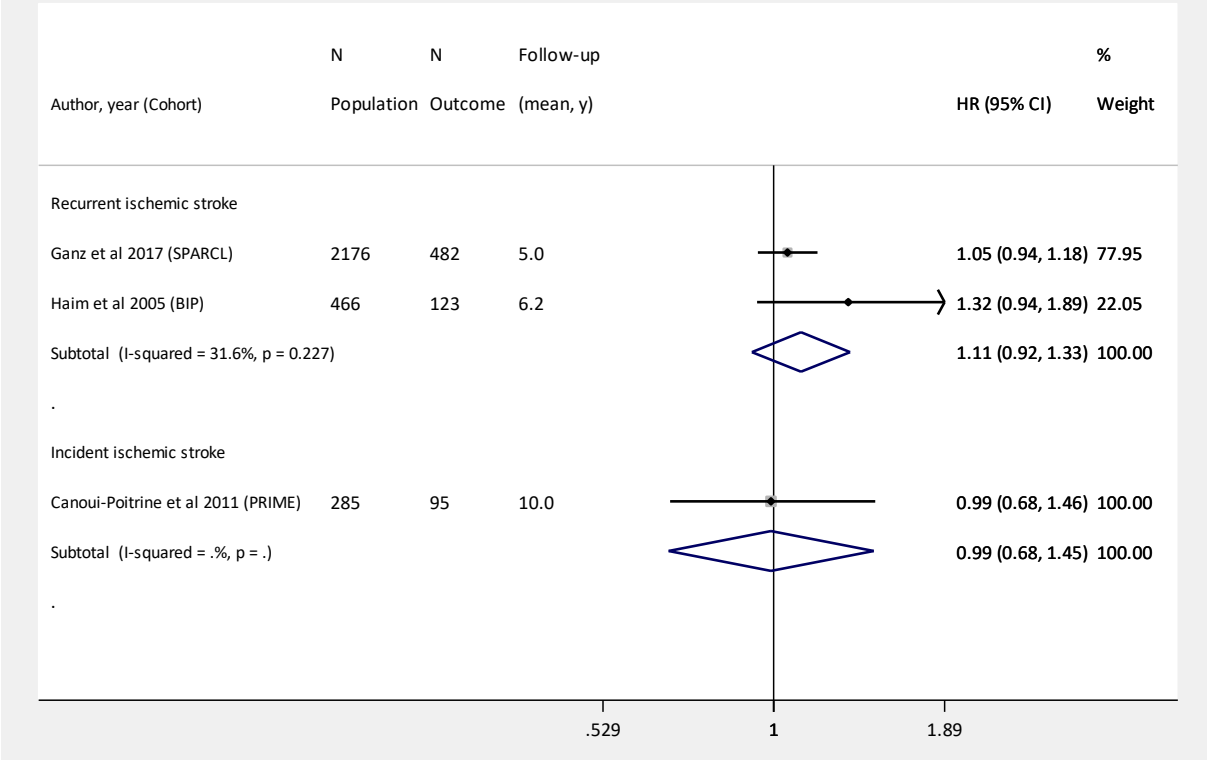


Supplemental Figure 7. Forest plots of the meta-analyses of circulating MCP-1 levels (1 SD increase) and risk of any ischemic stroke, as derived from published observational studies. (A) Case-control studies, (B) cohort studies.

A



B



SUPPLEMENTAL MATERIAL

Georgakis *et al*. Circulating monocyte chemoattractant protein-1 and risk of stroke: a meta-analysis of population-based studies involving 17,180 individuals.

Appendix I. Search strategy.

Online Table I. Summary of the study design, population characteristics, methods used for quantifying circulating MCP-1 levels, stroke outcome definitions, and assessments in the cohorts included in the meta-analysis.

Online Table II. Quality characteristics of the included studies according to the Newcastle-Ottawa Scale.

Online Table III. Associations between baseline circulating MCP-1 levels and risk of any stroke. Shown are the results from random-effects meta-analyses across the different models in the pooled sample consisting of six population-based studies.

Online Table IV. Associations between baseline circulating MCP-1 levels and risk of ischemic stroke. Shown are the results from random-effects meta-analyses across the different models in the pooled sample consisting of six population-based studies.

Online Table V. Associations between baseline circulating MCP-1 levels and risk of hemorrhagic stroke. Shown are the results from random-effects meta-analyses across the different models in the pooled sample consisting of six population-based studies.

Online Table VI. Meta-regression analyses for the effect of different study characteristics on the association between ln-transformed MCP-1 circulating levels at baseline (1 SD increment) with any stroke and etiological stroke subtypes (ischemic and hemorrhagic stroke).

Online Table VII. Associations between baseline circulating hsCRP, IL-6, and MCP-1 levels and risk of any stroke, ischemic stroke, and hemorrhagic stroke. Shown are the results from random-effects meta-analyses of the pooled sample consisting of four population-based studies, where both hsCRP and IL-6 levels were available.

Online Figure I. Flowchart of the study selection for the systematic review.

Online Figure II. Study-specific and pooled hazard ratios for incident any stroke per standard deviation increase in ln-transformed circulating MCP-1 levels and across MCP-1 level quartiles. Shown are the results from random-effects meta-analyses.

Online Figure III. Study-specific and pooled hazard ratios for incident ischemic stroke per standard deviation increase in ln-transformed circulating MCP-1 levels and across MCP-1 level quartiles. Shown are the results from random-effects meta-analyses (Model 2).

Online Figure IV. Study-specific and pooled hazard ratios for incident hemorrhagic stroke per standard deviation increase in ln-transformed circulating MCP-1 levels and across MCP-1 level quartiles. Shown are the results from random-effects meta-analyses (Model 2).

Online Figure V. Pooled hazard ratios for incident fatal and non-fatal stroke per circulating MCP-1 levels, as derived from random-effects meta-analyses (Model 2).

Online Figure VI. Pooled hazard ratios for incident any stroke per standard deviation increase in ln-transformed circulating MCP-1 levels and across MCP-1 level quartiles in sensitivity analyses omitting one study per time. Shown are the results from random-effects meta-analyses.

Online Figure VII. Pooled hazard ratios for incident ischemic stroke per standard deviation increase in ln-transformed circulating MCP-1 levels and across MCP-1 level quartiles in sensitivity analyses omitting one study per time. Shown are the results from random-effects meta-analyses.

Online Figure VIII. Pooled hazard ratios for incident hemorrhagic stroke per standard deviation increase in ln-transformed circulating MCP-1 levels and across MCP-1 level quartiles in sensitivity analyses omitting one study per time. Shown are the results from random-effects meta-analyses.

Online Figure IX. Pooled hazard ratios for incident ischemic stroke per standard deviation increase in ln-transformed circulating MCP-1 levels, as derived from random-effects meta-analyses stratified by pre-defined study variables.

Online References.

Appendix I. Search strategy.

(CCL2 OR MCP1 OR CCL-2 OR MCP-1 OR “monocyte chemoattractant protein 1” OR “small inducible cytokine A2” OR “chemokine (C-C motif) ligand 2” OR “C-C motif ligand 2”) AND (stroke OR cerebrovascular OR (coronary AND artery AND disease) OR (ischemic AND heart AND disease) OR (myocardial AND infarction))

1303 results in PubMed by March 15th 2019

Online Table I. Summary of the study design, population characteristics, methods used for quantifying circulating MCP-1 levels, stroke outcome definitions, and assessments in the cohorts included in the meta-analysis.

Cohort	Study design	Population characteristics	MCP-1 quantification	Definition-assessment of stroke
Atherosclerosis Risk in Communities (ARIC)	A sub-sample of the population-based prospective ARIC cohort study with available measurements on MCP-1[1]	Inhabitants of 4 US communities (Forsyth County, North Carolina; Jackson, Mississippi; the northwestern suburbs of Minneapolis, Minnesota; and Washington County, Maryland) aged 45-64 years	Duplicate measurements using direct sandwich ELISA (Amersham Pharmacia Biotech Inc., Piscataway, NJ, USA) in fasting plasma samples (stored at -70 °C)	Non-fatal and fatal stroke were defined through linkage with the hospital records for possible stroke-related hospitalizations (International Classification of Diseases, Ninth Revision [ICD-9] codes 430–438 until 1997 and codes 430–436 afterwards) and the National Death Index for stroke deaths; physician reviewers adjudicated all possible strokes and classified them as definite or probable ischemic and hemorrhagic events[2]
Dallas Heart Study (DHS)	A sub-sample of a population-based prospective cohort study designed to study cardiovascular disease with available measurements on MCP-1[3]	Multi-ethnic stratified random sample of Dallas County, US, residents aged 30-65 years	Duplicate measurements using immunoassay (BIOSITE Inc., San Diego, CA) on a high-throughput robotic platform (TECAN Genesis RSP 200/8) in fasting plasma samples (stored at -80 °C)	Non-fatal stroke was defined by either assessment of medical records during annual follow-up assessments or by tracking hospital admissions through the Dallas–Fort Worth Hospital Council Data Initiative database (coverage 90% of the study region) using the ICD 9 codes 430-438; fatal stroke was defined by death certification using the National Death Index according to the ICD 10 codes I60-I69[4]
European Prospective Investigation of Cancer (EPIC) - Norfolk study	Secondary analysis of a nested case-control study within the prospective population-based EPIC-Norfolk cohort of cases with coronary artery disease and healthy controls[5]	Inhabitants of Norfolk, UK, aged 45-79 years who were free of stroke and myocardial infarction at baseline	Multiplex assay using the Bioplex Suspension Array (Bio-Rad, Veenendaal, the Netherlands) in non-fasting serum samples (stored at -80 °C)	Non-fatal stroke was defined by hospital admission record linkage with the NHS hospital information system and ENCORE (East Norfolk COmmission Record; fatal stroke was defined by death certification derived from the Office of National Statistics, and was defined according to the ICD 9 codes 430-438, or the ICD 10 codes I60-I69[6]
Framingham Heart Study (FHS) - Offspring Cohort	Participants of the community-based prospective cohort FHS study who attended the examination cycle 7 (1998-2001)[7]	Offspring of the participants of the Original Cohort of the FHS and their spouses aged 33-90 years	Duplicate measurements using a commercially available ELISA (R&D Systems) in fasting serum samples (stored at -70 °C)[8]	Stroke was defined as rapidly developing signs of focal neurologic disturbance of presumed vascular etiology lasting more than 24 hours as part of an ongoing clinic and hospital surveillance including medical record review; laboratory testing; imaging; autopsy findings; and collaboration with general practitioners, emergency departments, and imaging facilities in the area[9]
Monitoring of Trends and Determinants in Cardiovascular Disease sub-cohort of the Cooperative Health Research in the Region of Augsburg (MONICA/ KORA)	Secondary analysis of a case-cohort study within the prospective population-based MONICA/KORA cohort of incident cases with coronary artery disease and a representative sub-cohort of MONICA/KORA sample[10]	Inhabitants of Augsburg and surrounding counties, Germany, aged 25-74 years	Luminex multiplex technology using a Luminex 100 analyzer (Luminex Corporation, Austin, TX, recombinant proteins and antibodies purchased from R&D systems) in non-fasting serum samples (stored at -80 °C)	Non-fatal stroke was defined by self-report validated by cross-linkage with hospital records and information gathered from the treating physicians of the participants; fatal stroke was defined by death certification derived from local health authorities and was defined according to the ICD 9 codes 430-434 (German modified version)[11]
Malmö Diet and Cancer Study (MDCS) - Cardiovascular (CV) sub-cohort	A random 50% sub-sample of the population-based prospective cohort MDCS study were included in the MDCS-CV sub-cohort designed to examine cardiovascular disease[12]	Inhabitants of Malmö, Sweden, aged 45-64 years	Proximity Extension Assay technique using the Proseek Multiplex CVD96x96 reagents kit (Olink Bioscience) in fasting plasma samples (stored at -80 °C)	Non-fatal and fatal stroke were defined by record linkage with the National Inpatient Register, the Swedish Causes of Death Register, and the Stroke Register of Malmö (STROMA) and was defined according to the ICD 9 codes 430-438[13]

Online Table II. Quality characteristics of the included studies according to the Newcastle-Ottawa Scale.

Cohort	ARIC	DHS	EPIC-Norfolk	FHS Offspring	MONICA/KORA	MDCS-CV
Selection items						
Representativeness of exposed cohort (general population study)	*	*	*	*	*	*
Selection of the non-exposed cohort (patients selected independently of MCP-1 levels)	*	*	*	*	*	*
Ascertainment of exposure (serum/plasma MCP-1 levels assessed with validated assay)	*	*	*	*	*	*
Outcome not present a start of study (exclusion of prevalent stroke cases from analysis)	*	*	*	*	*	*
Comparability items						
Adjustments on age, sex, race	*	*	*	*	*	*
Adjustments on vascular risk factors	*	*	*	*	*	*
Outcome items						
Assessment of outcome (assessment through medical records, hospital admission records, and death certificates)	*	*	*	*	*	*
Length of follow-up (>5 years)	*	*	*	*	*	*
Adequacy of follow-up cohorts (<10% lost to follow-up rates)	*	*	*	*	*	*
Total score	**9/9**	**9/9**	**9/9**	**9/9**	**9/9**	**9/9**

Online Table III. Associations between baseline circulating MCP-1 levels and risk of any stroke. Shown are the results from random-effects meta-analyses across the different models in the pooled sample consisting of six population-based studies.

	Model 1			Model 2			Alternative Model 2			Model 3		
Variables in the models	**HR**	**95%CI**	**p**	**HR**	**95%CI**	**p**	**HR**	**95%CI**	**p**	**HR**	**95%CI**	**p**
Age (1-yr increment)	1.09	(1.07-1.12)	7E-13	1.08	(1.05-1.11)	7E-8	1.07	(1.04-1.11)	2E-6	1.08	(1.05-1.11)	2E-7
Sex (males vs. females)	1.26	(0.98-1.62)	0.067	1.21	(1.00-1.48)	0.056	1.13	(0.93-1.36)	0.214	1.22	(1.00-1.48)	0.051
Hypertension (yes vs. no)				1.80	(1.58-2.04)	2E-19				1.78	(1.57-2.03)	1E-20
SBP (10 mmHg-increment)							1.16	(1.12-1.19)	3E-18			
Intake of antihypertensive medication							1.47	(1.29-1.67)	5E-9			
Diabetes (yes vs. no)				1.739	(1.27-2.38)	0.001				1.79	(1.26-2.53)	0.001
Fasting glucose levels (10 mg/dl increment)							1.03	(1.00-1.07)	0.04			
Intake of glucose-lowering medication							1.33	(0.93-1.91)	0.117			
Smoking (current vs. non-current)				1.594	(0.99-2.56)	0.054	1.52	(0.94-2.46)	0.086	1.51	(0.98-2.34)	0.062
Hypercholesterolemia (yes vs. no)				1.021	(0.88-1.19)	0.784				1.02	(0.89-1.16)	0.804
LDL-C levels (10 mg/dl increment)							1.01	(0.99-1.02)	0.406			
HDL-C levels (5 mg/dl increment)							0.98	(0.95-1.01)	0.269			
Intake of lipid-lowering medication							1.05	(0.82-1.35)	0.694			
Chronic kidney disease (yes vs. no)				1.00	(0.89-1.12)	0.999				0.97	(0.89-1.06)	0.546
eGFR (10 ml/min/1.73 m2 increment)							1.00	(0.99-1.00)	0.48			
BMI (5 kg/m2 increment)				1.01	(0.91-1.11)	0.896	0.96	(0.87-1.05)	0.336	0.97	(0.95-1.00)	0.044
Heart failure (yes vs. no)				1.18	(0.80-1.73)	0.402	1.35	(0.91-1.99)	0.134	1.18	(0.80-1.76)	0.405
Coronary artery disease (yes vs. no)				1.80	(1.38-2.34)	2E-5	1.74	(1.32-2.29)	8E-5	1.76	(1.35-2.31)	4E-5
Atrial fibrillation (yes vs. no)				1.50	(0.94-2.39)	0.091	1.48	(0.92-2.36)	0.106	1.51	(0.94-2.41)	0.086
ln-hsCRP (1-SD increment)										1.12	(1.05-1.19)	0.0003
ln-MCP1 (1-SD increment)	1.10	(1.01-1.19)	0.018	1.07	(1.01-1.14)	0.028	1.07	(1.00-1.15)	0.035	1.07	(1.00-1.14)	0.053
1st quartile		reference			reference			reference			Reference	
2nd quartile	1.17	(1.00-1.37)	0.058	1.16	(0.99-1.36)	0.075	1.16	(0.98-1.38)	0.079	1.18	(1.00-1.38)	0.048
3rd quartile	1.35	(1.16-1.57)	0.0001	1.31	(1.12-1.53)	0.001	1.35	(1.14-1.58)	0.0003	1.32	(1.13-1.55)	0.0004
4th quartile	1.43	(1.10-1.86)	0.004	1.33	(1.05-1.68)	0.008	1.37	(1.09-1.72)	0.005	1.34	(1.08-1.65)	0.007

All models are additionally adjusted for race, but following study-specific classifications that precluded meta-analysis for this variable.

Abbreviations: SBP, systolic blood pressure; DBP, diastolic blood pressure; LDL-C, low-density lipoprotein cholesterol; HDL-C, high-density lipoprotein cholesterol; eGFR, estimated glomerular filtration rate; BMI, body mass index; hsCRP, high-sensitivity C-reactive protein; MCP-1, monocyte-chemoattractant protein 1; HR, hazard ratio; SD, standard deviation.

Online Table IV. Associations between baseline circulating MCP-1 levels and risk of ischemic stroke. Shown are the results from random-effects meta-analyses across the different models in the pooled sample consisting of six population-based studies.

	Model 1			Model 2			Alternative Model 2			Model 3		
Variables in the models	**HR**	**95%CI**	**p**	**HR**	**95%CI**	**p**	**HR**	**95%CI**	**p**	**HR**	**95%CI**	**p**
Age (1-yr increment)	1.10	(1.07-1.12)	4E-13	1.08	(1.05-1.11)	7E-7	1.08	(1.04-1.11)	7E-6	1.08	(1.05-1.11)	4E-7
Sex (males vs. females)	1.28	(1.00-1.64)	0.050	1.22	(1.02-1.45)	0.029	1.12	(0.94-1.34)	0.193	1.23	(1.03-1.46)	0.022
Hypertension (yes vs. no)				1.80	(1.57-2.06)	3E-17				1.78	(1.55-2.05)	4E-16
SBP (10 mmHg-increment)							1.15	(1.10-1.20)	3E-11			
Intake of antihypertensive medication							1.52	(1.32-1.75)	3E-9			
Diabetes (yes vs. no)				1.88	(1.33-2.64)	0.0003				1.90	(1.32-2.72)	0.001
Fasting glucose levels (10 mg/dl increment)							1.04	(1.01-1.07)	0.013			
Intake of glucose-lowering medication							1.33	(0.90-1.96)	0.154			
Smoking (current vs. non-current)				1.55	(0.95-2.54)	0.082	1.47	(0.89-2.44)	0.137	1.48	(0.93-2.34)	0.097
Hypercholesterolemia (yes vs. no)				1.09	(0.92-1.28)	0.314				1.09	(0.94-1.26)	0.260
LDL-C levels (10 mg/dl increment)							1.01	(1.00-1.03)	0.112			
HDL-C levels (5 mg/dl increment)							0.98	(0.96-1.01)	0.243			
Intake of lipid-lowering medication							1.12	(0.86-1.47)	0.404			
Chronic kidney disease (yes vs. no)				0.97	(0.85-1.11)	0.664				0.94	(0.85-1.03)	0.198
eGFR (10 ml/min/1.73 m2 increment)							1.00	(0.99-1.00)	0.268			
BMI (5 kg/m2 increment)				1.01	(0.90-1.13)	0.877	0.95	(0.84-1.07)	0.412	0.99	(0.92-1.06)	0.721
Heart failure (yes vs. no)				1.16	(0.76-1.77)	0.501	1.29	(0.84-2.00)	0.246	1.16	(0.75-1.81)	0.508
Coronary artery disease (yes vs. no)				1.74	(1.22-2.48)	0.002	1.64	(1.13-2.38)	0.009	1.55	(0.97-2.48)	0.068
Atrial fibrillation (yes vs. no)				1.54	(0.94-2.54)	0.088	1.53	(0.93-2.54)	0.097	1.56	(0.95-2.56)	0.083
ln-hsCRP (1-SD increment)										1.14	(1.07-1.22)	0.0002
ln-MCP1 (1-SD increment)	1.12	(1.03-1.23)	0.007	1.11	(1.02-1.21)	0.009	1.11	(1.02-1.21)	0.011	1.10	(1.01-1.21)	0.018
1st quartile		reference			reference			reference			reference	
2nd quartile	1.19	(1.01-1.41)	0.039	1.19	(1.00-1.42)	0.047	1.17	(0.97-1.41)	0.089	1.22	(1.03-1.45)	0.022
3rd quartile	1.38	(1.17-1.63)	0.0001	1.35	(1.14-1.59)	0.0004	1.38	(1.16-1.65)	0.0003	1.36	(1.15-1.60)	0.0003
4th quartile	1.43	(1.11-1.85)	0.003	1.38	(1.07-1.77)	0.008	1.39	(1.10-1.76)	0.006	1.38	(1.10-1.74)	0.004

All models are additionally adjusted for race, but following study-specific classifications that precluded meta-analysis for this variable.

Abbreviations: SBP, systolic blood pressure; DBP, diastolic blood pressure; LDL-C, low-density lipoprotein cholesterol; HDL-C, high-density lipoprotein cholesterol; eGFR, estimated glomerular filtration rate; BMI, body mass index; hsCRP, high-sensitivity C-reactive protein; MCP-1, monocyte-chemoattractant protein 1; HR, hazard ratio; SD, standard deviation.

Online Table V. Associations between baseline circulating MCP-1 levels and risk of hemorrhagic stroke. Shown are the results from random-effects meta-analyses across the different models in the pooled sample consisting of six population-based studies.

	Model 1			Model 2			Alternative Model 2			Model 3		
Variables in the models	**HR**	**95%CI**	**p**	**HR**	**95%CI**	**p**	**HR**	**95%CI**	**p**	**HR**	**95%CI**	**p**
Age (1-yr increment)	1.08	(1.06-1.10)	0	1.08	(1.05-1.10)	7E-10	1.06	(1.03-1.09)	7E-5	1.07	(1.03-1.11)	0.0001
Sex (males vs. females)	1.05	(0.62-1.78)	0.847	1.04	(0.63-1.71)	0.879	0.82	(0.49-1.37)	0.446	0.89	(0.64-1.22)	0.453
Hypertension (yes vs. no)				1.94	(1.39-2.71)	0.0001				1.95	(1.39-2.73)	0.0001
SBP (10 mmHg-increment)							1.23	(1.14-1.34)	3E-7			
Intake of antihypertensive medication							1.32	(0.82-2.13)	0.250			
Diabetes (yes vs. no)				1.05	(0.67-1.65)	0.832				1.05	(0.66-1.65)	0.842
Fasting glucose levels (10 mg/dl increment)							0.95	(0.88-1.03)	0.224			
Intake of glucose-lowering medication							2.81	(0.94-8.38)	0.065			
Smoking (current vs. non-current)				1.57	(0.90-2.73)	0.110	1.49	(0.82-2.72)	0.193	1.36	(0.96-1.92)	0.087
Hypercholesterolemia (yes vs. no)				0.83	(0.59-1.17)	0.286				0.80	(0.56-1.13)	0.199
LDL-C levels (10 mg/dl increment)							0.98	(0.94-1.03)	0.465			
HDL-C levels (5 mg/dl increment)							1.05	(0.93-1.18)	0.417			
Intake of lipid-lowering medication							1.05	(0.48-2.32)	0.905			
Chronic kidney disease (yes vs. no)				1.17	(0.76-1.81)	0.474				1.17	(0.75-1.82)	0.487
eGFR (10 ml/min/1.73 m2 increment)							1.00	(0.92-1.10)	0.937			
BMI (5 kg/m2 increment)				0.93	(0.75-1.15)	0.493	0.94	(0.71-1.23)	0.645	0.94	(0.83-1.07)	0.330
Heart failure (yes vs. no)				6.93	(1.65-29.2)	0.008	12.0	(3.46-41.7)	9E-5	6.52	(1.24-34.2)	0.027
Coronary artery disease (yes vs. no)				1.30	(0.49-3.48)	0.601	1.37	(0.50-3.76)	0.547	1.42	(0.53-3.86)	0.488
Atrial fibrillation (yes vs. no)				3.97	(0.94-16.7)	0.061	3.83	(0.89-16.4)	0.071	3.90	(0.93-16.4)	0.064
ln-hsCRP (1-SD increment)										1.13	(0.96-1.34)	0.140
ln-MCP1 (1-SD increment)	1.05	(0.84-1.30)	0.669	1.02	(0.82-1.29)	0.833	1.04	(0.79-1.37)	0.776	1.02	(0.80-1.31)	0.844
1st quartile		reference			reference			reference			reference	
2nd quartile	0.96	(0.62-1.50)	0.873	0.95	(0.61-1.47)	0.807	0.97	(0.60-1.57)	0.907	0.96	(0.62-1.49)	0.860
3rd quartile	1.27	(0.84-1.92)	0.251	1.25	(0.82-1.91)	0.293	1.31	(0.80-2.15)	0.276	1.27	(0.84-1.93)	0.252
4th quartile	1.09	(0.71-1.66)	0.692	1.02	(0.66-1.56)	0.945	1.07	(0.67-1.71)	0.768	1.02	(0.67-1.57)	0.921

All models are additionally adjusted for race, but following study-specific classifications that precluded meta-analysis for this variable. The Dallas Heart Study (DHS) is not included in any of the analyses for hemorrhagic stroke due to the low number of events. The Atherosclerosis Risk in Community (ARIC) study is not included in the quartile analyses.

Abbreviations: SBP, systolic blood pressure; DBP, diastolic blood pressure; LDL-C, low-density lipoprotein cholesterol; HDL-C, high-density lipoprotein cholesterol; eGFR, estimated glomerular filtration rate; BMI, body mass index; hsCRP, high-sensitivity C-reactive protein; MCP-1, monocyte-chemoattractant protein 1; HR, hazard ratio; SD, standard deviation.

Online Table VI. Meta-regression analyses for the effect of different study characteristics on the association between ln-transformed MCP-1 circulating levels at baseline (1 SD increment) with any stroke and etiological stroke subtypes (ischemic and hemorrhagic stroke).

	Any stroke		Ischemic stroke		Hemorrhagic stroke	
Variable	**Exponentiated regression coefficient (95% CI)**	**p**	**Exponentiated regression coefficient (95% CI)**	**p**	**Exponentiated regression coefficient (95% CI)**	**p**
Age (1y-increment)	0.993 (0.979-1.007)	0.24	0.989 (0.974-1.005)	0.12	1.002 (0.914-1.099)	0.95
Males (5%-increment)	1.003 (0.941-1.068)	0.91	0.994 (0.919-1.075)	0.85	1.063 (0.950-1.190)	0.18
SBP (10 mmHg-increment)	0.932 (0.814-1.066)	0.22	0.897 (0.774-1.040)	0.11	1.065 (0.540-2.097)	0.79
Diabetes (5%-increment)	0.987 (0.903-1.079)	0.71	0:983 (0.877-1.102)	0.69	1.063 (0.857-1.320)	0.43
LDL-C (10 mg/dl-increment)	0.984 (0.933-1.037)	0.43	0.968 (0.919-1.020)	0.16	1.054 (0.833-1.335)	0.53
BMI (5kg/m^2-increment)	1.160 (0.776-1.734)	0.36	1.298 (0.856-1.970)	0.16	0.978 (0.098-9.707)	0.98
Current smokers (5%-increment)	0.997 (0.937-1.061)	0.91	0.994 (0.917-1.077)	0.84	1.076 (0.950-1.219)	0.16
eGFR (10ml/min/1.73m^2-increment)	1.064 (0.971-1.166)	0.13	1.090 (0.987-1.203)	0.07	1.016 (0.592-1.743)	0.93
Coronary artery disease (5%-increment)	1.033 (0.870-1.227)	0.63	1.058 (0.877-1.277)	0.45	0.830 (0.510-1.351)	0.31
hsCRP (1 unit-increment in ln(hsCRP))	1.028 (0.696-1.517)	0.84	1.125 (0.643-1.971)	0.55	0.992 (0.102-9.615)	0.99
Sample (serum vs. plasma)	0.985 (0.800-1.247)	0.88	0.943 (0.704-1.262)	0.61	1.043 (0.443-2.457)	0.89

Abbreviations: BMI, body mass index; hsCRP, high-sensitivity C-reactive protein; eGFR, estimated glomerular filtration rate; LDL, low-density liporprotein; MCP-1, monocyte chemoattractant protein- 1; SBP, systolic blood pressure.

Online Table VII. Associations between baseline circulating hsCRP, IL-6, and MCP-1 levels and risk of any stroke, ischemic stroke, and hemorrhagic stroke. Shown are the results from random-effects meta-analyses of the pooled sample consisting of four population-based studies, where both hsCRP and IL-6 levels were available.

			Any stroke				Ischemic stroke				Hemorrhagic stroke *			
Variables in the models	**Population**	**Follow-up (y)**	**Events**	**HR**	**95%CI**	**p**	**Events**	**HR**	**95%CI**	**p**	**Events**	**HR**	**95%CI**	**p**
Model adjusted for age, sex, race, vascular risk factors†														
ln-MCP1 (1-SD increment)	12686	15.6	777	1.08	(1.00-1.16)	0.056	634	1.12	(1.02-1.24)	0.020	108	0.90	(0.74-1.10)	0.298
1st quartile	3184	15.7	145		reference		114		reference		26		reference	
2nd quartile	3162	15.7	177	1.09	(0.87-1.37)	0.468	144	1.12	(0.87-1.43)	0.390	24	0.95	(0.51-1.79)	0.876
3rd quartile	3177	15.6	212	1.21	(0.98-1.50)	0.080	175	1.27	(1.01-1.62)	0.044	31	1.15	(0.58-2.28)	0.692
4th quartile	3163	15.3	243	1.33	(1.05-1.69)	0.014	201	1.43	(1.04-1.97)	0.022	27	0.91	(0.52-1.60)	0.745
Model adjusted for age, sex, race, vascular risk factors†, hsCRP levels														
ln-hsCRP (1-SD increment)	12519	15.6	773	1.11	(1.03-1.20)	0.009	616	1.14	(1.05-1.24)	0.003	107	1.03	(0.83-1.26)	0.803
ln-MCP1 (1-SD increment)	12519	15.6	773	1.06	(0.98-1.14)	0.098	616	1.12	(1.00-1.26)	0.048	107	0.91	(0.74-1.10)	0.321
1st quartile	3155	15.7	142		reference		110		reference		25		reference	
2nd quartile	3128	15.7	178	1.09	(0.87-1.36)	0.449	143	1.12	(0.87-1.44)	0.374	24	0.95	(0.51-1.77)	0.870
3rd quartile	3138	15.6	213	1.22	(0.98-1.51)	0.073	174	1.28	(1.01-1.63)	0.041	31	1.16	(0.59-2.29)	0.661
4th quartile	3098	15.3	240	1.32	(1.02-1.72)	0.039	189	1.42	(1.03-1.99)	0.037	27	0.92	(0.52-1.62)	0.777
Model adjusted for age, sex, race, vascular risk factors†, IL-6 levels														
ln-IL-6 (1-SD increment)	12516	15.6	758	1.12	(1.04-1.21)	0.003	614	1.17	(1.02-1.35)	0.025	107	1.12	(0.92-1.36)	0.251
ln-MCP1 (1-SD increment)	12516	15.6	769	1.05	(0.98-1.4)	0.146	614	1.12	(0.99-1.28)	0.064	107	0.88	(0.72-1.08)	0.210
1st quartile	3168	15.7	142		reference		109		reference		25		reference	
2nd quartile	3148	15.7	177	1.09	(0.87-1.36)	0.465	142	1.10	(0.86-1.42)	0.445	24	0.96	(0.49-1.88)	0.901
3rd quartile	3160	15.6	212	1.20	(0.96-1.49)	0.098	174	1.24	(0.97-1.58)	0.079	31	1.13	(0.56-2.27)	0.736
4th quartile	3141	15.3	238	1.31	(0.97-1.76)	0.086	189	1.39	(0.99-1.96)	0.052	27	0.86	(0.48-1.53)	0.611
Model adjusted for age, sex, race, vascular risk factors†, hsCRP, and IL-6 levels														
ln-hsCRP (1-SD increment)	12516	15.6	758	1.08	(1.00-1.19)	0.058	610	1.12	(1.02-1.23)	0.018	107	0.88	(0.79-1.23)	0.877
ln-IL-6 (1-SD increment)	12516	15.6	758	1.09	(1.00-1.19)	0.041	610	1.13	(0.96-1.35)	0.137	107	1.13	(0.92-1.40)	0.248
ln-MCP1 (1-SD increment)	12516	15.6	758	1.05	(0.98-1.13)	0.178	610	1.12	(0.98-1.29)	0.078	107	0.88	(0.72-1.08)	0.234
1st quartile	3168	15.7	141		reference		107		reference		25		reference	
2nd quartile	3148	15.7	176	1.10	(0.88-1.37)	0.422	141	1.12	(0.87-1.44)	0.398	24	0.96	(0.49-1.88)	0.914
3rd quartile	3160	15.6	211	1.21	(0.98-1.51)	0.078	173	1.26	(0.99-1.61)	0.059	31	1.14	(0.56-2.30)	0.718
4th quartile	3141	15.3	230	1.30	(0.97-1.76)	0.096	189	1.39	(0.98-1.99)	0.063	27	0.88	(0.49-1.56)	0.660

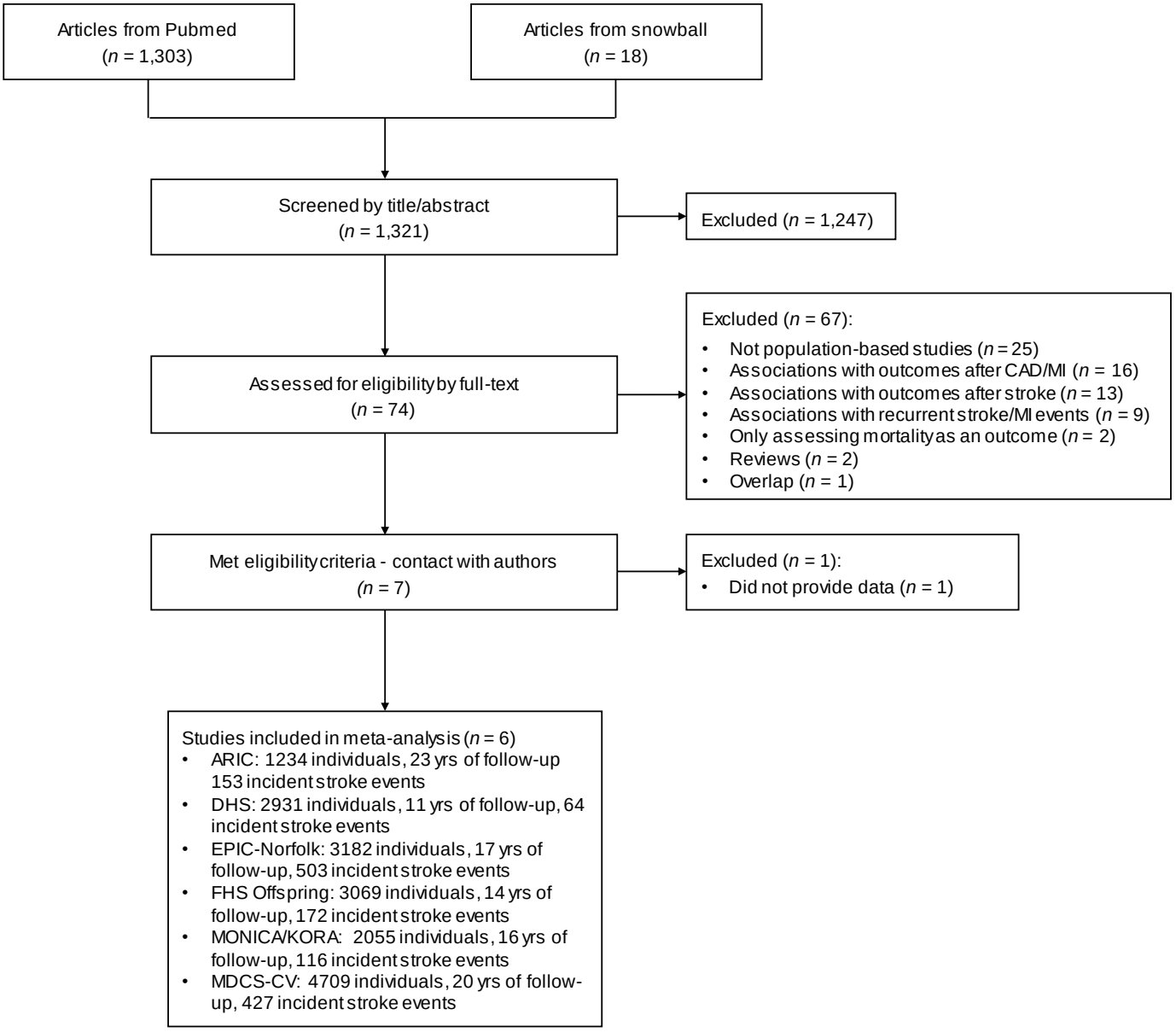
The Atherosclerosis Risk in Community (ARIC) and the European Prospective Investigation of Cancer-Norfolk (EPIC-Norfolk) studies are not included in these analyses because of non-availability of data on IL-6 levels.

* The Dallas Heart Study (DHS) is not included in any of the analyses for hemorrhagic stroke due to the low number of events.

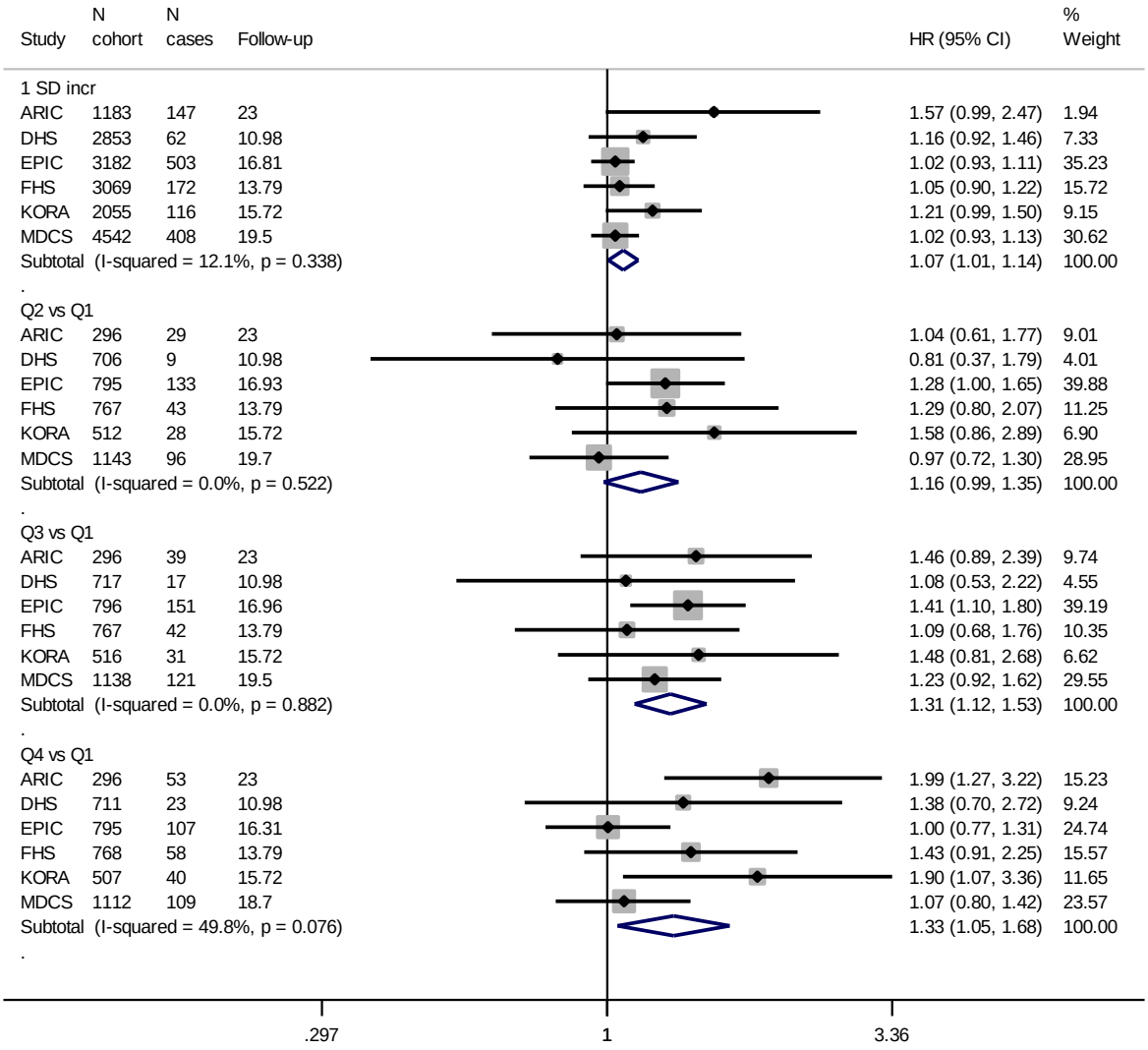
† Vascular risk factors included the models are: body mass index (1 kg/m^2 increment), smoking (current vs. non-current), estimated glomerular filtration rate (1 mL/min/1.73 m^2 increment), history of coronary artery disease, diabetes mellitus, hypertension, hypercholesterolemia, atrial fibrillation, and heart failure at baseline.

Abbreviations: MCP-1, monocyte-chemoattractant protein 1; hsCRP, high-sensitivity C-reactive protein; IL-6, interleukin-6; HR, hazard ratio; SD, standard deviation.

Online Figure I. Flowchart of the study selection for the systematic review.

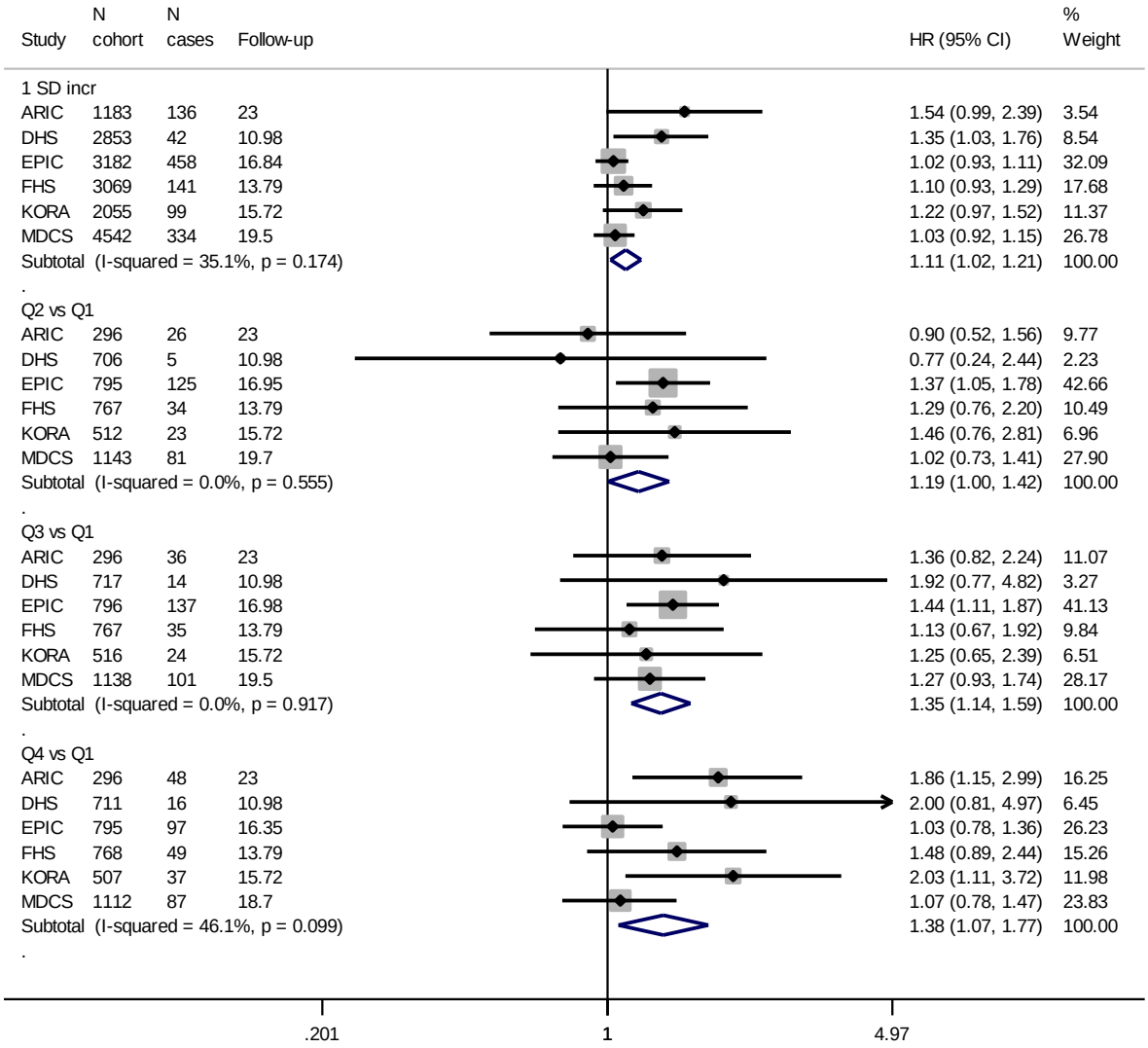


Online Figure II. Study-specific and pooled hazard ratios for incident any stroke per standard deviation increase in ln-transformed circulating MCP-1 levels and across MCP-1 level quartiles. Shown are the results from random-effects meta-analyses (Model 2).



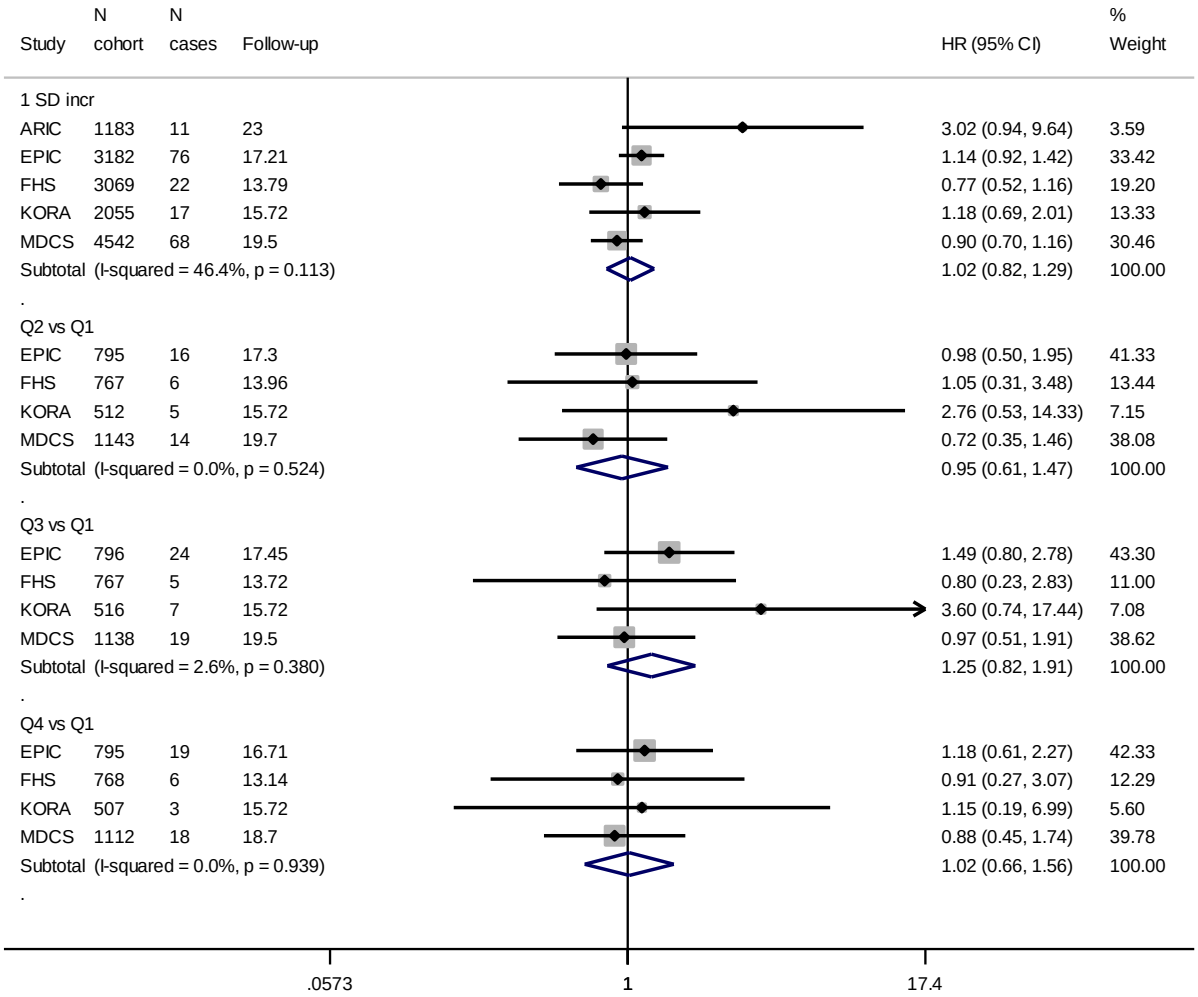
The results are derived from Cox proportional hazard models adjusted for age, sex, race, body mass index (1 kg/m2 increment), smoking (current vs. non-current), estimated glomerular filtration rate (1 mL/min/1.73 m² increment), history of coronary artery disease, diabetes mellitus, hypertension, hypercholesterolemia, atrial fibrillation, and heart failure at baseline.
Analyses for 1 SD increment correspond to ln-transformed MCP-1 levels.
The gray squares around the point estimates correspond to the weight of the included studies in the meta-analysis.
Abbreviations: ARIC, Atherosclerosis Risk in Communities Study; DHS, Dallas Heart Study; EPIC, European Prospective Investigation of Cancer; FHS Framingham Heart Study; KORA, Kooperative Gesundheitsforschung in der Region Augsburg; MDCS, Malmö Diet and Cancer Study.

Online Figure III. Study-specific and pooled hazard ratios for incident ischemic stroke per standard deviation increase in ln-transformed circulating MCP-1 levels and across MCP-1 level quartiles. Shown are the results from random-effects meta-analyses (Model 2).



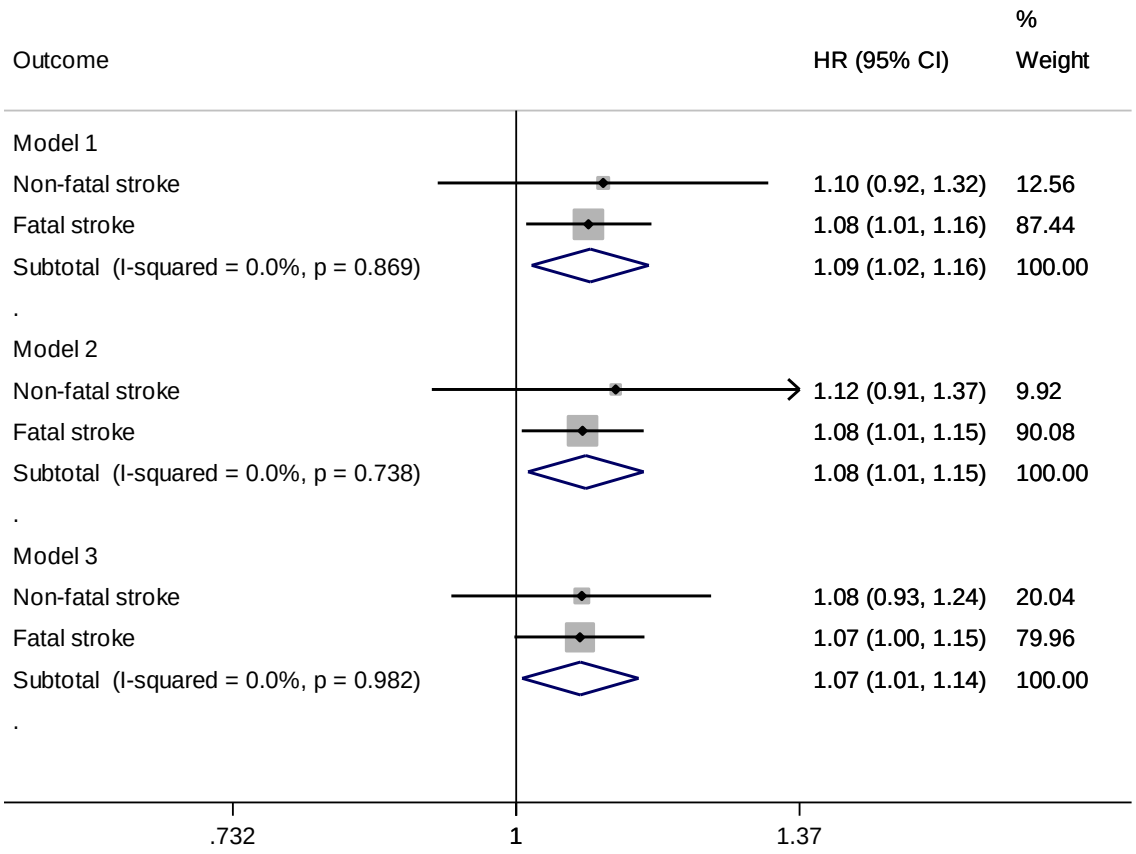
The results are derived from Cox proportional hazard models adjusted for age, sex, race, body mass index (1 kg/m2 increment), smoking (current vs. non-current), estimated glomerular filtration rate (1 mL/min/1.73 m² increment), history of coronary artery disease, diabetes mellitus, hypertension, hypercholesterolemia, atrial fibrillation, and heart failure at baseline.
Analyses for 1 SD increment correspond to ln-transformed MCP-1 levels.
The gray squares around the point estimates correspond to the weight of the included studies in the meta-analysis.
Abbreviations: ARIC, Atherosclerosis Risk in Communities Study; DHS, Dallas Heart Study; EPIC, European Prospective Investigation of Cancer; FHS Framingham Heart Study; KORA, Kooperative Gesundheitsforschung in der Region Augsburg; MDCS, Malmö Diet and Cancer Study.

Online Figure IV. Study-specific and pooled hazard ratios for incident hemorrhagic stroke per standard deviation increase in ln-transformed circulating MCP-1 levels and across MCP-1 level quartiles. Shown are the results from random-effects meta-analyses (Model 2).



The results are derived from Cox proportional hazard models adjusted for age, sex, race, body mass index (1 kg/m2 increment), smoking (current vs. non-current), estimated glomerular filtration rate (1 mL/min/1.73 m² increment), history of coronary artery disease, diabetes mellitus, hypertension, hypercholesterolemia, atrial fibrillation, and heart failure at baseline.
Analyses for 1 SD increment correspond to ln-transformed MCP-1 levels.
The Dallas Heart Study (DHS) is not included in any of the analyses for hemorrhagic stroke due to the low number of events.
The Atherosclerosis Risk in Community (ARIC) study is not included in the quartile analyses due to the low number of events.
The gray squares around the point estimates correspond to the weight of the included studies in the meta-analysis.
Abbreviations: ARIC, Atherosclerosis Risk in Communities Study; EPIC, European Prospective Investigation of Cancer; FHS Framingham Heart Study; KORA, Kooperative Gesundheitsforschung in der Region Augsburg; MDCS, Malmö Diet and Cancer Study.

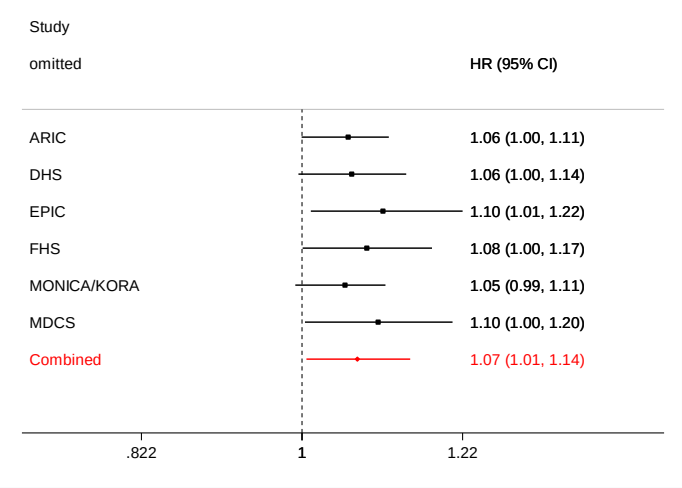
Online Figure V. Pooled hazard ratios for incident fatal and non-fatal stroke per circulating MCP-1 levels, as derived from random-effects meta-analyses.



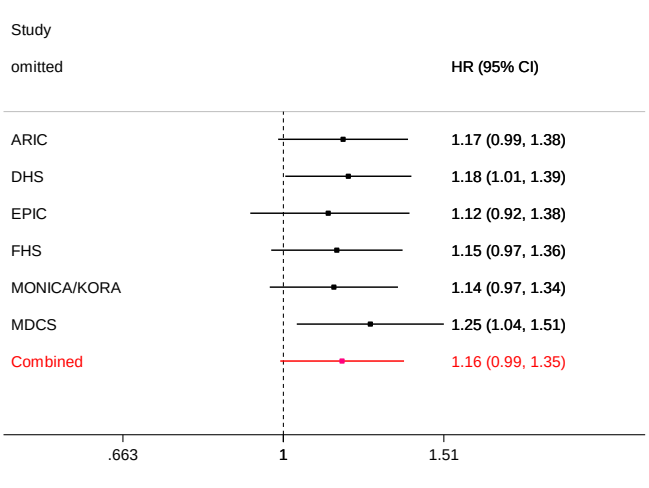
Analyses correspond to 1 SD increment in ln-transformed MCP-1 levels and represent pooled results of meta-analyses of all six studies. The results are derived from Cox proportional hazard models adjusted for age, sex, race, body mass index (1 kg/m2 increment), smoking (current vs. non-current), estimated glomerular filtration rate (1 mL/min/1.73 m² increment), history of coronary artery disease, diabetes mellitus, hypertension, hypercholesterolemia, atrial fibrillation, and heart failure at baseline (Model 2).

Online Figure VI. Pooled hazard ratios for incident any stroke per standard deviation increase in ln-transformed circulating MCP-1 levels and across MCP-1 level quartiles in sensitivity analyses omitting one study per time. Shown are the results from random-effects meta-analyses.

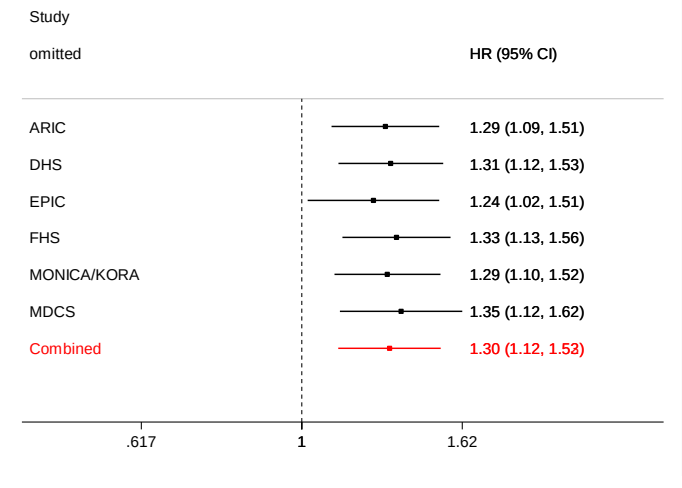
(A) 1 SD increment



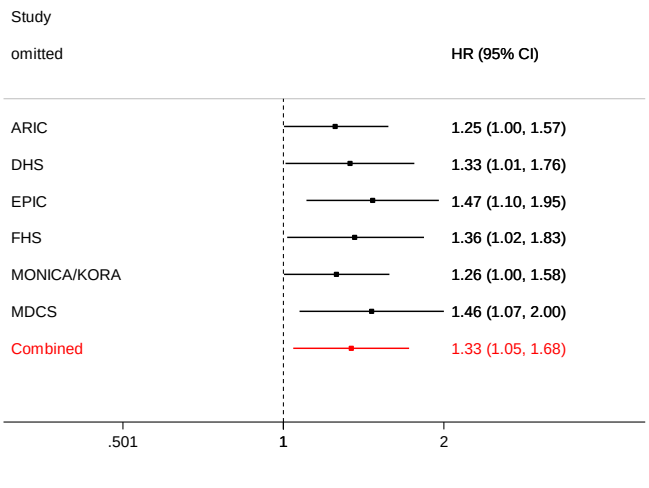
(B) Q2 vs. Q1



(C) Q3 vs. Q1



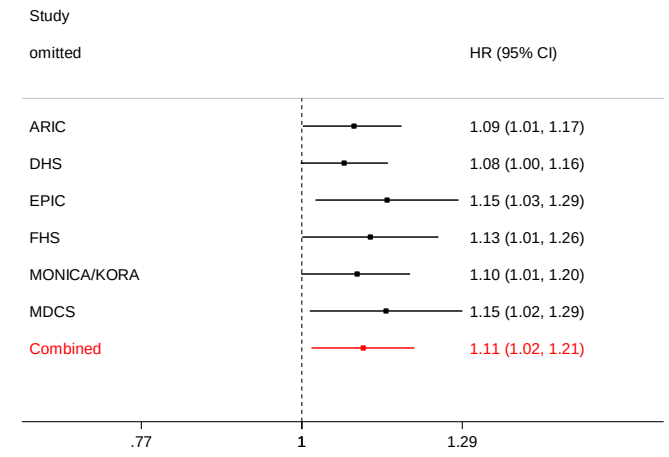
(D) Q4 vs. Q1



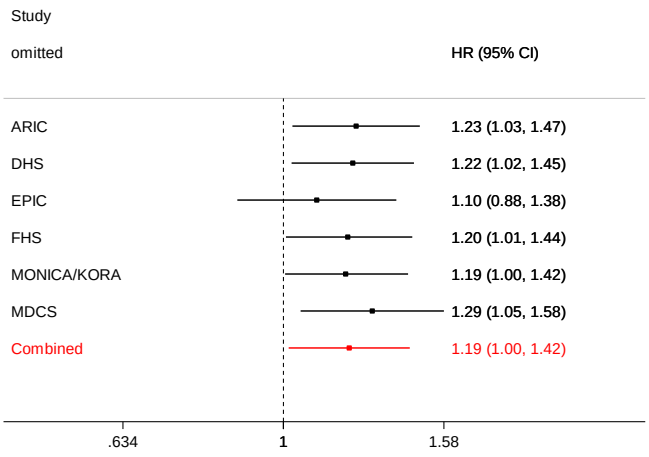
The results are derived from Cox proportional hazard models adjusted for age, sex, race, body mass index (1 kg/m2 increment), smoking (current vs. non-current), estimated glomerular filtration rate (1 mL/min/1.73 m² increment), history of coronary artery disease, diabetes mellitus, hypertension, hypercholesterolemia, atrial fibrillation, and heart failure at baseline.
Analyses for 1 SD increment correspond to ln-transformed MCP-1 levels.

Online Figure VII. Pooled hazard ratios for incident ischemic stroke per standard deviation increase in ln-transformed circulating MCP-1 levels and across MCP-1 level quartiles in sensitivity analyses omitting one study per time. Shown are the results from random-effects meta-analyses.

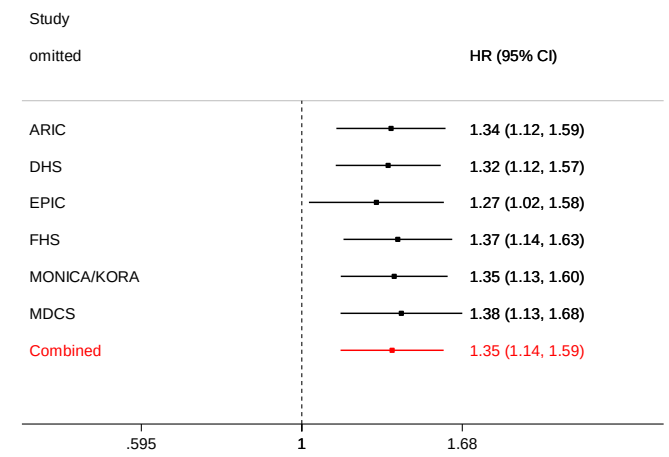
(B) 1 SD increment



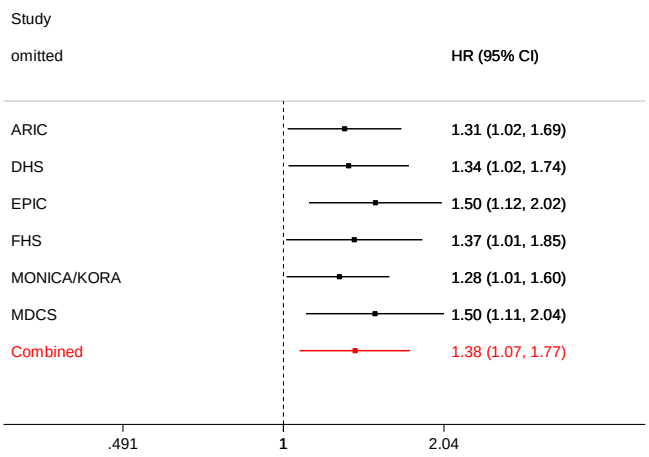
(B) Q2 vs. Q1



(D) Q3 vs. Q1

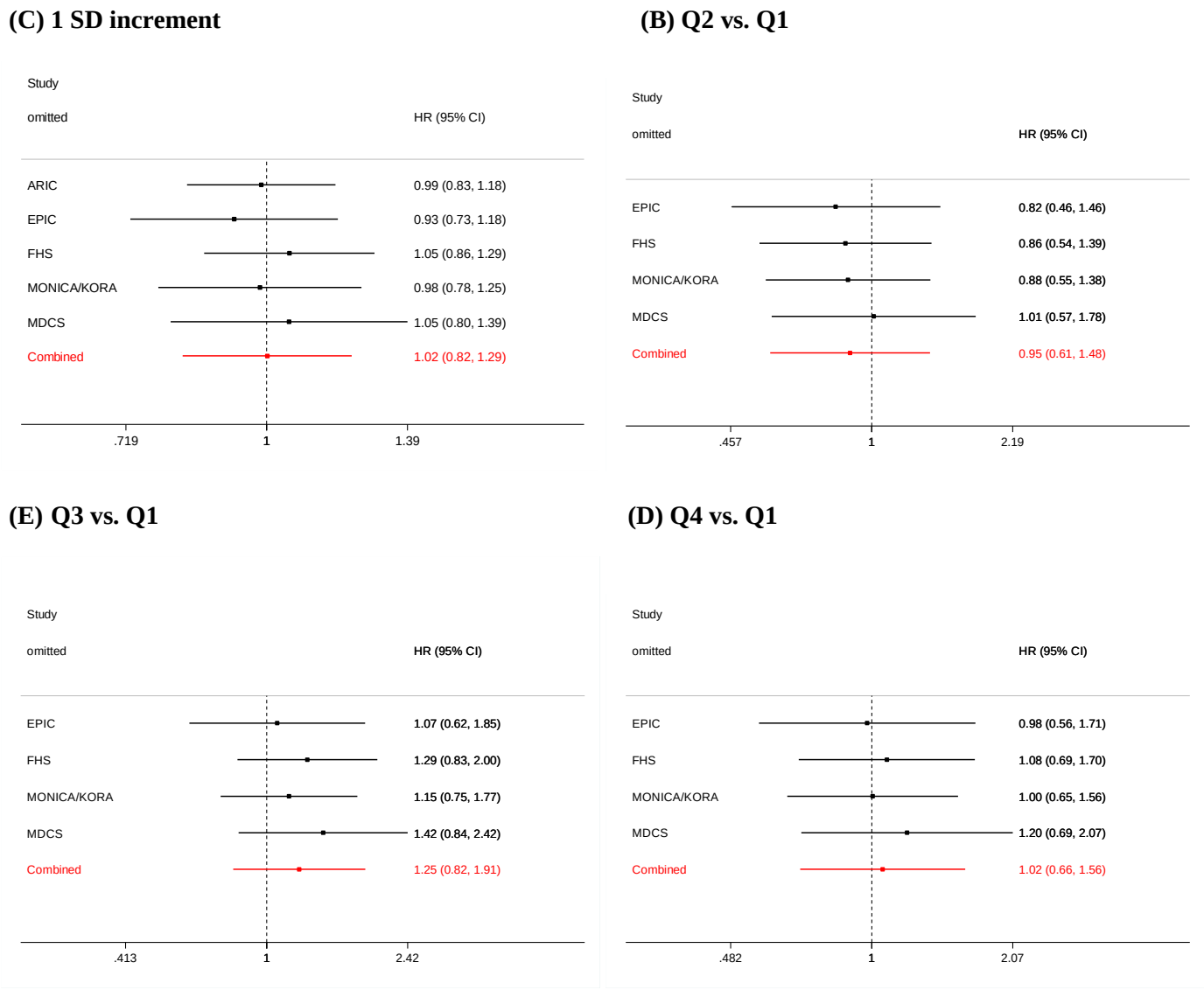


(D) Q4 vs. Q1



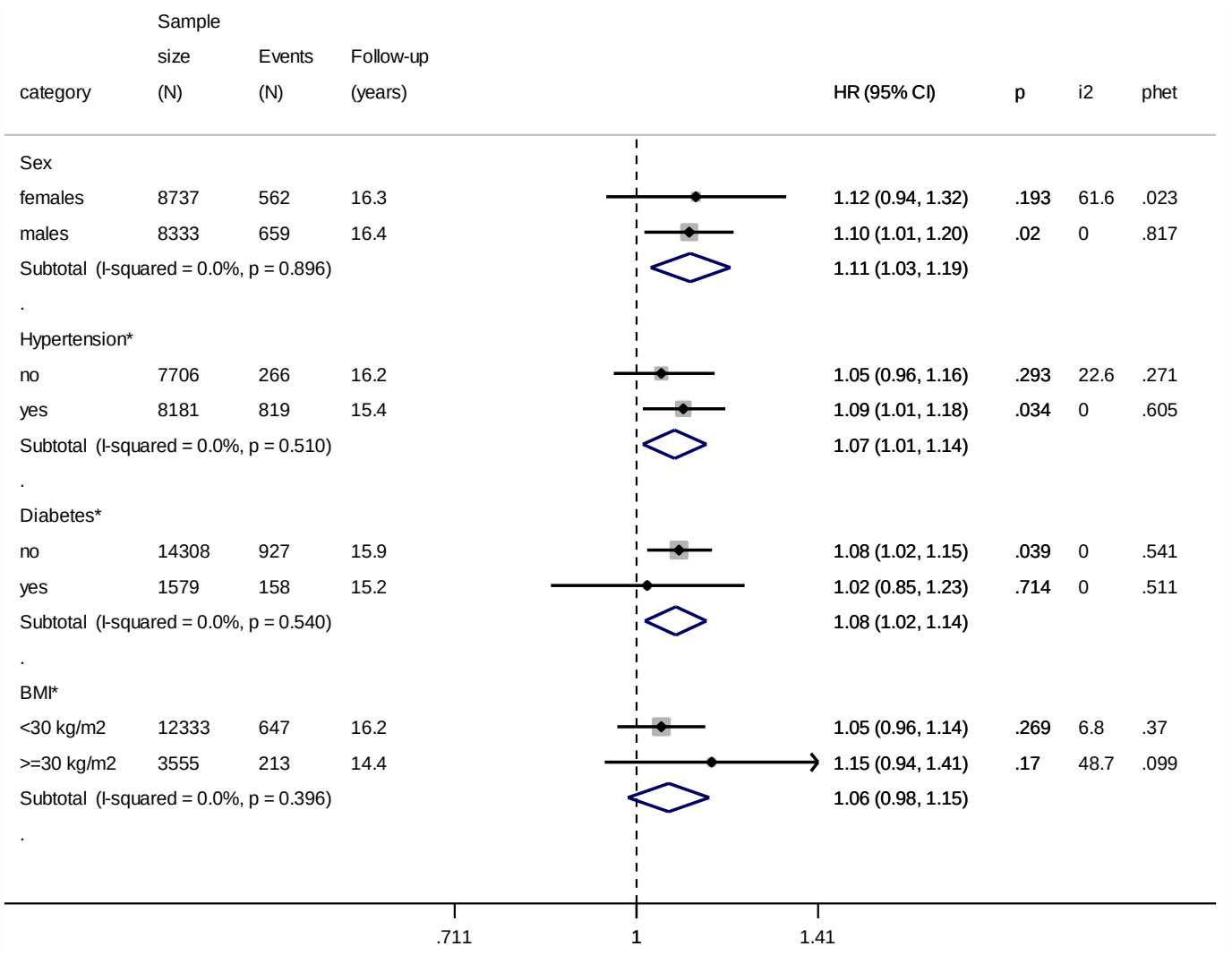
The results are derived from Cox proportional hazard models adjusted for age, sex, race, body mass index (1 kg/m2 increment), smoking (current vs. non-current), estimated glomerular filtration rate (1 mL/min/1.73 m² increment), history of coronary artery disease, diabetes mellitus, hypertension, hypercholesterolemia, atrial fibrillation, and heart failure at baseline.
Analyses for 1 SD increment correspond to ln-transformed MCP-1 levels.

Online Figure VIII. Pooled hazard ratios for incident hemorrhagic stroke per standard deviation increase in ln-transformed circulating MCP-1 levels and across MCP-1 level quartiles in sensitivity analyses omitting one study per time. Shown are the results from random-effects meta-analyses.



The results are derived from Cox proportional hazard models adjusted for age, sex, race, body mass index (1 kg/m2 increment), smoking (current vs. non-current), estimated glomerular filtration rate (1 mL/min/1.73 m² increment), history of coronary artery disease, diabetes mellitus, hypertension, hypercholesterolemia, atrial fibrillation, and heart failure at baseline.
Analyses for 1 SD increment correspond to ln-transformed MCP-1 levels.

Online Figure IX. Pooled hazard ratios for incident ischemic stroke per standard deviation increase in ln-transformed circulating MCP-1 levels, as derived from random-effects meta-analyses stratified by pre-defined study variables.



The p-values (p) correspond to the results of the random-effects meta-analyses and test statistical significance for the hazard ratios, whereas the p-values for heterogeneity (p-het) correspond to the Cochran Q test and test for statistical significance for the presence of heterogeneity in the respective meta-analysis. The results of heterogeneity between the pooled effects across the different variable categories are presented under the results for each variable.

The gray squares around the point estimates correspond to the weight of the included studies in the meta-analysis.

* ARIC has not been included in these analyses.

Online References

1. Hoogeveen RC, Morrison A, Boerwinkle E, Miles JS, Rhodes CE, Sharrett AR and Ballantyne CM. Plasma MCP-1 level and risk for peripheral arterial disease and incident coronary heart disease: Atherosclerosis Risk in Communities study. *Atherosclerosis*. 2005;183:301-7.
2. Rosamond WD, Folsom AR, Chambless LE, Wang CH, McGovern PG, Howard G, Copper LS and Shahar E. Stroke incidence and survival among middle-aged adults: 9-year follow-up of the Atherosclerosis Risk in Communities (ARIC) cohort. *Stroke*. 1999;30:736-43.
3. Deo R, Khera A, McGuire DK, Murphy SA, Meo Neto Jde P, Morrow DA and de Lemos JA. Association among plasma levels of monocyte chemoattractant protein-1, traditional cardiovascular risk factors, and subclinical atherosclerosis. *J Am Coll Cardiol*. 2004;44:1812-8.
4. Maroules CD, Rosero E, Ayers C, Peshock RM and Khera A. Abdominal aortic atherosclerosis at MR imaging is associated with cardiovascular events: the Dallas heart study. *Radiology*. 2013;269:84-91.
5. van Wijk DF, van Leuven SI, Sandhu MS, Tanck MW, Hutten BA, Wareham NJ, Kastelein JJ, Stroes ES, Khaw KT and Boekholdt SM. Chemokine ligand 2 genetic variants, serum monocyte chemoattractant protein-1 levels, and the risk of coronary artery disease. *Arterioscler Thromb Vasc Biol*. 2010;30:1460-6.
6. Sinha S, Myint PK, Luben RN and Khaw KT. Accuracy of death certification and hospital record linkage for identification of incident stroke. *BMC Med Res Methodol*. 2008;8:74.
7. Shoamanesh A, Preis SR, Beiser AS, Kase CS, Wolf PA, Vasan RS, Benjamin EJ, Seshadri S and Romero JR. Circulating biomarkers and incident ischemic stroke in the Framingham Offspring Study. *Neurology*. 2016;87:1206-11.
8. McDermott DH, Yang Q, Kathiresan S, Cupples LA, Massaro JM, Keaney JF, Jr., Larson MG, Vasan RS, Hirschhorn JN, O'Donnell CJ, Murphy PM and Benjamin EJ. CCL2 polymorphisms are associated with serum monocyte chemoattractant protein-1 levels and myocardial infarction in the Framingham Heart Study. *Circulation*. 2005;112:1113-20.
9. Carandang R, Seshadri S, Beiser A, Kelly-Hayes M, Kase CS, Kannel WB and Wolf PA. Trends in incidence, lifetime risk, severity, and 30-day mortality of stroke over the past 50 years. *JAMA*. 2006;296:2939-46.
10. Herder C, Baumert J, Thorand B, Martin S, Lowel H, Kolb H and Koenig W. Chemokines and incident coronary heart disease: results from the MONICA/KORA Augsburg case-cohort study, 1984-2002. *Arterioscler Thromb Vasc Biol*. 2006;26:2147-52.
11. Thiele I, Linseisen J, Heier M, Holle R, Kirchberger I, Peters A, Thorand B and Meisinger C. Time trends in stroke incidence and in prevalence of risk factors in Southern Germany, 1989 to 2008/09. *Sci Rep*. 2018;8:11981.
12. Schiopu A, Bengtsson E, Goncalves I, Nilsson J, Fredrikson GN and Bjorkbacka H. Associations Between Macrophage Colony-Stimulating Factor and Monocyte Chemotactic Protein 1 in Plasma and First-Time Coronary Events: A Nested Case-Control Study. *J Am Heart Assoc*. 2016;5.
13. Rosvall M, Janzon L, Berglund G, Engstrom G and Hedblad B. Incidence of stroke is related to carotid IMT even in the absence of plaque. *Atherosclerosis*. 2005;179:325-31.

Supplementary material online:

Interleukin-6 signaling effects on ischemic stroke and other cardiovascular outcomes: a Mendelian Randomization study. Georgakis *et al.*

Supplementary Methods. Secondary outcomes derived from the UK Biobank.

For some of the etiologically related outcomes, we examined the associations with the genetic variants in the IL6R gene that were used as instruments based on data from the UK Biobank. These included aortic aneurysm, peripheral artery disease, heart failure, deep vein thrombosis, and pulmonary embolism. To determine prevalent and incident cases for these phenotypes in the UK Biobank, we used diagnoses based on electronic health and hospital procedure codes, coded according to the 10th Edition of the International Classification of Diseases (ICD-10). Specifically, we extracted the ICD-10 codes from the fields "41270", "41202", and "41204" in the UK Biobank database. Aortic aneurysm was defined by the codes I71.1, I71.2, I71.3, and I71.4; peripheral artery disease by I73.8 and I73.9; heart failure by I11.0, I13.0, I13.2, and I.50; deep vein thrombosis by I80.2; and pulmonary embolism by I26. We then excluded related participants (pi-hat >0.0884 and participants of non-White-British descent and fit logistic regression models exploring the associations between the SNPs of interest and the respective outcomes. The models were further adjusted for age at baseline, sex, the first 10 principal components, and the genotyping chip for each SNP.

Table S1. Power calculations for the Mendelian randomization analyses performed in the current study.

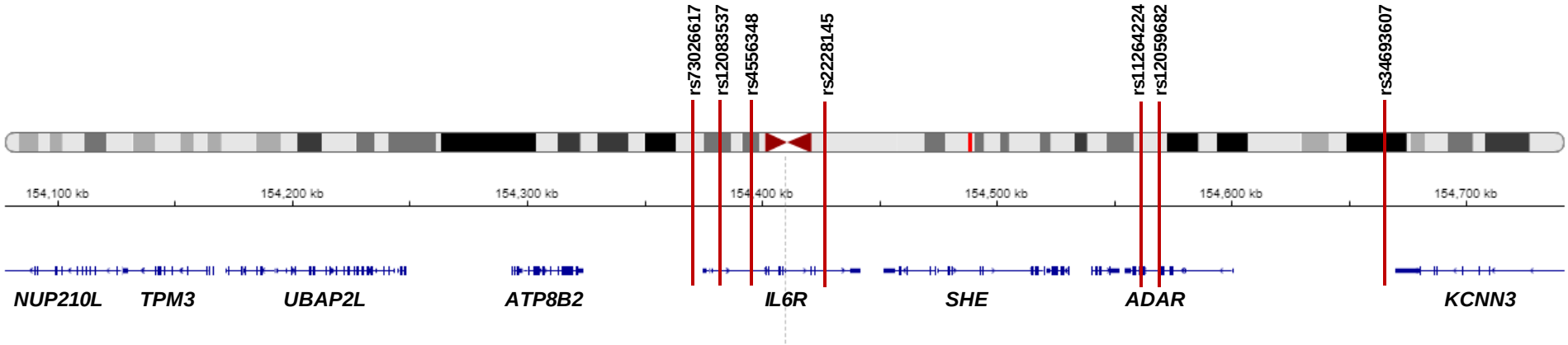
Phenotype	N total	% cases	Detectable OR* at 80% power
Coronary artery disease	184,305	33.0	≤0.93 or ≥1.09
Ischemic stroke	440,328	7.8	≤0.92 or ≥1.11
Large artery stroke	410,484	1.1	≤0.82 or ≥1.36
Cardioembolic stroke	413,304	1.7	≤0.84 or ≥1.28
Small vessel stroke	411,497	1.3	≤0.84 or ≥1.31
Myocardial infarction	167,180	26.1	≤0.92 or ≥1.1
Aortic aneurysm	316,142	0.6	≤0.75 or ≥1.92
Carotid plaque	48,434	44.5	≤0.86 or ≥1.16
Peripheral artery disease	317,717	1.3	≤0.81 or ≥1.41
Heart failure	321,406	2.8	≤0.86 or ≥1.23
Atrial fibrillation	129,831	14.2	≤0.89 or ≥1.16
Venous thromboembolism	60,139	12.5	≤0.84 or ≥1.27
Deep vein thrombosis	306,472	1.3	≤0.81 or ≥1.43
Pulmonary embolism	307,586	1.8	≤0.83 or ≥1.32

Power calculation were based on the online application "mRnd: Power calculations for Mendelian Randomization" (http://cnsgenomics.com/shiny/mRnd/).

R^2 was estimated using an additive model, after estimating the variance explained for each of the 7 variants from the associations estimates with CRP levels, according to the formula: [b] $R^2 = 2 \times MAF \times (1 - MAF) \times beta^2$, where MAF is the minimum allele frequency for the respective variant and beta the association estimate with circulating ln-CRP levels.

* Odds Ratios are scaled to the CRP-decreasing effects of tocilizumab (8 mg/kg).

Figure S1. The genomic region that was screened for identification of genetic instruments for IL-6 signaling and precise location of the identified single nucleotide polymorphisms (SNPs).



The region was selected as 300 kB upstream or downstream from the *IL6R* gene according to GRCh37/hg19 (chr1: 154,077,669-154,741,926). Gene name annotations are presented according to GENCODE (version 28) and accessed through the Integrative Genomics Viewer (https://igv.org/app/)[1].

Figure S2. Mendelian Randomization (MR) associations of genetically downregulated IL-6 signaling with ischemic stroke and coronary artery disease (positive control), as derived from fixed-effects inverse-variance-weighted (IVW), weighted median, contamination mixture (Con-Mix) and MR Pleiotropy Residual Sum and Outlier (MR-PRESSO) analyses.

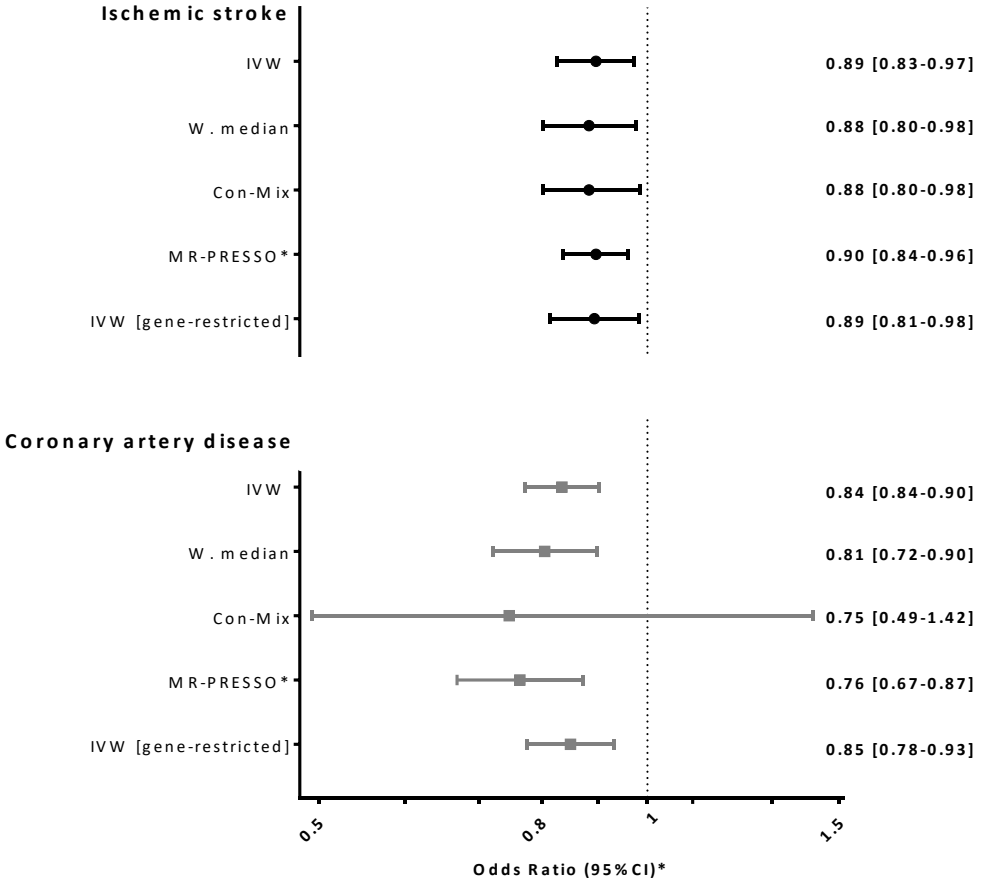
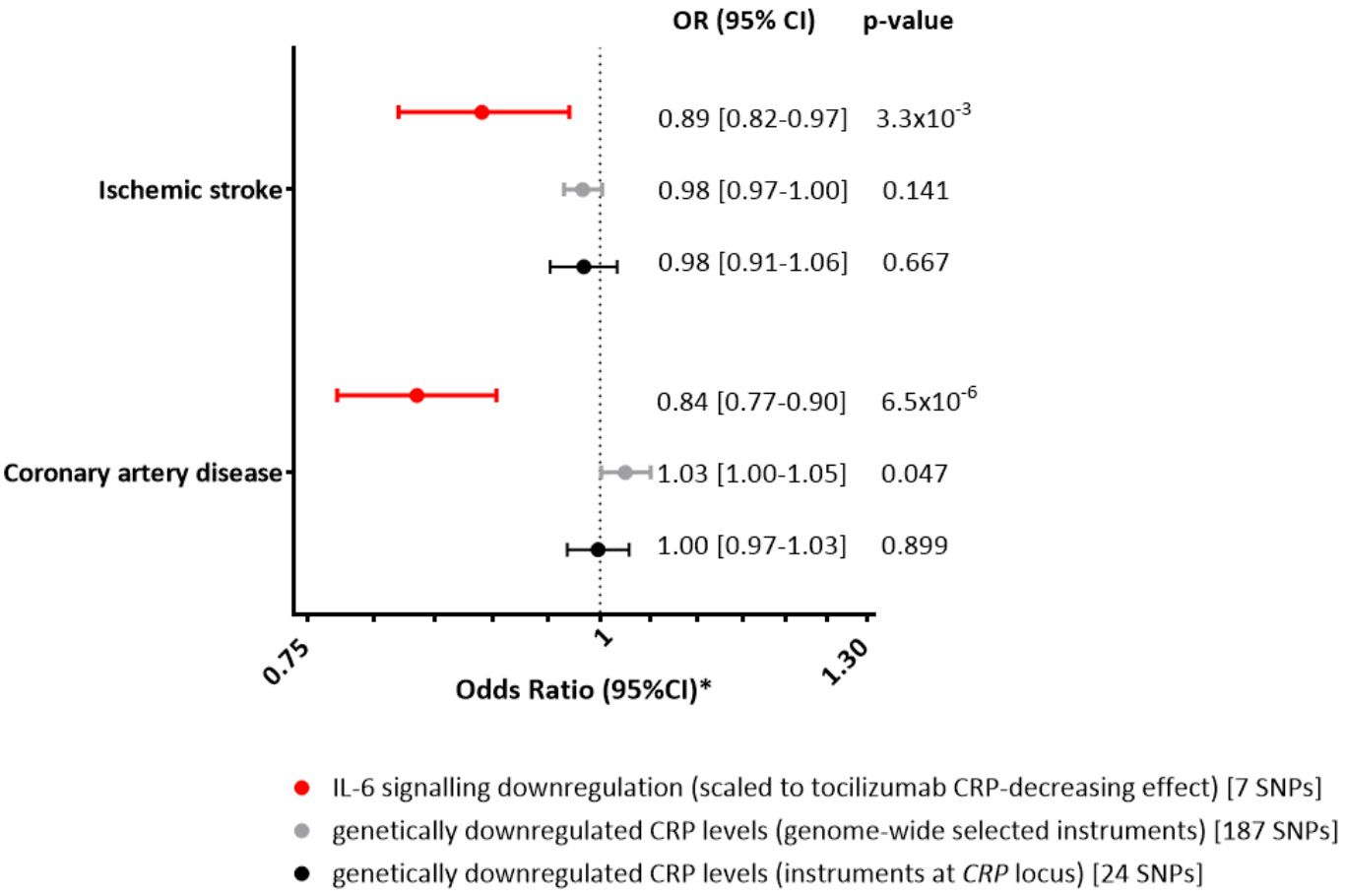
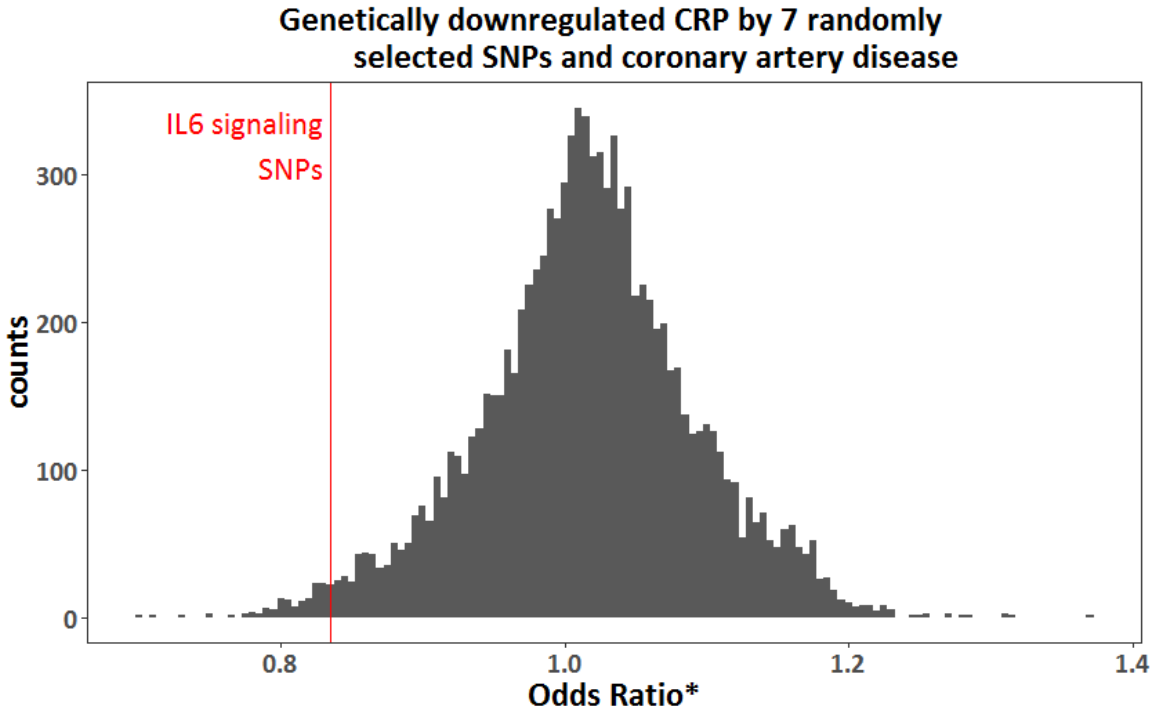


Figure S3. Mendelian Randomization associations of genetically downregulated IL-6 signaling and CRP levels with ischemic stroke and coronary artery disease. The effects represent Odds Ratios (OR) derived from inverse-variance-weighted MR analyses.



* Odds Ratios are scaled to the CRP-decreasing effects of tocilizumab (8 mg/kg).

Figure S4. Distribution of the effects of randomly selected sets of 7 CRP-decreasing SNPs on risk of coronary artery disease. The red line represents the effect of the 7 SNPs used as instruments for IL-6 signaling downregulation in the current study.



* Odds Ratios are scaled to the CRP-decreasing effects of tocilizumab (8 mg/kg).

Figure S5. Mendelian Randomization (MR) associations of genetically downregulated IL-6 signaling with large artery stroke, cardioembolic stroke, and small vessel stroke, as derived from fixed-effects inverse-variance-weighted (IVW), weighted median, contamination mixture (Con-Mix) and MR Pleiotropy Residual Sum and Outlier (MR-PRESSO) analyses.

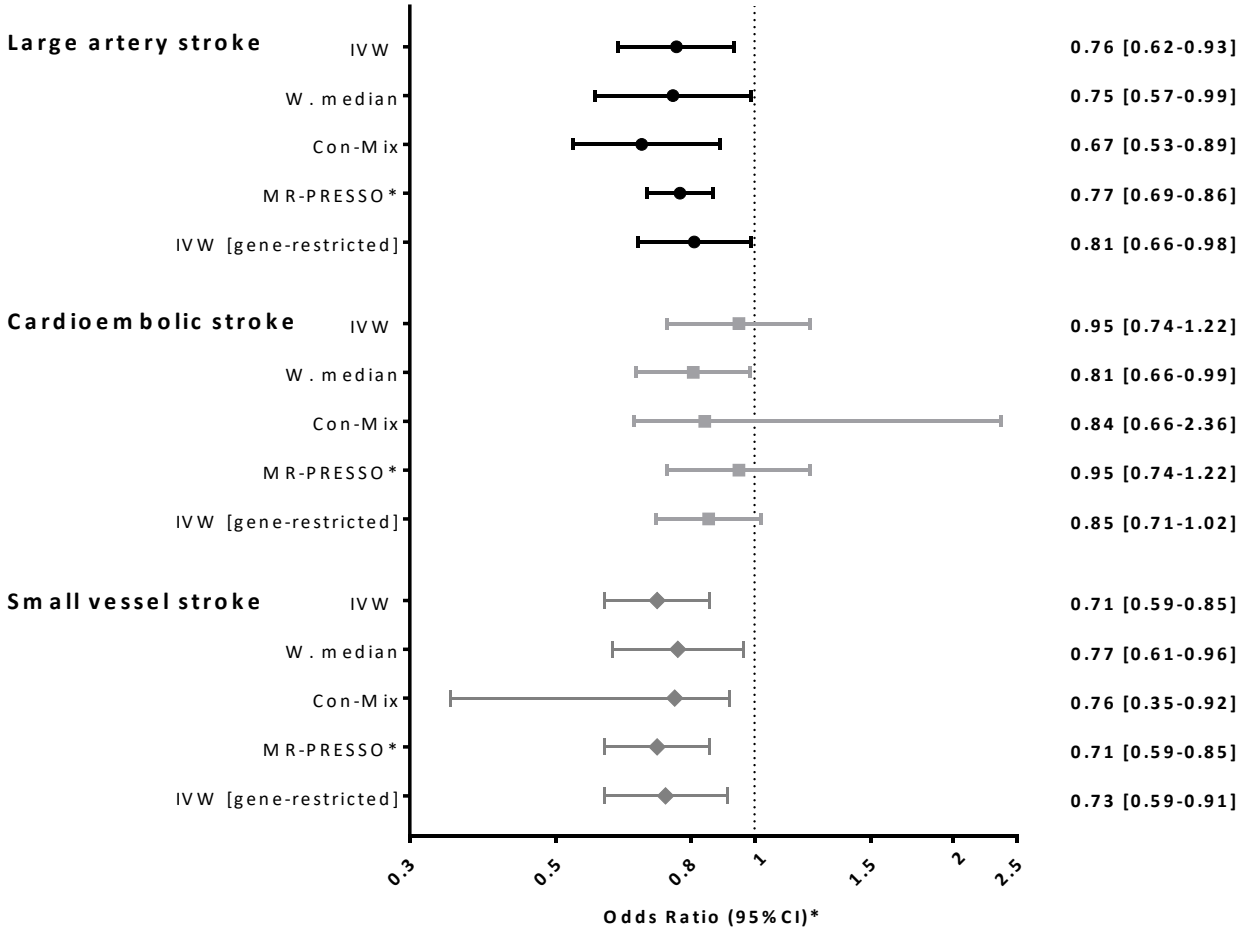
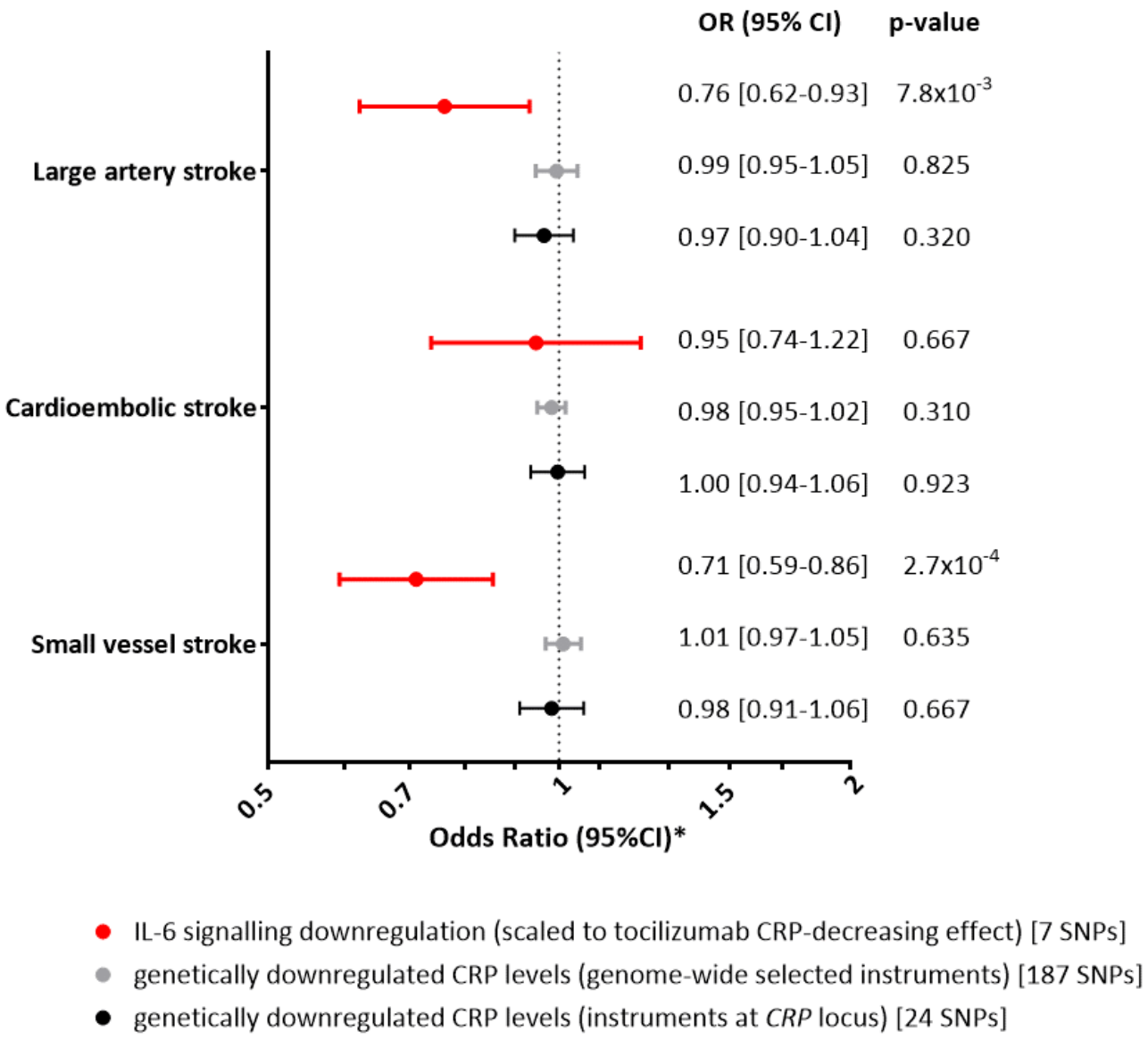


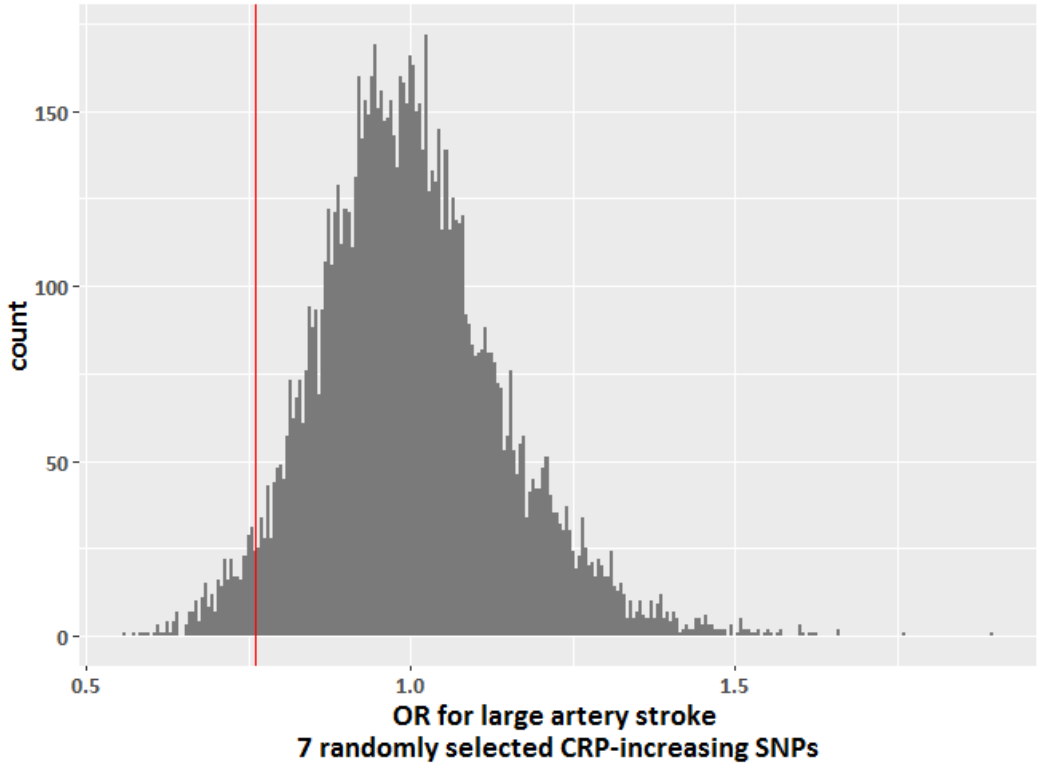
Figure S6. Mendelian Randomization associations of genetically downregulated IL-6 signaling and CRP levels with ischemic stroke subtypes. The effects represent Odds Ratios (OR) derived from inverse-variance-weighted MR analyses.



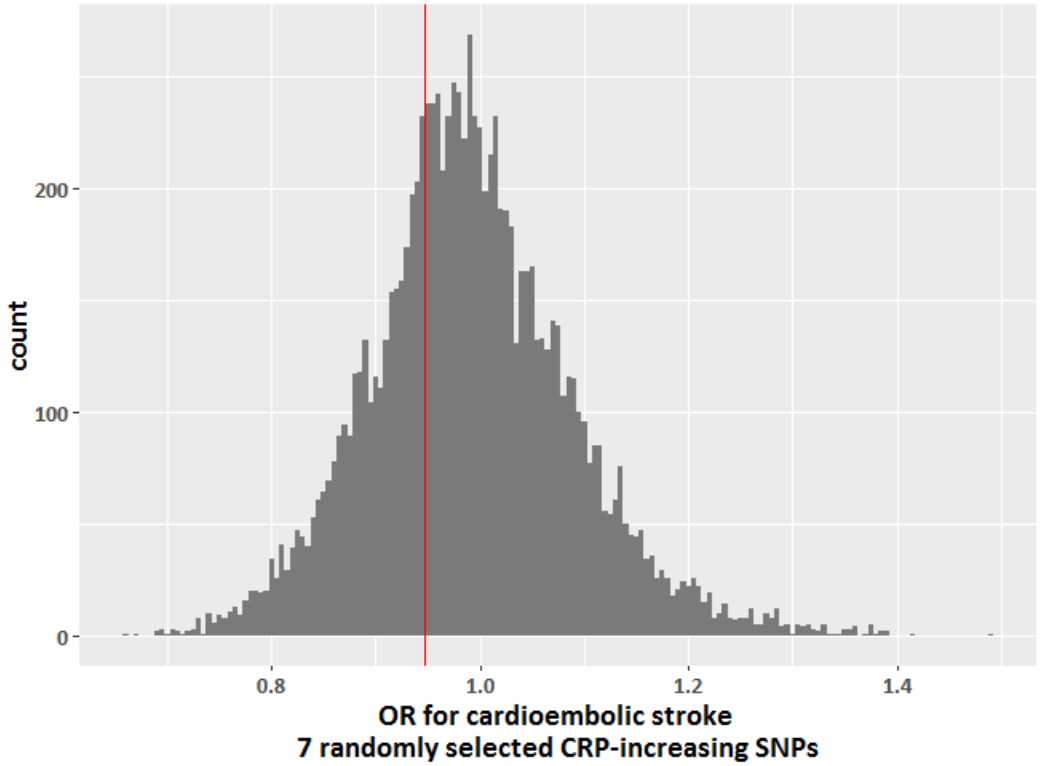
* Odds Ratios are scaled to the CRP-decreasing effects of tocilizumab (8 mg/kg).

Figure S7. Distribution of the effects of randomly selected sets of 7 CRP-decreasing SNPs on risk of (A) large artery, (B) cardioembolic, and (C) small vessel stroke. The red lines represent the effects of the 7 SNPs used as instruments for IL-6 signaling downregulation in the current study.

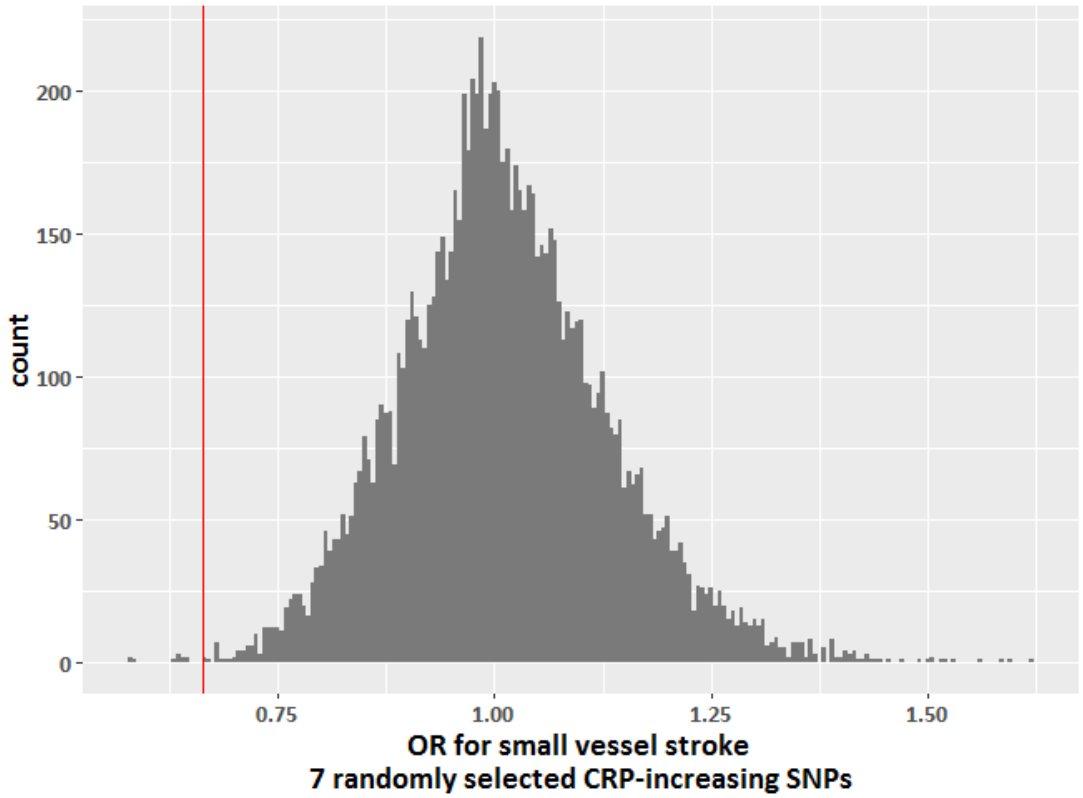
A



B



C



* Odds Ratios scaled to the CRP-decreasing effects of tocilizumab (8 mg/kg).

Supplementary References

1. Robinson JT, Thorvaldsdottir H, Winckler W, Guttman M, Lander ES, Getz G, Mesirov JP. Integrative genomics viewer. Nat Biotechnol 2011;**29**(1):24-6.

Consortia in the author list

The members of the **CHARGE Inflammation Working Group** are: Emelia Benjamin, Daniel I. Chasman, Abbas Dehghan, Tarunveer Singh Ahluwalia, James Meigs, Russell Tracy, Behrooz Z. Alizadeh, Symen Ligthart, Josh Bis, Gudny Eiriksdottir, Nathan Pankratz, Myron Gross, Alex Rainer, Harold Snieder, James G. Wilson, Bruce M. Psaty, Josee Dupuis, Bram Prins, Urmo Vaso, Maria Stathopoulou, Lude Franke, Terho Lehtimaki, Wolfgang Koenig, Yalda Jamshidi, Sophie Siest, Ali Abbasi, Andre G. Uitterlinden, Mohammadreza Abdollahi, Renate Schnabel, Ursula M. Schick, Ilja M. Nolte, Aldi Kraja, Yi-Hsiang Hsu, Daniel S. Tylee, Alyson Zwicker, Rudolf Uher, George Davey-Smith, Alanna C. Morrison, Andrew Hicks, Cornelia M. van Duijn, Cavin Ward-Caviness, Eric Boerwinkle, J. Rotter, Ken Rice, Leslie Lange, Markus Perola, Eco de Geus, Andrew P. Morris, Kari Matti Makela, David Stacey, Johan Eriksson, Tim M. Frayling, and Eline P. Slagboom.

The members of the **INVENT Consortium** are: Philippe Amouyel, Mariza de Andrade, Saonli Basu, Claudine Berr, Jennifer A Brody, Daniel I Chasman, Jean -Francois Dartigues, Aaron R Folsom, Marine Germain, Hugoline de Haan, John Heit, Jeanine Houwing-Duitermaat, Christopher Kabrhel, Peter Kraft, Grégoire Legal, Sara Lindström, Ramin Monajemi, Pierre-Emmanuel Morange, Bruce M Psaty, Pieter H Reitsma, Paul M Ridker, Lynda M Rose, Frits R Rosendaal, Noémie Saut, Eline Slagboom, David Smadja, Nicholas L Smith, Pierre Suchon, Weihong Tang, Kent D Taylor, David-Alexandre Trégouët, Christophe Tzourio, Marieke CH de Visser, Astrid van Hylckama Vlieg, Lu-Chen Weng, Kerri L Wiggins.

Supplemental Text

Genetic variants related to antihypertensive targets inform drug efficacy and side effects

Gill *et al*. Genetic variants inform drug effects

Dipender Gill MD*[1], Marios K. Georgakis MD*[2-4], Fotios Koskeridis MSc[5], Lan Jiang MS[6], Qiping Feng PhD[6], Wei-Qi Wei MD PhD[7], Evropi Theodoratou PhD[8], Paul Elliott FMedSci[1,9-12], Joshua C. Denny MD MS[7], Rainer Malik PhD[2-4], Evangelos Evangelou PhD[1,5], Abbas Dehghan MD PhD[1,9,11], Martin Dichgans MD†[2-4], Ioanna Tzoulaki PhD†[1,5,9,11]

*Contributed equally

†Contributed equally

I. Supplemental Methods

Mendelian randomization (MR)

In the main MR analysis, estimates for each single-nucleotide polymorphism (SNP) were derived using the Wald ratio method, with standard errors estimated using second order weights to allow for measurement error in both the exposure and outcome estimates (1). For drug targets with more than one related SNP, overall MR estimates were calculated by pooling individual MR estimates for each SNP using fixed-effects inverse-variance weighted (IVW) meta-analysis (1), and were scaled to the estimated effect of the corresponding drug target on systolic blood pressure (SBP) in randomized controlled trials (RCTs) (2), in order to reflect drug effect. After conversion of odds ratio estimates to relative risk (RR) using baseline incidences of CHD and stroke of 0.042 and 0.041 respectively from a systematic review of 613,815 participants enrolled in blood pressure lowering trials (3), MR results were compared with estimates from a recent Cochrane systematic review and meta-analysis of RCTs that investigated the effect of common antihypertensive drugs against placebo (2). Sensitivity analyses were also performed using MR RR estimates derived from baseline CHD and stroke incidences of 1%, 5% and 10%.

Investigation of pleiotropy

Heterogeneity in the MR estimates generated by different SNPs can be used to indicate such pleiotropy (4), which was identified through a significant Cochran's Q test (P<0.05) or an I^2 measure of heterogeneity >30%. MR statistical sensitivity analyses that are more robust to the inclusion of pleiotropic variants were also performed. Firstly, the weighted median estimator was used, which obtains an overall MR estimate by ordering individual SNP MR estimates by their magnitude weighted for their precision, and is reliable when more than half the information for the analysis comes from valid instruments (5). Secondly, the MR-Egger technique was performed, which regresses the SNP-outcome estimates against the SNP-exposure estimates, weighted for the

precision of the SNP-outcome estimates to give a reliable MR estimate and test for the presence of directional pleiotropy in scenarios where any pleiotropic effect of the genetic variants is independent of their association with the exposure (6). Finally, MR-PRESSO was conducted, which performs a zero-intercept regression of the SNP-outcome estimates against the SNP-exposure estimates to test, using residual errors, whether there are outlier SNPs (P<0.05), and whether removing these changes the MR estimates generated (7). MR-PRESSO generally requires that at least half of the genetic variants used do not relate to the outcome independently of the exposure (7). Statistical sensitivity analyses in MR suffer from low power (4), and as such no formal statistical significance threshold was set for these.

II. Supplemental Results

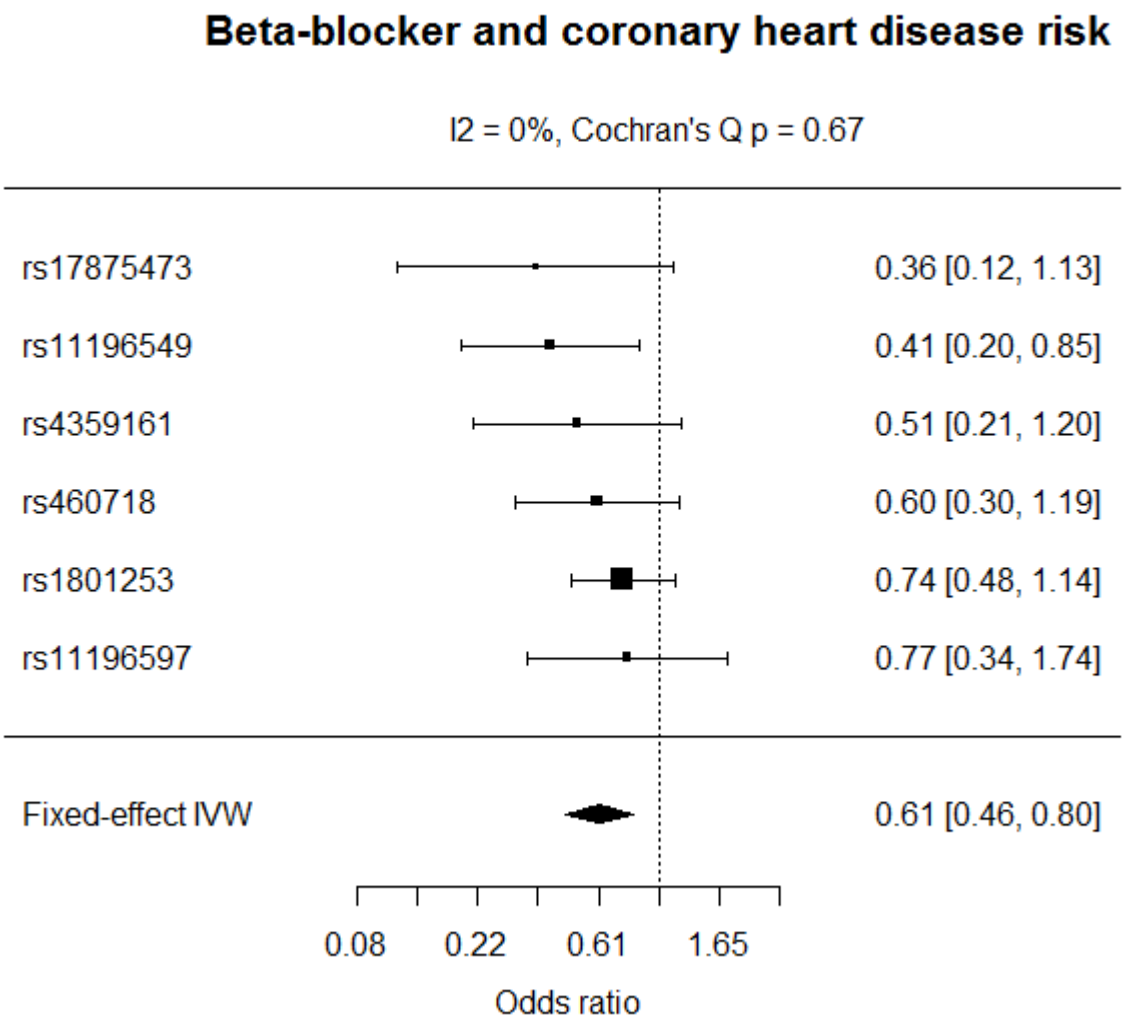
Mendelian randomization

The main variants used to proxy drug class effect were based on genetic association estimates that corrected for antihypertensive medication use and adjusted for body mass index (BMI) (8). To avoid possible bias related to medication non-compliance or introduction of collider effects respectively, sensitivity analyses were performed using the UK Biobank SBP GWAS that did not correct for medication use or adjust for BMI (9). No suitable variants were identified for ACEI, two SNPs were identified as variants for BB (Supplementary Table 7), and six SNPs as variants for CCB (Supplementary Table 8). IVW MR produced estimates that were comparable to the main analysis, but with wider confidence intervals (Supplementary Figures 5-8). Searching PhenoScanner (10), possible pleiotropic effects were identified for one BB SNP and five CCB SNPs (details are provided in Supplementary Table 9). Repeating the IVW MR analysis after excluding these SNPs also produced similar estimates to the main analysis (Supplementary Figures 5-8).

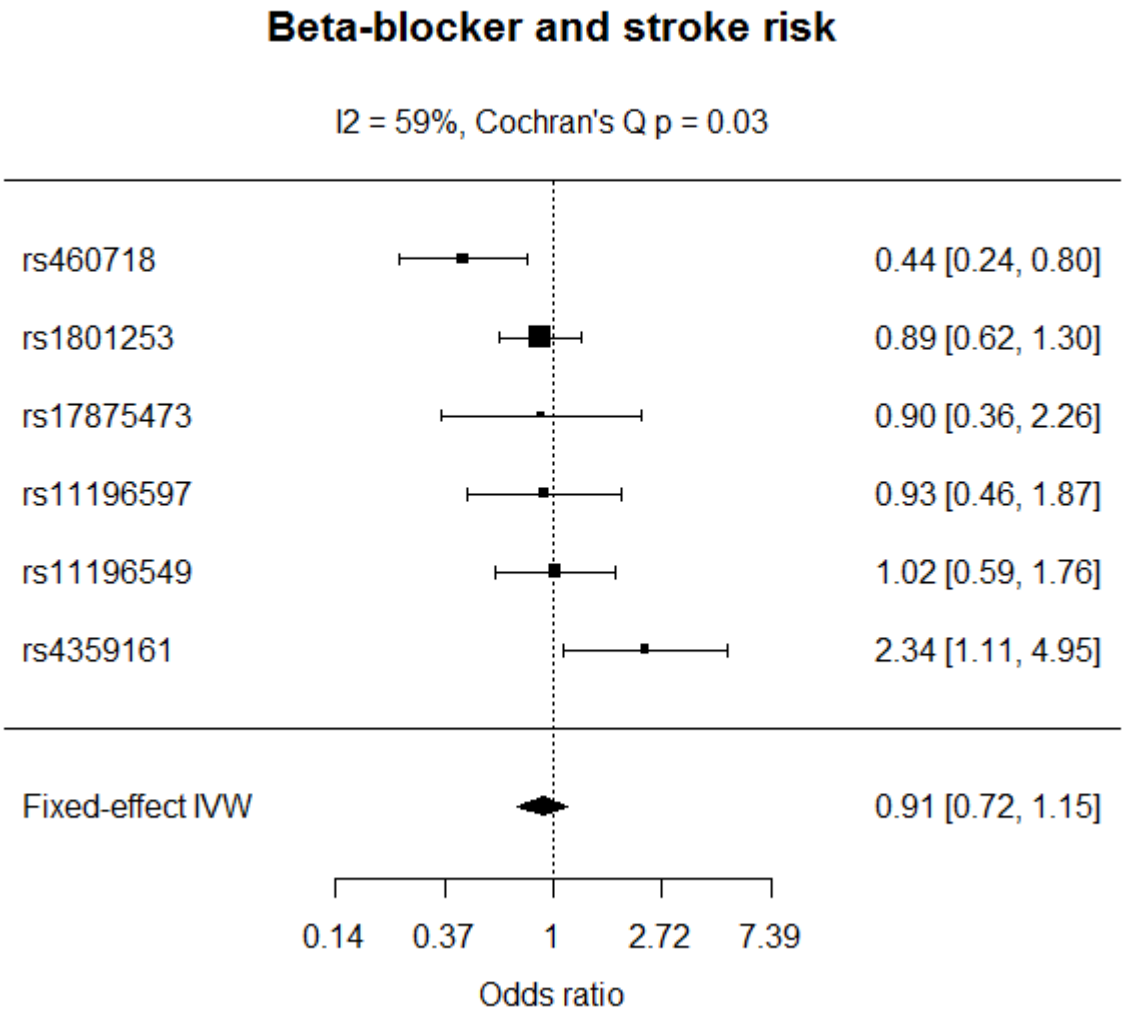
There was only evidence of heterogeneity, suggesting possible bias related to pleiotropic SNPs, in the MR analysis of BBs on stroke risk (I^2 59%, Cochran's Q *P*=0.03). The MR-Egger intercept was not significant for directional pleiotropy for either BBs (CHD *P*=0.87 and stroke *P*=0.89) or CCBs (CHD *P*=0.89 and stroke *P*=0.51). MR-PRESSO only detected outlier SNPs in the analysis of BBs on stroke risk (2 outliers), with MR-PRESSO estimates that excluded these SNPs consistent with the main analysis results (Supplementary Figure 6). Estimates using MR-Egger regression, the weighted median approach and MR-PRESSO also produced similar estimates to the main IVW MR analyses (Supplementary Figures 5-8).

III. Supplemental Figures

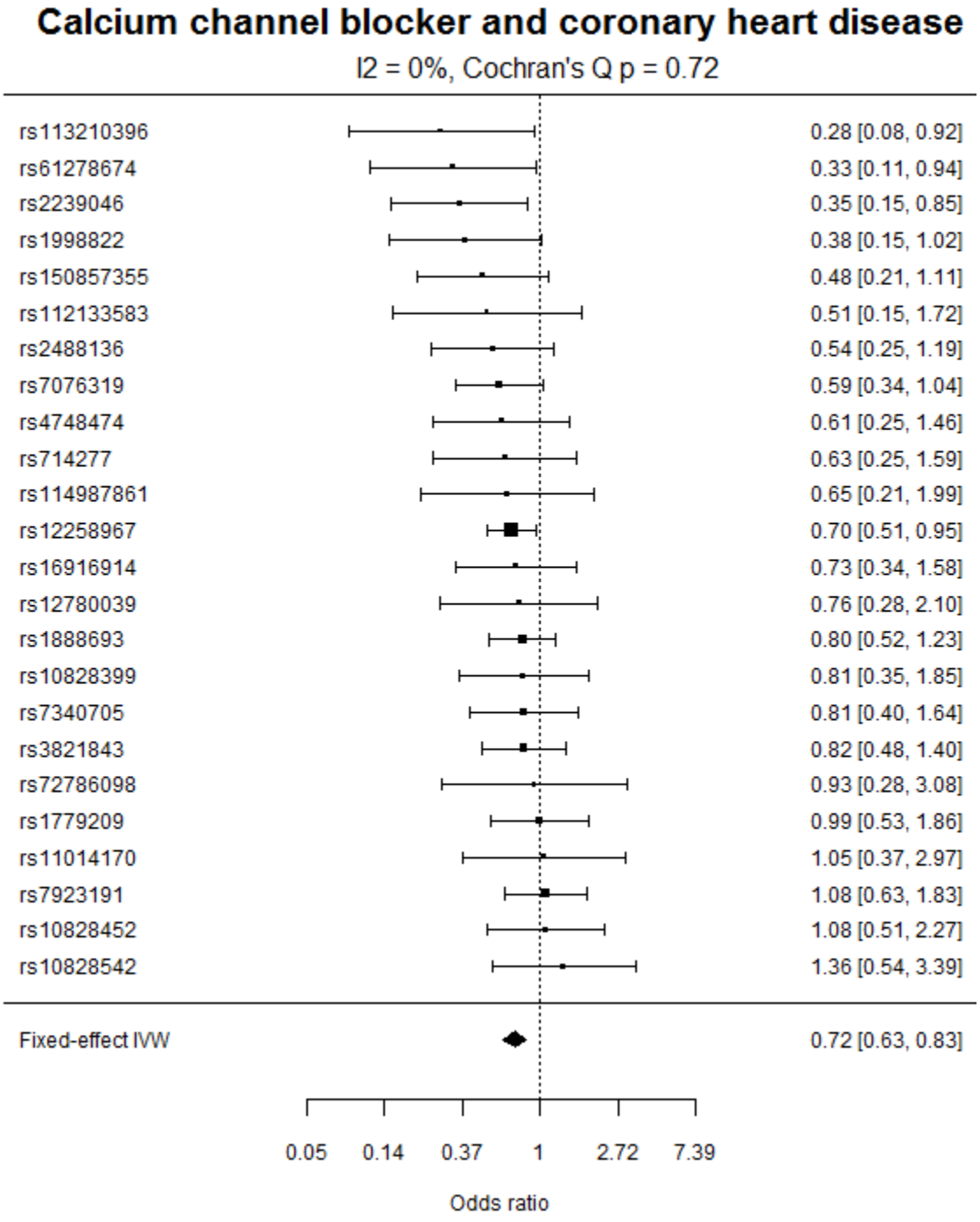
Supplementary Figure 1. Individual ratio method MR estimates for the analysis of BBs and CHD risk.



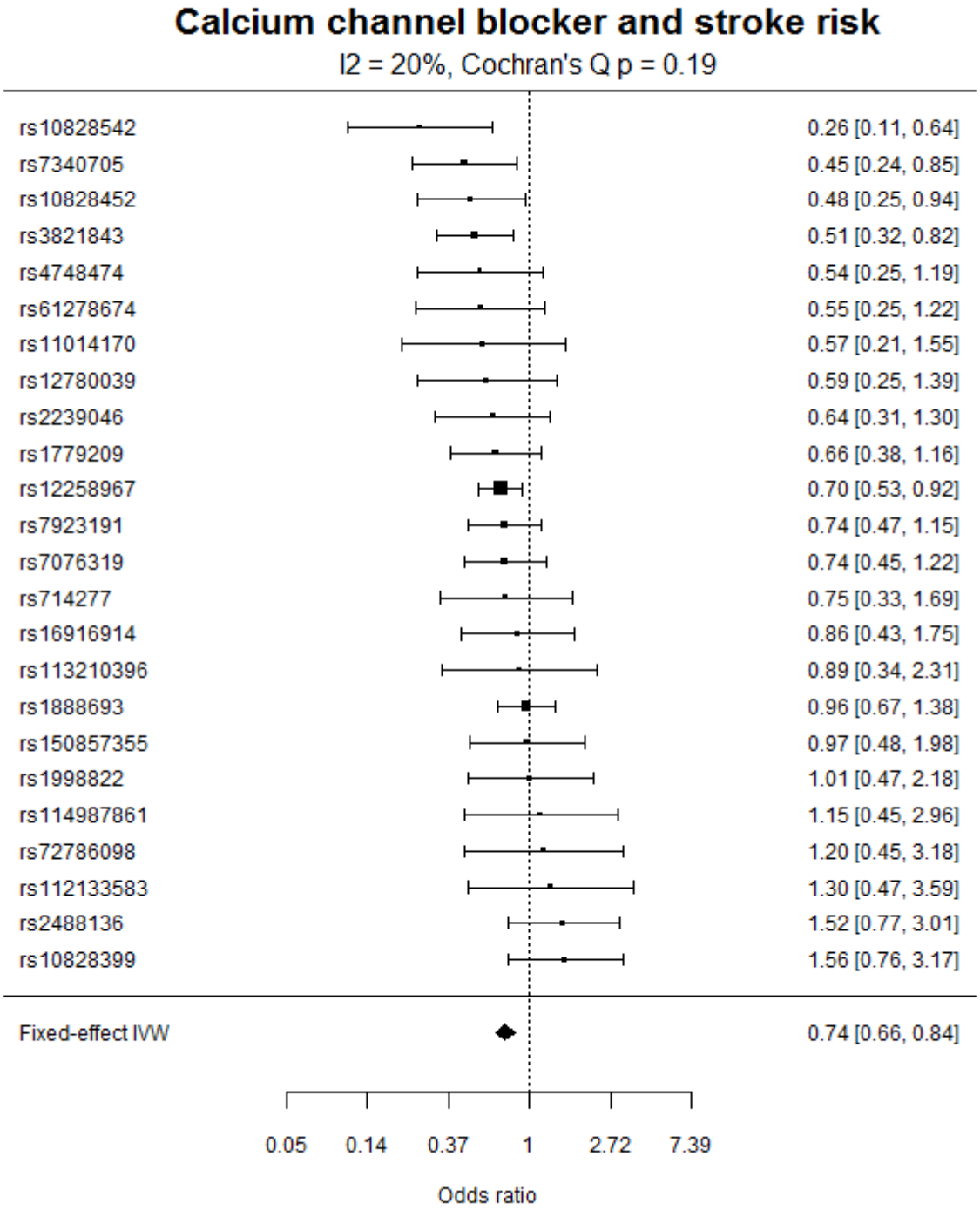
Supplementary Figure 2. Individual ratio method MR estimates for the analysis of BBs and stroke risk.



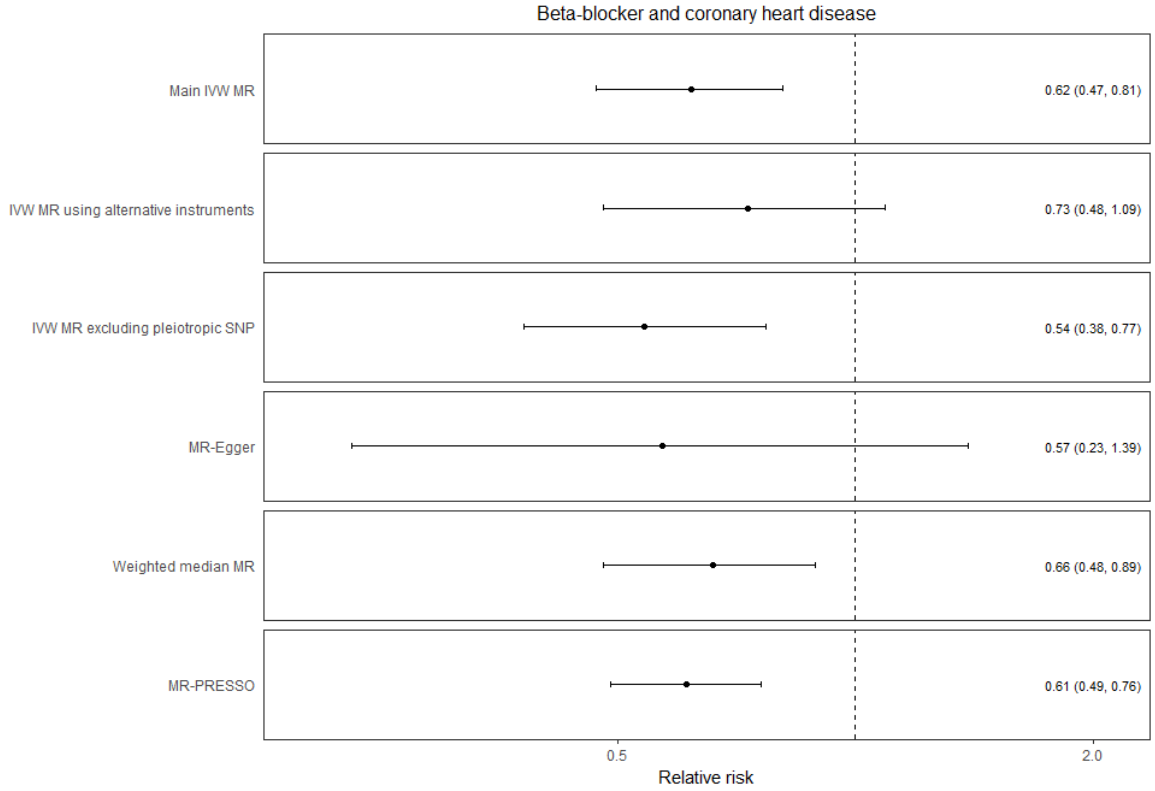
Supplementary Figure 3. Individual ratio method MR estimates for the analysis of CCBs and CHD risk.



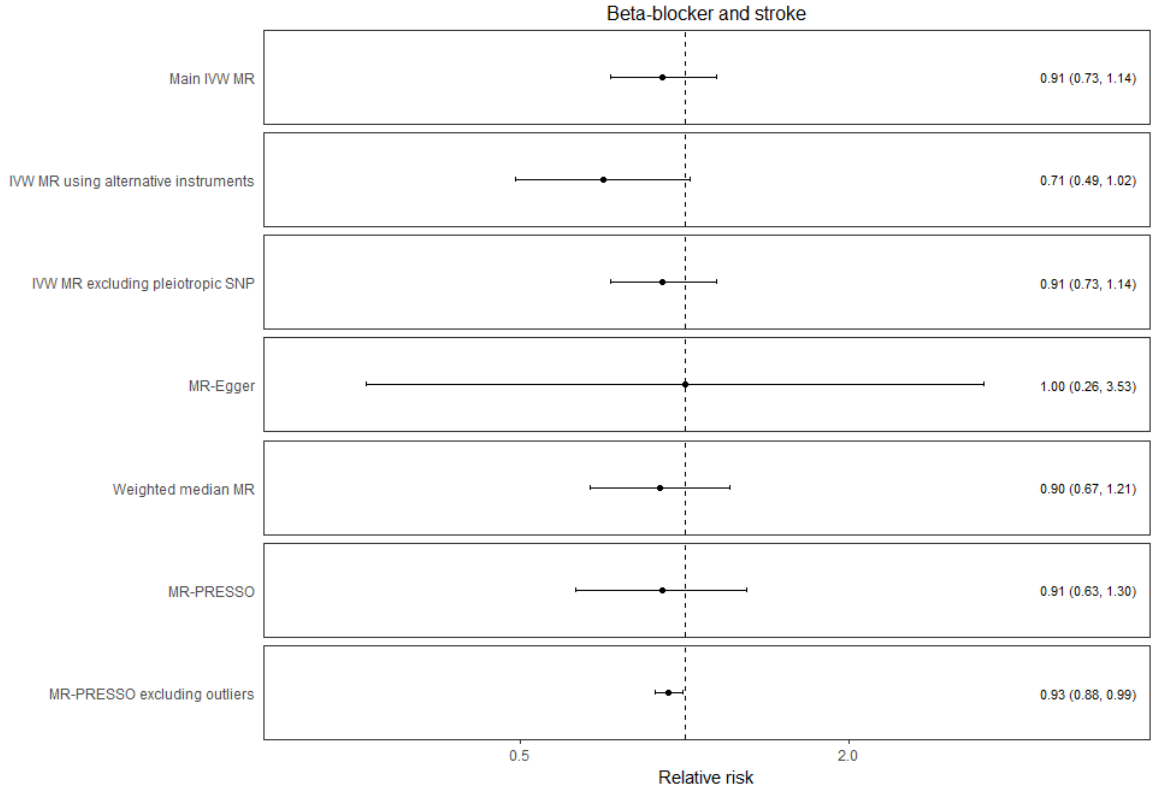
Supplementary Figure 4. Individual ratio method MR estimates for the analysis of CCBs and stroke risk.



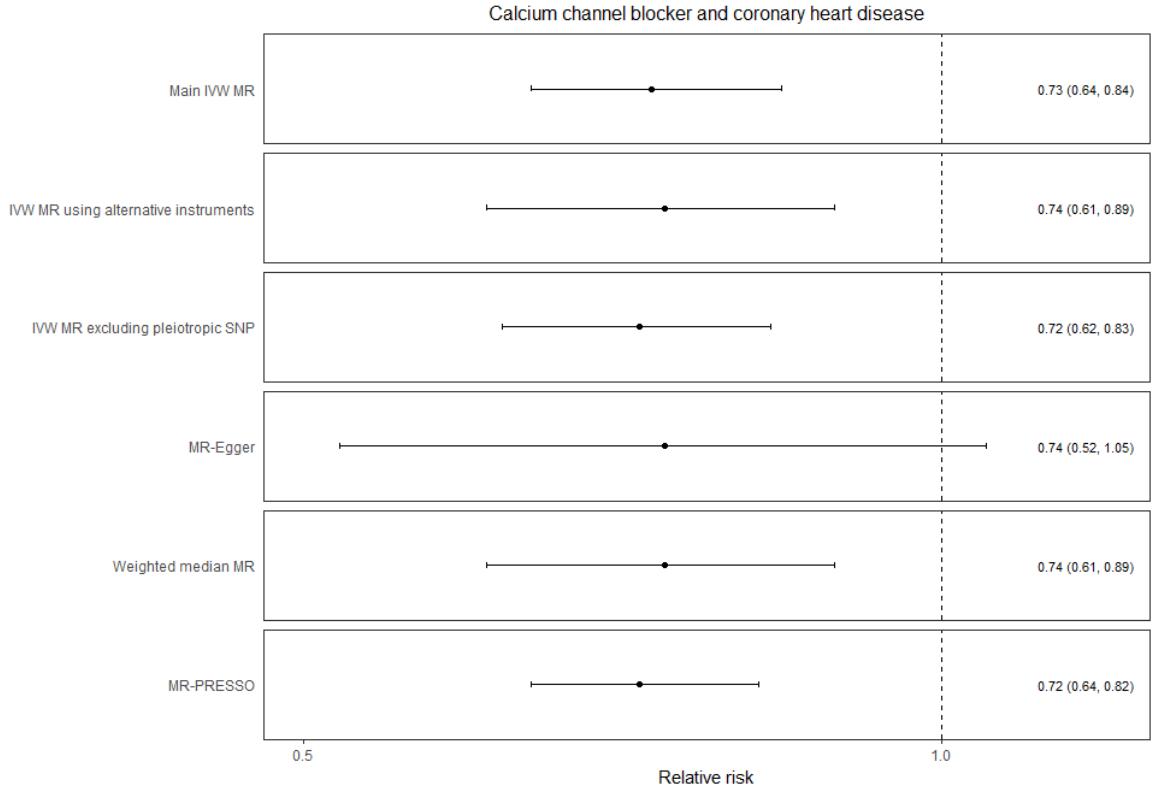
Supplementary Figure 5. MR sensitivity analyses for the analysis of BBs and CHD risk



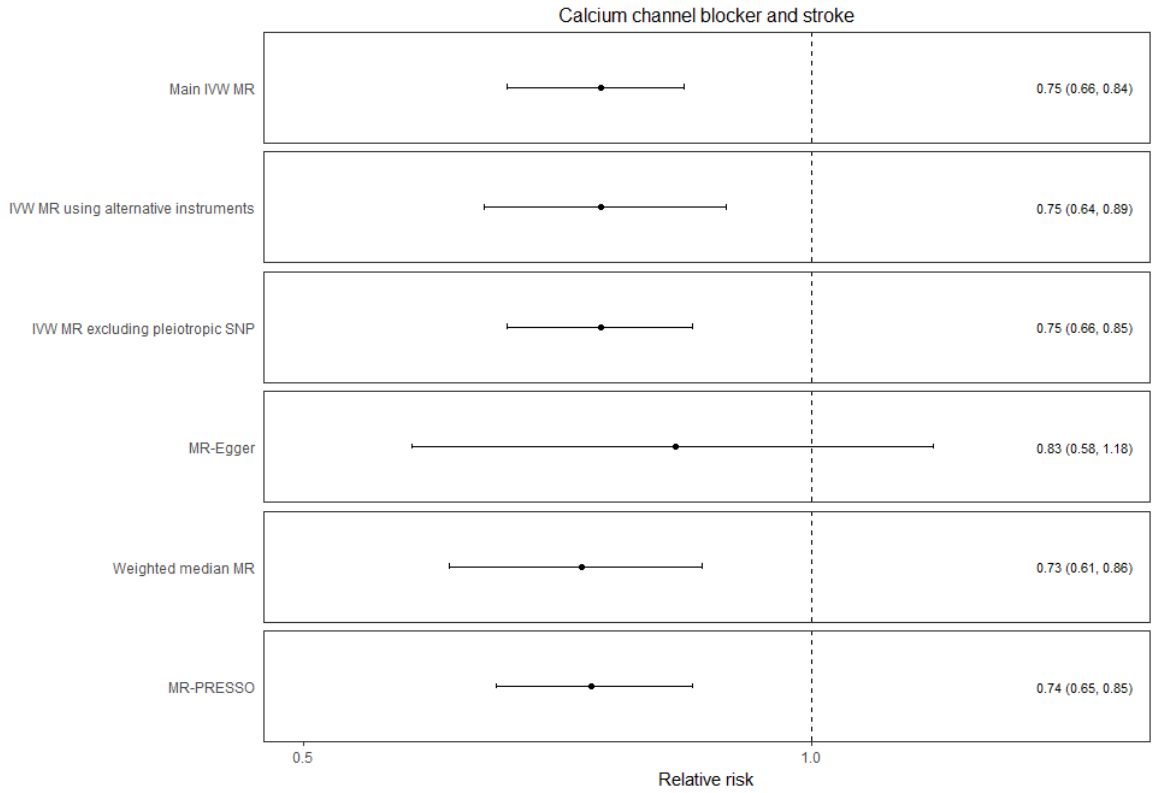
Supplementary Figure 6. MR sensitivity analyses for the analysis of BBs and stroke risk



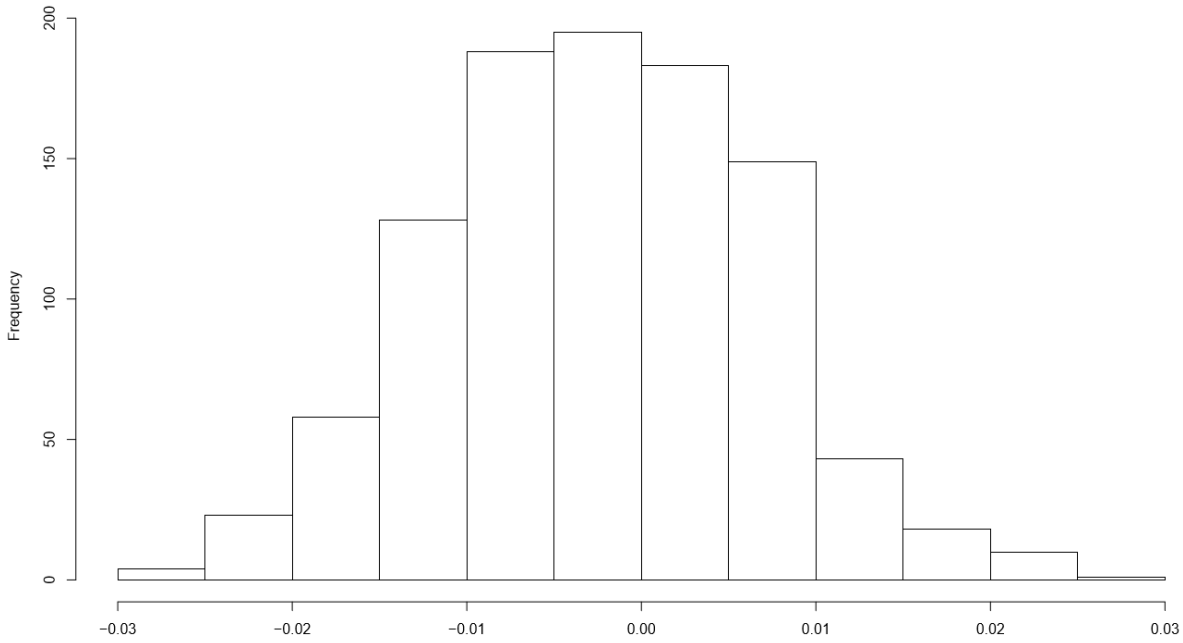
Supplementary Figure 7. MR sensitivity analyses for the analysis of CCBs and CHD risk



Supplementary Figure 8. MR sensitivity analyses for the analysis of CCBs and stroke risk



Supplementary Figure 9. Permutation analysis randomly sampling 24 SBP SNPs and investigating association with diverticulosis risk 1000 times



IV. Supplemental References

1. Burgess S, Butterworth A, Thompson SG. Mendelian randomization analysis with multiple genetic variants using summarized data. Genet Epidemiol. 2013;37(7):658-65.

2. Wright JM, Musini VM, Gill R. First-line drugs for hypertension. Cochrane Database Syst Rev. 2018;4:CD001841.

3. Ettehad D, Emdin CA, Kiran A, Anderson SG, Callender T, Emberson J, et al. Blood pressure lowering for prevention of cardiovascular disease and death: a systematic review and meta-analysis. Lancet. 2016;387(10022):957-67.

4. Burgess S, Bowden J, Fall T, Ingelsson E, Thompson SG. Sensitivity Analyses for Robust Causal Inference from Mendelian Randomization Analyses with Multiple Genetic Variants. Epidemiology. 2017;28(1):30-42.

5. Bowden J, Davey Smith G, Haycock PC, Burgess S. Consistent Estimation in Mendelian Randomization with Some Invalid Instruments Using a Weighted Median Estimator. Genet Epidemiol. 2016;40(4):304-14.

6. Bowden J, Davey Smith G, Burgess S. Mendelian randomization with invalid instruments: effect estimation and bias detection through Egger regression. Int J Epidemiol. 2015;44(2):512-25.

7. Verbanck M, Chen CY, Neale B, Do R. Detection of widespread horizontal pleiotropy in causal relationships inferred from Mendelian randomization between complex traits and diseases. Nat Genet. 2018;50(5):693-8.

8. Evangelou E, Warren HR, Mosen-Ansorena D, Mifsud B, Pazoki R, Gao H, et al. Genetic analysis of over 1 million people identifies 535 new loci associated with blood pressure traits. Nat Genet. 2018.

9. Neale Lab. 2019 January 16. Rapid GWAS of thousands of phenotypes for 337,000 samples in the UK Biobank 2018. http://www.nealelab.is/blog/2017/7/19/rapid-gwas-of-thousands-of-phenotypes-for-337000-samples-in-the-uk-biobank.

10. Staley JR, Blackshaw J, Kamat MA, Ellis S, Surendran P, Sun BB, et al. PhenoScanner: a database of human genotype-phenotype associations. Bioinformatics. 2016;32(20):3207-9.

V. Contributors

DG, IT, MKG and MD designed the study. DG, MKG, FK and LJ collectively had full access to the data and performed the analysis. All authors interpreted the results. DG and IT drafted the manuscript. All authors critically revised the manuscript for intellectual content. All authors approved the submitted version and are accountable for the integrity of the work.

VI. Acknowledgements

The authors acknowledge the contributors of the data used in this work: CARDIoGRAMplusC4D, International Consortium for Blood Pressure, MEGASTROKE, UK Biobank and Vanderbilt University Biobank. Links to the various web resources used are provided below.

CARDIoGRAMplusC4D: http://www.cardiogramplusc4d.org/

DrugBank: https://www.drugbank.ca/

GeneCards: https://www.genecards.org/

MEGATSROKE GWAS meta-analysis summary data: http://www.megastroke.org/

Neale Labe UK Biobank GWAS summary data: http://www.nealelab.is/blog/2017/7/19/rapid-gwas-of-thousands-of-phenotypes-for-337000-samples-in-the-uk-biobank

PhenoScanner: www.phenoscanner.medschl.cam.ac.uk/phenoscanner

UK Biobank: http://www.ukbiobank.ac.uk/

Vanderbilt University Biobank: https://victr.vanderbilt.edu/pub/biovu/

VII. Sources of Funding

DG is funded by the Wellcome 4i Clinical PhD Programme at Imperial College London. MG is funded by scholarships from the Deutscher Akademischer Austauschdienst and the Onassis Foundation. QF, W-QW and JCD are funded by National Institute of Health grants R01 HL133786, R01 GM120523, R01 LM010685. ET acknowledges support from Cancer Research UK (C31250/A22804). PE acknowledges support from the Medical Research Council and Public Health England (MR/L01341X/1) for the Medical Research Council-Public Health England Centre for Environment and Health; the National Institute for Health Research Imperial Biomedical Research Centre in collaboration with Imperial College NHS Healthcare Trust. PE is supported by the UK Dementia Research Institute, which receives funding from UK DRI Ltd funded by the UK Medical Research Council, Alzheimer's Society and Alzheimer's Research UK. PE is associate director of the Health Data Research UK London funded by a consortium led by the UK Medical Research Council. MD acknowledges funding from the European Union's Horizon 2020 research and innovation programme under grant agreements No 666881, SVDs@target and No 667375, CoSTREAM; the DFG as part of the Munich Cluster for Systems Neurology (EXC 1010 SyNergy) and the CRC 1123 (B3); the Corona Foundation; the Fondation Leducq (Transatlantic Network of Excellence on the Pathogenesis of Small Vessel Disease of the Brain); the e:Med program (e:AtheroSysMed) and the FP7/2007-2103 European Union project CVgenes@target (grant agreement number Health-F2-2013-601456). The MEGASTROKE project received funding from sources specified at http://www.megastroke.org/acknowledgments.html. Details of all MEGASTROKE authors are available at http://www.megastroke.org/authors.html.

VIII. Disclosures

All authors have no conflicts of interest to declare.

Supplementary Online Material

Georgakis MK, Gill D, Webb AJS, et al. Genetically determined blood pressure, antihypertensive drug classes and risk of stroke subtypes: a Mendelian Randomization Study.

eMethods

Table e-1. Genome-wide significant ($p<5x10^{-8}$) and independent ($r^2<0.001$) single nucleotide polymorphisms (SNP) that were used as instruments for systolic blood pressure (SBP).

Table e-2. Genome-wide significant ($p<5x10^{-8}$) and independent ($r^2<0.001$) single nucleotide polymorphisms (SNP) that were used as instruments for diastolic blood pressure (DBP).

Table e-3. Single nucleotide polymorphisms (SNP) that fulfilled our selection criteria to be used as proxies for the effects for antihypertensive drug classes.

Table e-4. Genomic regions of encoding genes and regulatory regions (promoters or enhances) of known antihypertensive drug targets, as identified via GeneHancer. These regions were screened for instrument selection of single nucleotide polymorphisms (SNP) that were associated with systolic blood pressure at genome-wide significance.

Table e-5. Sensitivity analyses for the Mendelian randomization associations between genetically determined systolic and diastolic blood pressure and risk of stroke and stroke subtypes.

Table e-6. Sensitivity analyses for the Mendelian randomization associations between geneic proxies for beta blockers and calcium channel blockers and risk of stroke, risk of stroke subtypes, and WMH volume.

eMethods

Genome-wide association analysis for WMH volume in the UK Biobank individual-level data

We performed a genome-wide association study (GWAS) analysis for total volume of white matter hyperintensities (WMH), derived from T1 and T2-FLAIR images in the UK Biobank data. Total WMH volume definition was based on the field 25781 from the UK Biobank dataset. We followed the methodology, as has been previously described.[1] Specifically, we log-transformed WMH volume to approximate a normal distribution. For the GWAS, we excluded related participants (pi-hat >0.1875) and participants of non-White-British descent. This resulted in 10,597 individuals with available data on WMH volume, who were included in the analyses. SNPs with MAF <0.01 were excluded, as were SNPs not imputed from the HRC panel. We fit a linear regression model with log(WMHV) ~ SNP + age at MRI + sex + PCs1-10 + genotyping chip for each SNP.

1. Rutten-Jacobs LCA, Tozer DJ, Duering M, et al. Genetic Study of White Matter Integrity in UK Biobank (N=8448) and the Overlap With Stroke, Depression, and Dementia. Stroke 2018;49:1340-1347.

Table e-1. Genome-wide significant ($p<5x10^{-8}$) and independent ($r^2<0.001$) single nucleotide polymorphisms (SNP) that were used as instruments for systolic blood pressure (SBP).

SNP	Chr	Position (GRCh37/hg19)	Effect allele	Other allele	EAF	beta	SE	*p*-value	$R^{2\,a}$	F^b
rs488834	1	10767902	t	c	0.765	-0.380	0.037	2.4E-25	3.8E-04	272.5
rs10776752	1	113044328	t	g	0.081	0.821	0.058	4.6E-46	3.4E-04	247.8
rs59980837	1	115827266	t	g	0.018	1.100	0.116	3.3E-21	1.1E-04	77.7
rs6699618	1	11881441	c	g	0.840	0.912	0.041	1.7E-109	6.7E-04	497.1
rs11585169	1	150572037	a	t	0.577	0.180	0.031	5.3E-09	2.4E-04	175.5
rs76719272	1	156129796	t	c	0.131	-0.274	0.046	3.0E-09	1.7E-04	126.5
rs75461554	1	15810172	t	c	0.201	-0.302	0.038	1.2E-15	2.7E-04	196.3
rs1889785	1	16348729	a	g	0.455	0.178	0.030	4.4E-09	2.4E-04	179.3
rs7796	1	1684169	c	g	0.511	0.339	0.031	5.0E-27	4.6E-04	338.1
rs12731646	1	169090660	t	c	0.409	-0.189	0.031	7.2E-10	2.5E-04	185.2
rs1043069	1	180859368	t	g	0.616	0.234	0.031	5.3E-14	3.0E-04	224.7
rs4651224	1	184585182	t	c	0.447	0.199	0.031	9.0E-11	2.7E-04	199.0
rs12042924	1	197297417	t	c	0.528	-0.181	0.030	2.6E-09	2.5E-04	182.7
rs11120093	1	207211326	t	c	0.408	-0.179	0.031	5.1E-09	2.4E-04	175.7
rs2724377	1	207974818	a	g	0.530	0.194	0.030	1.3E-10	2.7E-04	195.9
rs7555285	1	209970355	c	g	0.801	0.229	0.038	1.1E-09	2.0E-04	148.3
rs263532	1	2164116	t	c	0.576	0.180	0.031	4.7E-09	2.4E-04	177.5
rs68085857	1	217737629	t	c	0.234	0.274	0.036	1.7E-14	2.7E-04	199.3
rs72742507	1	221265336	t	c	0.300	-0.205	0.033	3.8E-10	2.4E-04	174.9
rs708117	1	228199902	a	g	0.520	0.287	0.030	1.6E-21	3.9E-04	291.1
rs699	1	230845794	a	g	0.593	-0.375	0.031	5.6E-34	5.0E-04	358.8
rs1565440	1	243387788	a	g	0.375	0.175	0.031	1.9E-08	2.2E-04	166.1
rs4926499	1	249155909	c	g	0.826	0.297	0.044	1.3E-11	2.3E-04	166.4
rs404100	1	25366987	t	c	0.451	0.194	0.030	1.7E-10	2.6E-04	194.2
rs34079867	1	27407850	t	c	0.266	0.199	0.035	1.8E-08	2.1E-04	157.6
rs4908348	1	28706949	t	g	0.694	0.237	0.033	8.1E-13	2.8E-04	203.5
rs2493296	1	3327032	t	c	0.143	0.418	0.044	3.1E-21	2.8E-04	203.5
rs11210029	1	41865293	a	g	0.632	-0.203	0.031	8.9E-11	2.6E-04	191.3
rs1408945	1	42364877	t	g	0.424	-0.320	0.030	8.3E-26	4.3E-04	316.4
rs1209384	1	43765089	a	g	0.388	0.256	0.031	2.8E-16	3.3E-04	244.8
rs778124	1	56606206	a	g	0.374	0.297	0.031	1.5E-21	3.8E-04	281.6
rs61772592	1	56979681	a	g	0.875	-0.318	0.046	2.9E-12	1.9E-04	141.7
rs12063372	1	59621911	a	g	0.385	0.199	0.032	3.9E-10	2.6E-04	190.8
rs10779795	1	6677064	a	g	0.661	0.219	0.032	7.4E-12	2.7E-04	196.9
rs12136922	1	67007389	a	g	0.495	0.203	0.030	2.7E-11	2.8E-04	200.3
rs658780	1	78555928	t	g	0.745	-0.203	0.035	5.3E-09	2.1E-04	156.2
rs786923	1	89242954	t	c	0.624	-0.308	0.031	2.8E-23	4.0E-04	293.1
rs7514579	1	94051350	a	c	0.771	0.224	0.036	5.5E-10	2.2E-04	160.6
rs1006545	10	102553647	t	g	0.887	0.685	0.048	3.5E-46	3.8E-04	278.1
rs11191580	10	104906211	t	c	0.918	1.100	0.055	7.7E-89	4.6E-04	337.4
rs117464403	10	107158054	a	g	0.018	0.864	0.120	5.8E-13	8.5E-05	62.1
rs12255372	10	114808902	t	g	0.288	0.236	0.034	1.9E-12	2.7E-04	194.1
rs1801253	10	115805056	c	g	0.734	0.463	0.034	2.8E-41	5.0E-04	366.8

rs72842207	10	121433675	t	c	0.214	-0.203	0.037	3.1E-08	1.9E-04	138.7
rs11592107	10	122968964	a	g	0.310	0.302	0.033	1.5E-20	3.6E-04	262.3
rs7093894	10	124234880	a	c	0.151	0.236	0.043	3.2E-08	1.7E-04	122.9
rs7912283	10	133773019	a	g	0.647	-0.214	0.032	2.9E-11	2.7E-04	198.2
rs1133400	10	134459388	a	g	0.786	-0.298	0.038	2.5E-15	2.8E-04	198.8
rs1623474	10	18471794	t	c	0.330	0.383	0.032	7.7E-33	4.7E-04	343.6
rs12258967	10	18727959	c	g	0.705	0.633	0.034	1.1E-78	7.2E-04	533.8
rs3802517	10	28233469	a	t	0.462	0.253	0.030	4.6E-17	3.5E-04	254.9
rs12264186	10	32289986	t	c	0.187	0.214	0.039	3.6E-08	1.8E-04	131.8
rs11252324	10	4124568	t	g	0.077	-0.416	0.057	3.6E-13	1.6E-04	120.2
rs4948643	10	45379759	t	c	0.282	0.226	0.034	2.4E-11	2.5E-04	185.2
rs34130368	10	48411796	t	g	0.117	-0.302	0.050	1.3E-09	1.7E-04	126.1
rs4245599	10	60365755	a	g	0.458	-0.179	0.031	4.0E-09	2.4E-04	180.7
rs57946343	10	63499951	t	c	0.853	0.716	0.043	2.1E-63	4.9E-04	364.0
rs2236295	10	64564892	t	g	0.398	-0.303	0.031	1.0E-22	4.0E-04	294.4
rs2177843	10	75409877	t	c	0.151	0.439	0.043	2.8E-24	3.1E-04	228.0
rs10749572	10	82136664	t	g	0.544	-0.203	0.030	1.9E-11	2.8E-04	204.3
rs111866816	10	94441507	t	c	0.071	0.357	0.060	2.3E-09	1.3E-04	95.3
rs2689690	10	95899706	t	c	0.368	-0.270	0.032	1.1E-17	3.5E-04	251.3
rs2274224	10	96039597	c	g	0.432	-0.452	0.030	6.0E-50	6.1E-04	449.4
rs604723	11	100610546	t	c	0.276	-0.655	0.034	2.5E-83	7.2E-04	530.2
rs7926110	11	107086143	t	g	0.673	0.260	0.032	5.7E-16	3.1E-04	232.4
rs641620	11	117074229	t	c	0.855	-0.319	0.044	3.7E-13	2.2E-04	159.0
rs573455	11	117267884	a	g	0.461	0.199	0.030	4.8E-11	2.7E-04	200.8
rs11222084	11	130273230	a	t	0.638	-0.336	0.032	1.8E-26	4.3E-04	314.9
rs7944927	11	130490917	t	c	0.782	0.224	0.039	1.2E-08	2.1E-04	154.4
rs2014408	11	16365282	t	c	0.209	0.517	0.037	1.3E-43	4.7E-04	346.5
rs7926335	11	16917869	t	c	0.269	0.314	0.034	2.5E-20	3.4E-04	249.5
rs569550	11	1887068	t	g	0.604	-0.577	0.032	1.3E-73	7.6E-04	544.8
rs74048190	11	2114221	t	c	0.952	-0.440	0.076	6.1E-09	1.1E-04	79.1
rs17762	11	22492454	a	g	0.078	0.412	0.057	5.6E-13	1.6E-04	119.7
rs1382472	11	27273967	a	g	0.404	-0.192	0.031	4.5E-10	2.5E-04	187.3
rs871004	11	28512458	a	g	0.348	0.234	0.032	1.6E-13	2.9E-04	215.1
rs1340030	11	30182068	t	c	0.635	0.194	0.031	5.8E-10	2.5E-04	182.2
rs11604310	11	45351420	t	c	0.166	-0.278	0.041	1.5E-11	2.1E-04	155.7
rs7107356	11	47676170	a	g	0.496	-0.460	0.030	1.6E-52	6.3E-04	466.6
rs2904315	11	48109948	a	g	0.313	-0.208	0.033	1.6E-10	2.5E-04	181.6
rs7125196	11	61272565	t	c	0.882	0.442	0.047	7.3E-21	2.5E-04	186.6
rs2306363	11	65405600	t	g	0.205	-0.436	0.038	5.2E-31	3.9E-04	287.7
rs7395791	11	69262916	a	g	0.442	-0.216	0.031	2.2E-12	2.9E-04	216.4
rs10501410	11	72088806	a	g	0.069	0.412	0.061	1.1E-11	1.5E-04	107.7
rs7927515	11	76125330	a	c	0.346	0.227	0.032	1.0E-12	2.8E-04	206.2
rs2289124	11	89224477	a	g	0.167	-0.308	0.042	1.1E-13	2.4E-04	173.9
rs360153	11	9762274	t	c	0.417	-0.345	0.031	1.7E-29	4.6E-04	339.8
rs67885470	11	99998431	t	c	0.209	-0.209	0.038	4.1E-08	1.9E-04	140.0
rs10207726	2	112744260	t	c	0.296	-0.214	0.033	8.1E-11	2.5E-04	181.1
rs6737318	2	114083120	a	g	0.778	0.235	0.036	1.1E-10	2.2E-04	164.5

rs2580350	2	121996007	a	g	0.561	0.177	0.031	8.4E-09	2.4E-04	176.6
rs17257081	2	135630498	a	g	0.807	0.227	0.039	6.4E-09	2.0E-04	140.9
rs55944332	2	145726621	a	g	0.763	-0.261	0.036	1.8E-13	2.6E-04	191.6
rs62170470	2	146989797	t	c	0.602	0.197	0.032	7.7E-10	2.6E-04	189.3
rs62187653	2	162469128	t	c	0.903	0.329	0.051	1.2E-10	1.6E-04	116.9
rs4667454	2	164867726	a	g	0.671	0.264	0.032	2.6E-16	3.2E-04	236.3
rs73029563	2	165008166	c	g	0.455	-0.514	0.030	4.2E-64	7.0E-04	516.7
rs11694601	2	174949358	a	g	0.597	-0.191	0.031	6.4E-10	2.5E-04	184.3
rs34727427	2	177016728	t	c	0.683	-0.235	0.032	4.0E-13	2.8E-04	206.7
rs1882212	2	182981968	a	g	0.779	0.275	0.036	3.3E-14	2.6E-04	191.9
rs13412750	2	191634958	a	g	0.271	-0.289	0.034	2.3E-17	3.1E-04	228.9
rs17760259	2	19744462	t	c	0.572	-0.265	0.030	2.3E-18	3.6E-04	263.6
rs12693982	2	204085635	t	c	0.402	0.258	0.031	7.5E-17	3.4E-04	250.3
rs3845811	2	208521512	c	g	0.566	-0.294	0.031	1.9E-21	4.0E-04	292.9
rs12694277	2	213188795	t	c	0.295	-0.202	0.034	1.8E-09	2.3E-04	170.2
rs2161967	2	218680529	t	g	0.428	0.284	0.031	2.9E-20	3.8E-04	281.8
rs3828282	2	218779144	c	g	0.428	0.186	0.032	5.3E-09	2.5E-04	184.2
rs10804330	2	227185749	t	c	0.567	0.235	0.031	1.6E-14	3.2E-04	233.6
rs1044822	2	230629138	t	c	0.148	-0.248	0.042	5.2E-09	1.7E-04	127.0
rs28365916	2	231280791	t	c	0.415	-0.171	0.031	2.2E-08	2.3E-04	168.5
rs139354822	2	242344695	t	c	0.970	0.612	0.098	3.5E-10	9.7E-05	69.5
rs2384063	2	25187115	t	c	0.761	0.327	0.036	6.3E-20	3.3E-04	240.9
rs1275988	2	26914364	t	c	0.611	-0.541	0.031	4.4E-69	7.1E-04	521.9
rs13420463	2	37517566	a	g	0.773	0.314	0.036	2.7E-18	3.0E-04	220.9
rs4952609	2	40555733	a	g	0.744	0.212	0.035	9.6E-10	2.2E-04	164.0
rs115262049	2	43196694	a	t	0.913	0.589	0.055	1.3E-26	2.6E-04	189.6
rs12464602	2	43397614	a	g	0.621	-0.244	0.032	1.0E-14	3.2E-04	232.8
rs13016772	2	55779476	t	c	0.765	0.252	0.036	1.2E-12	2.5E-04	183.9
rs2249105	2	65287896	a	g	0.632	0.293	0.031	7.6E-21	3.7E-04	273.2
rs10188003	2	66773469	t	c	0.393	0.188	0.031	8.8E-10	2.5E-04	182.0
rs6731373	2	68503044	a	g	0.349	0.191	0.033	4.2E-09	2.4E-04	176.2
rs6732123	2	69534650	c	g	0.417	-0.174	0.031	1.5E-08	2.3E-04	171.4
rs4577304	2	73403040	t	c	0.523	-0.177	0.030	5.0E-09	2.4E-04	178.9
rs72847885	2	86326717	a	g	0.663	0.241	0.032	3.1E-14	3.0E-04	218.5
rs9848170	3	11495983	c	g	0.597	0.323	0.031	7.0E-26	4.3E-04	315.5
rs12637573	3	121682388	a	g	0.472	-0.173	0.030	9.9E-09	2.4E-04	174.8
rs6438857	3	124557643	t	c	0.577	0.274	0.031	3.1E-19	3.7E-04	270.9
rs9880098	3	133949366	a	g	0.395	0.308	0.031	1.6E-23	4.0E-04	298.7
rs1199330	3	138101529	a	g	0.882	-0.265	0.047	1.7E-08	1.5E-04	110.5
rs9876694	3	141152017	t	c	0.058	0.471	0.065	4.6E-13	1.4E-04	105.0
rs11925504	3	14943965	a	g	0.572	-0.290	0.031	1.8E-21	3.9E-04	287.8
rs4408839	3	153729768	a	g	0.743	-0.230	0.035	2.4E-11	2.4E-04	178.2
rs79539362	3	154680449	t	c	0.899	0.400	0.050	2.1E-15	2.0E-04	147.2
rs17684859	3	158213841	t	c	0.734	-0.224	0.034	4.2E-11	2.4E-04	177.8
rs3980686	3	168697602	t	g	0.108	-0.500	0.049	1.0E-24	2.6E-04	194.6
rs1290784	3	169096900	t	c	0.448	0.412	0.030	3.0E-42	5.6E-04	412.9
rs2111557	3	169325621	t	c	0.468	0.176	0.030	5.2E-09	2.4E-04	178.2

rs4955575	3	169534538	a	c	0.746	0.216	0.035	5.6E-10	2.2E-04	165.9
rs262986	3	183435713	a	g	0.470	-0.237	0.031	7.7E-15	3.2E-04	239.7
rs13091418	3	185329756	c	g	0.666	-0.223	0.033	6.1E-12	2.7E-04	201.4
rs9869437	3	196228360	a	c	0.352	-0.200	0.032	3.2E-10	2.5E-04	185.0
rs189267552	3	20073193	a	t	0.013	-0.866	0.139	4.6E-10	6.2E-05	45.6
rs2643826	3	27562988	t	c	0.451	0.447	0.031	1.7E-48	6.1E-04	449.5
rs68115553	3	27704702	a	g	0.980	-0.645	0.114	1.7E-08	6.9E-05	50.3
rs743395	3	37598382	t	c	0.383	0.260	0.032	2.6E-16	3.4E-04	248.8
rs6788984	3	41107173	a	g	0.856	0.300	0.043	3.8E-12	2.0E-04	149.7
rs1052501	3	41925398	t	c	0.833	0.226	0.041	4.1E-08	1.7E-04	126.3
rs6771917	3	48108442	t	c	0.248	-0.379	0.036	1.4E-26	3.9E-04	286.9
rs7615099	3	53143901	a	g	0.668	0.189	0.032	3.9E-09	2.3E-04	170.1
rs6445583	3	53562894	a	g	0.747	0.277	0.035	1.9E-15	2.9E-04	213.0
rs3772219	3	56771251	a	c	0.682	0.273	0.032	3.1E-17	3.3E-04	240.4
rs7618284	3	66422246	c	g	0.339	-0.189	0.033	1.1E-08	2.3E-04	171.8
rs4499560	3	70920485	a	t	0.317	-0.220	0.033	1.5E-11	2.6E-04	193.0
rs9857362	3	74710462	a	c	0.529	0.173	0.031	1.6E-08	2.4E-04	170.6
rs1375564	3	85656311	t	c	0.640	0.258	0.032	2.8E-16	3.3E-04	240.6
rs13107325	4	103188709	t	c	0.074	-0.909	0.059	4.2E-53	3.4E-04	251.3
rs11097909	4	106911321	t	c	0.147	-0.363	0.043	3.4E-17	2.5E-04	184.1
rs1493132	4	108861082	t	c	0.660	-0.177	0.032	2.7E-08	2.2E-04	160.1
rs1814951	4	111408718	a	g	0.879	-0.323	0.047	3.9E-12	1.9E-04	139.4
rs4834792	4	120555696	a	t	0.480	0.197	0.030	7.2E-11	2.7E-04	199.0
rs7439567	4	138464842	t	c	0.411	0.254	0.031	2.3E-16	3.4E-04	247.8
rs72719160	4	144051276	a	t	0.683	-0.224	0.032	4.3E-12	2.7E-04	196.3
rs2353940	4	145740898	t	c	0.751	-0.208	0.036	6.8E-09	2.1E-04	156.4
rs73855810	4	148383424	a	g	0.141	0.273	0.043	3.0E-10	1.8E-04	134.0
rs7683728	4	156402654	t	c	0.531	-0.365	0.030	2.4E-33	5.0E-04	364.4
rs12643599	4	156639846	a	g	0.640	0.313	0.031	1.2E-23	4.0E-04	293.2
rs17035181	4	157678511	t	g	0.855	0.307	0.043	7.6E-13	2.1E-04	154.2
rs869396	4	169688000	a	c	0.466	-0.212	0.031	4.1E-12	2.9E-04	213.3
rs2610990	4	18008232	a	g	0.264	-0.290	0.034	2.9E-17	3.1E-04	228.0
rs34535756	4	2246927	t	c	0.039	0.478	0.079	1.2E-09	9.9E-05	73.3
rs1290933	4	2668217	a	c	0.692	-0.285	0.033	3.2E-18	3.3E-04	246.3
rs55924432	4	26812737	t	c	0.401	0.265	0.032	5.7E-17	3.5E-04	257.0
rs2498323	4	3451109	a	g	0.098	0.317	0.052	8.5E-10	1.5E-04	113.4
rs2291434	4	38387244	t	g	0.534	-0.262	0.030	5.1E-18	3.6E-04	263.8
rs12511987	4	46595623	t	g	0.823	-0.233	0.040	5.4E-09	1.9E-04	137.9
rs62309747	4	48713862	a	g	0.473	-0.224	0.030	1.6E-13	3.1E-04	226.7
rs60991988	4	54801228	t	g	0.893	0.379	0.050	2.8E-14	2.0E-04	145.1
rs13107261	4	63768826	a	g	0.369	-0.178	0.031	1.6E-08	2.3E-04	166.1
rs10008637	4	77414144	t	c	0.541	0.216	0.030	9.2E-13	2.9E-04	217.4
rs12509595	4	81182554	t	c	0.708	-0.837	0.033	2.6E-138	9.5E-04	701.9
rs60909079	4	83830244	c	g	0.249	-0.211	0.035	1.7E-09	2.2E-04	159.8
rs17010957	4	86719165	t	c	0.854	-0.534	0.043	1.8E-35	3.7E-04	269.6
rs10028284	4	89752913	a	t	0.818	0.294	0.040	1.7E-13	2.4E-04	176.2
rs11241313	5	114428167	t	c	0.311	-0.207	0.033	2.2E-10	2.4E-04	180.1

rs1624822	5	122475437	t	c	0.620	-0.336	0.031	5.1E-27	4.4E-04	321.7
rs9327297	5	122835051	c	g	0.668	0.275	0.032	8.1E-18	3.4E-04	247.4
rs758180	5	127354423	a	t	0.225	0.208	0.037	1.3E-08	2.0E-04	146.8
rs6892983	5	127845030	a	c	0.402	0.343	0.031	7.1E-29	4.5E-04	334.0
rs10069690	5	1279790	t	c	0.258	0.310	0.037	4.5E-17	3.3E-04	230.8
rs702395	5	140086677	t	c	0.437	0.232	0.031	3.2E-14	3.1E-04	231.1
rs2913920	5	141726983	t	c	0.765	0.242	0.036	1.6E-11	2.4E-04	175.7
rs7725413	5	15695987	t	c	0.770	-0.199	0.036	3.1E-08	1.9E-04	142.5
rs1957563	5	157474590	t	c	0.265	0.363	0.034	2.3E-26	3.9E-04	285.7
rs11960210	5	157817634	t	c	0.625	0.473	0.031	1.3E-51	6.1E-04	443.1
rs13358657	5	157938070	a	g	0.867	-0.388	0.045	3.0E-18	2.5E-04	181.0
rs3860770	5	173301427	a	g	0.292	-0.266	0.033	1.2E-15	3.0E-04	221.7
rs12153395	5	179411477	a	g	0.115	-0.330	0.049	1.1E-11	1.8E-04	135.4
rs12656497	5	32831939	t	c	0.403	-0.638	0.031	7.1E-96	8.4E-04	621.9
rs10941043	5	33194751	t	g	0.710	-0.259	0.033	6.4E-15	2.9E-04	216.1
rs4957026	5	361148	a	g	0.340	0.198	0.032	8.1E-10	2.4E-04	179.7
rs2113077	5	50799442	a	g	0.430	0.210	0.031	6.1E-12	2.8E-04	208.3
rs1694068	5	53283630	a	t	0.614	0.266	0.031	1.2E-17	3.5E-04	255.6
rs13179413	5	55868097	t	c	0.282	0.224	0.035	1.1E-10	2.5E-04	183.6
rs34496659	5	61798934	a	g	0.070	0.455	0.062	1.5E-13	1.6E-04	120.4
rs6870654	5	63831964	t	c	0.745	0.214	0.035	7.6E-10	2.2E-04	162.7
rs4286632	5	66291370	a	g	0.731	0.211	0.034	7.6E-10	2.3E-04	168.5
rs7703560	5	67678506	a	g	0.700	-0.225	0.033	1.5E-11	2.6E-04	189.1
rs246973	5	68007803	t	c	0.288	0.248	0.034	1.5E-13	2.8E-04	206.1
rs6452769	5	87389027	a	g	0.205	-0.314	0.038	7.8E-17	2.8E-04	207.8
rs76443575	5	96211594	c	g	0.036	-0.523	0.082	1.4E-10	1.0E-04	73.4
rs1871190	5	97953719	t	g	0.335	0.195	0.032	1.7E-09	2.4E-04	176.4
rs9486916	6	109013930	t	c	0.198	0.266	0.039	5.4E-12	2.3E-04	169.1
rs961764	6	117522156	c	g	0.425	-0.191	0.031	3.7E-10	2.6E-04	189.4
rs1630736	6	12295987	t	c	0.465	-0.171	0.031	3.5E-08	2.3E-04	172.0
rs10782230	6	126228512	a	g	0.485	0.211	0.030	2.9E-12	2.9E-04	213.5
rs9401913	6	127159982	a	g	0.439	0.520	0.031	3.7E-65	7.0E-04	520.0
rs9349379	6	12903957	a	g	0.593	0.266	0.031	1.3E-17	3.5E-04	260.6
rs9285476	6	134159976	c	g	0.707	0.184	0.033	3.1E-08	2.1E-04	155.0
rs13204703	6	140692862	t	c	0.751	0.197	0.035	1.9E-08	2.0E-04	149.2
rs8180684	6	143200936	t	c	0.290	0.213	0.034	1.8E-10	2.4E-04	177.9
rs7765526	6	147713764	a	g	0.463	0.201	0.031	5.9E-11	2.7E-04	202.5
rs17080102	6	151004770	c	g	0.069	-0.809	0.059	3.5E-42	2.9E-04	211.9
rs1293969	6	151959945	t	c	0.748	-0.199	0.035	1.0E-08	2.1E-04	151.9
rs509833	6	159711515	a	g	0.139	0.329	0.044	7.1E-14	2.2E-04	159.2
rs2745599	6	1613686	a	g	0.552	0.216	0.032	9.0E-12	2.9E-04	214.3
rs12661036	6	163737476	t	c	0.775	-0.210	0.037	1.8E-08	2.0E-04	148.7
rs7744902	6	166176722	a	g	0.077	-0.409	0.059	5.6E-12	1.6E-04	113.4
rs9368222	6	20686996	a	c	0.269	0.228	0.034	1.8E-11	2.5E-04	181.9
rs9393231	6	22123695	a	c	0.492	-0.215	0.031	3.4E-12	3.0E-04	217.6
rs7753826	6	26042239	a	t	0.190	0.428	0.039	1.0E-28	3.6E-04	267.0
rs2596498	6	31322688	t	c	0.638	-0.233	0.034	4.9E-12	3.0E-04	204.8

rs3132442	6	31839494	t	c	0.520	0.393	0.030	2.6E-38	5.4E-04	392.7
rs7763558	6	43349215	a	g	0.324	0.336	0.032	1.2E-25	4.0E-04	299.0
rs11967262	6	43760327	c	g	0.513	-0.172	0.031	3.4E-08	2.4E-04	172.0
rs78648104	6	50683009	t	c	0.908	-0.429	0.054	2.4E-15	2.0E-04	145.4
rs1575290	6	7715689	t	c	0.473	0.197	0.030	5.6E-11	2.7E-04	199.6
rs1984195	6	79657391	a	g	0.489	0.241	0.030	1.8E-15	3.3E-04	241.6
rs9361836	6	82235408	t	c	0.317	0.220	0.032	1.2E-11	2.6E-04	193.0
rs6921291	6	97066242	t	c	0.191	0.358	0.039	1.6E-20	3.0E-04	223.9
rs2392929	7	106414069	t	g	0.797	-0.751	0.038	2.0E-87	6.7E-04	491.9
rs34072724	7	130432469	a	g	0.489	-0.242	0.030	1.4E-15	3.3E-04	244.9
rs35680304	7	130973495	t	c	0.593	0.269	0.031	3.8E-18	3.6E-04	263.1
rs75672964	7	131321010	t	c	0.042	0.589	0.084	2.3E-12	1.3E-04	92.3
rs6957161	7	131361319	a	g	0.262	0.206	0.035	2.2E-09	2.2E-04	161.2
rs73727605	7	149474622	a	g	0.066	0.362	0.062	6.6E-09	1.2E-04	89.4
rs3918226	7	150690176	t	c	0.081	0.664	0.058	8.5E-31	2.7E-04	199.0
rs10224210	7	151413194	t	c	0.721	-0.383	0.034	1.6E-29	4.2E-04	312.7
rs1870735	7	155744303	c	g	0.453	0.206	0.031	3.6E-11	2.8E-04	206.9
rs3807925	7	18543250	a	g	0.650	-0.186	0.032	5.4E-09	2.3E-04	171.2
rs28688791	7	19039605	t	c	0.802	-0.322	0.038	2.3E-17	2.8E-04	207.8
rs6978112	7	1966841	t	c	0.411	0.229	0.031	1.3E-13	3.0E-04	223.7
rs112509803	7	24735004	c	g	0.114	-0.264	0.048	3.2E-08	1.5E-04	108.1
rs10282122	7	2529623	t	c	0.668	-0.302	0.033	2.5E-20	3.7E-04	270.5
rs3735533	7	27245893	t	c	0.074	-0.910	0.058	5.3E-56	3.4E-04	253.7
rs6961048	7	27328187	c	g	0.896	-0.530	0.050	1.4E-26	2.7E-04	199.5
rs11977526	7	46008110	a	g	0.401	-0.321	0.031	6.6E-25	4.2E-04	310.7
rs73049928	7	4669949	a	g	0.806	-0.238	0.039	1.2E-09	2.0E-04	150.8
rs12668436	7	47548893	t	c	0.754	-0.215	0.035	7.9E-10	2.2E-04	161.9
rs848445	7	77572461	t	c	0.285	-0.203	0.034	2.3E-09	2.3E-04	167.5
rs67617547	7	90297177	c	g	0.670	0.180	0.032	2.4E-08	2.2E-04	161.5
rs42032	7	92237426	a	g	0.264	-0.323	0.035	7.4E-21	3.5E-04	254.1
rs79069610	8	105921209	t	c	0.950	-0.401	0.073	3.7E-08	1.0E-04	77.2
rs35783704	8	105966258	a	g	0.104	-0.462	0.051	8.8E-20	2.4E-04	174.7
rs1821002	8	10640065	c	g	0.411	0.379	0.031	5.2E-35	5.0E-04	372.7
rs7830607	8	110097287	a	g	0.305	-0.206	0.033	3.1E-10	2.4E-04	177.1
rs2470004	8	120358445	t	c	0.818	-0.345	0.039	1.3E-18	2.8E-04	209.1
rs6986368	8	126513197	a	t	0.673	-0.213	0.033	9.6E-11	2.6E-04	187.5
rs2608029	8	129170126	c	g	0.665	0.181	0.032	1.6E-08	2.2E-04	162.9
rs4260863	8	129386613	c	g	0.616	0.191	0.031	1.2E-09	2.5E-04	180.3
rs7012866	8	135616959	t	g	0.499	-0.233	0.030	1.2E-14	3.2E-04	235.6
rs4440615	8	141057641	a	g	0.632	-0.220	0.031	1.9E-12	2.8E-04	207.7
rs4961293	8	141812374	t	c	0.451	0.227	0.030	7.4E-14	3.1E-04	227.9
rs7463212	8	143991858	a	t	0.545	-0.275	0.031	1.8E-19	3.8E-04	273.6
rs71499040	8	1711918	c	g	0.708	0.222	0.034	5.6E-11	2.5E-04	184.6
rs7844887	8	23402482	a	g	0.221	0.266	0.036	2.4E-13	2.5E-04	185.9
rs7821832	8	25889446	t	g	0.745	0.422	0.035	6.7E-34	4.4E-04	322.1
rs77375686	8	26043622	a	g	0.888	-0.347	0.049	8.4E-13	1.9E-04	139.6
rs1906672	8	38130025	a	g	0.232	0.297	0.036	1.2E-16	2.9E-04	214.4

rs4873492	8	51947549	t	c	0.172	0.343	0.040	1.6E-17	2.7E-04	198.7
rs2354862	8	64501744	a	c	0.641	0.251	0.032	2.4E-15	3.2E-04	231.4
rs13253358	8	68920135	t	c	0.298	0.213	0.033	1.1E-10	2.4E-04	180.5
rs2126474	8	76878957	t	g	0.413	-0.260	0.031	1.9E-17	3.5E-04	255.4
rs9918876	8	77681097	a	c	0.104	-0.298	0.050	2.3E-09	1.5E-04	112.2
rs148401029	8	81386066	a	c	0.035	-0.462	0.085	5.0E-08	8.6E-05	63.7
rs10091532	8	82853793	a	c	0.417	-0.207	0.031	1.3E-11	2.8E-04	203.9
rs843093	8	92528310	a	g	0.709	-0.209	0.034	7.0E-10	2.4E-04	174.4
rs2613203	8	95253197	a	t	0.815	-0.268	0.039	5.8E-12	2.2E-04	164.2
rs10980408	9	113249071	t	c	0.964	-0.761	0.083	3.8E-20	1.4E-04	107.9
rs7026176	9	116670743	t	g	0.512	-0.187	0.030	4.0E-10	2.6E-04	191.5
rs34025993	9	123516572	a	g	0.414	0.223	0.031	4.7E-13	3.0E-04	221.5
rs4838021	9	125657099	t	c	0.129	-0.301	0.045	3.1E-11	1.9E-04	136.9
rs13289468	9	128180332	a	c	0.574	0.249	0.031	3.9E-16	3.3E-04	249.1
rs6271	9	136522274	t	c	0.074	-0.555	0.061	1.2E-19	2.1E-04	153.0
rs11145807	9	139520789	a	g	0.406	0.214	0.032	3.5E-11	2.8E-04	204.2
rs9886665	9	22942770	t	c	0.267	0.205	0.034	2.5E-09	2.2E-04	162.2
rs4553000	9	34223553	t	c	0.514	-0.204	0.030	1.1E-11	2.8E-04	208.4
rs76452347	9	35906471	t	c	0.205	-0.297	0.040	7.1E-14	2.7E-04	198.2
rs927315	9	4117713	t	c	0.471	0.169	0.030	2.4E-08	2.3E-04	172.3
rs60191654	9	753648	a	g	0.812	-0.238	0.039	5.9E-10	2.0E-04	149.2
rs1410222	9	77239540	t	c	0.817	0.217	0.039	2.2E-08	1.8E-04	133.4
rs1332813	9	9350706	t	c	0.351	0.220	0.031	2.3E-12	2.8E-04	205.9
rs7045409	9	95201540	a	t	0.367	-0.186	0.031	2.5E-09	2.4E-04	176.4
rs5742643	12	102837863	t	c	0.249	-0.223	0.035	1.5E-10	2.3E-04	169.9
rs7310615	12	111865049	c	g	0.482	0.585	0.031	1.3E-81	8.0E-04	592.1
rs1896326	12	115342956	a	g	0.229	-0.280	0.037	4.4E-14	2.7E-04	202.0
rs35444	12	115552437	a	g	0.614	0.437	0.031	3.5E-45	5.7E-04	420.0
rs6490019	12	115920472	a	g	0.380	-0.290	0.031	6.6E-21	3.7E-04	279.8
rs1169078	12	122416254	c	g	0.688	-0.197	0.033	1.7E-09	2.3E-04	173.3
rs2024385	12	12888438	a	t	0.424	-0.264	0.031	5.9E-18	3.5E-04	264.6
rs117206641	12	133086888	t	c	0.111	0.315	0.050	2.7E-10	1.7E-04	123.9
rs1010064	12	20000315	a	c	0.816	0.357	0.039	3.0E-20	2.9E-04	219.6
rs73075659	12	20373541	a	g	0.665	0.396	0.032	5.5E-35	4.8E-04	361.2
rs3819532	12	2436837	t	c	0.391	-0.188	0.031	9.4E-10	2.5E-04	182.9
rs2129869	12	26457650	a	t	0.778	-0.264	0.036	2.4E-13	2.5E-04	186.8
rs78998485	12	434755	c	g	0.744	-0.245	0.035	1.5E-12	2.6E-04	190.8
rs61917655	12	48210787	t	c	0.101	0.343	0.051	2.7E-11	1.7E-04	128.0
rs57342147	12	50129422	a	g	0.903	0.279	0.051	4.2E-08	1.3E-04	100.4
rs12426261	12	50573037	a	g	0.379	0.378	0.031	2.3E-34	4.9E-04	364.4
rs7134440	12	53450097	t	c	0.082	0.479	0.056	1.6E-17	2.0E-04	148.1
rs7134677	12	54441498	t	c	0.298	-0.385	0.033	4.5E-31	4.4E-04	329.8
rs7306710	12	66376091	t	c	0.481	-0.243	0.030	1.0E-15	3.3E-04	248.6
rs4143175	12	67782397	t	c	0.241	0.219	0.035	5.1E-10	2.2E-04	163.7
rs7963801	12	79685226	t	c	0.422	-0.236	0.031	2.9E-14	3.2E-04	235.9
rs6539467	12	79955306	a	g	0.166	0.265	0.040	5.6E-11	2.0E-04	150.5
rs113695818	12	8837407	t	c	0.303	-0.184	0.033	2.6E-08	2.1E-04	158.9

rs17249754	12	90060586	a	g	0.168	-0.845	0.040	1.3E-97	6.5E-04	483.5
rs10777213	12	90349999	a	g	0.524	-0.179	0.030	2.5E-09	2.4E-04	182.6
rs9549627	13	113652369	a	g	0.118	0.285	0.050	1.2E-08	1.6E-04	118.3
rs7331680	13	115000650	t	g	0.149	0.410	0.042	3.4E-22	2.9E-04	212.5
rs483071	13	22294117	t	c	0.625	0.271	0.031	5.1E-18	3.5E-04	260.0
rs9507885	13	27951090	t	c	0.095	-0.321	0.054	3.2E-09	1.5E-04	112.6
rs7338758	13	30137828	t	c	0.245	0.355	0.035	7.0E-24	3.6E-04	266.1
rs4274337	13	41967193	a	g	0.170	-0.297	0.041	2.5E-13	2.3E-04	171.5
rs7491248	13	47180671	a	g	0.224	0.216	0.036	2.4E-09	2.1E-04	152.4
rs9526707	13	51489186	a	g	0.322	-0.204	0.032	2.8E-10	2.4E-04	182.1
rs75961402	13	56398286	a	g	0.153	0.266	0.042	1.9E-10	1.9E-04	141.6
rs17245822	13	73131694	a	c	0.627	-0.190	0.031	1.2E-09	2.4E-04	182.1
rs78474310	13	73826901	a	g	0.955	-0.470	0.073	1.5E-10	1.1E-04	82.4
rs6562778	13	74223828	a	g	0.459	0.178	0.030	5.0E-09	2.4E-04	181.0
rs17562391	14	100133250	t	c	0.419	0.197	0.031	1.3E-10	2.6E-04	196.3
rs75016974	14	100197940	t	c	0.142	-0.251	0.044	1.0E-08	1.7E-04	125.6
rs12885878	14	104007555	a	g	0.234	-0.229	0.037	4.3E-10	2.3E-04	168.0
rs365990	14	23861811	a	g	0.634	0.225	0.031	6.0E-13	2.9E-04	214.0
rs8904	14	35871217	a	g	0.368	0.306	0.031	1.7E-22	3.9E-04	289.5
rs7493678	14	39400917	a	t	0.651	-0.189	0.032	2.3E-09	2.4E-04	176.0
rs72683923	14	50735947	t	c	0.979	0.959	0.110	3.1E-18	1.1E-04	81.3
rs35413927	14	53420358	a	g	0.695	-0.300	0.033	5.3E-20	3.5E-04	261.1
rs12883810	14	68032235	t	c	0.146	-0.238	0.043	2.7E-08	1.6E-04	120.4
rs57786342	14	69260028	a	g	0.206	0.232	0.037	5.6E-10	2.1E-04	155.3
rs8003103	14	71451265	a	g	0.345	-0.176	0.032	3.6E-08	2.2E-04	162.5
rs3815460	14	73422259	c	g	0.898	-0.285	0.050	1.2E-08	1.4E-04	106.7
rs11159091	14	75074316	a	g	0.462	0.198	0.030	6.8E-11	2.7E-04	198.9
rs7154723	14	98590629	a	g	0.385	0.253	0.031	2.7E-16	3.3E-04	245.3
rs4606697	15	100087596	a	g	0.104	-0.320	0.052	9.7E-10	1.6E-04	121.3
rs8030856	15	40314967	c	g	0.605	-0.176	0.031	1.2E-08	2.3E-04	172.6
rs28866311	15	41442195	t	g	0.526	-0.276	0.030	5.5E-20	3.8E-04	282.4
rs4775769	15	48939888	t	g	0.095	-0.416	0.052	7.8E-16	2.0E-04	146.0
rs3098186	15	50810621	t	c	0.516	-0.242	0.030	1.4E-15	3.3E-04	248.0
rs2652812	15	63406170	t	c	0.754	-0.252	0.035	1.0E-12	2.6E-04	190.9
rs28429256	15	66931617	a	g	0.334	0.215	0.033	3.9E-11	2.6E-04	195.6
rs11636952	15	75114322	t	c	0.314	0.531	0.033	4.2E-59	6.3E-04	458.2
rs2627313	15	81006712	t	c	0.445	0.321	0.030	3.6E-26	4.4E-04	321.2
rs1994158	15	86064327	a	g	0.819	0.251	0.039	1.2E-10	2.0E-04	152.1
rs17807723	15	90023558	a	g	0.138	-0.272	0.044	8.4E-10	1.8E-04	131.3
rs4932373	15	91429287	a	c	0.674	-0.635	0.033	2.5E-83	7.7E-04	556.0
rs12906962	15	95312071	t	c	0.676	-0.265	0.033	3.3E-16	3.2E-04	237.3
rs2589218	15	96785017	t	c	0.730	-0.226	0.034	2.5E-11	2.4E-04	182.1
rs11075030	16	11976414	a	c	0.594	-0.175	0.031	1.7E-08	2.3E-04	168.7
rs11641374	16	1347717	a	c	0.600	-0.194	0.031	3.3E-10	2.6E-04	190.2
rs77924615	16	20392332	a	g	0.199	-0.408	0.039	1.1E-25	3.6E-04	265.6
rs12596630	16	2065666	t	c	0.090	0.428	0.055	5.0E-15	1.9E-04	139.7
rs7186298	16	21088031	t	c	0.430	-0.232	0.030	1.9E-14	3.1E-04	232.6

rs8044992	16	24811207	t	c	0.712	0.214	0.033	1.1E-10	2.4E-04	179.6
rs7189884	16	4145164	a	g	0.115	-0.314	0.048	4.2E-11	1.7E-04	130.5
rs12446456	16	4922201	t	c	0.427	-0.300	0.030	3.0E-23	4.0E-04	301.4
rs34941092	16	50550137	a	g	0.150	-0.323	0.043	3.2E-14	2.3E-04	168.4
rs4784541	16	51704452	t	c	0.475	-0.202	0.031	4.9E-11	2.8E-04	205.7
rs35098810	16	60635748	a	c	0.768	0.197	0.036	3.2E-08	1.9E-04	143.6
rs146550789	16	66781040	t	c	0.958	-0.482	0.078	5.6E-10	1.1E-04	79.0
rs62047964	16	70729954	t	c	0.062	0.512	0.069	9.3E-14	1.6E-04	120.8
rs1012089	16	74171973	c	g	0.475	-0.192	0.030	2.0E-10	2.6E-04	196.3
rs4888408	16	75432824	a	g	0.586	0.365	0.031	1.4E-32	4.9E-04	363.1
rs12926550	16	81510155	a	g	0.316	-0.255	0.032	3.4E-15	3.0E-04	225.7
rs8054587	16	86170044	t	c	0.527	0.167	0.030	3.4E-08	2.3E-04	169.9
rs3950627	16	86436343	a	c	0.531	0.185	0.031	1.8E-09	2.5E-04	187.2
rs6540119	16	87984477	a	t	0.334	0.202	0.032	3.9E-10	2.5E-04	183.9
rs908951	16	89697625	t	c	0.438	-0.226	0.032	7.1E-13	3.1E-04	224.6
rs8079811	17	1371473	c	g	0.348	-0.210	0.033	1.0E-10	2.6E-04	193.7
rs4925159	17	18185510	a	g	0.425	0.217	0.031	9.7E-13	2.9E-04	215.4
rs7211535	17	19922364	a	g	0.476	-0.178	0.030	4.6E-09	2.4E-04	179.9
rs2760748	17	2001604	a	t	0.098	0.363	0.051	1.1E-12	1.8E-04	131.3
rs1551355	17	30032420	t	c	0.233	0.210	0.036	3.9E-09	2.1E-04	153.9
rs9899540	17	30777924	a	t	0.400	0.201	0.032	1.9E-10	2.7E-04	197.6
rs7213273	17	43155914	a	g	0.655	-0.400	0.032	6.2E-37	5.0E-04	370.7
rs17608766	17	45013271	t	c	0.856	-0.690	0.043	2.5E-57	4.7E-04	346.1
rs3764400	17	46123932	t	c	0.864	0.375	0.045	3.7E-17	2.4E-04	180.9
rs9897429	17	47518378	a	g	0.520	0.265	0.032	1.2E-16	3.6E-04	270.3
rs1000423	17	59475642	t	c	0.732	0.414	0.035	6.5E-33	4.5E-04	329.3
rs56288724	17	60767135	a	g	0.583	-0.218	0.031	2.0E-12	2.9E-04	216.8
rs62076622	17	61090958	a	g	0.801	0.236	0.038	3.8E-10	2.1E-04	154.3
rs6504213	17	62381714	t	c	0.418	-0.298	0.031	1.2E-21	4.0E-04	297.1
rs113086489	17	7171356	t	c	0.553	0.325	0.031	3.8E-26	4.4E-04	329.0
rs4511593	17	7455536	t	c	0.653	-0.288	0.032	1.3E-19	3.6E-04	264.8
rs1436138	17	75316880	a	g	0.637	0.312	0.032	4.7E-23	4.0E-04	295.4
rs9302885	17	76799898	a	g	0.445	0.224	0.030	1.0E-13	3.0E-04	226.7
rs79930761	17	7815712	t	c	0.087	-0.469	0.056	4.9E-17	2.1E-04	153.0
rs11655604	17	79365861	t	c	0.358	-0.203	0.033	1.1E-09	2.6E-04	179.7
rs62082230	18	22676071	a	t	0.277	-0.188	0.035	4.7E-08	2.1E-04	154.6
rs1154214	18	24546824	t	g	0.396	-0.203	0.031	3.3E-11	2.7E-04	198.9
rs56407827	18	42179819	t	c	0.269	0.360	0.034	2.8E-26	3.9E-04	289.9
rs11874246	18	42596789	t	c	0.296	0.286	0.033	3.2E-18	3.3E-04	244.2
rs7236548	18	43097750	a	c	0.185	0.343	0.039	8.5E-19	2.8E-04	211.9
rs1437649	18	48132646	a	g	0.235	-0.219	0.036	8.6E-10	2.2E-04	161.1
rs665445	18	51842682	a	c	0.279	-0.191	0.033	1.2E-08	2.1E-04	157.6
rs10048404	18	54578482	t	c	0.370	-0.261	0.032	1.9E-16	3.3E-04	248.9
rs10460108	18	73034151	a	g	0.480	0.214	0.030	1.1E-12	2.9E-04	219.1
rs34413141	18	777282	a	t	0.182	-0.353	0.039	2.5E-19	2.9E-04	215.7
rs3816865	19	11507855	a	g	0.080	0.309	0.057	4.4E-08	1.3E-04	92.4
rs167479	19	11526765	t	g	0.473	-0.564	0.033	7.2E-67	7.7E-04	522.5

rs698748	19	1424888	a	g	0.421	0.187	0.033	8.9E-09	2.5E-04	181.0
rs8106184	19	17159779	a	c	0.745	-0.237	0.035	8.3E-12	2.5E-04	184.7
rs149339216	19	2144046	t	c	0.957	-0.691	0.078	6.9E-19	1.6E-04	114.2
rs4319878	19	21924452	t	c	0.561	0.169	0.031	3.8E-08	2.3E-04	170.9
rs8108027	19	22115901	c	g	0.295	0.195	0.033	3.7E-09	2.2E-04	166.0
rs28572357	19	31867447	a	c	0.602	-0.273	0.031	6.3E-19	3.6E-04	266.8
rs1433121	19	32591878	t	c	0.691	-0.228	0.033	2.7E-12	2.7E-04	199.7
rs33836	19	34008600	t	c	0.462	0.177	0.030	6.6E-09	2.4E-04	180.0
rs10420519	19	45298461	t	g	0.035	-0.492	0.089	2.9E-08	9.1E-05	66.9
rs7255933	19	45766729	a	g	0.257	0.231	0.035	2.4E-11	2.4E-04	180.7
rs11672660	19	46180184	t	c	0.200	0.221	0.038	6.3E-09	1.9E-04	143.2
rs571689	19	49207554	t	c	0.520	0.228	0.030	6.8E-14	3.1E-04	230.6
rs73046792	19	49605705	a	g	0.159	-0.355	0.043	7.2E-17	2.6E-04	192.4
rs68096471	19	5175709	a	g	0.266	-0.210	0.034	9.3E-10	2.3E-04	167.9
rs12985940	19	7262734	t	c	0.841	0.464	0.043	1.1E-26	3.4E-04	246.5
rs2423514	20	10693337	a	g	0.541	0.301	0.030	1.8E-23	4.1E-04	306.6
rs6108787	20	10967214	t	g	0.530	-0.427	0.030	5.4E-46	5.9E-04	435.5
rs6078093	20	11168669	a	g	0.428	-0.185	0.030	1.2E-09	2.5E-04	185.3
rs8125763	20	17883531	a	c	0.472	0.176	0.030	4.8E-09	2.4E-04	179.9
rs17812022	20	19007099	t	c	0.096	-0.361	0.053	5.6E-12	1.7E-04	128.1
rs6058088	20	30139886	t	g	0.844	0.283	0.042	1.1E-11	2.1E-04	153.0
rs79384779	20	31214944	t	c	0.151	0.318	0.043	1.1E-13	2.2E-04	167.3
rs6029756	20	40266681	a	g	0.323	-0.271	0.033	1.9E-16	3.3E-04	242.6
rs6031431	20	42795152	a	g	0.538	-0.262	0.030	7.0E-18	3.6E-04	266.0
rs2598	20	47241618	a	g	0.533	0.168	0.030	2.9E-08	2.3E-04	171.4
rs6090907	20	47410231	a	g	0.147	-0.385	0.043	1.3E-19	2.7E-04	198.1
rs234623	20	57488964	a	g	0.504	-0.180	0.030	2.4E-09	2.5E-04	183.8
rs6026744	20	57742388	a	t	0.877	-0.713	0.046	7.0E-54	4.2E-04	313.3
rs28374392	20	61189717	t	c	0.623	0.192	0.034	1.2E-08	2.5E-04	168.4
rs6062324	20	62446351	a	g	0.236	-0.329	0.036	1.2E-19	3.3E-04	241.5
rs6054139	20	6327810	a	g	0.606	0.209	0.031	8.2E-12	2.7E-04	205.0
rs2776037	21	16317933	t	c	0.415	-0.185	0.031	2.1E-09	2.5E-04	183.7
rs1882961	21	16556367	t	c	0.309	0.244	0.033	6.7E-14	2.9E-04	213.8
rs2833834	21	33814378	a	c	0.277	0.218	0.034	1.2E-10	2.4E-04	176.6
rs12627651	21	44760603	a	g	0.287	0.350	0.034	1.0E-24	3.9E-04	292.0
rs34487963	21	44838330	a	c	0.019	-0.882	0.124	1.4E-12	8.8E-05	63.0
rs7278003	21	44966069	t	c	0.438	-0.188	0.030	6.6E-10	2.5E-04	189.1
rs2238787	22	19976406	a	g	0.292	0.255	0.033	1.5E-14	2.9E-04	215.7
rs12321	22	29453193	c	g	0.433	-0.229	0.030	3.8E-14	3.1E-04	230.7
rs112854918	22	30588910	c	g	0.975	-0.558	0.100	2.8E-08	7.6E-05	56.7
rs8142376	22	32001037	t	c	0.491	0.168	0.030	2.2E-08	2.3E-04	171.7
rs148140538	22	50228044	t	c	0.081	-0.325	0.056	7.4E-09	1.3E-04	98.5
rs28578714	22	50727921	t	c	0.606	0.207	0.033	2.5E-10	2.7E-04	193.3

[a] $R^2 = \frac{2 \times EAF \times (1-EAF) \times beta^2}{SD^2}$,*wh*ere EAF is the effect allele frequency and beta is the effect estimate of the SNP on SBP (Shim 2015, PLoS One;10(4):e0120758).

[b] $F = \frac{R^2 \times (N-2)}{1-R^2}$ where R^2 is the variance of SBP explained by the specific SNP (as explained above) and *N* the number of individuals in the GWAS analysis (Palmer 2012, Stat Methods Med Res;21(3):223-42).

CHR: chromosome; EAF: effect allele frequency; SE: standard error; SNP: single nucleotide polymorphism.

Table e-2. Genome-wide significant ($p<5x10^{-8}$) and independent ($r^2<0.001$) single nucleotide polymorphisms (SNP) that were used as instruments for diastolic blood pressure (DBP).

SNP	Chr	Position (GRCh37/hg19)	Effect allele	Other allele	EAF	beta	SE	*p*-value	$R^{2\,a}$	F^b
rs488834	1	10767902	t	c	0.764	-0.193	0.021	1.9E-20	1.9E-04	142.4
rs10776752	1	1.13E+08	t	g	0.081	0.457	0.033	1.2E-43	1.9E-04	141.0
rs57748895	1	1.16E+08	a	t	0.982	-0.663	0.067	2.5E-23	6.4E-05	48.4
rs55857306	1	11895795	a	g	0.160	-0.522	0.024	5.1E-109	3.9E-04	292.0
rs72704264	1	1.46E+08	c	g	0.217	0.117	0.021	3.6E-08	1.1E-04	82.8
rs1819663	1	1.54E+08	a	g	0.507	0.115	0.017	4.6E-11	1.6E-04	118.0
rs11578696	1	1.56E+08	a	g	0.866	0.146	0.026	1.7E-08	9.3E-05	70.5
rs1889785	1	16348729	a	g	0.455	0.126	0.017	5.6E-13	1.7E-04	129.6
rs7524019	1	1.67E+08	t	c	0.492	0.104	0.017	2.6E-09	1.4E-04	107.8
rs12405515	1	1.72E+08	t	g	0.570	-0.170	0.017	1.9E-22	2.3E-04	172.8
rs34645159	1	1724366	a	g	0.501	-0.133	0.017	2.1E-14	1.8E-04	138.5
rs150816167	1	1.8E+08	t	c	0.955	-0.287	0.045	1.2E-10	6.8E-05	51.5
rs1999996	1	1.85E+08	a	g	0.559	-0.112	0.018	1.7E-10	1.5E-04	112.4
rs882624	1	2.02E+08	t	c	0.333	-0.157	0.019	2.3E-17	1.9E-04	143.6
rs2169137	1	2.04E+08	c	g	0.729	0.159	0.019	3.2E-16	1.7E-04	130.7
rs1502358	1	2.17E+08	a	g	0.681	-0.113	0.019	1.1E-09	1.3E-04	101.8
rs68085857	1	2.18E+08	t	c	0.234	0.191	0.021	9.8E-21	1.9E-04	142.6
rs35981664	1	2.19E+08	a	t	0.688	-0.161	0.019	2.0E-17	1.9E-04	143.4
rs2760061	1	2.28E+08	a	t	0.480	0.177	0.018	1.0E-23	2.4E-04	183.9
rs699	1	2.31E+08	a	g	0.593	-0.236	0.018	1.3E-40	3.1E-04	231.8
rs3943093	1	2.43E+08	t	c	0.323	0.248	0.018	3.9E-41	3.0E-04	225.7
rs4926499	1	2.49E+08	c	g	0.826	0.169	0.025	9.4E-12	1.3E-04	97.8
rs6686889	1	25030470	t	c	0.253	0.192	0.020	6.9E-22	2.0E-04	151.1
rs12728150	1	27268737	a	g	0.919	-0.205	0.032	1.3E-10	8.4E-05	63.4
rs2493296	1	3327032	t	c	0.142	0.250	0.025	7.5E-23	1.7E-04	124.2
rs2146315	1	42050366	t	c	0.232	-0.120	0.021	5.0E-09	1.2E-04	88.6
rs710249	1	43869235	c	g	0.426	0.150	0.017	6.4E-18	2.0E-04	152.9
rs4926901	1	48025824	a	g	0.355	0.098	0.018	4.8E-08	1.2E-04	93.8
rs4926923	1	48109225	t	c	0.912	0.192	0.031	4.7E-10	8.5E-05	63.6
rs78256308	1	50814474	t	g	0.981	0.411	0.066	3.7E-10	4.3E-05	32.5
rs10493408	1	66992054	a	c	0.133	0.158	0.026	5.1E-10	1.0E-04	75.3
rs34517439	1	78450517	a	c	0.120	-0.251	0.028	2.0E-19	1.5E-04	110.2
rs786921	1	89286673	a	g	0.596	-0.115	0.018	8.6E-11	1.5E-04	113.3
rs17396055	1	94730954	a	g	0.332	-0.115	0.018	4.1E-10	1.4E-04	105.1
rs1006545	10	1.03E+08	t	g	0.888	0.363	0.028	8.0E-40	2.0E-04	151.1
rs2273654	10	1.03E+08	t	c	0.561	0.117	0.018	2.8E-11	1.6E-04	118.2
rs12414028	10	1.05E+08	a	t	0.088	-0.514	0.031	1.6E-60	2.3E-04	172.0
rs2067831	10	1.06E+08	c	g	0.272	-0.128	0.020	5.1E-11	1.4E-04	105.6
rs2484294	10	1.16E+08	a	g	0.733	0.317	0.020	1.2E-58	3.4E-04	258.2
rs72842207	10	1.21E+08	t	c	0.215	-0.211	0.021	1.1E-23	2.0E-04	148.4
rs11592107	10	1.23E+08	a	g	0.309	0.120	0.019	1.2E-10	1.4E-04	107.0
rs10490923	10	1.24E+08	a	g	0.126	0.153	0.026	5.0E-09	9.3E-05	70.1
rs9419374	10	1.34E+08	a	g	0.354	0.116	0.019	3.4E-10	1.5E-04	110.6

rs1133400	10	1.34E+08	a	g	0.785	-0.132	0.022	8.3E-10	1.2E-04	90.7
rs6602177	10	17167141	t	c	0.707	-0.120	0.021	6.5E-09	1.4E-04	103.6
rs1623474	10	18471794	t	c	0.330	0.223	0.018	6.2E-34	2.7E-04	205.7
rs12258967	10	18727959	c	g	0.704	0.354	0.019	3.3E-75	4.1E-04	306.7
rs3802517	10	28233469	a	t	0.462	0.129	0.017	9.3E-14	1.8E-04	133.1
rs1265842	10	28924901	t	c	0.483	0.111	0.017	1.7E-10	1.5E-04	115.7
rs2487926	10	30300787	a	g	0.571	0.097	0.018	3.3E-08	1.3E-04	99.2
rs3006583	10	31280845	t	c	0.811	-0.130	0.022	4.7E-09	1.1E-04	83.0
rs11252324	10	4124568	t	g	0.077	-0.234	0.033	1.0E-12	9.1E-05	69.2
rs4948643	10	45379759	t	c	0.282	0.159	0.019	2.3E-16	1.8E-04	134.0
rs34130368	10	48411796	t	g	0.117	-0.203	0.028	8.8E-13	1.2E-04	87.1
rs72831343	10	63515681	t	g	0.858	0.494	0.025	4.8E-88	3.3E-04	249.0
rs2236295	10	64564892	t	g	0.399	-0.207	0.018	1.4E-31	2.7E-04	206.8
rs35506078	10	65210552	t	c	0.663	-0.135	0.018	1.5E-13	1.7E-04	125.4
rs12247028	10	75410052	a	g	0.632	-0.140	0.019	1.2E-13	1.8E-04	132.8
rs2274224	10	96039597	c	g	0.433	-0.279	0.017	1.2E-57	3.8E-04	284.6
rs604723	11	1.01E+08	t	c	0.275	-0.385	0.019	2.3E-87	4.2E-04	319.4
rs66682451	11	1.07E+08	a	g	0.725	0.135	0.019	3.4E-12	1.5E-04	111.8
rs7106104	11	1.12E+08	t	c	0.719	-0.119	0.019	7.7E-10	1.3E-04	99.8
rs12790943	11	1.2E+08	t	c	0.421	-0.100	0.018	1.1E-08	1.3E-04	101.7
rs12574332	11	1.23E+08	t	c	0.123	0.207	0.027	6.1E-15	1.2E-04	92.9
rs4936099	11	1.3E+08	a	c	0.599	0.175	0.018	1.2E-22	2.3E-04	174.1
rs7107711	11	13255538	c	g	0.778	0.126	0.021	1.7E-09	1.2E-04	90.3
rs7123705	11	14255043	c	g	0.816	-0.147	0.023	7.9E-11	1.2E-04	91.7
rs28570096	11	1616088	t	c	0.309	0.140	0.019	1.1E-13	1.6E-04	124.2
rs10832586	11	16304089	a	c	0.798	-0.308	0.022	2.5E-46	2.7E-04	206.7
rs7926335	11	16917869	t	c	0.270	0.180	0.020	2.0E-20	2.0E-04	147.6
rs79889784	11	1702117	t	g	0.018	-0.394	0.072	3.9E-08	3.7E-05	25.8
rs569550	11	1887068	t	g	0.605	-0.269	0.018	1.2E-49	3.5E-04	260.6
rs147081004	11	1919980	a	c	0.856	0.141	0.026	4.1E-08	9.6E-05	70.9
rs10500932	11	22501446	a	g	0.074	0.278	0.033	5.8E-17	1.1E-04	79.6
rs962369	11	27734420	t	c	0.699	0.168	0.019	6.0E-19	1.9E-04	146.0
rs7933758	11	31000774	t	c	0.305	-0.114	0.019	2.6E-09	1.3E-04	100.3
rs10838702	11	47410888	t	g	0.388	0.238	0.018	1.3E-40	3.1E-04	234.1
rs10839259	11	49321410	t	c	0.763	0.145	0.021	2.1E-12	1.4E-04	109.1
rs751984	11	61278246	t	c	0.883	0.394	0.028	1.4E-46	2.2E-04	167.6
rs35927325	11	63882495	t	c	0.061	0.222	0.036	1.0E-09	7.0E-05	53.1
rs2306363	11	65405600	t	g	0.205	-0.264	0.022	1.6E-34	2.4E-04	179.3
rs11228613	11	69068492	t	g	0.784	0.174	0.021	2.1E-16	1.6E-04	121.5
rs504217	11	72006086	t	c	0.074	0.275	0.034	2.5E-16	1.0E-04	77.1
rs7115331	11	76218590	t	g	0.714	-0.127	0.019	3.9E-11	1.4E-04	107.6
rs11021221	11	95308854	a	t	0.167	-0.188	0.023	6.9E-16	1.4E-04	108.6
rs61909958	11	96151677	c	g	0.812	0.128	0.023	2.2E-08	1.1E-04	81.0
rs360153	11	9762274	t	c	0.417	-0.220	0.018	4.4E-36	2.9E-04	222.6
rs28377357	2	1.13E+08	a	g	0.294	-0.124	0.019	6.0E-11	1.4E-04	107.2
rs62158170	2	1.14E+08	a	g	0.783	0.165	0.021	6.6E-15	1.5E-04	116.2
rs13021015	2	1.27E+08	a	c	0.160	0.152	0.024	2.3E-10	1.1E-04	84.7

rs4954192	2	1.36E+08	t	c	0.387	-0.123	0.018	8.1E-12	1.6E-04	118.5
rs55944332	2	1.46E+08	a	g	0.763	-0.237	0.020	3.3E-31	2.3E-04	178.0
rs12990959	2	1.49E+08	t	c	0.688	-0.127	0.019	1.1E-11	1.5E-04	113.7
rs2444769	2	1.58E+08	a	c	0.795	0.158	0.022	4.8E-13	1.4E-04	107.0
rs7572130	2	1.64E+08	a	g	0.896	-0.180	0.029	4.1E-10	9.2E-05	69.8
rs73029563	2	1.65E+08	c	g	0.455	-0.259	0.017	5.8E-50	3.5E-04	267.4
rs6735275	2	1.74E+08	t	c	0.729	0.123	0.019	2.6E-10	1.3E-04	100.8
rs6715901	2	1.8E+08	a	g	0.496	-0.138	0.017	2.8E-15	1.9E-04	141.7
rs12693302	2	1.83E+08	a	g	0.652	-0.238	0.018	2.2E-39	3.0E-04	224.8
rs7576060	2	1.88E+08	t	c	0.350	-0.102	0.018	2.1E-08	1.3E-04	96.3
rs7592578	2	1.91E+08	t	g	0.194	-0.200	0.022	4.7E-19	1.7E-04	129.8
rs1373780	2	19501029	c	g	0.185	0.125	0.022	2.6E-08	1.0E-04	77.7
rs824523	2	19707855	a	c	0.334	0.123	0.018	2.3E-11	1.5E-04	113.6
rs11692619	2	2.05E+08	t	c	0.361	-0.128	0.018	3.3E-12	1.6E-04	122.9
rs1263671	2	2.08E+08	t	c	0.837	-0.139	0.024	4.7E-09	1.0E-04	79.2
rs4675682	2	2.08E+08	t	c	0.538	-0.141	0.017	4.5E-16	1.9E-04	145.9
rs1035673	2	2.19E+08	t	c	0.397	0.163	0.018	3.0E-20	2.1E-04	162.0
rs13004222	2	2.2E+08	c	g	0.949	0.294	0.039	7.1E-14	7.8E-05	59.3
rs1039897	2	2.2E+08	a	g	0.650	-0.109	0.018	3.3E-09	1.4E-04	102.8
rs10804330	2	2.27E+08	t	c	0.567	0.133	0.018	4.6E-14	1.8E-04	135.7
rs1044822	2	2.31E+08	t	c	0.149	-0.133	0.024	4.1E-08	9.3E-05	70.4
rs4507125	2	2.4E+08	a	c	0.786	-0.124	0.021	3.6E-09	1.1E-04	87.0
rs11687089	2	25082926	t	c	0.583	0.174	0.018	2.8E-23	2.3E-04	176.1
rs1275988	2	26914364	t	c	0.611	-0.295	0.018	1.9E-62	3.8E-04	291.5
rs1468816	2	37595696	a	c	0.772	0.124	0.021	4.6E-09	1.2E-04	89.7
rs11124595	2	37887589	t	g	0.260	0.111	0.020	2.4E-08	1.2E-04	89.1
rs2160236	2	40557276	c	g	0.379	-0.142	0.018	4.3E-15	1.8E-04	137.5
rs76326501	2	43167878	a	c	0.909	0.362	0.031	2.2E-32	1.6E-04	124.6
rs4952668	2	43386568	a	g	0.624	-0.192	0.018	1.1E-26	2.5E-04	187.7
rs2586970	2	55829967	a	g	0.436	-0.149	0.018	1.6E-17	2.0E-04	149.9
rs2421200	2	61711815	t	g	0.488	-0.110	0.017	2.6E-10	1.5E-04	114.1
rs1876490	2	73052351	a	g	0.717	0.136	0.019	1.2E-12	1.5E-04	115.2
rs6546810	2	73389716	t	c	0.648	-0.120	0.018	3.2E-11	1.5E-04	114.1
rs311564	2	86293498	a	g	0.346	-0.133	0.018	4.2E-13	1.7E-04	125.2
rs62155750	2	96491456	a	g	0.693	-0.218	0.020	8.3E-29	2.5E-04	192.1
rs112393817	2	9807226	c	g	0.783	0.116	0.021	3.8E-08	1.1E-04	82.1
rs11923667	3	1.01E+08	a	t	0.407	0.118	0.018	3.1E-11	1.6E-04	118.0
rs28675079	3	1.12E+08	a	g	0.187	-0.144	0.022	8.3E-11	1.2E-04	91.3
rs347585	3	11286220	t	c	0.701	0.151	0.019	1.6E-15	1.7E-04	131.2
rs12152463	3	1.22E+08	t	c	0.425	0.101	0.017	8.0E-09	1.4E-04	102.4
rs4141663	3	1.25E+08	t	c	0.422	-0.150	0.018	1.4E-17	2.0E-04	151.7
rs4077158	3	1.34E+08	t	c	0.471	-0.183	0.017	3.1E-26	2.5E-04	190.1
rs9289557	3	1.38E+08	t	c	0.260	-0.119	0.021	8.7E-09	1.3E-04	95.1
rs6763931	3	1.41E+08	a	g	0.444	0.138	0.017	1.5E-15	1.9E-04	142.2
rs1687295	3	14889756	t	c	0.270	0.206	0.019	3.0E-26	2.2E-04	169.3
rs1527797	3	1.54E+08	t	c	0.740	-0.141	0.020	8.7E-13	1.5E-04	112.7
rs78809139	3	1.55E+08	a	g	0.101	-0.228	0.029	2.6E-15	1.1E-04	86.5

rs78151625	3	1.58E+08	t	c	0.834	-0.187	0.023	1.0E-15	1.4E-04	107.6
rs62234672	3	16592069	a	c	0.175	0.125	0.023	4.9E-08	9.9E-05	75.1
rs16853198	3	1.69E+08	a	g	0.924	0.339	0.033	4.4E-25	1.3E-04	99.3
rs1528293	3	1.69E+08	a	t	0.492	0.276	0.017	1.5E-57	3.8E-04	287.0
rs62294352	3	1.69E+08	t	c	0.216	-0.161	0.022	6.1E-13	1.5E-04	110.0
rs6779368	3	1.85E+08	a	g	0.658	-0.179	0.018	2.3E-22	2.2E-04	167.9
rs147501096	3	1.86E+08	c	g	0.072	-0.196	0.034	9.9E-09	7.2E-05	54.4
rs4244200	3	1.96E+08	c	g	0.280	-0.122	0.019	3.2E-10	1.3E-04	101.8
rs6777317	3	1.97E+08	a	g	0.290	0.125	0.020	1.5E-10	1.4E-04	105.7
rs2643826	3	27562988	t	c	0.451	0.186	0.018	2.8E-26	2.5E-04	191.5
rs7427249	3	37572489	a	g	0.580	-0.110	0.018	4.3E-10	1.5E-04	111.4
rs114714860	3	41882905	c	g	0.168	0.330	0.024	1.4E-44	2.5E-04	192.1
rs6442105	3	48182326	a	g	0.327	-0.249	0.019	3.1E-41	3.0E-04	227.9
rs61018691	3	50538600	c	g	0.135	0.160	0.026	3.4E-10	1.0E-04	77.9
rs1401494	3	53696955	t	c	0.473	-0.138	0.017	1.8E-15	1.9E-04	142.0
rs3772219	3	56771251	a	c	0.681	0.175	0.019	2.9E-21	2.1E-04	158.8
rs11130602	3	57947168	a	g	0.443	0.147	0.018	4.3E-17	2.0E-04	150.7
rs3774702	3	63856870	a	g	0.177	0.147	0.023	1.2E-10	1.2E-04	89.1
rs6795735	3	64705365	t	c	0.411	-0.144	0.018	3.1E-16	1.9E-04	143.1
rs7623706	3	74712754	a	g	0.565	0.098	0.018	2.8E-08	1.3E-04	98.6
rs11923343	3	85668570	a	g	0.360	-0.114	0.018	3.1E-10	1.4E-04	108.9
rs13107325	4	1.03E+08	t	c	0.074	-0.675	0.034	3.7E-88	2.5E-04	192.3
rs12503341	4	1.07E+08	a	g	0.039	-0.299	0.046	9.4E-11	6.2E-05	46.8
rs4245930	4	1.09E+08	a	g	0.632	-0.122	0.018	1.1E-11	1.6E-04	117.6
rs13118687	4	1.11E+08	a	g	0.470	-0.150	0.018	1.4E-17	2.0E-04	154.2
rs66887589	4	1.21E+08	t	c	0.522	-0.161	0.017	1.8E-20	2.2E-04	166.6
rs9286351	4	1.38E+08	a	g	0.581	-0.141	0.018	1.6E-15	1.9E-04	142.4
rs72719149	4	1.44E+08	t	c	0.684	-0.128	0.019	6.3E-12	1.5E-04	114.7
rs13124515	4	1.45E+08	t	c	0.313	-0.105	0.019	2.0E-08	1.2E-04	93.8
rs1123037	4	1.57E+08	a	t	0.477	-0.161	0.017	1.6E-20	2.2E-04	167.1
rs13139571	4	1.57E+08	a	c	0.237	-0.241	0.020	2.3E-32	2.4E-04	181.1
rs1425486	4	1.58E+08	t	c	0.321	-0.133	0.019	1.1E-12	1.6E-04	118.0
rs16896276	4	18015156	a	t	0.263	-0.131	0.020	3.8E-11	1.4E-04	105.1
rs61789369	4	2265295	a	g	0.957	-0.304	0.044	3.1E-12	6.9E-05	52.7
rs28667801	4	26785356	a	t	0.593	-0.162	0.018	1.9E-19	2.2E-04	162.2
rs11721984	4	38343935	t	c	0.453	-0.141	0.018	1.9E-15	1.9E-04	143.0
rs62301873	4	40603821	a	g	0.894	-0.173	0.028	1.1E-09	9.0E-05	68.2
rs11945489	4	56463775	t	c	0.291	-0.139	0.019	4.0E-13	1.6E-04	119.4
rs13152154	4	77417756	t	c	0.729	-0.119	0.020	1.2E-09	1.3E-04	96.4
rs12509595	4	81182554	t	c	0.708	-0.497	0.019	1.6E-148	5.7E-04	428.0
rs72976750	4	86725684	t	c	0.860	-0.172	0.025	7.4E-12	1.1E-04	85.5
rs7694000	4	95324968	a	t	0.539	-0.097	0.018	3.5E-08	1.3E-04	99.5
rs9326869	5	1.12E+08	t	c	0.249	0.110	0.020	4.0E-08	1.1E-04	85.3
rs335170	5	1.22E+08	a	c	0.408	0.113	0.018	1.6E-10	1.5E-04	112.3
rs1582931	5	1.23E+08	a	g	0.475	0.216	0.018	4.5E-35	3.0E-04	224.1
rs17677603	5	1.28E+08	a	g	0.616	-0.200	0.018	3.9E-29	2.6E-04	196.7
rs10069690	5	1279790	t	c	0.258	0.162	0.021	1.4E-14	1.7E-04	123.6

rs11745207	5	1.32E+08	c	g	0.742	0.113	0.020	1.3E-08	1.2E-04	90.2
rs1212061	5	1.42E+08	c	g	0.732	0.128	0.020	7.9E-11	1.4E-04	104.1
rs3776299	5	1.43E+08	a	g	0.456	0.127	0.018	5.1E-13	1.7E-04	128.8
rs78909293	5	1.48E+08	t	c	0.955	0.321	0.043	7.3E-14	7.6E-05	57.1
rs2921604	5	14867948	t	c	0.537	-0.096	0.018	4.5E-08	1.3E-04	99.4
rs3117736	5	1.57E+08	t	c	0.266	0.237	0.020	9.7E-34	2.5E-04	192.3
rs11960210	5	1.58E+08	t	c	0.625	0.247	0.018	3.4E-43	3.2E-04	237.9
rs13358657	5	1.58E+08	a	g	0.867	-0.224	0.026	1.7E-18	1.4E-04	107.3
rs6556384	5	1.58E+08	a	c	0.811	-0.152	0.022	5.9E-12	1.3E-04	96.8
rs114503346	5	1.72E+08	t	c	0.046	-0.268	0.043	3.1E-10	6.5E-05	48.8
rs55993676	5	1.73E+08	t	g	0.292	-0.210	0.019	3.8E-28	2.4E-04	179.7
rs1177764	5	32829975	c	g	0.405	-0.307	0.018	1.4E-67	4.1E-04	307.9
rs10941043	5	33194751	t	g	0.709	-0.127	0.019	2.5E-11	1.4E-04	108.9
rs4645335	5	3704761	a	g	0.336	0.114	0.019	7.0E-10	1.4E-04	106.0
rs1467049	5	42440062	t	g	0.805	0.124	0.022	1.4E-08	1.1E-04	81.3
rs6875967	5	50878292	a	g	0.352	0.134	0.018	1.2E-13	1.7E-04	127.7
rs10054208	5	55688992	t	c	0.362	0.119	0.019	1.5E-10	1.5E-04	114.0
rs12515541	5	57095011	t	g	0.607	0.116	0.018	6.2E-11	1.5E-04	114.8
rs1848510	5	57754005	a	g	0.362	0.126	0.018	4.1E-12	1.6E-04	119.9
rs10062049	5	61553881	t	c	0.136	0.221	0.026	4.5E-18	1.4E-04	106.8
rs2307111	5	75003678	t	c	0.603	-0.174	0.018	1.6E-22	2.3E-04	169.7
rs4704514	5	77820081	t	c	0.283	0.109	0.019	1.7E-08	1.2E-04	91.9
rs62380354	5	89484911	a	c	0.890	0.183	0.029	3.7E-10	9.8E-05	74.2
rs13355146	5	92023661	t	c	0.383	0.122	0.018	6.4E-12	1.6E-04	120.5
rs55770741	5	96220087	t	c	0.561	-0.128	0.018	2.2E-13	1.7E-04	131.4
rs1871190	5	97953719	t	g	0.334	0.108	0.019	6.6E-09	1.3E-04	99.8
rs72613227	6	1.06E+08	a	t	0.873	-0.188	0.029	3.9E-11	1.1E-04	80.1
rs7767235	6	1.16E+08	a	c	0.353	-0.118	0.018	7.9E-11	1.5E-04	112.4
rs509067	6	1.17E+08	t	c	0.414	-0.144	0.018	2.6E-16	1.9E-04	145.1
rs11153730	6	1.19E+08	t	c	0.509	0.155	0.017	2.6E-19	2.1E-04	161.0
rs76785130	6	1.22E+08	a	g	0.980	-0.429	0.066	9.4E-11	4.6E-05	34.4
rs13215166	6	1.27E+08	a	g	0.559	-0.309	0.017	1.8E-70	4.2E-04	317.8
rs9399137	6	1.35E+08	t	c	0.738	0.115	0.020	5.8E-09	1.2E-04	92.3
rs636202	6	1.4E+08	t	c	0.482	0.102	0.017	4.4E-09	1.4E-04	106.2
rs9791312	6	1.43E+08	a	c	0.655	-0.123	0.018	2.9E-11	1.5E-04	115.2
rs62434124	6	1.51E+08	t	c	0.071	-0.485	0.034	7.8E-47	1.8E-04	133.5
rs9478282	6	1.52E+08	t	c	0.112	-0.199	0.028	8.7E-13	1.1E-04	82.2
rs2569882	6	1620147	t	c	0.566	0.120	0.018	4.3E-11	1.6E-04	122.5
rs9365555	6	1.64E+08	a	g	0.674	0.125	0.019	2.0E-11	1.5E-04	114.6
rs11961593	6	1.66E+08	t	c	0.069	-0.316	0.035	1.5E-19	1.1E-04	81.6
rs1322639	6	1.7E+08	a	g	0.777	-0.158	0.021	3.9E-14	1.5E-04	114.3
rs67077402	6	20658978	a	c	0.683	-0.108	0.019	6.3E-09	1.3E-04	97.2
rs6934891	6	22139729	a	g	0.426	0.128	0.018	5.2E-13	1.7E-04	129.6
rs2744133	6	22392260	a	g	0.725	0.144	0.019	1.2E-13	1.6E-04	119.1
rs9467545	6	25638464	a	t	0.843	-0.255	0.024	7.4E-27	1.9E-04	140.4
rs198851	6	26104632	t	g	0.150	0.389	0.024	2.9E-57	2.7E-04	203.6
rs6922353	6	26465768	a	t	0.852	-0.164	0.024	1.4E-11	1.1E-04	86.5

rs389883	6	31947460	t	g	0.686	0.249	0.019	4.8E-40	2.9E-04	220.2
rs115447786	6	34354073	t	c	0.043	0.290	0.046	1.7E-10	6.5E-05	49.1
rs10947786	6	39156410	a	g	0.221	-0.138	0.021	4.6E-11	1.3E-04	98.8
rs6905288	6	43758873	a	g	0.568	0.176	0.018	7.8E-23	2.4E-04	178.4
rs881858	6	43806609	a	g	0.694	0.155	0.019	4.7E-16	1.8E-04	136.2
rs2397060	6	51611470	t	c	0.860	-0.161	0.025	1.5E-10	1.1E-04	79.1
rs1114347	6	51834297	a	g	0.518	-0.179	0.017	3.3E-25	2.5E-04	186.3
rs62413546	6	56012664	t	c	0.085	-0.188	0.032	4.6E-09	8.0E-05	60.5
rs504691	6	72206620	a	c	0.400	-0.118	0.018	3.1E-11	1.6E-04	117.4
rs1984195	6	79657391	a	g	0.488	0.174	0.017	1.4E-23	2.4E-04	178.7
rs9406076	6	8023804	t	c	0.328	0.101	0.019	4.6E-08	1.2E-04	92.7
rs16875357	6	85652904	t	g	0.757	-0.121	0.020	2.7E-09	1.2E-04	91.3
rs3798293	6	97033370	a	g	0.784	-0.133	0.021	2.7E-10	1.2E-04	93.8
rs4556017	7	1.01E+08	t	c	0.852	-0.160	0.025	9.7E-11	1.1E-04	83.3
rs2191046	7	1.08E+08	t	g	0.735	0.118	0.020	1.8E-09	1.3E-04	95.9
rs73033340	7	1195692	a	g	0.964	0.531	0.053	5.1E-24	1.0E-04	72.7
rs11556924	7	1.3E+08	t	c	0.383	-0.181	0.018	1.8E-23	2.4E-04	175.8
rs13237249	7	1.31E+08	t	c	0.398	0.137	0.018	1.0E-14	1.8E-04	136.3
rs75511781	7	1.31E+08	a	g	0.958	-0.372	0.047	2.5E-15	8.3E-05	60.8
rs7800558	7	1.4E+08	t	c	0.578	0.096	0.018	4.5E-08	1.3E-04	97.5
rs1044608	7	1.51E+08	c	g	0.923	-0.202	0.034	2.8E-09	7.9E-05	59.3
rs3918226	7	1.51E+08	t	c	0.081	0.612	0.033	5.3E-77	2.5E-04	188.6
rs310597	7	1.51E+08	a	g	0.631	-0.115	0.018	2.2E-10	1.5E-04	111.6
rs6464165	7	1.51E+08	t	c	0.719	-0.217	0.020	7.3E-29	2.4E-04	182.6
rs1534338	7	1.56E+08	a	g	0.603	-0.114	0.018	1.2E-10	1.5E-04	113.9
rs17432462	7	18548613	t	c	0.623	-0.104	0.018	7.3E-09	1.3E-04	101.2
rs12699415	7	1909479	a	g	0.413	0.124	0.018	3.3E-12	1.6E-04	124.4
rs4507656	7	22156538	c	g	0.693	-0.149	0.020	8.7E-14	1.7E-04	124.3
rs2906152	7	2523003	a	g	0.630	-0.187	0.018	5.5E-25	2.4E-04	181.0
rs7805035	7	25965890	a	t	0.412	0.134	0.018	2.4E-14	1.8E-04	135.2
rs3735533	7	27245893	t	c	0.074	-0.487	0.033	6.3E-49	1.8E-04	139.1
rs6961048	7	27328187	c	g	0.896	-0.273	0.029	1.3E-21	1.4E-04	105.2
rs342977	7	35459888	a	g	0.772	-0.158	0.021	1.7E-14	1.5E-04	115.8
rs2854746	7	45960645	c	g	0.400	0.113	0.018	3.3E-10	1.5E-04	112.0
rs17454517	7	50915776	a	g	0.494	0.122	0.017	2.7E-12	1.7E-04	125.2
rs58407878	7	7260161	a	t	0.150	0.157	0.024	1.2E-10	1.1E-04	83.4
rs1178979	7	72856430	t	c	0.805	0.150	0.022	1.0E-11	1.3E-04	97.2
rs3807101	7	80393418	t	c	0.123	-0.174	0.027	4.6E-11	1.0E-04	78.1
rs1449596	7	96395096	c	g	0.355	-0.109	0.018	1.9E-09	1.4E-04	103.4
rs7788746	7	99612405	t	g	0.669	-0.164	0.018	3.2E-19	2.0E-04	151.4
rs2978098	8	1.02E+08	a	c	0.547	0.155	0.018	1.3E-18	2.1E-04	157.9
rs142449193	8	1.03E+08	t	c	0.046	-0.257	0.043	1.5E-09	6.2E-05	47.0
rs2957468	8	1.06E+08	a	g	0.335	0.138	0.019	8.4E-14	1.7E-04	127.6
rs35091929	8	10693492	t	c	0.397	0.183	0.018	6.5E-25	2.4E-04	182.2
rs722783	8	1.2E+08	a	g	0.222	-0.209	0.021	9.0E-24	2.0E-04	150.4
rs9918907	8	1.25E+08	a	g	0.784	-0.119	0.021	1.6E-08	1.1E-04	83.8
rs7012891	8	1.27E+08	t	c	0.763	-0.139	0.021	1.2E-11	1.4E-04	103.5

rs4909314	8	1.36E+08	a	t	0.395	0.134	0.018	3.4E-14	1.8E-04	133.2
rs4074812	8	1.42E+08	a	g	0.554	-0.134	0.018	2.1E-14	1.8E-04	135.9
rs3802230	8	1.44E+08	a	c	0.545	-0.161	0.017	2.8E-20	2.2E-04	165.6
rs62503324	8	23400615	t	c	0.240	0.203	0.020	2.1E-23	2.0E-04	152.5
rs951914	8	25878995	c	g	0.713	0.190	0.019	5.1E-23	2.1E-04	162.2
rs17832905	8	26038759	a	c	0.072	0.192	0.035	2.8E-08	7.0E-05	53.0
rs17321041	8	26445194	t	c	0.063	0.231	0.036	1.8E-10	7.5E-05	57.0
rs1906672	8	38130025	a	g	0.232	0.140	0.021	8.5E-12	1.4E-04	104.2
rs10087280	8	49391836	a	g	0.832	0.138	0.023	2.5E-09	1.1E-04	80.5
rs4873492	8	51947549	t	c	0.173	0.140	0.023	1.3E-09	1.1E-04	83.3
rs2442618	8	6379832	t	c	0.572	-0.132	0.018	1.2E-13	1.8E-04	133.9
rs11778153	8	64503942	t	c	0.643	0.119	0.018	5.8E-11	1.5E-04	112.6
rs6983239	8	72507296	t	g	0.219	0.116	0.021	3.7E-08	1.1E-04	81.4
rs148401029	8	81386066	a	c	0.035	-0.312	0.049	1.3E-10	5.8E-05	44.1
rs56345595	8	82814156	a	g	0.585	0.133	0.018	5.2E-14	1.8E-04	134.2
rs73276406	8	96021760	c	g	0.146	0.156	0.025	2.0E-10	1.1E-04	81.1
rs4743021	9	1.09E+08	t	c	0.685	-0.108	0.019	2.4E-08	1.3E-04	95.7
rs10980408	9	1.13E+08	t	c	0.964	-0.375	0.048	4.2E-15	7.1E-05	53.8
rs10759697	9	1.17E+08	a	g	0.491	0.131	0.017	3.9E-14	1.8E-04	135.9
rs2133386	9	1.28E+08	a	c	0.433	-0.132	0.018	5.2E-14	1.8E-04	135.0
rs507666	9	1.36E+08	a	g	0.187	-0.285	0.022	2.3E-37	2.4E-04	178.9
rs6271	9	1.37E+08	t	c	0.074	-0.431	0.035	1.7E-34	1.6E-04	121.2
rs11145807	9	1.4E+08	a	g	0.406	0.155	0.018	4.1E-17	2.1E-04	152.0
rs4615669	9	21818674	a	g	0.560	-0.114	0.017	6.1E-11	1.5E-04	117.0
rs10491713	9	2506236	t	g	0.198	-0.122	0.022	2.0E-08	1.1E-04	80.4
rs1243876	9	35693104	t	c	0.701	-0.106	0.019	2.1E-08	1.2E-04	92.7
rs76452347	9	35906471	t	c	0.205	-0.225	0.023	9.4E-23	2.0E-04	152.2
rs12337056	9	628670	t	c	0.176	0.136	0.023	2.2E-09	1.1E-04	82.4
rs11141731	9	89888472	t	c	0.228	-0.126	0.021	1.3E-09	1.2E-04	92.0
rs1332812	9	9350986	a	t	0.647	-0.115	0.018	2.7E-10	1.4E-04	108.9
rs11112548	12	1.06E+08	a	t	0.956	0.274	0.044	5.8E-10	6.4E-05	48.4
rs116063464	12	1.1E+08	a	g	0.060	0.202	0.037	4.7E-08	6.3E-05	47.3
rs7137828	12	1.12E+08	t	c	0.518	-0.503	0.018	4.8E-180	6.9E-04	516.9
rs35443	12	1.16E+08	c	g	0.386	-0.266	0.018	1.2E-50	3.5E-04	262.6
rs7299936	12	1.16E+08	a	g	0.578	0.176	0.018	1.1E-23	2.4E-04	178.7
rs1790123	12	1.24E+08	t	c	0.803	0.199	0.022	6.9E-20	1.7E-04	131.1
rs2271139	12	1.25E+08	a	c	0.286	-0.125	0.019	8.2E-11	1.4E-04	105.7
rs61912333	12	19554817	c	g	0.496	0.119	0.018	1.1E-11	1.6E-04	123.6
rs4306343	12	20190630	a	t	0.279	-0.317	0.019	8.2E-61	3.5E-04	265.5
rs6487076	12	20470857	a	g	0.777	0.174	0.021	8.7E-17	1.7E-04	125.6
rs12229480	12	26472908	t	c	0.723	0.136	0.019	2.1E-12	1.5E-04	113.2
rs1669907	12	42777933	t	g	0.303	0.116	0.019	1.4E-09	1.3E-04	101.6
rs61917655	12	48210787	t	c	0.101	0.225	0.030	3.7E-14	1.1E-04	85.0
rs7967705	12	50511408	t	c	0.380	0.269	0.018	1.5E-51	3.5E-04	264.5
rs7134440	12	53450097	t	c	0.083	0.228	0.032	1.8E-12	9.5E-05	71.9
rs75507123	12	5417856	t	g	0.127	-0.143	0.026	3.9E-08	8.8E-05	66.4
rs6580970	12	54434277	t	c	0.299	-0.166	0.019	4.0E-18	1.9E-04	143.3

rs6581101	12	57136374	a	c	0.604	-0.126	0.018	2.0E-12	1.7E-04	125.5
rs7959649	12	67783108	t	c	0.242	0.117	0.020	8.1E-09	1.2E-04	89.2
rs521033	12	69951428	a	g	0.864	-0.180	0.025	1.1E-12	1.2E-04	88.4
rs710698	12	70369918	a	g	0.587	0.106	0.018	1.9E-09	1.4E-04	105.8
rs7132012	12	8832203	a	g	0.675	0.156	0.019	3.2E-17	1.9E-04	142.9
rs2681485	12	90025622	a	g	0.598	0.295	0.018	1.3E-62	3.9E-04	295.0
rs11108209	12	96109855	t	c	0.907	-0.190	0.030	2.4E-10	8.8E-05	66.9
rs544012	13	1.11E+08	t	g	0.266	0.114	0.020	1.4E-08	1.2E-04	92.6
rs36169093	13	1.14E+08	a	g	0.504	0.123	0.018	5.6E-12	1.7E-04	127.7
rs7321688	13	1.15E+08	a	c	0.233	0.151	0.021	2.0E-13	1.5E-04	111.6
rs682681	13	22294062	t	c	0.334	-0.145	0.019	4.5E-15	1.8E-04	134.4
rs61948065	13	25255052	a	c	0.879	-0.174	0.027	1.2E-10	1.0E-04	76.9
rs7338758	13	30137828	t	c	0.244	0.194	0.020	1.7E-21	2.0E-04	147.3
rs56256111	13	41478963	a	g	0.144	0.193	0.026	2.6E-13	1.3E-04	98.0
rs7992292	13	41968013	a	g	0.824	0.137	0.023	3.2E-09	1.1E-04	82.6
rs9526707	13	51489186	a	g	0.322	-0.122	0.019	6.6E-11	1.5E-04	110.5
rs9563529	13	58316637	t	g	0.204	0.122	0.022	1.4E-08	1.1E-04	82.6
rs3861113	13	72364382	a	c	0.083	0.213	0.032	3.9E-11	8.8E-05	66.2
rs12866098	13	73119617	a	g	0.342	0.103	0.019	2.7E-08	1.3E-04	96.7
rs1215469	13	80707408	a	c	0.230	-0.138	0.021	5.2E-11	1.3E-04	101.6
rs55684003	13	97988689	a	g	0.696	0.122	0.019	1.0E-10	1.4E-04	107.5
rs8014182	14	1.04E+08	t	c	0.132	-0.194	0.026	3.9E-14	1.2E-04	92.6
rs7350752	14	21841154	a	g	0.124	-0.150	0.027	2.0E-08	9.0E-05	68.0
rs17880989	14	23313633	a	g	0.026	0.401	0.059	1.1E-11	5.6E-05	40.3
rs1950500	14	24830850	t	c	0.292	0.140	0.019	2.2E-13	1.6E-04	120.2
rs4424827	14	35110857	t	c	0.567	-0.098	0.018	2.1E-08	1.3E-04	100.3
rs7155504	14	36158828	t	c	0.912	0.229	0.032	5.2E-13	1.0E-04	75.4
rs72683923	14	50735947	t	c	0.979	0.533	0.064	5.0E-17	6.1E-05	45.9
rs35413927	14	53420358	a	g	0.695	-0.127	0.019	1.8E-11	1.5E-04	112.4
rs194742	14	69287483	t	c	0.169	0.128	0.023	3.2E-08	9.9E-05	74.9
rs227426	14	70456664	t	g	0.562	0.112	0.018	1.7E-10	1.5E-04	114.6
rs2239268	14	72469591	a	g	0.701	0.110	0.019	7.4E-09	1.3E-04	95.7
rs4903064	14	73279420	t	c	0.765	0.154	0.021	7.8E-14	1.5E-04	115.4
rs10873612	15	26105602	t	c	0.596	-0.110	0.018	9.5E-10	1.5E-04	109.6
rs11070245	15	40317792	t	g	0.468	-0.129	0.017	1.6E-13	1.8E-04	132.0
rs2925345	15	41311799	t	c	0.468	0.189	0.017	1.6E-27	2.6E-04	195.7
rs2305654	15	42136977	a	c	0.345	0.171	0.019	4.0E-20	2.1E-04	156.5
rs7169864	15	53902901	t	c	0.232	-0.113	0.021	3.4E-08	1.1E-04	83.9
rs28429256	15	66931617	a	g	0.334	0.164	0.019	2.8E-18	2.0E-04	151.2
rs2469141	15	66967398	t	c	0.837	0.135	0.024	1.4E-08	1.0E-04	76.4
rs3743111	15	71587373	a	g	0.613	0.152	0.018	1.6E-17	2.0E-04	149.9
rs11636952	15	75114322	t	c	0.313	0.400	0.019	5.2E-99	4.7E-04	349.6
rs57708073	15	79066653	a	g	0.739	0.191	0.021	4.7E-19	2.0E-04	137.9
rs2627313	15	81006712	t	c	0.446	0.151	0.018	5.9E-18	2.1E-04	153.6
rs17807723	15	90023558	a	g	0.138	-0.176	0.026	7.4E-12	1.1E-04	86.0
rs4932373	15	91429287	a	c	0.674	-0.366	0.019	7.7E-84	4.4E-04	325.9
rs3743369	15	92707569	a	g	0.628	0.104	0.018	6.8E-09	1.3E-04	100.9

rs12906962	15	95312071	t	c	0.677	-0.238	0.019	8.7E-37	2.9E-04	215.8
rs2589218	15	96785017	t	c	0.730	-0.121	0.020	6.9E-10	1.3E-04	98.7
rs77924615	16	20392332	a	g	0.198	-0.316	0.022	3.7E-45	2.8E-04	208.8
rs12596630	16	2065666	t	c	0.091	0.261	0.031	1.0E-16	1.2E-04	86.9
rs9937801	16	21088130	t	c	0.569	0.155	0.017	4.8E-19	2.1E-04	158.5
rs80095680	16	30902353	a	g	0.737	-0.157	0.020	2.8E-15	1.7E-04	126.5
rs917522	16	4097222	t	c	0.885	0.167	0.027	1.0E-09	9.3E-05	70.4
rs12446456	16	4922201	t	c	0.427	-0.181	0.018	4.0E-25	2.4E-04	184.5
rs7192407	16	49783926	t	c	0.472	0.102	0.017	4.5E-09	1.4E-04	105.8
rs62030049	16	50572709	a	g	0.760	0.134	0.021	1.5E-10	1.3E-04	101.5
rs9932220	16	51758116	a	g	0.218	-0.159	0.021	3.8E-14	1.5E-04	112.8
rs12919839	16	56859216	t	c	0.284	-0.110	0.019	1.0E-08	1.2E-04	93.0
rs45474499	16	66914492	t	c	0.047	0.356	0.042	8.5E-18	8.8E-05	66.8
rs28544928	16	69329268	t	g	0.747	0.154	0.020	9.1E-15	1.6E-04	121.6
rs12444212	16	71437689	t	c	0.817	0.129	0.023	1.1E-08	1.1E-04	80.2
rs11859505	16	74195719	a	g	0.420	-0.104	0.018	9.8E-09	1.4E-04	103.8
rs8046697	16	75442144	t	c	0.417	-0.129	0.018	6.1E-13	1.7E-04	130.0
rs12929303	16	81602264	a	g	0.533	0.157	0.017	1.6E-19	2.2E-04	163.0
rs79286081	16	86555837	a	g	0.102	-0.163	0.030	4.8E-08	8.2E-05	61.9
rs908951	16	89697625	t	c	0.437	-0.198	0.018	7.7E-28	2.7E-04	200.0
rs9893005	17	16225506	c	g	0.535	-0.121	0.018	7.9E-12	1.6E-04	124.7
rs12938803	17	19204432	a	c	0.189	-0.160	0.022	8.8E-13	1.3E-04	100.1
rs4362428	17	2090341	a	c	0.409	-0.113	0.018	1.5E-10	1.5E-04	113.4
rs76954792	17	30033514	t	c	0.232	0.121	0.021	5.1E-09	1.2E-04	90.1
rs28661492	17	30609932	t	c	0.202	-0.136	0.022	9.6E-10	1.2E-04	91.2
rs9895032	17	3951946	t	c	0.498	0.107	0.018	1.2E-09	1.5E-04	111.5
rs2239917	17	43165887	t	c	0.425	0.173	0.018	9.7E-23	2.3E-04	176.0
rs55671319	17	43548424	a	g	0.817	0.137	0.023	5.4E-09	1.1E-04	84.2
rs8078510	17	47045862	a	g	0.269	-0.128	0.020	9.8E-11	1.4E-04	104.6
rs9889262	17	47398070	a	t	0.367	0.228	0.018	7.1E-37	2.9E-04	220.5
rs3785837	17	59468942	a	g	0.764	0.145	0.021	9.6E-12	1.4E-04	107.9
rs6504163	17	61545779	t	c	0.624	-0.184	0.018	6.3E-24	2.4E-04	179.5
rs1867624	17	62387091	t	c	0.615	0.141	0.018	2.1E-15	1.8E-04	139.3
rs55868524	17	7170665	a	g	0.611	0.144	0.018	5.5E-16	1.9E-04	142.8
rs1436138	17	75316880	a	g	0.637	0.199	0.018	7.3E-28	2.5E-04	191.5
rs7217916	17	76769434	a	g	0.385	0.111	0.018	5.6E-10	1.4E-04	109.6
rs138420351	17	7700063	t	c	0.016	0.557	0.085	7.1E-11	4.8E-05	35.2
rs74439044	17	7781019	t	c	0.902	-0.350	0.029	1.4E-32	1.7E-04	129.0
rs11077961	17	81012749	a	g	0.632	0.107	0.019	8.5E-09	1.4E-04	102.3
rs11665020	18	10879503	c	g	0.322	-0.142	0.019	2.8E-14	1.7E-04	129.2
rs11664194	18	20021031	a	t	0.460	-0.108	0.018	8.7E-10	1.5E-04	111.4
rs10164193	18	31161426	t	g	0.922	-0.220	0.033	1.9E-11	8.6E-05	65.5
rs11661473	18	42177123	a	g	0.268	0.201	0.020	1.5E-24	2.2E-04	163.6
rs4890499	18	42585761	a	g	0.255	0.113	0.020	1.3E-08	1.2E-04	89.4
rs58693787	18	48141710	a	g	0.754	0.158	0.020	3.8E-15	1.6E-04	122.1
rs1523871	18	51950877	c	g	0.566	-0.119	0.018	1.5E-11	1.6E-04	121.3
rs10048404	18	54578482	t	c	0.369	-0.110	0.018	2.0E-09	1.4E-04	106.2

rs7235890	18	55732115	t	g	0.896	-0.169	0.029	4.1E-09	8.7E-05	65.9
rs1903752	18	7129327	t	c	0.539	-0.099	0.018	3.2E-08	1.3E-04	102.0
rs4891258	18	72995537	a	g	0.683	-0.116	0.019	5.7E-10	1.4E-04	104.6
rs7227492	18	772064	t	c	0.818	0.181	0.023	1.4E-15	1.5E-04	112.3
rs387865	19	11284539	t	c	0.306	-0.106	0.019	3.2E-08	1.2E-04	92.5
rs167479	19	11526765	t	g	0.472	-0.362	0.019	1.7E-82	5.0E-04	340.5
rs73504817	19	17167723	t	c	0.713	0.177	0.019	1.7E-20	2.0E-04	151.3
rs72999033	19	19366632	t	c	0.066	0.279	0.036	5.9E-15	9.4E-05	70.9
rs7257694	19	30314666	t	c	0.400	0.184	0.018	6.3E-25	2.4E-04	182.3
rs1353532	19	31867132	a	t	0.602	-0.127	0.018	1.1E-12	1.7E-04	125.4
rs1433121	19	32591878	t	c	0.691	-0.135	0.019	6.9E-13	1.6E-04	120.3
rs73036520	19	45749484	c	g	0.254	0.156	0.020	1.3E-14	1.6E-04	122.4
rs2548459	19	49209339	t	c	0.481	-0.132	0.018	5.9E-14	1.8E-04	135.0
rs73046792	19	49605705	a	g	0.159	-0.152	0.025	5.9E-10	1.1E-04	83.3
rs10424224	19	7240481	t	c	0.358	0.104	0.018	1.0E-08	1.3E-04	99.8
rs7258382	19	7262569	t	c	0.839	0.262	0.025	3.0E-26	1.9E-04	142.9
rs2009733	19	8398714	a	g	0.500	0.122	0.018	5.1E-12	1.7E-04	123.9
rs693974	20	10557252	t	c	0.604	-0.185	0.018	1.8E-25	2.4E-04	184.1
rs1327235	20	10969030	a	g	0.529	-0.302	0.017	4.8E-68	4.1E-04	313.3
rs6078393	20	11908101	t	g	0.589	0.121	0.018	7.7E-12	1.6E-04	121.3
rs6081555	20	19245723	t	g	0.343	-0.102	0.018	3.1E-08	1.3E-04	95.3
rs2376997	20	30319199	a	c	0.249	-0.139	0.022	1.9E-10	1.4E-04	108.0
rs13042148	20	32298286	t	c	0.154	-0.167	0.024	7.2E-12	1.2E-04	90.7
rs7265695	20	40043096	t	c	0.804	0.197	0.022	2.5E-19	1.7E-04	129.3
rs6031431	20	42795152	a	g	0.538	-0.115	0.018	4.9E-11	1.6E-04	119.0
rs2598	20	47241618	a	g	0.533	0.139	0.018	1.9E-15	1.9E-04	143.8
rs79044887	20	47427831	c	g	0.852	0.243	0.025	4.0E-23	1.7E-04	127.2
rs234623	20	57488964	a	g	0.504	-0.119	0.017	8.6E-12	1.6E-04	123.2
rs6026739	20	57739469	a	t	0.877	-0.503	0.027	1.5E-79	3.0E-04	224.1
rs79208229	20	62516236	t	g	0.088	0.213	0.033	6.5E-11	9.3E-05	70.4
rs35213536	20	62694319	t	g	0.247	0.204	0.021	2.5E-23	2.1E-04	154.2
rs6108168	20	8626271	a	c	0.255	-0.190	0.020	1.1E-21	2.0E-04	150.2
rs1882961	21	16556367	t	c	0.309	0.127	0.019	1.4E-11	1.5E-04	113.1
rs2070527	21	40067495	a	c	0.249	-0.147	0.020	3.1E-13	1.5E-04	113.4
rs12627514	21	44759440	c	g	0.710	-0.216	0.020	2.0E-28	2.4E-04	184.7
rs34487963	21	44838330	a	c	0.019	-0.573	0.071	8.2E-16	5.7E-05	41.6
rs7278003	21	44966069	t	c	0.439	-0.129	0.018	1.8E-13	1.7E-04	132.4
rs5992929	22	18451977	t	c	0.283	0.168	0.019	3.1E-18	1.9E-04	142.4
rs134041	22	28056338	t	c	0.436	0.122	0.018	3.1E-12	1.7E-04	125.1
rs12321	22	29453193	c	g	0.433	-0.149	0.018	1.4E-17	2.0E-04	152.6
rs5753630	22	31861950	a	g	0.562	0.107	0.018	8.8E-10	1.4E-04	109.7

[a] $R^2 = \frac{2 \times EAF \times (1-EAF) \times beta^2}{SD^2}$,where EAF is the effect allele frequency and beta is the effect estimate of the SNP on SBP (Shim 2015, PLoS One;10(4):e0120758).

[b] $F = \frac{R^2 \times (N-2)}{1-R^2}$ where R^2 is the variance of SBP explained by the specific SNP (as explained above) and N the number of individuals in the GWAS analysis (Palmer 2012, Stat Methods Med Res;21(3):223-42).

CHR: chromosome; EAF: effect allele frequency; SE: standard error; SNP: single nucleotide polymorphism.

Table e-3. Single nucleotide polymorphisms (SNP) that fulfilled our selection criteria to be used as proxies for the effects for antihypertensive drug classes.

SNP	Chr	Position (GRCh37/hg19)	Gene	Effect allele	EAF	Beta	SE	p-value	R^2 [a]	F [b]
ACE inhibitors										
rs4291	17	61554194	*ACE*	a	0.615	-0.2839	0.0312	8.65E-20	3.7E-04	275.5
Beta blockers										
rs11196549 [c]	10	115707298	*ADRB1*	a	0.042	0.6884	0.0784	1.58E-18	1.5E-04	113.7
rs460718 [c]	10	115721364	*ADRB1*	a	0.326	-0.2764	0.0324	1.36E-17	3.3E-04	246.7
rs11196597 [c]	10	115788094	*ADRB1*	a	0.133	0.2858	0.0458	4.23E-10	1.8E-04	133.5
rs79850079	10	115790006	*ADRB1*	a	0.031	-0.5804	0.0905	1.45E-10	9.8E-05	72.2
rs17875473 [c]	10	115800294	*ADRB1*	t	0.087	0.3283	0.0552	2.66E-09	1.4E-04	105.9
rs2429511	10	115801253	*ADRB1*	t	0.520	-0.3728	0.0303	7.39E-35	5.1E-04	377.7
rs1801253 [c]	10	115805056	*ADRB1*	c	0.733	0.4626	0.0344	2.84E-41	5.0E-04	366.8
rs4359161 [c]	10	115826508	*ADRB1*	a	0.181	-0.2662	0.0391	9.46E-12	2.2E-04	160.3
Calcium channel blockers										
rs116556102	12	2303850	*CACNA1C*	c	0.983	-0.6853	0.1252	4.42E-08	6.0E-05	43.8
rs2239046 [c]	12	2434419	*CACNA1C*	a	0.681	0.2082	0.0322	9.58E-11	2.5E-04	185.2
rs714277 [c]	12	2514270	*CACNA1C*	t	0.283	0.1986	0.0333	2.38E-09	2.2E-04	165.4
rs2488136 [c]	10	18334521	*CACNB2*	a	0.287	0.2261	0.0334	1.22E-11	2.5E-04	187.9
rs1888693 [c]	10	18440444	*CACNB2*	a	0.344	0.3858	0.0317	4.69E-34	4.8E-04	352.8
rs17604757	10	18442940	*CACNB2*	a	0.932	-0.5022	0.0606	1.12E-16	1.7E-04	126.8
rs12571593	10	18443222	*CACNB2*	a	0.907	-0.3996	0.0521	1.71E-14	1.8E-04	136.2
rs12414844	10	18451994	*CACNB2*	t	0.266	0.2790	0.0342	3.15E-16	3.0E-04	220.8
rs7076319 [c]	10	18459450	*CACNB2*	a	0.733	-0.3210	0.0341	5.07E-21	3.4E-04	254.0
rs17662793	10	18465479	*CACNB2*	a	0.712	0.2363	0.0338	2.65E-12	2.7E-04	196.5
rs10828295	10	18466094	*CACNB2*	a	0.747	-0.3397	0.0349	2.18E-22	3.5E-04	257.6
rs16916922	10	18467744	*CACNB2*	a	0.858	-0.3662	0.0433	2.86E-17	2.4E-04	180.5
rs61278674 [c]	10	18481737	*CACNB2*	a	0.906	-0.3298	0.0540	1.03E-09	1.5E-04	113.6
rs4748444	10	18494482	*CACNB2*	t	0.663	0.1939	0.0327	3.13E-09	2.4E-04	175.1
rs1539680	10	18502889	*CACNB2*	c	0.792	-0.3259	0.0375	3.37E-18	2.9E-04	214.6
rs1779209 [c]	10	18514561	*CACNB2*	t	0.287	0.2736	0.0336	4.23E-16	3.1E-04	224.8
rs1757213	10	18537594	*CACNB2*	a	0.112	0.3084	0.0507	1.15E-09	1.7E-04	124.3
rs10828399 [c]	10	18553968	*CACNB2*	a	0.521	-0.1947	0.0302	1.10E-10	2.7E-04	197.2
rs10828452 [c]	10	18592450	*CACNB2*	a	0.793	0.3046	0.0388	4.20E-15	2.7E-04	202.6
rs17610275	10	18621630	*CACNB2*	t	0.926	0.3868	0.0613	2.87E-10	1.4E-04	106.4
rs10828542 [c]	10	18627285	*CACNB2*	a	0.613	0.1817	0.0311	5.18E-09	2.4E-04	174.8
rs112701401	10	18644811	*CACNB2*	c	0.969	0.5026	0.0921	4.92E-08	8.2E-05	60.3
rs7072277	10	18658707	*CACNB2*	a	0.471	-0.1746	0.0301	6.88E-09	2.4E-04	176.6
rs11013938	10	18669271	*CACNB2*	c	0.255	-0.3265	0.0350	1.17E-20	3.4E-04	251.6
rs12780039 [c]	10	18678987	*CACNB2*	c	0.121	0.2852	0.0470	1.26E-09	1.7E-04	123.1
rs79253631 [c]	10	18694223	*CACNB2*	a	0.986	-0.7774	0.1392	2.32E-08	5.7E-05	41.4
rs112133583 [c]	10	18695681	*CACNB2*	t	0.029	-0.5546	0.0973	1.18E-08	8.8E-05	65.2
rs7909027 [c]	10	18695892	*CACNB2*	t	0.649	-0.3312	0.0318	2.01E-25	4.1E-04	302.8
rs10828662	10	18703097	*CACNB2*	t	0.558	-0.2879	0.0304	2.54E-21	3.9E-04	288.1
rs982003	10	18707296	*CACNB2*	t	0.756	-0.2414	0.0351	6.21E-12	2.4E-04	180.3
rs1325990	10	18707352	*CACNB2*	a	0.470	-0.3873	0.0302	1.09E-37	5.3E-04	391.6
rs11014170 [c]	10	18710991	*CACNB2*	a	0.020	-0.6701	0.1150	5.61E-09	7.4E-05	54.4
rs72786085	10	18713206	*CACNB2*	c	0.079	-0.5309	0.0595	4.46E-19	2.1E-04	156.9

rs10828689	10	18721957	*CACNB2*	c	0.443	-0.3634	0.0304	6.94E-33	4.9E-04	364.1
rs67214975	10	18727251	*CACNB2*	a	0.456	-0.4144	0.0307	1.42E-41	5.7E-04	416.8
rs7923191 [c]	10	18727901	*CACNB2*	a	0.791	-0.3690	0.0376	1.10E-22	3.3E-04	246.5
rs12258967 [c]	10	18727959	*CACNB2*	c	0.704	0.6327	0.0337	1.08E-78	7.2E-04	533.8
rs72786098 [c]	10	18729855	*CACNB2*	a	0.032	-0.5033	0.0883	1.18E-08	8.6E-05	63.5
rs116936375	10	18737135	*CACNB2*	a	0.040	-0.5739	0.0810	1.40E-12	1.2E-04	90.4
rs12256244	10	18750045	*CACNB2*	a	0.626	0.4246	0.0315	2.09E-41	5.5E-04	402.8
rs1998822 [c]	10	18755664	*CACNB2*	a	0.723	-0.1958	0.0343	1.15E-08	2.2E-04	156.6
rs7070582	10	18755942	*CACNB2*	t	0.383	0.2502	0.0317	2.86E-15	3.3E-04	239.8
rs7076100	10	18759537	*CACNB2*	a	0.406	-0.3569	0.0308	5.51E-31	4.7E-04	349.1
rs7076247	10	18759629	*CACNB2*	t	0.388	0.2557	0.0309	1.33E-16	3.3E-04	246.6
rs11014494	10	18780705	*CACNB2*	a	0.492	0.1676	0.0304	3.37E-08	2.3E-04	170.0
rs10828784	10	18788273	*CACNB2*	c	0.663	0.2021	0.0345	4.49E-09	2.5E-04	182.9
rs12416030	10	18789075	*CACNB2*	t	0.796	-0.2088	0.0381	4.32E-08	1.9E-04	136.9
rs12416052	10	18789267	*CACNB2*	t	0.594	0.1987	0.0311	1.59E-10	2.6E-04	194.1
rs4748476	10	18792875	*CACNB2*	t	0.777	0.2166	0.0365	2.89E-09	2.1E-04	152.2
rs150857355 [c]	12	49209340	*CACNB3*	c	0.021	0.9406	0.1122	5.20E-17	1.1E-04	80.3
rs312487	3	53545622	*CACNA1D*	t	0.478	0.2194	0.0307	9.65E-13	3.0E-04	222.2
rs3821843 [c]	3	53558012	*CACNA1D*	a	0.680	0.3373	0.0335	6.56E-24	4.0E-04	296.6
rs9311502	3	53560321	*CACNA1D*	t	0.760	-0.2463	0.0355	3.87E-12	2.5E-04	181.8
rs1547950	3	53568283	*CACNA1D*	t	0.537	-0.2151	0.0307	2.33E-12	2.9E-04	217.0
rs11709630	3	53577164	*CACNA1D*	t	0.637	0.1931	0.0320	1.61E-09	2.5E-04	180.9
rs114987861 [c]	3	53605712	*CACNA1D*	a	0.028	0.5289	0.0958	3.36E-08	8.0E-05	59.1
rs113210396 [c]	3	53612327	*CACNA1D*	t	0.045	-0.4338	0.0770	1.76E-08	1.0E-04	75.7
rs3774475	3	53650483	*CACNA1D*	a	0.417	0.1854	0.0307	1.60E-09	2.5E-04	183.0
rs7340705 [c]	3	53734443	*CACNA1D*	t	0.673	-0.2425	0.0322	4.87E-14	2.9E-04	216.5
rs2633731	3	53738424	*CACNA1D*	t	0.396	-0.1963	0.0309	2.21E-10	2.6E-04	190.6

[a] $R^2 = \frac{2\times EAF\times(1-EAF)\times beta^2}{SD^2}$,*where* EAF is the effect allele frequency and beta is the effect estimate of the SNP on SBP (Shim 2015, PLoS One;10(4):e0120758).

[b] $F = \frac{R^2\times(N-2)}{1-R^2}$ where R^2 is the variance of SBP explained by the specific SNP (as explained above) and *N* the number of individuals in the GWAS analysis (Palmer 2012, Stat Methods Med Res;21(3):223-42).

[C] SNPs also included in the sensitivity analyses of the instruments correlated at a lower LD threshold (r^2<0.1).

ACE: angiotensin converting enzyme; CHR: chromosome; EAF: effect allele frequency; SE: standard error.

Table e-4. Genomic regions of encoding genes and regulatory regions (promoters or enhances) of known antihypertensive drug targets, as identified via GeneHancer. These regions were screened for instrument selection of single nucleotide polymorphisms (SNP) that were associated with systolic blood pressure at genome-wide significance.

Drug	Gene	**Genomic region (GRCh37/hg19)**	**Function**
ACEIs	*ACE*	chr17:61554422-61599205	Encoding gene
ACEIs	*ACE*	chr17:61551058-61556950	Promoter/Enhancer
ACEIs	*ACE*	chr17:61562201-61562303	Promoter/Enhancer
ACEIs	*ACE*	chr17:61508611-61515166	Promoter/Enhancer
ACEIs	*ACE*	chr17:61626418-61630304	Promoter/Enhancer
ACEIs	*ACE*	chr17:61431510-61431613	Enhancer
ACEIs	*ACE*	chr17:62090924-62103850	Promoter/Enhancer
ACEIs	*ACE*	chr17:61497048-61498662	Enhancer
ACEIs	*ACE*	chr17:61505277-61506104	Enhancer
ACEIs	*ACE*	chr17:61689560-61689960	Enhancer
ACEIs	*ACE*	chr17:61594421-61594870	Enhancer
ACEIs	*ACE*	chr17:61502881-61503030	Enhancer
ACEIs	*ACE*	chr17:61656647-61657871	Enhancer
ACEIs	*ACE*	chr17:61500762-61501161	Enhancer
ACEIs	*ACE*	chr17:60855121-60860435	Enhancer
ACEIs	*ACE*	chr17:61574731-61577281	Enhancer
ACEIs	*ACE*	chr17:60972606-60973907	Enhancer
ARBs	*AGTR1*	chr3:148415571-148460795	Encoding gene
ARBs	*AGTR1*	chr3:148415061-148416388	Promoter/Enhancer
ARBs	*AGTR1*	chr3:148366071-148367473	Enhancer
ARBs	*AGTR1*	chr3:148441102-148442130	Enhancer
ARBs	*AGTR1*	chr3:148360847-148362186	Enhancer
ARBs	*AGTR1*	chr3:148360520-148360788	Enhancer
ARBs	*AGTR1*	chr3:148899476-148899525	Enhancer
BBs	*ADRB1*	chr10:115803806-115806667	Enhancer
BBs	*ADRB1*	chr10:115802241-115807338	Promoter/Enhancer
BBs	*ADRB1*	chr10:115716558-115722360	Enhancer
BBs	*ADRB1*	chr10:115706609-115708137	Enhancer
BBs	*ADRB1*	chr10:115824009-115824850	Enhancer
BBs	*ADRB1*	chr10:115548188-115549279	Enhancer
BBs	*ADRB1*	chr10:115833610-115834154	Enhancer
BBs	*ADRB1*	chr10:115704333-115705870	Enhancer
BBs	*ADRB1*	chr10:116441242-116446390	Promoter/Enhancer
BBs	*ADRB1*	chr10:115842035-115843254	Enhancer
BBs	*ADRB1*	chr10:115784258-115788102	Enhancer
BBs	*ADRB1*	chr10:115725160-115725959	Enhancer
BBs	*ADRB1*	chr10:115697701-115697810	Enhancer
BBs	*ADRB1*	chr10:115741347-115744110	Enhancer
BBs	*ADRB1*	chr10:115782141-115782270	Enhancer
BBs	*ADRB1*	chr10:115559441-115559610	Enhancer
BBs	*ADRB1*	chr10:115841821-115841970	Enhancer
BBs	*ADRB1*	chr10:115826224-115827332	Enhancer

BBs	*ADRB1*	chr10:115827575-115828874	Enhancer
BBs	*ADRB1*	chr10:115910708-115912162	Enhancer
BBs	*ADRB1*	chr10:115752960-115753842	Enhancer
BBs	*ADRB1*	chr10:115651401-115651550	Enhancer
BBs	*ADRB1*	chr10:115683621-115684573	Enhancer
BBs	*ADRB1*	chr10:115758960-115759559	Enhancer
BBs	*ADRB1*	chr10:116457630-116458961	Enhancer
BBs	*ADRB1*	chr10:115561321-115561530	Enhancer
BBs	*ADRB1*	chr10:115625638-115626159	Enhancer
BBs	*ADRB1*	chr10:116437959-116439282	Enhancer
BBs	*ADRB1*	chr10:115789385-115790216	Enhancer
BBs	*ADRB1*	chr10:115800822-115802086	Enhancer
BBs	*ADRB1*	chr10:115799814-115800507	Enhancer
CCBs	*CACNA1S*	chr1:201008640-201081694	Encoding gene
CCBs	*CACNA1S*	chr1:201082861-201084129	Promoter/Enhancer
CCBs	*CACNA1S*	chr1:201079942-201082115	Enhancer
CCBs	*CACNA1S*	chr1:201122647-201124394	Promoter/Enhancer
CCBs	*CACNA1S*	chr1:201274996-201282487	Enhancer
CCBs	*CACNA1S*	chr1:201012141-201012270	Enhancer
CCBs	*CACNA1S*	chr1:201263489-201273426	Enhancer
CCBs	*CACNA1S*	chr1:200941201-200941350	Enhancer
CCBs	*CACNA1S*	chr1:201106054-201107629	Enhancer
CCBs	*CACNA1S*	chr1:201056384-201057751	Enhancer
CCBs	*CACNA1S*	chr1:201057880-201061411	Enhancer
CCBs	*CACNA1S*	chr1:201111781-201111890	Enhancer
CCBs	*CACNA1S*	chr1:201063580-201070116	Enhancer
CCBs	*CACNA1S*	chr1:201071203-201078371	Enhancer
CCBs	*CACNA1S*	chr1:201032066-201032442	Enhancer
CCBs	*CACNA1D*	chr3:53528683-53847760	Encoding gene
CCBs	*CACNA1D*	chr3:53526751-53529027	Promoter/Enhancer
CCBs	*CACNA1D*	chr3:53529053-53530500	Promoter/Enhancer
CCBs	*CACNA1D*	chr3:53361979-53363570	Enhancer
CCBs	*CACNA1D*	chr3:53551784-53554581	Enhancer
CCBs	*CACNA1D*	chr3:53558098-53559174	Enhancer
CCBs	*CACNA1D*	chr3:53379987-53382717	Promoter/Enhancer
CCBs	*CACNA1D*	chr3:53859957-53860202	Enhancer
CCBs	*CACNA1D*	chr3:53384627-53385227	Enhancer
CCBs	*CACNA1D*	chr3:53388806-53389878	Enhancer
CCBs	*CACNA1D*	chr3:53354541-53355561	Enhancer
CCBs	*CACNA1D*	chr3:53511553-53514862	Enhancer
CCBs	*CACNA1D*	chr3:53457125-53457810	Enhancer
CCBs	*CACNA1D*	chr3:53539797-53541150	Enhancer
CCBs	*CACNA1D*	chr3:53531730-53534232	Enhancer
CCBs	*CACNA1D*	chr3:53405028-53405227	Enhancer
CCBs	*CACNA1D*	chr3:53559666-53560346	Enhancer
CCBs	*CACNA1D*	chr3:53698941-53699090	Enhancer
CCBs	*CACNA1D*	chr3:53604273-53605793	Enhancer

CCBs	*CACNA1D*	chr3:53742028-53742227	Enhancer
CCBs	*CACNA1D*	chr3:53664741-53664930	Enhancer
CCBs	*CACNA1D*	chr3:53714181-53714330	Enhancer
CCBs	*CACNA1D*	chr3:53664161-53664290	Enhancer
CCBs	*CACNA1D*	chr3:53647109-53647870	Enhancer
CCBs	*CACNA1D*	chr3:53568043-53568300	Enhancer
CCBs	*CACNA1D*	chr3:53707221-53707290	Enhancer
CCBs	*CACNA1D*	chr3:53723669-53724036	Enhancer
CCBs	*CACNA1D*	chr3:53741428-53741627	Enhancer
CCBs	*CACNA1D*	chr3:53787436-53788581	Enhancer
CCBs	*CACNA1D*	chr3:53793207-53793702	Enhancer
CCBs	*CACNA1D*	chr3:53796777-53798080	Enhancer
CCBs	*CACNA1D*	chr3:53798222-53799233	Enhancer
CCBs	*CACNA1D*	chr3:53810959-53811945	Enhancer
CCBs	*CACNA1D*	chr3:53809709-53810892	Enhancer
CCBs	*CACNA1D*	chr3:53817819-53818839	Enhancer
CCBs	*CACNA1D*	chr3:53828693-53829307	Enhancer
CCBs	*CACNA1D*	chr3:53829341-53829430	Enhancer
CCBs	*CACNA1D*	chr3:53838172-53842050	Enhancer
CCBs	*CACNA1D*	chr3:53744028-53744627	Enhancer
CCBs	*CACNA1D*	chr3:53759658-53759955	Enhancer
CCBs	*CACNA1D*	chr3:53843873-53844716	Enhancer
CCBs	*CACNA1D*	chr3:53802862-53803908	Enhancer
CCBs	*CACNA1D*	chr3:53806640-53808437	Enhancer
CCBs	*CACNA1D*	chr3:53799850-53800909	Enhancer
CCBs	*CACNA1D*	chr3:53801465-53802329	Enhancer
CCBs	*CACNA1D*	chr3:53782002-53784852	Enhancer
CCBs	*CACNA1D*	chr3:53777569-53779058	Enhancer
CCBs	*CACNA1D*	chr3:53764428-53765080	Enhancer
CCBs	*CACNA1D*	chr3:53766775-53767435	Enhancer
CCBs	*CACNA2D1*	chr7:81575760-82073114	Encoding gene
CCBs	*CACNA2D1*	chr7:82071056-82074330	Promoter/Enhancer
CCBs	*CACNA2D1*	chr7:82039597-82041108	Enhancer
CCBs	*CACNA2D1*	chr7:82223184-82225724	Enhancer
CCBs	*CACNA2D1*	chr7:82171489-82172576	Enhancer
CCBs	*CACNA2D1*	chr7:82164161-82164310	Enhancer
CCBs	*CACNA2D1*	chr7:82063702-82063952	Enhancer
CCBs	*CACNA2D1*	chr7:81584968-81585322	Enhancer
CCBs	*CACNA2D1*	chr7:81858656-81858971	Enhancer
CCBs	*CACNA2D1*	chr7:82057141-82058356	Enhancer
CCBs	*CACNA2D1*	chr7:82059961-82060110	Enhancer
CCBs	*CACNA2D1*	chr7:82013345-82013689	Enhancer
CCBs	*CACNA2D1*	chr7:81785601-81785750	Enhancer
CCBs	*CACNA2D1*	chr7:81948701-81949880	Enhancer
CCBs	*CACNA2D1*	chr7:81833679-81834445	Enhancer
CCBs	*CACNA2D1*	chr7:81734523-81735564	Enhancer
CCBs	*CACNA2D1*	chr7:82034101-82034250	Enhancer

CCBs	*CACNA2D1*	chr7:81920961-81922045	Enhancer
CCBs	*CACNA2D1*	chr7:81709702-81710455	Enhancer
CCBs	*CACNA2D1*	chr7:81914377-81914723	Enhancer
CCBs	*CACNA2D1*	chr7:81808741-81808930	Enhancer
CCBs	*CACNA2D1*	chr7:81739157-81739440	Enhancer
CCBs	*CACNA2D1*	chr7:81916621-81916770	Enhancer
CCBs	*CACNA2D1*	chr7:81787741-81787827	Enhancer
CCBs	*CACNA2D1*	chr7:81946361-81947530	Enhancer
CCBs	*CACNA2D1*	chr7:81941894-81943091	Enhancer
CCBs	*CACNA2D1*	chr7:81915266-81916265	Enhancer
CCBs	*CACNA2D1*	chr7:81914861-81914999	Enhancer
CCBs	*CACNA2D1*	chr7:81809481-81810272	Enhancer
CCBs	*CACNA2D1*	chr7:81739641-81739790	Enhancer
CCBs	*CACNA2D1*	chr7:81841421-81842618	Enhancer
CCBs	*CACNA2D1*	chr7:81813081-81813230	Enhancer
CCBs	*CACNA2D1*	chr7:81590210-81590492	Enhancer
CCBs	*CACNA2D1*	chr7:81678694-81681663	Enhancer
CCBs	*CACNA2D1*	chr7:81664197-81666962	Enhancer
CCBs	*CACNA2D2*	chr3:50400230-50541675	Encoding gene
CCBs	*CACNA2D2*	chr3:50540431-50541530	Promoter/Enhancer
CCBs	*CACNA2D2*	chr3:50535234-50535293	Promoter
CCBs	*CACNA2D2*	chr3:51420877-51430387	Promoter/Enhancer
CCBs	*CACNA2D2*	chr3:50510252-50511323	Enhancer
CCBs	*CACNA2D2*	chr3:50624952-50631056	Enhancer
CCBs	*CACNA2D2*	chr3:50486750-50489034	Enhancer
CCBs	*CACNA2D2*	chr3:50552257-50555213	Enhancer
CCBs	*CACNA2D2*	chr3:50557041-50557190	Enhancer
CCBs	*CACNA2D2*	chr3:50560739-50564041	Enhancer
CCBs	*CACNA2D2*	chr3:50483138-50484199	Enhancer
CCBs	*CACNA2D2*	chr3:50484337-50485546	Enhancer
CCBs	*CACNA2D2*	chr3:50473601-50473790	Enhancer
CCBs	*CACNA2D2*	chr3:50464650-50466328	Enhancer
CCBs	*CACNA2D2*	chr3:50401766-50403767	Promoter/Enhancer
CCBs	*CACNA2D2*	chr3:50479739-50481683	Enhancer
CCBs	*CACNA2D2*	chr3:50472754-50473413	Enhancer
CCBs	*CACNA2D2*	chr3:50467315-50469064	Enhancer
CCBs	*CACNA2D2*	chr3:50427643-50429192	Enhancer
CCBs	*CACNA2D2*	chr3:50410601-50411305	Enhancer
CCBs	*CACNA2D2*	chr3:50491422-50491591	Enhancer
CCBs	*CACNA2D2*	chr3:50425346-50425879	Enhancer
CCBs	*CACNA2D2*	chr3:50411624-50411893	Enhancer
CCBs	*CACNB1*	chr17:37329709-37353956	Encoding gene
CCBs	*CACNB1*	chr17:37347421-37359080	Promoter/Enhancer
CCBs	*CACNB1*	chr17:37330363-37331930	Enhancer
CCBs	*CACNB1*	chr17:37170207-37173852	Enhancer
CCBs	*CACNB1*	chr17:37392822-37395535	Enhancer
CCBs	*CACNB1*	chr17:37512839-37515343	Enhancer

CCBs	*CACNB1*	chr17:37401081-37401270	Enhancer
CCBs	*CACNB1*	chr17:37401361-37401510	Enhancer
CCBs	*CACNB1*	chr17:37174255-37175468	Enhancer
CCBs	*CACNB1*	chr17:37332121-37332270	Enhancer
CCBs	*CACNB1*	chr17:37333681-37333850	Enhancer
CCBs	*CACNB1*	chr17:37395941-37396110	Enhancer
CCBs	*CACNB1*	chr17:37364621-37367146	Enhancer
CCBs	*CACNB1*	chr17:37427341-37427490	Enhancer
CCBs	*CACNB1*	chr17:37326315-37327866	Enhancer
CCBs	*CACNB1*	chr17:37325454-37325653	Enhancer
CCBs	*CACNB1*	chr17:37328301-37328450	Enhancer
CCBs	*CACNB1*	chr17:37329881-37330030	Enhancer
CCBs	*CACNB1*	chr17:37420122-37420730	Enhancer
CCBs	*CACNB1*	chr17:37566364-37566971	Enhancer
CCBs	*CACNB1*	chr17:37749329-37749495	Enhancer
CCBs	*CACNB1*	chr17:37334901-37335070	Enhancer
CCBs	*CACNB1*	chr17:37334100-37334787	Enhancer
CCBs	*CACNB2*	chr10:18429606-18830798	Encoding gene
CCBs	*CACNB2*	chr10:18428674-18431380	Promoter/Enhancer
CCBs	*CACNB2*	chr10:18688989-18690673	Promoter/Enhancer
CCBs	*CACNB2*	chr10:18629529-18630129	Promoter
CCBs	*CACNB2*	chr10:18549618-18549677	Promoter
CCBs	*CACNB2*	chr10:18452390-18454590	Enhancer
CCBs	*CACNB2*	chr10:18467498-18470172	Enhancer
CCBs	*CACNB2*	chr10:18451281-18451430	Enhancer
CCBs	*CACNB2*	chr10:18493061-18493210	Enhancer
CCBs	*CACNB2*	chr10:18592794-18593791	Enhancer
CCBs	*CACNB2*	chr10:18725955-18730327	Enhancer
CCBs	*CACNB2*	chr10:18473833-18475245	Enhancer
CCBs	*CACNB2*	chr10:18383961-18384110	Enhancer
CCBs	*CACNB2*	chr10:18475337-18476696	Enhancer
CCBs	*CACNB2*	chr10:18406301-18406450	Enhancer
CCBs	*CACNB2*	chr10:18759506-18760793	Enhancer
CCBs	*CACNB2*	chr10:18550812-18550951	Enhancer
CCBs	*CACNB2*	chr10:18775187-18776352	Enhancer
CCBs	*CACNB2*	chr10:18700589-18701887	Enhancer
CCBs	*CACNB2*	chr10:18578722-18579070	Enhancer
CCBs	*CACNB2*	chr10:18433444-18435058	Enhancer
CCBs	*CACNB2*	chr10:18447501-18448892	Enhancer
CCBs	*CACNB2*	chr10:18446528-18447484	Enhancer
CCBs	*CACNB2*	chr10:18534018-18534028	Enhancer
CCBs	*CACNB2*	chr10:18435549-18436688	Enhancer
CCBs	*CACNB2*	chr10:18504001-18504170	Enhancer
CCBs	*CACNB2*	chr10:18486006-18486731	Enhancer
CCBs	*CACNB2*	chr10:18707269-18708134	Enhancer
CCBs	*CACNB2*	chr10:18527921-18528030	Enhancer
CCBs	*CACNB2*	chr10:18522241-18522992	Enhancer

CCBs	*CACNB2*	chr10:18505561-18505670	Enhancer
CCBs	*CACNB2*	chr10:18601402-18602376	Enhancer
CCBs	*CACNB2*	chr10:18436916-18441361	Enhancer
CCBs	*CACNB2*	chr10:18482789-18485238	Enhancer
CCBs	*CACNB2*	chr10:18333406-18335050	Enhancer
CCBs	*CACNB2*	chr10:18790055-18790529	Enhancer
CCBs	*CACNB2*	chr10:18691881-18692030	Enhancer
CCBs	*CACNB2*	chr10:18688181-18688330	Enhancer
CCBs	*CACNB2*	chr10:18567741-18568674	Enhancer
CCBs	*CACNB2*	chr10:18621941-18623677	Enhancer
CCBs	*CACNB2*	chr10:18762421-18762610	Enhancer
CCBs	*CACNB2*	chr10:18760801-18761292	Enhancer
CCBs	*CACNB2*	chr10:18712923-18713793	Enhancer
CCBs	*CACNB2*	chr10:18794315-18795372	Enhancer
CCBs	*CACNB2*	chr10:18745609-18746373	Enhancer
CCBs	*CACNB2*	chr10:18514101-18514250	Enhancer
CCBs	*CACNB2*	chr10:18348427-18349711	Enhancer
CCBs	*CACNB2*	chr10:18583664-18583897	Enhancer
CCBs	*CACNB2*	chr10:18738269-18739296	Enhancer
CCBs	*CACNB2*	chr10:18744481-18744630	Enhancer
CCBs	*CACNB2*	chr10:18744181-18744330	Enhancer
CCBs	*CACNB2*	chr10:18528875-18530208	Enhancer
CCBs	*CACNB2*	chr10:18718065-18718645	Enhancer
CCBs	*CACNB2*	chr10:18751830-18756160	Enhancer
CCBs	*CACNB2*	chr10:18749922-18751579	Enhancer
CCBs	*CACNB2*	chr10:18579741-18580614	Enhancer
CCBs	*CACNB2*	chr10:18567265-18567424	Enhancer
CCBs	*CACNB2*	chr10:18648500-18649316	Enhancer
CCBs	*CACNB2*	chr10:18702212-18703692	Enhancer
CCBs	*CACNB2*	chr10:18703721-18704168	Enhancer
CCBs	*CACNB2*	chr10:18778915-18780659	Enhancer
CCBs	*CACNB2*	chr10:18787912-18789147	Enhancer
CCBs	*CACNB2*	chr10:18792942-18793535	Enhancer
CCBs	*CACNB2*	chr10:18817341-18817490	Enhancer
CCBs	*CACNB2*	chr10:18816741-18816890	Enhancer
CCBs	*CACNB2*	chr10:18740308-18740990	Enhancer
CCBs	*CACNB2*	chr10:18741628-18742721	Enhancer
CCBs	*CACNB2*	chr10:18732026-18732639	Enhancer
CCBs	*CACNB2*	chr10:18723647-18724379	Enhancer
CCBs	*CACNB3*	chr12:49207577-49222726	Encoding gene
CCBs	*CACNB3*	chr12:49203763-49210435	Promoter/Enhancer
CCBs	*CACNB3*	chr12:49211575-49213450	Promoter/Enhancer
CCBs	*CACNB3*	chr12:49448916-49455655	Promoter/Enhancer
CCBs	*CACNB3*	chr12:49108721-49111783	Promoter/Enhancer
CCBs	*CACNB3*	chr12:49522862-49527437	Promoter/Enhancer
CCBs	*CACNB3*	chr12:49657029-49663594	Promoter/Enhancer
CCBs	*CACNB3*	chr12:49180347-49184467	Promoter/Enhancer

CCBs	*CACNB3*	chr12:48225635-48230585	Enhancer
CCBs	*CACNB3*	chr12:49687781-49689222	Promoter/Enhancer
CCBs	*CACNB3*	chr12:49147321-49147490	Enhancer
CCBs	*CACNB3*	chr12:49173178-49175941	Enhancer
CCBs	*CACNB3*	chr12:49159161-49159930	Enhancer
CCBs	*CACNB3*	chr12:49580149-49584425	Promoter/Enhancer
CCBs	*CACNB3*	chr12:49162061-49162210	Enhancer
CCBs	*CACNB3*	chr12:49144534-49144559	Enhancer
CCBs	*CACNB3*	chr12:49160229-49161861	Enhancer
CCBs	*CACNB3*	chr12:49179679-49180345	Enhancer
CCBs	*CACNB3*	chr12:49217901-49218190	Enhancer
CCBs	*CACNB3*	chr12:49267159-49267283	Enhancer
CCBs	*CACNB4*	chr2:152689285-152955593	Encoding gene
CCBs	*CACNB4*	chr2:152954056-152956046	Promoter/Enhancer
CCBs	*CACNB4*	chr2:152830115-152830847	Promoter/Enhancer
CCBs	*CACNB4*	chr2:152918555-152920030	Enhancer
CCBs	*CACNB4*	chr2:152894160-152895545	Enhancer
CCBs	*CACNB4*	chr2:152851019-152853401	Enhancer
CCBs	*CACNB4*	chr2:152910872-152912587	Enhancer
CCBs	*CACNB4*	chr2:152855681-152855810	Enhancer
CCBs	*CACNB4*	chr2:152882049-152882769	Enhancer
CCBs	*CACNB4*	chr2:152779338-152780439	Enhancer
CCBs	*CACNB4*	chr2:152803715-152805070	Enhancer
CCBs	*CACNB4*	chr2:152906315-152907630	Enhancer
CCBs	*CACNB4*	chr2:153308651-153311667	Enhancer
CCBs	*CACNB4*	chr2:152564775-152568412	Enhancer
CCBs	*CACNB4*	chr2:152573345-152574228	Enhancer
CCBs	*CACNB4*	chr2:152700971-152701203	Enhancer
CCBs	*CACNB4*	chr2:153319291-153319615	Enhancer
CCBs	*CACNB4*	chr2:152757130-152757527	Enhancer
CCBs	*CACNB4*	chr2:152860915-152861914	Enhancer
CCBs	*CACNB4*	chr2:152945115-152946314	Enhancer
CCBs	*CACNB4*	chr2:152899381-152899610	Enhancer
CCBs	*CACNB4*	chr2:152036987-152037136	Enhancer
CCBs	*CACNB4*	chr2:152735004-152736098	Enhancer
CCBs	*CACNB4*	chr2:152742479-152743543	Enhancer
CCBs	*CACNB4*	chr2:152887915-152888514	Enhancer
CCBs	*CACNB4*	chr2:152971717-152971973	Enhancer
CCBs	*CACNB4*	chr2:152699171-152700353	Enhancer
CCBs	*CACNB4*	chr2:152714201-152714370	Enhancer
CCBs	*CACNB4*	chr2:152789657-152790028	Enhancer
CCBs	*CACNB4*	chr2:152748315-152748714	Enhancer
CCBs	*CACNB4*	chr2:152774315-152775314	Enhancer
CCBs	*CACNB4*	chr2:152800230-152802596	Enhancer
CCBs	*CACNB4*	chr2:152800061-152800210	Enhancer
CCBs	*CACNG1*	chr17:65040652-65052913	Encoding gene
CCBs	*CACNG1*	chr17:65038563-65044391	Promoter/Enhancer

CCBs	*CACNG1*	chr17:65239492-65244198	Promoter/Enhancer
CCBs	*CACNG1*	chr17:65049530-65051511	Enhancer
CCBs	*CACNG1*	chr17:65106597-65109013	Enhancer
CCBs	*CACNG1*	chr17:65090722-65092662	Enhancer
CCBs	*CACNG1*	chr17:65253360-65257469	Enhancer
CCBs	*CACNG1*	chr17:65154098-65154970	Enhancer
CCBs	*CACNG1*	chr17:65276135-65278586	Enhancer
CCBs	*CACNG1*	chr17:65052236-65053547	Enhancer
CCBs	*CACNG1*	chr17:65234500-65237415	Enhancer
CCBs	*CACNG1*	chr17:65045559-65048695	Enhancer
CCBs	*CACNG1*	chr17:65103906-65106201	Enhancer
CCBs	*CACNG1*	chr17:65211601-65212541	Enhancer
CCBs	*CACNG1*	chr17:65072516-65072985	Enhancer
CCBs	*CACNG1*	chr17:65055208-65056211	Enhancer
TDs	*SLC12A3*	chr16:56899119-56949762	Encoding gene
TDs	*SLC12A3*	chr16:56899070-56899129	Promoter
TDs	*SLC12A3*	chr16:56964403-56974458	Promoter/Enhancer
TDs	*SLC12A3*	chr16:56839748-56841942	Enhancer
TDs	*SLC12A3*	chr16:56834256-56834865	Enhancer
TDs	*SLC12A3*	chr16:56893506-56897709	Enhancer
TDs	*SLC12A3*	chr16:56903512-56904197	Enhancer
TDs	*SLC12A3*	chr16:56901241-56901712	Enhancer
TDs	*SLC12A3*	chr16:56905112-56905513	Enhancer
TDs	*SLC12A3*	chr16:56948424-56953992	Enhancer
TDs	*SLC12A3*	chr16:56942554-56948010	Enhancer
TDs	*SLC12A3*	chr16:56908210-56909649	Enhancer
TDs	*SLC12A3*	chr16:56932921-56933070	Enhancer
TDs	*SLC12A3*	chr16:56922841-56922990	Enhancer
CCBs	*CACNA1C*	chr12:2079952-2807115	Encoding gene
CCBs	*CACNA1C*	chr12:2161566-2164566	Promoter/Enhancer
CCBs	*CACNA1C*	chr12:2081194-2082566	Enhancer
CCBs	*CACNA1C*	chr12:2079295-2081006	Enhancer
CCBs	*CACNA1C*	chr12:2691972-2692849	Enhancer
CCBs	*CACNA1C*	chr12:2143967-2144936	Enhancer
CCBs	*CACNA1C*	chr12:2183654-2186279	Enhancer
CCBs	*CACNA1C*	chr12:2372777-2379299	Enhancer
CCBs	*CACNA1C*	chr12:2216896-2226195	Enhancer
CCBs	*CACNA1C*	chr12:1767798-1773105	Enhancer
CCBs	*CACNA1C*	chr12:2161154-2161552	Enhancer
CCBs	*CACNA1C*	chr12:2419663-2421680	Enhancer
CCBs	*CACNA1C*	chr12:2228156-2230922	Enhancer
CCBs	*CACNA1C*	chr12:2206541-2209473	Enhancer
CCBs	*CACNA1C*	chr12:2364427-2364812	Enhancer
CCBs	*CACNA1C*	chr12:2444050-2449090	Enhancer
CCBs	*CACNA1C*	chr12:2181896-2183390	Enhancer
CCBs	*CACNA1C*	chr12:2210028-2211778	Enhancer
CCBs	*CACNA1C*	chr12:2200243-2201838	Enhancer

CCBs	*CACNA1C*	chr12:2112379-2114467	Promoter/Enhancer
CCBs	*CACNA1C*	chr12:2101946-2105129	Enhancer
CCBs	*CACNA1C*	chr12:2044799-2047397	Enhancer
CCBs	*CACNA1C*	chr12:2118861-2119010	Enhancer
CCBs	*CACNA1C*	chr12:2121501-2121650	Enhancer
CCBs	*CACNA1C*	chr12:2048444-2049910	Enhancer
CCBs	*CACNA1C*	chr12:2122654-2123350	Enhancer
CCBs	*CACNA1C*	chr12:2075648-2076741	Enhancer
CCBs	*CACNA1C*	chr12:2058121-2060041	Enhancer
CCBs	*CACNA1C*	chr12:2055375-2057136	Enhancer
CCBs	*CACNA1C*	chr12:2120201-2121450	Enhancer
CCBs	*CACNA1C*	chr12:2117981-2118130	Enhancer
CCBs	*CACNA1C*	chr12:2173006-2173941	Enhancer
CCBs	*CACNA1C*	chr12:2082932-2084071	Enhancer
CCBs	*CACNA1C*	chr12:2069591-2073188	Enhancer
CCBs	*CACNA1C*	chr12:2089853-2091243	Enhancer
CCBs	*CACNA1C*	chr12:2125886-2127966	Enhancer
CCBs	*CACNA1C*	chr12:2166481-2166670	Enhancer
CCBs	*CACNA1C*	chr12:2466656-2469074	Enhancer
CCBs	*CACNA1C*	chr12:2403410-2405816	Enhancer
CCBs	*CACNA1C*	chr12:2356637-2359010	Enhancer
CCBs	*CACNA1C*	chr12:2057237-2057501	Enhancer
CCBs	*CACNA1C*	chr12:2431967-2434188	Enhancer
CCBs	*CACNA1C*	chr12:2352323-2354776	Enhancer
CCBs	*CACNA1C*	chr12:2365074-2366654	Enhancer
CCBs	*CACNA1C*	chr12:2391640-2398088	Enhancer
CCBs	*CACNA1C*	chr12:2450181-2451861	Enhancer
CCBs	*CACNA1C*	chr12:2164767-2165166	Enhancer
CCBs	*CACNA1C*	chr12:2277277-2279377	Enhancer
CCBs	*CACNA1C*	chr12:2271349-2274081	Enhancer
CCBs	*CACNA1C*	chr12:2322103-2323450	Enhancer
CCBs	*CACNA1C*	chr12:2110661-2111403	Enhancer
CCBs	*CACNA1C*	chr12:2288839-2290765	Enhancer
CCBs	*CACNA1C*	chr12:2347936-2349143	Enhancer
CCBs	*CACNA1C*	chr12:2415078-2416557	Enhancer
CCBs	*CACNA1C*	chr12:2306940-2308560	Enhancer
CCBs	*CACNA1C*	chr12:2292498-2294028	Enhancer
CCBs	*CACNA1C*	chr12:2453367-2455170	Enhancer
CCBs	*CACNA1C*	chr12:2084572-2086775	Enhancer
CCBs	*CACNA1C*	chr12:2087127-2087591	Enhancer
CCBs	*CACNA1C*	chr12:2095160-2096573	Enhancer
CCBs	*CACNA1C*	chr12:2139487-2143377	Enhancer
CCBs	*CACNA1C*	chr12:2175103-2177148	Enhancer
CCBs	*CACNA1C*	chr12:2262602-2264090	Enhancer
CCBs	*CACNA1C*	chr12:2193334-2194490	Enhancer
CCBs	*CACNA1C*	chr12:2409918-2412424	Enhancer
CCBs	*CACNA1C*	chr12:2158809-2159784	Enhancer

CCBs	*CACNA1C*	chr12:2359329-2360982	Enhancer
CCBs	*CACNA1C*	chr12:2428657-2429566	Enhancer
CCBs	*CACNA1C*	chr12:2400461-2400630	Enhancer
CCBs	*CACNA1C*	chr12:2119384-2120057	Enhancer
CCBs	*CACNA1C*	chr12:2203341-2204336	Enhancer
CCBs	*CACNA1C*	chr12:2170143-2171251	Enhancer
CCBs	*CACNA1C*	chr12:2281781-2281930	Enhancer
CCBs	*CACNA1C*	chr12:2260436-2262389	Enhancer
CCBs	*CACNA1C*	chr12:2302013-2306687	Enhancer
CCBs	*CACNA1C*	chr12:2368400-2369290	Enhancer
CCBs	*CACNA1C*	chr12:2442171-2442390	Enhancer
CCBs	*CACNA1C*	chr12:2270781-2270930	Enhancer
CCBs	*CACNA1C*	chr12:2339165-2339784	Enhancer
CCBs	*CACNA1C*	chr12:2238381-2238550	Enhancer
CCBs	*CACNA1C*	chr12:2268934-2269887	Enhancer
CCBs	*CACNA1C*	chr12:2254615-2256378	Enhancer
CCBs	*CACNA1C*	chr12:2342039-2342990	Enhancer
CCBs	*CACNA1C*	chr12:2428367-2428566	Enhancer
CCBs	*CACNA1C*	chr12:2427157-2427717	Enhancer
CCBs	*CACNA1C*	chr12:2398761-2400405	Enhancer
CCBs	*CACNA1C*	chr12:2335414-2336748	Enhancer
CCBs	*CACNA1C*	chr12:2167767-2169880	Enhancer
CCBs	*CACNA1C*	chr12:2148495-2149324	Enhancer
CCBs	*CACNA1C*	chr12:2138706-2139344	Enhancer
CCBs	*CACNA1C*	chr12:2171351-2172258	Enhancer
CCBs	*CACNA1C*	chr12:2245167-2245366	Enhancer
CCBs	*CACNA1C*	chr12:2246831-2248330	Enhancer
CCBs	*CACNA1C*	chr12:2280498-2280632	Enhancer
CCBs	*CACNA1C*	chr12:2281401-2281550	Enhancer
CCBs	*CACNA1C*	chr12:2265561-2265750	Enhancer
CCBs	*CACNA1C*	chr12:2407617-2408956	Enhancer
CCBs	*CACNA1C*	chr12:2192366-2192566	Enhancer
CCBs	*CACNA1C*	chr12:2274261-2274410	Enhancer
CCBs	*CACNA1C*	chr12:2281201-2281350	Enhancer
CCBs	*CACNA1C*	chr12:2177211-2178762	Enhancer
CCBs	*CACNA1C*	chr12:2442567-2442967	Enhancer
CCBs	*CACNA1C*	chr12:2441541-2441690	Enhancer
CCBs	*CACNA1C*	chr12:2451967-2452766	Enhancer
CCBs	*CACNA1C*	chr12:2416801-2416950	Enhancer
CCBs	*CACNA1C*	chr12:2323730-2325500	Enhancer
CCBs	*CACNA1C*	chr12:2349249-2351414	Enhancer
CCBs	*CACNA1C*	chr12:2380803-2386142	Enhancer
CCBs	*CACNA1C*	chr12:2336889-2337604	Enhancer
CCBs	*CACNA1C*	chr12:2457223-2458492	Enhancer
CCBs	*CACNA1C*	chr12:2249789-2251721	Enhancer
CCBs	*CACNA1C*	chr12:2294781-2295709	Enhancer
CCBs	*CACNA1C*	chr12:2186647-2187626	Enhancer

CCBs	*CACNA1C*	chr12:2438360-2439517	Enhancer
CCBs	*CACNA1C*	chr12:2407095-2407603	Enhancer
CCBs	*CACNA1C*	chr12:2386355-2387332	Enhancer
CCBs	*CACNA1C*	chr12:2328750-2331231	Enhancer
CCBs	*CACNA1C*	chr12:2400774-2402962	Enhancer
CCBs	*CACNA1C*	chr12:2245404-2246734	Enhancer
CCBs	*CACNA1C*	chr12:2274438-2275487	Enhancer
CCBs	*CACNA1C*	chr12:2195393-2196970	Enhancer
CCBs	*CACNA1C*	chr12:2233400-2237679	Enhancer
CCBs	*CACNA1C*	chr12:2283997-2286741	Enhancer
CCBs	*CACNA1C*	chr12:2287176-2288132	Enhancer
CCBs	*CACNA1C*	chr12:2258324-2259667	Enhancer
CCBs	*CACNA1C*	chr12:2343203-2345601	Enhancer
CCBs	*CACNA1C*	chr12:2299019-2299959	Enhancer
CCBs	*CACNA1C*	chr12:2412745-2414119	Enhancer
CCBs	*CACNA1C*	chr12:2188468-2189111	Enhancer
CCBs	*CACNA1C*	chr12:2434584-2435528	Enhancer
CCBs	*CACNA1C*	chr12:2316947-2318292	Enhancer
CCBs	*CACNA1C*	chr12:2333218-2334248	Enhancer
CCBs	*CACNA1C*	chr12:2331290-2332420	Enhancer
CCBs	*CACNA1C*	chr12:2440488-2441051	Enhancer
CCBs	*CACNA1C*	chr12:2320137-2320914	Enhancer
CCBs	*CACNA1C*	chr12:2326691-2328736	Enhancer
CCBs	*CACNA1C*	chr12:2649836-2650311	Enhancer
CCBs	*CACNA1C*	chr12:2506981-2507050	Enhancer
CCBs	*CACNA1C*	chr12:2659879-2661404	Enhancer
CCBs	*CACNA1C*	chr12:2515793-2516578	Enhancer
CCBs	*CACNA1C*	chr12:2522460-2525715	Enhancer
CCBs	*CACNA1C*	chr12:2526621-2527392	Enhancer
CCBs	*CACNA1C*	chr12:2695181-2696063	Enhancer
CCBs	*CACNA1C*	chr12:2543714-2545518	Enhancer
CCBs	*CACNA1C*	chr12:2721754-2724057	Enhancer
CCBs	*CACNA1C*	chr12:2547538-2548942	Enhancer
CCBs	*CACNA1C*	chr12:2497459-2500942	Enhancer
CCBs	*CACNA1C*	chr12:2733321-2734829	Enhancer
CCBs	*CACNA1C*	chr12:2555271-2555641	Enhancer
CCBs	*CACNA1C*	chr12:2558507-2559370	Enhancer
CCBs	*CACNA1C*	chr12:2649619-2649812	Enhancer
CCBs	*CACNA1C*	chr12:2736852-2737810	Enhancer
CCBs	*CACNA1C*	chr12:2504526-2506285	Enhancer
CCBs	*CACNA1C*	chr12:2740641-2741724	Enhancer
CCBs	*CACNA1C*	chr12:2511126-2512729	Enhancer
CCBs	*CACNA1C*	chr12:2512831-2514124	Enhancer
CCBs	*CACNA1C*	chr12:2565222-2568247	Enhancer
CCBs	*CACNA1C*	chr12:2749071-2750838	Enhancer
CCBs	*CACNA1C*	chr12:2518209-2521853	Enhancer
CCBs	*CACNA1C*	chr12:2463531-2463940	Enhancer

CCBs	*CACNA1C*	chr12:2792481-2792530	Enhancer
CCBs	*CACNA1C*	chr12:2792101-2792350	Enhancer
CCBs	*CACNA1C*	chr12:2791561-2791670	Enhancer
CCBs	*CACNA1C*	chr12:2791761-2791970	Enhancer
CCBs	*CACNA1C*	chr12:2473710-2474314	Enhancer
CCBs	*CACNA1C*	chr12:2799725-2801764	Promoter/Enhancer
CCBs	*CACNA1C*	chr12:2603721-2603850	Enhancer
CCBs	*CACNA1C*	chr12:2724729-2726058	Enhancer
CCBs	*CACNA1C*	chr12:2500386-2500884	Enhancer
CCBs	*CACNA1C*	chr12:2552550-2553765	Enhancer
CCBs	*CACNA1C*	chr12:2561981-2563825	Enhancer
CCBs	*CACNA1C*	chr12:2559627-2561813	Enhancer
CCBs	*CACNA1C*	chr12:2488061-2488210	Enhancer
CCBs	*CACNA1C*	chr12:2531341-2532583	Enhancer
CCBs	*CACNA1C*	chr12:2537218-2542473	Enhancer
CCBs	*CACNA1C*	chr12:2489067-2491096	Enhancer
CCBs	*CACNA1C*	chr12:2471978-2473410	Enhancer
CCBs	*CACNA1C*	chr12:2608221-2608370	Enhancer
CCBs	*CACNA1C*	chr12:2613061-2613207	Enhancer
CCBs	*CACNA1C*	chr12:2610308-2611812	Enhancer
CCBs	*CACNA1C*	chr12:2481105-2484809	Enhancer
CCBs	*CACNA1C*	chr12:2486081-2486390	Enhancer
CCBs	*CACNA1C*	chr12:2491894-2495082	Enhancer

ACEI: angiotensin converting enzyme inhibitors; ARB: angiotensin II receptor blockers; BB: beta blockers; CCB: calcium channel blockers; TD: thiazide diuretics.

Table e-5. Sensitivity analyses for the Mendelian randomization associations between genetically determined systolic and diastolic blood pressure and risk of stroke and stroke subtypes.

Outcome	SBP (10 mm Hg increment)			DBP (5 mm Hg increment)		
Any stroke	**OR**	**95%CI**	**p**	**OR**	**95%CI**	**p**
IVW (primary analysis)	1.39	(1.33-1.44)	1.9E-60	1.27	(1.23-1.32)	1.2E-42
MR Egger	1.54	(1.40-1.71)	6.0E-17	1.36	(1.25-1.48)	5.2E-13
Egger Intercept	1.00	(0.99-1.00)	0.056	1.00	(1.00-1.00)	0.136
Weighted median	1.42	(1.35-1.50)	2.0E-38	1.28	(1.22-1.34)	1.3E-27
Weighted modal	1.44	(1.30-1.60)	1.2E-11	1.32	(1.21-1.44)	9.2E-10
IVW after exclusion of MR-PRESSO outliers	1.38	(1.33-1.43)	1.2E-63	1.28	(1.24-1.32)	9.2E-54
IVW restricted in Europeans	1.37	(1.32-1.43)	4.5E-47	1.26	(1.21-1.31)	1.8E-35
IVW based on UKB summary statistics not adjusted for antihypertensive medication use or BMI	1.43	(1.36-1.50)	2.4E-51	1.32	(1.26-1.37)	8.5E-38
Ischemic stroke	**OR**	**95%CI**	**p**	**OR**	**95%CI**	**p**
IVW (primary analysis)	1.41	(1.35-1.47)	1.3E-53	1.28	(1.24-1.33)	2.6E-40
MR Egger	1.56	(1.40-1.74)	5.9E-16	1.40	(1.28-1.53)	3.0E-13
Egger Intercept	1.00	(0.99-1.00)	0.134	1.00	(1.00-1.00)	0.134
Weighted median	1.45	(1.37-1.54)	4.7E-38	1.30	(1.25-1.36)	2.0E-33
Weighted modal	1.51	(1.35-1.69)	5.6E-13	1.37	(1.22-1.53)	1.0E-07
IVW after exclusion of MR-PRESSO outliers	1.40	(1.35-1.46)	5.9E-59	1.28	(1.24-1.32)	1.2E-47
IVW restricted in Europeans	1.40	(1.34-1.47)	3.6E-49	1.27	(1.22-1.32)	3.6E-33
IVW based on UKB summary statistics not adjusted for antihypertensive medication use or BMI	1.45	(1.38-1.52)	6.4E-50	1.32	(1.26-1.38)	6.5E-35
Large artery stroke	**OR**	**95%CI**	**p**	**OR**	**95%CI**	**p**
IVW (primary analysis)	1.68	(1.54-1.84)	5.2E-30	1.34	(1.25-1.44)	1.0E-14
MR Egger	1.69	(1.35-2.12)	5.2E-06	1.41	(1.18-1.69)	1.7E-04
Egger Intercept	1.00	(0.99-1.01)	0.999	1.00	(1.00-1.00)	0.505
Weighted median	1.70	(1.48-1.95)	1.0E-13	1.37	(1.24-1.52)	1.3E-09
Weighted modal	1.73	(1.30-2.29)	1.5E-04	1.28	(0.94-1.74)	1.2E-01
IVW after exclusion of MR-PRESSO outliers	1.67	(1.53-1.82)	2.1E-31	1.36	(1.26-1.46)	2.4E-17
IVW restricted in Europeans	1.81	(1.63-2.00)	7.1E-29	1.35	(1.24-1.47)	2.0E-11
IVW based on UKB summary statistics not adjusted for antihypertensive medication use or BMI	1.81	(1.64-2.00)	4.5E-32	1.38	(1.27-1.51)	3.1E-13
Cardioembolic stroke	**OR**	**95%CI**	**p**	**OR**	**95%CI**	**p**
IVW (primary analysis)	1.24	(1.16-1.34)	9.9E-09	1.17	(1.10-1.24)	2.7E-06
MR Egger	1.49	(1.24-1.80)	2.3E-05	1.33	(1.14-1.56)	3.2E-04
Egger Intercept	0.99	(0.99-1.00)	0.066	1.00	(0.99-1.00)	0.096
Weighted median	1.30	(1.17-1.45)	1.1E-06	1.24	(1.13-1.36)	2.3E-06
Weighted modal	1.31	(0.99-1.75)	0.060	1.54	(1.26-1.89)	3.5E-05
IVW after exclusion of MR-PRESSO outliers	1.25	(1.17-1.35)	1.0E-09	1.18	(1.11-1.25)	1.8E-07
IVW restricted in Europeans	1.21	(1.12-1.31)	1.2E-06	1.15	(1.08-1.23)	4.4E-05
IVW based on UKB summary statistics not adjusted for antihypertensive medication use or BMI	1.27	(1.17-1.38)	8.8E-09	1.19	(1.10-1.28)	8.0E-06
Small vessel stroke	**OR**	**95%CI**	**p**	**OR**	**95%CI**	**p**
IVW (primary analysis)	1.47	(1.36-1.58)	3.5E-22	1.36	(1.27-1.45)	7.8E-19
MR Egger	1.44	(1.19-1.76)	2.7E-04	1.39	(1.17-1.64)	1.3E-04
Egger Intercept	1.00	(1.00-1.01)	0.739	1.00	(1.00-1.00)	0.739
Weighted median	1.54	(1.37-1.73)	9.5E-14	1.43	(1.31-1.56)	9.5E-16

Weighted modal	1.74	(1.38-2.18)	1.9E-06	1.54	(1.24-1.92)	9.8E-05
IVW after exclusion of MR-PRESSO outliers	1.47	(1.36-1.58)	7.1E-23	1.38	(1.29-1.47)	4.5E-23
IVW restricted in Europeans	1.57	(1.43-1.72)	1.3E-21	1.39	(1.28-1.50)	1.5E-15
IVW based on UKB summary statistics not adjusted for antihypertensive medication use or BMI	1.52	(1.39-1.66)	4.8E-21	1.41	(1.30-1.53)	5.1E-17
Intracerebral hemorrhage	**OR**	**95%CI**	**p**	**OR**	**95%CI**	**p**
IVW (primary analysis)	1.41	(1.11-1.79)	8.3E-03	1.29	(1.05-1.57)	0.019
MR Egger	1.49	(0.81-2.74)	0.201	1.22	(0.72-2.07)	0.457
Egger Intercept	1.00	(0.98-1.02)	0.824	1.00	(0.99-1.01)	0.841
Weighted median	1.49	(1.09-2.03)	0.012	1.31	(1.00-1.71)	0.047
Weighted modal	1.56	(0.80-3.04)	0.192	1.81	(0.89-3.71)	0.103
IVW after exclusion of MR-PRESSO outliers	1.41	(1.11-1.79)	0.005	1.29	(1.05-1.57)	0.014
IVW based on UKB summary statistics not adjusted for antihypertensive medication use or BMI	1.43	(1.09-1.87)	0.009	1.36	(1.07-1.73)	0.013
Lobar intracerebral hemorrhage	**OR**	**95%CI**	**p**	**OR**	**95%CI**	**p**
IVW (primary analysis)	1.04	(0.77-1.40)	0.389	0.97	(0.76-1.25)	0.391
MR Egger	1.03	(0.48-2.21)	0.936	1.02	(0.53-1.97)	0.960
Egger Intercept	1.00	(0.98-1.02)	0.999	1.00	(0.99-1.01)	0.868
Weighted median	1.26	(0.85-1.87)	0.259	1.12	(0.77-1.63)	0.563
Weighted modal	1.19	(0.62-2.31)	0.596	1.14	(0.62-2.08)	0.671
IVW after exclusion of MR-PRESSO outliers	1.04	(0.77-1.40)	0.819	0.97	(0.76-1.25)	0.837
IVW based on UKB summary statistics not adjusted for antihypertensive medication use or BMI	1.06	(0.79-1.48)	0.738	0.98	(0.71-1.33)	0.889
Deep intracerebral hemorrhage	**OR**	**95%CI**	**p**	**OR**	**95%CI**	**p**
IVW (primary analysis)	1.73	(1.30-2.32)	8.3E-04	1.54	(1.21-1.97)	8.2E-04
MR Egger	1.86	(0.89-3.87)	0.097	1.24	(0.66-2.34)	0.506
Egger Intercept	1.00	(0.98-1.02)	0.856	1.00	(0.99-1.02)	0.505
Weighted median	1.59	(1.01-2.48)	0.043	1.45	(0.99-2.12)	0.055
Weighted modal	1.15	(0.38-3.52)	0.800	1.40	(0.49-3.96)	0.530
IVW after exclusion of MR-PRESSO outliers	1.77	(1.33-2.35)	8.2E-05	1.54	(1.21-1.97)	4.3E-04
IVW based on UKB summary statistics not adjusted for antihypertensive medication use or BMI	1.79	(1.30-2.47)	4.1E-04	1.66	(1.24-2.23)	6.6E-04
WMH volume	**β**	**95%CI**	**p**	**β**	**95%CI**	**p**
IVW (primary analysis)	0.101	(0.053, 0.149)	3.8E-05	0.107	(0.064, 0.150)	1.1E-06
MR Egger	0.159	(0.034, 0.284)	0.013	0.113	(0.007, 0.218)	0.036
Egger Intercept	-0.002	(-0.006, 0.002)	0.323	0.000	(-0.004, 0.004)	0.909
Weighted median	0.130	(0.057, 0.202)	1.5E-06	0.115	(0.051, 0.179)	4.9E-04
Weighted modal	0.211	(-0.026, 0.447)	0.081	0.172	(-0.011, 0.355)	0.065
IVW after exclusion of MR-PRESSO outliers	0.108	(0.059, 0.156)	1.6E-05	0.110	(0.068, 0.151)	3.7E-07
IVW based on UKB summary statistics not adjusted for antihypertensive medication use or BMI	0.088	(0.042, 0.134)	1.9E-04	0.097	(0.053, 0.141)	1.6E-05

BMI: body mass index, DBP: diastolic blood pressure; IVW: inverse variance weighted; MR-PRESSO: Mendelian randomization pleiotropy residual sum and outlier; OR: Odds ratio; SBP: systolic blood pressure; WMH: white matter hyperintensities.

Table e-6. Sensitivity analyses for the Mendelian randomization associations between geneic proxies for beta blockers and calcium channel blockers and risk of stroke, risk of stroke subtypes, and WMH volume.

Outcome	BB (10 mm Hg decrease in SBP)			CCB (10 mm Hg decrease in SBP)		
Any stroke	**OR**	**95%CI**	**p**	**OR**	**95%CI**	**p**
Primary MR analysis with SNPs clumped at r^2<0.4 & adjusted for LD correlation	1.00	(0.84-1.18)	0.997	0.69	(0.64-0.74)	1.7E-26
IVW (SNPs clumped at r^2<0.1)	0.90	(0.71-1.15)	0.397	0.67	(0.59-0.76)	1.4E-08
MR Egger	1.00	(0.23-4.27)	0.997	0.78	(0.56-1.11)	0.179
Egger Intercept	1.00	(0.95-1.05)	0.885	0.99	(0.98-1.01)	0.318
Weighted median	0.89	(0.65-1.23)	0.485	0.67	(0.55-0.81)	4.5E-05
Weighted modal	0.90	(0.61-1.33)	0.600	0.66	(0.53-0.83)	2.6E-04
IVW after exclusion of MR-PRESSO outliers	0.93*	(0.70-1.23)	0.597	0.67	(0.59-0.76)	1.4E-08
Ischemic stroke	**OR**	**95%CI**	**p**	**OR**	**95%CI**	**p**
Primary MR analysis with SNPs clumped at r^2<0.4 & adjusted for LD correlation	**1.02**	(0.86-1.23)	0.795	0.71	(0.66-0.76)	4.3E-20
IVW (SNPs clumped at r^2<0.1)	**0.89**	(0.72-1.15)	0.439	0.69	(0.60-0.79)	6.7E-07
MR Egger	0.98	(0.17-5.62)	0.978	0.80	(0.56-1.14)	0.220
Egger Intercept	1.00	(0.94-1.06)	0.909	1.00	(0.98-1.01)	0.404
Weighted median	0.95	(0.68-1.32)	0.757	0.71	(0.58-0.86)	7.3E-04
Weighted modal	0.94	(0.64-1.38)	0.752	0.71	(0.57-0.88)	1.5E-03
IVW after exclusion of MR-PRESSO outliers*	0.94*	(0.70-1.27)	0.690	0.69	(0.60-0.79)	2.1E-07
Large artery stroke	**OR**	**95%CI**	**p**	**OR**	**95%CI**	**p**
Primary MR analysis with SNPs clumped at r^2<0.4 & adjusted for LD correlation	0.89	(0.57-1.37)	0.586	0.85	(0.73-0.99)	0.037
IVW (SNPs clumped at r^2<0.1)	0.91	(0.48-1.75)	0.783	0.83	(0.59-1.17)	0.277
MR Egger	2.12	(0.25-18.1)	0.491	0.75	(0.29-1.94)	0.546
Egger Intercept	0.97	(0.90-1.05)	0.422	1.00	(0.97-1.03)	0.789
Weighted median	1.04	(0.49-2.21)	0.917	0.82	(0.50-1.33)	0.421
Weighted modal	1.13	(0.45-2.85)	0.797	0.85	(0.54-1.33)	0.469
IVW after exclusion of MR-PRESSO outliers*	0.91**	(0.48-1.75)	0.783	0.83**	(0.57-1.20)	0.312
Cardioembolic stroke	**OR**	**95%CI**	**p**	**OR**	**95%CI**	**p**
Primary MR analysis with SNPs clumped at r^2<0.4 & adjusted for LD correlation	1.31	(0.90-1.91)	0.153	0.88	(0.82-0.95)	3.6E-04
IVW (SNPs clumped at r^2<0.1)	1.03	(0.60-1.77)	0.919	0.82	(0.61-1.10)	0.183
MR Egger	0.72	(0.06-9.22)	0.801	0.84	(0.40-1.77)	0.645
Egger Intercept	1.01	(0.92-1.11)	0.774	1.00	(0.98-1.02)	0.933
Weighted median	1.07	(0.52-2.21)	0.858	0.82	(0.52-1.31)	0.408
Weighted modal	1.57	(0.62-4.00)	0.345	0.77	(0.46-1.26)	0.296
IVW after exclusion of MR-PRESSO outliers*	1.03**	(0.60-1.77)	0.919	0.82**	(0.61-1.10)	0.180
Small vessel stroke	**OR**	**95%CI**	**p**	**OR**	**95%CI**	**p**
Primary MR analysis with SNPs clumped at r^2<0.4 & adjusted for LD correlation	1.09	(0.75-1.59)	0.646	0.60	(0.52-0.71)	4.4E-10
IVW (SNPs clumped at r^2<0.1)	0.77	(0.45-1.33)	0.344	0.63	(0.46-0.85)	2.9E-03
MR Egger	0.87	(0.02-34.78)	0.942	0.69	(0.25-1.90)	0.474
Egger Intercept	1.00	(0.87-1.13)	0.941	1.00	(0.97-1.03)	0.841
Weighted median	0.79	(0.4-1.57)	0.497	0.55	(0.35-0.87)	0.010
Weighted modal	0.83	(0.43-1.59)	0.567	0.50	(0.31-0.81)	4.9E-03
IVW after exclusion of MR-PRESSO outliers*	0.89**	(0.47-1.69)	0.726	0.63**	(0.44-0.90)	0.010

Intracerebral hemorrhage	**OR**	**95%CI**	**p**	**OR**	**95%CI**	**p**
Primary MR analysis with SNPs clumped at r^2<0.4 & adjusted for LD correlation	1.25	(0.40-3.88)	0.704	1.09	(0.65-1.83)	0.746
IVW (SNPs clumped at r^2<0.1)	2.56	(0.29-22.4)	0.396	1.23	(0.49-3.08)	0.665
MR Egger	34.8	(0.08-15930)	0.256	3.93	(0.40-38.5)	0.240
Egger Intercept	0.91	(0.73-1.12)	0.372	0.96	(0.90-1.03)	0.274
Weighted median	3.89	(0.27-55.5)	0.316	1.04	(0.27-4.06)	0.951
Weighted modal	5.71	(0.35-86.7)	0.23	1.00	(0.21-5.08)	0.960
IVW after exclusion of MR-PRESSO outliers*	2.56**	(0.29-22.4)	0.396	1.23**	(0.49-3.08)	0.665
Lobar intracerebral hemorrhage	**OR**	**95%CI**	**p**	**OR**	**95%CI**	**p**
Primary MR analysis with SNPs clumped at r^2<0.4 & adjusted for LD correlation	1.66	(0.05-57.49)	0.779	1.70	(0.67-4.35)	0.379
IVW (SNPs clumped at r^2<0.1)	5.98	(0.33-108)	0.225	1.26	(0.38-4.18)	0.706
MR Egger	4060	(1.22-13537849)	0.045	8.26	(0.42-161)	0.164
Egger Intercept	0.78	(0.59-1.04)	0.092	0.94	(0.86-1.03)	0.175
Weighted median	11.6	(0.36-369)	0.166	1.52	(0.26-8.87)	0.641
Weighted modal	30.7	(0.12-9537)	0.228	1.50	(0.21-10.6)	0.676
IVW after exclusion of MR-PRESSO outliers*	5.98**	(0.33-108)	0.225	1.26**	(0.38-4.18)	0.706
Deep intracerebral hemorrhage	**OR**	**95%CI**	**p**	**OR**	**95%CI**	**p**
Primary MR analysis with SNPs clumped at r^2<0.4 & adjusted for LD correlation	1.74	(0.07-41.9)	0.733	0.66	(0.31-1.40)	0.167
IVW (SNPs clumped at r^2<0.1)	2.28	(0.18-29.6)	0.528	0.83	(0.28-2.44)	0.734
MR Egger	1.41	(0.01-1905)	0.926	0.95	(0.07-13.5)	0.971
Egger Intercept	1.02	(0.79-1.31)	0.888	1.01	(0.93-1.09)	0.848
Weighted median	3.03	(0.14-67.0)	0.482	0.80	(0.18-3.59)	0.772
Weighted modal	5.72	(0.08-395)	0.416	0.91	(0.15-5.72)	0.911
IVW after exclusion of MR-PRESSO outliers*	2.28**	(0.18-29.6)	0.528	0.83**	(0.28-2.44)	0.734
WMH volume	**β**	**95%CI**	**p**	**β**	**95%CI**	**p**
Primary MR analysis with SNPs clumped at r2<0.4 & adjusted for LD correlation	-0.146	(-0.448, 0.157)	0.345	-0.491	(-0.591, -0.391)	3.5E-07
IVW (SNPs clumped at r2<0.1)	-0.345	(-1.279, 0.588)	0.469	-0.510	(-0.701, -0.319)	1.5E-07
MR Egger	-1.390	(-4.620, 1.840)	0.399	-0.831	(-1.284, -0.377)	3.2E-04
Egger Intercept	0.040	(-0.078, 0.158)	0.505	0.012	(-0.003, 0.027)	0.128
Weighted median	-0.682	(-1.152, -0.213)	0.004	-0.553	(-0.841, -0.265)	1.7E-04
Weighted modal	-0.744	(-1.315, -0.173)	0.011	-0.537	(-0.858, -0.216)	9.1E-04
IVW after exclusion of MR-PRESSO outliers*	-0.515*	(-1.142, 0.112)	0.249	-0.510**	(-0.701, -0.319)	1.5E-07

* rs4359161 and rs460718 were identified as outliers with the MR-PRESSO approach.

** No outliers identified with the MR-PRESSO approach.

IVW: inverse-variance weigted; LD: linkage disequilibrium; MR-PRESSO: Mendelian randomization pleiotropy residual sum and outlier; OR: odds ratio; SBP: systolic blood pressure; SNP: single nucleotide polymorphism; WMH: white matter hyperintensities.

SUPPLEMENTARY ONLINE CONTENT

Genetic determinants of blood lipids and cerebral small vessel disease: role of HDL cholesterol

Table of contents

Supplementary Table 1. Descriptive characteristics of the genome-wide association studies (GWAS) that were included in our Mendelian randomization analysis.

GWAS	Phenotype	Sample size	Ancestry	Adjustments[a]
Instrument selection				
GLGC & MVP	HDL-C, LDL-C, TG	617,303 individuals	Multi-ancestry	age, age^2, sex
GLGC	HDL-C, LDL-C, TG (for lipid-modifying drug targets & sensitivity analyses)	188,577 individuals	Multi-ancestry	age, age^2, sex
NMR-measured metabolite GWAS	Lipoprotein particle components	24,925 individuals	European	age, sex, time from last meal
Examined outcomes				
MEGASTROKE	Small vessel stroke	11,710 cases; 287,067 controls	Multi-ancestry	age, sex
UK Biobank	WMH volume	10,597 individuals	European (White British)	age, sex
ISGC ICH	ICH and subtypes (lobar, deep ICH)	1,537 cases/ 1,490 controls	European	age, sex

[a] All GWAS studies have further adjusted for principal components.

Abbreviations. GLGC, global lipids genetics consortium; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; MVP, Million Veteran Program; NMR, Nuclear Magnetic Resonance; TG, triglycerides.

Supplementary Table 2. Genetic instruments for blood lipid levels selected from the meta-analyzed datasets of the Million Veterans Program (MVP) and the Global Lipids Genetics Consortium (GLGC).

Phenotype	SNP	Chr	Position (hg18)	Eff_allele	Effect	SE	P-value	R2	F
HDL-C	rs1767141	1	23734350	a	-0.026	0.004	1.4E-10	0.0001	64.5
HDL-C	rs6668958	1	26902388	t	0.020	0.003	2.3E-12	0.0001	81.0
HDL-C	rs17162330	1	27236212	t	0.041	0.003	1.5E-35	0.0004	258.0
HDL-C	rs12144891	1	28344980	a	0.017	0.003	1.3E-10	0.0001	69.4
HDL-C	rs4660293	1	40028180	a	0.040	0.002	2.5E-62	0.0006	342.9
HDL-C	rs1168089	1	63113719	t	-0.015	0.002	1.8E-10	0.0001	63.6
HDL-C	rs4847399	1	93584606	a	-0.022	0.002	8.3E-25	0.0002	136.2
HDL-C	rs2878349	1	107549245	a	-0.015	0.003	2.1E-09	0.0001	61.3
HDL-C	rs12740374	1	109817590	t	0.045	0.002	9.6E-79	0.0007	430.3
HDL-C	rs7550711	1	110082886	t	-0.049	0.006	1.6E-16	0.0001	82.5
HDL-C	rs28362581	1	110163879	a	-0.028	0.004	4.0E-13	0.0001	67.1
HDL-C	rs333947	1	110470764	a	-0.027	0.003	4.3E-15	0.0002	113.6
HDL-C	rs267738	1	150940625	t	-0.024	0.003	9.9E-22	0.0002	109.5
HDL-C	rs12145743	1	156700651	t	-0.014	0.002	2.0E-10	0.0001	53.0
HDL-C	rs1011731	1	172346548	a	0.013	0.002	1.9E-11	0.0001	54.5
HDL-C	rs4650994	1	178515312	a	-0.019	0.002	4.7E-22	0.0002	115.1
HDL-C	rs2243976	1	182157235	a	0.029	0.002	2.0E-33	0.0004	234.2
HDL-C	rs16856110	1	205631767	a	0.020	0.003	5.0E-10	0.0001	84.0
HDL-C	rs6694509	1	219631981	t	-0.019	0.002	2.6E-17	0.0002	111.2
HDL-C	rs2807834	1	220970593	t	-0.024	0.003	1.8E-21	0.0002	145.1
HDL-C	rs4846914	1	230295691	a	0.047	0.002	9.2E-110	0.0011	663.0
HDL-C	rs1043900	1	230416744	a	0.023	0.003	8.3E-16	0.0002	109.6
HDL-C	rs558971	1	234853406	a	-0.014	0.002	8.1E-09	0.0001	59.1
HDL-C	rs11553746	2	272203	t	0.016	0.002	5.7E-14	0.0001	68.7
HDL-C	rs2867125	2	622827	t	0.017	0.003	3.2E-11	0.0001	52.4
HDL-C	rs4850047	2	3634753	t	0.020	0.003	1.0E-09	0.0001	63.3
HDL-C	rs676210	2	21231524	a	0.062	0.002	4.7E-143	0.0013	787.0
HDL-C	rs562338	2	21288321	a	0.018	0.003	1.8E-12	0.0001	60.7
HDL-C	rs36020289	2	53992622	c	0.046	0.008	2.0E-09	0.0001	43.0
HDL-C	rs12990465	2	65281401	t	0.026	0.003	1.9E-18	0.0003	164.3
HDL-C	rs13389219	2	165528876	t	0.033	0.002	1.2E-54	0.0005	314.9
HDL-C	rs6435161	2	203519783	t	0.018	0.002	1.6E-13	0.0001	82.2
HDL-C	rs2943641	2	227093745	t	0.039	0.002	2.5E-77	0.0007	427.1
HDL-C	rs11712666	3	11619958	a	-0.014	0.002	1.0E-09	0.0001	60.7
HDL-C	rs2067819	3	12359049	a	0.021	0.003	5.6E-14	0.0002	93.9
HDL-C	rs2292101	3	12434901	t	-0.044	0.007	1.8E-11	0.0002	95.1
HDL-C	rs6777217	3	36979042	a	-0.013	0.002	1.1E-10	0.0001	51.0
HDL-C	rs2305637	3	47045846	t	-0.026	0.003	1.1E-19	0.0002	119.1
HDL-C	rs6762477	3	50093209	a	0.023	0.002	2.1E-28	0.0003	162.8
HDL-C	rs13326165	3	52532118	a	0.021	0.003	1.3E-16	0.0001	89.4
HDL-C	rs11242	3	53125922	t	0.015	0.002	2.3E-10	0.0001	68.0
HDL-C	rs35000036	3	123190731	t	-0.013	0.002	3.3E-10	0.0001	50.5
HDL-C	rs1279840	3	136006576	t	0.028	0.003	3.4E-25	0.0003	177.9
HDL-C	rs3773910	3	152171870	c	-0.017	0.003	4.1E-10	0.0001	65.9
HDL-C	rs900399	3	156798732	a	-0.021	0.002	1.4E-26	0.0002	134.2

HDL-C	rs7633675	3	185510613	t	0.013	0.002	2.2E-10	0.0001	49.0
HDL-C	rs4234589	3	185818882	a	0.022	0.003	6.2E-11	0.0001	72.3
HDL-C	rs11248051	4	858332	t	-0.019	0.003	8.5E-09	0.0001	40.8
HDL-C	rs10019888	4	26062990	a	0.024	0.003	4.6E-15	0.0002	102.8
HDL-C	rs293429	4	69591612	t	-0.013	0.002	1.5E-09	0.0001	45.5
HDL-C	rs10023050	4	88064431	a	-0.015	0.002	3.7E-11	0.0001	68.9
HDL-C	rs3822072	4	89741269	a	-0.021	0.002	1.4E-20	0.0002	141.0
HDL-C	rs2602836	4	100014805	a	0.013	0.002	2.0E-10	0.0001	48.7
HDL-C	rs112519623	4	103184239	a	-0.047	0.008	8.8E-09	0.0001	39.8
HDL-C	rs13107325	4	103188709	t	-0.078	0.004	6.3E-80	0.0009	540.0
HDL-C	rs6855363	4	157670537	t	-0.019	0.002	1.3E-14	0.0002	93.2
HDL-C	rs7735253	5	53297611	a	-0.021	0.003	6.5E-16	0.0002	105.7
HDL-C	rs459193	5	55806751	a	0.026	0.002	9.2E-33	0.0003	168.5
HDL-C	rs9686661	5	55861786	t	-0.034	0.003	3.5E-42	0.0004	229.0
HDL-C	rs4976033	5	67714246	a	0.013	0.002	1.8E-11	0.0001	55.1
HDL-C	rs10057967	5	74997756	t	-0.021	0.003	2.8E-14	0.0002	121.3
HDL-C	rs4705986	5	132349654	t	-0.035	0.006	2.0E-09	0.0001	87.9
HDL-C	rs390299	5	153363334	a	-0.015	0.002	4.6E-12	0.0001	60.4
HDL-C	rs2434612	5	158022041	a	0.023	0.003	8.1E-16	0.0002	104.0
HDL-C	rs7730898	5	170459675	a	-0.018	0.003	1.9E-12	0.0001	81.4
HDL-C	rs1265099	6	31105413	a	0.017	0.002	1.2E-16	0.0001	86.2
HDL-C	rs184070214	6	31526080	a	-0.045	0.007	1.5E-09	0.0001	46.8
HDL-C	rs9332739	6	31903804	c	-0.029	0.005	1.8E-09	0.0001	43.9
HDL-C	rs3135006	6	32667119	t	-0.019	0.003	3.0E-13	0.0001	84.9
HDL-C	rs2894342	6	33774394	a	0.015	0.002	1.3E-09	0.0001	46.4
HDL-C	rs1759645	6	34194866	t	0.024	0.003	3.3E-13	0.0002	97.4
HDL-C	rs16885998	6	34268107	t	0.051	0.006	3.0E-16	0.0002	138.8
HDL-C	rs11755393	6	34824636	a	0.030	0.002	5.9E-45	0.0004	262.9
HDL-C	rs41270076	6	35467891	t	0.039	0.006	6.2E-10	0.0001	48.2
HDL-C	rs4711698	6	41987451	t	0.018	0.003	2.8E-11	0.0001	76.4
HDL-C	rs2274517	6	42932715	t	0.016	0.002	1.4E-11	0.0001	74.5
HDL-C	rs6905288	6	43758873	a	-0.030	0.002	5.5E-49	0.0004	262.8
HDL-C	rs35349911	6	43785255	t	-0.014	0.002	1.0E-11	0.0001	57.9
HDL-C	rs881858	6	43806609	a	-0.014	0.002	9.5E-11	0.0001	49.3
HDL-C	rs2754820	6	109246891	a	0.020	0.003	3.1E-09	0.0001	76.6
HDL-C	rs884366	6	109574095	a	-0.014	0.002	8.1E-11	0.0001	54.0
HDL-C	rs3756772	6	116325142	t	0.013	0.002	1.6E-10	0.0001	49.4
HDL-C	rs2745353	6	127452935	t	-0.020	0.002	6.5E-24	0.0002	125.5
HDL-C	rs6925103	6	137076010	t	-0.012	0.002	1.3E-09	0.0001	45.5
HDL-C	rs643381	6	139839423	a	0.021	0.002	2.5E-25	0.0002	133.5
HDL-C	rs41272114	6	161006077	t	0.066	0.006	3.4E-27	0.0003	181.0
HDL-C	rs1652507	6	161082461	t	0.046	0.003	9.6E-51	0.0006	365.4
HDL-C	rs1997243	7	1083777	a	-0.022	0.003	2.2E-15	0.0001	80.2
HDL-C	rs2303361	7	6449496	t	-0.024	0.002	4.1E-23	0.0002	118.6
HDL-C	rs10282707	7	17911038	t	-0.026	0.002	7.6E-32	0.0003	200.3
HDL-C	rs1534696	7	26397239	a	0.019	0.003	1.3E-13	0.0002	104.1
HDL-C	rs2726070	7	36170883	a	-0.017	0.003	2.0E-10	0.0001	90.4
HDL-C	rs4917014	7	50305863	t	-0.014	0.002	5.0E-11	0.0001	50.2

HDL-C	rs1178979	7	72856430	t	-0.034	0.003	6.2E-41	0.0003	206.0
HDL-C	rs11556924	7	129663496	t	0.013	0.002	2.0E-09	0.0001	44.8
HDL-C	rs972283	7	130466854	a	0.028	0.002	2.9E-43	0.0004	233.6
HDL-C	rs3735080	7	150217309	t	-0.014	0.002	2.7E-09	0.0001	41.1
HDL-C	rs7787577	7	150521026	a	0.031	0.005	8.3E-12	0.0002	102.0
HDL-C	rs11774381	8	9183339	t	-0.028	0.003	5.1E-24	0.0003	192.0
HDL-C	rs4841132	8	9183596	a	-0.108	0.003	1.9E-215	0.0020	1207.7
HDL-C	rs9657541	8	10643164	t	-0.025	0.003	3.6E-17	0.0002	124.0
HDL-C	rs1801177	8	19805708	a	-0.120	0.009	2.3E-44	0.0005	324.7
HDL-C	rs264	8	19813180	a	0.033	0.003	6.1E-23	0.0003	161.6
HDL-C	rs268	8	19813529	a	0.238	0.008	5.5E-213	0.0018	1108.5
HDL-C	rs13702	8	19824492	t	-0.059	0.005	4.4E-32	0.0014	890.5
HDL-C	rs17091872	8	19831977	a	-0.056	0.004	2.6E-36	0.0009	573.9
HDL-C	rs2410622	8	19854773	t	-0.026	0.004	1.4E-10	0.0002	113.1
HDL-C	rs6983170	8	19860161	t	0.093	0.010	5.6E-19	0.0005	281.3
HDL-C	rs6651485	8	19861854	a	0.054	0.004	1.9E-36	0.0011	650.1
HDL-C	rs2083637	8	19865175	a	-0.048	0.005	3.5E-22	0.0009	557.7
HDL-C	rs7837677	8	19889872	t	-0.034	0.004	3.8E-16	0.0005	317.9
HDL-C	rs10106652	8	19928160	a	0.031	0.005	4.6E-11	0.0004	230.1
HDL-C	rs34859606	8	19930682	c	-0.019	0.003	9.0E-09	0.0001	70.4
HDL-C	rs6586892	8	19941145	a	0.036	0.004	5.5E-22	0.0006	377.0
HDL-C	rs6983999	8	19955920	a	0.024	0.003	2.3E-15	0.0003	176.9
HDL-C	rs4512408	8	71099094	t	0.026	0.004	5.5E-12	0.0001	80.8
HDL-C	rs2957447	8	106357374	a	0.014	0.002	8.9E-09	0.0001	58.2
HDL-C	rs2293889	8	116599199	t	-0.030	0.002	2.7E-52	0.0004	275.0
HDL-C	rs4871137	8	121868551	t	-0.021	0.002	4.4E-22	0.0002	127.3
HDL-C	rs17405319	8	126449406	t	-0.025	0.003	9.0E-16	0.0002	110.9
HDL-C	rs2954026	8	126484526	t	-0.049	0.002	1.3E-108	0.0010	622.1
HDL-C	rs581080	9	15305378	c	0.037	0.003	1.5E-49	0.0004	266.9
HDL-C	rs13292026	9	107557315	a	-0.044	0.007	4.9E-09	0.0001	66.2
HDL-C	rs2230808	9	107562804	t	-0.024	0.002	3.8E-23	0.0002	129.8
HDL-C	rs76881554	9	107578620	a	-0.164	0.025	3.4E-11	0.0001	56.4
HDL-C	rs2066714	9	107586753	t	-0.051	0.003	3.5E-69	0.0006	398.0
HDL-C	rs3905000	9	107657070	a	-0.055	0.003	6.8E-56	0.0008	477.7
HDL-C	rs10120087	9	107661150	a	0.044	0.004	1.5E-26	0.0004	228.8
HDL-C	rs1800978	9	107665978	c	0.057	0.004	3.7E-52	0.0007	460.3
HDL-C	rs13284054	9	107669073	t	0.054	0.005	1.5E-27	0.0006	375.3
HDL-C	rs1800977	9	107690450	a	0.026	0.003	3.2E-24	0.0003	186.2
HDL-C	rs10733608	9	117148430	t	0.014	0.002	6.3E-09	0.0001	59.9
HDL-C	rs635634	9	136155000	t	0.015	0.003	6.6E-09	0.0001	40.9
HDL-C	rs11255744	10	8601074	t	-0.018	0.003	1.3E-09	0.0001	80.4
HDL-C	rs10904908	10	17260290	a	-0.012	0.002	4.1E-09	0.0001	42.0
HDL-C	rs970548	10	46013277	a	-0.025	0.002	1.7E-26	0.0002	137.0
HDL-C	rs2068888	10	94839642	a	0.021	0.002	1.1E-24	0.0002	128.9
HDL-C	rs2862954	10	101912064	t	-0.017	0.002	6.0E-18	0.0001	90.6
HDL-C	rs2792751	10	113940329	t	0.028	0.002	2.5E-36	0.0003	197.6
HDL-C	rs2148489	10	114048792	t	0.022	0.003	1.8E-15	0.0002	108.0
HDL-C	rs7076938	10	115789375	t	0.017	0.002	5.2E-15	0.0001	73.1
HDL-C	rs140201358	11	823586	c	0.052	0.009	2.8E-09	0.0001	43.3

HDL-C	rs16928809	11	2936952	a	-0.025	0.004	7.7E-12	0.0001	62.1
HDL-C	rs6486121	11	13355770	t	-0.015	0.002	2.5E-10	0.0001	65.3
HDL-C	rs2303975	11	14276999	a	0.019	0.003	1.4E-10	0.0001	49.5
HDL-C	rs925946	11	27667202	t	-0.012	0.002	6.6E-09	0.0001	37.8
HDL-C	rs7927401	11	32481177	t	-0.021	0.003	1.2E-11	0.0002	98.1
HDL-C	rs3824866	11	47258853	t	0.033	0.004	4.4E-19	0.0003	178.2
HDL-C	rs326214	11	47298360	a	-0.025	0.003	1.0E-20	0.0003	162.9
HDL-C	rs10838738	11	47663049	a	0.019	0.002	1.6E-17	0.0002	103.0
HDL-C	rs61897793	11	61599347	a	0.024	0.004	3.7E-09	0.0002	98.3
HDL-C	rs174583	11	61609750	t	-0.042	0.002	1.1E-68	0.0008	498.3
HDL-C	rs35169799	11	64031241	t	-0.040	0.004	5.0E-22	0.0002	109.7
HDL-C	rs644740	11	65561468	t	0.016	0.002	3.9E-12	0.0001	78.0
HDL-C	rs622082	11	68703959	a	0.015	0.002	1.6E-13	0.0001	63.3
HDL-C	rs499974	11	75455021	a	-0.025	0.003	1.5E-22	0.0002	116.6
HDL-C	rs746463	11	109995944	t	-0.017	0.002	6.3E-14	0.0001	71.9
HDL-C	rs180349	11	116611827	a	0.017	0.003	1.8E-10	0.0001	75.4
HDL-C	rs10488698	11	116633947	a	0.045	0.004	1.2E-25	0.0002	131.4
HDL-C	rs964184	11	116648917	c	0.132	0.003	0.0E+00	0.0044	2730.3
HDL-C	rs138326449	11	116701354	a	0.716	0.026	2.1E-170	0.0031	1896.5
HDL-C	rs138407155	11	116707044	a	0.356	0.042	2.4E-17	0.0002	93.6
HDL-C	rs12281729	11	116838130	a	0.077	0.005	3.5E-49	0.0007	402.5
HDL-C	rs10892063	11	116896155	a	0.042	0.003	9.4E-45	0.0008	515.0
HDL-C	rs12269901	11	116973929	c	0.027	0.003	3.3E-23	0.0003	191.3
HDL-C	rs593245	11	117183650	t	0.015	0.002	2.0E-09	0.0001	64.9
HDL-C	rs7941030	11	122522375	t	-0.023	0.002	3.3E-30	0.0002	151.4
HDL-C	rs4937122	11	126228659	t	0.032	0.004	6.0E-13	0.0002	93.7
HDL-C	rs7134375	12	20473758	a	0.022	0.002	6.2E-27	0.0002	139.1
HDL-C	rs7134150	12	20591332	a	-0.031	0.005	6.3E-11	0.0001	76.1
HDL-C	rs4963975	12	26443030	a	-0.020	0.003	2.3E-13	0.0001	88.5
HDL-C	rs1126930	12	49399132	c	-0.037	0.006	2.9E-11	0.0001	52.5
HDL-C	rs784563	12	53866619	t	0.015	0.002	3.1E-11	0.0001	71.5
HDL-C	rs11613352	12	57792580	t	0.023	0.003	5.4E-17	0.0002	110.3
HDL-C	rs2373459	12	101873956	t	0.016	0.002	2.9E-11	0.0001	70.2
HDL-C	rs7298565	12	109937534	a	0.028	0.002	1.8E-44	0.0004	240.9
HDL-C	rs3184504	12	111884608	t	-0.024	0.002	5.0E-31	0.0003	182.5
HDL-C	rs72650673	12	111885310	a	-0.172	0.028	1.4E-09	0.0001	47.2
HDL-C	rs1183910	12	121420807	a	0.013	0.002	1.7E-09	0.0001	43.1
HDL-C	rs12369179	12	122963550	t	-0.031	0.005	3.8E-10	0.0001	92.3
HDL-C	rs1798192	12	123200768	t	-0.019	0.002	3.6E-21	0.0002	113.5
HDL-C	rs940904	12	123491572	a	-0.023	0.003	1.8E-16	0.0002	121.0
HDL-C	rs4759375	12	123796238	t	0.049	0.004	6.0E-35	0.0004	257.2
HDL-C	rs12317176	12	124404718	t	-0.022	0.002	5.2E-20	0.0002	141.9
HDL-C	rs863750	12	124505444	t	-0.017	0.002	3.1E-13	0.0001	88.0
HDL-C	rs12230272	12	125083696	a	0.022	0.003	5.7E-11	0.0002	102.9
HDL-C	rs838880	12	125261593	t	-0.025	0.002	6.3E-31	0.0003	173.3
HDL-C	rs10773105	12	125283766	t	-0.026	0.002	3.6E-27	0.0003	201.1
HDL-C	rs150728540	12	125292360	a	0.476	0.058	1.6E-16	0.0001	83.8
HDL-C	rs5891	12	125299542	t	0.078	0.009	2.2E-18	0.0001	91.1
HDL-C	rs7306660	12	125327384	a	-0.029	0.002	2.4E-32	0.0004	232.8

HDL-C	rs7298751	12	125380232	a	-0.043	0.004	5.1E-32	0.0004	244.7
HDL-C	rs17532301	13	41609047	a	-0.027	0.005	3.4E-09	0.0001	59.4
HDL-C	rs10483776	14	65914867	a	0.015	0.003	3.9E-09	0.0001	40.4
HDL-C	rs8021180	14	70783943	a	0.014	0.002	3.3E-09	0.0001	61.9
HDL-C	rs13379043	14	74250126	t	-0.018	0.002	1.4E-15	0.0001	78.1
HDL-C	rs4983559	14	105277209	a	-0.025	0.002	2.2E-36	0.0003	190.0
HDL-C	rs9944249	15	41847176	t	-0.013	0.002	9.1E-09	0.0001	53.0
HDL-C	rs55707100	15	43820717	t	-0.093	0.006	3.1E-51	0.0004	277.0
HDL-C	rs4622454	15	58646332	t	0.021	0.003	1.0E-14	0.0002	119.4
HDL-C	rs4775041	15	58674695	c	0.039	0.004	9.1E-24	0.0006	381.0
HDL-C	rs16940147	15	58676119	a	0.050	0.008	1.1E-09	0.0002	146.8
HDL-C	rs117901517	15	58678869	t	0.038	0.006	3.2E-10	0.0002	111.4
HDL-C	rs34718390	15	58682690	a	0.053	0.006	1.1E-18	0.0003	204.5
HDL-C	rs1532085	15	58683366	a	0.062	0.004	2.9E-68	0.0018	1137.3
HDL-C	rs7165077	15	58686809	t	-0.027	0.004	8.5E-13	0.0002	104.1
HDL-C	rs6494003	15	58690048	a	0.093	0.011	5.0E-18	0.0004	247.8
HDL-C	rs16940233	15	58702941	t	-0.048	0.006	7.0E-18	0.0003	198.5
HDL-C	rs12912415	15	58721447	a	0.040	0.004	2.6E-25	0.0004	246.5
HDL-C	rs6494006	15	58730571	t	0.044	0.006	1.7E-11	0.0002	145.5
HDL-C	rs17301746	15	58731395	t	0.088	0.010	8.6E-20	0.0003	169.8
HDL-C	rs936960	15	58751877	t	0.052	0.004	7.9E-32	0.0005	286.4
HDL-C	rs1869138	15	58779039	t	-0.028	0.004	3.1E-11	0.0001	88.4
HDL-C	rs6083	15	58838010	a	-0.016	0.003	5.0E-09	0.0001	77.0
HDL-C	rs17269397	15	58857378	a	-0.026	0.003	6.4E-18	0.0003	208.7
HDL-C	rs424346	15	59010962	t	0.054	0.009	2.2E-09	0.0002	133.3
HDL-C	rs181181625	15	59377940	t	0.348	0.033	1.3E-26	0.0003	208.9
HDL-C	rs12148597	15	61955338	a	-0.016	0.003	5.3E-09	0.0001	54.7
HDL-C	rs34317102	15	63414083	a	-0.019	0.002	1.5E-16	0.0001	80.0
HDL-C	rs2228510	15	63970456	t	-0.012	0.002	4.6E-09	0.0001	42.4
HDL-C	rs139271800	15	90214777	a	-0.248	0.037	3.2E-11	0.0001	53.2
HDL-C	rs7202647	16	985891	t	0.017	0.003	4.6E-09	0.0001	70.1
HDL-C	rs12928822	16	11403893	t	0.018	0.003	1.6E-10	0.0001	53.3
HDL-C	rs1421085	16	53800954	t	0.028	0.002	3.4E-45	0.0004	236.0
HDL-C	rs12929759	16	54410447	a	0.018	0.003	3.0E-13	0.0001	89.4
HDL-C	rs4238772	16	55029160	a	0.029	0.005	1.2E-09	0.0001	84.2
HDL-C	rs8044753	16	56883438	a	0.025	0.003	3.9E-19	0.0003	189.0
HDL-C	rs36049418	16	56921840	a	0.066	0.009	1.4E-14	0.0002	101.5
HDL-C	rs11648751	16	56937262	t	-0.030	0.004	1.0E-11	0.0002	117.6
HDL-C	rs37029	16	56949168	a	-0.024	0.003	2.0E-21	0.0003	177.8
HDL-C	rs9989419	16	56985139	a	-0.034	0.002	1.4E-41	0.0005	334.5
HDL-C	rs72786786	16	56985514	a	0.058	0.004	3.0E-40	0.0014	869.0
HDL-C	rs76315536	16	56986976	t	0.161	0.016	1.1E-24	0.0005	290.3
HDL-C	rs4783961	16	56994894	a	0.026	0.004	2.1E-12	0.0003	208.7
HDL-C	rs1800775	16	56995236	a	0.032	0.005	3.7E-11	0.0005	310.2
HDL-C	rs34065661	16	56995935	c	-0.485	0.019	1.5E-138	0.0021	1273.2
HDL-C	rs7203984	16	56999258	a	0.074	0.005	2.2E-56	0.0018	1104.4
HDL-C	rs1532624	16	57005479	a	0.113	0.005	1.7E-121	0.0062	3859.4
HDL-C	rs12708974	16	57005550	t	-0.086	0.005	4.5E-73	0.0015	956.9
HDL-C	rs5883	16	57007353	t	0.167	0.006	4.7E-186	0.0030	1877.3

HDL-C	rs289714	16	57007451	a	0.082	0.005	8.4E-70	0.0021	1312.3
HDL-C	rs5880	16	57015091	c	-0.138	0.005	1.5E-147	0.0018	1113.2
HDL-C	rs506829	16	57383759	t	0.019	0.003	2.2E-09	0.0001	78.3
HDL-C	rs16962034	16	60419220	t	-0.016	0.003	6.2E-09	0.0001	72.5
HDL-C	rs7202185	16	67714560	a	-0.037	0.005	3.4E-14	0.0002	115.6
HDL-C	rs16942887	16	67928042	a	0.074	0.003	3.1E-134	0.0012	765.1
HDL-C	rs1345868	16	71669624	a	0.014	0.003	7.8E-09	0.0001	55.0
HDL-C	rs12443634	16	81524274	a	-0.031	0.003	2.0E-25	0.0004	244.8
HDL-C	rs3803800	17	7462969	a	-0.013	0.002	5.7E-09	0.0001	40.4
HDL-C	rs2071379	17	26695832	a	0.014	0.002	5.1E-10	0.0001	61.3
HDL-C	rs11078915	17	37715426	t	-0.027	0.004	6.4E-13	0.0003	157.4
HDL-C	rs11869286	17	37813856	c	0.024	0.003	1.8E-18	0.0003	161.7
HDL-C	rs11556624	17	37815304	c	0.087	0.007	8.9E-37	0.0003	213.2
HDL-C	rs4794822	17	38156712	t	-0.018	0.002	1.9E-16	0.0001	91.1
HDL-C	rs72836561	17	41926126	t	-0.187	0.006	7.6E-228	0.0021	1293.7
HDL-C	rs231539	17	41942109	t	-0.036	0.004	4.2E-23	0.0004	221.1
HDL-C	rs17679445	17	46022065	a	0.030	0.004	6.1E-13	0.0001	66.0
HDL-C	rs11652146	17	47422363	a	-0.017	0.003	6.6E-12	0.0001	78.1
HDL-C	rs12602912	17	65870073	t	-0.018	0.002	2.2E-13	0.0001	59.4
HDL-C	rs10852765	17	66884879	a	0.016	0.002	1.1E-11	0.0001	73.8
HDL-C	rs2292642	17	76395430	t	0.032	0.002	1.6E-55	0.0005	297.9
HDL-C	rs2289750	17	76437343	a	-0.028	0.005	6.5E-09	0.0001	77.2
HDL-C	rs1788783	18	21161134	t	0.015	0.002	7.0E-11	0.0001	69.4
HDL-C	rs8093249	18	47097398	a	0.034	0.004	9.5E-22	0.0003	184.7
HDL-C	rs77960347	18	47109955	a	-0.250	0.009	7.7E-166	0.0017	1033.2
HDL-C	rs117623631	18	47113165	t	0.343	0.025	1.0E-41	0.0005	333.8
HDL-C	rs3786248	18	47118219	t	-0.059	0.006	2.8E-26	0.0004	221.8
HDL-C	rs7241918	18	47160953	t	0.074	0.003	4.3E-120	0.0015	902.9
HDL-C	rs4939886	18	47176793	a	0.020	0.003	8.8E-10	0.0002	110.5
HDL-C	rs11660468	18	47209143	t	0.025	0.002	1.2E-25	0.0003	190.2
HDL-C	rs9956279	18	57942799	t	-0.017	0.003	1.0E-10	0.0001	71.5
HDL-C	rs12975319	19	3414088	a	-0.015	0.003	3.7E-09	0.0001	62.1
HDL-C	rs10408844	19	7244884	t	0.018	0.003	5.2E-10	0.0001	63.6
HDL-C	rs4804833	19	7970635	a	-0.015	0.002	4.9E-10	0.0001	66.5
HDL-C	rs116843064	19	8429323	a	0.246	0.007	1.3E-252	0.0031	1913.2
HDL-C	rs2913968	19	8467235	t	0.017	0.003	2.0E-10	0.0001	68.3
HDL-C	rs6511720	19	11202306	t	0.020	0.003	1.1E-10	0.0001	49.4
HDL-C	rs17616661	19	11303554	a	0.026	0.004	1.2E-12	0.0001	62.1
HDL-C	rs737337	19	11347493	t	0.057	0.003	1.2E-62	0.0006	375.5
HDL-C	rs2111504	19	32917455	a	0.017	0.003	1.4E-10	0.0001	48.1
HDL-C	rs731839	19	33899065	a	0.018	0.002	2.4E-17	0.0001	89.5
HDL-C	rs2075650	19	45395619	a	0.038	0.003	7.7E-28	0.0003	209.7
HDL-C	rs77301115	19	45396973	a	-0.066	0.009	2.0E-13	0.0002	134.3
HDL-C	rs7412	19	45412079	t	0.086	0.004	3.0E-85	0.0011	650.4
HDL-C	rs439401	19	45414451	t	0.014	0.002	7.9E-11	0.0001	58.3
HDL-C	rs4420638	19	45422946	a	0.042	0.004	2.9E-29	0.0005	318.0
HDL-C	rs5167	19	45448465	t	-0.038	0.002	2.4E-78	0.0007	405.2
HDL-C	rs8111071	19	46307406	a	0.028	0.004	1.1E-14	0.0001	72.3
HDL-C	rs2303108	19	47589895	t	0.014	0.002	7.9E-11	0.0001	49.7

HDL-C	rs3752125	19	52327784	t	-0.020	0.003	3.4E-16	0.0002	105.8
HDL-C	rs12975366	19	54759361	t	0.021	0.002	1.4E-19	0.0002	132.8
HDL-C	rs386000	19	54792761	c	0.044	0.003	5.7E-48	0.0006	395.1
HDL-C	rs12979085	19	54837165	a	0.020	0.003	1.1E-12	0.0002	105.2
HDL-C	rs1132274	20	17596155	a	-0.016	0.003	3.9E-09	0.0001	42.6
HDL-C	rs2268086	20	32648738	a	0.014	0.002	3.4E-09	0.0001	60.2
HDL-C	rs1415771	20	33734493	a	-0.013	0.002	1.9E-10	0.0001	51.3
HDL-C	rs1800961	20	43042364	t	-0.142	0.006	5.1E-137	0.0012	745.5
HDL-C	rs3827066	20	44586023	t	0.050	0.004	4.2E-37	0.0007	413.4
HDL-C	rs8123864	20	44598670	t	0.048	0.003	2.2E-55	0.0010	643.8
HDL-C	rs1211644	20	45592842	t	-0.018	0.003	3.2E-09	0.0001	73.7
HDL-C	rs11700063	20	46153148	a	0.021	0.003	3.3E-14	0.0002	101.2
HDL-C	rs4239651	20	46340596	t	-0.021	0.003	8.4E-10	0.0001	82.2
HDL-C	rs6025606	20	56098733	t	0.012	0.002	2.9E-09	0.0001	40.6
HDL-C	rs310631	20	62196253	a	0.013	0.002	7.4E-09	0.0001	52.9
HDL-C	rs6062343	20	62695931	a	0.014	0.002	7.6E-12	0.0001	57.7
HDL-C	rs235314	21	46271452	t	-0.015	0.002	7.2E-11	0.0001	69.0
HDL-C	rs12482088	21	46901973	a	0.019	0.003	5.0E-10	0.0001	77.3
HDL-C	rs181362	22	21932068	t	-0.030	0.002	1.3E-36	0.0003	202.2
HDL-C	rs4823006	22	29451671	a	-0.013	0.002	9.1E-11	0.0001	51.4
HDL-C	rs17738540	22	30888527	t	-0.018	0.002	1.6E-14	0.0001	71.1
HDL-C	rs738322	22	38569006	a	-0.020	0.002	4.2E-23	0.0002	121.0
HDL-C	rs738409	22	44324727	c	0.016	0.002	1.5E-11	0.0001	51.9
LDL-C	*rs2992753*	1	18808292	a	-0.012	0.002	3.9E-09	0.0001	44.0
LDL-C	rs35172831	1	25850206	t	0.019	0.003	8.1E-12	0.0002	111.8
LDL-C	rs12748152	1	27138393	t	0.026	0.004	7.6E-12	0.0001	57.3
LDL-C	rs17111483	1	55485098	t	0.040	0.005	1.9E-16	0.0003	190.5
LDL-C	rs11206510	1	55496039	t	0.068	0.003	2.7E-111	0.0013	832.9
LDL-C	rs2479409	1	55504650	a	-0.022	0.002	1.1E-22	0.0002	140.7
LDL-C	rs11583680	1	55505668	t	0.027	0.004	1.9E-12	0.0002	105.8
LDL-C	rs10888896	1	55509213	c	0.029	0.003	1.0E-20	0.0003	200.2
LDL-C	rs693668	1	55521109	a	0.029	0.004	4.5E-15	0.0004	232.2
LDL-C	rs562556	1	55524237	a	0.130	0.008	4.9E-61	0.0047	2926.2
LDL-C	rs61739739	1	55548991	t	0.080	0.011	1.7E-12	0.0001	81.2
LDL-C	rs1165222	1	55638075	a	-0.135	0.008	1.7E-63	0.0047	2908.7
LDL-C	rs1475701	1	55638546	t	-0.068	0.006	1.8E-26	0.0003	211.8
LDL-C	rs7551981	1	55719166	t	0.030	0.003	7.2E-27	0.0004	258.6
LDL-C	rs10489488	1	55792722	a	-0.093	0.011	5.3E-17	0.0002	152.8
LDL-C	rs12742537	1	63346976	a	0.015	0.002	2.5E-14	0.0001	70.2
LDL-C	rs10874746	1	93323971	t	-0.015	0.002	2.3E-13	0.0001	66.4
LDL-C	rs1730859	1	107617707	a	-0.019	0.003	2.6E-13	0.0002	98.5
LDL-C	rs12740374	1	109817590	t	-0.160	0.002	0.0E+00	0.0086	5379.2
LDL-C	rs4745	1	155106227	a	0.012	0.002	5.7E-10	0.0001	47.5
LDL-C	rs867772	1	220972343	a	-0.026	0.003	5.8E-19	0.0003	173.2
LDL-C	rs558971	1	234853406	a	-0.036	0.002	1.2E-51	0.0007	406.6
LDL-C	rs1473886	2	20368519	t	-0.015	0.002	2.1E-10	0.0001	65.9
LDL-C	rs12710745	2	21112689	a	0.020	0.003	1.0E-14	0.0002	119.0
LDL-C	rs6547409	2	21190209	t	-0.095	0.006	8.9E-62	0.0009	573.7
LDL-C	rs1801702	2	21225485	c	0.097	0.007	1.5E-47	0.0005	281.4

LDL-C	rs1042023	2	21229446	c	0.086	0.011	5.5E-15	0.0001	85.9
LDL-C	rs12713843	2	21238367	t	-0.188	0.016	8.2E-33	0.0003	191.3
LDL-C	rs12713844	2	21238413	c	0.074	0.011	5.0E-12	0.0001	63.7
LDL-C	rs679899	2	21250914	a	-0.029	0.002	7.0E-37	0.0004	267.5
LDL-C	rs515135	2	21286057	t	-0.088	0.003	1.4E-202	0.0024	1489.3
LDL-C	rs62122515	2	21295227	a	0.026	0.003	8.2E-14	0.0003	183.0
LDL-C	rs4635554	2	21389659	t	-0.022	0.003	2.9E-18	0.0002	133.9
LDL-C	rs1260327	2	27711893	a	0.015	0.002	2.1E-11	0.0001	67.5
LDL-C	rs814295	2	27743215	a	0.031	0.003	1.2E-19	0.0002	153.4
LDL-C	rs11556157	2	44028013	a	-0.020	0.002	2.0E-18	0.0002	94.7
LDL-C	rs72796748	2	44080310	t	0.059	0.007	2.6E-19	0.0003	189.1
LDL-C	rs4077440	2	44081042	t	0.117	0.005	1.1E-122	0.0067	4167.0
LDL-C	rs6718187	2	44082362	a	-0.070	0.005	2.0E-43	0.0024	1501.7
LDL-C	rs4148218	2	44099582	a	-0.060	0.003	7.7E-70	0.0011	676.7
LDL-C	rs11125936	2	62871225	t	0.025	0.003	1.1E-13	0.0001	66.5
LDL-C	rs10185855	2	101642260	a	0.014	0.002	8.9E-09	0.0001	55.2
LDL-C	rs10490626	2	118835841	a	-0.045	0.004	2.4E-30	0.0003	168.7
LDL-C	rs1808458	2	118879253	t	0.031	0.005	2.6E-10	0.0001	74.7
LDL-C	rs6706968	2	121310269	a	-0.023	0.003	3.9E-16	0.0003	157.6
LDL-C	rs2198562	2	158465673	c	0.035	0.006	2.9E-10	0.0001	68.8
LDL-C	rs2287623	2	169830155	a	-0.018	0.002	1.4E-19	0.0002	97.5
LDL-C	rs6435161	2	203519783	t	0.024	0.003	4.3E-21	0.0002	146.1
LDL-C	rs1048013	2	204154552	t	0.012	0.002	8.8E-10	0.0001	46.2
LDL-C	rs887829	2	234668570	t	-0.021	0.002	1.7E-22	0.0002	115.3
LDL-C	rs7616006	3	12267648	a	0.022	0.002	2.3E-20	0.0002	149.7
LDL-C	rs7640978	3	32533010	t	-0.032	0.004	4.7E-20	0.0002	104.4
LDL-C	rs2251219	3	52584787	t	-0.014	0.002	7.6E-11	0.0001	54.9
LDL-C	rs13315871	3	58381287	a	-0.032	0.004	3.2E-19	0.0002	101.4
LDL-C	rs1979848	3	132165178	a	0.026	0.004	2.3E-09	0.0001	69.6
LDL-C	rs10513551	3	160086055	t	-0.015	0.002	1.4E-10	0.0001	73.0
LDL-C	rs3748034	4	3446091	t	0.018	0.003	2.4E-09	0.0001	49.1
LDL-C	rs3816873	4	100504664	t	0.014	0.002	4.1E-09	0.0001	43.3
LDL-C	rs13107325	4	103188709	t	-0.029	0.004	9.9E-13	0.0001	76.5
LDL-C	rs870992	5	52193237	a	-0.027	0.004	1.8E-10	0.0001	71.5
LDL-C	rs10062361	5	74565153	t	0.025	0.003	1.9E-13	0.0002	132.1
LDL-C	rs3846662	5	74651084	a	-0.057	0.002	2.1E-131	0.0016	980.6
LDL-C	rs11955819	5	74782412	a	-0.058	0.008	4.8E-13	0.0002	108.9
LDL-C	rs4530754	5	122855416	a	0.016	0.002	1.8E-14	0.0001	79.1
LDL-C	rs10065787	5	131436486	t	-0.019	0.003	2.2E-09	0.0002	109.2
LDL-C	rs2522056	5	131801726	a	-0.018	0.002	1.2E-13	0.0001	70.1
LDL-C	rs4704825	5	156382308	a	-0.028	0.003	5.1E-25	0.0004	221.9
LDL-C	rs2235215	6	16131156	t	0.025	0.003	1.8E-18	0.0003	163.8
LDL-C	rs1800562	6	26093141	a	-0.049	0.004	6.6E-29	0.0003	159.9
LDL-C	rs129128	6	26125342	t	0.021	0.004	3.4E-09	0.0001	62.5
LDL-C	rs2249741	6	31240712	a	-0.020	0.003	7.8E-14	0.0002	126.2
LDL-C	rs13192471	6	32671103	t	-0.034	0.003	5.8E-33	0.0003	179.2
LDL-C	rs3800406	6	35133074	a	0.028	0.004	1.1E-12	0.0002	95.6
LDL-C	rs1129187	6	42932200	t	-0.012	0.002	8.5E-10	0.0001	45.9

LDL-C	rs2239619	6	52453220	a	0.016	0.002	4.0E-14	0.0001	73.3
LDL-C	rs17789218	6	100600097	t	0.022	0.003	4.0E-16	0.0002	109.7
LDL-C	rs9390698	6	101296389	a	0.013	0.002	1.3E-10	0.0001	50.0
LDL-C	rs3798236	6	116309649	t	0.018	0.003	1.8E-13	0.0002	93.5
LDL-C	rs9376090	6	135411228	t	0.028	0.002	5.6E-33	0.0003	172.8
LDL-C	rs1044418	6	139229872	t	0.018	0.003	1.2E-10	0.0001	52.6
LDL-C	rs12208357	6	160543148	t	0.062	0.004	2.7E-52	0.0005	319.8
LDL-C	rs34130495	6	160560824	a	0.048	0.007	2.5E-13	0.0001	66.9
LDL-C	rs62440901	6	160569068	t	0.036	0.004	1.1E-20	0.0003	199.5
LDL-C	rs3798220	6	160961137	t	-0.136	0.008	1.2E-64	0.0007	410.3
LDL-C	rs10455872	6	161010118	a	-0.088	0.006	4.6E-54	0.0010	610.7
LDL-C	rs12175867	6	161019138	t	0.025	0.003	1.8E-14	0.0002	145.5
LDL-C	rs1652507	6	161082461	t	0.027	0.003	1.2E-19	0.0002	125.7
LDL-C	rs10263252	7	1049949	a	-0.023	0.003	6.3E-12	0.0002	107.4
LDL-C	rs1997243	7	1083777	a	-0.016	0.003	6.7E-09	0.0001	43.9
LDL-C	rs144787122	7	2296552	a	-0.104	0.018	4.0E-09	0.0001	44.1
LDL-C	rs2282889	7	21476188	a	-0.016	0.003	5.0E-09	0.0001	80.4
LDL-C	rs12670798	7	21607352	t	-0.029	0.002	1.4E-34	0.0003	196.1
LDL-C	rs4722551	7	25991826	t	-0.039	0.003	3.2E-42	0.0004	256.4
LDL-C	rs2391211	7	26008233	t	0.023	0.004	7.7E-11	0.0002	106.7
LDL-C	rs4302748	7	36191699	a	0.015	0.003	1.2E-09	0.0001	44.7
LDL-C	rs35803101	7	44578500	a	-0.139	0.017	1.1E-15	0.0001	82.8
LDL-C	rs10260606	7	44584551	c	0.037	0.003	1.2E-32	0.0004	260.1
LDL-C	rs1014283	7	87076587	a	-0.019	0.003	2.0E-09	0.0001	63.9
LDL-C	rs330093	8	9175958	c	0.029	0.003	2.0E-20	0.0003	187.1
LDL-C	rs11774381	8	9183339	t	-0.029	0.003	1.9E-24	0.0003	203.9
LDL-C	rs11782386	8	9201787	t	-0.025	0.003	8.5E-13	0.0001	72.3
LDL-C	rs4921914	8	18272438	t	-0.018	0.002	4.9E-14	0.0001	73.3
LDL-C	rs9298506	8	55437524	a	-0.022	0.003	8.4E-16	0.0002	92.9
LDL-C	rs2081687	8	59388565	t	0.027	0.002	2.5E-37	0.0003	202.4
LDL-C	rs2737245	8	116658583	t	-0.018	0.003	2.7E-09	0.0001	76.6
LDL-C	rs2954029	8	126490972	a	0.024	0.003	4.1E-18	0.0003	178.7
LDL-C	rs4870941	8	126498828	c	0.044	0.004	3.3E-33	0.0007	422.8
LDL-C	rs2954038	8	126507389	a	-0.037	0.003	8.5E-32	0.0006	344.9
LDL-C	rs3780181	9	2640759	a	0.033	0.004	4.1E-17	0.0001	89.4
LDL-C	rs67710536	9	19376255	a	-0.027	0.003	3.7E-15	0.0001	80.5
LDL-C	rs10757272	9	22088260	t	-0.020	0.003	2.9E-10	0.0002	117.8
LDL-C	rs3905000	9	107657070	a	-0.018	0.003	6.7E-10	0.0001	51.0
LDL-C	rs635634	9	136155000	t	0.075	0.003	1.5E-182	0.0018	1094.2
LDL-C	rs3812594	9	139368953	a	-0.014	0.002	1.6E-09	0.0001	44.9
LDL-C	rs7080366	10	17254832	t	0.019	0.003	8.5E-15	0.0002	115.9
LDL-C	rs41274050	10	52573772	t	0.077	0.011	4.8E-12	0.0002	142.2
LDL-C	rs2068888	10	94839642	a	-0.017	0.002	1.1E-17	0.0001	89.7
LDL-C	rs2274224	10	96039597	c	-0.014	0.002	3.0E-09	0.0001	61.7
LDL-C	rs2792751	10	113940329	t	0.026	0.002	1.1E-32	0.0003	176.5
LDL-C	rs1891110	10	124610027	a	0.021	0.002	3.0E-26	0.0002	137.3
LDL-C	rs4752805	11	48018355	a	-0.015	0.002	7.0E-10	0.0001	50.7
LDL-C	rs174449	11	61640379	a	0.027	0.003	1.1E-26	0.0003	204.8
LDL-C	rs2521567	11	61699055	a	-0.015	0.002	5.9E-10	0.0001	66.6

LDL-C	rs3816492	11	66297363	t	-0.017	0.002	3.4E-12	0.0001	61.4
LDL-C	rs11603023	11	118486067	t	0.013	0.002	3.0E-10	0.0001	47.9
LDL-C	rs7941030	11	122522375	t	-0.014	0.002	5.7E-12	0.0001	55.0
LDL-C	rs10893500	11	126250774	t	-0.043	0.003	4.0E-36	0.0004	271.7
LDL-C	rs1521516	12	51055708	t	-0.016	0.003	1.2E-10	0.0001	73.2
LDL-C	rs61754230	12	72179446	t	0.052	0.008	2.3E-11	0.0001	55.4
LDL-C	rs3184504	12	111884608	t	-0.026	0.002	1.4E-34	0.0003	204.5
LDL-C	rs1169288	12	121416650	a	-0.035	0.002	1.1E-55	0.0005	326.9
LDL-C	rs10773003	12	123775127	a	0.024	0.004	3.1E-09	0.0001	56.7
LDL-C	rs11571836	13	32973439	a	0.021	0.003	4.1E-10	0.0002	94.1
LDL-C	rs3742318	13	33017043	t	0.020	0.003	9.2E-16	0.0001	78.6
LDL-C	rs4773173	13	111025118	a	0.016	0.003	5.4E-11	0.0001	75.4
LDL-C	rs9646133	14	71096344	t	-0.019	0.002	2.0E-20	0.0002	101.4
LDL-C	rs13379043	14	74250126	t	0.014	0.002	5.5E-11	0.0001	52.7
LDL-C	rs28929474	14	94844947	t	0.071	0.008	5.0E-19	0.0002	102.5
LDL-C	rs3812945	15	75289722	t	-0.015	0.002	4.1E-10	0.0001	68.7
LDL-C	rs35259348	16	72003952	c	0.020	0.003	7.7E-13	0.0001	90.1
LDL-C	rs7197453	16	72079127	c	0.025	0.004	1.5E-12	0.0003	177.3
LDL-C	rs217181	16	72114002	t	-0.049	0.003	1.3E-48	0.0008	492.6
LDL-C	rs9302635	16	72144174	t	0.055	0.004	1.9E-41	0.0009	559.9
LDL-C	rs28555129	16	83984776	a	0.013	0.002	1.1E-09	0.0001	48.7
LDL-C	rs8069974	17	4670972	c	0.015	0.003	3.4E-09	0.0001	62.2
LDL-C	rs314253	17	7091650	t	0.020	0.002	4.6E-22	0.0002	115.1
LDL-C	rs871841	17	8216468	t	-0.015	0.002	2.5E-13	0.0001	66.1
LDL-C	rs6502640	17	18122485	a	0.019	0.003	3.6E-09	0.0001	75.4
LDL-C	rs704	17	26694861	a	0.020	0.002	2.2E-23	0.0002	124.4
LDL-C	rs12601110	17	27035335	a	0.027	0.005	4.9E-09	0.0001	71.0
LDL-C	rs1487971	17	28572753	t	-0.016	0.002	8.3E-11	0.0001	71.9
LDL-C	rs11080150	17	29629326	a	0.015	0.002	2.3E-11	0.0001	57.7
LDL-C	rs72836561	17	41926126	t	-0.037	0.006	2.5E-10	0.0001	51.2
LDL-C	rs4968318	17	45451894	a	0.022	0.002	1.7E-27	0.0002	142.7
LDL-C	rs118004742	17	45468858	t	0.029	0.005	2.2E-09	0.0001	52.4
LDL-C	rs12939848	17	65370808	t	-0.014	0.002	5.6E-09	0.0001	56.8
LDL-C	rs12602912	17	65870073	t	0.016	0.002	7.2E-11	0.0001	46.9
LDL-C	rs77542162	17	67081278	a	-0.177	0.009	1.9E-84	0.0011	667.1
LDL-C	rs4968839	17	67125840	t	0.039	0.003	5.8E-47	0.0006	395.5
LDL-C	rs72852601	17	67149972	t	0.048	0.008	2.5E-09	0.0001	48.2
LDL-C	rs2886232	17	67150176	t	0.041	0.005	6.6E-17	0.0003	202.3
LDL-C	rs4485425	17	73767437	a	-0.019	0.002	2.2E-16	0.0002	94.0
LDL-C	rs4129767	17	76403984	a	0.016	0.002	5.1E-15	0.0001	75.7
LDL-C	rs77960347	18	47109955	a	-0.080	0.009	1.1E-17	0.0002	104.4
LDL-C	rs7241918	18	47160953	t	0.019	0.003	9.0E-12	0.0001	60.0
LDL-C	rs941408	19	2814181	t	0.016	0.003	3.7E-10	0.0001	67.5
LDL-C	rs1982074	19	10668673	a	0.023	0.003	3.2E-18	0.0002	97.1
LDL-C	rs892010	19	11038843	c	0.040	0.005	2.3E-13	0.0002	120.0
LDL-C	rs10417443	19	11129429	c	0.032	0.003	1.5E-31	0.0005	322.9
LDL-C	rs1122608	19	11163601	t	-0.048	0.003	9.6E-63	0.0008	511.3
LDL-C	rs4300767	19	11163689	a	-0.095	0.006	3.8E-63	0.0017	1063.1
LDL-C	rs6511721	19	11206575	a	-0.037	0.003	1.1E-31	0.0007	425.7

LDL-C	rs73015030	19	11207516	a	-0.079	0.009	4.8E-20	0.0004	223.7
LDL-C	rs3745677	19	11211077	a	0.077	0.008	4.0E-20	0.0007	432.9
LDL-C	rs11669576	19	11222300	a	0.064	0.007	3.6E-22	0.0004	238.5
LDL-C	rs45508991	19	11233886	t	0.106	0.015	3.5E-13	0.0001	82.0
LDL-C	rs5927	19	11233941	a	-0.030	0.004	7.9E-15	0.0003	208.9
LDL-C	rs2569538	19	11238548	a	-0.061	0.007	2.1E-20	0.0006	389.1
LDL-C	rs892115	19	11263650	t	-0.023	0.004	2.3E-10	0.0002	143.1
LDL-C	rs6511727	19	11315817	t	0.020	0.003	1.8E-15	0.0002	119.4
LDL-C	rs4804579	19	11358700	t	0.024	0.004	2.8E-10	0.0002	99.9
LDL-C	rs58542926	19	19379549	t	-0.098	0.004	7.5E-146	0.0013	816.7
LDL-C	rs150090162	19	44536189	a	0.161	0.019	1.0E-16	0.0002	115.4
LDL-C	rs8103315	19	45254168	a	0.028	0.004	2.0E-11	0.0002	102.2
LDL-C	rs35106910	19	45284266	a	0.032	0.005	1.3E-11	0.0001	58.6
LDL-C	rs1135062	19	45322744	a	-0.017	0.002	4.5E-13	0.0001	73.4
LDL-C	rs3852856	19	45361574	a	0.061	0.005	5.9E-38	0.0012	747.4
LDL-C	rs12610605	19	45370838	a	0.126	0.005	4.9E-164	0.0043	2678.6
LDL-C	rs8104483	19	45372354	t	-0.087	0.004	3.4E-116	0.0031	1927.4
LDL-C	rs6859	19	45382034	a	0.078	0.003	8.7E-132	0.0030	1842.4
LDL-C	rs11669338	19	45382984	t	0.050	0.006	8.0E-16	0.0004	255.7
LDL-C	rs3852861	19	45383061	t	-0.056	0.004	1.1E-45	0.0015	950.4
LDL-C	rs187706273	19	45385488	a	-0.108	0.016	4.4E-11	0.0001	76.1
LDL-C	rs157580	19	45395266	a	0.075	0.003	2.2E-113	0.0027	1659.5
LDL-C	rs157582	19	45396219	t	-0.032	0.005	7.3E-11	0.0004	226.2
LDL-C	rs115881343	19	45403216	t	0.195	0.009	6.8E-94	0.0018	1136.5
LDL-C	rs10119	19	45406673	a	0.067	0.004	1.9E-53	0.0018	1103.7
LDL-C	rs405509	19	45408836	t	0.173	0.004	0.0E+00	0.0149	9325.2
LDL-C	rs769450	19	45410444	a	0.060	0.005	2.9E-37	0.0018	1093.3
LDL-C	rs769452	19	45411110	t	-0.149	0.020	4.3E-14	0.0001	71.4
LDL-C	rs439401	19	45414451	t	0.029	0.003	4.6E-22	0.0004	249.3
LDL-C	rs59325138	19	45416291	t	0.098	0.004	1.0E-119	0.0046	2845.8
LDL-C	rs732841	19	46207810	a	0.041	0.006	6.0E-13	0.0002	103.2
LDL-C	rs17651629	19	46406463	t	-0.032	0.004	1.3E-17	0.0002	127.7
LDL-C	rs492602	19	49206417	a	-0.028	0.002	4.3E-40	0.0004	238.7
LDL-C	rs641738	19	54676763	t	0.014	0.002	1.0E-09	0.0001	59.7
LDL-C	rs35350976	19	59023174	a	-0.017	0.003	7.2E-09	0.0001	51.8
LDL-C	rs2143544	20	17789221	t	-0.016	0.003	5.5E-09	0.0001	80.9
LDL-C	rs2745865	20	17847735	t	0.039	0.004	2.1E-26	0.0004	239.8
LDL-C	rs6058302	20	34290037	t	-0.027	0.003	1.8E-15	0.0002	106.0
LDL-C	rs6016373	20	39154095	a	0.023	0.002	9.2E-30	0.0002	150.9
LDL-C	rs926663	20	39245775	a	0.014	0.003	8.3E-09	0.0001	62.1
LDL-C	rs6072328	20	39913996	t	-0.026	0.003	1.7E-22	0.0003	203.1
LDL-C	rs6062343	20	62695931	a	-0.015	0.002	2.9E-13	0.0001	65.6
LDL-C	rs2833487	21	33087863	a	-0.035	0.006	4.3E-10	0.0001	71.1
LDL-C	rs2183573	21	40574305	a	-0.014	0.002	4.0E-10	0.0001	61.7
LDL-C	rs138777	22	35711098	a	0.013	0.002	2.3E-09	0.0001	45.0
LDL-C	rs738409	22	44324727	c	0.015	0.002	6.8E-10	0.0001	47.2
LDL-C	rs13268	22	45996298	a	0.042	0.007	3.6E-10	0.0001	48.6
TG	rs1077514	1	23766233	t	0.021	0.003	2.4E-14	0.0001	71.0
TG	*rs16826069*	1	39797055	a	-0.024	0.002	5.4E-23	0.0002	116.7

TG	rs2055491	1	50852769	t	-0.012	0.002	3.6E-09	0.0001	43.0
TG	rs10889353	1	63118196	a	0.075	0.002	2.0E-280	0.0025	1552.9
TG	rs2613503	1	72839774	a	0.020	0.003	1.5E-11	0.0001	74.0
TG	rs12740374	1	109817590	t	-0.015	0.002	1.8E-10	0.0001	48.9
TG	rs12043350	1	153854380	t	0.015	0.003	9.4E-10	0.0001	61.4
TG	rs1011731	1	172346548	a	-0.013	0.002	4.1E-11	0.0001	52.6
TG	rs78444298	1	184672098	a	-0.052	0.009	3.0E-09	0.0001	51.3
TG	rs2821231	1	203518382	t	0.016	0.003	3.9E-09	0.0001	77.8
TG	rs765751	1	219669226	t	-0.020	0.002	2.3E-17	0.0002	109.9
TG	rs10489615	1	230304988	a	0.039	0.002	3.2E-85	0.0007	461.5
TG	rs2273967	1	230415293	t	-0.018	0.003	8.8E-11	0.0001	69.0
TG	rs1473886	2	20368519	t	-0.017	0.002	1.2E-13	0.0001	89.8
TG	rs1801701	2	21228827	t	-0.030	0.004	2.2E-17	0.0001	89.0
TG	rs676210	2	21231524	a	-0.076	0.002	3.7E-211	0.0020	1208.9
TG	rs541041	2	21294975	a	0.023	0.003	1.2E-18	0.0002	102.0
TG	rs3208747	2	24431127	t	-0.150	0.022	6.3E-12	0.0001	66.5
TG	rs1049817	2	27550967	a	0.046	0.003	5.2E-49	0.0010	623.5
TG	rs11689803	2	27566520	a	-0.021	0.004	1.0E-09	0.0002	111.0
TG	rs11891554	2	27613617	a	-0.096	0.006	7.2E-63	0.0008	472.4
TG	rs79593977	2	27702663	a	0.269	0.029	2.0E-20	0.0013	806.8
TG	rs780090	2	27718474	t	0.048	0.005	1.9E-23	0.0004	237.5
TG	rs147073127	2	27726437	a	-0.213	0.019	2.7E-28	0.0003	162.7
TG	rs814295	2	27743215	a	0.077	0.004	9.3E-90	0.0016	982.0
TG	rs1919128	2	27801759	a	-0.046	0.003	1.4E-47	0.0008	499.0
TG	rs115289288	2	28006500	t	-0.222	0.019	1.4E-31	0.0022	1336.3
TG	rs4245791	2	44074431	t	-0.017	0.002	1.1E-14	0.0001	75.1
TG	rs1861410	2	58933591	t	-0.016	0.003	2.0E-09	0.0001	74.3
TG	rs2723062	2	65280220	a	-0.023	0.003	1.6E-17	0.0002	151.0
TG	rs2049019	2	66671858	a	-0.014	0.002	9.5E-09	0.0001	50.4
TG	rs13396091	2	146371961	a	0.014	0.002	2.0E-09	0.0001	57.8
TG	rs13389219	2	165528876	t	-0.035	0.002	2.1E-62	0.0006	364.9
TG	rs16849863	2	165728290	t	0.056	0.008	1.6E-12	0.0001	71.7
TG	rs3769823	2	202122995	a	-0.014	0.002	3.2E-10	0.0001	50.0
TG	rs6435161	2	203519783	t	0.015	0.002	1.4E-09	0.0001	55.5
TG	rs1344642	2	219555262	a	-0.013	0.002	5.1E-11	0.0001	52.5
TG	rs2943650	2	227105921	t	0.042	0.003	2.9E-48	0.0008	523.1
TG	rs1801282	3	12393125	c	0.034	0.003	3.7E-26	0.0002	149.4
TG	rs17819328	3	12489342	t	-0.025	0.002	3.3E-26	0.0003	191.2
TG	rs13326165	3	52532118	a	-0.017	0.003	1.4E-11	0.0001	56.1
TG	rs7621025	3	136272246	t	-0.021	0.003	4.1E-10	0.0002	96.8
TG	rs4683438	3	142652559	t	-0.017	0.002	8.1E-13	0.0001	82.8
TG	rs382534	3	155547274	t	0.015	0.003	4.5E-09	0.0001	54.4
TG	rs9822326	3	156803565	a	0.016	0.002	3.8E-11	0.0001	73.7
TG	rs10513687	3	170725730	t	0.022	0.003	6.7E-11	0.0001	67.4
TG	rs17600346	3	172223982	t	-0.046	0.007	3.3E-11	0.0002	101.7
TG	rs6599389	4	939113	a	0.022	0.004	6.9E-09	0.0001	45.5
TG	rs11248060	4	964359	t	0.022	0.003	6.1E-13	0.0001	64.6
TG	rs3748034	4	3446091	t	0.028	0.003	2.9E-19	0.0002	119.8

TG	rs16844401	4	3449652	a	0.032	0.004	1.0E-14	0.0001	80.5
TG	rs6831256	4	3473139	a	-0.014	0.002	1.7E-11	0.0001	60.4
TG	rs9884830	4	26027797	t	0.019	0.003	2.4E-10	0.0001	62.0
TG	rs1037814	4	88049850	t	-0.024	0.002	2.8E-24	0.0003	177.9
TG	rs10029254	4	88160140	t	0.021	0.003	1.1E-13	0.0002	93.7
TG	rs13133548	4	89740128	a	0.012	0.002	3.3E-09	0.0001	43.2
TG	rs1126673	4	100045616	t	0.018	0.002	5.6E-16	0.0001	81.2
TG	rs13107325	4	103188709	t	0.033	0.004	1.7E-15	0.0002	95.3
TG	rs41278045	4	110638764	a	-0.199	0.029	4.0E-12	0.0001	58.7
TG	rs6054	4	155489608	t	0.123	0.018	7.2E-12	0.0001	67.4
TG	rs6855363	4	157670537	t	0.017	0.002	5.9E-12	0.0001	74.3
TG	rs4311394	5	53300662	a	-0.020	0.002	3.4E-20	0.0002	98.2
TG	rs459193	5	55806751	a	-0.035	0.002	3.7E-52	0.0005	300.7
TG	rs2448428	5	55844049	t	0.015	0.002	4.3E-10	0.0001	68.3
TG	rs9686661	5	55861786	t	0.044	0.003	2.3E-67	0.0006	374.1
TG	rs4976033	5	67714246	a	-0.016	0.002	1.2E-15	0.0001	78.1
TG	rs1045241	5	118729286	t	-0.017	0.003	3.7E-11	0.0001	71.3
TG	rs26008	5	131008194	t	0.024	0.004	2.3E-10	0.0001	49.1
TG	rs4705986	5	132349654	t	0.034	0.006	4.4E-09	0.0001	84.1
TG	rs4704820	5	156334681	t	0.022	0.003	1.0E-11	0.0001	88.6
TG	rs6882076	5	156390297	t	-0.042	0.002	6.7E-83	0.0008	501.2
TG	rs1650527	5	158022724	t	0.025	0.003	5.1E-16	0.0002	138.9
TG	rs2524060	6	31267422	a	-0.029	0.003	3.3E-20	0.0003	185.6
TG	rs2442719	6	31320538	t	-0.024	0.002	7.5E-32	0.0003	178.9
TG	rs17207867	6	31938412	t	0.027	0.004	5.0E-13	0.0001	70.2
TG	rs9271366	6	32586854	a	0.028	0.003	7.4E-19	0.0002	119.2
TG	rs9273368	6	32626475	a	-0.018	0.003	9.3E-09	0.0001	78.8
TG	rs11752643	6	32669373	t	0.066	0.006	3.7E-26	0.0003	155.8
TG	rs2395655	6	36645696	a	0.017	0.002	6.6E-13	0.0001	80.0
TG	rs6458349	6	43759789	a	0.038	0.003	4.1E-37	0.0006	353.7
TG	rs78807370	6	43761091	a	-0.043	0.004	3.9E-31	0.0005	293.1
TG	rs881858	6	43806609	a	0.020	0.002	1.8E-20	0.0002	107.2
TG	rs4715316	6	52628998	t	-0.016	0.003	8.5E-09	0.0001	68.8
TG	rs2745353	6	127452935	t	0.019	0.002	1.9E-20	0.0002	106.0
TG	rs9388768	6	130374102	a	0.013	0.002	1.2E-09	0.0001	45.4
TG	rs643381	6	139839423	a	-0.024	0.002	3.2E-32	0.0003	172.5
TG	rs12208357	6	160543148	t	0.030	0.004	1.1E-13	0.0001	75.1
TG	rs2665357	6	160848167	a	-0.018	0.002	1.9E-15	0.0002	104.3
TG	rs645718	6	161406239	a	-0.034	0.005	2.1E-10	0.0001	62.5
TG	rs12699758	7	15964238	a	0.016	0.003	3.6E-09	0.0001	58.1
TG	rs4410790	7	17284577	t	-0.012	0.002	1.4E-09	0.0001	43.2
TG	rs10235225	7	25905599	a	0.015	0.002	1.3E-09	0.0001	64.9
TG	rs4722551	7	25991826	t	0.019	0.003	1.1E-11	0.0001	63.7
TG	rs1534696	7	26397239	a	-0.019	0.002	2.9E-15	0.0002	110.6
TG	rs2070971	7	44197583	t	0.023	0.003	7.2E-12	0.0001	84.4
TG	rs3757838	7	44231310	a	0.030	0.005	3.8E-11	0.0001	71.5
TG	rs1178979	7	72856430	t	0.044	0.004	1.1E-30	0.0006	353.9
TG	rs799158	7	73019074	t	0.076	0.008	4.0E-21	0.0004	273.6
TG	rs3812316	7	73020337	c	0.087	0.005	4.3E-82	0.0016	987.5
TG	rs287621	7	130435181	t	0.022	0.003	4.7E-19	0.0002	123.0

TG	rs3735080	7	150217309	t	0.016	0.002	2.6E-11	0.0001	56.2
TG	rs16884656	7	150512307	t	-0.024	0.004	8.3E-09	0.0001	75.6
TG	rs4240624	8	9184231	a	-0.034	0.004	1.2E-17	0.0002	108.7
TG	rs11776767	8	10683929	c	0.024	0.002	1.3E-27	0.0003	160.1
TG	rs2686187	8	11654796	a	-0.014	0.002	3.7E-09	0.0001	60.5
TG	rs3947	8	11702375	a	0.019	0.002	7.1E-15	0.0001	86.5
TG	rs1495741	8	18272881	a	-0.036	0.002	3.4E-56	0.0005	296.3
TG	rs1801177	8	19805708	a	0.095	0.009	1.1E-26	0.0003	202.0
TG	rs264	8	19813180	a	-0.028	0.003	7.9E-17	0.0002	120.1
TG	rs268	8	19813529	a	-0.232	0.008	5.5E-202	0.0017	1058.9
TG	rs301	8	19816934	t	0.062	0.005	2.8E-36	0.0015	904.3
TG	rs312	8	19817997	c	0.048	0.006	1.4E-13	0.0005	302.0
TG	rs326	8	19819439	a	0.078	0.005	6.7E-63	0.0026	1582.2
TG	rs12545984	8	19847259	t	0.044	0.006	2.1E-13	0.0005	316.6
TG	rs17091905	8	19849757	a	-0.042	0.006	1.5E-13	0.0004	237.7
TG	rs10105418	8	19875365	a	0.082	0.009	1.9E-21	0.0003	208.6
TG	rs4637851	8	19922610	a	-0.029	0.003	8.5E-22	0.0004	236.0
TG	rs13256965	8	19962962	a	0.017	0.002	7.5E-12	0.0001	84.7
TG	rs3736147	8	22471824	a	0.018	0.003	1.7E-10	0.0001	86.9
TG	rs1982768	8	25898565	a	-0.022	0.004	3.0E-09	0.0001	74.8
TG	rs2081687	8	59388565	t	0.019	0.002	9.5E-20	0.0002	102.6
TG	rs4738141	8	72469742	a	-0.017	0.003	1.6E-09	0.0001	67.1
TG	rs17730649	8	126465305	a	0.021	0.003	3.8E-16	0.0002	137.4
TG	rs6982502	8	126479362	t	-0.028	0.005	5.1E-09	0.0004	238.0
TG	rs2980876	8	126481694	t	-0.091	0.005	1.4E-73	0.0036	2230.5
TG	rs8180991	8	126500350	c	-0.034	0.003	2.6E-24	0.0004	260.5
TG	rs4871624	8	126629328	t	-0.020	0.003	1.0E-14	0.0002	100.1
TG	rs3927680	9	16887366	a	-0.016	0.002	1.2E-15	0.0001	78.1
TG	rs4120895	9	33787532	t	-0.018	0.003	4.3E-11	0.0001	92.3
TG	rs1800978	9	107665978	c	0.027	0.004	2.9E-13	0.0002	101.9
TG	rs17134592	10	5260682	c	0.021	0.003	4.3E-10	0.0001	64.6
TG	rs41274050	10	52573772	t	0.094	0.011	2.7E-17	0.0003	209.2
TG	rs7923609	10	65133822	a	0.030	0.003	1.2E-21	0.0004	270.8
TG	rs2298117	10	70346740	t	0.012	0.002	1.0E-09	0.0001	45.7
TG	rs7901016	10	74637326	t	-0.040	0.004	3.5E-20	0.0002	114.7
TG	rs10748579	10	94090498	a	-0.018	0.002	1.9E-18	0.0002	95.4
TG	rs7081888	10	94764660	t	-0.033	0.005	1.7E-11	0.0002	97.4
TG	rs2068888	10	94839642	a	-0.037	0.002	5.8E-71	0.0007	415.5
TG	rs2792751	10	113940329	t	-0.018	0.002	7.5E-16	0.0001	81.0
TG	rs11195943	10	114154815	t	-0.023	0.004	2.0E-10	0.0001	60.3
TG	rs10886863	10	122929493	t	-0.058	0.010	4.3E-09	0.0002	110.1
TG	rs7940646	11	10669228	t	-0.015	0.002	4.4E-12	0.0001	59.5
TG	rs10832027	11	13357183	a	0.016	0.002	1.8E-11	0.0001	68.7
TG	rs546383	11	18065663	t	-0.013	0.002	7.6E-09	0.0001	52.6
TG	rs925946	11	27667202	t	0.016	0.002	7.6E-14	0.0001	68.9
TG	rs326214	11	47298360	a	0.022	0.002	4.1E-26	0.0002	130.6
TG	rs2727271	11	61603358	a	-0.065	0.003	1.8E-78	0.0009	586.7
TG	rs174587	11	61612830	t	0.048	0.003	6.9E-50	0.0008	467.7

TG	rs35169799	11	64031241	t	0.041	0.004	4.8E-24	0.0002	120.4
TG	rs4014195	11	65506822	c	-0.015	0.002	2.6E-12	0.0001	60.8
TG	rs2229738	11	68562328	t	0.024	0.004	1.6E-10	0.0001	50.8
TG	rs11237471	11	78082604	t	-0.022	0.003	1.7E-10	0.0001	83.7
TG	rs4938289	11	116458785	t	0.047	0.006	1.3E-14	0.0003	164.7
TG	rs12799766	11	116558427	a	0.042	0.005	3.7E-19	0.0006	377.5
TG	rs74360954	11	116582542	t	-0.113	0.008	1.5E-43	0.0013	804.6
TG	rs2000571	11	116585533	a	0.063	0.006	1.5E-26	0.0013	811.0
TG	rs180357	11	116599504	t	-0.049	0.005	4.2E-24	0.0010	626.6
TG	rs4938307	11	116604514	a	-0.073	0.007	4.7E-28	0.0011	683.7
TG	rs61730763	11	116631482	a	0.190	0.023	2.9E-16	0.0002	128.9
TG	rs17120029	11	116650118	t	0.142	0.009	1.2E-56	0.0025	1576.5
TG	rs11604424	11	116651115	t	-0.066	0.006	1.5E-27	0.0015	930.8
TG	rs619054	11	116660813	a	-0.057	0.004	1.1E-36	0.0012	716.3
TG	rs143292359	11	116661001	a	0.241	0.040	1.5E-09	0.0001	57.6
TG	rs662799	11	116663707	a	-0.106	0.008	1.2E-43	0.0015	914.6
TG	rs9804646	11	116665079	t	-0.060	0.006	2.8E-23	0.0006	355.6
TG	rs5104	11	116692334	t	-0.035	0.004	1.6E-17	0.0003	190.7
TG	rs11216157	11	116711180	a	0.055	0.007	1.2E-16	0.0007	434.1
TG	rs888246	11	116724232	t	0.071	0.006	9.5E-29	0.0009	550.3
TG	rs2075292	11	116732512	t	-0.063	0.005	8.4E-40	0.0009	545.8
TG	rs11216168	11	116741553	a	-0.069	0.006	5.3E-27	0.0012	726.5
TG	rs2000615	11	116915819	t	0.057	0.006	4.0E-21	0.0007	430.3
TG	rs490262	11	117222592	a	-0.019	0.003	2.2E-13	0.0001	67.1
TG	rs7134375	12	20473758	a	-0.014	0.002	7.4E-13	0.0001	61.9
TG	rs4149056	12	21331549	t	-0.032	0.003	3.3E-30	0.0003	159.4
TG	rs718314	12	26453283	a	-0.019	0.003	1.4E-14	0.0001	87.3
TG	rs7979398	12	46086708	t	0.015	0.002	9.3E-11	0.0001	68.1
TG	rs11613352	12	57792580	t	-0.025	0.003	5.8E-20	0.0002	131.4
TG	rs2075260	12	109696838	a	0.015	0.003	4.6E-09	0.0001	41.5
TG	rs3742004	12	111798553	a	0.016	0.003	9.8E-09	0.0001	53.4
TG	rs940904	12	123491572	a	0.015	0.003	4.2E-09	0.0001	55.3
TG	rs11057408	12	124464836	t	-0.022	0.002	2.7E-20	0.0002	138.5
TG	rs10846744	12	125312425	c	0.027	0.003	1.3E-15	0.0002	121.2
TG	rs2298058	13	95248566	t	0.024	0.003	5.4E-17	0.0003	157.7
TG	rs7400722	13	114527838	a	0.016	0.002	1.9E-12	0.0001	71.6
TG	rs7157785	14	64235556	t	0.020	0.003	7.1E-13	0.0001	71.3
TG	rs11634257	15	40388492	a	0.018	0.003	1.5E-09	0.0001	74.5
TG	rs17747633	15	40916237	a	-0.015	0.002	4.6E-12	0.0001	63.2
TG	rs16949992	15	44238869	c	-0.075	0.006	5.7E-34	0.0005	293.5
TG	rs11858955	15	44246293	a	-0.115	0.016	3.8E-13	0.0002	133.3
TG	rs493258	15	58687880	t	0.017	0.002	1.4E-17	0.0001	89.9
TG	rs12913346	15	63530965	a	0.024	0.004	4.1E-11	0.0001	83.5
TG	rs17184382	15	63792486	a	0.017	0.002	3.9E-13	0.0001	88.9
TG	rs2415168	15	73109629	a	0.015	0.003	3.2E-09	0.0001	55.8
TG	rs10152471	15	101890913	a	-0.016	0.003	4.9E-09	0.0001	74.6
TG	rs143076454	16	921179	a	0.048	0.008	6.8E-09	0.0001	48.6
TG	rs11075253	16	15148646	a	-0.027	0.003	2.6E-15	0.0003	185.3
TG	rs2032915	16	31117413	t	-0.017	0.002	5.4E-14	0.0001	87.0

TG	rs9939609	16	53820527	a	0.019	0.002	2.0E-21	0.0002	106.8
TG	rs1800775	16	56995236	a	-0.023	0.002	3.4E-27	0.0003	162.2
TG	rs7203984	16	56999258	a	-0.022	0.003	5.2E-12	0.0002	97.0
TG	rs9940315	16	69876164	a	-0.019	0.003	4.7E-13	0.0002	107.5
TG	rs2000999	16	72108093	a	0.020	0.003	7.3E-15	0.0001	81.3
TG	rs12443634	16	81524274	a	0.018	0.003	9.3E-10	0.0001	84.4
TG	rs1053328	16	85711860	t	0.014	0.002	8.8E-09	0.0001	54.3
TG	rs3853818	17	7346302	t	0.016	0.002	6.5E-12	0.0001	75.9
TG	rs897453	17	17425631	t	-0.014	0.002	1.1E-12	0.0001	61.2
TG	rs1563631	17	18221134	t	-0.017	0.002	6.7E-15	0.0001	75.2
TG	rs3110454	17	28651363	t	-0.017	0.003	7.4E-10	0.0001	83.3
TG	rs2306590	17	34854280	a	-0.013	0.002	5.0E-10	0.0001	50.3
TG	rs2079005	17	41865627	t	-0.022	0.003	6.2E-15	0.0002	104.3
TG	rs1662750	17	42011823	a	-0.018	0.003	1.4E-11	0.0002	95.0
TG	rs2074108	17	42336149	t	-0.015	0.002	1.1E-09	0.0001	64.6
TG	rs11871606	17	45732774	a	0.016	0.002	3.4E-16	0.0001	82.2
TG	rs8075803	17	47346529	t	-0.019	0.002	1.3E-16	0.0002	111.7
TG	rs12602912	17	65870073	t	0.023	0.002	1.2E-21	0.0002	100.9
TG	rs2125345	17	73782191	t	0.013	0.002	1.8E-09	0.0001	46.5
TG	rs2292642	17	76395430	t	-0.021	0.002	4.3E-25	0.0002	125.4
TG	rs1652343	18	21131929	t	0.016	0.003	2.0E-09	0.0001	75.1
TG	rs17178414	19	4945250	t	0.019	0.003	1.0E-10	0.0002	97.1
TG	rs1799816	19	7125518	t	-0.068	0.011	6.4E-10	0.0001	47.6
TG	rs7248104	19	7224431	a	-0.019	0.002	1.5E-21	0.0002	108.6
TG	rs116843064	19	8429323	a	-0.265	0.007	3.8E-295	0.0036	2223.6
TG	rs140744493	19	8436373	t	-0.121	0.019	1.5E-10	0.0001	50.5
TG	rs1862644	19	18724315	a	-0.017	0.003	2.2E-09	0.0001	86.6
TG	rs117877390	19	19378416	t	-0.103	0.011	1.7E-21	0.0006	357.1
TG	rs10401969	19	19407718	t	0.095	0.004	5.2E-150	0.0013	824.6
TG	rs145702982	19	19579726	a	0.119	0.021	6.2E-09	0.0003	172.3
TG	rs8182584	19	33909710	t	0.017	0.002	2.7E-15	0.0001	81.6
TG	rs1688030	19	35556744	t	-0.033	0.005	5.5E-13	0.0001	78.8
TG	rs2018519	19	35559787	t	-0.021	0.003	8.9E-11	0.0001	74.7
TG	rs28399653	19	45315445	a	0.041	0.006	5.5E-12	0.0001	60.1
TG	rs4803760	19	45333834	t	0.021	0.003	1.3E-11	0.0001	83.5
TG	rs157582	19	45396219	t	0.042	0.004	7.5E-24	0.0006	383.4
TG	rs439401	19	45414451	t	-0.080	0.002	1.2E-229	0.0030	1869.3
TG	rs59325138	19	45416291	t	-0.040	0.003	1.2E-31	0.0008	469.6
TG	rs7259004	19	45432557	c	0.064	0.005	2.3E-45	0.0008	463.7
TG	rs2287922	19	49232226	a	0.018	0.002	4.0E-17	0.0002	96.4
TG	rs1132990	19	50028163	a	-0.018	0.003	4.2E-09	0.0001	63.0
TG	rs6029143	20	39118662	t	-0.035	0.005	8.6E-13	0.0001	92.4
TG	rs6016381	20	39180436	t	0.018	0.002	8.4E-15	0.0001	92.0
TG	rs1997833	20	39690342	t	-0.016	0.003	1.2E-09	0.0001	61.0
TG	rs3827066	20	44586023	t	-0.043	0.004	4.3E-28	0.0005	308.0
TG	rs8123864	20	44598670	t	-0.044	0.003	2.0E-46	0.0009	535.9
TG	rs1211644	20	45592842	t	0.024	0.003	2.8E-15	0.0002	131.0
TG	rs2426428	20	50886412	t	-0.031	0.005	2.3E-11	0.0001	92.2

TG	rs6025606	20	56098733	t	-0.013	0.002	7.4E-10	0.0001	48.3
TG	rs41302559	20	56140439	a	-0.145	0.021	1.8E-12	0.0001	61.9
TG	rs114139997	21	46875775	a	-0.365	0.039	2.9E-21	0.0003	180.4
TG	rs200559406	21	46875817	a	0.282	0.036	6.6E-15	0.0001	78.5
TG	rs35665085	22	17625915	a	0.032	0.005	1.9E-12	0.0001	61.4
TG	rs738322	22	38569006	a	0.021	0.002	9.2E-26	0.0002	135.9
TG	rs5757161	22	38990662	a	0.016	0.003	1.6E-10	0.0001	72.5
TG	rs738409	22	44324727	c	0.017	0.002	4.0E-12	0.0001	59.6

Supplementary Table 3. Trait-specific genetic instruments for blood lipid levels selected from the Global Lipids Genetics Consortium (GLGC) dataset. Variants were selected on the basis of their association with the respective trait at p<5E-8 and a p>0.01 regarding their association with the other two traits.

			Associations with HDL-C			Associations with LDL-C			Associations with TG		
Phenotype	**SNP**	**Eff_allele**	**Effect**	**SE**	**P-value**	**Effect**	**SE**	**P-value**	**Effect**	**SE**	**P-value**
HDL-C	rs103294	t	0.052	0.004	4.00E-30	0.007	0.005	0.123	-0.002	0.004	0.752
HDL-C	rs10773105	t	-0.036	0.004	3.20E-24	0.006	0.004	0.122	0.004	0.003	0.509
HDL-C	rs11246602	c	0.034	0.005	1.68E-10	0.002	0.006	0.526	-0.009	0.005	0.192
HDL-C	rs12226802	g	0.033	0.005	1.29E-09	0	0.005	0.619	-0.007	0.005	0.23
HDL-C	rs16942887	a	0.083	0.005	8.28E-54	0.001	0.005	0.798	-0.012	0.005	0.0296
HDL-C	rs17695224	g	0.029	0.004	2.42E-13	0.011	0.004	0.0125	-0.012	0.004	0.0113
HDL-C	rs181362	c	0.038	0.004	9.24E-18	0.007	0.005	0.0793	0.009	0.004	0.0281
HDL-C	rs205262	a	0.028	0.004	3.88E-13	0.009	0.004	0.0313	-0.003	0.004	0.803
HDL-C	rs2240327	g	0.024	0.003	1.11E-11	0.001	0.004	0.971	-0.002	0.003	0.867
HDL-C	rs2241210	g	0.033	0.004	2.49E-20	0.008	0.004	0.0855	0.003	0.003	0.247
HDL-C	rs2290547	a	-0.03	0.005	3.69E-09	0.001	0.005	0.793	0.01	0.004	0.0221
HDL-C	rs2472509	g	0.023	0.004	1.21E-09	0	0.004	0.708	-0.002	0.004	0.722
HDL-C	rs2602836	g	-0.019	0.003	4.96E-08	0.001	0.004	0.831	0.009	0.003	0.0212
HDL-C	rs4650994	a	-0.021	0.003	6.70E-09	0.003	0.004	0.338	0.002	0.003	0.398
HDL-C	rs4917014	g	0.022	0.004	1.03E-08	0.005	0.004	0.246	-0.001	0.004	0.887
HDL-C	rs4983559	g	0.02	0.004	9.57E-09	0.003	0.004	0.583	0	0.004	0.971
HDL-C	rs499974	a	-0.026	0.004	1.12E-08	0.001	0.005	0.826	-0.009	0.004	0.0541
HDL-C	rs702485	g	0.024	0.003	6.45E-12	0.001	0.004	0.787	-0.002	0.003	0.475
HDL-C	rs838876	g	-0.049	0.004	7.33E-33	0.003	0.004	0.442	0.005	0.004	0.377
LDL-C	rs1010167	g	-0.004	0.004	0.396	0.025	0.004	6.22E-11	0.002	0.004	0.808
LDL-C	rs11563251	t	0.006	0.006	0.365	0.035	0.006	4.50E-08	0.008	0.006	0.0826
LDL-C	rs1250229	c	-0.003	0.004	0.404	0.024	0.004	3.13E-08	0.009	0.004	0.0139
LDL-C	rs12670798	c	-0.001	0.004	0.733	0.034	0.004	4.81E-14	0.01	0.004	0.0168
LDL-C	rs16831243	t	0.011	0.005	0.039	0.038	0.006	9.06E-12	-0.001	0.005	0.987
LDL-C	rs17508045	t	-0.009	0.006	0.0466	0.049	0.007	4.91E-12	-0.008	0.006	0.4
LDL-C	rs1800562	g	0.007	0.007	0.242	0.062	0.008	8.25E-14	-0.013	0.007	0.172
LDL-C	rs2030746	t	-0.003	0.004	0.306	0.021	0.004	8.61E-09	0.003	0.004	0.491
LDL-C	rs2294261	a	-0.009	0.004	0.0206	0.033	0.004	6.57E-17	0.002	0.003	0.587
LDL-C	rs2328223	c	0	0.005	0.859	0.03	0.005	5.63E-09	-0.007	0.005	0.115
LDL-C	rs314253	t	-0.003	0.004	0.353	0.024	0.004	3.44E-10	0.009	0.003	0.0298
LDL-C	rs364585	g	0.001	0.004	0.822	0.025	0.004	4.28E-10	-0.002	0.003	0.44
LDL-C	rs3780181	a	0.004	0.007	0.542	0.045	0.007	1.76E-09	-0.007	0.007	0.491
LDL-C	rs4148218	g	-0.003	0.004	0.456	0.044	0.005	6.76E-21	0.004	0.004	0.295
LDL-C	rs4530754	a	0.001	0.003	0.934	0.028	0.004	3.58E-12	0.002	0.003	0.742
LDL-C	rs6065311	c	0.002	0.003	0.437	0.042	0.004	1.66E-30	0.006	0.003	0.0227
LDL-C	rs6489818	a	0	0.005	0.928	0.028	0.005	4.57E-09	-0.004	0.004	0.54
LDL-C	rs6603981	t	0.004	0.004	0.381	0.034	0.004	3.10E-13	0.007	0.004	0.174
LDL-C	rs7225700	c	0.01	0.004	0.0235	0.03	0.004	3.56E-13	-0.005	0.004	0.236
LDL-C	rs7703051	a	0.002	0.004	0.421	0.073	0.004	1.40E-77	0.006	0.003	0.163
LDL-C	rs7832643	t	-0.001	0.004	0.595	0.034	0.004	2.67E-17	0.002	0.003	0.472
LDL-C	rs7832643	t	-0.001	0.004	0.595	0.034	0.004	2.67E-17	0.002	0.003	0.472
LDL-C	rs8017377	a	-0.004	0.004	0.434	0.03	0.004	2.52E-15	0.006	0.004	0.142
LDL-C	rs8176720	t	0.001	0.004	0.943	0.033	0.004	1.59E-17	-0.007	0.004	0.0609
LDL-C	rs903319	c	0.01	0.004	0.0122	0.027	0.004	5.22E-11	-0.005	0.004	0.138

TG	rs10029254	t	-0.009	0.004	0.0487	0.006	0.004	0.205	0.027	0.004	7.55E-09
TG	rs1781930	g	0.002	0.005	0.625	0.01	0.005	0.057	0.031	0.004	2.51E-11
TG	rs603446	c	-0.002	0.004	0.873	0.009	0.004	0.0114	0.05	0.003	3.92E-43
TG	rs9693857	c	0.004	0.004	0.527	0.005	0.004	0.298	-0.02	0.003	1.69E-08

Supplementary Table 4. Genetic instruments for the circulating cholesterol and triglyceride concentrations of the lipoprotein particles as selected and extracted from the dataset of the GWAS meta-analysis on Nuclear Magnetic Resonance (NMR)-measured circulating metabolites.

Metabolite	SNP	Chr	Position (hg18)	Eff_allele	Effect	SE	P-value	R2	F
L.HDL.C	rs1077835	15	58723426	G	0.181	0.012	1.39E-53	0.0122	265.5
L.HDL.C	rs11076174	16	57003146	C	-0.134	0.016	1.17E-16	0.0035	76.7
L.HDL.C	rs11076176	16	57007446	G	-0.171	0.014	1.62E-34	0.0081	176.2
L.HDL.C	rs111543310	15	59531818	C	0.338	0.049	6.88E-12	0.0036	69.2
L.HDL.C	rs112835635	15	59351989	G	0.263	0.035	2.13E-13	0.0031	63.2
L.HDL.C	rs112884731	15	59504897	C	0.544	0.057	2.75E-21	0.0055	98.4
L.HDL.C	rs113298164	15	58855748	T	0.584	0.047	2.74E-34	0.0092	165.4
L.HDL.C	rs116142092	15	59751872	T	0.383	0.050	5.61E-14	0.0034	61.4
L.HDL.C	rs11633043	15	58837722	A	0.078	0.014	2.57E-08	0.0017	36.4
L.HDL.C	rs117901517	15	58678869	C	-0.142	0.023	5.22E-10	0.0030	58.2
L.HDL.C	rs1318175	15	58586129	T	-0.087	0.013	5.41E-11	0.0022	47.1
L.HDL.C	rs1367117	2	21263900	A	-0.063	0.011	1.50E-08	0.0016	35.1
L.HDL.C	rs1373657	15	58717762	T	0.165	0.030	4.42E-08	0.0021	38.2
L.HDL.C	rs138690293	15	59310760	C	0.722	0.107	2.61E-11	0.0039	62.4
L.HDL.C	rs142855631	15	59286876	T	0.737	0.108	1.33E-11	0.0039	63.1
L.HDL.C	rs146842281	15	59356659	T	0.154	0.022	3.53E-12	0.0027	52.8
L.HDL.C	rs148902553	15	59776836	C	0.388	0.051	3.48E-14	0.0036	62.1
L.HDL.C	rs16940472	15	58835317	A	0.130	0.020	2.49E-10	0.0026	56.9
L.HDL.C	rs16940810	15	59115159	T	0.252	0.030	5.86E-17	0.0038	77.2
L.HDL.C	rs17301746	15	58731395	T	0.265	0.038	5.18E-12	0.0029	63.1
L.HDL.C	rs174583	11	61609750	T	-0.078	0.010	2.92E-14	0.0029	62.4
L.HDL.C	rs17821274	15	58684478	C	-0.086	0.010	3.91E-16	0.0033	72.3
L.HDL.C	rs17821298	15	58690738	A	0.092	0.012	7.20E-15	0.0030	65.7
L.HDL.C	rs1800777	16	57017319	A	-0.234	0.035	4.04E-11	0.0027	51.6
L.HDL.C	rs181412360	15	59158953	C	0.378	0.038	6.01E-23	0.0059	106.4
L.HDL.C	rs182776276	15	59254589	G	0.573	0.060	2.84E-21	0.0056	96.2
L.HDL.C	rs183975744	15	59052479	T	0.747	0.120	7.21E-10	0.0028	43.1
L.HDL.C	rs185241689	15	59143155	G	0.810	0.114	1.96E-12	0.0039	64.0
L.HDL.C	rs185481	15	58666679	C	0.069	0.010	2.68E-11	0.0024	51.0
L.HDL.C	rs186924495	20	44686926	T	-0.207	0.037	3.38E-08	0.0018	32.6
L.HDL.C	rs1883025	9	107664301	T	-0.072	0.012	9.85E-09	0.0016	35.1
L.HDL.C	rs189375934	15	60196526	G	0.310	0.053	6.61E-09	0.0022	38.1
L.HDL.C	rs189418461	15	59725202	G	0.379	0.050	5.52E-14	0.0034	61.2
L.HDL.C	rs192630343	15	59286102	A	0.708	0.107	5.45E-11	0.0038	62.1
L.HDL.C	rs247617	16	56990716	A	0.210	0.011	1.93E-82	0.0184	403.3
L.HDL.C	rs261291	15	58680178	C	0.179	0.010	2.39E-68	0.0151	329.4
L.HDL.C	rs28370984	15	58629308	C	0.177	0.032	3.98E-08	0.0017	33.7
L.HDL.C	rs291	8	19815852	C	0.101	0.012	1.45E-17	0.0036	77.8
L.HDL.C	rs34718390	15	58682690	A	0.156	0.024	6.31E-11	0.0028	61.5
L.HDL.C	rs35547826	15	58720405	A	-0.084	0.014	7.63E-09	0.0017	37.6
L.HDL.C	rs429358	19	45411941	C	-0.092	0.013	1.46E-11	0.0024	51.1
L.HDL.C	rs435306	20	44538484	T	-0.073	0.011	2.88E-10	0.0020	42.2
L.HDL.C	rs517755	19	45009036	C	-0.229	0.041	3.58E-08	0.0019	36.8
L.HDL.C	rs55995508	16	56827946	A	0.159	0.025	4.93E-10	0.0023	44.6
L.HDL.C	rs6065904	20	44534651	A	-0.135	0.012	1.72E-30	0.0065	140.2

L.HDL.C	rs6499857	16	56935090	C	-0.067	0.012	3.70E-08	0.0015	32.8
L.HDL.C	rs6507939	18	47176261	C	0.095	0.013	3.34E-12	0.0024	51.2
L.HDL.C	rs6544366	2	21204025	T	0.064	0.011	8.14E-09	0.0016	34.8
L.HDL.C	rs67053123	12	125353810	A	0.092	0.015	3.90E-10	0.0021	46.3
L.HDL.C	rs73959582	18	47148886	C	0.079	0.014	4.62E-08	0.0016	35.2
L.HDL.C	rs7412	19	45412079	T	0.158	0.025	8.48E-10	0.0026	48.2
L.HDL.C	rs75835816	8	19885513	C	-0.293	0.038	2.87E-14	0.0033	63.3
L.HDL.C	rs76083992	20	44544798	T	-0.229	0.032	2.01E-12	0.0026	51.8
L.HDL.C	rs76116860	15	59834938	C	0.277	0.041	2.66E-11	0.0027	52.2
L.HDL.C	rs79844529	15	58445279	T	0.190	0.032	3.43E-09	0.0024	45.8
L.HDL.C	rs8042174	15	58685970	C	-0.112	0.020	2.10E-08	0.0018	38.0
L.HDL.C	rs938507	15	58582034	A	0.101	0.014	2.89E-12	0.0024	51.2
L.HDL.C	rs964184	11	116648917	C	0.082	0.014	8.07E-09	0.0016	34.5
M.LDL.C	rs10424477	19	10636051	T	-0.071	0.011	1.53E-09	0.0021	46.4
M.LDL.C	rs111740198	19	44878217	A	-0.321	0.052	1.22E-09	0.0033	48.4
M.LDL.C	rs112635299	14	94838142	T	0.237	0.040	4.03E-09	0.0020	39.4
M.LDL.C	rs11587071	1	55522674	T	-0.092	0.014	1.95E-11	0.0023	49.1
M.LDL.C	rs116054287	1	56401689	C	-0.397	0.038	4.41E-25	0.0058	126.0
M.LDL.C	rs117261169	19	45491032	T	-0.361	0.055	1.44E-10	0.0025	48.6
M.LDL.C	rs117569256	19	45423330	G	-0.821	0.107	4.27E-14	0.0068	91.9
M.LDL.C	rs11878174	19	45723379	C	0.073	0.012	6.11E-10	0.0026	51.0
M.LDL.C	rs12043403	1	55431933	C	-0.138	0.018	3.27E-14	0.0038	73.8
M.LDL.C	rs12086676	1	55738663	T	-0.076	0.013	2.68E-09	0.0018	38.8
M.LDL.C	rs12916	5	74656539	C	0.084	0.010	1.86E-16	0.0034	73.7
M.LDL.C	rs137992968	19	11239696	T	-0.204	0.034	3.32E-09	0.0018	39.1
M.LDL.C	rs138270540	4	75353427	C	0.226	0.036	9.95E-10	0.0025	46.3
M.LDL.C	rs138287365	4	74781004	C	0.373	0.050	2.70E-13	0.0037	68.3
M.LDL.C	rs138525976	1	55960656	A	-0.103	0.018	2.24E-08	0.0017	35.7
M.LDL.C	rs140339333	4	75396456	A	0.288	0.045	2.32E-10	0.0030	55.3
M.LDL.C	rs140411770	19	45356517	A	-0.542	0.088	1.58E-09	0.0049	76.2
M.LDL.C	rs142130958	19	11190652	A	-0.221	0.016	2.19E-40	0.0092	200.6
M.LDL.C	rs143413051	4	75560225	T	0.381	0.055	1.05E-11	0.0034	57.9
M.LDL.C	rs143736900	4	72871285	C	0.594	0.083	2.35E-12	0.0045	71.5
M.LDL.C	rs144591518	19	10518992	T	-0.194	0.034	3.05E-08	0.0016	34.0
M.LDL.C	rs144721118	1	54196340	A	-0.270	0.040	1.94E-11	0.0034	65.0
M.LDL.C	rs146568567	1	54824117	A	-0.315	0.032	1.24E-22	0.0054	104.9
M.LDL.C	rs147319495	2	20912953	G	-0.064	0.011	8.09E-09	0.0017	36.9
M.LDL.C	rs147825223	19	45479553	C	0.173	0.028	1.92E-09	0.0022	39.8
M.LDL.C	rs148359521	2	21414212	T	-0.187	0.032	8.31E-09	0.0020	39.0
M.LDL.C	rs148382396	1	54639713	A	-0.390	0.051	5.53E-14	0.0044	70.1
M.LDL.C	rs149048538	19	45053024	A	-0.282	0.044	1.91E-10	0.0025	47.8
M.LDL.C	rs149844719	1	54519237	T	-0.183	0.028	1.18E-10	0.0023	50.7
M.LDL.C	rs149944945	1	56129361	G	-0.292	0.034	3.47E-17	0.0043	82.3
M.LDL.C	rs150785555	1	56005603	A	-0.451	0.036	1.28E-35	0.0090	175.6
M.LDL.C	rs150966173	19	45421204	T	0.225	0.039	1.01E-08	0.0020	39.4
M.LDL.C	rs150985779	19	45147992	T	-0.269	0.038	2.86E-12	0.0031	59.1
M.LDL.C	rs151193598	4	73303394	A	0.634	0.087	8.45E-13	0.0061	82.8
M.LDL.C	rs157594	19	45425175	G	0.133	0.012	4.31E-29	0.0086	167.0

M.LDL.C	rs17111503	1	55503448	G	0.075	0.013	1.34E-08	0.0018	39.6
M.LDL.C	rs17395160	1	55085141	G	-0.086	0.012	2.42E-12	0.0025	53.1
M.LDL.C	rs180961170	1	57012269	G	-0.364	0.052	4.27E-12	0.0037	65.6
M.LDL.C	rs181066897	4	73499882	C	0.515	0.080	2.30E-10	0.0041	66.1
M.LDL.C	rs181169081	2	21312870	A	-0.185	0.032	1.07E-08	0.0020	38.2
M.LDL.C	rs181594442	1	57006537	A	-0.364	0.052	4.33E-12	0.0037	65.6
M.LDL.C	rs181847072	4	73134560	G	0.592	0.083	1.85E-12	0.0046	72.3
M.LDL.C	rs182300850	1	54389320	C	-0.371	0.060	1.06E-09	0.0032	48.4
M.LDL.C	rs182318839	19	45747128	T	0.300	0.054	3.77E-08	0.0027	47.3
M.LDL.C	rs184566992	19	44887996	T	-0.353	0.052	2.52E-11	0.0037	58.6
M.LDL.C	rs184650103	4	74850649	T	0.406	0.047	2.78E-17	0.0045	77.6
M.LDL.C	rs185049786	4	74644512	C	0.335	0.054	1.08E-09	0.0034	62.6
M.LDL.C	rs185415345	1	56625395	A	-0.168	0.027	1.48E-09	0.0025	48.2
M.LDL.C	rs185802315	19	10777054	G	-0.231	0.036	2.50E-10	0.0025	44.8
M.LDL.C	rs185886292	19	45565918	T	0.184	0.032	1.51E-08	0.0021	37.3
M.LDL.C	rs186538116	1	56840574	C	-0.421	0.045	1.49E-20	0.0059	105.9
M.LDL.C	rs188099946	19	45189605	T	-0.296	0.045	1.06E-10	0.0022	48.6
M.LDL.C	rs189409600	19	45341066	T	-0.749	0.103	8.32E-13	0.0060	83.6
M.LDL.C	rs189718275	19	45063850	A	-0.296	0.044	3.75E-11	0.0028	53.2
M.LDL.C	rs190217562	4	75180409	C	0.251	0.035	1.36E-12	0.0059	106.0
M.LDL.C	rs190934192	1	55334001	A	-0.385	0.040	2.23E-21	0.0069	134.8
M.LDL.C	rs191210370	1	54236244	G	-0.212	0.038	3.42E-08	0.0021	40.7
M.LDL.C	rs191404723	1	54636232	T	-0.390	0.051	4.44E-14	0.0041	71.1
M.LDL.C	rs191448950	1	55584844	A	-0.484	0.032	1.80E-51	0.0111	243.0
M.LDL.C	rs192012905	19	44463485	G	-0.340	0.053	1.93E-10	0.0029	52.4
M.LDL.C	rs192570155	1	55246601	C	-0.492	0.045	4.04E-27	0.0085	152.5
M.LDL.C	rs193084249	1	26987646	G	0.182	0.031	1.04E-08	0.0021	39.7
M.LDL.C	rs2007708	19	45410420	A	-0.831	0.104	3.41E-15	0.0054	99.4
M.LDL.C	rs207176	1	55791846	T	0.121	0.017	2.14E-12	0.0026	55.4
M.LDL.C	rs2207132	20	39142516	A	0.137	0.025	3.69E-08	0.0021	40.8
M.LDL.C	rs2927472	19	45349369	C	0.151	0.018	6.13E-17	0.0041	88.8
M.LDL.C	rs2965149	19	45190766	C	-0.069	0.011	1.57E-10	0.0022	47.7
M.LDL.C	rs2967668	19	45302951	G	-0.178	0.017	4.32E-25	0.0071	137.2
M.LDL.C	rs2980875	8	126481747	G	-0.058	0.010	5.86E-09	0.0017	36.1
M.LDL.C	rs312030	2	21462743	C	0.108	0.018	1.25E-09	0.0019	41.9
M.LDL.C	rs3185010	19	11275842	A	0.064	0.011	5.50E-09	0.0017	37.0
M.LDL.C	rs34722314	2	21271707	A	-0.124	0.014	6.82E-18	0.0037	80.2
M.LDL.C	rs3741298	11	116657561	T	-0.072	0.012	1.49E-09	0.0018	39.1
M.LDL.C	rs429358	19	45411941	C	0.220	0.013	2.46E-59	0.0136	298.2
M.LDL.C	rs4609471	1	55493584	A	-0.382	0.030	8.83E-37	0.0113	220.5
M.LDL.C	rs4803748	19	45247048	T	-0.080	0.011	1.32E-13	0.0030	65.5
M.LDL.C	rs55810502	5	74380959	G	0.070	0.012	8.93E-09	0.0017	36.5
M.LDL.C	rs58826447	19	45328379	A	0.070	0.011	1.05E-10	0.0022	48.3
M.LDL.C	rs62117161	19	45233385	G	-0.206	0.020	1.98E-24	0.0054	117.5
M.LDL.C	rs62120794	2	21100426	T	-0.177	0.024	4.41E-13	0.0028	56.8
M.LDL.C	rs629301	1	109818306	T	0.126	0.012	1.62E-25	0.0055	118.9
M.LDL.C	rs6511721	19	11206575	A	-0.092	0.011	6.23E-17	0.0042	91.7
M.LDL.C	rs6663252	1	55630151	C	-0.084	0.012	1.01E-11	0.0024	51.1

M.LDL.C	rs6732011	2	21146521	T	-0.059	0.010	6.73E-09	0.0016	35.4
M.LDL.C	rs6859	19	45382034	G	-0.080	0.010	1.10E-15	0.0032	69.1
M.LDL.C	rs7255743	19	46018119	A	-0.178	0.032	4.50E-08	0.0016	35.0
M.LDL.C	rs73048351	19	45160086	A	-0.415	0.064	2.47E-10	0.0030	58.3
M.LDL.C	rs73066442	7	21592973	G	0.071	0.011	8.03E-10	0.0019	40.9
M.LDL.C	rs73556990	19	44888175	G	-0.326	0.052	8.98E-10	0.0036	50.7
M.LDL.C	rs73564218	19	45665952	C	0.173	0.030	1.27E-08	0.0020	35.4
M.LDL.C	rs74073060	1	55638930	A	-0.478	0.037	1.28E-36	0.0098	190.3
M.LDL.C	rs7412	19	45412079	T	-0.565	0.025	#######	0.0338	638.4
M.LDL.C	rs75647206	1	56947591	T	-0.365	0.050	6.08E-13	0.0035	67.7
M.LDL.C	rs7604788	2	21190024	T	-0.176	0.022	3.01E-15	0.0033	72.3
M.LDL.C	rs76670936	19	45196581	A	-0.145	0.018	1.18E-14	0.0034	74.4
M.LDL.C	rs76866386	2	44075483	C	-0.142	0.018	1.42E-14	0.0029	63.2
M.LDL.C	rs77021821	4	75684215	T	0.251	0.041	1.71E-09	0.0023	42.2
M.LDL.C	rs78620068	2	21524000	A	-0.115	0.017	7.72E-11	0.0022	47.6
M.LDL.C	rs79668907	19	11257169	T	-0.091	0.012	7.58E-13	0.0031	66.3
M.LDL.C	rs79890446	19	45723446	T	-0.234	0.033	2.89E-12	0.0034	65.8
M.LDL.C	rs8106814	19	45441608	C	-0.101	0.014	4.47E-12	0.0035	68.2
M.LDL.C	rs8111962	19	10915324	T	-0.095	0.014	1.59E-11	0.0023	49.6
M.LDL.C	rs934197	2	21267461	A	0.113	0.011	3.50E-24	0.0052	113.4
M.LDL.C	rs984976	5	74910870	G	0.075	0.010	1.30E-13	0.0027	59.0
IDL.TG	rs10495713	2	21200519	G	-0.062	0.010	2.48E-09	0.0019	37.2
IDL.TG	rs113105798	15	59301460	A	0.198	0.036	4.06E-08	0.0018	35.2
IDL.TG	rs113298164	15	58855748	T	0.395	0.047	2.42E-16	0.0042	75.3
IDL.TG	rs113531395	17	4886829	T	-0.246	0.036	2.44E-11	0.0031	59.4
IDL.TG	rs114822153	4	73238544	A	0.244	0.043	1.68E-08	0.0025	48.0
IDL.TG	rs115849089	8	19912370	A	-0.098	0.017	2.05E-08	0.0017	33.7
IDL.TG	rs116054287	1	56401689	C	-0.282	0.038	5.00E-13	0.0031	60.0
IDL.TG	rs116302332	4	75370891	T	0.257	0.040	1.90E-10	0.0028	54.7
IDL.TG	rs11633043	15	58837722	A	0.079	0.014	4.38E-08	0.0018	34.4
IDL.TG	rs116802199	17	4801101	C	-0.235	0.032	3.08E-13	0.0031	58.1
IDL.TG	rs1168041	1	62960250	C	0.084	0.012	1.96E-12	0.0028	53.9
IDL.TG	rs117749052	15	58749309	C	0.216	0.037	9.78E-09	0.0027	51.9
IDL.TG	rs118095054	19	19621301	G	-0.137	0.024	1.34E-08	0.0022	42.5
IDL.TG	rs12043403	1	55431933	C	-0.101	0.018	3.23E-08	0.0020	39.2
IDL.TG	rs1268353	11	116639692	T	-0.064	0.011	1.66E-09	0.0020	37.9
IDL.TG	rs1318175	15	58586129	T	-0.088	0.014	2.89E-10	0.0022	42.6
IDL.TG	rs13329672	15	58699937	T	0.086	0.012	9.82E-13	0.0029	55.6
IDL.TG	rs138195472	15	58672107	T	0.223	0.035	3.10E-10	0.0025	44.3
IDL.TG	rs138287365	4	74781004	C	0.407	0.049	3.37E-16	0.0044	85.3
IDL.TG	rs140250995	4	73723860	C	0.636	0.100	3.37E-10	0.0035	64.6
IDL.TG	rs140339333	4	75396456	A	0.309	0.044	4.64E-12	0.0034	65.8
IDL.TG	rs143413051	4	75560225	T	0.404	0.055	3.05E-13	0.0037	66.9
IDL.TG	rs143736900	4	72871285	C	0.692	0.082	1.08E-16	0.0060	100.2
IDL.TG	rs145347194	15	58670135	C	0.135	0.023	3.51E-09	0.0024	46.2
IDL.TG	rs146568567	1	54824117	A	-0.208	0.032	1.02E-10	0.0024	45.7
IDL.TG	rs146842281	15	59356659	T	0.146	0.022	6.31E-11	0.0024	47.1
IDL.TG	rs149297353	19	20115517	G	-0.192	0.030	2.02E-10	0.0026	50.9

IDL.TG	rs149944945	1	56129361	G	-0.248	0.034	9.23E-13	0.0031	59.1
IDL.TG	rs150392353	2	21320317	C	-0.228	0.032	1.93E-12	0.0030	58.2
IDL.TG	rs150536132	19	19679560	T	-0.183	0.030	1.13E-09	0.0028	50.4
IDL.TG	rs150785555	1	56005603	A	-0.328	0.036	1.74E-19	0.0048	92.2
IDL.TG	rs151193598	4	73303394	A	0.658	0.085	3.13E-14	0.0066	94.0
IDL.TG	rs1532085	15	58683366	G	-0.156	0.010	8.71E-49	0.0117	229.0
IDL.TG	rs157594	19	45425175	G	0.126	0.012	4.34E-26	0.0077	148.6
IDL.TG	rs1663255	15	58514242	T	-0.071	0.011	2.41E-10	0.0022	42.6
IDL.TG	rs16940213	15	58695337	T	0.126	0.013	1.70E-20	0.0046	89.8
IDL.TG	rs17001002	19	10948031	A	-0.094	0.016	8.44E-09	0.0022	35.2
IDL.TG	rs17216525	19	19662220	T	-0.132	0.021	4.39E-10	0.0021	40.7
IDL.TG	rs181066897	4	73499882	C	0.523	0.080	1.17E-10	0.0042	68.2
IDL.TG	rs181169081	2	21312870	A	-0.227	0.032	2.17E-12	0.0030	57.6
IDL.TG	rs181181625	15	59377940	T	0.388	0.058	3.00E-11	0.0027	48.4
IDL.TG	rs181275587	19	20486755	A	-0.167	0.029	2.02E-08	0.0022	39.0
IDL.TG	rs181412360	15	59158953	C	0.255	0.038	3.52E-11	0.0027	48.4
IDL.TG	rs1815786	11	116921390	C	-0.095	0.016	1.47E-09	0.0021	41.5
IDL.TG	rs181807530	17	4774814	G	-0.232	0.032	9.01E-13	0.0032	55.0
IDL.TG	rs181847072	4	73134560	G	0.692	0.082	5.83E-17	0.0062	102.2
IDL.TG	rs183162020	4	73690263	G	0.861	0.115	1.99E-13	0.0086	85.0
IDL.TG	rs183305631	19	19597444	A	-0.218	0.032	1.54E-11	0.0033	59.0
IDL.TG	rs1838504	15	58666410	T	0.099	0.010	1.20E-20	0.0049	94.4
IDL.TG	rs184650103	4	74850649	T	0.430	0.046	6.76E-20	0.0051	90.4
IDL.TG	rs1848922	2	21471603	C	0.113	0.013	7.08E-19	0.0044	84.5
IDL.TG	rs185049786	4	74644512	C	0.358	0.053	3.00E-11	0.0039	74.6
IDL.TG	rs1872741	15	59450895	T	0.073	0.013	3.91E-08	0.0017	32.2
IDL.TG	rs1883711	20	39179822	C	0.155	0.025	6.67E-10	0.0026	49.6
IDL.TG	rs189741280	19	19624481	G	-0.199	0.030	5.27E-11	0.0029	56.1
IDL.TG	rs190121281	19	19252779	A	-0.231	0.033	3.24E-12	0.0036	63.8
IDL.TG	rs190217562	4	75180409	C	0.327	0.040	1.00E-15	0.0040	72.1
IDL.TG	rs190934192	1	55334001	A	-0.246	0.040	1.37E-09	0.0028	55.0
IDL.TG	rs191448950	1	55584844	A	-0.319	0.032	4.51E-23	0.0053	103.5
IDL.TG	rs192570155	1	55246601	C	-0.335	0.045	2.46E-13	0.0039	70.4
IDL.TG	rs193092110	15	58730460	A	0.206	0.036	1.26E-08	0.0020	35.6
IDL.TG	rs247617	16	56990716	A	-0.084	0.011	4.51E-13	0.0029	56.3
IDL.TG	rs261334	15	58726744	C	-0.197	0.012	1.09E-56	0.0138	270.1
IDL.TG	rs2642636	15	58363242	G	-0.062	0.011	1.49E-08	0.0018	34.5
IDL.TG	rs28370984	15	58629308	C	0.215	0.032	3.09E-11	0.0026	49.7
IDL.TG	rs28395406	15	58629349	G	-0.108	0.016	2.50E-11	0.0026	49.6
IDL.TG	rs2954029	8	126490972	T	-0.083	0.010	1.36E-15	0.0035	67.2
IDL.TG	rs3005923	1	56801542	A	-0.250	0.036	1.13E-11	0.0033	64.7
IDL.TG	rs4075673	2	21150787	C	-0.102	0.010	5.92E-22	0.0050	97.5
IDL.TG	rs429358	19	45411941	C	0.122	0.014	3.02E-18	0.0042	81.1
IDL.TG	rs4609471	1	55493584	A	-0.254	0.030	3.64E-17	0.0050	97.2
IDL.TG	rs61999891	15	58299599	A	0.103	0.017	5.16E-09	0.0022	42.0
IDL.TG	rs6511720	19	11202306	T	-0.176	0.017	1.76E-23	0.0056	107.6
IDL.TG	rs6511721	19	11206575	A	-0.070	0.011	5.20E-10	0.0025	47.4
IDL.TG	rs660240	1	109817838	C	0.079	0.012	5.70E-10	0.0021	40.1

IDL.TG	rs74073060	1	55638930	A	-0.323	0.037	1.80E-17	0.0044	86.1
IDL.TG	rs7412	19	45412079	T	-0.263	0.026	2.70E-23	0.0073	116.9
IDL.TG	rs7604788	2	21190024	T	-0.195	0.022	4.58E-18	0.0044	84.8
IDL.TG	rs77021821	4	75684215	T	0.266	0.040	7.83E-11	0.0025	49.2
IDL.TG	rs79192207	2	21417897	C	0.087	0.014	7.29E-10	0.0021	40.5
IDL.TG	rs79225634	5	74619639	T	0.062	0.011	1.60E-08	0.0018	34.0
IDL.TG	rs79660716	15	58521171	G	0.205	0.027	1.68E-13	0.0035	66.9
IDL.TG	rs8042174	15	58685970	C	-0.132	0.020	2.42E-10	0.0024	45.4
IDL.TG	rs8100204	19	19393714	A	-0.118	0.016	1.88E-12	0.0032	61.1
IDL.TG	rs9302635	16	72144174	C	-0.078	0.013	1.27E-08	0.0017	33.7
IDL.TG	rs964184	11	116648917	C	-0.149	0.015	7.59E-24	0.0054	104.8
L.LDL.C	rs10424477	19	10636051	T	-0.082	0.011	1.82E-12	0.0029	63.0
L.LDL.C	rs10449300	1	109381904	G	-0.060	0.011	3.73E-08	0.0016	34.3
L.LDL.C	rs111740198	19	44878217	A	-0.341	0.052	1.13E-10	0.0037	54.4
L.LDL.C	rs114664261	2	21410015	T	0.264	0.046	2.19E-08	0.0027	52.8
L.LDL.C	rs11587071	1	55522674	T	-0.099	0.014	7.48E-13	0.0026	56.0
L.LDL.C	rs116054287	1	56401689	C	-0.397	0.038	3.95E-25	0.0058	126.0
L.LDL.C	rs117261169	19	45491032	T	-0.396	0.055	1.99E-12	0.0030	58.4
L.LDL.C	rs117569256	19	45423330	G	-0.867	0.107	1.42E-15	0.0076	102.6
L.LDL.C	rs11878174	19	45723379	C	0.077	0.012	9.53E-11	0.0029	55.8
L.LDL.C	rs12043403	1	55431933	C	-0.141	0.018	1.20E-14	0.0039	76.2
L.LDL.C	rs12086676	1	55738663	T	-0.081	0.013	2.28E-10	0.0020	44.0
L.LDL.C	rs13014768	2	21514796	G	-0.117	0.017	2.48E-12	0.0025	54.8
L.LDL.C	rs137992968	19	11239696	T	-0.213	0.034	6.08E-10	0.0020	42.8
L.LDL.C	rs138270540	4	75353427	C	0.220	0.036	1.62E-09	0.0023	45.1
L.LDL.C	rs138287365	4	74781004	C	0.346	0.049	4.07E-12	0.0032	61.6
L.LDL.C	rs138525976	1	55960656	A	-0.107	0.018	7.47E-09	0.0018	38.1
L.LDL.C	rs140339333	4	75396456	A	0.266	0.044	2.49E-09	0.0025	48.8
L.LDL.C	rs140411770	19	45356517	A	-0.519	0.088	7.65E-09	0.0044	69.6
L.LDL.C	rs142130958	19	11190652	A	-0.233	0.016	1.07E-44	0.0102	222.7
L.LDL.C	rs143413051	4	75560225	T	0.363	0.055	6.07E-11	0.0030	53.8
L.LDL.C	rs143736900	4	72871285	C	0.540	0.082	1.08E-10	0.0037	60.7
L.LDL.C	rs144064722	4	73406173	G	0.232	0.034	1.82E-11	0.0027	51.3
L.LDL.C	rs144591518	19	10518992	T	-0.196	0.034	1.88E-08	0.0016	35.0
L.LDL.C	rs144721118	1	54196340	A	-0.292	0.040	3.96E-13	0.0039	75.9
L.LDL.C	rs144900553	19	10798974	T	-0.293	0.052	2.35E-08	0.0022	47.8
L.LDL.C	rs146568567	1	54824117	A	-0.332	0.032	4.40E-25	0.0060	116.9
L.LDL.C	rs147319495	2	20912953	G	-0.066	0.011	3.19E-09	0.0018	38.9
L.LDL.C	rs147825223	19	45479553	C	0.166	0.028	8.18E-09	0.0021	36.6
L.LDL.C	rs148359521	2	21414212	T	-0.194	0.032	2.43E-09	0.0022	41.8
L.LDL.C	rs148382396	1	54639713	A	-0.416	0.051	1.04E-15	0.0050	79.7
L.LDL.C	rs149048538	19	45053024	A	-0.310	0.044	2.45E-12	0.0030	57.7
L.LDL.C	rs149844719	1	54519237	T	-0.203	0.028	8.40E-13	0.0029	62.4
L.LDL.C	rs149944945	1	56129361	G	-0.298	0.034	8.05E-18	0.0044	85.5
L.LDL.C	rs150785555	1	56005603	A	-0.470	0.036	1.20E-38	0.0098	190.9
L.LDL.C	rs150966173	19	45421204	T	0.228	0.039	6.44E-09	0.0021	40.3
L.LDL.C	rs150985779	19	45147992	T	-0.284	0.038	1.53E-13	0.0034	66.0

L.LDL.C	rs151330717	19	45196964	A	-0.329	0.058	2.07E-08	0.0025	48.4
L.LDL.C	rs157594	19	45425175	G	0.138	0.012	3.63E-31	0.0092	179.3
L.LDL.C	rs17111503	1	55503448	G	0.077	0.013	6.09E-09	0.0019	41.4
L.LDL.C	rs180961170	1	57012269	G	-0.380	0.052	4.36E-13	0.0040	71.6
L.LDL.C	rs181169081	2	21312870	A	-0.192	0.032	2.83E-09	0.0021	41.2
L.LDL.C	rs181594442	1	57006537	A	-0.380	0.052	4.36E-13	0.0040	71.6
L.LDL.C	rs181847072	4	73134560	G	0.537	0.082	9.04E-11	0.0037	61.4
L.LDL.C	rs182300850	1	54389320	C	-0.401	0.060	4.12E-11	0.0038	56.6
L.LDL.C	rs183162020	4	73690263	G	0.722	0.115	6.81E-10	0.0060	59.6
L.LDL.C	rs183383492	19	11232974	C	-0.352	0.063	3.52E-08	0.0019	33.5
L.LDL.C	rs184229638	19	45671925	A	-0.688	0.115	3.24E-09	0.0042	50.6
L.LDL.C	rs184566992	19	44887996	T	-0.371	0.052	2.06E-12	0.0041	64.9
L.LDL.C	rs184650103	4	74850649	T	0.381	0.046	5.88E-16	0.0040	71.0
L.LDL.C	rs185049786	4	74644512	C	0.323	0.053	2.07E-09	0.0031	60.6
L.LDL.C	rs185415345	1	56625395	A	-0.171	0.027	6.61E-10	0.0026	50.1
L.LDL.C	rs185802315	19	10777054	G	-0.235	0.036	1.30E-10	0.0026	46.1
L.LDL.C	rs186538116	1	56840574	C	-0.441	0.045	2.06E-22	0.0065	116.1
L.LDL.C	rs188099946	19	45189605	T	-0.276	0.045	1.65E-09	0.0020	42.3
L.LDL.C	rs189409600	19	45341066	T	-0.746	0.103	1.00E-12	0.0059	82.8
L.LDL.C	rs189718275	19	45063850	A	-0.323	0.044	4.55E-13	0.0033	63.6
L.LDL.C	rs190217562	4	75180409	C	0.293	0.040	6.82E-13	0.0032	57.7
L.LDL.C	rs190934192	1	55334001	A	-0.399	0.040	7.10E-23	0.0075	144.8
L.LDL.C	rs191210370	1	54236244	G	-0.236	0.038	7.05E-10	0.0026	50.7
L.LDL.C	rs191404723	1	54636232	T	-0.415	0.051	9.32E-16	0.0046	80.4
L.LDL.C	rs191448950	1	55584844	A	-0.498	0.032	1.03E-54	0.0119	258.4
L.LDL.C	rs192012905	19	44463485	G	-0.362	0.053	1.19E-11	0.0033	59.4
L.LDL.C	rs192570155	1	55246601	C	-0.511	0.045	3.38E-29	0.0092	164.7
L.LDL.C	rs193084249	1	26987646	G	0.180	0.031	1.48E-08	0.0020	38.8
L.LDL.C	rs2007708	19	45410420	A	-0.852	0.104	6.39E-16	0.0057	104.5
L.LDL.C	rs207176	1	55791846	T	0.125	0.017	4.57E-13	0.0027	58.7
L.LDL.C	rs2207132	20	39142516	A	0.142	0.025	1.13E-08	0.0023	43.8
L.LDL.C	rs261334	15	58726744	C	-0.070	0.012	6.63E-09	0.0017	37.2
L.LDL.C	rs2722641	19	44892775	A	-0.262	0.044	5.32E-09	0.0029	52.2
L.LDL.C	rs2954022	8	126482621	A	-0.058	0.010	6.03E-09	0.0017	36.1
L.LDL.C	rs2965149	19	45190766	C	-0.068	0.011	1.82E-10	0.0022	47.3
L.LDL.C	rs2967668	19	45302951	G	-0.187	0.017	1.28E-27	0.0078	151.7
L.LDL.C	rs312030	2	21462743	C	0.101	0.018	1.72E-08	0.0017	36.0
L.LDL.C	rs34042070	16	72101525	G	0.069	0.012	2.68E-08	0.0015	33.1
L.LDL.C	rs34722314	2	21271707	A	-0.138	0.014	1.22E-21	0.0046	98.5
L.LDL.C	rs3741298	11	116657561	T	-0.069	0.012	8.86E-09	0.0016	35.4
L.LDL.C	rs3935470	5	74352180	G	0.058	0.011	4.76E-08	0.0015	32.7
L.LDL.C	rs404935	19	45372794	A	-0.157	0.017	2.82E-19	0.0044	94.4
L.LDL.C	rs429358	19	45411941	C	0.208	0.013	2.54E-53	0.0122	266.6
L.LDL.C	rs4426495	2	21143982	T	-0.064	0.010	3.82E-10	0.0019	41.4
L.LDL.C	rs4609471	1	55493584	A	-0.401	0.030	1.78E-40	0.0125	243.3
L.LDL.C	rs4803748	19	45247048	T	-0.080	0.011	1.64E-13	0.0030	64.9
L.LDL.C	rs4804573	19	11277232	A	-0.057	0.010	2.60E-08	0.0016	35.3
L.LDL.C	rs495828	9	136154867	T	0.072	0.012	1.61E-09	0.0018	38.5

L.LDL.C	rs58446550	19	45328380	A	0.070	0.011	9.41E-11	0.0022	48.4
L.LDL.C	rs58996925	1	56267033	G	-0.062	0.011	2.38E-08	0.0016	34.0
L.LDL.C	rs61770425	1	55085125	G	-0.083	0.012	1.26E-11	0.0023	50.1
L.LDL.C	rs62117161	19	45233385	G	-0.217	0.020	5.38E-27	0.0060	130.5
L.LDL.C	rs62120794	2	21100426	T	-0.185	0.024	3.51E-14	0.0031	62.1
L.LDL.C	rs629301	1	109818306	T	0.128	0.012	4.36E-26	0.0056	121.6
L.LDL.C	rs6511721	19	11206575	A	-0.091	0.011	1.76E-16	0.0041	88.9
L.LDL.C	rs6663252	1	55630151	C	-0.087	0.012	2.33E-12	0.0025	54.2
L.LDL.C	rs6756629	2	44065090	A	-0.141	0.018	2.03E-14	0.0029	62.2
L.LDL.C	rs6859	19	45382034	G	-0.079	0.010	2.22E-15	0.0031	67.6
L.LDL.C	rs7255743	19	46018119	A	-0.183	0.032	1.93E-08	0.0017	36.8
L.LDL.C	rs73048351	19	45160086	A	-0.419	0.064	1.57E-10	0.0031	59.4
L.LDL.C	rs73066442	7	21592973	G	0.073	0.011	2.90E-10	0.0020	43.0
L.LDL.C	rs73556990	19	44888175	G	-0.346	0.052	8.47E-11	0.0040	56.9
L.LDL.C	rs73564218	19	45665952	C	0.167	0.030	4.33E-08	0.0018	32.8
L.LDL.C	rs74073060	1	55638930	A	-0.496	0.037	2.63E-39	0.0105	204.6
L.LDL.C	rs7412	19	45412079	T	-0.573	0.025	#######	0.0347	657.1
L.LDL.C	rs75647206	1	56947591	T	-0.382	0.050	4.90E-14	0.0038	74.1
L.LDL.C	rs7604788	2	21190024	T	-0.183	0.022	2.55E-16	0.0036	77.8
L.LDL.C	rs76488675	1	56885874	G	-0.134	0.024	2.5E-08	0.0020	38.9
L.LDL.C	rs76670936	19	45196581	A	-0.144	0.018	1.9E-14	0.0034	73.1
L.LDL.C	rs77021821	4	75684215	T	0.242	0.040	3.3E-09	0.0021	40.7
L.LDL.C	rs79225634	5	74619639	T	0.086	0.010	4.4E-16	0.0033	72.0
L.LDL.C	rs79668907	19	11257169	T	-0.092	0.012	4E-13	0.0031	67.8
L.LDL.C	rs79890446	19	45723446	T	-0.248	0.033	1.5E-13	0.0038	73.4
L.LDL.C	rs8106814	19	45441608	C	-0.103	0.014	1.6E-12	0.0037	70.9
L.LDL.C	rs8111962	19	10915324	T	-0.102	0.014	4E-13	0.0027	57.4
L.LDL.C	rs934197	2	21267461	A	0.114	0.011	1.7E-24	0.0053	114.8
L.LDL.C	rs9749236	19	45524553	C	-0.441	0.075	8.4E-09	0.0024	45.2
L.LDL.C	rs984976	5	74910870	G	0.071	0.010	1.5E-12	0.0025	53.8
M.HDL.C	rs117040820	16	57005762	T	0.320	0.052	1.1E-09	0.0027	52.4
M.HDL.C	rs1800777	16	57017319	A	-0.233	0.035	5.1E-11	0.0026	50.8
M.HDL.C	rs2126259	8	9185146	C	0.094	0.014	1.4E-10	0.0020	43.1
M.HDL.C	rs247617	16	56990716	A	0.165	0.011	7.6E-52	0.0114	249.2
M.HDL.C	rs286	8	19815256	T	0.124	0.021	4.2E-09	0.0018	39.2
M.HDL.C	rs28888131	16	56991624	A	-0.123	0.013	1.3E-19	0.0042	91.9
M.HDL.C	rs289743	16	57017796	A	-0.060	0.010	1.1E-08	0.0016	34.7
M.HDL.C	rs34932218	8	19855661	G	0.064	0.011	1.2E-08	0.0016	34.2
M.HDL.C	rs429358	19	45411941	C	-0.083	0.013	7.3E-10	0.0020	42.3
M.HDL.C	rs4939883	18	47167214	C	0.073	0.013	2.3E-08	0.0015	32.4
M.HDL.C	rs590820	1	230309619	G	-0.058	0.010	1.7E-08	0.0016	35.3
M.HDL.C	rs7499892	16	57006590	T	-0.171	0.013	1.5E-38	0.0085	184.0
M.HDL.C	rs75835816	8	19885513	C	-0.238	0.038	6.3E-10	0.0022	41.6
IDL.C	rs10424477	19	10636051	T	-0.083	0.012	5.1E-12	0.0029	56.7
IDL.C	rs10449300	1	109381904	G	-0.066	0.011	6E-09	0.0019	37.2
IDL.C	rs111740198	19	44878217	A	-0.327	0.052	6.2E-10	0.0034	50.1
IDL.C	rs114664261	2	21410015	T	-0.259	0.047	4.1E-08	0.0026	50.8
IDL.C	rs11579068	1	55780213	C	-0.106	0.014	9.1E-14	0.0030	57.6

IDL.C	rs116054287	1	56401689	C	-0.400	0.038	1.1E-24	0.0062	120.8
IDL.C	rs117261169	19	45491032	T	-0.367	0.055	6.9E-11	0.0026	50.3
IDL.C	rs11748027	5	74909972	T	0.072	0.010	6.2E-12	0.0026	49.6
IDL.C	rs117569256	19	45423330	G	-0.780	0.107	7.2E-13	0.0061	82.9
IDL.C	rs11878174	19	45723379	C	0.076	0.012	1.3E-10	0.0028	55.0
IDL.C	rs12043403	1	55431933	C	-0.132	0.018	4.5E-13	0.0035	67.1
IDL.C	rs12086676	1	55738663	T	-0.084	0.013	1.8E-10	0.0022	43.2
IDL.C	rs137992968	19	11239696	T	-0.228	0.035	1.3E-10	0.0023	44.4
IDL.C	rs138287365	4	74781004	C	0.305	0.049	1E-09	0.0025	47.8
IDL.C	rs138525976	1	55960656	A	-0.115	0.019	1.6E-09	0.0021	40.7
IDL.C	rs142130958	19	11190652	A	-0.238	0.017	4.2E-42	0.0103	201.3
IDL.C	rs143413051	4	75560225	T	0.313	0.055	1.7E-08	0.0022	40.0
IDL.C	rs144545816	2	21413077	A	0.093	0.012	1.7E-14	0.0033	63.1
IDL.C	rs144721118	1	54196340	A	-0.297	0.040	1.8E-13	0.0040	78.2
IDL.C	rs144900553	19	10798974	T	-0.346	0.053	1.6E-10	0.0030	58.5
IDL.C	rs146568567	1	54824117	A	-0.328	0.032	1.7E-24	0.0059	114.2
IDL.C	rs148359521	2	21414212	T	-0.203	0.032	4E-10	0.0024	46.0
IDL.C	rs148382396	1	54639713	A	-0.419	0.051	6.7E-16	0.0051	80.8
IDL.C	rs149048538	19	45053024	A	-0.311	0.044	2.2E-12	0.0030	58.0
IDL.C	rs149844719	1	54519237	T	-0.207	0.028	4.6E-13	0.0032	62.4
IDL.C	rs149944945	1	56129361	G	-0.289	0.034	7.9E-17	0.0042	80.5
IDL.C	rs150966173	19	45421204	T	0.221	0.039	1.9E-08	0.0020	37.9
IDL.C	rs150985779	19	45147992	T	-0.271	0.038	1.8E-12	0.0031	60.1
IDL.C	rs151193598	4	73303394	A	0.507	0.085	4.8E-09	0.0039	55.7
IDL.C	rs1532085	15	58683366	G	-0.091	0.011	2.1E-17	0.0040	76.5
IDL.C	rs157594	19	45425175	G	0.136	0.012	3.7E-30	0.0089	173.3
IDL.C	rs16940213	15	58695337	T	0.080	0.013	4.7E-09	0.0018	35.7
IDL.C	rs180961170	1	57012269	G	-0.375	0.052	9.5E-13	0.0039	69.6
IDL.C	rs181169081	2	21312870	A	-0.202	0.032	4E-10	0.0024	45.7
IDL.C	rs181594442	1	57006537	A	-0.375	0.052	9.4E-13	0.0039	69.6
IDL.C	rs182300850	1	54389320	C	-0.419	0.060	5.6E-12	0.0041	61.7
IDL.C	rs182896710	19	10962613	T	-0.182	0.032	2.5E-08	0.0021	40.7
IDL.C	rs183162020	4	73690263	G	0.651	0.115	2.8E-08	0.0049	48.4
IDL.C	rs183305631	19	19597444	A	-0.186	0.032	1E-08	0.0024	42.6
IDL.C	rs183383492	19	11232974	C	-0.362	0.063	1.5E-08	0.0020	35.5
IDL.C	rs184229638	19	45671925	A	-0.663	0.115	1.2E-08	0.0038	46.9
IDL.C	rs184566992	19	44887996	T	-0.353	0.052	2.4E-11	0.0037	58.7
IDL.C	rs184650103	4	74850649	T	0.327	0.046	3.8E-12	0.0029	52.4
IDL.C	rs185415345	1	56625395	A	-0.164	0.027	3.2E-09	0.0024	46.2
IDL.C	rs186538116	1	56840574	C	-0.439	0.045	3E-22	0.0064	115.3
IDL.C	rs188026950	1	55939497	A	-0.463	0.036	9.3E-38	0.0096	186.5
IDL.C	rs1883711	20	39179822	C	0.149	0.025	3E-09	0.0024	45.7
IDL.C	rs189409600	19	45341066	T	-0.650	0.103	5.3E-10	0.0045	62.8
IDL.C	rs189524519	19	11002852	G	-0.263	0.041	2.7E-10	0.0026	47.1
IDL.C	rs189718275	19	45063850	A	-0.322	0.044	5.5E-13	0.0033	63.2
IDL.C	rs190121281	19	19252779	A	-0.197	0.033	2.8E-09	0.0026	46.4
IDL.C	rs190217562	4	75180409	C	0.254	0.040	5.1E-10	0.0024	43.3

IDL.C	rs190425759	19	10644246	A	-0.219	0.035	1.1E-09	0.0022	41.6
IDL.C	rs190934192	1	55334001	A	-0.388	0.040	1.2E-21	0.0070	136.7
IDL.C	rs191210370	1	54236244	G	-0.250	0.038	6.5E-11	0.0029	56.9
IDL.C	rs191404723	1	54636232	T	-0.416	0.051	8.9E-16	0.0046	80.5
IDL.C	rs191448950	1	55584844	A	-0.484	0.032	2.1E-51	0.0123	240.3
IDL.C	rs192012905	19	44463485	G	-0.347	0.053	8.1E-11	0.0031	54.6
IDL.C	rs192570155	1	55246601	C	-0.500	0.045	5.2E-28	0.0088	158.0
IDL.C	rs2007708	19	45410420	A	-0.751	0.104	1.1E-12	0.0044	80.9
IDL.C	rs2287029	19	10916684	T	-0.109	0.015	2.2E-13	0.0029	56.6
IDL.C	rs2479410	1	55505861	A	-0.081	0.012	8.1E-11	0.0026	50.3
IDL.C	rs261334	15	58726744	C	-0.119	0.012	2E-21	0.0050	97.0
IDL.C	rs2722641	19	44892775	A	-0.253	0.044	2E-08	0.0027	48.4
IDL.C	rs2965149	19	45190766	C	-0.070	0.011	4.6E-10	0.0023	44.3
IDL.C	rs2967668	19	45302951	G	-0.172	0.017	1.4E-23	0.0066	128.2
IDL.C	rs2980860	8	126485337	G	-0.065	0.010	5.2E-10	0.0021	40.6
IDL.C	rs312030	2	21462743	C	0.106	0.018	8.7E-09	0.0019	36.0
IDL.C	rs35913552	2	21272896	A	-0.143	0.015	3.9E-21	0.0049	94.6
IDL.C	rs395908	19	45373565	A	-0.161	0.018	2.6E-18	0.0044	85.7
IDL.C	rs429358	19	45411941	C	0.183	0.014	5.9E-39	0.0094	182.2
IDL.C	rs4609471	1	55493584	A	-0.394	0.030	4.5E-39	0.0120	234.6
IDL.C	rs4803748	19	45247048	T	-0.071	0.011	2.4E-10	0.0024	45.7
IDL.C	rs4804573	19	11277232	A	-0.059	0.011	4.5E-08	0.0017	33.2
IDL.C	rs565436	1	55524601	A	0.089	0.012	3.8E-13	0.0029	56.4
IDL.C	rs58446550	19	45328380	A	0.065	0.011	2.9E-09	0.0020	38.0
IDL.C	rs61770425	1	55085125	G	-0.086	0.013	1.3E-11	0.0026	49.3
IDL.C	rs62117161	19	45233385	G	-0.195	0.021	6.2E-21	0.0049	95.3
IDL.C	rs62120794	2	21100426	T	-0.182	0.024	1.9E-13	0.0032	56.9
IDL.C	rs62523994	8	145026582	A	0.060	0.011	2.3E-08	0.0017	33.2
IDL.C	rs629301	1	109818306	T	0.127	0.012	7.5E-24	0.0055	105.7
IDL.C	rs635634	9	136155000	T	0.083	0.013	2.1E-10	0.0022	42.4
IDL.C	rs6511721	19	11206575	A	-0.086	0.011	1.9E-14	0.0037	71.9
IDL.C	rs6663252	1	55630151	C	-0.091	0.012	5E-13	0.0029	55.1
IDL.C	rs6732011	2	21146521	T	-0.074	0.011	3.6E-12	0.0026	50.3
IDL.C	rs6756629	2	44065090	A	-0.129	0.019	1.7E-11	0.0025	47.8
IDL.C	rs6859	19	45382034	G	-0.071	0.010	1.1E-11	0.0025	49.1
IDL.C	rs7255743	19	46018119	A	-0.181	0.033	4.6E-08	0.0018	34.2
IDL.C	rs72740818	15	58654303	C	0.058	0.010	4.4E-08	0.0017	32.0
IDL.C	rs73048351	19	45160086	A	-0.377	0.064	8.5E-09	0.0025	48.1
IDL.C	rs73556990	19	44888175	G	-0.329	0.052	6.4E-10	0.0036	51.6
IDL.C	rs74073060	1	55638930	A	-0.487	0.037	6.7E-38	0.0101	197.0
IDL.C	rs7412	19	45412079	T	-0.533	0.026	5.1E-92	0.0298	490.8
IDL.C	rs75647206	1	56947591	T	-0.379	0.050	7.9E-14	0.0038	72.9
IDL.C	rs7604788	2	21190024	T	-0.189	0.022	3.8E-17	0.0041	80.1
IDL.C	rs76488675	1	56885874	G	-0.147	0.024	1.1E-09	0.0024	46.6
IDL.C	rs76670936	19	45196581	A	-0.142	0.019	2.1E-13	0.0033	63.0
IDL.C	rs7786322	7	21592766	T	0.070	0.012	9E-09	0.0018	35.1
IDL.C	rs78620068	2	21524000	A	-0.131	0.019	4.7E-12	0.0027	52.3

IDL.C	rs79225634	5	74619639	T	0.090	0.011	2.5E-16	0.0037	71.5
IDL.C	rs79668907	19	11257169	T	-0.084	0.013	1.1E-10	0.0026	50.6
IDL.C	rs79890446	19	45723446	T	-0.238	0.033	1.3E-12	0.0035	67.8
IDL.C	rs8106814	19	45441608	C	-0.095	0.014	9.4E-11	0.0031	59.6
IDL.C	rs952275	2	21221399	G	0.109	0.010	3.6E-25	0.0058	112.4
XS.VLDL.TG	rs11076176	16	57007446	G	0.090	0.014	8.7E-10	0.0022	42.0
XS.VLDL.TG	rs11096689	2	21140540	T	-0.097	0.012	1.6E-16	0.0037	71.6
XS.VLDL.TG	rs113531395	17	4886829	T	-0.215	0.036	5.3E-09	0.0024	45.6
XS.VLDL.TG	rs115849089	8	19912370	A	-0.156	0.017	3.4E-19	0.0044	86.0
XS.VLDL.TG	rs116802199	17	4801101	C	-0.197	0.032	1.1E-09	0.0022	40.7
XS.VLDL.TG	rs1168041	1	62960250	C	0.094	0.012	4E-15	0.0035	67.2
XS.VLDL.TG	rs1260326	2	27730940	C	-0.081	0.011	6.6E-14	0.0030	58.8
XS.VLDL.TG	rs1268353	11	116639692	T	-0.081	0.011	2.5E-14	0.0031	60.7
XS.VLDL.TG	rs12747477	1	55448248	A	-0.219	0.033	9.9E-11	0.0027	52.5
XS.VLDL.TG	rs138287365	4	74781004	C	0.313	0.049	3.9E-10	0.0026	50.3
XS.VLDL.TG	rs143413051	4	75560225	T	0.308	0.055	2.9E-08	0.0022	38.8
XS.VLDL.TG	rs143736900	4	72871285	C	0.593	0.082	1.3E-12	0.0044	73.5
XS.VLDL.TG	rs146695330	19	20139610	A	-0.215	0.035	1.5E-09	0.0027	48.0
XS.VLDL.TG	rs150536132	19	19679560	T	-0.181	0.030	1.8E-09	0.0028	49.2
XS.VLDL.TG	rs150617279	19	20139234	A	-0.118	0.017	1.9E-11	0.0027	52.7
XS.VLDL.TG	rs150785555	1	56005603	A	-0.229	0.036	3.2E-10	0.0023	44.8
XS.VLDL.TG	rs151007118	11	116583864	T	0.221	0.033	8.6E-11	0.0025	48.6
XS.VLDL.TG	rs151193598	4	73303394	A	0.531	0.086	9.5E-10	0.0043	61.2
XS.VLDL.TG	rs157594	19	45425175	G	0.111	0.012	1.1E-20	0.0060	116.0
XS.VLDL.TG	rs17216525	19	19662220	T	-0.143	0.021	1.4E-11	0.0025	47.9
XS.VLDL.TG	rs174418	15	58687603	C	-0.088	0.010	1.5E-16	0.0038	73.1
XS.VLDL.TG	rs181169081	2	21312870	A	-0.213	0.032	5.1E-11	0.0026	50.5
XS.VLDL.TG	rs181847072	4	73134560	G	0.595	0.082	6.8E-13	0.0045	75.4
XS.VLDL.TG	rs183162020	4	73690263	G	0.711	0.116	1.5E-09	0.0059	57.8
XS.VLDL.TG	rs183305631	19	19597444	A	-0.212	0.032	6.1E-11	0.0031	55.7
XS.VLDL.TG	rs1838504	15	58666410	T	0.069	0.010	7E-11	0.0024	46.3
XS.VLDL.TG	rs184650103	4	74850649	T	0.340	0.046	5.7E-13	0.0032	56.5
XS.VLDL.TG	rs1848922	2	21471603	C	0.093	0.013	4E-13	0.0029	56.6
XS.VLDL.TG	rs1883711	20	39179822	C	0.149	0.025	3.3E-09	0.0024	45.6
XS.VLDL.TG	rs188651594	11	116673091	A	0.267	0.042	5.5E-10	0.0025	45.2
XS.VLDL.TG	rs189741280	19	19624481	G	-0.188	0.030	5.5E-10	0.0026	50.2
XS.VLDL.TG	rs190121281	19	19252779	A	-0.210	0.033	2.3E-10	0.0030	53.0
XS.VLDL.TG	rs190217562	4	75180409	C	0.251	0.040	7.5E-10	0.0024	42.5
XS.VLDL.TG	rs191164477	2	21267593	T	-0.212	0.032	3.8E-11	0.0026	51.0
XS.VLDL.TG	rs193260502	11	116611138	A	0.216	0.038	2.6E-08	0.0023	44.1
XS.VLDL.TG	rs247617	16	56990716	A	-0.116	0.011	2.2E-23	0.0055	106.9
XS.VLDL.TG	rs261334	15	58726744	C	-0.118	0.012	2.9E-21	0.0050	96.3
XS.VLDL.TG	rs2878419	5	74640490	T	0.061	0.010	9E-09	0.0018	35.0
XS.VLDL.TG	rs2954029	8	126490972	T	-0.086	0.010	2.8E-16	0.0037	70.6
XS.VLDL.TG	rs34041051	19	45442349	C	0.067	0.011	8.9E-09	0.0021	39.7
XS.VLDL.TG	rs34346326	7	73016181	C	-0.076	0.014	3.6E-08	0.0017	31.9
XS.VLDL.TG	rs429358	19	45411941	C	0.108	0.014	1.7E-14	0.0033	63.1
XS.VLDL.TG	rs4609471	1	55493584	A	-0.173	0.030	1E-08	0.0023	45.0

XS.VLDL.TG	rs58542926	19	19379549	T	-0.153	0.021	7.9E-13	0.0028	53.6
XS.VLDL.TG	rs62123892	2	21084445	T	-0.090	0.016	2.2E-08	0.0018	33.9
XS.VLDL.TG	rs6511720	19	11202306	T	-0.145	0.017	1.9E-16	0.0038	73.1
XS.VLDL.TG	rs6511721	19	11206575	A	-0.064	0.011	1.4E-08	0.0020	39.5
XS.VLDL.TG	rs6544366	2	21204025	T	-0.121	0.011	2.9E-25	0.0058	112.3
XS.VLDL.TG	rs7115242	11	116908283	G	-0.125	0.016	5E-15	0.0034	66.7
XS.VLDL.TG	rs72660594	1	55636240	C	-0.201	0.029	7.2E-12	0.0027	52.9
XS.VLDL.TG	rs74073060	1	55638930	A	-0.229	0.037	1.8E-09	0.0022	43.2
XS.VLDL.TG	rs77182215	11	116942366	A	0.165	0.029	1.6E-08	0.0019	37.6
XS.VLDL.TG	rs79202680	17	4692640	T	-0.217	0.035	7.9E-10	0.0023	44.9
XS.VLDL.TG	rs964184	11	116648917	C	-0.216	0.015	9.4E-49	0.0114	222.7
S.VLDL.TG	rs10401845	19	11191536	C	-0.078	0.013	5.2E-09	0.0018	38.2
S.VLDL.TG	rs1042034	2	21225281	T	0.105	0.011	1.3E-20	0.0042	91.0
S.VLDL.TG	rs111648015	8	19724434	T	-0.168	0.030	3.3E-08	0.0018	38.3
S.VLDL.TG	rs112030397	19	8582383	G	-0.082	0.014	8.4E-09	0.0017	35.8
S.VLDL.TG	rs113560866	11	117015189	C	0.082	0.014	1.3E-08	0.0017	37.5
S.VLDL.TG	rs115849089	8	19912370	A	-0.176	0.017	1.6E-25	0.0059	129.0
S.VLDL.TG	rs1168041	1	62960250	C	0.086	0.011	3.8E-14	0.0030	64.1
S.VLDL.TG	rs116843064	19	8429323	A	-0.211	0.035	2.5E-09	0.0025	48.9
S.VLDL.TG	rs117001569	8	19574920	G	-0.232	0.041	2.6E-08	0.0017	33.7
S.VLDL.TG	rs1240659	11	116493950	G	0.071	0.013	4.5E-08	0.0016	34.2
S.VLDL.TG	rs1260326	2	27730940	C	-0.099	0.010	6.2E-22	0.0046	98.8
S.VLDL.TG	rs1268353	11	116639692	T	-0.085	0.010	5.1E-17	0.0034	74.0
S.VLDL.TG	rs12997242	2	21381177	A	0.062	0.010	2.7E-09	0.0017	37.7
S.VLDL.TG	rs145106713	8	19942183	T	-0.257	0.042	2.3E-09	0.0020	38.8
S.VLDL.TG	rs150617279	19	20139234	A	-0.106	0.017	1.3E-09	0.0022	42.8
S.VLDL.TG	rs151007118	11	116583864	T	0.268	0.033	2.5E-15	0.0037	71.6
S.VLDL.TG	rs17216525	19	19662220	T	-0.137	0.020	1E-11	0.0023	49.5
S.VLDL.TG	rs188651594	11	116673091	A	0.312	0.042	3.1E-13	0.0035	61.8
S.VLDL.TG	rs2980853	8	126478350	C	-0.072	0.010	4.1E-13	0.0026	56.3
S.VLDL.TG	rs34346326	7	73016181	C	-0.113	0.014	1.1E-14	0.0031	67.0
S.VLDL.TG	rs36229786	16	56993901	C	0.086	0.014	1.3E-09	0.0020	43.0
S.VLDL.TG	rs3826688	19	45418961	C	0.090	0.011	1.4E-15	0.0035	76.5
S.VLDL.TG	rs429358	19	45411941	C	0.106	0.013	5.8E-15	0.0032	68.6
S.VLDL.TG	rs4341893	2	21135577	G	-0.066	0.010	4.7E-10	0.0019	41.1
S.VLDL.TG	rs5167	19	45448465	G	0.058	0.010	3.8E-08	0.0016	31.8
S.VLDL.TG	rs579674	11	116528224	G	0.081	0.013	2.8E-10	0.0020	43.7
S.VLDL.TG	rs6065904	20	44534651	A	0.076	0.012	1.6E-10	0.0020	43.8
S.VLDL.TG	rs61905067	11	116578982	G	0.235	0.039	2E-09	0.0021	45.5
S.VLDL.TG	rs7115242	11	116908283	G	-0.127	0.016	8.8E-16	0.0033	71.9
S.VLDL.TG	rs72836561	17	41926126	T	0.215	0.036	3.1E-09	0.0023	43.5
S.VLDL.TG	rs72999033	19	19366632	T	-0.155	0.021	3.4E-13	0.0027	57.8
S.VLDL.TG	rs77182215	11	116942366	A	0.205	0.029	1.6E-12	0.0030	58.3
S.VLDL.TG	rs7826306	8	19900671	C	-0.064	0.010	1.4E-09	0.0018	39.1
S.VLDL.TG	rs821840	16	56993886	G	-0.109	0.012	3.1E-20	0.0046	99.1
S.VLDL.TG	rs9472125	6	43756169	T	-0.095	0.016	5.6E-09	0.0025	48.6
S.VLDL.TG	rs964184	11	116648917	C	-0.242	0.014	7.6E-66	0.0140	305.7
S.VLDL.C	rs1042034	2	21225281	T	0.107	0.011	2.1E-21	0.0044	94.6

S.VLDL.C	rs115849089	8	19912370	A	-0.125	0.017	1.6E-13	0.0030	64.4
S.VLDL.C	rs11591147	1	55505647	T	-0.338	0.035	1E-21	0.0068	109.3
S.VLDL.C	rs116054287	1	56401689	C	-0.242	0.038	2.7E-10	0.0022	46.6
S.VLDL.C	rs1168041	1	62960250	C	0.076	0.011	1.9E-11	0.0023	50.4
S.VLDL.C	rs118146573	16	57000938	A	0.120	0.017	2.5E-12	0.0025	53.9
S.VLDL.C	rs11881315	19	10909953	T	-0.074	0.013	1.2E-08	0.0016	35.1
S.VLDL.C	rs1260326	2	27730940	C	-0.074	0.010	7.6E-13	0.0025	54.5
S.VLDL.C	rs1268353	11	116639692	T	-0.066	0.010	5.4E-11	0.0021	45.2
S.VLDL.C	rs1367117	2	21263900	A	0.098	0.011	1E-18	0.0040	85.5
S.VLDL.C	rs138287365	4	74781004	C	0.301	0.049	1.5E-09	0.0024	46.5
S.VLDL.C	rs140339333	4	75396456	A	0.259	0.044	5.8E-09	0.0024	46.3
S.VLDL.C	rs142130958	19	11190652	A	-0.173	0.016	2.4E-25	0.0056	122.0
S.VLDL.C	rs143413051	4	75560225	T	0.345	0.055	4.3E-10	0.0027	48.6
S.VLDL.C	rs143736900	4	72871285	C	0.520	0.082	4.3E-10	0.0034	56.4
S.VLDL.C	rs146568567	1	54824117	A	-0.221	0.032	5.8E-12	0.0027	51.5
S.VLDL.C	rs150103689	1	56105434	G	-0.327	0.044	1.5E-13	0.0036	63.7
S.VLDL.C	rs150617279	19	20139234	A	-0.107	0.017	8.5E-10	0.0023	43.6
S.VLDL.C	rs151007118	11	116583864	T	0.214	0.033	2.4E-10	0.0024	45.8
S.VLDL.C	rs151193598	4	73303394	A	0.528	0.085	9.5E-10	0.0043	60.4
S.VLDL.C	rs157594	19	45425175	G	0.102	0.012	1.1E-17	0.0050	96.9
S.VLDL.C	rs17216525	19	19662220	T	-0.131	0.020	7.3E-11	0.0021	45.3
S.VLDL.C	rs17414716	1	55759138	G	-0.281	0.033	1.4E-17	0.0038	82.6
S.VLDL.C	rs181847072	4	73134560	G	0.522	0.082	2.6E-10	0.0035	57.9
S.VLDL.C	rs183162020	4	73690263	G	0.682	0.115	5.2E-09	0.0054	53.1
S.VLDL.C	rs183305631	19	19597444	A	-0.184	0.032	1.3E-08	0.0023	41.7
S.VLDL.C	rs184650103	4	74850649	T	0.346	0.046	1.6E-13	0.0033	58.6
S.VLDL.C	rs188357577	4	75417188	G	0.335	0.051	9.4E-11	0.0026	50.8
S.VLDL.C	rs188651594	11	116673091	A	0.254	0.042	3.1E-09	0.0023	40.8
S.VLDL.C	rs190934192	1	55334001	A	-0.250	0.040	6.7E-10	0.0029	56.5
S.VLDL.C	rs192570155	1	55246601	C	-0.325	0.045	9.3E-13	0.0037	66.4
S.VLDL.C	rs2980875	8	126481747	G	-0.067	0.010	1.8E-11	0.0022	47.9
S.VLDL.C	rs3005923	1	56801542	A	-0.222	0.036	1.5E-09	0.0026	50.9
S.VLDL.C	rs312030	2	21462743	C	0.110	0.018	5.4E-10	0.0020	43.4
S.VLDL.C	rs3764261	16	56993324	A	-0.103	0.011	4E-21	0.0044	95.4
S.VLDL.C	rs429358	19	45411941	C	0.138	0.013	2.2E-24	0.0054	116.6
S.VLDL.C	rs4609471	1	55493584	A	-0.251	0.030	8.4E-17	0.0049	94.3
S.VLDL.C	rs562338	2	21288321	G	0.083	0.013	9.7E-11	0.0021	44.8
S.VLDL.C	rs61905067	11	116578982	G	0.222	0.039	1.5E-08	0.0019	40.6
S.VLDL.C	rs62123892	2	21084445	T	-0.092	0.015	4.6E-09	0.0018	37.9
S.VLDL.C	rs646776	1	109818530	T	0.070	0.012	5.6E-09	0.0017	36.8
S.VLDL.C	rs6511721	19	11206575	A	-0.068	0.011	5.3E-10	0.0023	50.2
S.VLDL.C	rs6720307	2	20921334	C	-0.061	0.010	2.9E-09	0.0018	38.1
S.VLDL.C	rs7115242	11	116908283	G	-0.103	0.016	6.2E-11	0.0022	47.5
S.VLDL.C	rs72999033	19	19366632	T	-0.147	0.021	4.8E-12	0.0024	52.1
S.VLDL.C	rs74073060	1	55638930	A	-0.308	0.037	4E-16	0.0040	78.3
S.VLDL.C	rs7412	19	45412079	T	-0.235	0.025	6.6E-20	0.0058	107.2
S.VLDL.C	rs77021821	4	75684215	T	0.231	0.040	1.5E-08	0.0019	37.0
S.VLDL.C	rs79225634	5	74619639	T	0.078	0.010	1.7E-13	0.0027	58.9

S.VLDL.C	rs964184	11	116648917	C	-0.188	0.014	2.3E-40	0.0084	183.7
S.VLDL.C	rs984976	5	74910870	G	0.071	0.010	1.4E-12	0.0025	53.7
M.VLDL.TG	rs1168001	1	62933758	A	0.072	0.011	2.6E-11	0.0022	46.5
M.VLDL.TG	rs116843064	19	8429323	A	-0.210	0.035	3.4E-09	0.0025	47.5
M.VLDL.TG	rs1260326	2	27730940	C	-0.095	0.010	6.8E-20	0.0041	88.1
M.VLDL.TG	rs1268353	11	116639692	T	-0.079	0.010	1E-14	0.0029	62.4
M.VLDL.TG	rs149611002	8	19986935	T	-0.248	0.042	6.4E-09	0.0022	42.0
M.VLDL.TG	rs150617279	19	20139234	A	-0.096	0.017	4.1E-08	0.0018	34.6
M.VLDL.TG	rs151007118	11	116583864	T	0.246	0.034	4.7E-13	0.0031	59.2
M.VLDL.TG	rs17120347	11	116996539	A	0.091	0.015	7.4E-10	0.0019	40.5
M.VLDL.TG	rs17216525	19	19662220	T	-0.120	0.020	2.6E-09	0.0018	37.5
M.VLDL.TG	rs188632579	11	116611098	C	0.256	0.046	3.7E-08	0.0017	33.6
M.VLDL.TG	rs188651594	11	116673091	A	0.277	0.043	1.2E-10	0.0027	47.6
M.VLDL.TG	rs34121855	7	73040814	G	-0.117	0.013	2E-18	0.0039	83.4
M.VLDL.TG	rs42121	7	72842267	T	-0.107	0.018	7.7E-09	0.0024	45.3
M.VLDL.TG	rs439401	19	45414451	C	0.080	0.011	1.2E-12	0.0027	57.8
M.VLDL.TG	rs579674	11	116528224	G	0.081	0.013	3.6E-10	0.0020	42.7
M.VLDL.TG	rs59007384	19	45396665	T	0.069	0.012	1.2E-08	0.0017	36.2
M.VLDL.TG	rs6065904	20	44534651	A	0.077	0.012	9.1E-11	0.0021	44.5
M.VLDL.TG	rs61905067	11	116578982	G	0.222	0.039	1.7E-08	0.0019	40.0
M.VLDL.TG	rs6586886	8	19875408	A	-0.058	0.010	2.4E-08	0.0016	33.1
M.VLDL.TG	rs673548	2	21237544	A	-0.080	0.011	2.4E-12	0.0024	51.1
M.VLDL.TG	rs7115242	11	116908283	G	-0.114	0.016	4.1E-13	0.0027	57.8
M.VLDL.TG	rs72999033	19	19366632	T	-0.138	0.021	9E-11	0.0021	45.4
M.VLDL.TG	rs77182215	11	116942366	A	0.205	0.029	2.4E-12	0.0030	56.7
M.VLDL.TG	rs77697917	17	41840849	T	0.209	0.037	2.8E-08	0.0020	37.7
M.VLDL.TG	rs79236614	8	19860460	G	-0.166	0.017	1.6E-21	0.0045	96.2
M.VLDL.TG	rs821840	16	56993886	G	-0.070	0.012	3.3E-09	0.0019	40.4
M.VLDL.TG	rs9472125	6	43756169	T	-0.091	0.016	2.6E-08	0.0023	43.9
M.VLDL.TG	rs964184	11	116648917	C	-0.228	0.014	2.4E-58	0.0124	266.9
S.LDL.C	rs10180633	2	21144829	T	-0.065	0.010	1.6E-10	0.0020	42.8
S.LDL.C	rs10402524	19	45329344	C	0.065	0.010	1.1E-09	0.0019	41.6
S.LDL.C	rs10424477	19	10636051	T	-0.072	0.011	5E-10	0.0023	48.9
S.LDL.C	rs111740198	19	44878217	A	-0.293	0.052	2.8E-08	0.0028	40.1
S.LDL.C	rs112635299	14	94838142	T	0.249	0.040	6.6E-10	0.0021	43.1
S.LDL.C	rs116054287	1	56401689	C	-0.364	0.038	1.9E-21	0.0049	105.8
S.LDL.C	rs117261169	19	45491032	T	-0.320	0.055	1.2E-08	0.0020	38.2
S.LDL.C	rs117569256	19	45423330	G	-0.734	0.107	1.3E-11	0.0054	73.3
S.LDL.C	rs11878174	19	45723379	C	0.070	0.012	3.2E-09	0.0024	46.4
S.LDL.C	rs12043403	1	55431933	C	-0.131	0.018	5.5E-13	0.0034	66.3
S.LDL.C	rs12086676	1	55738663	T	-0.071	0.013	3E-08	0.0015	33.4
S.LDL.C	rs1260326	2	27730940	C	-0.061	0.010	4.3E-09	0.0017	36.7
S.LDL.C	rs137992968	19	11239696	T	-0.195	0.034	1.3E-08	0.0017	36.0
S.LDL.C	rs138270540	4	75353427	C	0.219	0.036	1.8E-09	0.0023	44.6
S.LDL.C	rs138287365	4	74781004	C	0.357	0.049	7.9E-13	0.0034	65.4
S.LDL.C	rs140339333	4	75396456	A	0.260	0.044	5.4E-09	0.0024	46.5
S.LDL.C	rs140411770	19	45356517	A	-0.527	0.088	4E-09	0.0046	72.0
S.LDL.C	rs142130958	19	11190652	A	-0.207	0.016	7.8E-36	0.0081	175.8

S.LDL.C	rs143413051	4	75560225	T	0.364	0.055	4.7E-11	0.0030	54.1
S.LDL.C	rs143736900	4	72871285	C	0.584	0.082	2.5E-12	0.0043	71.1
S.LDL.C	rs144064722	4	73406173	G	0.252	0.034	2.5E-13	0.0031	60.6
S.LDL.C	rs144721118	1	54196340	A	-0.249	0.040	5.4E-10	0.0029	55.2
S.LDL.C	rs146568567	1	54824117	A	-0.288	0.032	2.6E-19	0.0045	87.8
S.LDL.C	rs146982841	19	10771544	T	-0.223	0.036	9.4E-10	0.0023	41.6
S.LDL.C	rs147825223	19	45479553	C	0.165	0.028	1E-08	0.0020	36.0
S.LDL.C	rs148359521	2	21414212	T	-0.180	0.032	2.9E-08	0.0019	36.0
S.LDL.C	rs148382396	1	54639713	A	-0.364	0.051	1.9E-12	0.0038	61.1
S.LDL.C	rs149048538	19	45053024	A	-0.242	0.044	4.5E-08	0.0018	35.0
S.LDL.C	rs149844719	1	54519237	T	-0.172	0.028	1.2E-09	0.0021	44.9
S.LDL.C	rs149944945	1	56129361	G	-0.286	0.034	1.3E-16	0.0041	78.7
S.LDL.C	rs150785555	1	56005603	A	-0.435	0.036	1.9E-33	0.0084	163.3
S.LDL.C	rs150966173	19	45421204	T	0.222	0.039	1.5E-08	0.0020	38.2
S.LDL.C	rs150985779	19	45147992	T	-0.245	0.038	1.9E-10	0.0025	48.8
S.LDL.C	rs157594	19	45425175	G	0.130	0.012	6.3E-28	0.0082	158.9
S.LDL.C	rs17111503	1	55503448	G	0.072	0.013	4.3E-08	0.0017	36.6
S.LDL.C	rs17395160	1	55085141	G	-0.081	0.012	3.5E-11	0.0022	47.3
S.LDL.C	rs180961170	1	57012269	G	-0.349	0.052	2.9E-11	0.0034	60.1
S.LDL.C	rs181169081	2	21312870	A	-0.178	0.032	3.5E-08	0.0018	35.3
S.LDL.C	rs181594442	1	57006537	A	-0.348	0.052	3E-11	0.0034	60.0
S.LDL.C	rs181847072	4	73134560	G	0.581	0.082	2.1E-12	0.0043	71.8
S.LDL.C	rs182300850	1	54389320	C	-0.350	0.060	7.7E-09	0.0029	43.1
S.LDL.C	rs182318839	19	45747128	T	0.298	0.054	4E-08	0.0027	46.8
S.LDL.C	rs183162020	4	73690263	G	0.760	0.115	7E-11	0.0067	66.2
S.LDL.C	rs184566992	19	44887996	T	-0.318	0.052	1.5E-09	0.0030	47.7
S.LDL.C	rs184650103	4	74850649	T	0.396	0.046	3.5E-17	0.0043	76.6
S.LDL.C	rs185049786	4	74644512	C	0.336	0.053	3.9E-10	0.0034	65.8
S.LDL.C	rs185415345	1	56625395	A	-0.159	0.027	8.5E-09	0.0022	43.4
S.LDL.C	rs186538116	1	56840574	C	-0.405	0.045	3.2E-19	0.0055	97.8
S.LDL.C	rs189409600	19	45341066	T	-0.713	0.103	8.8E-12	0.0054	75.6
S.LDL.C	rs189718275	19	45063850	A	-0.256	0.044	9.3E-09	0.0021	39.8
S.LDL.C	rs190217562	4	75180409	C	0.300	0.040	1.7E-13	0.0034	60.4
S.LDL.C	rs190934192	1	55334001	A	-0.359	0.040	6.8E-19	0.0060	117.1
S.LDL.C	rs191404723	1	54636232	T	-0.365	0.051	1.5E-12	0.0036	62.1
S.LDL.C	rs191448950	1	55584844	A	-0.469	0.032	1E-48	0.0105	228.0
S.LDL.C	rs192012905	19	44463485	G	-0.308	0.053	7.5E-09	0.0024	42.9
S.LDL.C	rs192570155	1	55246601	C	-0.458	0.045	7.8E-24	0.0074	132.0
S.LDL.C	rs193084249	1	26987646	G	0.177	0.031	2.3E-08	0.0019	37.6
S.LDL.C	rs2007708	19	45410420	A	-0.798	0.104	3.4E-14	0.0050	91.6
S.LDL.C	rs207176	1	55791846	T	0.114	0.017	3.8E-11	0.0023	48.8
S.LDL.C	rs2479408	1	55504188	G	-0.082	0.014	2E-08	0.0019	41.5
S.LDL.C	rs2927472	19	45349369	C	0.124	0.018	5.6E-12	0.0028	59.9
S.LDL.C	rs2954027	8	126485294	A	-0.069	0.010	5.2E-12	0.0023	50.7
S.LDL.C	rs2965149	19	45190766	C	-0.066	0.011	8.1E-10	0.0020	43.7
S.LDL.C	rs2967668	19	45302951	G	-0.163	0.017	1.8E-21	0.0060	115.3
S.LDL.C	rs34042070	16	72101525	G	0.076	0.012	8.6E-10	0.0019	40.0
S.LDL.C	rs35913552	2	21272896	A	-0.115	0.014	9.2E-16	0.0032	70.0

S.LDL.C	rs429358	19	45411941	C	0.220	0.013	1.2E-59	0.0136	297.6
S.LDL.C	rs4609471	1	55493584	A	-0.355	0.030	3.2E-32	0.0098	190.5
S.LDL.C	rs4614977	2	44087024	G	-0.130	0.018	3.2E-12	0.0024	51.9
S.LDL.C	rs4703667	5	74613906	C	0.090	0.010	2.6E-18	0.0039	84.6
S.LDL.C	rs4803748	19	45247048	T	-0.083	0.011	2.1E-14	0.0032	69.4
S.LDL.C	rs533617	2	21233972	C	-0.172	0.022	4E-15	0.0032	68.5
S.LDL.C	rs562556	1	55524237	A	0.082	0.013	6.7E-10	0.0019	41.7
S.LDL.C	rs61457016	19	41085400	G	-0.317	0.056	3.1E-08	0.0019	39.2
S.LDL.C	rs62117161	19	45233385	G	-0.178	0.020	9.2E-19	0.0041	87.7
S.LDL.C	rs62120794	2	21100426	T	-0.165	0.024	1.5E-11	0.0024	49.1
S.LDL.C	rs629301	1	109818306	T	0.123	0.012	2E-24	0.0052	112.8
S.LDL.C	rs6511721	19	11206575	A	-0.084	0.011	2.1E-14	0.0035	76.1
S.LDL.C	rs6663252	1	55630151	C	-0.078	0.012	2.2E-10	0.0020	44.2
S.LDL.C	rs6859	19	45382034	G	-0.070	0.010	1.6E-12	0.0025	53.3
S.LDL.C	rs73048351	19	45160086	A	-0.390	0.064	2.4E-09	0.0027	51.5
S.LDL.C	rs73556990	19	44888175	G	-0.295	0.052	2.8E-08	0.0029	41.4
S.LDL.C	rs74073060	1	55638930	A	-0.455	0.037	2E-33	0.0088	171.7
S.LDL.C	rs7412	19	45412079	T	-0.492	0.025	5.5E-83	0.0256	480.1
S.LDL.C	rs75647206	1	56947591	T	-0.349	0.050	5.2E-12	0.0032	61.8
S.LDL.C	rs76670936	19	45196581	A	-0.129	0.018	5.7E-12	0.0027	58.9
S.LDL.C	rs77021821	4	75684215	T	0.243	0.040	2.7E-09	0.0021	40.9
S.LDL.C	rs78620068	2	21524000	A	-0.116	0.017	4.2E-11	0.0023	48.6
S.LDL.C	rs79668907	19	11257169	T	-0.076	0.012	1.9E-09	0.0021	46.3
S.LDL.C	rs79890446	19	45723446	T	-0.219	0.033	6.2E-11	0.0030	57.3
S.LDL.C	rs8106814	19	45441608	C	-0.097	0.014	2.6E-11	0.0032	62.8
S.LDL.C	rs8111962	19	10915324	T	-0.087	0.014	5.7E-10	0.0019	41.7
S.LDL.C	rs934197	2	21267461	A	0.103	0.011	2.2E-20	0.0043	93.6
S.LDL.C	rs964184	11	116648917	C	-0.106	0.014	6.7E-14	0.0027	58.4
S.LDL.C	rs984976	5	74910870	G	0.078	0.010	1E-14	0.0030	63.9
S.HDL.TG	rs11076174	16	57003146	C	0.096	0.016	2.7E-09	0.0018	39.8
S.HDL.TG	rs11076176	16	57007446	G	0.133	0.014	1.9E-21	0.0049	106.7
S.HDL.TG	rs117241420	8	19770344	A	-0.247	0.041	2.3E-09	0.0020	37.9
S.HDL.TG	rs1260326	2	27730940	C	-0.069	0.010	3.6E-11	0.0022	46.8
S.HDL.TG	rs1268353	11	116639692	T	-0.083	0.010	2.2E-16	0.0033	71.1
S.HDL.TG	rs138287365	4	74781004	C	0.288	0.049	7.2E-09	0.0022	42.7
S.HDL.TG	rs140339333	4	75396456	A	0.248	0.044	2.4E-08	0.0022	42.6
S.HDL.TG	rs151007118	11	116583864	T	0.202	0.034	2.7E-09	0.0021	40.5
S.HDL.TG	rs151193598	4	73303394	A	0.511	0.086	3.6E-09	0.0040	56.5
S.HDL.TG	rs157594	19	45425175	G	0.079	0.012	2.7E-11	0.0030	58.7
S.HDL.TG	rs1800777	16	57017319	A	0.202	0.035	1.4E-08	0.0020	38.2
S.HDL.TG	rs1815786	11	116921390	C	-0.118	0.015	3.6E-14	0.0031	67.4
S.HDL.TG	rs183365738	4	72954415	A	0.524	0.082	2.1E-10	0.0035	58.5
S.HDL.TG	rs184650103	4	74850649	T	0.313	0.046	2.8E-11	0.0027	47.9
S.HDL.TG	rs1848922	2	21471603	C	0.074	0.012	1.4E-09	0.0019	40.5
S.HDL.TG	rs188651594	11	116673091	A	0.236	0.042	3.7E-08	0.0020	35.3
S.HDL.TG	rs190217562	4	75180409	C	0.240	0.040	3.6E-09	0.0022	38.8
S.HDL.TG	rs2954029	8	126490972	T	-0.074	0.010	8.5E-14	0.0028	59.9
S.HDL.TG	rs34356624	8	19903935	C	0.222	0.039	2.3E-08	0.0018	35.1

S.HDL.TG	rs3764261	16	56993324	A	-0.148	0.011	3.6E-42	0.0092	199.9
S.HDL.TG	rs429358	19	45411941	C	0.100	0.013	1.5E-13	0.0028	61.5
S.HDL.TG	rs4296389	2	21142994	T	-0.077	0.011	5.1E-13	0.0026	55.4
S.HDL.TG	rs6065904	20	44534651	A	0.090	0.012	2.8E-14	0.0029	62.0
S.HDL.TG	rs6511720	19	11202306	T	-0.098	0.017	7.3E-09	0.0018	37.9
S.HDL.TG	rs6586886	8	19875408	A	-0.057	0.010	3.7E-08	0.0015	32.7
S.HDL.TG	rs6957745	7	73056750	C	-0.090	0.013	2.4E-11	0.0023	50.6
S.HDL.TG	rs79236614	8	19860460	G	-0.165	0.017	4.4E-21	0.0044	95.3
S.HDL.TG	rs9472125	6	43756169	T	-0.092	0.016	1.9E-08	0.0023	45.2
S.HDL.TG	rs964184	11	116648917	C	-0.199	0.014	1.2E-44	0.0094	204.8
XL.HDL.C	rs11076174	16	57003146	C	-0.093	0.016	7.1E-09	0.0017	36.8
XL.HDL.C	rs111543310	15	59531818	C	0.322	0.049	4.2E-11	0.0033	63.1
XL.HDL.C	rs112835635	15	59351989	G	0.218	0.035	7.6E-10	0.0022	43.6
XL.HDL.C	rs112884731	15	59504897	C	0.527	0.057	2.3E-20	0.0052	92.3
XL.HDL.C	rs112925355	15	59125988	A	0.210	0.029	3.6E-13	0.0027	57.8
XL.HDL.C	rs113298164	15	58855748	T	0.554	0.047	2E-31	0.0083	148.4
XL.HDL.C	rs116142092	15	59751872	T	0.378	0.050	7.6E-14	0.0033	59.7
XL.HDL.C	rs12708967	16	56993211	C	-0.100	0.013	1.4E-13	0.0028	59.7
XL.HDL.C	rs138690293	15	59310760	C	0.638	0.107	2.9E-09	0.0030	48.8
XL.HDL.C	rs139066754	20	44224606	A	-0.106	0.018	6E-09	0.0018	39.9
XL.HDL.C	rs142855631	15	59286876	T	0.652	0.108	1.7E-09	0.0030	49.3
XL.HDL.C	rs142887188	15	60132580	G	0.246	0.042	3.9E-09	0.0023	39.0
XL.HDL.C	rs146842281	15	59356659	T	0.148	0.022	1.8E-11	0.0025	48.5
XL.HDL.C	rs148527372	3	159734448	A	-0.554	0.095	5.7E-09	0.0026	39.8
XL.HDL.C	rs148902553	15	59776836	C	0.382	0.051	5E-14	0.0035	60.2
XL.HDL.C	rs1532624	16	57005479	A	0.127	0.010	1.1E-37	0.0079	170.7
XL.HDL.C	rs174547	11	61570783	C	-0.085	0.010	3.9E-17	0.0034	74.5
XL.HDL.C	rs17821274	15	58684478	C	-0.075	0.010	8.3E-13	0.0025	54.9
XL.HDL.C	rs17821298	15	58690738	A	0.066	0.012	2E-08	0.0016	33.5
XL.HDL.C	rs181412360	15	59158953	C	0.344	0.038	1.5E-19	0.0049	88.0
XL.HDL.C	rs182776276	15	59254589	G	0.549	0.060	5.7E-20	0.0051	88.4
XL.HDL.C	rs183975744	15	59052479	T	0.671	0.120	2.4E-08	0.0022	34.9
XL.HDL.C	rs185241689	15	59143155	G	0.756	0.114	3.8E-11	0.0034	55.6
XL.HDL.C	rs185481	15	58666679	C	0.058	0.010	1.7E-08	0.0017	35.9
XL.HDL.C	rs189375934	15	60196526	G	0.318	0.053	1.9E-09	0.0023	40.2
XL.HDL.C	rs189418461	15	59725202	G	0.375	0.050	6E-14	0.0034	59.9
XL.HDL.C	rs192630343	15	59286102	A	0.619	0.107	7.6E-09	0.0029	47.4
XL.HDL.C	rs1943973	18	47179516	A	0.087	0.013	1.3E-10	0.0020	42.4
XL.HDL.C	rs2070895	15	58723939	A	0.168	0.012	5.8E-47	0.0104	226.0
XL.HDL.C	rs2575876	9	107665739	A	-0.100	0.013	2.7E-15	0.0030	65.2
XL.HDL.C	rs261291	15	58680178	C	0.154	0.010	3.3E-51	0.0110	240.3
XL.HDL.C	rs34718390	15	58682690	A	0.153	0.024	1.1E-10	0.0027	58.9
XL.HDL.C	rs4810479	20	44545048	T	0.117	0.011	1.2E-25	0.0053	115.5
XL.HDL.C	rs60439253	15	58874532	T	0.230	0.028	4E-16	0.0036	72.5
XL.HDL.C	rs61803025	1	161600591	C	-0.104	0.019	1.9E-08	0.0021	41.5
XL.HDL.C	rs67053123	12	125353810	A	0.081	0.015	2.7E-08	0.0017	35.9
XL.HDL.C	rs686030	9	15304782	A	0.083	0.015	1.5E-08	0.0015	33.1
XL.HDL.C	rs76116860	15	59834938	C	0.265	0.041	1.2E-10	0.0025	47.8
XL.HDL.C	rs7873387	9	107595602	C	0.093	0.016	9.2E-09	0.0016	34.4
XL.HDL.C	rs79844529	15	58445279	T	0.182	0.032	1.3E-08	0.0022	41.6

M.VLDL.C	rs10401845	19	11191536	C	-0.088	0.013	2.4E-11	0.0023	49.7
M.VLDL.C	rs1042034	2	21225281	T	0.109	0.011	4.9E-22	0.0045	97.4
M.VLDL.C	rs113560866	11	117015189	C	0.083	0.014	5.9E-09	0.0018	39.1
M.VLDL.C	rs115849089	8	19912370	A	-0.164	0.017	2.7E-22	0.0051	111.0
M.VLDL.C	rs1168041	1	62960250	C	0.094	0.011	1.4E-16	0.0035	76.1
M.VLDL.C	rs117001569	8	19574920	G	-0.238	0.041	1.1E-08	0.0018	35.3
M.VLDL.C	rs1260326	2	27730940	C	-0.094	0.010	5E-20	0.0041	89.1
M.VLDL.C	rs1268353	11	116639692	T	-0.087	0.010	8.2E-18	0.0036	77.4
M.VLDL.C	rs145106713	8	19942183	T	-0.252	0.042	4.2E-09	0.0019	37.3
M.VLDL.C	rs146695330	19	20139610	A	-0.203	0.035	8.3E-09	0.0024	43.0
M.VLDL.C	rs150536132	19	19679560	T	-0.192	0.030	1.4E-10	0.0031	55.3
M.VLDL.C	rs150617279	19	20139234	A	-0.128	0.017	1.8E-13	0.0032	62.6
M.VLDL.C	rs151007118	11	116583864	T	0.263	0.033	7.1E-15	0.0036	68.9
M.VLDL.C	rs17145738	7	72982874	T	-0.110	0.015	4.3E-13	0.0027	57.3
M.VLDL.C	rs17216525	19	19662220	T	-0.153	0.020	2.6E-14	0.0029	61.8
M.VLDL.C	rs183130	16	56991363	T	-0.105	0.012	2.4E-18	0.0045	83.3
M.VLDL.C	rs183305631	19	19597444	A	-0.212	0.032	4.7E-11	0.0031	55.6
M.VLDL.C	rs188651594	11	116673091	A	0.317	0.042	1.1E-13	0.0036	63.8
M.VLDL.C	rs189741280	19	19624481	G	-0.180	0.030	2.6E-09	0.0024	45.7
M.VLDL.C	rs190121281	19	19252779	A	-0.196	0.033	2.6E-09	0.0026	46.0
M.VLDL.C	rs2954021	8	126482077	G	-0.069	0.010	2.8E-12	0.0024	51.4
M.VLDL.C	rs36229786	16	56993901	C	0.079	0.014	1.9E-08	0.0017	36.7
M.VLDL.C	rs3826688	19	45418961	C	0.092	0.011	2.2E-16	0.0037	80.4
M.VLDL.C	rs3846661	5	74639178	G	0.066	0.010	9E-11	0.0021	45.3
M.VLDL.C	rs429358	19	45411941	C	0.111	0.013	3.4E-16	0.0034	74.5
M.VLDL.C	rs579674	11	116528224	G	0.078	0.013	9.8E-10	0.0019	40.8
M.VLDL.C	rs61905067	11	116578982	G	0.239	0.039	1.1E-09	0.0022	46.9
M.VLDL.C	rs6586891	8	19914598	A	-0.065	0.010	8.1E-10	0.0018	39.9
M.VLDL.C	rs7115242	11	116908283	G	-0.117	0.016	8.9E-14	0.0028	61.5
M.VLDL.C	rs71480307	11	116516873	A	0.084	0.015	2.7E-08	0.0016	34.8
M.VLDL.C	rs72660594	1	55636240	C	-0.175	0.029	1.8E-09	0.0021	40.2
M.VLDL.C	rs72836561	17	41926126	T	0.208	0.036	9.9E-09	0.0021	40.4
M.VLDL.C	rs72999033	19	19366632	T	-0.176	0.021	1E-16	0.0035	74.8
M.VLDL.C	rs7533354	1	63217503	C	-0.082	0.015	3E-08	0.0016	33.5
M.VLDL.C	rs7575840	2	21273490	T	0.074	0.011	1.9E-11	0.0023	50.0
M.VLDL.C	rs77182215	11	116942366	A	0.206	0.029	1.2E-12	0.0030	58.6
M.VLDL.C	rs964184	11	116648917	C	-0.234	0.014	8.7E-62	0.0130	284.8
M.VLDL.C	rs984976	5	74910870	G	0.063	0.010	3.5E-10	0.0019	41.9
L.VLDL.C	rs10889331	1	62943007	T	0.086	0.012	3.8E-13	0.0032	55.9
L.VLDL.C	rs117241420	8	19770344	A	-0.232	0.041	2.2E-08	0.0017	32.9
L.VLDL.C	rs1260326	2	27730940	C	-0.089	0.010	8.5E-18	0.0037	78.1
L.VLDL.C	rs1268353	11	116639692	T	-0.073	0.010	8.6E-13	0.0025	53.3
L.VLDL.C	rs150617279	19	20139234	A	-0.109	0.017	4.7E-10	0.0023	44.6
L.VLDL.C	rs151007118	11	116583864	T	0.256	0.034	5E-14	0.0034	64.2
L.VLDL.C	rs17120347	11	116996539	A	0.091	0.015	6.2E-10	0.0019	40.9
L.VLDL.C	rs17216525	19	19662220	T	-0.134	0.020	3.1E-11	0.0022	46.8
L.VLDL.C	rs181583353	11	39151067	G	0.249	0.044	2.4E-08	0.0024	41.5
L.VLDL.C	rs188651594	11	116673091	A	0.289	0.043	2.1E-11	0.0029	51.5

L.VLDL.C	rs191238346	11	39167052	A	0.247	0.044	2.9E-08	0.0023	40.6
L.VLDL.C	rs2001945	8	126477978	C	-0.058	0.010	7E-09	0.0017	35.2
L.VLDL.C	rs34482346	7	72915521	C	-0.128	0.015	1.4E-16	0.0035	74.5
L.VLDL.C	rs4296389	2	21142994	T	-0.074	0.011	4.1E-12	0.0024	50.4
L.VLDL.C	rs438811	19	45416741	T	0.094	0.012	1.8E-14	0.0032	67.5
L.VLDL.C	rs579674	11	116528224	G	0.088	0.013	1E-11	0.0024	50.4
L.VLDL.C	rs7115242	11	116908283	G	-0.092	0.016	5.1E-09	0.0018	37.5
L.VLDL.C	rs71480307	11	116516873	A	0.083	0.015	4E-08	0.0016	33.8
L.VLDL.C	rs72999033	19	19366632	T	-0.150	0.021	2.3E-12	0.0025	53.2
L.VLDL.C	rs76975037	8	19851508	A	-0.148	0.018	6.4E-17	0.0035	74.2
L.VLDL.C	rs77182215	11	116942366	A	0.200	0.029	7.1E-12	0.0029	54.2
L.VLDL.C	rs821840	16	56993886	G	-0.092	0.012	6.1E-15	0.0033	70.3
L.VLDL.C	rs9472125	6	43756169	T	-0.093	0.016	1.4E-08	0.0024	45.5
L.VLDL.C	rs964184	11	116648917	C	-0.214	0.014	2.7E-51	0.0109	233.8
L.VLDL.TG	rs10455872	6	161010118	G	-0.165	0.028	5.4E-09	0.0021	40.6
L.VLDL.TG	rs10889360	1	63173918	T	-0.064	0.011	3E-08	0.0016	33.7
L.VLDL.TG	rs1260326	2	27730940	C	-0.094	0.010	9.8E-20	0.0041	87.3
L.VLDL.TG	rs1268353	11	116639692	T	-0.068	0.010	3E-11	0.0022	46.0
L.VLDL.TG	rs13030345	2	28003174	T	0.070	0.013	4.2E-08	0.0015	32.6
L.VLDL.TG	rs145106713	8	19942183	T	-0.243	0.044	3.4E-08	0.0018	32.6
L.VLDL.TG	rs151007118	11	116583864	T	0.226	0.034	3.4E-11	0.0026	49.6
L.VLDL.TG	rs16996148	19	19658472	T	-0.123	0.020	6.9E-10	0.0019	40.4
L.VLDL.TG	rs17120347	11	116996539	A	0.086	0.015	4.8E-09	0.0017	36.6
L.VLDL.TG	rs17411024	8	19852134	A	-0.154	0.018	9.6E-18	0.0038	78.0
L.VLDL.TG	rs188651594	11	116673091	A	0.241	0.043	2.3E-08	0.0020	35.8
L.VLDL.TG	rs34346326	7	73016181	C	-0.110	0.013	9.4E-17	0.0035	74.1
L.VLDL.TG	rs4350231	1	62922660	A	-0.071	0.011	5.5E-11	0.0021	45.2
L.VLDL.TG	rs438811	19	45416741	T	0.090	0.012	2.9E-13	0.0029	61.2
L.VLDL.TG	rs579674	11	116528224	G	0.078	0.013	1.4E-09	0.0019	39.9
L.VLDL.TG	rs7115242	11	116908283	G	-0.099	0.016	3.9E-10	0.0020	43.0
L.VLDL.TG	rs72999033	19	19366632	T	-0.133	0.021	4.6E-10	0.0020	41.9
L.VLDL.TG	rs77182215	11	116942366	A	0.202	0.029	4.3E-12	0.0029	55.3
L.VLDL.TG	rs9472125	6	43756169	T	-0.091	0.016	3.1E-08	0.0023	43.4
L.VLDL.TG	rs964184	11	116648917	C	-0.206	0.014	7E-48	0.0101	217.5
XL.VLDL.TG	rs10455872	6	161010118	G	-0.181	0.028	1.2E-10	0.0026	49.3
XL.VLDL.TG	rs1168041	1	62960250	C	0.084	0.011	1E-13	0.0028	61.2
XL.VLDL.TG	rs1260326	2	27730940	C	-0.093	0.010	1.4E-19	0.0040	86.3
XL.VLDL.TG	rs1268353	11	116639692	T	-0.057	0.010	1.2E-08	0.0016	33.7
XL.VLDL.TG	rs13234157	7	72971728	A	-0.111	0.015	3.2E-13	0.0027	58.5
XL.VLDL.TG	rs151007118	11	116583864	T	0.209	0.033	5E-10	0.0023	43.7
XL.VLDL.TG	rs17120347	11	116996539	A	0.082	0.014	1.8E-08	0.0016	33.8
XL.VLDL.TG	rs17216525	19	19662220	T	-0.129	0.021	9.2E-10	0.0019	40.1
XL.VLDL.TG	rs17411024	8	19852134	A	-0.125	0.017	8.2E-13	0.0025	54.1
XL.VLDL.TG	rs181583353	11	39151067	G	0.242	0.044	4.6E-08	0.0022	39.8
XL.VLDL.TG	rs186696265	6	161111700	T	-0.326	0.057	1.9E-08	0.0020	35.1
XL.VLDL.TG	rs4296389	2	21142994	T	-0.059	0.011	2.4E-08	0.0015	32.5
XL.VLDL.TG	rs438811	19	45416741	T	0.081	0.013	5E-10	0.0023	44.6
XL.VLDL.TG	rs579674	11	116528224	G	0.081	0.013	2.6E-10	0.0020	43.4

XL.VLDL.TG	rs72999033	19	19366632	T	-0.144	0.022	1.3E-10	0.0021	45.3
XL.VLDL.TG	rs77182215	11	116942366	A	0.182	0.029	3.1E-10	0.0024	45.7
XL.VLDL.TG	rs964184	11	116648917	C	-0.179	0.014	7E-37	0.0076	165.4
XXL.VLDL.TG	rs10455872	6	161010118	G	-0.194	0.028	4.8E-12	0.0029	56.6
XXL.VLDL.TG	rs1168041	1	62960250	C	0.077	0.011	7.7E-12	0.0024	51.6
XXL.VLDL.TG	rs1260326	2	27730940	C	-0.093	0.010	8E-20	0.0040	87.2
XXL.VLDL.TG	rs1268353	11	116639692	T	-0.056	0.010	3.1E-08	0.0015	31.7
XXL.VLDL.TG	rs13233571	7	72971231	T	-0.094	0.015	6.4E-10	0.0019	41.4
XXL.VLDL.TG	rs151007118	11	116583864	T	0.201	0.033	2.1E-09	0.0021	40.5
XXL.VLDL.TG	rs17217098	19	19702384	A	-0.117	0.021	1.9E-08	0.0015	33.3
XXL.VLDL.TG	rs483082	19	45416178	T	0.097	0.012	1.2E-15	0.0034	72.9
XXL.VLDL.TG	rs72999033	19	19366632	T	-0.126	0.021	2.3E-09	0.0018	38.3
XXL.VLDL.TG	rs77182215	11	116942366	A	0.167	0.029	6.8E-09	0.0020	38.6
XXL.VLDL.TG	rs77729186	8	19826318	G	-0.116	0.017	9.5E-12	0.0022	48.5
XXL.VLDL.TG	rs821840	16	56993886	G	-0.065	0.012	2.2E-08	0.0017	35.9
XXL.VLDL.TG	rs964184	11	116648917	C	-0.153	0.014	1.8E-27	0.0056	120.6
XL.HDL.TG	rs11096689	2	21140540	T	-0.073	0.011	9.9E-11	0.0021	44.4
XL.HDL.TG	rs111543310	15	59531818	C	0.492	0.049	1.5E-23	0.0076	147.3
XL.HDL.TG	rs112835635	15	59351989	G	0.331	0.035	2.3E-20	0.0050	100.3
XL.HDL.TG	rs112884731	15	59504897	C	0.712	0.057	1.8E-35	0.0094	169.1
XL.HDL.TG	rs112925355	15	59125988	A	0.290	0.029	1.9E-23	0.0051	110.7
XL.HDL.TG	rs113298164	15	58855748	T	0.750	0.047	1.1E-55	0.0152	273.8
XL.HDL.TG	rs113531395	17	4886829	T	-0.204	0.036	2.4E-08	0.0021	41.0
XL.HDL.TG	rs114716552	15	58600902	G	-0.152	0.021	9E-13	0.0031	60.6
XL.HDL.TG	rs116142092	15	59751872	T	0.473	0.050	1.5E-20	0.0052	93.6
XL.HDL.TG	rs11632970	15	58837515	C	0.078	0.012	5.5E-10	0.0022	48.3
XL.HDL.TG	rs11638718	15	58079462	G	-0.071	0.012	7.4E-09	0.0016	35.0
XL.HDL.TG	rs116802199	17	4801101	C	-0.217	0.032	1.1E-11	0.0027	49.8
XL.HDL.TG	rs116869421	15	58709436	C	-0.360	0.051	4.1E-12	0.0034	66.0
XL.HDL.TG	rs117386336	15	58568077	T	0.218	0.039	3.3E-08	0.0018	35.6
XL.HDL.TG	rs117459981	15	58619066	C	-0.279	0.047	4.3E-09	0.0020	43.1
XL.HDL.TG	rs117597286	15	58587369	C	-0.260	0.036	1.3E-12	0.0030	57.2
XL.HDL.TG	rs117749052	15	58749309	C	0.276	0.037	1.6E-13	0.0044	85.0
XL.HDL.TG	rs117806344	15	58693213	T	-0.377	0.051	2.6E-13	0.0037	70.8
XL.HDL.TG	rs118078695	15	58686409	A	0.295	0.051	1.3E-08	0.0025	48.2
XL.HDL.TG	rs12442723	15	59458663	C	0.102	0.013	1.2E-15	0.0032	69.7
XL.HDL.TG	rs1268353	11	116639692	T	-0.061	0.010	2.3E-09	0.0017	37.5
XL.HDL.TG	rs12899090	15	59901576	G	0.096	0.015	5.8E-10	0.0028	54.8
XL.HDL.TG	rs1318175	15	58586129	T	-0.157	0.013	1.3E-32	0.0072	155.1
XL.HDL.TG	rs13329672	15	58699937	T	0.156	0.011	3.3E-41	0.0095	206.9
XL.HDL.TG	rs138195472	15	58672107	T	0.357	0.035	2.6E-24	0.0064	114.3
XL.HDL.TG	rs138690293	15	59310760	C	0.719	0.107	3.1E-11	0.0038	62.0
XL.HDL.TG	rs142538594	15	58192308	G	0.383	0.055	7E-12	0.0057	91.8
XL.HDL.TG	rs142855631	15	59286876	T	0.728	0.108	2.5E-11	0.0038	61.4
XL.HDL.TG	rs142887188	15	60132580	G	0.257	0.042	1.1E-09	0.0025	42.5
XL.HDL.TG	rs144149061	15	58500098	T	0.838	0.131	2.3E-10	0.0049	66.7
XL.HDL.TG	rs145347194	15	58670135	C	0.221	0.022	4.4E-23	0.0063	135.9
XL.HDL.TG	rs146842281	15	59356659	T	0.256	0.022	4.4E-31	0.0075	146.3
XL.HDL.TG	rs148828254	15	58571224	A	0.209	0.031	5.1E-11	0.0023	50.7

XL.HDL.TG	rs148902553	15	59776836	C	0.477	0.050	8.5E-21	0.0055	94.2
XL.HDL.TG	rs150536132	19	19679560	T	-0.166	0.030	3.2E-08	0.0023	41.1
XL.HDL.TG	rs1532085	15	58683366	G	-0.264	0.010	9E-155	0.0336	748.0
XL.HDL.TG	rs1540037	18	47182664	G	0.092	0.012	7.9E-14	0.0029	63.3
XL.HDL.TG	rs16939881	15	58471979	C	0.315	0.028	1E-29	0.0084	164.1
XL.HDL.TG	rs1711062	15	58508790	C	-0.105	0.011	4.9E-22	0.0052	112.0
XL.HDL.TG	rs17231506	16	56994528	T	0.069	0.011	3.1E-10	0.0020	43.2
XL.HDL.TG	rs181412360	15	59158953	C	0.483	0.038	1.1E-36	0.0097	174.4
XL.HDL.TG	rs181450801	15	59326120	A	0.463	0.076	1.9E-09	0.0031	58.6
XL.HDL.TG	rs181835401	1	63135955	A	-0.090	0.011	4.4E-15	0.0031	67.2
XL.HDL.TG	rs182776276	15	59254589	G	0.732	0.060	5.7E-34	0.0091	157.9
XL.HDL.TG	rs182785673	15	58073964	T	-0.466	0.076	1.3E-09	0.0028	50.4
XL.HDL.TG	rs183276229	15	58742906	C	0.310	0.035	6.3E-18	0.0046	81.3
XL.HDL.TG	rs183975744	15	59052479	T	0.803	0.120	3.4E-11	0.0032	50.0
XL.HDL.TG	rs185241689	15	59143155	G	0.836	0.114	3.8E-13	0.0042	68.1
XL.HDL.TG	rs185533289	15	58782289	C	-0.307	0.055	3.5E-08	0.0019	32.2
XL.HDL.TG	rs186603838	15	58865534	A	0.166	0.030	4E-08	0.0018	31.5
XL.HDL.TG	rs188131745	15	58553702	A	-0.464	0.079	6.2E-09	0.0027	51.3
XL.HDL.TG	rs189375934	15	60196526	G	0.429	0.053	8.4E-16	0.0042	73.1
XL.HDL.TG	rs189418461	15	59725202	G	0.470	0.050	9.9E-21	0.0053	94.0
XL.HDL.TG	rs190121281	19	19252779	A	-0.196	0.033	2.7E-09	0.0026	46.0
XL.HDL.TG	rs190548956	15	59985051	A	0.266	0.042	3.8E-10	0.0023	43.9
XL.HDL.TG	rs191448950	1	55584844	A	-0.217	0.032	1.3E-11	0.0022	48.4
XL.HDL.TG	rs192060595	15	58907990	C	0.329	0.057	9.8E-09	0.0025	43.1
XL.HDL.TG	rs192630343	15	59286102	A	0.709	0.107	5.1E-11	0.0038	62.3
XL.HDL.TG	rs192924868	15	59231939	C	0.115	0.021	3.1E-08	0.0020	37.9
XL.HDL.TG	rs193092110	15	58730460	A	0.303	0.035	2.8E-17	0.0044	77.7
XL.HDL.TG	rs1998013	1	55958030	T	-0.230	0.035	1.1E-10	0.0022	48.1
XL.HDL.TG	rs2044332	15	58646641	A	0.122	0.014	8.2E-17	0.0038	81.5
XL.HDL.TG	rs2070895	15	58723939	A	0.302	0.012	3E-148	0.0337	749.9
XL.HDL.TG	rs2217970	15	60090978	A	-0.130	0.019	3.8E-11	0.0025	49.2
XL.HDL.TG	rs2414585	15	58785756	G	-0.308	0.048	1.6E-10	0.0028	54.5
XL.HDL.TG	rs2642636	15	58363242	G	-0.072	0.010	3.7E-12	0.0024	52.5
XL.HDL.TG	rs28370984	15	58629308	C	0.344	0.032	9.9E-27	0.0066	127.5
XL.HDL.TG	rs28601761	8	126500031	G	-0.068	0.010	1.8E-11	0.0023	49.2
XL.HDL.TG	rs2881925	2	20390694	A	-0.056	0.010	1.4E-08	0.0016	34.0
XL.HDL.TG	rs2932196	15	57912338	T	0.056	0.010	2.5E-08	0.0015	32.4
XL.HDL.TG	rs34101191	15	58793567	A	0.075	0.010	1.4E-12	0.0028	59.8
XL.HDL.TG	rs35138338	15	58744481	T	-0.089	0.013	3E-11	0.0033	70.4
XL.HDL.TG	rs35684611	15	58721302	G	-0.138	0.014	2.4E-21	0.0047	100.8
XL.HDL.TG	rs426684	15	58662280	T	0.066	0.010	4.1E-11	0.0022	47.3
XL.HDL.TG	rs439401	19	45414451	C	0.086	0.011	1.3E-14	0.0032	68.3
XL.HDL.TG	rs4775039	15	58670897	G	0.160	0.011	1.4E-45	0.0129	280.9
XL.HDL.TG	rs479084	15	58666087	G	-0.120	0.010	1.1E-29	0.0067	145.7
XL.HDL.TG	rs490098	15	58691225	A	-0.179	0.013	1.9E-42	0.0093	202.4
XL.HDL.TG	rs4939873	18	47062054	T	0.133	0.023	1.4E-08	0.0017	36.4
XL.HDL.TG	rs55817218	15	58562006	A	-0.170	0.020	6.2E-17	0.0041	88.9
XL.HDL.TG	rs55861554	15	58761235	C	0.067	0.011	6.3E-10	0.0020	43.5
XL.HDL.TG	rs56296027	2	21134011	C	-0.062	0.011	3.2E-08	0.0015	32.7

XL.HDL.TG	rs572107	15	59055810	C	-0.069	0.010	5.2E-11	0.0022	46.9
XL.HDL.TG	rs61999891	15	58299599	A	0.122	0.017	3.5E-12	0.0030	58.9
XL.HDL.TG	rs62001693	15	58614892	A	-0.221	0.034	1.3E-10	0.0028	59.5
XL.HDL.TG	rs6589592	11	116957907	G	-0.105	0.016	2E-11	0.0023	50.6
XL.HDL.TG	rs7178935	15	59368167	A	0.064	0.011	1E-08	0.0016	34.6
XL.HDL.TG	rs72739708	15	57733779	T	0.114	0.021	5E-08	0.0015	32.3
XL.HDL.TG	rs73424577	15	58869185	G	0.305	0.028	5.5E-27	0.0064	128.9
XL.HDL.TG	rs73959582	18	47148886	C	0.097	0.014	2.4E-11	0.0024	52.5
XL.HDL.TG	rs74073060	1	55638930	A	-0.218	0.037	8.1E-09	0.0020	39.1
XL.HDL.TG	rs74537322	15	58342102	G	-0.406	0.065	7.2E-10	0.0028	49.6
XL.HDL.TG	rs75870978	15	58177266	G	0.391	0.066	4.6E-09	0.0055	60.8
XL.HDL.TG	rs76116860	15	59834938	C	0.339	0.041	3.1E-16	0.0040	78.2
XL.HDL.TG	rs76212899	15	58263295	G	-0.157	0.027	5.2E-09	0.0022	42.6
XL.HDL.TG	rs76438892	15	58687932	G	-0.271	0.047	1.1E-08	0.0018	35.7
XL.HDL.TG	rs78321025	15	58108078	A	0.248	0.034	9.6E-13	0.0035	67.9
XL.HDL.TG	rs79202680	17	4692640	T	-0.242	0.035	5.2E-12	0.0029	55.8
XL.HDL.TG	rs8025975	15	59696602	G	0.097	0.015	4.2E-10	0.0021	44.8
XL.HDL.TG	rs8042174	15	58685970	C	-0.206	0.020	6.8E-25	0.0059	128.8
XL.HDL.TG	rs8043310	15	58731818	A	0.360	0.039	1.2E-19	0.0050	96.6
XL.HDL.TG	rs8100204	19	19393714	A	-0.096	0.016	4.3E-09	0.0021	44.9
XL.HDL.TG	rs935202	15	58457569	A	0.083	0.011	1.1E-14	0.0031	67.2
XL.HDL.TG	rs938507	15	58582034	A	0.139	0.014	6.8E-22	0.0045	97.2
XL.HDL.TG	rs964184	11	116648917	C	-0.155	0.014	1.1E-27	0.0057	123.4
XL.HDL.TG	rs97384	11	61624181	C	0.084	0.010	1.2E-15	0.0034	74.0

Supplementary Table 5. Genetic instruments for lipid drug targets, as selected and extracted from Global Lipids Genetics Consortium (GLGC) GWAS dataset.

Phenotype	Target	Proxy treatment	SNP	Chr	Position (hg18)	Eff_allele	Effect	SE	P-value	R2	F
HDL-C	CETP	CETP inhibitors	rs12446867	16	57052901	G	0.034	0.004	1.8E-16	0.0005	84.0
HDL-C	CETP	CETP inhibitors	rs12448528	16	56985555	G	0.199	0.005	2.1E-344	0.0139	2577.3
HDL-C	CETP	CETP inhibitors	rs12597002	16	57002404	C	0.085	0.004	1.1E-102	0.0029	535.4
HDL-C	CETP	CETP inhibitors	rs12720917	16	57019392	C	0.098	0.006	5.9E-68	0.0023	407.0
HDL-C	CETP	CETP inhibitors	rs12928552	16	57048707	G	0.050	0.008	2.2E-09	0.0002	35.6
HDL-C	CETP	CETP inhibitors	rs13306673	16	56900931	C	0.098	0.006	2.8E-48	0.0016	288.0
HDL-C	CETP	CETP inhibitors	rs13306677	16	56926195	A	0.090	0.006	1.4E-50	0.0014	268.4
HDL-C	CETP	CETP inhibitors	rs1566439	16	57024662	C	0.027	0.004	3.5E-15	0.0004	67.9
HDL-C	CETP	CETP inhibitors	rs16963520	16	56936563	A	0.071	0.007	3.1E-25	0.0014	130.7
HDL-C	CETP	CETP inhibitors	rs17290922	16	57024317	G	0.055	0.008	3.4E-13	0.0006	58.9
HDL-C	CETP	CETP inhibitors	rs17370142	16	57050348	T	0.034	0.006	1.2E-11	0.0002	43.8
HDL-C	CETP	CETP inhibitors	rs1864163	16	56997233	G	0.225	0.004	3.6E-573	0.0198	3745.5
HDL-C	CETP	CETP inhibitors	rs1875236	16	57033696	A	0.059	0.007	1.4E-18	0.0006	113.9
HDL-C	CETP	CETP inhibitors	rs247615	16	56984763	A	0.076	0.004	2.9E-62	0.0020	371.2
HDL-C	CETP	CETP inhibitors	rs247616	16	56989590	T	0.243	0.004	1.2E-802	0.0245	4650.2
HDL-C	CETP	CETP inhibitors	rs289719	16	57009941	T	0.113	0.004	5.4E-173	0.0056	1048.7
HDL-C	CETP	CETP inhibitors	rs289726	16	57074451	T	0.036	0.004	1.3E-22	0.0006	112.2
HDL-C	CETP	CETP inhibitors	rs289745	16	57019532	A	0.028	0.004	2.3E-20	0.0004	66.6
HDL-C	CETP	CETP inhibitors	rs4329913	16	56905432	C	0.041	0.006	1.4E-11	0.0006	54.1
HDL-C	CETP	CETP inhibitors	rs5883	16	57007353	T	0.115	0.008	1.8E-31	0.0015	254.0
HDL-C	CETP	CETP inhibitors	rs7188963	16	56931565	C	0.059	0.004	3.2E-39	0.0012	220.1
HDL-C	CETP	CETP inhibitors	rs7204290	16	56968039	G	0.030	0.004	1.5E-14	0.0004	71.4
HDL-C	CETP	CETP inhibitors	rs7499911	16	57036440	G	0.060	0.011	1.3E-08	0.0004	40.4
HDL-C	CETP	CETP inhibitors	rs9938160	16	56984590	C	0.060	0.005	3.2E-25	0.0014	125.6
LDL-C	HMGCR	Statins	rs10066707	5	74560579	A	0.050	0.005	3.0E-19	0.0012	108.1
LDL-C	HMGCR	Statins	rs10515198	5	74641560	A	0.060	0.006	6.0E-22	0.0007	114.7
LDL-C	HMGCR	Statins	rs3857388	5	74620377	C	0.042	0.006	2.2E-11	0.0004	68.5
LDL-C	HMGCR	Statins	rs7703051	5	74625487	A	0.073	0.004	1.4E-77	0.0026	443.6
LDL-C	NPC1L1	Ezetimibe	rs2073547	7	44582331	G	0.049	0.005	1.9E-21	0.0007	125.0
LDL-C	NPC1L1	Ezetimibe	rs217386	7	44600695	G	0.036	0.004	1.2E-19	0.0006	110.2
LDL-C	NPC1L1	Ezetimibe	rs7798185	7	44570717	A	0.041	0.007	1.9E-09	0.0004	37.3
LDL-C	PCSK9	PCSK9 inhibitors	rs10493176	1	55538552	T	0.078	0.010	2.5E-14	0.0012	105.4
LDL-C	PCSK9	PCSK9 inhibitors	rs11206510	1	55496039	T	0.083	0.005	2.4E-53	0.0018	312.2
LDL-C	PCSK9	PCSK9 inhibitors	rs11583974	1	55551718	A	0.065	0.012	4.0E-09	0.0002	24.5
LDL-C	PCSK9	PCSK9 inhibitors	rs12067569	1	55528629	A	0.089	0.010	2.0E-17	0.0005	85.3
LDL-C	PCSK9	PCSK9 inhibitors	rs2479394	1	55486064	G	0.039	0.004	1.6E-19	0.0006	105.1
LDL-C	PCSK9	PCSK9 inhibitors	rs2479409	1	55504650	G	0.064	0.004	2.5E-50	0.0018	317.0
LDL-C	PCSK9	PCSK9 inhibitors	rs2483205	1	55518316	C	0.051	0.005	4.7E-20	0.0013	102.6
LDL-C	PCSK9	PCSK9 inhibitors	rs4927193	1	55509872	T	0.035	0.006	4.3E-11	0.0003	48.7
LDL-C	PCSK9	PCSK9 inhibitors	rs502576	1	55512882	G	0.065	0.007	2.9E-22	0.0015	122.3
LDL-C	PCSK9	PCSK9 inhibitors	rs585131	1	55524116	T	0.064	0.005	2.7E-35	0.0012	205.3
LDL-C	PCSK9	PCSK9 inhibitors	rs7552841	1	55518752	T	0.037	0.004	5.4E-15	0.0006	88.1
LDL-C	ABCG5G8	Bile acid resins	rs10208987	2	44043135	T	0.049	0.007	2.4E-12	0.0004	66.8
LDL-C	ABCG5G8	Bile acid resins	rs1025447	2	44022970	C	0.042	0.005	3.8E-16	0.0005	80.2
LDL-C	ABCG5G8	Bile acid resins	rs4148214	2	44079004	T	0.039	0.004	3.8E-25	0.0007	129.8
LDL-C	ABCG5G8	Bile acid resins	rs4953023	2	44074000	G	0.131	0.007	1.7E-66	0.0027	454.5
LDL-C	ABCG5G8	Bile acid resins	rs6544713	2	44073881	T	0.081	0.004	4.8E-83	0.0027	467.8
LDL-C	ABCG5G8	Bile acid resins	rs75279593	2	44085035	A	0.074	0.012	1.4E-08	0.0005	42.7

LDL-C	LDLR	LDL receptor	rs1010679	19	11207102	T	0.102	0.006	3.5E-54	0.0030	253.7
LDL-C	LDLR	LDL receptor	rs3786721	19	11146499	T	0.047	0.004	2.9E-31	0.0011	178.3
LDL-C	LDLR	LDL receptor	rs3786722	19	11161537	C	0.075	0.004	5.5E-63	0.0021	358.8
LDL-C	LDLR	LDL receptor	rs379309	19	11284302	C	0.031	0.004	1.4E-13	0.0005	81.8
LDL-C	LDLR	LDL receptor	rs5742911	19	11243445	A	0.061	0.006	4.8E-24	0.0014	111.9
LDL-C	LDLR	LDL receptor	rs5927	19	11233941	G	0.035	0.005	2.8E-13	0.0005	75.9
LDL-C	LDLR	LDL receptor	rs6511720	19	11202306	G	0.221	0.006	3.8E-262	0.0086	1479.6
LDL-C	LDLR	LDL receptor	rs688	19	11227602	T	0.054	0.004	1.0E-43	0.0014	240.8

Supplementary Table 6. Power calculations for the Mendelian randomization analyses performed in the current study.

Lipid trait	Variance explained (R^2) [a]	Maximum / Minimum association estimate [b] for 1-β >0.80		
		Small vessel stroke	WMH volume	Intracerebral hemorrhage
Sample		298,777	10,597	3,026
Number of cases		11,710		1,545
Blood lipid levels		OR	beta (max / min)	OR (max / min)
HDL-C	12.4%	≤0.93 & ≥1.08	≤-0.06 / ≥0.06	≤0.75 / ≥1.33
LDL-C	11.6%	≤0.93 / ≥1.08	≤-0.07/ ≥0.07	≤0.74 / ≥1.35
TG	9.31%	≤0.92 / ≥1.09	≤-0.09 / ≥0.09	≤0.71 / ≥1.40
Lipid drug targets [c]				
CETP	9.43%	≤0.92 / ≥1.09	≤-0.09 / ≥0.09	≤0.72 / ≥1.39
HMGCR	0.48%	≤0.72 / ≥1.39	≤-0.39 / ≥0.39	≤0.25 / ≥3.97
NPC1L1	0.18%	≤0.58 / ≥1.68	≤-0.64 / ≥0.64	≤0.12 / ≥8.03
PCSK9	1.11%	≤0.79 / ≥1.26	≤-0.26 / ≥0.26	≤0.39 / ≥2.57
ABCG5/G8	0.74%	≤0.76 / ≥1.31	≤-0.32 / ≥0.32	≤0.32 / ≥3.13
LDLR	1.87%	≤0.83 / ≥1.20	≤-0.20 / ≥0.20	≤0.48 / ≥2.08
PPARA	-	-	-	-
ANGPTL3	0.21%	≤0.63 / ≥1.59	≤-0.60 / ≥0.60	≤0.14 / ≥7.09
ANGPTL4	-	-	-	-
APOC3	2.25%	≤0.85 / ≥1.18	≤-0.19 / ≥0.19	≤0.51 / ≥1.95
LPL	1.65%	≤0.83 / ≥1.21	≤-0.22 / ≥0.22	≤0.46 / ≥2.18
Lipoprotein particle components				
S.HDL.TG	8.89%	≤0.92 / ≥1.09	≤-0.09 / ≥0.09	≤0.71 / ≥1.41
M.HDL.C	4.37%	≤0.88 / ≥1.13	≤-0.13 / ≥0.13	≤0.62 / ≥1.62
L.HDL.C	21.5%	≤0.94 / ≥1.06	≤-0.05 / ≥0.05	≤0.81 / ≥1.25
XL.HDL.C	13.9%	≤0.93 / ≥1.07	≤-0.06 / ≥0.06	≤0.76 / ≥1.32
XL.HDL.TG	47.8%	≤0.96 / ≥1.04	≤-0.04 / ≥0.04	≤0.86 / ≥1.16
S.LDL.C	35.9%	≤0.95 / ≥1.05	≤-0.05 / ≥0.05	≤0.84 / ≥1.19
M.LDL.C	42.2%	≤0.95 / ≥1.05	≤-0.04 / ≥0.04	≤0.85 / ≥1.17
L.LDL.C	45.7%	≤0.96 / ≥1.04	≤-0.04 / ≥0.04	≤0.86 / ≥1.16
IDL.C	42.1%	≤0.95 / ≥1.05	≤-0.04 / ≥0.04	≤0.85 / ≥1.17
IDL.TG	30.8%	≤0.95 / ≥1.05	≤-0.05 / ≥0.05	≤0.83 / ≥1.21
XS.VLDL.TG	17.4%	≤0.93 / ≥1.07	≤-0.06 / ≥0.06	≤0.78 / ≥1.28
S.VLDL.C	16.3%	≤0.93 / ≥1.07	≤-0.06 / ≥0.06	≤0.78 / ≥1.29
S.VLDL.TG	10.8%	≤0.93 / ≥1.08	≤-0.08 / ≥0.08	≤0.73 / ≥1.37
M.VLDL.C	11.6%	≤0.93 / ≥1.08	≤-0.07 / ≥0.07	≤0.74 / ≥1.35
M.VLDL.TG	7.86%	≤0.91 / ≥1.10	≤-0.10 / ≥0.10	≤0.69 / ≥1.44
L.VLDL.C	7.04%	≤0.91 / ≥1.10	≤-0.11 / ≥0.11	≤0.68 / ≥1.47
L.VLDL.TG	5.49%	≤0.89 / ≥1.12	≤-0.12 / ≥0.12	≤0.65 / ≥1.54
XL.VLDL.TG	4.40%	≤0.88 / ≥1.13	≤-0.13 / ≥0.13	≤0.62 / ≥1.62
XXL.VLDL.TG	3.30%	≤0.87 / ≥1.15	≤-0.15 / ≥0.15	≤0.57 / ≥1.74

Shown are the rages of associations estimates that could be detected with a power of 1-β>0.8 and at a statistical significance threshold of α<0.05.

[a] $R^2 = (beta\ x\ \sqrt{2\ x\ MAF(1-MAF)})^2$, where MAF is the minimum allele frequency and beta is the effect of the SNP on the respective cytokine levels (Park et al 2010, Nat. Genet. 42, 570–575).

[b] Odds Ratios are presented for the binary outcomes (small vessel stroke, intracerebral hemorrhage) and beta coefficients for the continuous outcomes (white matter hyperintensities volume).

[c] The R^2 for CETP, HMGCR, NPC1L1, PCSK9, ABCG5/G8, LDLR, PPARA, ANGPTL3, ANGPTL4, APOC3, and LPL drug targets correspond to the variance explained by variants in these loci for HDL-C, LDL-C, TG levels, as appropriately.

Supplementary Table 7. Mendelian randomization (MR) association estimates of cholesterol and triglyceride concentrations across lipoprotein particles with small vessel stroke and WMH volume. Shown are the results derived from IVW MR analyses.

	Small vessel stroke				WMH volume			
Outcome	**SNPs (N)**	**OR (95%CI)**	**p-value**	**p-het**	**SNPs (N)**	**beta (95%CI)**	**p-value**	**p-het**
Cholesterol in HDL particles								
M.HDL.C	8	0.84 (0.73-0.96)	**0.007**	0.183	10	-0.09 (-0.16 to -0.02)	**0.009**	0.232
L.HDL.C	42	0.99 (0.94-1.05)	0.781	0.509	54	0.00 (-0.03 to 0.03)	0.930	0.443
XL.HDL.C	30	0.98 (0.91-1.06)	0.646	0.044	36	0.00 (-0.04 to 0.04)	0.884	0.915
Cholesterol in LDL and larger particles								
S.LDL.C	36	1.08 (1.02-1.15)	**0.010**	0.539	46	0.06 (0.02 to 0.11)	**0.002**	0.012
M.LDL.C	36	1.07 (1.01-1.14)	0.030	0.716	49	0.06 (0.02 to 0.10)	**0.002**	0.027
L.LDL.C	40	1.07 (1.01-1.13)	0.022	0.614	57	0.06 (0.02 to 0.09)	**0.002**	0.095
IDL.C	42	1.08 (1.02-1.14)	**0.008**	0.589	42	0.05 (0.01 to 0.09)	**0.008**	0.027
S.VLDL.C	26	1.08 (1.00-1.18)	0.057	0.186	31	0.05 (-0.01 to 0.10)	0.091	0.367
M.VLDL.C	24	1.09 (1.01-1.19)	0.033	0.137	24	-0.01 (-0.07 to 0.06)	0.866	0.472
L.VLDL.C	17	1.05 (0.95-1.16)	0.354	0.116	17	-0.05 (-0.12 to 0.03)	0.215	0.791
Triglycerides in lipoprotein particles								
S.HDL.TG	25	**1.14 (1.04-1.23)**	**0.003**	0.175	31	-0.04 (-0.09 to 0.02)	0.200	0.018
XL.HDL.TG	76	1.03 (0.99-1.07)	0.165	0.437	88	0.00 (-0.02 to 0.03)	0.744	0.846
IDL.TG	41	1.07 (1.01-1.14)	0.027	0.685	41	0.02 (-0.02 to 0.06)	0.355	0.013
XS.VLDL.TG	27	1.07 (0.99-1.15)	0.095	0.325	28	-0.02 (-0.07 to 0.04)	0.568	0.039
S.VLDL.TG	26	1.08 (0.99-1.17)	0.074	0.164	26	-0.03 (-0.09 to 0.03)	0.264	0.177
M.VLDL.TG	18	1.03 (0.94-1.14)	0.509	0.162	21	-0.03 (-0.10 to 0.05)	0.486	0.857
L.VLDL.TG	15	1.04 (0.94-1.17)	0.432	0.029	15	-0.06 (-0.14 to 0.02)	0.131	0.663
XL.VLDL.TG	12	1.01 (0.89-1.15)	0.829	0.108	14	-0.04 (-0.13 to 0.04)	0.315	0.878
XXL.VLDL.TG	8	1.03 (0.89-1.21)	0.673	0.213	10	-0.04 (-0.14 to 0.06)	0.432	0.751

Bold indicates p-values <0.05 after adjustment for false discovery rate (FDR).

Odds Ratios correspond to 1 SD increment in the corresponding variable. P-het values correspond to the p-value of the Cochran's Q statistic exploring heterogeneity across the estimates.

Supplementary Table 8. Multivariable Mendelian randomization (MR) association estimates of cholesterol and triglyceride concentrations across lipoprotein particles with small vessel stroke and WMH volume.

	Small vessel stroke		WMH volume	
Outcome	**OR (95%CI)**	**p-value**	**beta (95%CI)**	**p-value**
Cholesterol in HDL particles [adjusted for LDL-C and TG]				
M.HDL.C	0.83 (0.76-0.91)	**$4.3x10^{-5}$**	-0.08 (-0.13 to -0.02)	**0.008**
L.HDL.C	0.95 (0.90-1.00)	0.063	-0.03 (-0.07 to 0.01)	0.145
XL.HDL.C	0.95 (0.89-1.01)	0.090	-0.02 (-0.07 to 0.02)	0.314
Cholesterol in LDL and larger particles [adjusted for HDL-C and TG]				
S.LDL.C	1.11 (1.03-1.19)	**0.004**	0.02 (-0.02 to 0.06)	0.366
M.LDL.C	1.11 (1.04-1.18)	**0.002**	0.02 (-0.02 to 0.06)	0.325
L.LDL.C	1.11 (1.04-1.19)	**0.001**	0.02 (-0.02 to 0.06)	0.338
IDL.C	1.11 (1.04-1.18)	**0.001**	0.02 (-0.02 to 0.05)	0.413
S.VLDL.C	1.15 (1.04-1.26)	**0.005**	-0.01 (-0.07 to 0.05)	0.821
M.VLDL.C	1.17 (1.02-1.35)	0.024	-0.05 (-0.14 to 0.05)	0.326
L.VLDL.C	1.08 (0.90-1.30)	0.420	-0.11 (-0.23 to 0.00)	0.058
TG in any lipoprotein particles [adjusted for HDL-C and LDL-C]				
S.HDL.TG	1.11 (1.02-1.22)	0.016	-0.06 (-0.12 to 0.00)	0.047
XL.HDL.TG	1.07 (1.02-1.14)	0.010	-0.03 (-0.07 to 0.01)	0.132
IDL.TG	1.09 (1.01-1.17)	0.018	-0.05 (-0.09 to 0.00)	0.051
XS.VLDL.TG	1.07 (1.00-1.15)	0.052	-0.04 (-0.08 to 0.01)	0.117
S.VLDL.TG	1.05 (0.97-1.13)	0.242	-0.04 (-0.09 to 0.00)	0.072
M.VLDL.TG	1.04 (0.96-1.13)	0.362	-0.06 (-0.10 to -0.01)	0.030
L.VLDL.TG	1.03 (0.94-1.12)	0.524	-0.05 (-0.11 to 0.00)	0.063
XL.VLDL.TG	1.00 (0.91-1.09)	0.937	-0.06 (-0.11 to 0.00)	0.038
XXL.VLDL.TG	1.02 (0.93-1.13)	0.646	-0.05 (-0.11 to 0.01)	0.117

Bold indicates p-values <0.05 after adjustment for false discovery rate (FDR).

Odds Ratios correspond to 1 SD increment in the corresponding variable.

For the multivariable MR analyses we used a genetic score for HDL-C, LDL-C and larger particle cholesterol, and for TGT, by combining all unique instruments significantly associated with HDL-C or cholesterol concentrations in HDL particles, LDL-C or cholesterol concentrations in LDL and larger ApoB particles, and with TG or triglyceride concentrations in any TG particles, respectively.

Supplementary Table 9. Mendelian randomization (MR) association estimates of cholesterol and triglyceride concentrations across lipoprotein particles with small vessel stroke and WMH volume, as derived from alternative MR methods in case of heterogeneity (p<0.10 in Cochran's Q) in the IVW analysis. Shown are the results derived from weighed median and MR-Egger analyses.

Small vessel stroke	MR method	OR	SE	95%CI		p-value
XL.HDL.C	IVW	0.96	0.062	0.85	1.09	0.537
	Weighted median	1.02	0.068	0.90	1.17	0.731
	MR Egger	0.95	0.212	0.63	1.44	0.812
	Egger Intercept	1.00	0.023	0.96	1.05	0.952
L.VLDL.TG	IVW	1.05	0.077	0.90	1.22	0.536
	Weighted median	1.01	0.079	0.86	1.18	0.910
	MR Egger	1.13	0.206	0.76	1.69	0.549
	Egger Intercept	0.99	0.022	0.95	1.03	0.690
WMH volume	**MR method**	**beta**	**SE**	**95%CI**		**p-value**
S.LDL.C	IVW	0.066	0.027	0.013	0.118	0.014
	Weighted median	0.054	0.03	-0.005	0.114	0.072
	MR Egger	0.098	0.05	0.000	0.197	0.050
	Egger Intercept	-0.005	0.006	-0.017	0.008	0.438
M.LDL.C	IVW	0.063	0.024	0.017	0.110	0.008
	Weighted median	0.088	0.032	0.026	0.150	0.006
	MR Egger	0.101	0.043	0.017	0.185	0.019
	Egger Intercept	-0.006	0.006	-0.017	0.005	0.292
L.LDL.C	IVW	0.058	0.021	0.017	0.099	0.005
	Weighted median	0.065	0.03	0.005	0.124	0.033
	MR Egger	0.099	0.037	0.027	0.171	0.007
	Egger Intercept	-0.007	0.005	-0.016	0.003	0.172
IDL.C	IVW	0.050	0.023	0.005	0.095	0.028
	Weighted median	0.053	0.027	0.000	0.106	0.051
	MR Egger	0.114	0.041	0.033	0.195	0.006
	Egger Intercept	-0.009	0.005	-0.018	0.001	0.067
IDL.TG	IVW	0.021	0.027	-0.031	0.073	0.428
	Weighted median	0.013	0.036	-0.056	0.083	0.710
	MR Egger	0.082	0.066	-0.047	0.211	0.214
	Egger Intercept	-0.008	0.008	-0.023	0.007	0.313
XS.VLDL.TG	IVW	-0.016	0.037	-0.088	0.056	0.667
	Weighted median	0.002	0.044	-0.084	0.088	0.964
	MR Egger	-0.036	0.119	-0.268	0.196	0.762
	Egger Intercept	0.002	0.012	-0.022	0.026	0.858

Odds Ratios correspond to 1 SD increment in the corresponding variable.

Supplementary Table 10. Mendelian randomization (MR) association estimates of HDL-C raising variants in/close to the *CEPT* locus, and risk of small vessel stroke, as derived from alternative MR methods. Shown are the results derived from weighed median and MR-Egger analyses, because the IVW MR analysis showed significant heterogeneity.

Small vessel stroke	MR method	OR	95%CI	p-value
HDL-C raising variants in *CETP*	IVW MR	0.82	(0.75-0.89)	0.001
	Weighted median	0.81	(0.68-0.96)	0.014
	MR Egger	0.71	(0.57-0.89)	0.003
	Egger Intercept	1.01	(0.99-1.03)	0.404

Odds Ratios correspond to 1 SD increment in HDL-C for the *CETP* variants.

Supplementary Table 11. Mendelian randomization (MR) association estimates of HDL-C raising variants in/close to the *CEPT* locus, and risk of small vessel stroke, WMH volume, and risk of intracerebral hemorrhage, as derived from IVW MR analyses and multivariable MR analyses adjusting for the effects of the variants on both HDL-C and LDL-C. Shown are the results derived from the IVW MR and multivariable MR analyses.

	IVW MR		Multivariable MR	
Outcome	**Effect estimate (95%CI)**	**p-value**	**Effect estimate (95%CI)**	**p-value**
Small vessel stroke	**OR**		**OR**	
HDL-C (1 SD increment)	0.82 (0.75-0.89)	9.0×10^{-6}	0.87 (0.74-1.02)	0.094
LDL-C (1 SD decrement)	0.41 (0.23-0.73)	0.003	1.40 (0.64-3.06)	0.414
WMH volume	**beta**		**beta**	
HDL-C (1 SD increment)	-0.08 (-0.13 to -0.02)	0.008	-0.27 (-0.64 to 0.11)	0.166
LDL-C (1 SD decrement)	-0.28 (0.00 to 0.56)	0.048	0.34 (-2.15 to 1.47)	0.714

Odds Ratios and beta coefficients correspond to 1 SD increment in HDL-C and 1 SD decrement in LDL-C.

Supplementary Table 12. Mendelian randomization (MR) association estimates of cholesterol and triglyceride concentrations across lipoprotein particles with intracerebral hemorrhage. Shown are the results derived from IVW MR and multivariable MR analyses.

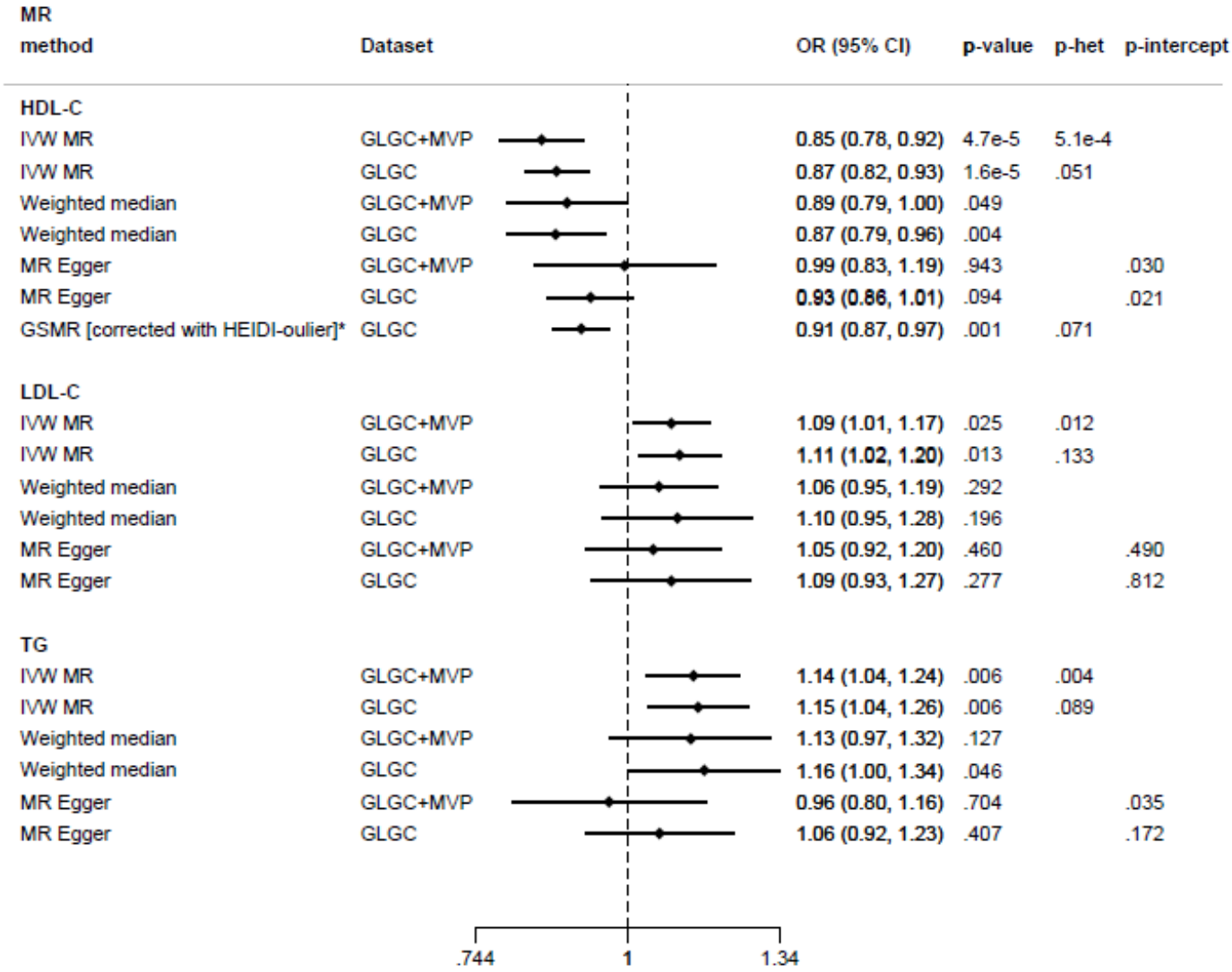
	IVW MR				Multivariable MR	
Outcome	**SNPs (N)**	**OR (95%CI)**	**p-value**	**p-het**	**OR (95%CI)**	**p-value**
Cholesterol in HDL particles [adjusted for LDL-C and TG]						
M.HDL.C	8	1.33 (0.92-1.92)	0.124	0.183	1.51 (1.17-1.95)	**0.001**
L.HDL.C	42	1.06 (0.89-1.26)	0.534	0.509	1.09 (0.93-1.29)	0.295
XL.HDL.C	30	1.07 (0.85-1.34)	0.570	0.044	1.15 (0.94-1.40)	0.172
Cholesterol in LDL and larger particles [adjusted for HDL-C and TG]						
S.LDL.C	36	0.76 (0.63-0.93)	**0.007**	0.702	0.84 (0.69-1.01)	0.066
M.LDL.C	36	0.81 (0.67-0.97)	0.019	0.472	0.85 (0.71-1.02)	0.088
L.LDL.C	40	0.79 (0.67-0.94)	**0.008**	0.283	0.83 (0.70-0.99)	0.036
IDL.C	42	0.78 (0.65-0.92)	**0.004**	0.589	0.78 (0.66-0.93)	**0.004**
S.VLDL.C	26	0.86 (0.66-1.12)	0.264	0.040	0.80 (0.61-1.05)	0.106
M.VLDL.C	24	1.01 (0.78-1.32)	0.923	0.074	0.90 (0.60-1.34)	0.592
L.VLDL.C	17	1.21 (0.86-1.69)	0.267	0.035	0.87 (0.51-1.49)	0.614
Triglycerides in any lipoprotein particle [adjusted for HDL-C and LDL-C]						
S.HDL.TG	25	0.85 (0.66-1.10)	0.223	0.175	1.14 (0.87-1.50)	0.346
XL.HDL.TG	76	0.93 (0.82-1.06)	0.291	0.437	0.94 (0.79-1.12)	0.477
IDL.TG	41	0.84 (0.68-1.04)	0.117	0.171	0.88 (0.70-1.10)	0.265
S.VLDL.TG	26	1.07 (0.82-1.39)	0.620	0.095	1.05 (0.84-1.31)	0.665
XS.VLDL.TG	27	0.96 (0.74-1.24)	0.732	0.156	1.09 (0.87-1.37)	0.447
M.VLDL.TG	18	1.13 (0.82-1.54)	0.458	0.050	1.23 (0.97-1.54)	0.085
L.VLDL.TG	15	1.40 (0.98-2.00)	0.065	0.249	1.26 (0.99-1.60)	0.055
XL.VLDL.TG	12	1.46 (0.99-2.14)	0.056	0.145	1.25 (0.97-1.60)	0.081
XXL.VLDL.TG	8	1.19 (0.77-1.86)	0.434	0.124	1.22 (0.92-1.61)	0.165

Bold indicates p-values <0.05 after adjustment for false discovery rate (FDR).

Odds Ratios correspond to 1 SD increment in the corresponding variable. P-het values correspond to the p-value of the Cochran's Q statistic exploring heterogeneity across the estimates.

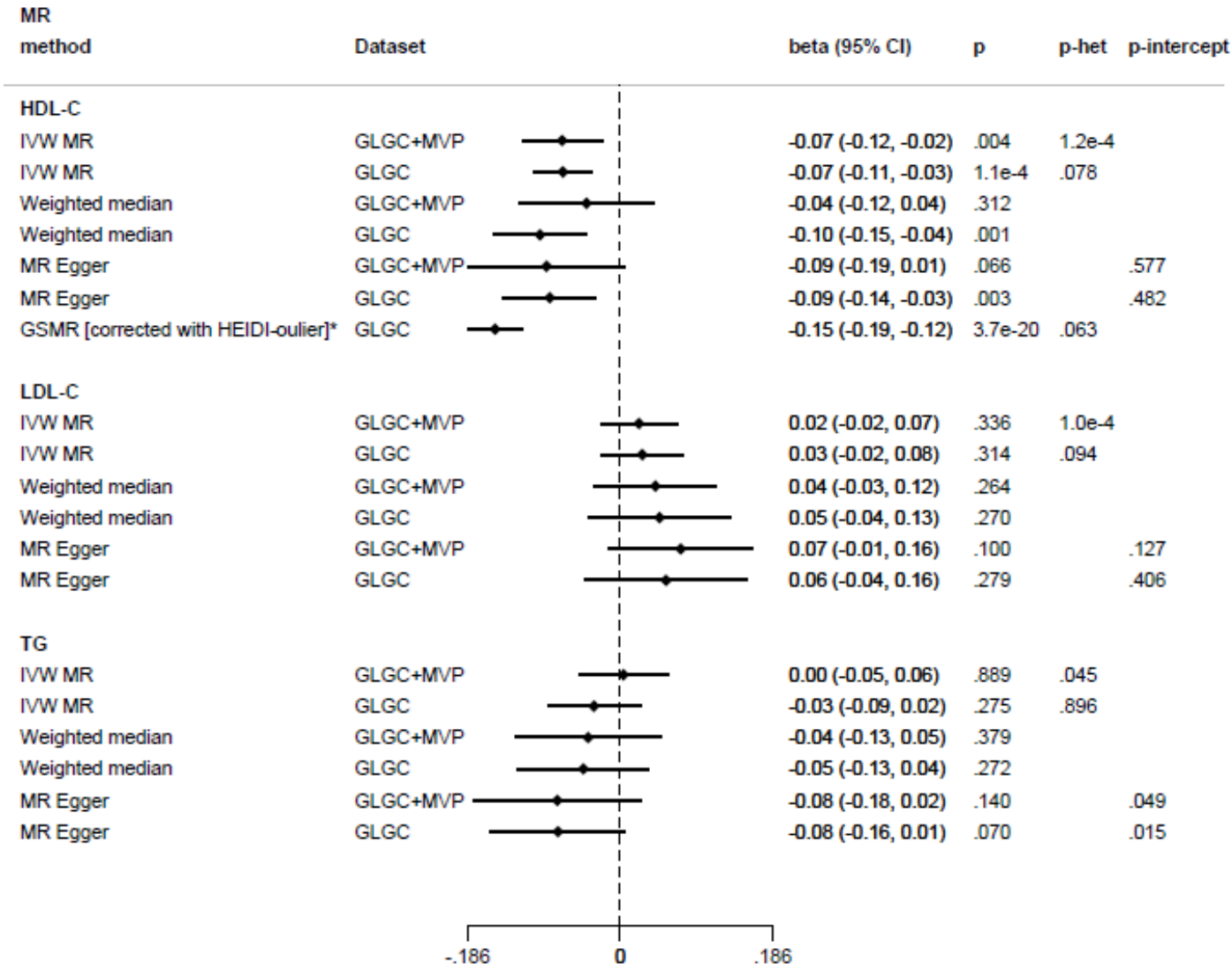
For the multivariable MR analyses we used a genetic score for HDL-C, LDL-C and larger particle cholesterol, and for triglycerides, by combining all unique instruments significantly associated with HDL-C or cholesterol concentrations in HDL particles, LDL-C or cholesterol concentrations in LDL and larger ApoB particles, and with TG or triglyceride concentrations in any TG particles. For the concentration of cholesterol in HDL particles, we have performed adjustments for LDL-C and TG, for concentration of cholesterol in LDL and larger ApoB particles, we have performed adjustments for HDL-C and TG, and for triglyceride concentrations we have performed adjustments for HDL-C and LDL-C.

Supplementary Figure 1. Sensitivity Mendelian randomization (MR) analyses between genetic determinants of blood lipid levels (HDL-C, LDL-C, TG) with risk of small vessel stroke. Shown are the results derived from IVW MR, weighted median, MR-Egger, and the GSMR approach incorporated with the HEIDI-outlier detector. The genetic instruments are weighted either based on the estimates derived from the full GLGC+MVP sample, or form those derived from the restricted GLGC dataset, where patients under lipid-modifying treatment had been excluded.



GLGC: global lipids genetics consortium; MVP: millions veteran program.
* the HEIDI-outlier approach detected rs11875988, rs11065979, and rs12417015 as outliers and excluded them from the analyses
p-het: derived from the Cochran Q statistic for the IVW analyses and from the global heterogeneity test for the GSMR analyses.
p-interecept: statistical significance of the intercept derived from MR-Egger regression analyses

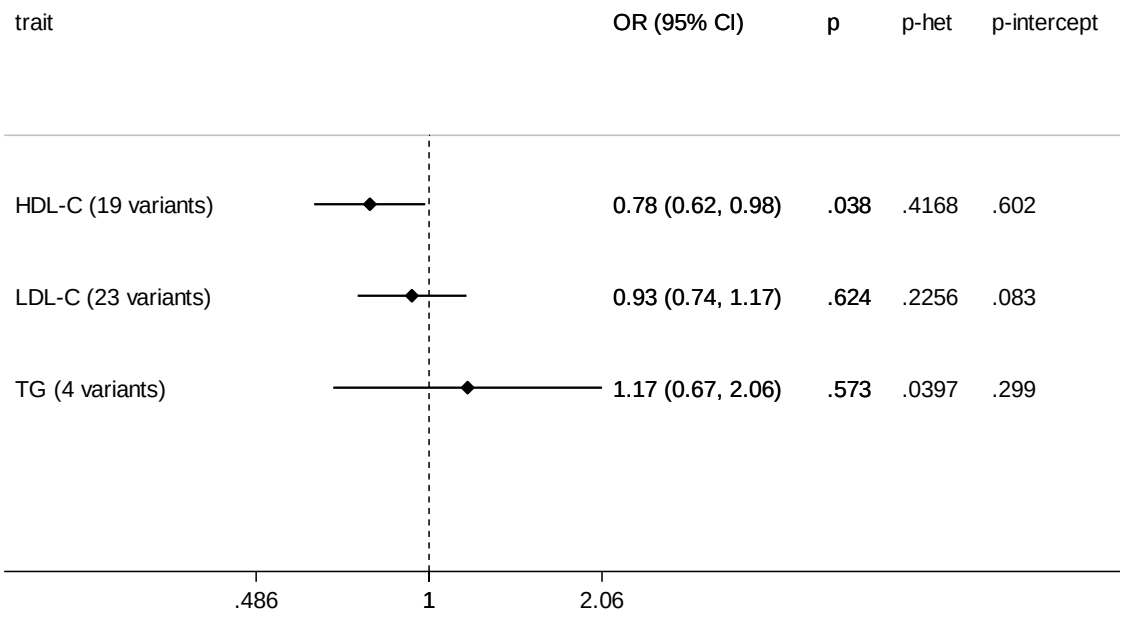
Supplementary Figure 2. Sensitivity Mendelian randomization (MR) analyses between genetic determinants of blood lipid levels (HDL-C, LDL-C, TG) with WMH volume.
Shown are the results derived from IVW MR, weighted median, MR-Egger, and the GSMR approach incorporated with the HEIDI-outlier detector. The genetic instruments are weighted either based on the estimates derived from the full GLGC+MVP sample, or form those derived from the restricted GLGC dataset, where patients under lipid-modifying treatment had been excluded.



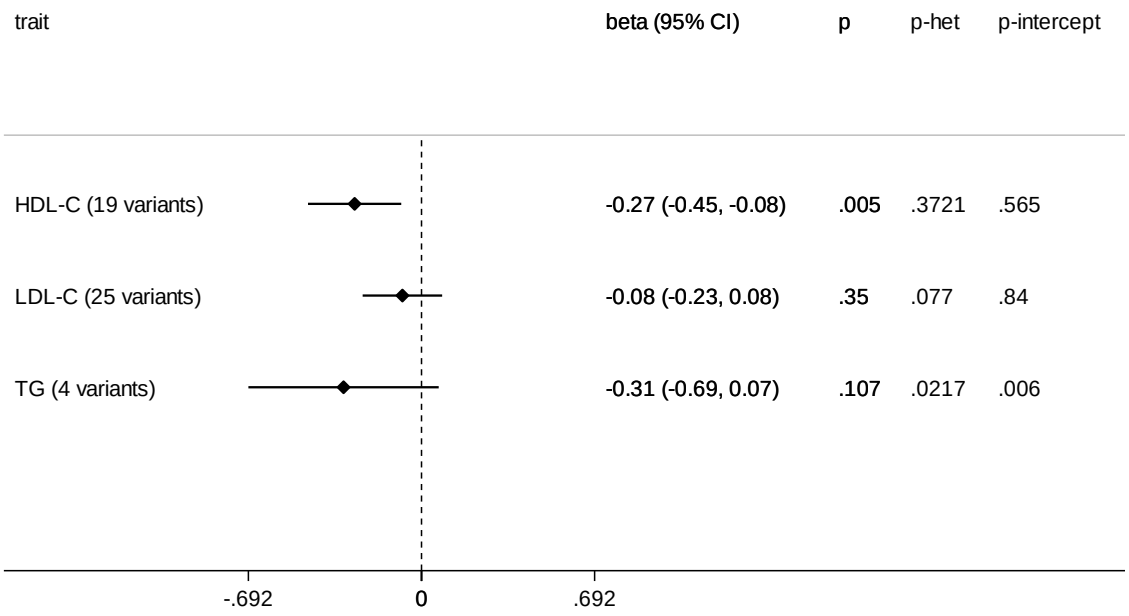
GLGC: global lipids genetics consortium; MVP: millions veteran program.
* the HEIDI-outlier approach detected rs701106 and rs13116385 as outliers and excluded them from the analyses
p-het: derived from the Cochran Q statistic for the IVW analyses and from the global heterogeneity test for the GSMR analyses.
p-interecept: statistical significance of the intercept derived from MR-Egger regression analyses

Supplementary Figure 3. Mendelian randomization (MR) associations between genetic determinants of blood lipid levels (HDL-C, LDL-C, TG) with (A) risk of small vessel stroke and (B) WMH volume when restricting instrument selection to those specific for the respective traits. Shown are the results derived from IVW MR.

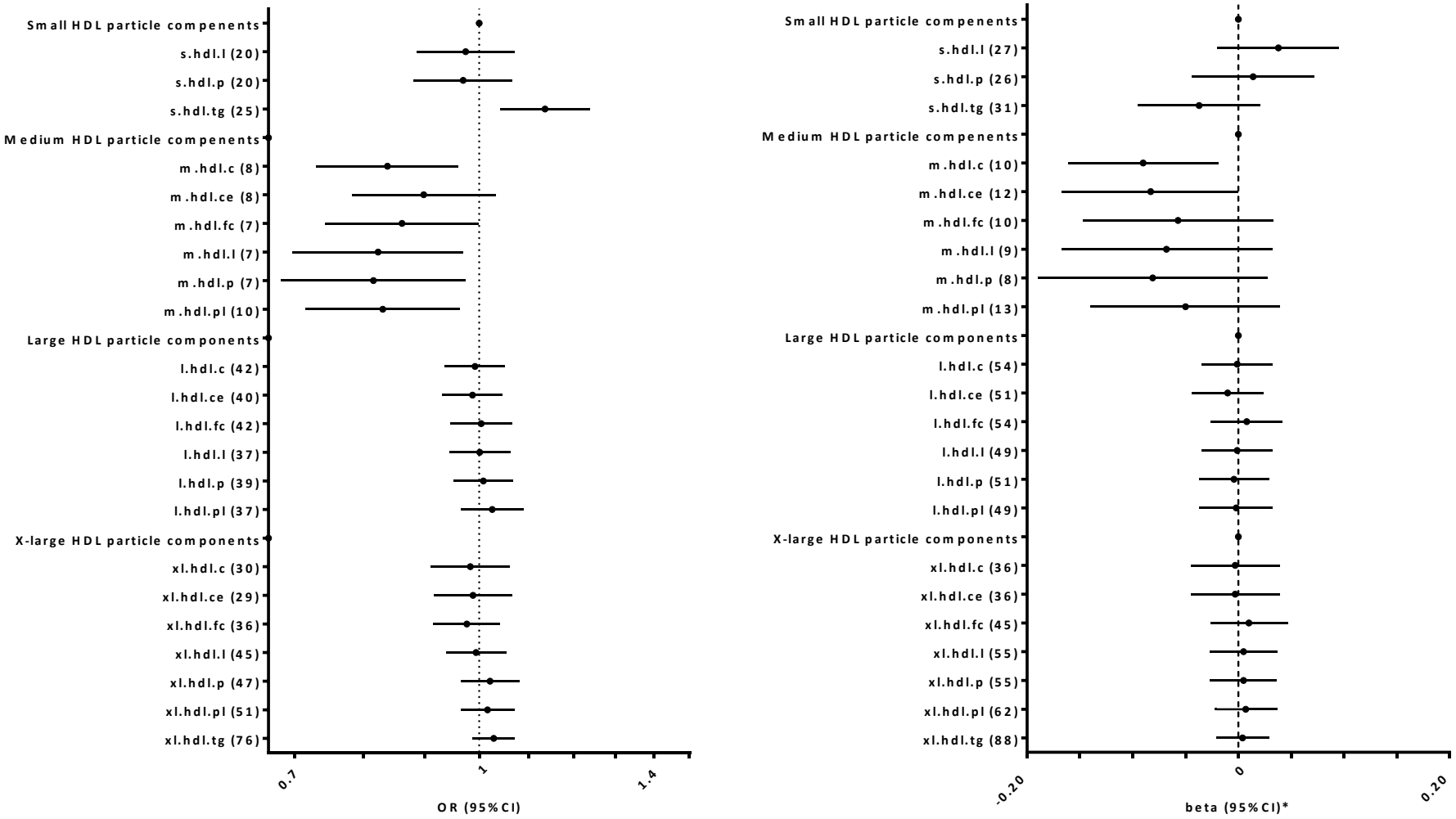
(A) Small vessel stroke



(B) WMH volume

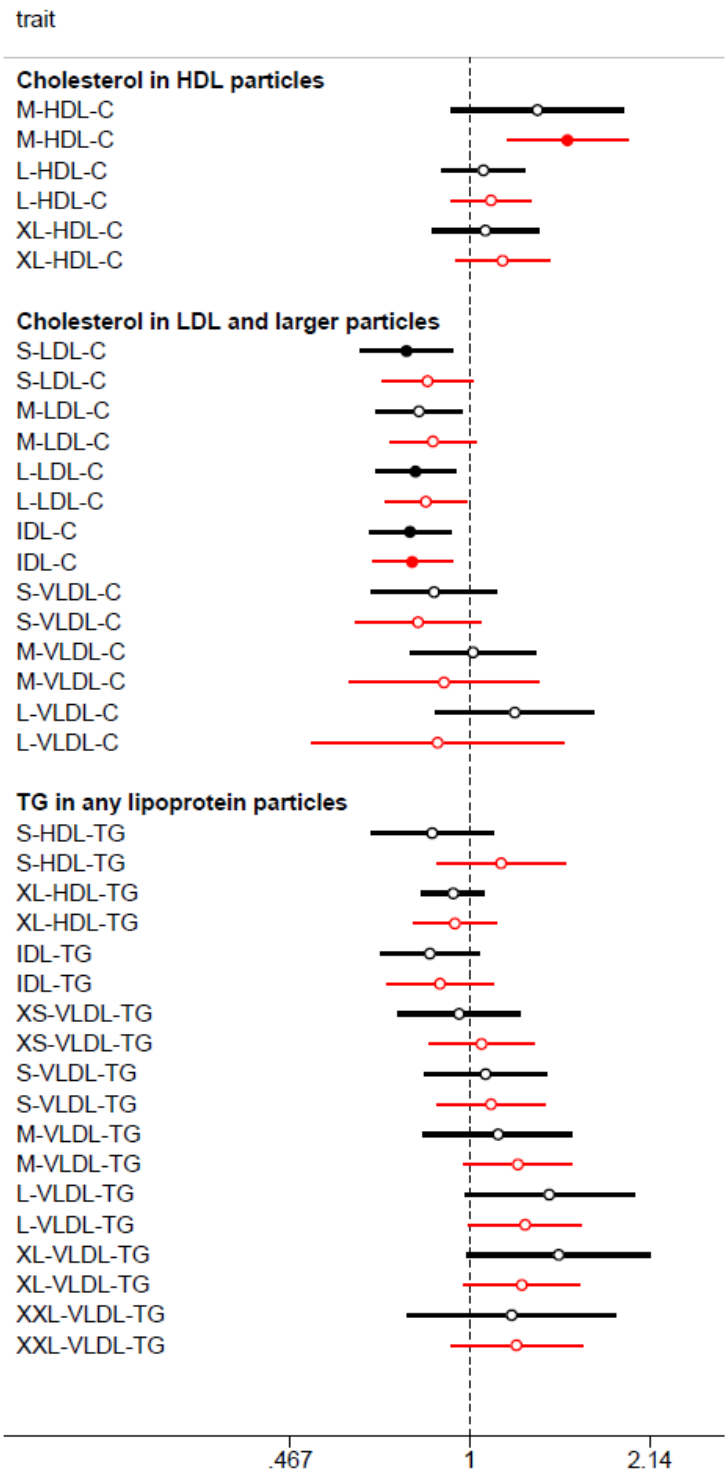


Supplementary Figure 4. Mendelian randomization (MR) associations of all components of the HDL lipoprotein particles defined by size with (A) risk of small vessel stroke and (B) WMH volume. Shown are the results derived from IVW MR analyses.



The first part of the abbreviations for the particle components indicates the size of the particles (small, medium, large, extra-large); the second refers to the type of lipoprotein particles (here HDL); the third part indicates the measured component in the respective size-defined particle: c, total cholesterol; ce, cholesterol-esters; fc, free cholesterol; l, total lipids; p, concentration of this particle class; pl, phospholipids; tg, triglycerides.

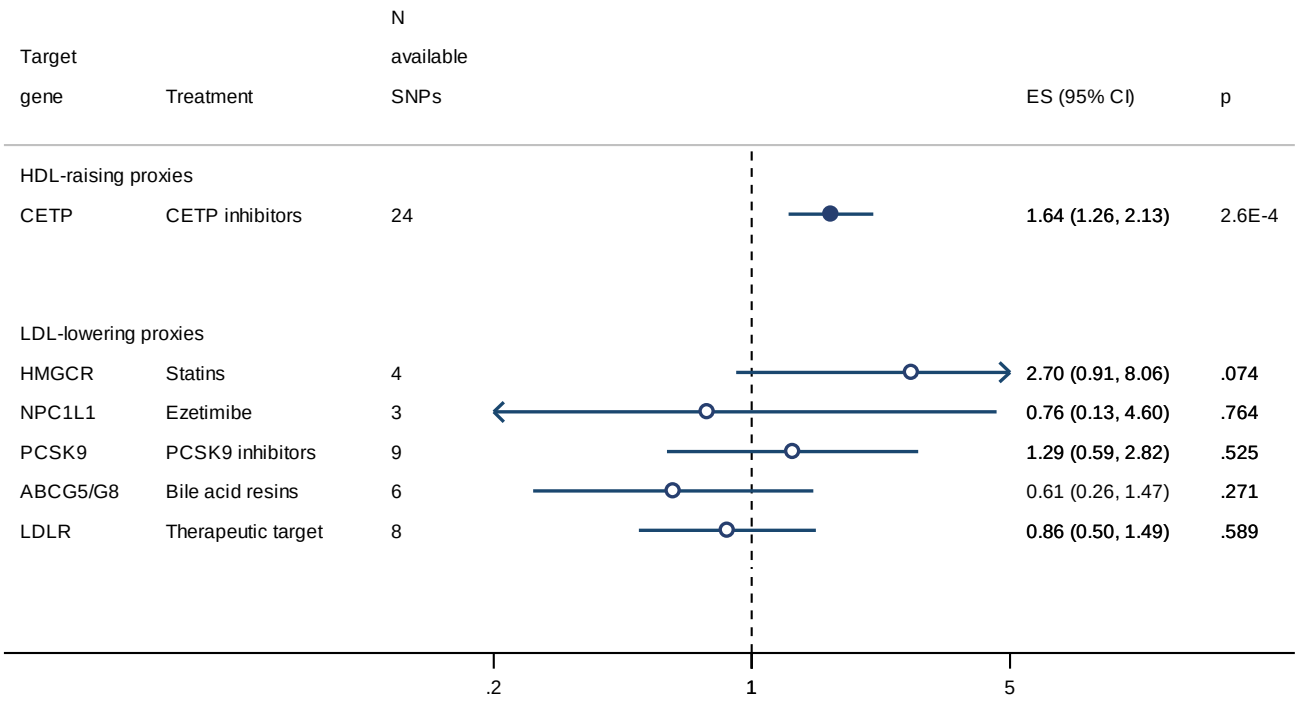
Supplementary Figure 5. Mendelian randomization (MR) associations of cholesterol (C) and triglyceride (TG) concentrations in lipoprotein particles defined by size with intracerebral hemorrhage. Shown are the results derived from IVW MR analyses.



Solid-centre circles indicate association estimates with a q-value <0.05 after adjusting for multiple testing comparisons with the false discovery rate (FDR).

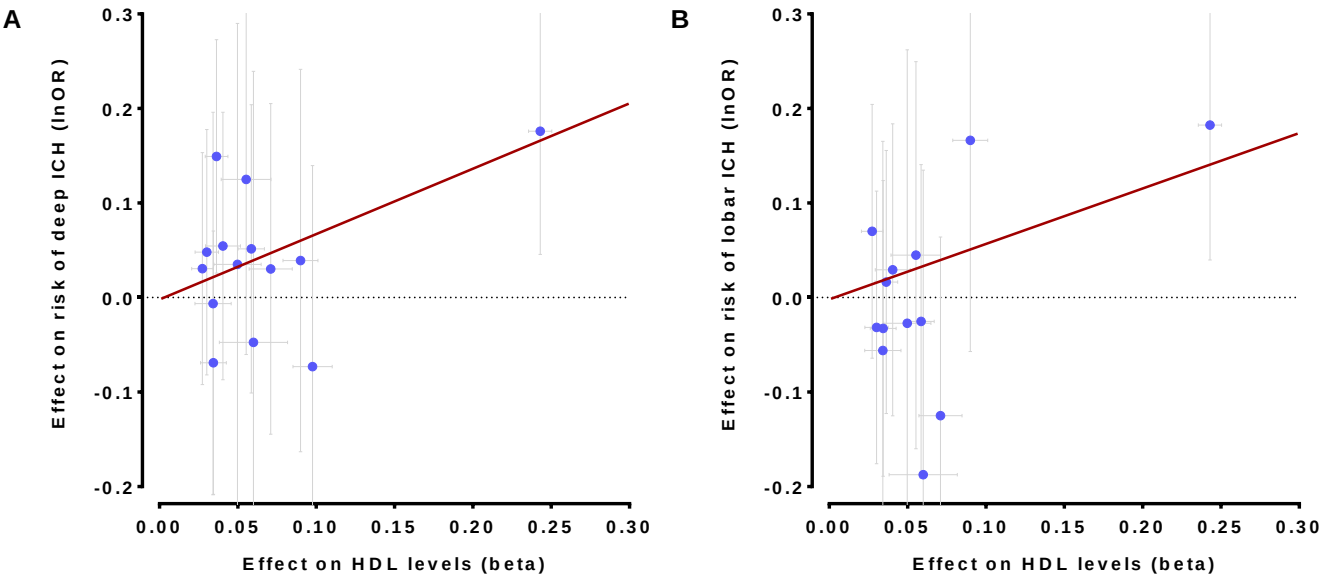
The black point estimates and confidence interval lines correspond to the results of the random-effects IVW MR analyses, whereas the red point estimates and confidence interval lines to the multivariable MR analyses.

Supplementary Figure 6. Mendelian randomization associations between HDL-C-raising and LDL-C-lowering genetic variants in the loci of known lipid-modifying drug targets and risk of intracerebral hemorrhage. Shown are the results derived from IVW MR analyses.



The results are scaled per 1 SD increment in circulating HDL-C levels for the HDL-C-raising drug targets and per 1 SD increment in circulating LDL- for the LDL-C-lowering drug targets, respectively.

Supplementary Figure 7. Mendelian randomization (MR) associations between HDL-C-raising genetic variants in the *CETP* locus and risk of (A) deep and (B) lobar intracerebral hemorrhage (ICH). Shown are the results from the IVW MR analyses. The results are scaled per 1 SD increment in circulating HDL-C levels.



CURRICULUM VITAE

Marios Georgakis, MD, DSc

Date/place of birth: Lefkada, Greece, October 31st 1991
Current position: **Neurology Resident physician & Postdoctoral research fellow**, Institute for Stroke and Dementia Research (ISD), LMU University Hospital
Feodor-Lynen-Straße 17, 81377 Munich, Germany,
e-mail: marios.georgakis@med.uni-muenchen.de, tel: +49 (0)89 4400 – 46126

EDUCATION

10/2017-04/2020 ***Doctoral studies (Ph.D.)*** in Neurosciences, Graduate School of Systemic Neurosciences, LMU, Munich

09/2015-09/2019 ***Doctoral studies (D.Sc.)*** in Epidemiology, National & Kapodistrian University of Athens
Graduation grade: "Honors"

09/2015-09/2017 ***Master studies (M.Sc.):*** Neurosciences, National & Kapodistrian University of Athens
Graduation grade: 9.8/10 ("Honors"), top 5% of class

09/2009-08/2015 ***Medical studies (M.D.):*** Medical School, National & Kapodistrian University of Athens
Graduation grade: 8.8/10 ("Honors"), top 10% of class

09/2003-06/2009 ***Secondary education*****:** 1st Gymnasium & 1st Lyceum of Lefkas, Greece
Panhellenic exams grade: 19.47 out of 20 ("Honors")

CLINICAL POSITIONS

01/2020-today **Neurology Resident,** Institute for Stroke and Dementia Research & Neurology Department, LMU University Hospital, Munich, Germany

RESEARCH EXPERIENCE

01/2020-today **Postdoctoral research fellow** in Institute for Stroke and Dementia Research, University Hospital of Munich, Ludwig-Maximillian University (LMU), Germany
PI: Prof. Martin Dichgans
Research focus: Genetic and clinical epidemiology of cerebrovascular disease and vascular dementia

10/2017-12/2019 **Doctoral researcher** in Institute for Stroke and Dementia Research, University Hospital of Munich, Ludwig-Maximillian University (LMU), Germany
Supervisor: Prof. Martin Dichgans
Research focus: Genome-phenome interactions on the pathogenesis of stroke and its subtypes

06/2013-09/2017 **Undergraduate research fellow and postdoctoral researcher** in Department of Epidemiology, Medical School, National and Kapodistrian University of Athens, Greece
Supervisor: Prof. Eleni Th. Petridou
Research focus: Epidemiology of neuropsychiatric disorders of the elderly, neuro-oncology

09/2016-09/2017 **Research fellow (Master thesis student)** in Lab of Neurodegenerative diseases, Bio-academy of Athens, Greece
Supervisor: Prof. Leonidas Stefanis
Topic: The role of chaperone-mediated autophagy in Parkinson's disease (basic science project)

09/2015-09/2017 **Research fellow** (long-distance), Department of Women's Health, Uppsala University, Sweden
PI: Prof. Alkistis Skalkidou
Research focus: postpartum depression

AWARDS/ACHIEVEMENTS

10/2019 **Best Oral Presentation Award** at the 2019 Meeting of International Stroke Genetics Consortium, St. Louis, US

05/2019 **Best Poster Award** at 5th European Stroke Organization Conference, Milan, Italy

10/2018-09/2020 **Scholarship** for Doctoral studies by the Onassis Public Benefit Foundation

10/2018-09/2019 **Research Grant** for Doctoral studies by the German Academic Exchange Service (DAAD)

09/2018 **Travel Award** - Neurepiomics Summer School 2018, Bordeaux, France

09/2015- 08/2017 **Scholarship** for Master studies by the "Bodossaki Foundation"

09/2015 **Best Poster Award** at 27th Greek Conference of Social Pediatrics and Health Promotion, Sparti-Monemvasia

09/2010-08/2015 **Scholarship** for Medical studies by the legacy of "Antonios Papadakis"

04/2008 **Third Award by Greek Mathematical Society** in national mathematical "Euclid" exams contest as a 2nd year High School student

OTHER ACADEMIC ACTIVITIES

2017-today **Invited reviewer for scientific journals:**
J Amer Col Cardiol, Neurology, JACC Heart Failure, J Amer Ger Soc, J Neurol, J Neurol Sciences, J Affect Dis, J Psych Res, Front Neurol

Member in scientific societies:
2018-today - ISGC: International Stroke Genetics Consortium
2019-today - ESOC: European Stroke Organization
2019-today - Hellenic Society of Cerebrovascular Diseases
2020-today - HIAAD: Hellenic Initiative Against Alzheimer's Disease

Teaching activities:

2018-today	- Supervising PhD and MD students in the context of their theses, LMU Munich, Germany
2016-2017	- Lectures to MD students in the context of their Epidemiology, Preventive Medicine, and English Medical Terminology courses, University of Athens, Greece
2014-2017	- Supervising of MD students in the context of elective projects for their Epidemiology course, University of Athens, Greece

Continuing education:

2017-2019	- Advanced courses on Statistics, Advanced Epidemiology, and Genetic Epidemiology offered by the Master in Epidemiology and Public Health of LMU Munich
09/2018	- Neurepiomics Summer School on “Epidemiology of -omics data”, Bordeaux, France
01-04/2015	- Medical Neuroscience 12-week online course, Duke University, North Carolina, US

OTHER SKILLS

Languages	English (C2 level), German (C1 level), Greek (native)
Computer skills	Statistical analysis programming (R, SAS, STATA, SPSS)

LIST OF PUBLICATIONS

54 peer-reviewed publications and 14 manuscripts accepted/in revision/submitted (1st author in 41; 2nd author in 7; last author in 1)

Citations: 562; h-index: 14 (Google Scholar, as of 24 April 2020: https://scholar.google.gr/citations?user=_Td2rBwAAAAJ&hl=el)

- **Georgakis MK,** Malik R, Björkbacka H, Pana TA, … Dichgans M. Circulating monocyte chemoattractant protein-1 and risk of stroke: a meta-analysis of population-based studies involving 17,180 individuals. ***Circ Res***. 2019 Sep 27;125(8):773-782. doi: 10.1161/CIRCRESAHA.119.315380. Epub 2019 Sep 3. ***[IF: 15.9***]
- **Georgakis MK,** Malik R, Gill D, Franceschini N, Sudlow CLM, INVENT Consortium, CHARGE Inflammation Working Group, Dichgans M. Interleukin-6 signaling effects on ischemic stroke and other cardiovascular outcomes: a Mendelian Randomization study. [Accepted in ***Circ Gen Prec Med***] ***[IF: 6.1]***
- **Georgakis MK,** Malik R, Anderson CD, Parhofer KG, Hopewell JC, Dichgans M. Genetically determined blood lipids and cerebral small vessel disease: role of HDL cholesterol. ***Brain***. 2020 Feb 1;143(2):597-610. doi: 10.1093/brain/awz413. ***[IF: 11.8]***
- Gill D*, **Georgakis MK*,** Koskeridis F, Jiang Feng Theodoratou Elliott Denny JC, Malik R, Evangelou E, Dehghan A, Dichgans M, Tzoulaki I. Genetic variants related to antihypertensive targets inform drug efficacy and side effects. ***Circulation***. 2019 Jul 23;140(4):270-279. ***[IF: 23.1]***
- **Georgakis MK*,** Gill D*, Webb AJS, Evangelou E, Elliott P, Sudlow CLM, Dehghan A, Malik R, Tzoulaki I, Dichgans M. Genetically determined blood pressure, antihypertensive drug classes, and risk of stroke: a Mendelian Randomization Study. [Accepted in ***Neurology***] *(*equally contributed*) ***[IF: 8.7]***
- **Georgakis MK,** Gill D, Rannikmäe K, Traylor M, Anderson CD, MEGASTROKE consortium (ISGC), Lee JM, Kamatani Y, Hopewell JC, Worrall BB, Bernhagen J, Sudlow CLM, Malik R, Dichgans M. Genetically Determined Levels of Circulating Cytokines and Risk of Stroke: Role of Monocyte Chemoattractant Protein-1. ***Circulation***. 2019 Jan 8;139(2):256-268. ***[IF: 23.1]***

- Marini S*, **Georgakis MK*,** Chung J, Henry JQA, Dichgans M, Rosand J, Malik R, Andreson CD. Genetic overlap and causal inferences between impaired kidney function and cerebrovascular diseases. [Accepted in ***Neurology***] *(*equally contributed*) ***[IF: 8.7]***
- Katsanos AH, Palaiodimou L, Price C, Giannopoulos S, Lemmens R, Kosmidou M, **Georgakis MK,** Weimar C, Kelly PJ, Tsivgoulis G. Colchicine for stroke prevention in patients with coronary artery disease: a systematic review and meta-analysis. ***Eur J Neurol.*** 2020 Mar 5. doi: 10.1111/ene.14198. ***[IF: 4.4]***
- Zumel-Marne A, Kundi M, Castaño-Vinyals G, Alguacil J, Petridou ET, **Georgakis MK,** …, Cardis E. Clinical presentation of young people (10-24 years old) with brain tumors: results from the international MOBI-Kids study. ***J Neurooncol.*** 2020 Apr;147(2):427-440. doi: 10.1007/s11060-020-03437-4. ***[IF: 3.1]***
- **Georgakis MK,** Beskou-Kontou T, Theodoridis I, Skalkidou A, Petridou ET. Surgical menopause in association with cognitive function and risk of dementia: A systematic review and meta-analysis. ***Psychoneuroendocrinology***. 2019 Mar 19;106:9-19. ***[IF: 4.7]***
- Nyquist P, **Georgakis MK**. Remote ischemic preconditioning effects on brain vasculature: don't cry because it's over, smile because it happened. ***Neurology.*** 2019 Jul 2;93(1):15-16. [Editorial] ***[IF: 8.7]***
- **Georgakis MK,** Duering M, Wardlaw JM, Dichgans M. White matter hyperintensities and long-term outcomes in ischemic stroke: a systematic review and meta-analysis. ***Neurology***. 2019 Mar 19;92(12):e1298-e1308. ***[IF: 8.7]***
- Weaver NA, Zhao L, Biesbroek JM, Kuijf HJ, Aben HP, Bae HJ, Caballero MÁA, Chappell FM, Chen CPLH, Dichgans M, Duering M, **Georgakis MK,** … Biessels GJ. The Meta VCI Map consortium for meta-analyses on strategic lesion locations for vascular cognitive impairment using lesion-symptom mapping: Design and multicenter pilot study. ***Alzheimers Dement (Amst).*** 2019 Apr 12;11:310-326. ***[IF: 14.4]***
- Malik R, Rannikmäe K, Traylor M, **Georgakis MK**, …, Dichgans M. Genome-Wide Meta-analysis identifies three novel loci associated with stroke. ***Ann Neurol*.** 2018 Dec;84(6):934-939. ***[IF: 9.5]***
- Zietemann V*, **Georgakis MK*,** Dondaine T, Müller C, Mendyk AM, Kopczak A, Hénon H, Bombois S, Wollenweber FA, Bordet R, Dichgans M. Early MoCA predicts long-term

cognitive and functional outcome and mortality after stroke. ***Neurology.*** 2018 Nov 13;91(20):e1838-e1850. *(*equally contributed*) ***[IF: 8.7]***

- Gill D, **Georgakis MK,** Laffan M, Sabater-Lleal M, Malik R, Tzoulaki I, Veltkamp R, Denhgan A. Genetically determined factor XI levels and risk of cardiovascular disease. ***Stroke*****.** 2018 Nov;49(11):2761-2763. ***[IF: 6.0]***
- Gill D, Monori G, **Georgakis MK,** Tzoulaki I, Laffan M. Genetically Determined Platelet Count and Risk of Cardiovascular Disease. ***Arterioscler Thromb Vasc Biol.*** 2018 Dec;38(12):2862-2869. ***[IF: 6.7]***
- Papadopoulos A, Palaiopanos K, Protogerou AP, Paraskeuas G, Tsivgoulis G, **Georgakis MK**. Left ventricular hypertrophy and cerebral small vessel disease: a systematic review and meta-analysis. [Submitted]
- Malsch C, Liman T, Wiedmann S, Siegerink B, **Georgakis MK,** Tiedt S, Endres M, Heuschmann PU. Outcome after stroke attributable to baseline factors-The PROSpective Cohort with Incident Stroke (PROSCIS). ***PLoS One.*** 2018 Sep 26;13(9):e0204285. ***[IF: 2.8]***
- Karalexi MA, Dessypris N, **Georgakis MK,** Ryzhov A, … Petridou ET. Birth seasonality patterns of childhood central nervous system tumors: analysis of 6000 cases from 16 Southern-Eastern European population-based cancer registries. [In revision in ***Int J Cancer***] ***[IF: 7.4]***
- **Georgakis MK,** Dessypris N, Papadakis V, Tragiannidis A, Bouka E, …, Petridou ET; NARECHEM-ST CNS tumors Working Group. Perinatal and early life risk factors for childhood brain tumors: Is instrument-assisted delivery associated with higher risk? ***Cancer Epidemiol***. 2019 Apr;59:178-184. ***[IF: 2.7]***
- **Georgakis MK,** Tsivgoulis G, Spinos D, Liaskas A, Herrlinger U, Petridou ET. Prognostic Factors and Survival of Gliomatosis Cerebri: A Systematic Review and Meta-Analysis. ***World Neurosurg*****.** 2018 Dec;120:e818-e854. ***[IF: 1.9]***
- Petridou ET, **Georgakis MK,** Erdmann F, Ma X, Heck JE, ... , Skalkidou A. Advanced parental age as risk factor for childhood acute lymphoblastic leukemia: results from studies of the Childhood Leukemia International Consortium. ***Eur J Epidemiol*****.** 2018 Oct;33(10):965-976. ***[IF: 6.5]***

- Benetou DR, Stergianos E, Geropeppa M, Ntinopoulou E, Tzanni M, Pourtsidis A, Petropoulos AC, **Georgakis MK**, Tousoulis D, Petridou ET. Late-onset cardiomyopathy among survivors of childhood lymphoma treated with anthracyclines: a systematic review. ***Hellenic J Cardiol*.** 2018 Sep 29. ***[IF: 2.3]***

- Cato K, Sylvén SM, **Georgakis MK**, Kollia N, Rubertsson C, Skalkidou A. Antenatal depressive symptoms and early initiation of breastfeeding in association with exclusive breastfeeding six weeks postpartum: a longitudinal population-based study. ***BMC Pregnancy Childbirth*.** 2019 Jan 29;19(1):49. ***[IF: 2.3]***

- **Georgakis MK,** Tsivgoulis G, Spinos D, Dimitriou NG, Kyritsis AP, Herrlinger U, Petridou ET. Clinical, neuroimaging and histopathological features of gliomatosis cerebri: a systematic review based on synthesis of published individual patient data. ***J Neurooncol***. 2018 Nov;140(2):467-475. ***[IF: 3.1]***

- **Georgakis MK,** Tsivgoulis G, Pourtsidis A, Petridou ET. Gliomatosis Cerebri Among Children and Adolescents: An Individual-Patient Data Meta-analysis of 182 Patients. ***J Child Neurol*.** 2019 Jun;34(7):394-401. ***[IF: 2.1]***

- **Georgakis MK**, Spinos D, Pourtsidis A, Psyrri A, Panourias IG, Sgouros S, Petridou ET. Incidence and survival of gliomatosis cerebri: a population-based cancer registration study. ***J Neurooncol*.** 2018 Jun;138(2):341-349. ***[IF: 3.1]***

- **Georgakis MK,** Chatzopoulou D, Tsivgoulis G, Petridou ET. Albuminuria and cerebral small-vessel disease: a meta-analysis. ***J Am Geriatr Soc*.** 2018 Mar;66(3):509-517. ***[IF: 4.1]***

- Panagopoulou P, **Georgakis MK**, Baka M, Moschovi M, … , Petridou ET. Persisting inequalities in survival patterns of childhood neuroblastoma in Southern and Eastern Europe and the effect of socio-economic development compared with those of the US. ***Eur J Cancer*.** 2018 Jun;96:44-53. ***[IF: 6.7]***

- **Georgakis MK**, Dessypris N, Baka M, Moschovi M, … , Petridou ET. Neuroblastoma among children in Southern and Eastern European cancer registries: Variations in incidence and temporal trends compared to US. ***Int J Cancer*.** 2018 May 15;142(10):1977-1985. ***[IF: 7.4]***

- Doganis D, Panagopoulou P, Tragiannidis A, **Georgakis MK**, … , Petridou ET. Childhood nephroblastoma in Southern and Eastern Europe and the US: Incidence

variations and temporal trends by human development index. ***Cancer Epidemiol*.** 2018 Jun;54:75-81. ***[IF: 2.7]***

- Skalkidou A, Kullinger M, **Georgakis MK**, Kieler H, Kesmodel US. Systematic misclassification of gestational age by ultrasound biometry: implications for clinical practice and research methodology in the Nordic countries. ***Acta Obstet Gynecol Scand*.** 2018 Apr;97(4):440-444. ***[IF: 2.7]***
- **Georgakis MK,** Papathoma P, Ryzhov A, Zivkovic-Perisic S, …, Petridou ET. Malignant central nervous system tumors in adolescents and young adults (15-39 years) in Southern-Eastern Europe and SEER, US: mortality and survival patterns. ***Cancer*.** 2017 Nov 15;123(22):4458-4471. ***[IF: 6.1]***
- Iliadis SI, Skalkidou A, Ranstrand H, **Georgakis MK,** Axfors C, Papadopoulos FC. Self-Harm Thoughts Postpartum as a Marker for Long-Term Morbidity. ***Front Public Health*.** 2018 Feb 19;6:34. ***[IF: 2.5]***
- **Georgakis MK**, Dimitriou NG, Karalexi MA, Mihas C, Nasothimiou EG, Tousoulis D, Tsivgoulis G, Petridou ET. Albuminuria in association with dementia and cognitive function: a systematic review and meta-analysis***. J Am Ger Soc*.** 2017 Jun;65(6):1190-1198. ***[IF: 4.1]***
- Eckerdal P, **Georgakis MK**, Kollia N, Wikström AK, Högberg U, Skalkidou A. Delineating the association between mode of delivery and postpartum depression symptoms: a longitudinal study. ***Acta Obstet Gynecol Scand.*** 2018 Mar;97(3):301-311. ***[IF: 2.7]***
- **Georgakis MK**, Panagopoulou P, Papathoma P, Tragiannidis A, … , Petridou ET. Central nervous system tumours among adolescents and young adults (15-39 years) in Southern and Eastern Europe: Registration improvements reveal higher incidence rates compared to the US. ***Eur J Cancer*.** 2017 Nov; 86:46-58. ***[IF: 6.7]***
- Gorgui J, Gasbarrino K, **Georgakis MK,** Karalexi MA, Nauche B, Petridou ET, Daskalopoulou SS. Circulating adiponectin levels in relation to carotid atherosclerotic plaque presence, ischemic stroke risk, and mortality: A systematic review and meta-analyses. ***Metabolism*.** 2017 Apr;69 :51-66. ***[IF: 6.5]***
- **Georgakis MK,** Papadopoulos FC, Beratis I, Michelakos T, … , Petridou ET. Validation of TICS for detection of dementia and mild cognitive impairment among individuals

characterized by low levels of education or illiteracy: a population-based study in rural Greece. ***Clin Neuropsychol*.** 2017 Jan-Dec;31(sup1):61-71. ***[IF: 2.0]***

- **Georgakis MK,** Kalogirou EI, Liaskas A, Karalexi MA, Papathoma P, Ladopoulou K, Kantzanou M, Tsivgoulis G; NARECHEM-BT Working Group, Petridou ET. Anthropometrics at birth and risk of a primary central nervous system tumour: A systematic review and meta-analysis. ***Eur J Cancer*.** 2017 Apr;75:117-131. ***[IF: 6.7]***
- Salih Joelsson L, Tydén T, Wanggren K, **Georgakis MK,** Stern J, Berglund A, Skalkidou A. Anxiety and depression symptoms among sub-fertile women, women pregnant after infertility treatment, and naturally pregnant women. ***Eur Psychiatry*.** 2017 Sep;45:212-219. ***[IF: 3.9]***
- **Georgakis MK**, Protogerou AD, Kalogirou EI, Tousoulis D, Petridou ET. Advanced statistical methodologies to address inherent study limitations. Author Response to Ayubi and Saeid. ***J Clin Hypertens (Greenwich)*.** 2017 Sep;19(9):923-924. ***[IF: 2.4]***
- **Georgakis MK**, Synetos A, Mihas A, Karalexi MA, Seshadri S, Petridou ET. Left ventricular hypertrophy in association with cognitive impairment: a systematic review and meta-analysis. ***Hypertens Res.*** 2017 Jul;40(7):696-709. ***[IF: 3.2]***
- Sundström Poromaa I, Comasco E, **Georgakis MK**, Skalkidou A. Sex differences in depression during pregnancy and the postpartum period. ***J Neurosci Res*.** 2017 Jan 2;95(1-2):719-730. ***[IF: 4.1]***
- **Georgakis MK,** Ntinopoulou E, Chatzopoulou D, Petridou ET. Season of birth and primary central nervous system tumors: a systematic review of the literature with critical appraisal of underlying mechanisms. ***Ann Epidemiol*.** 2017 Sep;27(9):593-602.e3. ***[IF: 2.6]***
- Skalkidou A, Sergentanis TN, Gialamas SP, **Georgakis MK**, Psaltopoulou T, Trivella M, Siristatidis CS, Evangelou E, Petridou E. Risk of endometrial cancer in women treated with ovary-stimulating drugs for subfertility. ***Cochrane Database Syst Rev*.** 2017 Mar 25;3:CD010931. ***[IF: 7.8]***
- **Georgakis MK**, Protogerou AD, Kalogirou EI, Kontogeorgi E, … , Petridou ET. Blood Pressure and All-Cause Mortality by Level of Cognitive Function in the Elderly: Results From a Population-Based Study in Rural Greece. ***J Clin Hypertens (Greenwich)*.** 2017 Sep;19(9):923-924. ***[IF: 2.4]***

- **Georgakis MK**, Kalogirou EI, Diamantaras AA, Daskalopoulou SS, Munro CA, Lyketsos CG, Skalkidou A, Petridou ET. Age at menopause and duration of reproductive period in association with dementia and cognitive function: A systematic review and meta-analysis. ***Psychoneuroendocrinology*.** 2016 3;73:224-243. ***[IF: 4.7]***
- **Georgakis MK,** Karalexi MA, Kalogirou EI, Ryzhov A, ... , Petridou ET. Incidence, time trends and survival patterns of childhood pilocytic astrocytomas in Southern-Eastern Europe and SEER, US. ***J Neurooncol.*** 2017 Jan;131(1):163-175. ***[IF: 3.1]***
- Karalexi MA, **Georgakis MK**, Dessypris N, Ryzhov A, ... , Petridou ET. Mortality and survival patterns of childhood lymphomas: geographic and age-specific patterns in Southern-Eastern European and SEER/US registration data. ***Hematol Oncol*.** 2017 Dec;35(4):608-618. ***[IF: 3.4]***
- **Georgakis MK**, Papadopoulos FC, Protogerou AD, Pagonari I, … , Petridou ET. Comorbidity of cognitive impairment and late -life depression increase mortality: results from a cohort of community-dwelling elderly in rural Greece. ***J Geriatr Psychiatry Neurol.*** 2016 Jul;29(4):195-204. ***[IF: 2.7]***
- **Georgakis MK**, Thomopoulos TP, Diamantaras AA, Kalogirou EI, Skalkidou A, Daskalopoulou SS, Petridou ET. Age at menopause and duration of reproductive period in association with depression in postmenopausal women: a systematic review and meta-analysis*. **JAMA Psychiatry.*** 2016;73(2):139-149. ***[IF: 15.9]***
- **Georgakis MK,** Skalkidou A, Petridou ET. Estrogen-Based Therapies and Depression in Women Who Naturally Enter Menopause Before Population Average-Reply. ***JAMA Psychiatry*.** 2016 Aug 1;73(8):874-5. ***[IF: 15.9]***
- **Georgakis MK**, Karalexi MA, Agius D, Antunes L, ... , Petridou ET. Incidence and time trends of childhood lymphomas: findings from 14 Southern and Eastern European cancer registries and the Surveillance, Epidemiology and End Results, USA. ***Cancer Causes Control.*** 2016 Nov;27(11):1381-1394. ***[IF: 2.7]***
- Tousoulis D*, **Georgakis MK*,** Oikonomou E, Papageorgiou N, … , Siasos G. Asymmetric Dimethylarginine: Clinical Significance and Novel Therapeutic Approaches. ***Curr Med Chem*.** 2015;22(24):2871-901. *(*equally contributed*) ***[IF: 3.5]***
- **Georgakis MK**, Anagnostopoulou S. Epileptogenesis after central nervous system infections. ***Encephalos*.** 2014; 51, 81-93.

Presentations, Posters and Invited Talks in Scientific Meetings/ Conferences

- **Georgakis MK.** Prediction of Drug Responses by Mendelian Randomization. ESSW 2019: 5th European Stroke Science Workshop. Garmisch-Partenkirchen, Germany. November 2019 [Invited Talk].
- **Georgakis MK,** Malik R, … Dichgans M. Interleukin-6 signaling effects on ischemic stroke and other cardiovascular outcomes: a Mendelian Randomization study. *ISGC Fall 2019: Investigators Meeting of the International Stroke Genetics Consortium.* St. Louis, MO, USA. October 2019 [Oral Presentation].
- **Georgakis MK,** Malik R, … Dichgans M. Circulating Monocyte Chemoattractant Protein-1 (MCP-1) and Risk of Incident Stroke: Meta-analysis of Population-Based Studies. *ISGC Fall 2019: Investigators Meeting of the International Stroke Genetics Consortium.* St. Louis, MO, USA. October 2019 [Oral Presentation].
- **Georgakis MK,** Duering M, Wardlaw JM, Dichgans M. Leukoaraiosis in association with long-term outcomes after ischemic stroke: a systematic review and meta-analysis. *ESOC 2019: European Stroke Organization Conference,* Milan, Italy, May 2019 [Oral Presentation].
- **Georgakis MK,** Gill D, … , Malik R, Dichgans M. Circulating Monocyte Chemoattractant Protein-1 as a Novel Risk Factor of Stroke. *ESOC 2019: European Stroke Organization Conference*, Milan, Italy, May 2019 [Poster].
- **Georgakis MK,** Gill D, Evangelou E, … , Tzoulaki I, Dichgans M. Genetically determined blood pressure, blood pressure lowering drugs, and risk of stroke and stroke subtypes: a Mendelian Randomization study. *ISGC Spring 2019: Investigators Meeting of the International Stroke Genetics Consortium.* Cambridge, UK, April 2019 [Oral Presentation].
- **Georgakis MK.** Monocyte chemoattractant protein-1 and risk of stroke: a Mendelian Randomization study followed by validation in a population-based cohort. *XXVII: Symposium "Forschung in der Neurologie"* (Organized by the Neurology Department of the University Hospital of LMU Munich). Munich, Germany, November 2018 [Invited Talk].
- **Georgakis MK,** Zietemann V, ... , Bordet R, Dichgans M. Early post-admission Montreal Cognitive Assessment predicts long-term cognitive and functional outcome and mortality after stroke Neurology. *VasCog 2018: The 9th International Conference of The*

International Society of Vascular Behavioural and Cognitive Disorders. Hong Kong, China, November 2018 [Poster].

- **Georgakis MK,** Xilouri M, Stefanis L. Alpha-synuclein-independent dopaminergic neurodegeneration following in vivo inhibition of chaperone-mediated autophagy in mice. *20 YEARS of alpha-synuclein in Parkinson's disease & related synucleionopathies*. Athens, Greece, September 2017 [Poster].
- **Georgakis MK.** Methodology of Systematic Reviews and Meta-analyses. *Annual Summer Retreat of the Department of Women's and Children's Health of the Uppsala University Hospital.* Spetses, Greece, September 2016 [Invited Speaker].
- **Georgakis MK,** Kalogirou EI, …, NARECHEM-BT Working Group, Petridou ET. Anthropometric measurements at birth and risk of primary central nervous system tumors: a systematic review and meta-analysis. *28th Annual Conference of the Greek Society for Social Pediatrics and Health Promotion.* Trikala, Greece, October 2016 [Oral presentation].
- **Georgakis MK,** Kalogirou EI, Liaskas A, …, NARECHEM-BT Working Group, Petridou ET. Anthropometric measurements at birth and risk of primary central nervous system tumors: a systematic review and meta-analysis. *Children with Cancer.* London, UK, September 2016 [Oral presentation].
- **Georgakis MK,** Karalexi MA, … , SEE Working Group, NARECHEM-BT Working Group, Petridou ET. Childhood pilocytic astrocytomas in Southern-Eastern Europe and US: incidence, survival and outcome discrepancies. *European Network of Cancer Registries (ENCR) Scientific meeting and General Assembly.* Baveno, Italy, October 2016 [Poster].
- **Georgakis MK,** Petridou ET. Genetics of temporal lobe epilepsy. *27th Annual Conference of the Greek Society for Social Pediatrics and Health Promotion.* Monemvasia-Sparti, Greece, October 2016 [Poster].

EIDESSTATTLICHE VERSICHERUNG/AFFIDAVIT

Hiermit versichere ich an Eides statt, dass ich die vorliegende Dissertation „**Using Genetics to Explore Novel Risk Factors and Drug Targets for Cerebrovascular Disease“** selbstständig angefertigt habe, mich außer der angegebenen keiner weiteren Hilfsmittel bedient und alle Erkenntnisse, die aus dem Schrifttum ganz oder annähernd übernommen sind, als solche kenntlich gemacht und nach ihrer Herkunft unter Bezeichnung der Fundstelle einzeln nachgewiesen habe.

I hereby confirm that the dissertation “**Using Genetics to Explore Novel Risk Factors and Drug Targets for Cerebrovascular Disease”** is the result of my own work and that I have only used sources or materials listed and specified in the dissertation.

München, den 21.11.2019

Marios K. Georgakis

DECLARATION OF AUTHOR CONTRIBUTIONS

The authors contributed to the manuscript as follows:

(1) **Georgakis MK,** Gill D, Rannikmäe K, Traylor M, Anderson CD, Lee JM, Kamatani Y, Hopewell JC, Worrall BB, Bernhagen J, Sudlow CLM, Malik R*, Dichgans M*. Genetically Determined Levels of Circulating Cytokines and Risk of Stroke. ***Circulation***. 2019 Jan 8;139(2):256-268. * equally contributed

MKG, RM, and MD conceptualized and designed the study. MKG and RM performed the statistical analyses. All authors interpreted the results. MKG and MD drafted the manuscript. All authors critically revised the manuscript for intellectual content. All authors approved the submitted version and are accountable for the integrity of the work.

(2) **Georgakis MK,** Malik R, Björkbacka H, Pana TA, Demissie S, Ayers C, Elhadad MA, Fornage M, Beiser AS, Benjamin EJ, Boekholdt SM, Engström G, Herder C, Hoogeveen RC, Koenig W, Melander O, Orho-Melander M, Schiopu A, Söderholm M, Wareham N, Ballantyne CM, Peters A, Seshadri S, Myint PK, Nilsson J, de Lemos JA, Dichgans M. Circulating Monocyte Chemoattractant Protein-1 and Risk of Stroke: Meta-Analysis of Population-Based Studies Involving 17 180 Individuals. ***Circ Res***. 2019 Sep 27;125(8):773-782.

MKG and MD conceptualized and designed the study. MKG performed the systematic review and the meta-analyses of the pooled data that were received from the individual studies. HB, TAP, SD, CA, MAE, ASB, EJB, SMB, GE, CH, RCH, WK, OM, MOM, AS, MS, NW, CMB, AP, SS, PKM, JN, and JAdL performed the statistical analyses of the individual studies and provided the summary data. All authors interpreted the results. MKG and MD drafted the manuscript. All authors critically revised the manuscript for intellectual content. All authors approved the submitted version and are accountable for the integrity of the work.

(3) **Georgakis MK,** Malik R, Gill D, Franceschini N, Sudlow CLM, INVENT Consortium, CHARGE Inflammation Working Group, Dichgans M. Interleukin-6 signaling effects on

ischemic stroke and other cardiovascular outcomes: a Mendelian Randomization study. ***MedRxiv*.** [Preprint – In press in ***Circ Gen Prec Med***]

MKG, RM, and MD conceptualized and designed the study. MKG performed the statistical analyses. All authors interpreted the results. MKG and MD drafted the manuscript. All authors critically revised the manuscript for intellectual content. All authors approved the submitted version and are accountable for the integrity of the work.

(4) Gill D*, **Georgakis MK*,** Koskeridis F, Jiang L, Feng Q, Wei WQ, Theodoratou E, Elliott P, Denny JC, Malik R, Evangelou E, Dehghan A, Dichgans M†, Tzoulaki I†. Use of Genetic Variants Related to Antihypertensive Drugs to Inform on Efficacy and Side Effects. ***Circulation***. 2019 Jul 23;140(4):270-279. *,† equally contributed

DG and MKG conceptualized the study. DG, IT, MKG and MD designed the study. DG, MKG, FK and LJ collectively had full access to the data and performed the analysis. All authors interpreted the results. DG and IT drafted the manuscript. All authors critically revised the manuscript for intellectual content. All authors approved the submitted version and are accountable for the integrity of the work.

(5) **Georgakis MK*,** Gill D*, Webb AJS, Evangelou E, Elliott P, Sudlow CLM, Dehghan A, Malik R, Tzoulaki I†, Dichgans D†. Genetically determined blood pressure, antihypertensive drug classes, and risk of stroke subtypes. [In press in ***Neurology***] *,† equally contributed

MKG, DG, RM, IT, and MD conceptualized and designed the study. MKG and DG performed the statistical analysis. All authors interpreted the results. MKG and MD drafted the manuscript. All authors critically revised the manuscript for intellectual content. All authors approved the submitted version and are accountable for the integrity of the work.

(6) **Georgakis MK,** Malik R, Anderson CD, Parhofer KG, Hopewell JC, Dichgans M. Genetically determined blood lipids and cerebral small vessel disease: role of HDL cholesterol. [Accepted] ***Brain***. 2020 Feb 1;143(2):597-610.

MKG, RM, and MD conceptualized the study. MKG, RM, CDA, JCH, and MD designed the study. MKG and RM performed the statistical analysis. All authors interpreted the results. MKG and

MD drafted the manuscript. All authors critically revised the manuscript for intellectual content. All authors approved the submitted version and are accountable for the integrity of the work.

Signatures:

Marios K. Georgakis	Martin Dichgans	Dipender Gill
PhD candidate	Supervisor	Shared 1st author