Impact of atopy on risk of glioma: A Mendelian randomization study

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ABSTRACT

BACKGROUND: An inverse relationship between allergies with glioma risk has been reported in several but not all epidemiological observational studies. We performed an analysis of genetic variants associated with atopy to assess the relationship with glioma risk using Mendelian randomization (MR), an approach unaffected by biases from temporal variability and reverse causation that might have affected earlier investigations.

METHODS: Two-sample MR was undertaken using genome-wide association study data. We used single nucleotide polymorphisms (SNPs) associated with atopic dermatitis, asthma and hay fever, IgE levels and self-reported allergy as instrumental variables. We calculated MR estimates for the odds ratio (OR) for each risk factor with glioma using SNP-glioma estimates from 12,488 cases and 18,169 controls, using inverse-variance weighted (IVW), maximum likelihood estimation (MLE), weighted median estimate (WME) and mode-based estimate (MBE) methods. Violation of MR assumptions due to directional pleiotropy were sought using MR-Egger regression and HEIDI-outlier analysis.

RESULTS: Under IVW, MLE, WME and MBE methods, associations between glioma risk with asthma and hay fever, self-reported allergy and IgE levels were non-significant. An inverse relationship between atopic dermatitis and glioma risk was found by IVW (OR=0.96, 95% confidence interval [CI]: 0.93-1.00, P=0.041) and MLE (OR=0.96, 95% CI=0.94-0.99, *P*=0.003) but not by WME (OR=0.96, 95% CI: 0.91-1.01, *P*=0.114) or MBE (OR=0.97, 95% CI: 0.92-1.02, *P*=0.194).

CONCLUSIONS: Our investigation does not provide strong evidence for relationship between atopy and the risk of developing glioma, but findings do not preclude a small effect in relation to atopic dematitis. Our analysis also serves to illustrate the value of using several MR methods to derive robust conclusions.

KEYWORDS: Mendelian randomisation; allergy; cancer; glioma; risk

BACKGROUND

Although glioma accounts for around 80% of malignant primary brain tumours [1], to date few aetiological risk factors are well established for the disease [2]. Over the past three decades the search for an immune-mediated risk factor that might influence risk has led to studies of a possible relationship between multiple allergic conditions and autoimmune disorders with glioma [3].

Several case-control studies have shown that self-reported allergic conditions may protect against glioma [4]. For example, in the International Adult Brain Tumour Study, based on 1,178 glioma patients, an odds ratio (OR) of 0.59 was found for any self-reported allergy [5]. Other case-control studies have reported similar ORs, however most have been reliant on substantial numbers of proxy informants (up to 44%) [4, 6], and potential bias as a consequence of how controls were ascertained, thereby casting doubt on findings. In contrast to case-control studies, evidence for an association between glioma and allergy from cohort-based analyses has been less forthcoming [7], although such studies have been poorly powered to demonstrate a relationship.

Assaying IgE potentially reduces bias stemming from self-reporting despite levels not necessarily corresponding to specific allergies or equating to a single allergic response. Nevertheless measurement of IgE has been explored by a number of researchers seeking to identify risk factors for glioma [8-10]. In a case-control study of 228 cases and 289 controls performed in 2004, self-reported allergies and IgE levels were both inversely associated with glioma, but concordance between the two outcomes was poor [8]. In a larger study of 535 cases and 532 controls, both self-reported allergies and IgE levels were inversely related to

glioma risk, however IgE levels in patients were affected by temozolomide treatment [11]. A case-control study nested within the European Prospective Investigation into Cancer and Nutrition cohort based on prospectively collected serum IgE levels reported a non-significant OR of 0.73 [9]. A similar nested case-control study performed in the USA based on 181 cases reported non-significant OR of 0.72 for high serum IgE [10].

Several mechanisms have been proposed to explain a possible association between atopic disease and glioma [12]. The findings could reflect a true causal effect of the heightened immune function reported for atopy on tumour development. Alternatively, the associations observed might be non-causal, arising as a consequence of methodological biases inherent in the study design. Imprecisely defined exposures such as allergic disease are likely to have affected the validity of the findings of both case-control and cohort studies. The heterogeneous description of allergy in studies and different levels of detail in self-reporting on individual allergies complicate interpretation of the results. Additional biases include possible selection bias in controls, recall bias from self-reported allergy assessment and reverse causation or confounding from unmeasured effects. Finally, the high frequency of exposure ascertainment by proxy for cases is also likely to have systematically biased findings.

Mendelian randomization (MR) analysis can be used to minimise potential biases in conventional observational studies and to determine the causal association of an exposure with an outcome, such as disease risk [13]. The causal association can also be manifested by common genetic and biological pathways that determine two sequentially developed phenotypes, such as an atopic trait and glioma risk. Atopy has a strong heritable basis [14, 15], and thus far genome-wide association studies (GWAS) have identified over 50 loci

associated with different atopy-related traits [16]. The alleles associated with atopy should be randomly assigned to offspring from parents during mitosis, a process analogous to the random assignment of subjects to an exposure of interest in randomised clinical trials. Thus, genetic scores summarising the effects of single nucleotide polymorphisms (SNPs) associated with atopy-related traits can serve as instrumental variables (IVs) in a MR analysis of atopy and glioma risk.

To examine the nature of the association between atopy and glioma, we implemented two-sample MR [17] to estimate associations between atopy-associated SNPs and glioma risk using summary data from the recent GWAS meta-analysis performed by the Glioma International Case-Control Consortium study (GICC) [18].

METHODS

Two-sample MR was undertaken using GWAS data. Ethical approval was not sought for this specific project because all data came from the summary statistics of published GWAS, and no individual-level data were used.

Glioma genotyping data

Glioma genotyping data were derived from the most recent meta-analysis of GWAS in glioma, which related >10 million genetic variants (after imputation) to glioma, in 12,488 glioma patients and 18,169 controls from eight independent GWAS datasets of individuals of European descent [18] (**Additional file 2: Table S1**). Comprehensive details of the genotyping and quality control of the seven GWAS have been previously reported [18].

Genetic variant instruments for atopic traits

SNPs associated with each of the atopy-related traits investigated – atopic dermatitis (eczema), asthma and hay fever, IgE level, and self-reported allergy by the NHGRI-EBI GWAS Catalog [19-26] at genome-wide significance (*i.e.* $P \leq 5.0 \times 10^{-8}$) in individuals with European ancestry were used as IVs. To avoid co-linearity between SNPs for each trait, we excluded SNPs that were correlated (*i.e.* r^2 value of ≥0.001) within each trait, and only considered the SNPs with the strongest effect on the trait for use as IVs (**Additional file 3: Table S2**). For each SNP, we recovered the chromosome position, risk allele, association estimates (*per*-allele log-OR) and standard errors (SEs), summarised in **Table 1**. The allele that was associated with increased risk of the exposure was considered the effect allele. For IgE level, the allele associated with an increase in serum IgE was considered the effect allele. Allele frequencies for these SNPs were compared between the atopy-related trait and glioma

datasets to ensure that the effect estimates were recorded with respect to the same allele. Gliomas are heterogeneous and different tumour subtypes, defined in part by malignancy grade (for example, pilocytic astrocytoma World Health Organization (WHO) grade I, diffuse 'low-grade' glioma WHO grade II, anaplastic glioma WHO grade III and glioblastoma [GBM] WHO grade IV) can be distinguished [27]. For the sake of brevity we considered gliomas as being either GBM or non-GBM.

Two-sample Mendelian randomization method

The association between each atopy-related trait and glioma was examined using MR on summary statistics using the inverse variance weighted (IVW) method and maximum likelihood estimation (MLE) as *per* Burgess *et al.* [28]. The IVW ratio estimate ($\hat{\beta}$) of all SNPs associated with each atopy-related trait on glioma risk was calculated as follows:

$$\hat{\beta} = \frac{\sum_k X_k Y_k \sigma_{Y_k}^{-2}}{\sum_k X_k^2 \sigma_{Y_k}^{-2}} \quad .$$

Where X_k corresponds to the association of SNP *k* (as log of the OR per risk allele) with the atopy-related trait *Y,* Y_k is the association between SNP *k* and glioma risk (as log OR) with standard error σ_{Y_k}. The estimate for ($\hat{\beta}$) represents the causal increase in the log odds of glioma for each trait. The standard error of the combined ratio estimate is given by:

$$\text{se}(\hat{\beta}) = \sqrt{\frac{1}{\sum_k X_k^2 \sigma_{Y_k}^{-2}}} \quad .$$

For the maximum likelihood estimate, a bivariate normal distribution for the genetic associations was assumed, and the R function *optim* was used to estimate β. se($\hat{\beta}$) was calculated using observed information. The correlation between the errors of Y_k and X_k was taken to be 0 as they were derived from independent studies.

A central tenet in MR is the absence of pleiotropy (*i.e.* a gene influencing multiple traits) between the SNPs influencing the exposure and outcome disease risk [13]. This would be revealed as deviation from a linear relationship between SNPs and their effect size for atopy and glioma risk. To examine for violation of the standard IV assumptions in our analysis we first performed MR-Egger regression, as well as HEIDI-outlier analysis as per Zhu, Z *et al*. [29] imposing the advocated threshold of *P* ≤0.01. Additionally we derived weighted median estimates (WME) [30] and mode-based estimates (MBE) [31] to establish the robustness of findings.

Atopic dermatitis, asthma and hay fever, and self-reported allergyand all of the disease outcomes (all glioma, GBM and non-GBM glioma) are binary The causal effect estimates therefore represent the odds for outcome disease risk per unit increase in the log OR of the exposure disease [32]. These ORs have been converted to represent the OR for the outcome disease per doubling in odds of the exposure disease to aid interpretation [32].

For each statistical test we considered a global significance level of *P* <0.05 as being satisfactory to derive conclusions. To assess the robustness of our conclusions, we initially imposed a conservative Bonferroni-corrected significance threshold of 0.0125 (*i.e*. 0.05/4 atopy-related traits). We considered a *P* value ≥0.05 as non-significant (*i.e*. no association), a *P* value <0.05 as evidence for a potential causal association, and a *P* <0.0125 as significant

evidence for an association. All statistical analyses were undertaken using R software (Version 3.1.2). The meta and gsmr packages were used to generate forest plots and perform HEIDI-outlier analysis [29].

The power of a MR investigation depends greatly on the proportion of variance in the risk factor that is explained by the IV. We estimated study power *a priori* using the methodology of Burgess *et al.* [33], making use of published estimates of the heritability of trait associated IV SNPs [34-36], as well as estimates found by direct calculation (**Additional file 4: Table S3**), and the reported effect of each trait on glioma risk reported in meta-analysis of epidemiological studies [18]. **Additional file 5: Table S4** shows the range of ORs, for which we had less than 80% power to detect for each of the four atopy-related traits.

Simulation model

Through simulation we evaluated the suitability of using each employed MR method in a two-sample setting with binary-exposure and binary-outcome data. For *i* index *N* genetic variants and *j* index individuals genetic variants g_{ij} were generated independently by sampling from a Binomial(2,p_j) distribution with probability p_j drawn from a Uniform(0.1,0.9) distribution, to mimic bi-allelic SNPs in Hardy-Weinberg equilibrium. Let w_j correspond to the per-allele OR for the exposure disease, sampled from ORs reported for genome-wide significant SNPs reported in the GWAS Catalog [37], and *v* be the OR for the outcome disease per doubling in odds of the exposure disease. For each individual, exposure disease odds x_j, outcome disease odds y_j, exposure disease status a_j, and outcome disease status b_j were determined as follows:

$$x_j = x_0 \prod_{i=1}^{N} {w_i}^{g_{ij}}$$

$$y_j = y_0 \times 2^{\log_2 x_j \times \log_2 v}$$

$$a_j \sim \text{Binomial}(1, \frac{x_j}{1 + x_j})$$

$$b_j \sim \text{Binomial}(1, \frac{y_j}{1 + y_j})$$

Data for 1,000,000 individuals were simulated and partitioned at random to reflect the two-sample setting. Cases and controls for the exposure and outcome GWAS were sampled from each half of the dataset using the exposure and outcome disease statuses of each individual, and association statistics computed under an additive logistic regression model. To ensure the simulated data closely resembled the atopy-related trait and glioma data, the simulation analysis was repeated for each binary atopy-related trait using the same number of genetic variants as IVs and the same numbers of case and control individuals as used to estimate the atopy-related trait and glioma association statistics (**Additional file 6: Table S5**). Parameters x_0=0.0005 and y_0=0.01 were chosen to ensure the prevalence of the simulated exposure and outcome diseases were similar to that of the atopy-related traits and glioma respectively (**Additional file 6: Table S5**). To determine the suitability of each MR method we considered two scenarios: (i) no causal relationship between exposure and outcome (v=1.00) and (ii) a causal relationship between exposure and outcome (v=1.33). We performed 100 simulations for each scenario for each binary atopy-related trait.

RESULTS

The atopic dermatitis risk SNP rs909341, which is highly correlated with the chromosome 20q13.33 glioma risk SNP rs2297440 (D'=0.89, r^2=0.77), was strongly associated with risk of glioma (P=2.10x10^{-34}). Testing for pleiotropy using HEIDI outlier-analysis formally identified rs909341 as violating the assumption of the instrument on the outcome. Henceforth we confined our analysis of the relationship between atopic dermatitis and glioma to a dataset excluding this SNP.

Figure 1 shows forest plots of ORs for glioma generated from the SNPs. There was minimal evidence of heterogeneity between variants for asthma and hay fever, atopic dermatitis, IgE levels and self-reported allergy (respective I^2 and P_{het} values being 28% and 0.192, 8% and 0.377, 0% and 0.444, and 0% and 0.707). Including rs909341 in the analysis for atopic dermatitis, the I^2 value was 90% and $P_{het} < 10^{-4}$ (**Additional file 1: Figure S1**), providing further evidence that inclusion of this SNP would invalidate the MR analysis.

The results of the IVW, MLE, WME, MBE and MR-Egger methods are summarised in **Table 2**. Using the IVW method to pool results from individual SNPs, no associations (*i.e*. $P \geq 0.05$) were identified between genetically conferred risk of raised IgE level (OR=0.88, 95% CI=0.69-1.13, P=0.319), asthma and hay fever (OR=0.96, 95% CI=0.90-1.03, P=0.248), or self-reported allergy (OR=1.03, 95% CI=0.95-1.11, P=0.534) with risk of all glioma. There was some support for an inverse relationship between atopic dermatitis and glioma risk (OR=0.96, 95% CI=0.93-1.00, P=0.041), albeit not significant after adjustment for multiple testing.

Using MLE, no associations were identified between asthma and hay fever (OR=0.96, 95% CI=0.93-1.00, *P*=0.066), IgE levels (OR=0.88, 95% CI=0.74-1.05, *P*=0.157) or self-reported allergy (OR=1.02, 95% CI=0.97-1.08, *P*=0.429) with risk of all glioma. For atopic dermatitis, an OR of 0.96 (95% CI=0.94-0.99, *P*=0.003) was shown, which remained significant after adjusting for multiple testing. **Figure 2** shows relaxation of the assumption that the correlation between the errors in X_k and Y_k is zero for each of the atopy-related traits demonstrating the consistency of findings. Specifically, for a correlation in the range -0.15 to 0.15 the association between atopic dermatitis and glioma risk remained significant.

In contrast to findings from IVW and MLE, no significant support was provided by either the WME or MBE for an association between any of the atopy-related traits and glioma risk, including atopic dermatitis (WME: OR=0.96, 95% CI=0.91-1.01, *P*=0.114; MBE: OR=0.97, 95% CI: 0.92-1.02, *P*=0.194; **Table 2**).

The respective effect estimated from MR-Egger regression (**Figure 3**) for atopic dermatitis, IgE, asthma and hay fever, and self-reported allergy were 0.97 for atopic dermatitis (95% CI=0.92-1.03; *P*=0.375) 0.63 for IgE levels (95% CI=0.32-1.25; *P*=0.184), 0.99 for asthma and hay fever (95% CI=0.72-1.36, *P*=0.951) and 0.92 for self-reported allergy (95% CI=0.69-1.22; *P*=0.540), with intercepts of -0.004 (95% CI=-0.014-0.006, *P*=0.396), 0.027 (95% CI=-0.001-0.053, *P*=0.042), -0.007 (95% CI=-0.030-0.016, *P*=0.542) and 0.017 (95% CI=0.003-0.031, *P*=0.018). Collectively these findings provide possible evidence of systematic bias in the IVW estimate for IgE level and self-reported allergy, which might have arisen through overall unbalanced horizontal pleiotropy. There was no such evidence for such pleiotropy in respect of atopic dermatitis.

We explored the possibility that a relationship between atopy and glioma might be subtype specific, considering GBM and non-GBM separately. Imposing a stronger significance threshold of *P*=0.00625 (0.05/8, to correct for testing four traits over two outcomes), no histology-specific associations were shown by the IVW method between asthma and hay fever, IgE levels and self-reported allergy and glioma risk, respective ORs for the IVW method being 0.97, 0.92, and 1.04 for GBM tumours, 0.96, 0.97, and 1.04 for non-GBM tumours (**Additional file 7: Table S6**). For atopic dermatitis, a significant OR of 0.94 (95% CI=0.90-0.98, *P*=0.004) was shown for GBM but not for non-GBM (OR=0.98, 95% CI= 0.93-1.03, *P*=0.421). The association between atopic dermatitis and risk of GBM was also apparent in the MLE analysis, which provided an OR of 0.94 (95% CI=0.91-0.97, *P*=2.17 x 10^{-4}). MR-Egger regression provided for an intercept of -0.007 (95% CI=-0.019-0.005, *P*=0.247). As with the analysis of all glioma the association between atopic dermatitis and GBM was weaker under the WME (OR=0.96, 95% CI=0.91-1.02, *P*=0.172) and MBE (OR=0.95, 95% CI= 0.90-1.01, *P*=0.096) frameworks.

Although previously implemented in other studies [32, 38], ratio estimators may not fully recapitulate an estimate of the causal OR in the case of binary-exposures such as atopic dermatitis, and binary-outcomes such as glioma [39]. We therefore evaluated through simulation whether the IVW, MLE, WME, MBE and MR-Egger methods provide reliable estimates of causal ORs. When no causal relationship between exposure and outcome was simulated, each MR method provided accurate estimates of the null relationship (**Additional file 6: Table S5**). Conversely when a causal relationship was simulated the magnitudes of the relationship estimates were weakly inflated in some instances (**Additional file 6: Table S5**), indicating the importance of considering additional evidence when evaluating causal relationships between binary exposures and binary outcomes.

DISCUSSION

To our knowledge, this is the first MR study evaluating a range of atopy related traits with glioma risk undertaken. Overall our results provide evidence for a causal protective effect of atopic dermatitis with GBM tumours, but do not provide evidence that asthma and hay fever, raised IgE levels, or self-reported allergy is protective against the risk of developing glioma.

Possible mechanisms explaining an observed inverse relation between the risk of atopic dermatitis and the risk of glioma have been suggested in previous papers [12], postulated to be the consequence of the hyperactivity of the immune system. The question thus arises as to how such divergent findings for other atopic traits can be explained or reconciled, when they have been previously reported in high numbers.

A key assumption in MR is that the instrument affects glioma risk through its effect on a specific phenotype/exposure (*i.e.* atopic traits), and does not have a direct effect on glioma risk. We tested this assumption using MR-Egger regression and HEIDI outlier analysis and found possible evidence of violation of this assumption for IgE and self-reported allergy. It is notable that self-reported allergy does not show an approximately quadratic response to correlation, in contrast to asthma and hay fever, atopic dermatitis, and IgE level. This is likely to be a consequence of imprecise estimates of the association between SNPs and allergy, illustrating the inherent issue in attempting to make use of self-reported allergy data as an atopy-related trait.

The meta-analyses of published epidemiological observational studies have indeed provided strong evidence for an inverse relationship between atopy and glioma risk [40]. However, most of the support for such a relationship comes from the case-control studies [4]. A common limitation in retrospective studies of glioma has been the use of proxy respondents for cases who have cognitive impairment. A related issue is that glioma cases may not remember past exposures accurately due to cognitive deficits [4]. Such issues are compounded by the fact that across studies multiple atopic traits have been assessed. The strength of support for a relationship seen across case-control studies contrasts markedly with the limited the evidence for a relationship from prospective cohort-based analyses [7].

By inference, a relationship between long-term antihistamine use could theoretically provide supporting evidence, albeit indirect, that atopic mediated mechanisms influence glioma risk. However, the impact of antihistamine use is difficult to disentangle from that of allergies, as these factors are highly correlated, and few individuals without allergies use antihistamines regularly. Paradoxically an increased risk for glioma associated with antihistamine, particularly among individuals with allergic conditions, has been found in some studies [41, 42].

Raised IgE levels and self-reported allergy suffer limitations as traits used to assess the effect of atopy on glioma risk as they are both variable over short time scales in their level of expression (in contrast to clinical diagnosis of atopic dermatitis). Further, allergies may develop later in life, and patients may not necessarily exhibit symptoms. This introduces the possibility of bias and error due to time varying association of SNPs with the exposure. However, it has been suggested that seasonality does not have a significant effect [11]. Th

An additional possible explanation for the lack of causal association between IgE levels and glioma risk seen in this study is that the causality is in fact reversed, which could result in epidemiological observational studies reporting inverse relationships [8, 9], but would not affect an MR analysis. Immunosuppression caused by glioblastoma is well documented [43, 44] and may lead to reduced expression of atopy. Furthermore, in addition to steroids, temozolomide therapy, routinely used to treat GBM nowadays, leads to reduced blood IgE levels [11].

Using data from large genetic consortia for multiple atopy-related traits and glioma risk has enabled us to more precisely test our study hypotheses than if we had used individual-level data from a smaller study. Through simulation scenarios, the IVW, MLE, WME, MBE and MR-Egger methods have been demonstrated to accurately estimate causal effects using summary-level data [28, 30, 31, 45]. However, using summary-level data instead of individual-level data limits the approaches that can be used to test the validity of genetic variants as IVs, as adjusting for measured covariates and assessing gene-environment interactions is generally not possible using summary-level data [46]. The first-stage F statistic was large (>25 for all traits), and so weak instrument bias is unlikely.

Epidemiological observational studies have reported inverse relationships between atopy-related traits and glioma risk, with ORs in the range 0.43-0.96 for asthma [6, 47], 0.42-0.90 for atopic dermatitis [6, 47], 0.37-0.73 for IgE levels [8-10] and 0.47-0.69 for self-reported allergies [4, 5, 8]. Odds ratios for binary exposures estimated in this MR study represent the OR for the outcome disease per doubling in odds of the exposure disease, and the magnitudes of these causal effect estimates are therefore not directly comparable to those reported in observational studies.

Our MR analysis has several strengths. Firstly, by utilising the random allocation of genetic variants, we were able to overcome potential confounding and reverse causation that may bias estimates from observational studies, Secondly, given that a poor outcome from glioma is almost universal, it is unlikely that survival bias will have influenced study findings. Lastly, the findings from this study represent the association of a lifelong atopy with glioma in the general European population.

Our study does however have limitations. Firstly, while it is entirely appropriate to implement different MR methods to assess the robustness of finding, they have different differing power to demonstrate associations, with the WME, MBE and MR-Egger methods having less power than IVW and MLE. Irrespective of such factors our study had only had 80% power to detect ORs of 1.16, 1.09, 1.16 and 1.22 for asthma and hay fever, atopic dermatitis, IgE level and self-reported allergy respectively (**Additional file 5: Table S4**). This is a result of the very low proportion of variability in the atopy-related traits explained by the SNPs used. Hence, we cannot exclude the possibility that these traits influence glioma risk, albeit modestly. To explore this possibility will require additional IVs and larger sample sizes affording increased power. Furthermore, it is possible that an effect of atopy on glioma risk might be mediated through mechanisms associated with a trait that we have not captured by using MR to assess asthma and hay fever, and self-reported allergy. Secondly, a weakness of the two-sample MR strategy is that it does not allow examination of non-linear relationships between exposures and outcomes. Finally, we have sought to examine whether bias could be introduced when considering a binary exposure for a binary outcome. Although in our simulation study we found no evidence of bias when estimating non-causal

relationships, we did not extend our analysis to consider the potential impact of invalid SNPs.

CONCLUSIONS

In conclusion, our investigation does not provide strong evidence for a relationship between atopy-related diseases and risk of developing glioma, but findings do not preclude a small effect for atopic dermatitis. Our analysis also serves to illustrate the value of using several MR methods to derive robust conclusions.

LIST OF ABBREVIATIONS

CI: Confidence interval

GBM: Glioblastoma

GICC: Glioma International Case-Control Consortium Study

GWAS: Genome-wide association study

IV: Instrumental variable

IVW: Inverse-variance weighted

MBE: Mode-based estimate

MLE: Maximum likelihood estimation

MR: Mendelian randomization

OR: Odds ratio

SE: Standard error

SNP: Single nucleotide polymorphism

WHO: World Health Organization

WME: Weighted median estimate

DECLARATIONS

Ethics approval and consent to participate

Two-sample MR was undertaken using GWAS data. Ethical approval was not sought for this specific project because all data came from the summary statistics of published GWAS, and no individual-level data were used.

Consent for publication

Not applicable

Availability of data and material

Genotype data from the GICC GWAS are available from the database of Genotypes and Phenotypes (dbGaP) under accession phs001319.v1.p1. Additionally, genotypes from the GliomaScan GWAS can be accessed through dbGaP accession phs000652.v1.p1.

Competing interests

The authors declare that they have no competing interests

Funding

L D-H was supported by a Wellcome Trust Summer Student bursary. A.S. is supported by a Cancer Research UK clinical Fellowship. In the UK, funding was provided by Cancer Research UK (C1298/A8362) supported by the Bobby Moore Fund. The GICC was supported by grants from the National Institutes of Health, Bethesda, Maryland (R01CA139020, R01CA52689, P50097257, P30CA125123). The UK Interphone Study was supported by the European Commission Fifth Framework Program "Quality of Life and Management of Living Resources" and the UK Mobile Telecommunications and Health Programme. The Mobile

Manufacturers Forum and the GSM Association provided funding for the study through the scientifically independent International Union against Cancer (UICC).

Author contributions

R.S.H. and A.J.C. managed the project. L.D-H., A.J.C., A.S., P.J.L. and R.S.H. drafted the manuscript. L.D-H. and A.J.C. performed statistical analyses. B.K., K.L., M.J.S. and R.H.S. acquired and analysed the U.K. data. M. Simon, P.H., M.M.N. and K.-H.J. acquired and analysed the German data. D.I.J., Q.T.O., J.E.E.-P., G.N.A., E.B.C., D.I., J.S., J.S.B.-S., S.H.O., J.L.B., R.K.L., C.J., R.B.J., B.S.M., M.R.W., M.L.B. and R.S.H. acquired and analysed the GICC data. S.C. and P.R. acquired and analysed the National Cancer Institute (NCI) data. M. Sanson acquired and analysed the French data. All authors reviewed the final manuscript.

Acknowledgements

Not applicable

612

613 **Table 1: Variant and effect allele with frequencies and magnitude of effect on each atopy-**

614 **related trait and strength of association with glioma**

615

Region	SNP	Position (bp)*	Alleles**	MAF	Hay Fever and Asthma	Glioma
					OR (95% CI)	OR (95% CI)
2q12.1	rs10197862	102966549	G/A	G=0.161	1.24 (1.16-1.32)	0.98 (0.93–1.03)
4p14	rs4833095	38799710	C/T	T=0.425	1.20 (1.14-1.26)	1.03 (0.99–1.08)
5q22.1	rs1837253	110401872	T/C	T=0.382	1.17 (1.11-1.23)	0.96 (0.93–1.00)
8q21.13	rs7009110	81291879	C/T	C=0.467	1.14 (1.09-1.19)	0.98 (0.94–1.01)
9p24.1	rs72699186	6175855	A/T	T=0.110	1.26 (1.17-1.36)	0.97 (0.93–1.02)
11q13.5	rs2155219	76299194	G/T	G=0.468	1.17 (1.13-1.21)	1.01 (0.97–1.05)
15q22.33	rs17294280	67468285	A/G	G=0.120	1.18 (1.12-1.25)	0.98 (0.94–1.03)
16p13.13	rs62026376	11228712	T/C	T=0.144	1.17 (1.11-1.23)	0.97 (0.93–1.01)
17q21.1	rs7212938	38122680	T/G	G=0.473	1.16 (1.11-1.22)	1.00 (0.97–1.04)

616

Region	SNP	Position*	Alleles**	MAF	Atopic Dermatitis	Glioma
					OR (95% CI)	OR (95% CI)
1q21.3	rs11205006	152440176	T/A	A=0.265	1.62 (1.48-1.77)	0.96 (0.91–1.02)
1q21.3	rs2228145	154426970	A/C	C=0.293	1.15 (1.10-1.20)	0.99 (0.96–1.03)
2p25.1	rs10199605	8495097	A/G	A=0.244	1.04 (1.03-1.06)	1.01 (0.97–1.05)
2p13.3	rs112111458	71100105	G/A	G=0.224	1.08 (1.05-1.10)	0.98 (0.92–1.03)
2q24.3	rs6720763	167992286	T/C	C=0.320	1.29 (1.18-1.41)	1.02 (0.97–1.06)
5p13.2	rs10214237	35883734	C/T	C=0.176	1.06 (1.05-1.08)	0.98 (0.94–1.02)
5q31.1	rs1295686	131995843	C/T	T=0.422	1.35 (1.22-1.49)	0.99 (0.95–1.03)
6p21.32	rs12153855	32074804	T/C	C=0.125	1.58 (1.40-1.78)	0.97 (0.92–1.03)
8q21.13	rs6473227	81285892	A/C	A=0.473	1.06 (1.05-1.08)	0.98 (0.94–1.02)
9p21.3	rs10738626	22373457	C/T	C=0.397	1.23 (1.15-1.32)	0.96 (0.93–1.00)
10p15.1	rs6602364	6038853	G/C	G=0.492	1.05 (1.03-1.07)	1.03 (0.99–1.07)
11q13.1	rs10791824	65559266	A/G	G=0.490	1.15 (1.12-1.19)	0.99 (0.95–1.02)
11q24.3	rs7127307	128187383	C/T	C=0.488	1.09 (1.07-1.11)	0.99 (0.95–1.03)
11q13.5	rs7130588	76270683	G/A	G=0.216	1.29 (1.20-1.38)	1.02 (0.98–1.06)
14q13.2	rs2143950	35572357	C/T	T=0.215	1.08 (1.06-1.10)	1.01 (0.97–1.06)
16p13.13	rs2041733	11229589	C/T	T=0.496	1.09 (1.06-1.11)	0.97 (0.94–1.01)
19p13.2	rs2164983	8789381	C/A	A=0.169	1.16 (1.10-1.22)	0.95 (0.90–1.00)
20q13.33	rs909341	62328742	T/C	T=0.262	1.32 (1.21-1.44)	1.32 (1.26–1.37)

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Region	SNP	Position*	Alleles**	MAF	IgE level***	Glioma
					OR (95% CI)	OR (95% CI)
1q23.2	rs2251746	159272060	C/T	C=0.015	1.09 (1.08-1.11)	0.98 (0.95–1.02)
5q31.1	rs20541	131995964	A/G	A=0.270	1.08 (1.06-1.10)	1.01 (0.97–1.06)
6p22.1	rs2571391	29923838	C/A	C=0.303	1.06 (1.05-1.08)	0.97 (0.94–1.01)
6p21.32	rs2858331	32681277	A/G	G=0.490	1.04 (1.03-1.06)	1.02 (0.98–1.06)
12q13.3	rs1059513	57489709	C/T	C=0.070	1.13 (1.09-1.17)	0.97 (0.92–1.03)

618

Region	SNP	Position*	Alleles**	MAF	Self-reported allergy	Glioma
					OR (95% CI)	OR (95% CI)
2q12.1	rs10189699	102879464	A/C	A=0.143	1.16 (1.12-1.20)	0.99 (0.94–1.04)
2q33.1	rs10497813	198914072	T/G	T=0.401	1.08 (1.05-1.11)	0.99 (0.96–1.03)
3q28	rs9860547	188128979	G/A	A=0.272	1.08 (1.05-1.11)	1.02 (0.98–1.06)
4p14	rs2101521	38811551	A/G	A=0.475	1.15 (1.12-1.18)	1.02 (0.98–1.07)
4q27	rs17388568	123329369	G/A	A=0.141	1.08 (1.05-1.11)	1.01 (0.97–1.05)
5p13.1	rs7720838	40486896	G/T	T=0.362	1.08 (1.06-1.11)	1.02 (0.99–1.06)
5q22.1	rs1438673	110467499	T/C	C=0.296	1.12 (1.09-1.15)	0.97 (0.94–1.01)
6p21.33	rs9266772	31352113	T/C	C=0.175	1.11 (1.08-1.14)	1.03 (0.98–1.08)
9p24.1	rs7032572	6172380	A/G	G=0.114	1.12 (1.08-1.16)	0.97 (0.93–1.02)
10p14	rs962993	9053132	T/C	T=0.106	1.07 (1.05-1.10)	1.02 (0.98–1.06)
11q13.5	rs2155219	76999194	G/T	G=0.468	1.11 (1.09-1.14)	1.01 (0.97–1.05)
15q22.33	rs17228058	67450305	A/G	G=0.100	1.08 (1.05-1.11)	1.00 (0.96–1.04)
17q21.1	rs9303280	38074031	T/C	T=0.346	1.07 (1.05-1.09)	0.98 (0.94–1.02)
20q13.2	rs6021270	50141264	C/T	T=0.346	1.16 (1.10-1.22)	1.02 (0.94–1.10)

* NCBI build 37; ** Reference allele/effect allele; *** per standard deviation; MAF= minor allele frequency; OR= odds ratio

Table 2: IVW, MLE, WME, MBE and MR-Egger test results for combined atopy-related instrumental variables

Trait	IVW		MLE		WME		MBE		MR-Egger slope		MR-Egger intercept	
	OR (95% CI)	*P*	OR (95% CI)	*P*	OR (95% CI)	*P*	OR (95% CI)	*P*	OR (95% CI)	*P*	Estimate (95% CI)	*P*
Asthma and hay fever	0.96 (0.90-1.03)	0.248	0.96 (0.93-1.00)	0.066	0.93 (0.86-1.01)	0.087	0.91 (0.80-1.04)	0.191	0.99 (0.72-1.36)	0.951	-0.007 (-0.030-0.016)	0.542
Atopic dermatitis	0.96 (0.93-1.00)	0.041	0.96 (0.94-0.99)	0.003	0.96 (0.91-1.01)	0.114	0.97 (0.92-1.02)	0.194	0.97 (0.92-1.03)	0.375	0.004 (-0.014-0.006)	0.396
IgE level	0.88 (0.69-1.13)	0.319	0.88 (0.74-1.05)	0.157	0.83 (0.61-1.12)	0.218	0.82 (0.57-1.19)	0.355	0.63 (0.32-1.25)	0.184	0.027 (0.001-0.053)	0.042
Self-reported allergy	1.03 (0.95-1.11)	0.534	1.02 (0.97-1.08)	0.429	1.08 (0.97-1.20)	0.184	1.12 (0.92-1.36)	0.275	0.92 (0.69-1.22)	0.540	0.017 (0.003-0.031)	0.018

IVW= inverse variance weighted; MLE= maximum likelihood estimation; WME= weighted median estimate; MBE= mode-based estimate

Figure 1: Forest plot of Wald ORs and 95% confidence intervals generated from SNPs associated with atopy-related traits. ORs for individual SNPs are listed according to magnitude of effect in the instrumental variable analysis and are presented with pooled effects using the IVW method. Squares represent the point estimate, and the bars are the 95% confidence intervals. (a) Asthma and hay fever, (b) atopic dermatitis, (c) IgE level, (d) self-reported allergy.

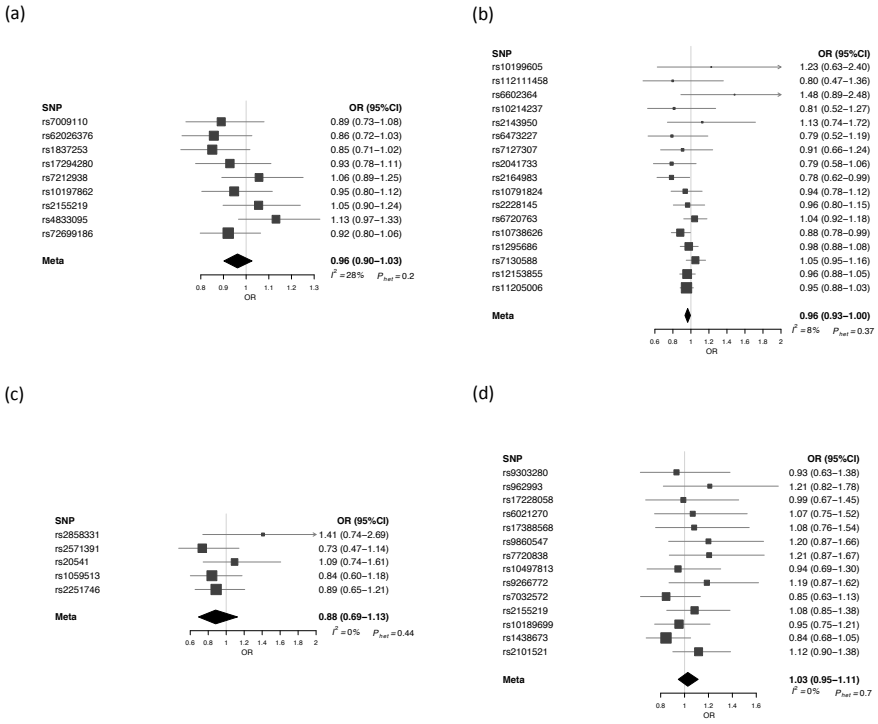


Figure 2: Plot of *P* value of MLE associations with glioma against correlation between errors in X_k and Y_k. (a) Asthma and hay fever, (b) atopic dermatitis, (c) IgE level, (d) self-reported allergy.

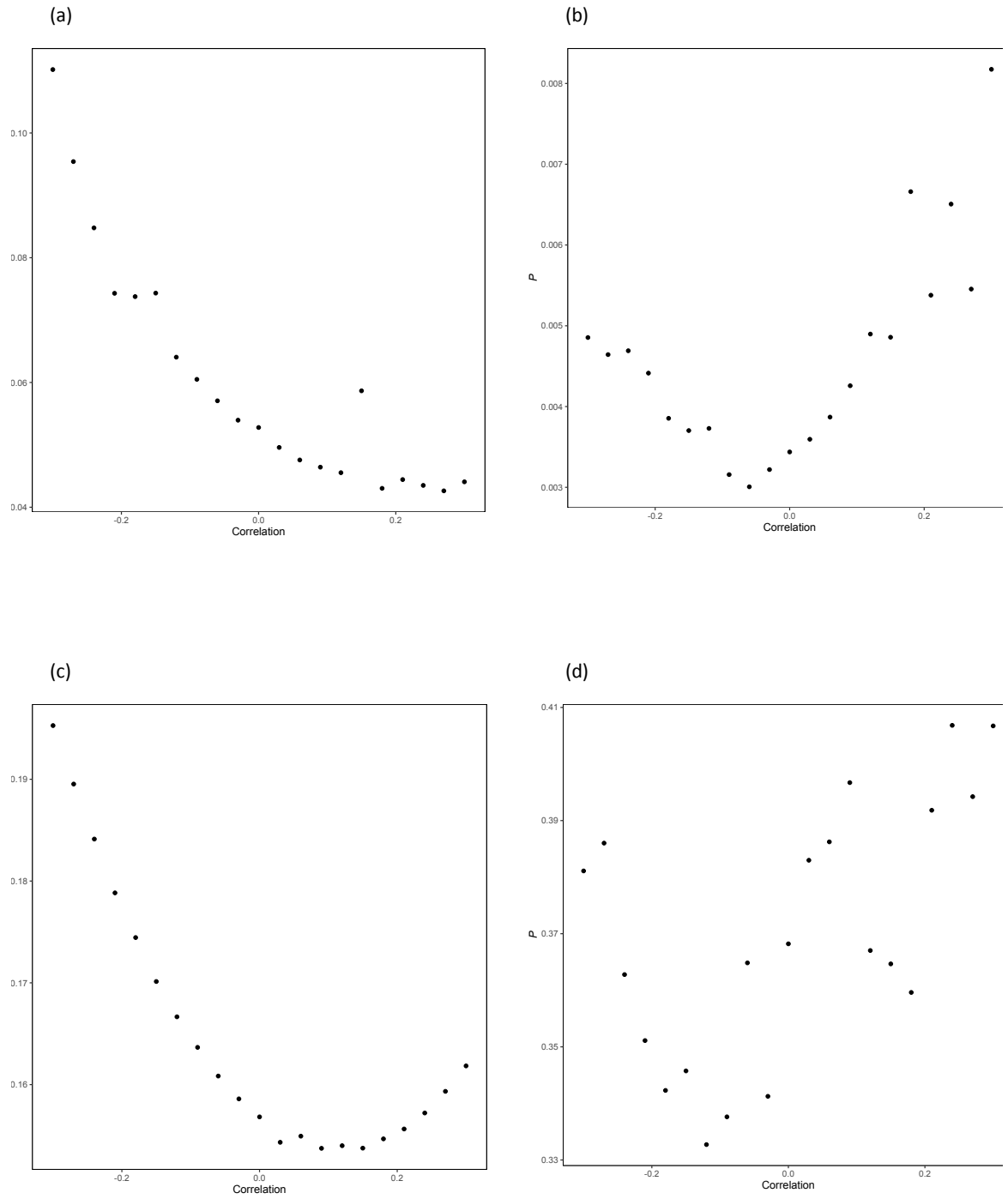
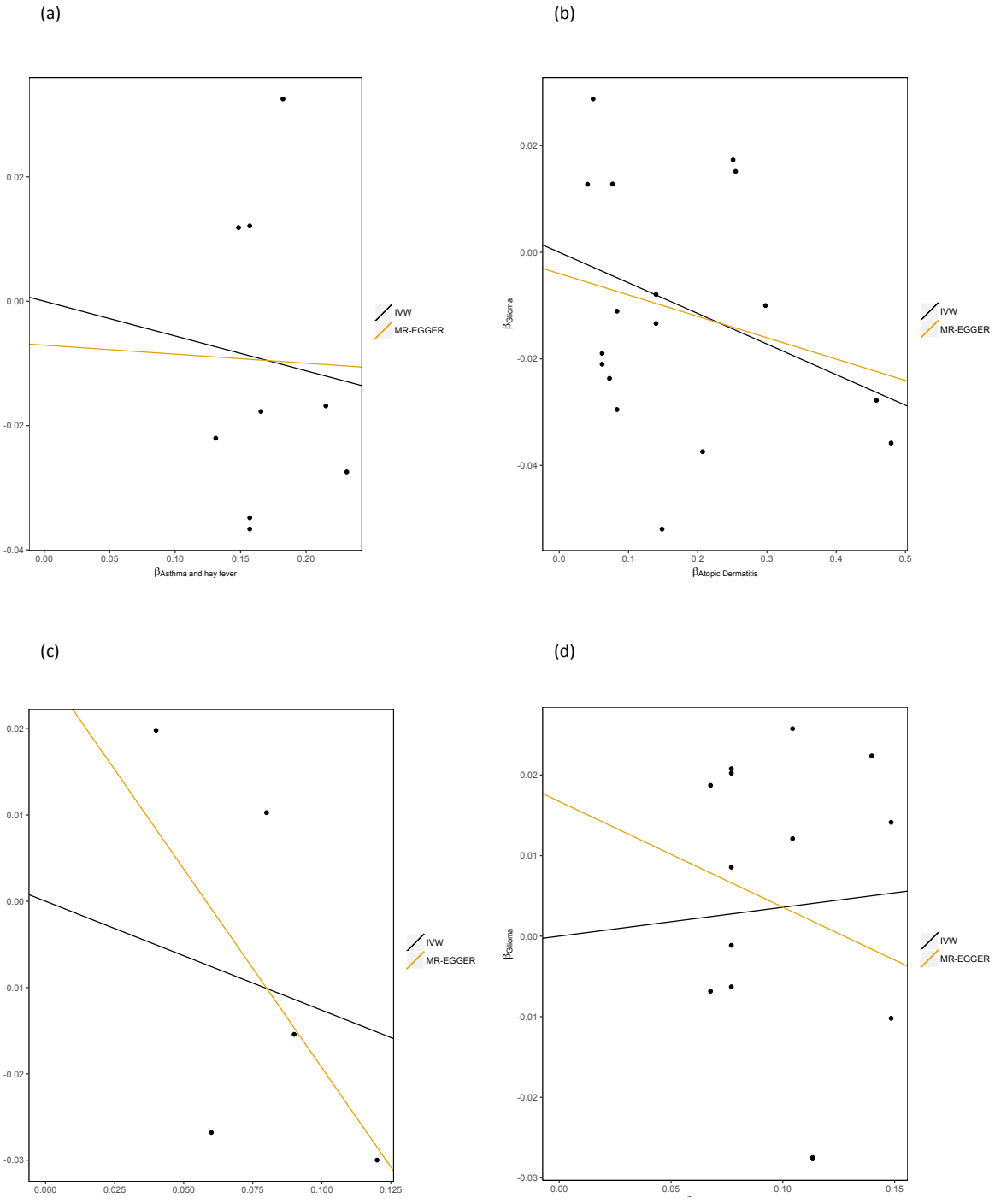


Figure 3: Scatter plots of genetic associations with glioma against genetic associations with the exposure. (a) Asthma and hay fever, (b) atopic dermatitis, (c) IgE level, (d) self-reported allergy.



ADDITIONAL FILES

Additional file 1: Figure S1: Forest plot of Wald ORs and 95% confidence intervals generated from SNPs associated with atopic dermatitis, including rs909341. ORs for individual SNPs are listed according to magnitude of effect in the instrumental variable analysis and are presented with pooled effects using the IVW method. Squares represent the point estimate, and the bars are the 95% confidence intervals. (DOCX 92 kb)

Additional file 2: Table S1: Summary of the eight glioma genome-wide association studies. (XLSX 30 kb)

Additional file 3: Table S2: Table of SNPs reported in the NHGRI-EBI GWAS Catalog for each trait, with correlations between SNPs. (XLSX 50 kb)

Additional file 4: Table S3: Percentage of variance explained by the combined sets of SNPs used as IVs. (XLSX 34 kb)

Additional file 5: Table S4: Range of ORs for which study had <80% power, for each atopy-related trait (P=0.05, two-sided). (XLSX 9 kb)

Additional file 6: Table S5: Simulation analyses. (XLSX 30 kb)

Additional file 7: Table S6: IVW, MLE, WME, MBE and MR-Egger test results for combined atopy-related instrumental variables and glioma subtypes. (XLSX 40kb)

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igure S1: Forest plot of Wald ORs and 95% confidence intervals generated from SNPs associated with atopic dermatitis, including rs909341. ORs for individual SNPs re listed according to magnitude of effect in the instrumental variable analysis and are presented with pooled effects using the IVW method. Squares represent the oint estimate, and the bars are the 95% confidence intervals.

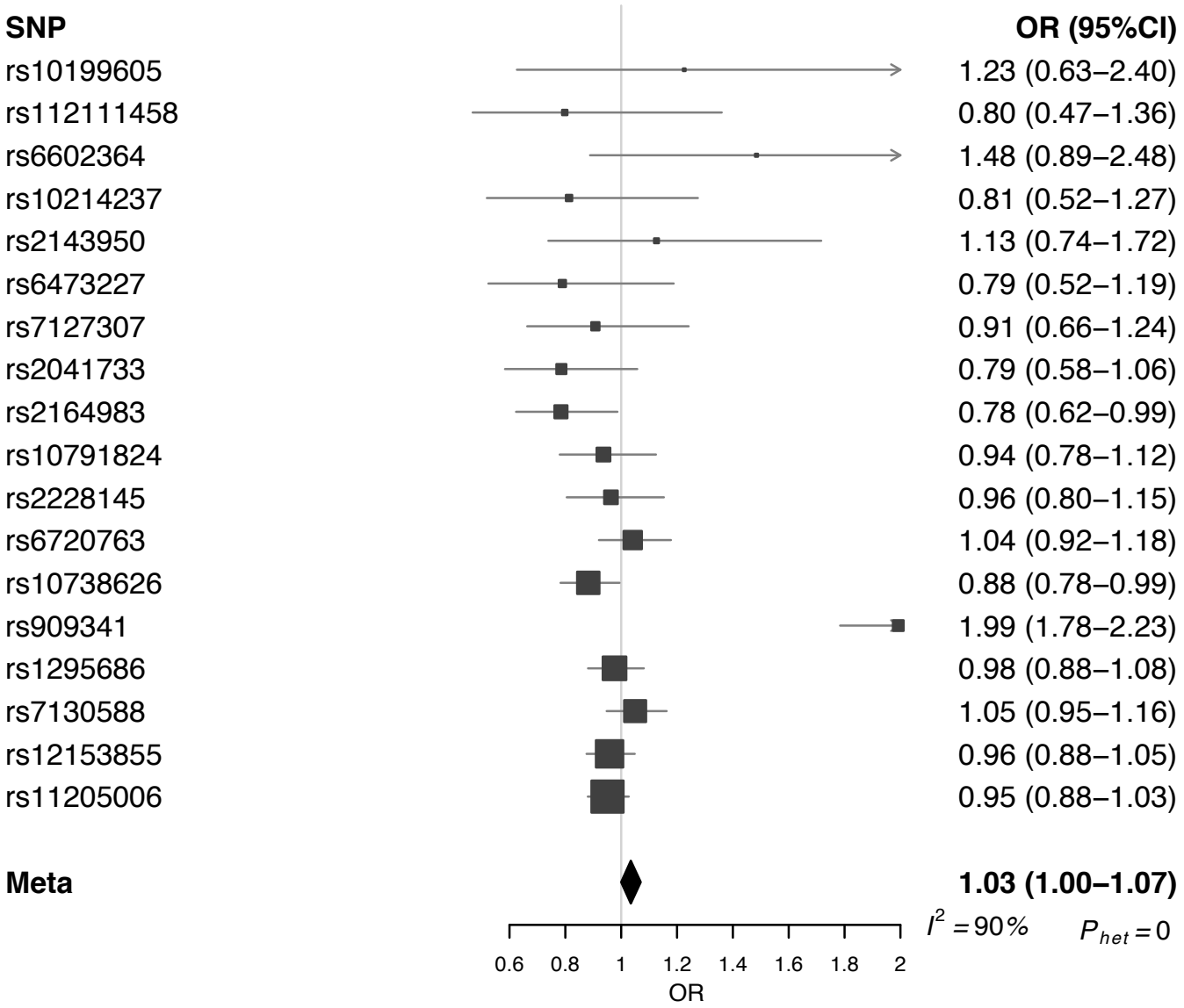


Table S1: Summary of the eight glioma genome-wide assoc

Series	Study centre
FRE	Groupe Hospitalier Pitié-Salpêtrière Paris
GER	University of Bonn
GICC	GLIOGENE Consortium
MDA	The University of Texas M.D. Anderson Cancer Center
GiomaScan (NIH)	National Cancer Institute
UCSF-Mayo	Mayo Clinic
UCSF (SFAGS)	University of California, San Francisco
UK	INTERPHONE
Total	

iation studies

Sampling
Patients with glioma were ascertained through the Service de Neurologie Mazarin, Groupe Hospitalier Pitié-Salpêtrière Paris. Controls were ascertained from the SU.VI.MAX (Supplementation en Vitamines et MineralsAntioXydants) study.
Comprised of patients who had undergone surgery for a glioma at the Department of Neurosurgery, University of Bonn Medical Center, between 1996 and 2008. Control subjects were taken from three population studies: KORA (Co- operative Health Research in the Region of Augsburg); POPGEN (Population Genetic Cohort) and the Heinz Nixdorf Recall study.
Comprise glioma cases and controls that were ascertained through Brigham and Women's Hospital (Boston, Massachusetts), Case Western Reserve University (Cleveland, Ohio), Columbia University (New York, New York), the Danish Cancer Society Research Centre (Copenhagen, Denmark), the Gertner Institute (Tel Hashomer, Israel), Duke University (Durham, North Carolina), the University of Texas MD Anderson Cancer Center (Houston, Texas), Memorial Sloan Kettering Cancer Center (New York, New York), the Mayo Clinic (Rochester, Minnesota), NorthShore HealthSystem (Chicago, Illinois), Umeå University (Umeå, Sweden), the University of California, San Francisco (San Francisco, California), the University of Southern California (Los Angeles, California), and the Institute of Cancer Research (London, United Kingdom). Cases had newly diagnosed glioma, and controls had no personal history of central nervous system tumor at the time of ascertainment
Cases were ascertained through the MD Anderson Cancer Center, Texas, between 1990 and 2008. Individuals from the Cancer Genetic Markers of Susceptibility studies served as controls. Cases were newly diagnosed glioma [ICDO-3 codes 9380-9480 or equivalent], and controls were cancer-free at the time of glioma diagnosis.
Comprised of Mayo cases, UCSF cases, and Mayo Clinic Biobank control data
Cases were adults with newly diagnosed, histologically confirmed glioma. Population-based cases who were diagnosed between 1991 and 2009 and who were residing in the six San Francisco Bay area counties were ascertained using the Cancer Prevention Institute of California's early-case ascertainment system. Clinic-based cases who were diagnosed between 2002 and 2012 were recruited from the UCSF Neuro-oncology Clinic, regardless of the place of residence. From 1991 to 2010, population-based controls from the same residential area as the population-based cases were identified using random digit-dialing and were frequency matched to population-based cases for age, gender and ethnicity. Between 2010 and 2012, all controls were selected from the UCSF general medicine phlebotomy clinic. Clinic-based controls were matched to clinic-based glioma cases for age, gender and ethnicity.
Cases were ascertained through the INTERPHONE study. Individuals from the 1958 Birth Cohort served as a source of controls.

No. cases	No. controls
1,423	1,190
846	1,310
4,564	3,265
1,175	2,236
1,653	2,725
1,519	804
677	3,940
631	2,699
12,488	18,169

Table S2: Table of SNPs reported in the NHGRI-EBI GWAS Catalog for each trait, with correlations between SNPs

Asthma and hay fever Chr	Chr rsid	2 rs10197862	4 rs4833095	5 rs1438673	5 rs1837253	8 rs7009110	9 rs72699186	11 rs2155219	15 rs17294280	16 rs62026376
2	rs10197862	1								
4	rs4833095	0	1							
5	rs1438673	0	0	1						
5	rs1837253	0	0	0.0290264	1					
8	rs7009110	0	0	0	0	1				
9	rs72699186	0	0	0	0	0	1			
11	rs2155219	0	0	0	0	0	0	1		
15	rs17294280	0	0	0	0	0	0	0	1	
16	rs62026376	0	0	0	0	0	0	0	0	1
17	rs7212938	0	0	0	0	0	0	0	0	0

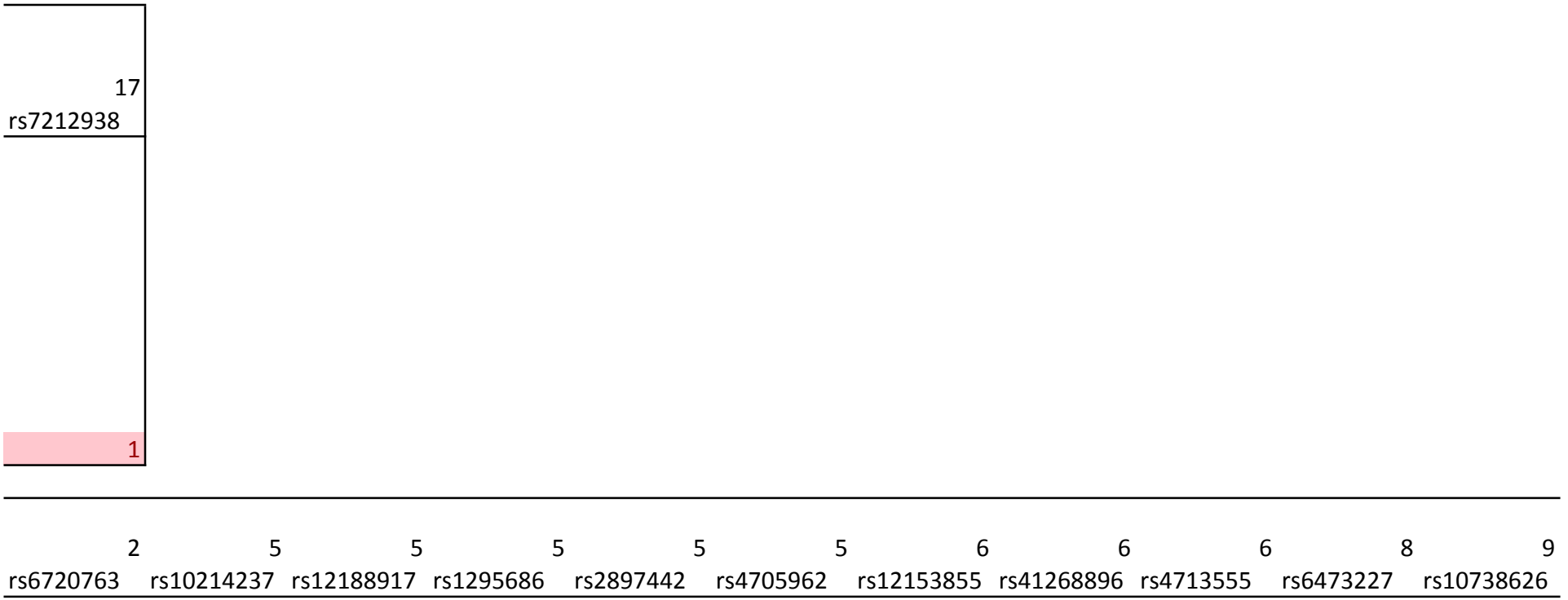
Atopic Dermatitis Chr	Chr rsid	1 rs11205006	1 rs12144049	1 rs2228145	1 rs61813875	1 rs6661961	1 rs7512552	2 rs10199605	2 rs112111458	2 rs6419573
1	rs11205006	1								
1	rs12144049	1	1							
1	rs2228145	0.00033237	9.48E-05	1						
1	rs61813875	0.0346025	0.0746275	2.95E-05	1					
1	rs6661961	0.477915	0.5082	0.00028748	0.0345781	1				
1	rs7512552	0.002187	0.00051363	1.61E-05	0.00016209	0.00036636	1			
2	rs10199605	0	0	0	0	0	0	1		
2	rs112111458	0	0	0	0	0	0	0.00011906	1	
2	rs6419573	0	0	0	0	0	0	2.76E-05	0.0033077	1

2	rs6720763	0	0	0	0	0	0	0.00011986	6.94E-05	0.00242265
5	rs10214237	0	0	0	0	0	0	0	0	0
5	rs12188917	0	0	0	0	0	0	0	0	0
5	rs1295686	0	0	0	0	0	0	0	0	0
5	rs2897442	0	0	0	0	0	0	0	0	0
5	rs4705962	0	0	0	0	0	0	0	0	0
6	rs12153855	0	0	0	0	0	0	0	0	0
6	rs41268896	0	0	0	0	0	0	0	0	0
6	rs4713555	0	0	0	0	0	0	0	0	0
8	rs6473227	0	0	0	0	0	0	0	0	0
9	rs10738626	0	0	0	0	0	0	0	0	0
10	rs6602364	0	0	0	0	0	0	0	0	0
11	rs10791824	0	0	0	0	0	0	0	0	0
11	rs2212434	0	0	0	0	0	0	0	0	0
11	rs479844	0	0	0	0	0	0	0	0	0
11	rs7127307	0	0	0	0	0	0	0	0	0
11	rs7130588	0	0	0	0	0	0	0	0	0
11	rs7927894	0	0	0	0	0	0	0	0	0
14	rs2143950	0	0	0	0	0	0	0	0	0
16	rs2041733	0	0	0	0	0	0	0	0	0
19	rs2164983	0	0	0	0	0	0	0	0	0
19	rs2918307	0	0	0	0	0	0	0	0	0
20	rs4809219	0	0	0	0	0	0	0	0	0
20	rs6010620	0	0	0	0	0	0	0	0	0
20	rs909341	0	0	0	0	0	0	0	0	0

IgE levels	Chr	1	1	1	5	6	6	6	12
Chr	rsid	rs4656784	rs13962	rs2251746	rs20541	rs2858331	rs2571391	rs2523809	rs1059513
1	rs4656784	1							

1	rs13962	0.3643	1							
1	rs2251746	0.462008	0.410714	1						
5	rs20541	0	0	0	1					
6	rs2858331	0	0	0	0	1				
6	rs2571391	0	0	0	0	0.00010884	1			
6	rs2523809	0	0	0	0	0.0130407	0.0648709	1		
12	rs1059513	0	0	0	0	0	0	0	1	

Self-reported allergy Chr	Chr	2	4	5	5	8	9	11	15	16
	rsid	rs10197862	rs4833095	rs1438673	rs1837253	rs7009110	rs72699186	rs2155219	rs17294280	rs62026376
2	rs10197862	1								
4	rs4833095	0	1							
5	rs1438673	0	0	1						
5	rs1837253	0	0	0.0290264	1					
8	rs7009110	0	0	0	0	1				
9	rs72699186	0	0	0	0	0	1			
11	rs2155219	0	0	0	0	0	0	1		
15	rs17294280	0	0	0	0	0	0	0	1	
16	rs62026376	0	0	0	0	0	0	0	0	1
17	rs7212938	0	0	0	0	0	0	0	0	0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rs7212938
1
2
rs6720763
5
rs10214237
5
rs12188917
5
rs1295686
5
rs2897442
5
rs4705962
6
rs12153855
6
rs41268896
6
rs4713555
8
rs6473227
9
rs10738626

1										
0	1									
0	0.00239469	1								
0	5.00E-06	0.24802	1							
0	0.0002101	0.0239946	0.162452	1						
0	0.00047095	0.0348378	0.180208	0.858099	1					
0	0	0	0	0	0	1				
0	0	0	0	0	0	0.0452232	1			
0	0	0	0	0	0	0.18826	0.0275876	1		
0	0	0	0	0	0	0	0	0	1	
0	0	0	0	0	0	0	0	0	0	1
0	0	0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0	0	0
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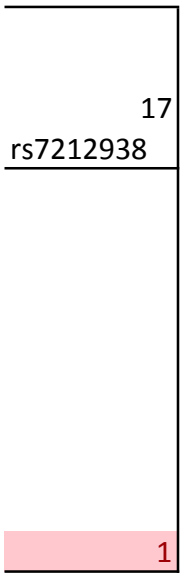
0	0	0	0	0	0	0	0	0	0	0


17
rs7212938
1

10	11	11	11	11	11	11	14	16	19	19
rs6602364	rs10791824	rs2212434	rs479844	rs7127307	rs7130588	rs7927894	rs2143950	rs2041733	rs2164983	rs2918307

1										
0	1									
0	0.00026763	1								
0	0.938237	0.00055208	1							
0	0.00037286	0.00059044	0.00025972	1						
0	0.00024611	0.674246	0.00036951	0.00047794	1					
0	0.00028194	0.597991	0.00067521	0.00095006	0.84696	1				
0	0	0	0	0	0	0	1			
0	0	0	0	0	0	0	0	1		
0	0	0	0	0	0	0	0	0	1	
0	0	0	0	0	0	0	0	0	1	1
0	0	0	0	0	0	0	0	0	0	0
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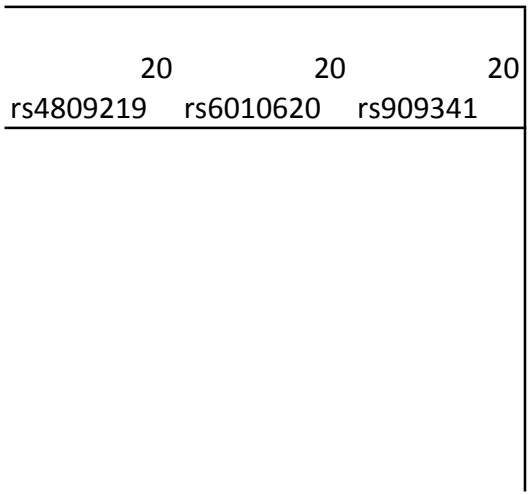
20
20
20
rs4809219
rs6010620
rs909341

1		
0.0985128	1	
0.748019	0.756937	1

Table S3: Percentage of variance explained by the combined sets of SNPs used as IVs.

Trait	Number of SNPs used as IV	Percentage of variance explained
Asthma and hay fever	9	2.1
Atopic dermatitis	18	6.9
IgE levels	5	4.8
Self-reported allergy	14	1.3

Table S4: Range of ORs for which study had <80% power, for each atopy-related trait (*P*=0.05, two-sided).

	<80% ORs		
Trait	**All glioma**	**GBM**	**non-GBM**
Asthma and hay fever	0.86-1.16	0.82-1.22	0.82-1.22
Atopic dermatitis	0.92-1.09	0.89-1.11	0.89-1.11
IgE levels	0.86-1.16	0.83-1.21	0.83-1.21
Self-reported allergy	0.82-1.22	0.78-1.29	0.77-1.29

Table S5: Simulation analyses.

		Simulation parameters						
Trait	Scenario	OR for outcome per doubling in odds of exposure (*v*)	Number of genetic variants used as IVs	Number of cases in exposure GWAS	Number of controls in exposure GWAS	Number of cases in outcome GWAS	Number of controls in outcome GWAS	Mean simulated d Exposure
Asthma and hay fever	i	1.00	9	6,685	14,091	12,488	18,169	0.010
Atopic dermatitis	i	1.00	18	21,399	95,464	12,488	18,169	0.092
Self-reported allergy	i	1.00	14	26,311	27,551	12,488	18,169	0.044
Asthma and hay fever	ii	1.33	9	6,685	14,091	12,488	18,169	0.015
Atopic dermatitis	ii	1.33	18	21,399	95,464	12,488	18,169	0.110
Self-reported allergy	ii	1.33	14	26,311	27,551	12,488	18,169	0.058

IVW: inverse variance weighted; MLE: maximum likelihood estimate; WME: weighted median estimate; MBE: mode-based estimate; OR

Simulation results					
lisease prevalence	Mean estimated OR (SD)				
Outcome	IVW	MLE	WME	MBE	MR-Egger
0.010	0.999 (0.015)	0.999 (0.014)	0.998 (0.018)	0.998 (0.019)	0.999 (0.027)
0.010	1.002 (0.010)	1.002 (0.009)	1.002 (0.012)	1.001 (0.014)	1.003 (0.018)
0.010	1.000 (0.014)	1.001 (0.012)	0.999 (0.015)	0.999 (0.015)	0.996 (0.023)
0.001	1.327 (0.022)	1.329 (0.025)	1.326 (0.027)	1.327 (0.026)	1.324 (0.046)
0.004	1.359 (0.029)	1.355 (0.031)	1.356 (0.029)	1.357 (0.029)	1.356 (0.030)
0.003	1.343 (0.027)	1.338 (0.028)	1.343 (0.028)	1.343 (0.028)	1.341 (0.033)

: odds ratio; SD: standard deviation

Table S6: IVW, MLE, MBE and MR-Egger test results for combined atopy-related instrumental variables and glioma subtypes

	GBM								
	IVW		MLE		WME		MBE		MR-Egge
Trait	OR (95% CI)	*P*-value	OR (95% CI)	*P*-value	OR (95% CI)	*P*-value	OR (95% CI)	*P*-value	OR (95% CI)
Asthma and hay fever	0.97 (0.89-1.06)	0.515	0.98 (0.93-1.03)	0.375	0.95 (0.86-1.05)	0.349	0.92 (0.75-1.12)	0.429	1.01 (0.68-1.49)
Atopic dermatitis	0.94 (0.90-0.98)	0.004	0.94 (0.91-0.97)	2.17E-04	0.96 (0.91-1.02)	0.172	0.95 (0.90-1.01)	0.096	0.96 (0.89-1.03)
IgE levels	0.92 (0.67-1.25)	0.587	0.92 (0.74-1.14)	0.427	0.78 (0.54-1.13)	0.191	0.77 (0.50-1.18)	0.300	0.46 (0.20-1.06)
Self-reported allergy	1.04 (0.94-1.15)	0.473	1.04 (0.96-1.11)	0.327	1.05 (0.92-1.20)	0.463	1.06 (0.88-1.27)	0.566	0.94 (0.66-1.33)

IVW: inverse variance weighted; MLE: maximum likelihood estimate; WME: weighted median estimate, MBE: mode-based estimate

	Non-GBM glioma									
r	IVW		MLE		WME		MBE		MR-Egger	
P-value	OR (95% CI)	*P*-value	OR (95% CI)	*P*-value	OR (95% CI)	*P*-value	OR (95% CI)	*P*-value	OR (95% CI)	*P*-value
0.985	0.96 (0.90-1.04)	0.325	0.96 (0.91-1.02)	0.161	0.95 (0.86-1.05)	0.343	0.92 (0.79-1.07)	0.313	0.91 (0.60-1.39)	0.656
0.237	0.98 (0.93-1.03)	0.421	0.99 (0.96-1.03)	0.602	0.96 (0.90-1.03)	0.267	0.96 (0.90-1.02)	0.236	0.96 (0.89-1.04)	0.320
0.067	0.97 (0.70-1.35)	0.853	0.97 (0.77-1.22)	0.791	1.09 (0.74-1.62)	0.651	1.11 (0.71-1.72)	0.670	1.21 (0.59-2.94)	0.668
0.718	1.04 (0.94-1.15)	0.473	1.04 (0.96-1.11)	0.327	1.05 (0.92-1.20)	0.446	1.06 (0.86-1.29)	0.601	0.77 (0.53-1.13)	0.178