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Genome-wide meta-analysis identifies new susceptibility loci for migraine

A full list of authors and affiliations appears at the end of the article.

These authors contributed equally to this work.

Migraine is the most common brain disorder, affecting approximately 14% of the adult population, but its molecular mechanisms are poorly understood. We report the results of a meta-analysis across 29 genome-wide association studies, including a total of 23 285 migraine cases and 95 425 population-matched controls. We identified 12 loci associated with migraine susceptibility ($P < 5 \times 10^{-8}$). Five loci are new (near *AJAP1* on 1p36, near *TSPAN2* on 1p13, within *FHL5* on 6q16, within *c7orf10* on 7p14, and near *MMP16* on 8q21). Three of these loci were identified in disease subgroup analyses. Brain tissue eQTL analysis suggests potential functional candidate genes at four loci: *APOA1BP*, *TBC1D7*, *FUT9*, *STAT6*, and *ATP5B*.

Recently, significant progress has been made in the identification of common genetic variants associated with migraine susceptibility through genome-wide association (GWA) studies of clinic-based migraine with aura (MA) patients¹, migraineurs from the general population^{2,3}, and clinic-based migraine without aura (MO) patients⁴. To further elucidate the genetic susceptibility of migraine, we performed a meta-analysis of 23 285 individuals with migraine from 29 clinic- and population-based studies (Fig. 1, Supplementary Fig. 1, Supplementary Note). These include 5 175 cases from five clinic-based patient collections,

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Corresponding authors: Aarno Palotie (ap8@sanger.ac.uk), Verner Anttila (anttila@atgu.mgh.harvard.edu).

⁵⁵Details appear in the Supplementary Note.

Author Contributions

Jointly supervised research: A.Palotie, D.C., D.R.N., M.Daly, K.S., G.D.S., M.W., M.Dichgans, A.M., M.Ferrari, C.K., T.K., D.S., L.C., J.A.Z., M.R.J., C.D., D.I.B., J.K., L.Q., G.T.

Conceived and designed the study: A.Palotie, D.C., D.R.N., V.Anttila, B.S.W., M.Daly, M.W., M.Dichgans, A.M., C.K., L.C., J.A.Z., M.R.J., C.D., D.I.B., J.K., T.K., M.Kallela, R.M., B.V., G.T., L.Q., M.A.I., L.L., E.H., M.S., H.S., K.S., F.J., T.F., B.M.M.

Performed the experiments: V.Anttila, B.S.W., A.Palotie, D.R.N., G.D.S., M.W., M.Dichgans, A.M., C.K., J.A.Z., M.R.J., O.R., C.D., D.I.B., J.K., E.B., M.Kallela, B.V., G.T., E.H., T.F., R.R.F., N.G.M., A.G.U., T.M., J.G.E.

Performed statistical analysis: V.Anttila, B.S.W., P.G., D.C., D.R.N., M.Daly, D.S., E.B., J.R.G., S.B.R.J., T.K., F.B., G.M., R.M., B.V., L.Q., M.A.I., L.L., I.D., P.P., M.S., S.Steinberg, T.F., B.M.M.

Analysed the data: V.Anttila, B.S.W., P.G., A.Palotie, D.C., D.R.N., L.C., J.R.G., S.B.R.J., K.L., T.K., F.B., G.M., R.M., B.V., S.E.M., L.Q., M.A.I., L.L., J.W., P.P., M.S., S.Steinberg, H.S., T.F., N.A., B.M.M., D.T.

Contributed reagents/materials/analysis tools: D.C., D.R.N., M.Daly, A.Palotie, G.D.S., M.W., M.Dichgans, A.M., M.Ferrari, C.K., D.S., L.C., J.A.Z., M.R.J., O.R., C.D., D.I.B., B.W.P., J.K., E.B., J.R.G., K.L., E.R., V.Anttila, B.S.W., P.G., T.K., F.B., G.M., M.Kallela, R.M., B.V., G.T., U.T., W.L.M., L.Q., M.Koiranen, M.A.I., T.L., A.H.S., L.L., I.D., B.N., M.S., L.M.R., J.E.B., P.M.R., S.Steinberg, H.S., F.J., D.A.L., D.M.E., S.M.R., M.Färkkilä, V.Arto, M.A.K., T.F., J.S., R.R.F., N.P., C.M.W., R.Z., A.C.H., P.A.F.M., G.W.M., N.G.M., G.B., H.G., A.Heinze, K.H.K., F.M.K.W., A.L.H., A.Pouta, J.E., A.G.U., A.Hofman, J.J.H., J.M.V., K.H., M.A., B.M.M., S.Schreiber, T.M., E.W., A.A., J.G.E., B.T., D.T.

Wrote the paper: V.Anttila, B.S.W., A.Palotie, D.C., D.R.N., A.M., C.K.

All authors contributed to the final version of the manuscript.

Competing Financial Interests

The authors declare no competing financial interests

matched to 13 972 population-based controls (Supplementary Table 1), as well as 18 110 cases from 14 population-based studies and 81 453 migraine-free or control individuals from the same studies (Supplementary Table 2). Results from GWA studies of the five clinic-based collections^{1,4} and four of the population-based collections^{2,3} have been previously reported (Supplementary Fig. 2).

In addition to the primary meta-analysis using all available genotype data, three subgroup analyses were performed in those cohorts where sufficient additional clinical information was available (Supplementary Table 3). The first two subgroups consisted of migraine cases fulfilling the International Headache Society diagnostic criteria⁵ for either MA or MO. The third subgroup included only the clinic-based samples, under the hypothesis that they represent a group of migraineurs more enriched for severe migraines than cases identified from the general population.

Results from the primary meta-analysis and the three subgroups identify 142 single nucleotide polymorphisms (SNPs), at a total of 12 loci, to be significantly associated with migraine susceptibility (Table 1, Supplementary Fig. 3,4). Eight of those loci contain SNPs that lie within a known transcript. In addition, 1 168 SNPs at 134 loci (Supplementary Table 4) showed suggestive association to migraine, again combining the primary analysis and the three subgroup analyses. The single most significant *P* value overall was observed for rs11172113 in the primary analysis (*P* value 2.69×10^{-19} ; Fig. 2a, Table 1) at the *LRPI* locus on 12q13.

Five of the 12 genome-wide significant loci are new (near *AJAPI*, near *TSPAN2*, *FHL5*, *c7orf10*, and near *MMP16*), while seven confirm previously reported migraine loci (*PRDM16*², *MEF2D*⁴, *TRPM8*^{1,2}, near *TGFBR2*⁴, *PHACTR1*⁴, *ASTN2*⁴, and *LRPI*²). All seven previously reported loci seen in this study remained significant (all $p < 6.25 \times 10^{-3}$, correcting for eight previously reported loci) in analyses that exclude samples used in previous reports^{1,2,4} (Supplementary Fig. 5 and Supplementary Table 5). Among the newly identified loci, two contain SNPs with significant association that are located within known transcripts. On 6q16, *FHL5* encodes a transcription factor that regulates cAMP responsive elements *CREM* and *CREB*⁶, which play a role in synaptic plasticity⁷ and memory formation⁸. The locus also overlaps KIAA0776, which encodes a hypothetical protein also known as UFL1. On 7p14, mutations in *c7orf10* have been found in phenotypically mild or even clinically asymptomatic forms of glutaric aciduria type III⁹, a rare metabolic abnormality leading to persistent excretion of glutaric acid.

The novel loci on 1p36, 1p13, and 8q21 are located outside known transcripts. On 1p36, rs10915437 is located approximately 500 kb telomeric from *AJAPI* and approximately 300kb centromeric from a gene cluster encoding the apoptosis-related proteins DFFB and TP73 as well as centrosomal protein CEP104. *AJAPI* is expressed in brain (Supplementary Fig. 6) and has been associated with tumor invasion and regulation of metalloproteinase activity¹⁰. On 1p13, rs12134493 is located 87 kb 5' of *TSPAN2*, a member of the tetraspanin family, encoding a cell surface protein that mediates signal transduction events involved in the regulation of cell development, activation, growth and motility¹¹. *TSPAN2* has further been shown to act as a regulator of metalloproteinase activity¹¹. On 8q21, rs10504861 is

located 200 kb telomeric from matrix metalloproteinase *MMP16*. Members of the metalloproteinase family are widely expressed in human tissues and are involved in the breakdown of extracellular matrix in normal physiological processes. Notably, the protein encoded by *MMP16* (MT-MMP2) cleaves LRP1¹², encoded by a previously reported migraine gene². In addition, *MMP16* has recently been shown to be involved in basal NgR1 (Nogo-66 receptor) shedding in cortical neurons, thereby increasing axonal and synaptic plasticity¹³.

Four of the twelve loci (near *AJAPI*, near *TGFBR2*, *PHACTR1*, near *MMP16*), including two of the novel associations, were identified exclusively in the subgroup analyses (Table 1 and Supplementary Table 4). Two of the loci (rs9349379 in *PHACTR1* on 6p21, and rs10504861 near *MMP16* on 8q21) reached genome-wide significance only in MO, while no SNPs reached genome-wide significance in MA (Fig. 2c, d). The lowest *P* value in MA was with SNP rs7015657 ($p = 7.88 \times 10^{-8}$), which is located approximately 582 kb 3' from *GFRA2*, a member of the glial cell line-derived neurotrophic factor family.

A similar subgroup analysis was performed in only those samples that originate from specialized migraine clinics. Two loci with suggestive association in the primary analysis, rs6790925 (near *TGFBR2*) and rs6478241 (*ASTN2*), reached genome-wide significance in the clinic-based subgroup (Table 1 and Supplementary Tables 6,7). All of the 12 genome-wide significant loci associated with migraine had larger estimated effect sizes in the clinic-based subgroup compared to the primary (All) analysis (Supplementary Table 6, 7). A two-tailed binomial test shows the chance of observing larger effects at all 12 loci is significantly different from that expected by chance ($P = 4.88 \times 10^{-4}$). Among all reported loci ($P < 1 \times 10^{-5}$), only the clinic-based group showed a number of associated SNPs with higher effect sizes ($OR > 1.2$) at low frequency ($MAF < 0.05$; Supplementary Fig. 7). Thus, clinic-based migraine samples may represent a promising subgroup to help prioritise loci in the search for low frequency variants with moderate effects. Overall, among the 146 loci identified, twice as many have causative minor alleles over protective ones (with the ratio increasing towards the lower minor allele frequencies).

To explore the biological context for the identified loci we examined the properties of the most proximal genes to the 12 genome-wide significant top SNPs (Table 1). In expression data from 55 269 samples profiled using the Affymetrix HG-U133 Plus 2.0 microarray (including 1 990 brain and 384 endothelial samples), 11 of the 12 genes nearest to the identified loci (all except *FHL5*) were at least moderately (>20% of samples of the tissue showing a normalized log₂ expression value greater than 6; see Methods) expressed in disease-relevant brain regions (Supplementary Fig. 6). In contrast, only *TGFBR2* and *MEF2D* show moderate or greater expression in the endothelial samples. Possibly reflecting known co-morbidity between migraine and cardiovascular disease¹⁴, two of the 12 most proximal genes (*TGFBR2* and *PHACTR1*) have also been associated with cardiovascular traits: *TGFBR2* mutations have been reported to cause monogenic Marfan's syndrome¹⁵ and to be involved in abdominal aortic aneurysms¹⁶, while *PHACTR1* is associated with early onset myocardial infarction¹⁷. *TSPAN2*¹⁸, *MEF2D*¹⁹, *TRPM8*²⁰, *TGFBR2*²¹, *PHACTR1*²², *MMP16*²³, *ASTN2*²⁴, and *LRP1*²⁵ have been suggested to have functions in synaptic

formation or regulation; *PRDM16* has been linked to oxidative stress response, and *AJAPI* in maintaining tissue borders (Supplementary Fig. 8).

To identify possible non-proximity based genes underlying these associations, we examined eQTL data among 394 samples of brain tissue from the North American Brain Expression Consortium and the UK Brain Expression Consortium. Among the 12 regions with significant association to migraine, four were found to contain significant eQTLs (see Methods) among the SNP-probe pairs within the tested brain samples (four in the frontal cortex, one in the cerebellum; Table 2). On chromosome 1, rs12136718 is an eQTL for *APOA1BP*, rather than the gene closest to it, *MEF2D*. *APOA1BP* is widely expressed and is potentially linked with cholesterol efflux from cells²⁶. On chromosome 6, rs35128104 is an eQTL for *FUT9*, encoding α 1,3-fucosyl transferase IX. The FUT9 enzyme synthesizes the Lewis X (Le^x) carbohydrate structure, which has been implicated in neurite outgrowth in several types of brain neuronal cells²⁷⁻²⁹. At the chromosome 12 locus, two different eQTLs in brain tissue were found within the peak, for *STAT6* (rs4559) and for *ATP5B* (rs113953523), the former at a very robust *P*-value (2.16×10^{-22}). The *STAT* genes are known for transducing activation signals to transcription factors in macrophages³⁰, and *STAT6* phosphorylation has recently been shown to sense oxidative stress in astrocytes resulting in prostaglandin release³¹. A second eQTL gene at the same locus, *ATP5B*, is the β subunit of the mitochondrial ATP synthase, but a potential specific role in neuronal or vascular cells is not known. Finally, on chromosome 6, rs9349379 is an eQTL in cerebellar tissue for *TBC1D7*, that potentially down regulates the tuberous sclerosis gene, *TSC1*, through a positive regulation of the mTOR-signalling pathway³². In the central nervous system, *TSC1/2* signalling contributes to neural connectivity via its multi-faceted roles³³. Based on the available data it is not possible to decide whether a gene identified by the eQTL analysis or the gene closest to the strongest positional association is the most relevant gene contributing to migraine pathogenesis.

In a hypersensitivity site analysis, associated SNPs in the migraine loci were found to occur significantly more often in DNase I hypersensitivity sites in a number of tissues (Supplementary Fig. 9). This suggests that the loci associated with migraine are enriched for actively transcribed regions, supporting a regulatory role for the variants. Both neuronal and vascular tissue types carry an enriched set of sites within the detected loci. In addition, querying the RegulomeDB (see URLs) showed that several of the associated SNPs were found to overlap directly with known transcription factor binding motifs (Supplementary Table 8).

The number of significantly associated loci is still modest to form a broad understanding of the disease susceptibility, and any proposed functional hypothesis from the identified loci must thus be taken with caution. Some functional hypotheses could be inferred from the results of this study, as the majority of the identified loci harbour genes that can be linked to neuronal function.

The eQTL analysis further supported that regulatory effects in brain tissue may underlie several of the association signals. However, the lack of replication across brain regions

suggests that our ability to use the eQTL data to pinpoint the functionally most significant gene within the locus is limited.

The observed difference between the number of significant loci in the MO and MA groups (6 vs 0, respectively; Fig. 2c, d) despite reasonably similar sample sizes was somewhat unexpected. MA has been shown to have a considerably higher heritability estimate and sibling recurrence risk than MO (3.8 vs 1.9), and has thus been considered to be the more heritable of the two common migraine types³⁴. One possible explanation could be that the genetic susceptibility to MA is mediated more by rare variants with larger effect sizes, although this remains speculative. Another explanation for the difference may be a higher degree of heterogeneity among the MA cases (due to genetically distinct subgroups, for example). No common variants specifically predisposing to aura were identified by this study (the lowest observed P-value was $p = 7.88 \times 10^{-8}$).

In summary, we conducted a large migraine meta-analysis and identified 12 loci associated with migraine susceptibility, including five loci not previously associated with migraine, as well as 134 additional suggestive loci. An eQTL analysis of brain tissue highlighted a further five genes potentially implicated in migraine susceptibility. Two of the 12 loci were observed only in the clinic-based sample group, possibly suggesting more specificity to severe migraine headache, and two only in the MO group. Seven previously reported loci for migraine susceptibility were replicated in independent samples in this study. The difference in the number of identified loci and the strength of association suggest that the genetic background of MA is considerably less influenced by common variants than that of MO, contrary to previous expectations. Finally, while pathway analysis of the 146 loci showed no concentration in any particular pathway or tissue, eight of the 12 identified loci are located in or immediately outside genes with known function in synaptic or neuronal regulation and several of them exert regulatory control on one another.

Methods

Overall study design

For this meta-analysis, we used SNP marker data from 23 285 cases and 95 425 controls of European descent from 29 studies, including five clinic-based studies compared to population-matched control samples with unknown migraine status, as well as 14 entirely population-based cohorts. Four of the population-based cohorts (the B58C, NFBC, Young Finns and FinnTwin; see Supplementary Note for further details) were birth cohorts. The datasets for the meta-analysis included previously genotyped genome-wide association (GWA) study data from migraine-specific studies by the International Headache Genetics Consortium (see URLs) studies and the Women's Genome Health Study, as well as a number of pre-existing population-based GWA studies (for complete list of references, see Supplementary Table 2). Local research ethics committees approved the individual studies, and informed consent was obtained from all participants when necessary (see the Supplementary Materials for full details of ethics and consent procedures for each study). Additional details on sample recruitment and phenotypes and summary details for each collection are given in the Supplementary Note and in Supplementary Tables 1 and 2. Genome-wide SNP genotyping was performed independently in each cohort with the use of

various standard genotyping technologies, and imputed for each study with reference to HapMap release 21 or 22 CEU phased genotypes³⁵.

Study phenotypes

The primary phenotype analysed was migraine of any type, regardless of source. This was followed by a subgroup analysis consisting of 1) analysing only the clinical samples, 2) only samples satisfying criteria for MA, and 3) for MO. Population-based samples were not analysed genome-wide as a subgroup, due to forming 78% of cases and 85% of controls in the main analysis, but associations were calculated for the significant SNPs for comparative purposes. In the clinical cohorts, headache specialist has assigned a migraine diagnosis based on direct or telephone interview or through the use of an extensive questionnaire. For the population studies, migraine status for individuals in a study sample has been determined by a questionnaire (see Supplementary Note).

Statistical analysis of GWA study data

Each study contributed summary statistic data from an association analysis performed using a frequentist additive model based on an expected allelic dosage model for SNP markers, adjusting for gender (using either SNPTEST or ProbABEL [see URLs]). SNPs were filtered on per-study level based on inclusion criteria of MAF>0.1% and imputation quality measures of $I_A > 0.6$ (IMPUTE 2³⁶) or $r^2 > 0.3$ (MACH³⁷). Four of the included studies contain novel genotyping (HUNT) or imputation (the Finnish, German and Dutch MA studies and HUNT). In the meta-analysis, combined association data for ~2.3 million imputed and genotyped autosomal SNPs were analysed in a fixed-effects model using GWAMA. At this stage, SNPs with a heterogeneity coefficient I^2 exceeding 75% or presence in only four or fewer studies were filtered out. In the meta-analysis, there was little evidence for population stratification at the study level (each genomic inflation factor $\lambda \leq 1.1$), though moderate inflation was observed at the meta-analysis level ($\lambda = 1.15$; Supplementary Fig. 4). For estimating genome-wide significance, we used the commonly accepted threshold of 5×10^{-8} for primary loci³⁸, and 1×10^{-5} for secondary loci, in accordance with the reporting threshold for the GWAS catalog³⁹. At secondary loci, to limit spurious associations, at least two SNPs were required to pass the significance threshold within a 50kb window. We also estimated the robustness of this threshold using the false discovery rate method of Benjamini & Hochberg⁴⁰, showing that the P value threshold corresponding to FDR<0.05 was 2.33×10^{-5} . The quantile-quantile plot (Supplementary Fig. 4) of the meta-analysis P values showed a marked excess of association signals well beyond those expected by chance below the suggestive reporting threshold. The significant loci were visualized using the LocusZoom interface⁴¹. For the heterogeneity analyses for migraine type, due to shared controls in some of the sets, the available study samples were divided into as equally-sized groups in terms of effective study size as possible, and then the data was analysed using the gender heterogeneity analysis method⁴² (-sex option) of GWAMA⁴³, with a dummy variable coding for MA and MO instead of gender. For the heterogeneity analyses for gender, the same method was used to compare P values from males-only and females-only analyses.

Pathway analyses

MAGENTA - The MAGENTA software⁴⁴ was used to conduct an analysis to evaluate whether *P* values for association with migraine are enriched to particular biological networks, using pathway lists from GO, PANTHER, INGENUITY, KEGG, REACTOME and BIOCARTA. In the gene set enrichment analysis, *P* values were estimated via 10,000 permutations of genes evaluated at 75th FDR percentile (due to assumption of high polygenicity), manually corrected to account for FDR across all pathway sets. **DAPPLE** - Using a refined database of high-confidence protein-protein interactions (InWeb^{45,46}) we used DAPPLE⁴⁷ to assess the amount of physical interactions connecting the genes within 50 kb of the 146 reported migraine loci, as well as an analysis of only the 16 proteins from the 12 genome-wide significant loci. Both direct and indirect (through 1st order common interaction partners) were measured and compared to a random expectation over 10,000 permutations, and the resulting network was plotted. **GRAIL** - The GRAIL web interface⁴⁸ was used to explore similarities in published PubMed articles (August 2012 freeze), using data from HapMap release 22 CEU and gene size correction set to on. From the GRAIL results, only genes with significant (*P* value < 0.05) are shown, and the list of similar genes was capped at genes within the top 200 highest ranks.

Overlap with DNase I hypersensitivity sites

The positions of SNPs from migraine-associated loci were overlapped with DNase I 'hotspot' regions from the ENCODE project that mark generalized chromatin accessibility mapped for each of 125 diverse cell lines and tissues⁴⁹. To assess the significance of overlap for the set of SNPs as a whole, 100 background sets of SNPs were chosen from the genome so that each migraine associated SNP was matched in each set by a SNP within the same decile for minor allele frequency, distance to the nearest transcription start site and GC content of the 100 base region surrounding the SNP. The background SNP sets were overlapped with the DNase I hotspots, and the enrichment for overlap with the migraine associated SNP set expressed as the Z score relative to the distribution of background SNP set overlaps on a per cell line basis. In addition migraine associated SNPs were analysed for other overlap with ENCODE data including transcription factor motifs using RegulomeDB (<http://regulome.stanford.edu>)⁵⁰.

Tissue-based gene expression analysis

For the tissue analysis, a microarray-based analysis of gene expression was performed on a dataset of 55 269 samples in the Gene Expression Omnibus (GEO) database that were measured on the Affymetrix U133 Plus 2.0 Array. Each sample in the raw expression data was first linearly transformed using a modified invariant set normalization method⁵¹ on a set of eighty control genes with stable expression on U133 Plus 2.0. The expression data was log₂ transformed to stabilize the variance and expression distribution. Finally, the data were quantile-normalized⁵² to match the expression distribution of each sample. Expression values for genes with multiple probe sets were calculated by taking the median value of all probe sets for that gene. Following normalization, a log₂ expression value of 4 is considered baseline and log₂ expression values greater than 6 are considered expressed. Sample annotations were curated based on GEO descriptions provided by depositors. To account for

variation in the number of samples representing each tissue in the dataset, expression of a gene was plotted to show the fraction of samples of a tissue that exceeds a log₂ expression value of 6, with higher fractions indicating more ubiquitous expression in the tissue in question.

eQTL analysis

Based on the meta-analysis results for association with migraine, 146 regions of interest were queried against the expression quantitative trait loci (eQTL) results from the North American Brain Expression and UK Brain Expression Consortium studies (GEO # GSE36192, dbGaP # phs000249). These eQTL results are based on Cerebellum and Frontal Cortex tissue mRNA expression levels from 394 human subjects (see Supplementary methods). Of the 146 associated regions 134 were represented in the eQTL analysis. Within these regions 831 mRNA expression traits and 222 668 *cis* SNP/expression trait pairs were considered in Cerebellum and 864 mRNA expression traits and 230 660 *cis* SNP/expression trait pairs were considered in within the Frontal Cortex. 45 SNPs within the migraine associated loci were found to have significant correlation (evaluated at FDR-corrected threshold; 0.0001668 for frontal cortex and 0.0002187 for cerebellum) with the expression of 12 mRNA transcripts in both the Cerebellum and the Frontal Cortex. A more detailed methods description can be found in Supplementary Materials. The extent of linkage disequilibrium between SNPs associated with migraine and the SNPs in the tested eQTL SNP-probe pairs was evaluated using SNAP (see [URLs](#)).

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Authors

Verner Anttila^{#1,2,3,4}, Bendik S. Winsvold^{#1,5}, Padhraig Gormley¹, Tobias Kurth^{6,7,8}, Francesco Bettella⁹, George McMahon¹⁰, Mikko Kallela¹¹, Rainer Malik¹², Boukje de Vries¹³, Gisela Terwindt¹⁴, Sarah E. Medland¹⁵, Unda Todt¹⁶, Wendy L. McArdle¹⁰, Lydia Quaye¹⁷, Markku Koironen¹⁸, M. Arfan Ikram^{19,20,21}, Terho Lehtimäki²², Anine H. Stam¹⁴, Lannie Ligthart^{23,24}, Juho Wedenoja²⁵, Ian Dunham²⁶, Benjamin M. Neale^{3,4}, Preet Palta^{1,2}, Eija Hamalainen^{1,2}, Markus Schürks²⁷, Lynda M Rose⁸, Julie E. Buring⁸, Paul M. Ridker^{8,28}, Stacy Steinberg⁹, Hreinn Stefansson⁹, Finnbog Jakobsson^{xxx}, Debbie A. Lawlor¹⁰, David M. Evans¹⁰, Susan M. Ring¹⁰, Markus Färkkilä¹¹, Ville Artto¹¹, Mari A Kaunisto^{2,29},

URLs.

DAPPLE - www.broadinstitute.org/mpg/dapple
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 IMPUTE2 - mathgen.stats.ox.ac.uk/impute
 International Headache Genetics Consortium- www.headachegenetics.org
 MAGENTA - www.broadinstitute.org/mpg/magenta
 ProbABEL - <http://www.genabel.org/packages/ProbABEL>
 RegulomeDB - <http://regulome.stanford.edu>
 SNAP - <http://www.broadinstitute.org/mpg/snap>
 SNPTTEST - https://mathgen.stats.ox.ac.uk/genetics_software/snptest

Tobias Freilinger^{12,30}, Jean Schoenen³¹, Rune R. Frants¹³, Nadine Pelzer¹⁴, Claudia M. Weller¹³, Ronald Zielman¹⁴, Andrew C. Heath³², Pamela A.F. Madden³², Grant W. Montgomery¹⁵, Nicholas G. Martin¹⁵, Guntram Borck¹⁶, Hartmut Göbel³³, Axel Heinze³³, Katja Heinze-Kuhn³³, Frances M.K. Williams¹⁷, Anna-Liisa Hartikainen³⁴, Anneli Pouta^{18,34,35}, Joyce van den Ende¹⁹, Andre G. Uitterlinden³⁶, Albert Hofman³⁷, Najaf Amin¹⁹, Jouke-Jan Hottenga²³, Jacqueline M. Vink²³, Kauko Heikkilä²⁵, Michael Alexander^{38,39}, Bertram Muller-Myhsok^{40,69}, Stefan Schreiber^{41,42}, Thomas Meitinger^{43,44}, Heinz Erich Wichmann^{45,46,47}, Arpo Aromaa⁴⁸, Johan G. Eriksson^{29,48,49,50,51}, Bryan Traynor⁵², Daniah Trabzuni^{53,54}, North American Brain Expression Consortium⁵⁵, UK Brain Expression Consortium⁵⁵, Elizabeth Rossin^{3,4,56}, Kasper Lage^{3,4,57,58,59}, Suzanne B.R. Jacobs⁴, J. Raphael Gibbs^{52,53}, Ewan Birney²⁶, Jaakko Kaprio^{2,60,25}, Brenda W. Penninx^{63,24,62,61}, Dorret I. Boomsma²³, Cornelia van Duijn¹⁹, Olli Raitakari^{64,65}, Marjo-Riitta Jarvelin^{66,18,67,35}, John-Anker Zwart⁵, Lynn Cherkas¹⁷, David P. Strachan⁶⁸, Christian Kubisch¹⁶, Michel D. Ferrari¹⁴, Arn M.J.M. van den Maagdenberg^{13,14}, Martin Dichgans^{12,69}, Maija Wessman^{2,29}, George Davey Smith¹⁰, Kari Stefansson^{9,xxx2}, Mark J. Daly^{3,4}, Dale R. Nyholt¹⁵, Daniel Chasman^{8,28}, and Aarno Palotie^{1,2,4} **for the International Headache Genetics Consortium⁵⁵**

Affiliations

¹Wellcome Trust Sanger Institute, Wellcome Trust Genome Campus, Cambridge, UK ²Institute for Molecular Medicine Finland (FIMM), University of Helsinki, Helsinki, Finland ³Analytical and Translational Genetics Unit, Department of Medicine, Massachusetts General Hospital and Harvard Medical School, Boston, MA, USA ⁴Program in Medical and Population Genetics, Broad Institute of MIT and Harvard, Cambridge, MA, USA ⁵Department of Neurology, Oslo University Hospital and University of Oslo, Oslo, Norway ⁶INSERM Unit 708 – Neuroepidemiology, F-33000 Bordeaux, France ⁷University of Bordeaux, F-33000 Bordeaux, France ⁸Division of Preventive Medicine, Department of Medicine, Brigham and Women's Hospital, Harvard Medical School, Boston, USA ⁹deCODE genetics, Reykjavik, Iceland ¹⁰MRC Centre for Causal Analyses in Translational Epidemiology, School of Social and Community Medicine, University of Bristol, Bristol, UK ¹¹Department of Neurology, Helsinki University Central Hospital, Helsinki, Finland ¹²Institute for Stroke and Dementia Research, Klinikum der Universität München, Ludwig-Maximilians-Universität, Munich, Germany ¹³Department of Human Genetics, Leiden University Medical Centre, Leiden, The Netherlands ¹⁴Department of Neurology, Leiden University Medical Centre, Leiden, The Netherlands ¹⁵Queensland Institute of Medical Research, Brisbane, Queensland, Australia ¹⁶Institute of Human Genetics, University of Ulm, Ulm, Germany ¹⁷Department of Twin Research and Genetic Epidemiology, King's College London, London, UK ¹⁸Institute of Health Sciences, University of Oulu, Oulu, Finland ¹⁹Genetic Epidemiology Unit, Department of Epidemiology, Erasmus University Medical Center, Rotterdam, the Netherlands ²⁰Department of Radiology Erasmus University Medical Centre, Rotterdam, The Netherlands ²¹Department of Neurology Erasmus

University Medical Centre, Rotterdam, The Netherlands ²²Department of Clinical Chemistry, Fimlab Laboratories and University of Tampere School of Medicine, Tampere, Finland ²³Department of Biological Psychology, VU University, Amsterdam, The Netherlands ²⁴EMGO+ Institute for Health and Care Research, VU University Medical Center, Amsterdam, The Netherlands ²⁵Department of Public Health, Hjelt Institute, University of Helsinki, Helsinki, Finland ²⁶European Bioinformatics Institute, Wellcome Trust Genome Campus, Cambridge, UK ²⁷Department of Neurology, University Hospital Essen, Essen, Germany ²⁸Harvard Medical School, Boston, MA 02215, USA ²⁹Folkhälsan Research Center, Helsinki, Finland ³⁰Department of Neurology, Klinikum der Universität München, Munich, Germany ³¹Headache Research Unit, Department of Neurology and Groupe Interdisciplinaire de Génoprotéomique Appliquée (GIGA)-Neurosciences, Liège University, Liège, Belgium ³²Department of Psychiatry, Washington University School of Medicine, St. Louis, Missouri, USA ³³Kiel Pain and Headache Center, Kiel, Germany ³⁴Department of Clinical Sciences/Obstetrics and Gynecology, University Hospital of Oulu, Oulu, Finland ³⁵Department of Children, Young People and Families, National Institute for Health and Welfare, Helsinki, Finland ³⁶Department of Internal Medicine, Erasmus Medical Center, Rotterdam, The Netherlands ³⁷Genetic Epidemiology Unit, Department of Clinical Genetics, Erasmus University Medical Centre, Rotterdam, The Netherlands ³⁸Department of Genomics, Life & Brain Center, University of Bonn, Bonn, Germany ³⁹Institute of Human Genetics, University of Bonn, Bonn, Germany ⁴⁰Max Planck Institute of Psychiatry, Munich, Germany ⁴¹Department of Clinical Molecular Biology, Christian Albrechts University, Kiel, Germany ⁴²Department of Internal Medicine I, Christian Albrechts University, Kiel, Germany ⁴³Institute of Human Genetics, Helmholtz Center Munich, Neuherberg, Germany ⁴⁴Institute of Human Genetics, Klinikum Rechts der Isar, Technische Universität München, Munich, Germany ⁴⁵Institut für Medizinische Informationsverarbeitung, Biometrie und Epidemiologie, Ludwig-Maximilians-Universität München, Munich, Germany ⁴⁶Institute of Epidemiology I, HelmholtzCenter Munich, Neuherberg, Germany ⁴⁷Klinikum Großhadern, Ludwig-Maximilians-Universität München, Munich, Germany ⁴⁸National Institute for Health and Welfare, Helsinki, Finland ⁴⁹Department of General Practice, Helsinki University Central Hospital, Helsinki, Finland ⁵⁰Vaasa Central Hospital, Vaasa, Finland ⁵¹Department of General Practice and Primary Health Care, University of Helsinki, Helsinki, Finland ⁵²Laboratory of Neurogenetics, National Institute on Aging, National Institutes of Health, Bethesda, MD, USA ⁵³Department of Molecular Neuroscience, Institute of Neurology, University College London, London, UK ⁵⁴Department of Genetics, King Faisal Specialist Hospital and Research Centre, Riyadh, Saudi Arabia ⁵⁵Center for Human Genetic Research, Massachusetts General Hospital, Boston, MA, USA ⁵⁶Pediatric Surgical Research Laboratories, Massachusetts General Hospital for Children, Massachusetts General Hospital, Boston, MA, USA ⁵⁷Center for Biological Sequence Analysis, Department of Systems Biology, Technical University of Denmark, Lyngby, Denmark ⁵⁸Center for Protein Research, University of Copenhagen, Copenhagen, Denmark ⁶⁰Department

of Mental Health and Alcohol Research, National Institute for Health and Welfare, Helsinki, Finland ⁶¹Department of Psychiatry, Leiden University Medical Center, Leiden, The Netherlands ⁶²Department of Psychiatry, University Medical Center Groningen, Groningen, The Netherlands ⁶³Department of Psychiatry, VU University Medical Center, Amsterdam, The Netherlands ⁶⁴Research Centre of Applied and Preventive Cardiovascular Medicine, University of Turku, Turku University Hospital, Turku, Finland ⁶⁵Department of Clinical Physiology and Nuclear Medicine, Turku University Hospital, Turku, Finland ⁶⁶Department of Epidemiology and Biostatistics, School of Public Health, MRC-HPA Centre for Environment and Health, Faculty of Medicine, Imperial College, London, UK ⁶⁷Biocenter Oulu, University of Oulu, Oulu, Finland ⁶⁸Division of Population Health Sciences and Education, St George's, University of London, London, UK ⁶⁹Munich Cluster for Systems Neurology (SyNergy), Munich, Germany ^{xxx}Department of Neurology, Landspítali University Hospital, Reykjavik, Iceland ^{xxx2}School of Medicine, University of Iceland, Reykjavik, Iceland

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All migraine (23 285 vs. 95 425)			Migraine with aura (5 118 vs. 74 239)			Migraine without aura (7 107 vs. 69 427)		
Study	Cases	Controls	Study	Cases	Controls	Study	Cases	Controls
ALSPAC	3 134	5 103	deCODE	120	34 617	deCODE	537	34 617
Australia	1 683	2 383	ERF	141	1 216	ERF	189	1 216
B58C	1 165	4 141	Finnish MA	1 032	3 513	German MO	1 208	2 564
deCODE	2 139	34 617	German MA	997	1 105	HUNT	1 175	1 097
ERF	330	1 216	HUNT	359	1 097	LUMINA MO	1 118	2 016
Finnish MA	1 032	3 513	LUMINA MA	820	4 774	NTR&NESDA	154	2 360
FinnTwin	189	580	NTR&NESDA	103	2 260	Rotterdam	275	1 647
German MA	997	1 105	Rotterdam	76	1 647	TWINS UK	468	3 837
German MO	1 208	2 564	TWINS UK	235	3 837	WGHS	1 826	18 108
HUNT	1 608	1 097	WGHS	1 177	18 108	Young Finns	157	2 065
LUMINA MA	820	4 774	Young Finns	58	2 065			
LUMINA MO	1 118	2 016						
NFBC	757	4 399						
NTR&NESDA	282	2 260						
Rotterdam	351	1 647						
TWINS UK	972	3 837						
WGHS	5 122	18 108						
Young Finns	378	2 065						

Clinic-based studies (5 175 vs. 13 972)		
Study	Cases	Controls
LUMINA MA	820	4 774
LUMINA MO	1 118	2 016
German MA	997	1 105
German MO	1 208	2 564
Finnish MA	1 032	3 513

Figure 1. Description of the studies comprising the International Migraine Genetics Meta-analysis Consortium, and their sample contributions to each analysis

Each coloured box corresponds to one analysed phenotype and lists the total number of cases and controls, as well as the sample contributions of individual cohorts. Participation in each analysis depended on the availability of the data in question and the recruitment method.

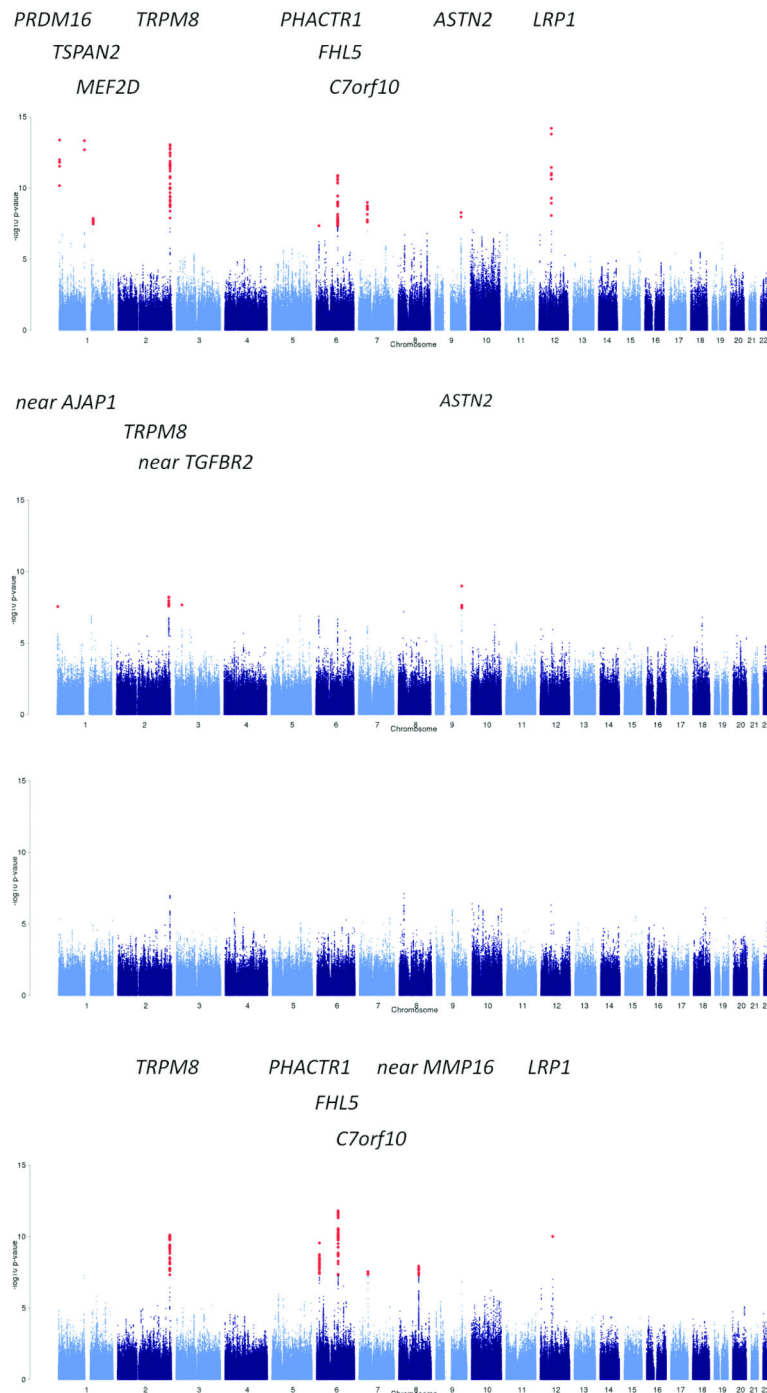


Figure 2. Manhattan plot of the results of the meta-analysis

Results of the meta-analysis of all migraine cases, of any migraine subtype or recruiting method, versus all available controls, adjusting for gender. Red dots indicate SNPs with significant ($< 5 \times 10^{-8}$) *P* values.

- A) All migraine (23 285 cases, 95 425 controls)
- B) Clinic-based studies only (5 175 cases, 13 972 controls)
- C) Migraine with aura (5 118 cases, 74 239 controls)

D) Migraine without aura (7 107 cases, 69 427 controls)

Table 1

Results of the association analyses

SNP	Chr	Position	Location	Gene	Minor allele	MAF	P-value	OR (95% CI)	q/p-value	I ²	Group or subgroup with the lowest p-value	Additional significance in
rs2651899	1	3,073,572	Genic	<i>PRDM16</i> *	C	0.41	3.28 × 10 ⁻¹⁴	1.09 (1.07-1.12)	0.214	20%	All samples	
rs10915437	1	4,082,866	Intergenic	<i>near AJAP1</i>	G	0.36	2.81 × 10 ⁻⁸	0.86 (0.82-0.91)	0.108	47%	Clinics only	
rs12134493	1	115,479,469	Intergenic	<i>near TSPAN2</i>	A	0.46	6.71 × 10 ⁻¹⁴	1.14 (1.10-1.18)	0.480	14%	All samples	
rs2274316	1	154,712,866	Genic	<i>MEF2D</i> *	C	0.37	3.14 × 10 ⁻⁸	1.07 (1.04-1.09)	0.021	45%	All samples	
rs7577262	2	234,483,608	Genic	<i>TRPM8</i> *	A	0.10	3.27 × 10 ⁻¹³	0.87 (0.84-0.90)	0.070	33%	All samples	Clinics only, MO only
rs6790925	3	30,455,089	Intergenic	<i>near TGFBR2</i> *	T	0.38	2.16 × 10 ⁻⁸	1.15 (1.10-1.21)	0.780	0%	Clinics only	
rs9349379	6	13,011,943	Genic	<i>PHACTR1</i> *	G	0.40	2.81 × 10 ⁻¹⁰	0.86 (0.82-0.90)	0.443	0%	MO only	All samples
rs13208321	6	96,967,075	Genic	<i>FHL5</i>	A	0.22	2.15 × 10 ⁻¹²	1.18 (1.13-1.24)	0.168	0%	All samples	MO only
rs4379368	7	40,432,725	Genic	<i>c7orf10</i>	T	0.12	1.46 × 10 ⁻⁹	1.11 (1.08-1.15)	0.441	2%	All samples	MO only
rs10504861	8	89,617,048	Intergenic	<i>near MMP16</i>	T	0.16	1.32 × 10 ⁻⁸	0.86 (0.81-0.90)	0.755	0%	MO only	
rs6478241	9	118,292,450	Genic	<i>ASTN2</i> *	A	0.38	1.04 × 10 ⁻⁹	1.16 (1.11-1.22)	0.646	0%	Clinics only	All samples
rs11172113	12	55,813,550	Genic	<i>LRP1</i> *	C	0.43	2.69 × 10 ⁻¹⁹	0.90 (0.88-0.92)	0.188	22%	All samples	MO only

When multiple subgroups show significant association, *P* values and odds ratios (OR) are shown for the analysis with the lowest *P* value. ORs reported for the minor allele. Chromosomal positions are based on NCBI build 36. In the Location and Gene columns, SNPs located within the gene transcript ('Genic') list that gene, while for intergenic SNPs ('Intergenic') the gene reported for that locus is listed.

* Previously identified migraine locus. MAF – minor allele frequency. q/p value - *P* value from Cochran's heterogeneity statistic. *I*² – heterogeneity index. MO – migraine without aura.

Table 2
Significant eQTLs observed in 394 samples of two different brain tissue at loci significantly associated with migraine

Chr	Position (bp)	Locus	Probe	Gene name	Frontal cortex				
					SNPs with sign. eQTL with probe	SNP with lowest eQTL p-value	Lowest eQTL p-value	Highest LD with reported SNP	SNPs with sign. eQTL with probe
1	154,830,352	<i>MEF2D</i>	ILMN_1729533	<i>APOA1BP</i>	1	rs12136718	2.18×10^{-5}	0.38	0
6	13,413,333	<i>PHACTR1</i>	ILMN_1661622	<i>TBC1D7</i>	0	-	-	-	1
6	96,769,539	<i>FHL5</i>	ILMN_1878007	<i>FUT9</i>	2	rs35128104	5.96×10^{-5}	0.74	0
12	55,775,568	<i>LRPI</i>	ILMN_1763198	<i>STAT6</i>	40	rs4559	2.16×10^{-22}	1	-
12	55,318,337	<i>LRPI</i>	ILMN_1772132	<i>ATP5B</i>	1	rs113953523	1.62×10^{-4}	0.39	0

-Chr - chromosome. All basepair positions are given for build 36/hg18. 'Highest LD...' column indicates the maximum extent of LD observed between a significant eQTL SNP-probe pair (see Methods), and a SNP passing the reporting threshold ($p < \times 10^{-5}$) for association with migraine. Due to multiple overlapping signals, the HLA region has been excluded from the analysis.