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Analysis of the correlation between inflammatory cytokines and glioblastoma

A Mendelian randomization study

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Abstract

Epidemiological studies have demonstrated that inflammatory cytokines are associated with cancer development. However, the causal relationship between inflammatory cytokines and glioblastoma remains unclear. We used a two-sample Mendelian randomization approach to determine the potential causal effects of inflammatory cytokines on glioblastoma. Genome-wide association study summary statistics for 41 inflammatory cytokines were obtained from the University of Bristol database, and 91 inflammatory cytokines were acquired from the genome-wide association study catalog database. Genetic data on glioblastoma were downloaded from the FinnGen consortium (R10). Mendelian randomization analysis and Bayesian weighted Mendelian randomization analysis were performed to investigate the causal relationship between inflammatory cytokines and glioblastoma risk. A combination of Mendelian randomization (MR)-Egger, MR-Pleiotropy Residual Sum and Outliers, and Radial MR methods was employed to assess horizontal pleiotropy, which is a potential bias in MR studies. The Cochran Q test was used to quantify the degree of heterogeneity. Finally, we conducted a comprehensive meta-analysis to confirm the robustness of our findings. Increased levels of tumor necrosis factor β (odds ratio [OR] = 1.597, 95% CI: 1.143–2.230, $P = .006$) and interleukin-10 (OR = 1.452, 95% CI: 1.059–1.992, $P = .021$) were associated with an increased risk of glioblastoma. Conversely, higher levels of circulating fibroblast growth factor 21 (OR = 0.456, 95% CI: 0.276–0.754, $P = .002$) and macrophage inflammatory protein 1a (OR = 0.743, 95% CI: 0.558–0.990, $P = .042$) were associated with a decreased risk of glioblastoma. No significant causal effect on inflammatory cytokines from glioblastoma was detected, and no significant heterogeneity in instrumental variables or horizontal pleiotropy was observed. Our findings indicate that specific inflammatory cytokines may play a role in glioblastoma development, acting as either protective factors or risk factors. This offers valuable insights into the disease mechanism and suggests that targeting these cytokines could be a potential strategy for glioblastoma prevention and treatment.

Abbreviations: BWMR = Bayesian weighted Mendelian randomization, CCL3 = chemokine (C-C motif) ligand 3, FGF21 = fibroblast growth factor 21, GBM = glioblastoma, GWAS = genome-wide association study, IL-10 = interleukin-10, IVs = instrumental variables, IVW = inverse variance weighted, LD = linkage disequilibrium, MR = Mendelian randomization, OR = odds ratio, SNPs = single nucleotide polymorphisms, TNFα = tumor necrosis factor alpha, TNFβ = tumor necrosis factor beta.

Keywords: genome-wide association study, glioblastoma, inflammatory cytokines, Mendelian randomization, tumor microenvironment

1. Introduction

Glioblastoma (GBM), a grade instrumental variable (IV) tumor, is the most common and aggressive brain tumor. It originates from glial cells of the central nervous system and accounts for approximately 49% of malignant brain tumors.[1] Despite advances in surgical resection, radiotherapy, chemotherapy, and tumor treatment (TTFields), the prognosis for patients with GBM remains extremely poor,[2] with a nearly 100% recurrence rate and a median overall survival of approximately 20 months.[3] Only a limited number of patients are capable of surviving beyond the age of 5 years.[4] Radiation exposure and some rare familial cancer syndromes such as neurofibromatosis type 1,[5] Lynch syndrome,[6] and Li-Fraumeni syndrome,[7] are associated with an increased risk of GBM. However, the exact cause of GBM remains unclear. Early identification and prompt treatment are crucial for improving the outcomes of patients diagnosed with GBM. The medical community continues to prioritize research on the prevention of GBM to increase survival

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The datasets generated during and/or analyzed during the current study are publicly available.

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rates and enhance the quality of life of individuals afflicted by GBM.

Increasing evidence suggests that inflammatory cytokines play a crucial role in the development and progression of tumors and have a significant impact on the host antitumor response.[8,9] These proteins can be categorized into different groups based on their roles, such as interleukins, interferons, chemokines, the tumor necrosis factor superfamily, colony-stimulating factors, and growth factors. Understanding the intricate connection between inflammatory cytokines and cancer has opened new avenues for investigation and potential therapeutic approaches. Given the significant impact of inflammatory cytokines on tumor development and the host antitumor response, there is increasing interest in targeting these molecules as a potential strategy for cancer treatment. Researchers are exploring ways to block pro-tumor cytokines and enhance the antitumor immune response. Moreover, some studies have demonstrated that altering the levels of certain cytokines within the tumor microenvironment can enhance the efficacy of existing anticancer therapies.

Mendelian randomization (MR) is a powerful epidemiological research method that can provide valuable insights into causal relationships between exposures and outcomes by using genetic variants as IVs.[10,11] The use of genetic variation as a proxy for exposure in MR circumvents the limitations of traditional observational studies, allowing researchers to assess the causal effects of an exposure on an outcome without the confusion caused by reverse causality or residual confusion.[12]

In this study, we extracted validated genetic variants from 2 published genome-wide association study (GWAS) summary datasets of 41 inflammatory cytokines[13] and 91 inflammatory cytokines.[14] We then performed MR analyses to test whether a range of circulating inflammatory cytokines could be causally associated with the risk of GBM (FinnGen R10) at onset. If MR studies identify certain inflammatory cytokines as having a causal relationship with GBM, this may help to develop new prevention and treatment strategies.

2. Methods

2.1. Study design

In our study, a bidirectional two-sample MR analysis was conducted to evaluate the causal relationships between 132 inflammatory cytokines and GBM, based on summary-level datasets from large-scale GWASs.

We used a two-sample MR analysis to evaluate the causal relationships between 132 circulating inflammatory cytokines and GBM. In the forward analysis, we used 132 inflammatory cytokines from the 2 databases as exposures and GBM as the outcome to explore the possibility that different inflammatory cytokines cause GBM. We evaluated the causal relationship between GBM and each inflammatory cytokine using a reverse analysis. Single nucleotide polymorphisms (SNPs) significantly associated with exposure were used as IVs.[15,16] The selected IVs must satisfy 3 fundamental assumptions.[17]

(1) Relevance assumption: IVs are strongly correlated with exposure.
(2) Independent assumption: IVs were not associated with any known confounders identified based on established biological and epidemiological knowledge regarding potential factors influencing the relationship between 132 inflammatory cytokines and GBM.
(3) Exclusion restriction assumption: There is no correlation between IVs and outcomes, except for their possible connection with exposure.

The study design is illustrated in Fig. 1. We strictly adhered to the recommendations outlined in the Strengthening the Reporting of Observational Studies in Epidemiology Mendelian Randomization (STROBE-MR) framework (Table S1, Supplemental Digital Content, https://links.lww.com/MD/O1000).[18]

2.2. Data sources

All IVs utilized in this study were derived from the summaries of GWASs. The data on 41 inflammatory cytokines were derived from a meta-analysis including 8293 Finnish individuals from 3 separate population-based cohorts [the Cardiovascular Risk in Young Finns Study (YFS), FINRISK 1997, and FINRISK 2002], which was published in 2017 by Ahola-Olli et al.[13] The University of Bristol provides publicly available GWAS summary statistics for each inflammatory cytokine (https://data.bris.ac.uk/data/dataset). GWAS summary statistics can also be downloaded from the IEU Open GWAS (accession numbers from ebi-a-GCST004420 to ebi-a-GCST004460) at https://gwas.mrcieu.ac.uk (Table S2, Supplemental Digital Content, https://links.lww.com/MD/O1000). The 91 inflammatory cytokines under investigation were derived from a meta-analysis involving 11 cohorts of 14,824 participants of European ancestry published in 2023 by Jing Hua Zhao et al.[14] The complete GWAS summary statistics for each cytokine were obtained from the OpenGWAS database (https://www.ebi.ac.uk/gwas/) under accession numbers GCST90274758-GCST90274848 (Table S3, Supplemental Digital Content, https://links.lww.com/MD/O1000). The FinnGen consortium provided summary statistics for GBM at the genus level (https://finngen.gitbook.io/documentation/, Documentation for the R10 release). This study included 314,193 European individuals, of whom 253 were patients with GBM and 314,446 were controls. Population selection ensured the absence of overlap between the exposure and outcome datasets. The details of the GWASs are summarized in Table 1.

2.3. Instrument selection

To ensure that SNPs selected as IVs were strongly associated with exposure, we implemented the following steps based on the 3 core assumptions of the MR analysis.

First, the genome-wide significance criterion of $P < 5 \times 10^{-8}$ was employed to identify SNPs significantly linked to both GBM and inflammatory cytokines. As only a few SNPs were found for certain inflammatory cytokines and GBM when they were considered for exposure, we adopted a higher cutoff ($P < 1 \times 10^{-5}$).[19]

Second, SNPs that showed potential linkage disequilibrium (LD) with an r^2 value of ≥0.001 and an LD distance of <10,000 kb were excluded to ensure the independence of SNPs using the clustering algorithm in the PLINK software (version v1.90) (https://www.cog-genomics.org/plink/1.9/).

Third, the LDtrait website (https://ldlink.nih.gov/?tab=ldtrait) was used to identify disease-related SNPs and to exclude SNPs associated with potential confounders.[20]

Finally, we calculated the F-statistics for each SNP using the established formula.[21] The R^2 value, which is indicative of the proportion of variance in the exposure variable explained by IV, was derived using the following formula:

$$R^2 = 2 \times EAF \times (1 - EAF) \times \beta^2$$

The F-statistic, which further assesses the strength of the association between SNP and exposure, was calculated as follows:

$$F = \frac{R^2 \times (N - 2)}{1 - R^2}$$

Here, EAF denotes the effect allele frequency, β is the effect size of the association between the SNP and inflammatory cytokine, and N is the sample size of the GWAS from which

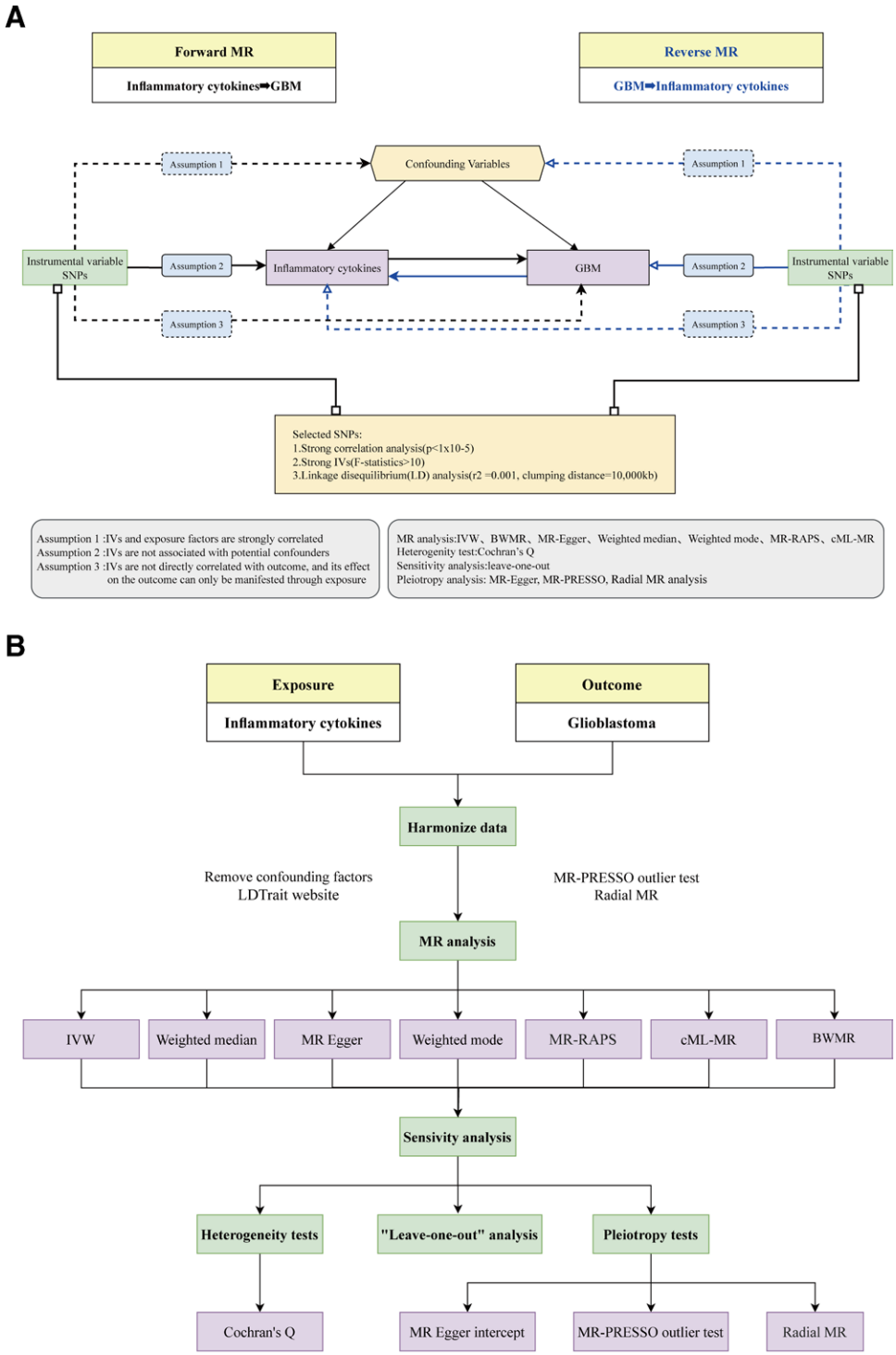


Figure 1. The study design of our study. (A) Three instrumental variable assumptions for Mendelian Randomization analysis; (B) the study design of two sample MR analysis.

Table 1

Baseline information for inflammatory cytokines and glioblastoma.

Traits	Catlog ID	Population	Year	Author	PMID	DOI
91 inflammatory proteins	GCST90274758~GCST90274848	European	2023	Zhao Jing Hua	37563310	10.1038/s41590-023-01588-w
41 inflammatory proteins	ebi-a-GCST004420~ebi-a-GCST004460	European	2021	Marita Kalaoja	33491305	10.1002/oby.23060
Phenotype	**Consortium**	**Population**	**Year**	**Cases**	**Sample size**	**Websites**
Brain glioblastoma	FinnGen (R10)	European	2023	253	314,446	https://www.finngen.fi/en

the SNPs were derived. SNPs with an F-statistic exceeding the threshold of 10 were deemed to have a strong association with exposure and, therefore, qualified as suitable IVs for MR analysis. We further refined our selection by conducting a Steiger test after excluding weak IVs.

2.4. MR analysis and Bayesian weighted Mendelian randomization analysis

In our investigation of the genetic causality between inflammatory cytokines and GBM based on GWAS data, we conducted a two-sample MR analysis using a variety of commonly employed MR methodologies. The MR–Egger,[22] weighted median,[23] inverse variance weighted (IVW),[24,25] weighted mode,[26] constrained maximum likelihood-based Mendelian randomization[27] and robust adjusted profile score[28] models were used. The IVW method, which is known for its efficiency and high statistical power, was used to evaluate causal associations. The IVW findings were considered to be suggestive of a meaningful association if they achieved statistical significance ($P < .05$) and were supported by a consistent trend across other approaches in instances where other methods, such as the weighted median method and the MR-Egger method, failed to produce meaningful outcomes.

Bayesian weighted Mendelian randomization (BWMR) analysis serves as a valuable complement to traditional two-sample MR analysis. BWMR analysis enhances the stability and reliability of the final results using a Bayesian framework. Moreover, BWMR analysis enhances the conventional two-sample MR by providing a more robust estimation in the presence of heterogeneity and potential violations of the assumptions underpinning the IV analysis.[29]

By employing a comprehensive array of MR methods, we assessed the genetic causality between inflammatory cytokines and GBM from multiple perspectives, ensuring that our research findings are both accurate and robust.

2.5. Sensitivity analyses

The Cochrane Q test was used to quantitatively evaluate heterogeneity among the Cochrane Q test ($P < .05$).[25] Furthermore, we applied the MR-Egger intercept test ($P < .05$) to examine whether there was evidence of directional pleiotropy within the IVs.[22] A significant deviation in the intercept from zero indicates directional pleiotropy.

Furthermore, we employed the MR-Pleiotropy Residual Sum and Outlier method[30] to detect and eliminate outliers that could introduce horizontal pleiotropy. In the event that outliers were identified, the SNPs associated with them were excluded from subsequent analyses.

Our research utilized an innovative methodology by utilizing modified second-order weights to identify and subsequently eliminate outliers in the MR analysis. This was accomplished by utilizing the "RadialMR" package (https://github.com/WSpiller/RadialMR) in the R programming environment. This method improves the robustness of the MR analysis by minimizing the potential impact of outliers, which could otherwise skew the results and lead to biased estimates of the causal effect.[31]

To identify potentially heterogeneous SNPs, we conducted leave-one-out sensitivity analysis. This approach involved systematically removing each SNP from the analysis and observing its impact on the overall results.

2.6. Meta-analysis

Finally, we evaluated the same inflammatory cytokine results from both databases after screening based on the "meta" package to confirm the reliability of the MR results. A random-effects model was used when the heterogeneity of the results was significant ($I^2 \geq 50\%$ or $P < .05$), and a fixed-effects model was used when the results were not significant ($I^2 < 50\%$ and $P \geq .05$).[19,32]

All the statistical analyses were performed using R version 4.3.3 (https://www.r-project.org/), along with the "TwoSampleMR," "RadialMR," and "MRPRESSO" packages. The data were visualized using the forestploter R package. The outcomes of the MR analysis were quantified using odds ratios with 95% confidence intervals, and a P value <.05 was considered to indicate statistical significance.

2.7. Ethics statement

Our research relied entirely on publicly available GWAS data that had already received ethical approval prior to its release. Because this study did not involve the collection of new data, it was not necessary to obtain additional ethical approval.

3. Results

3.1. The causal effect of inflammatory proteins on GBM

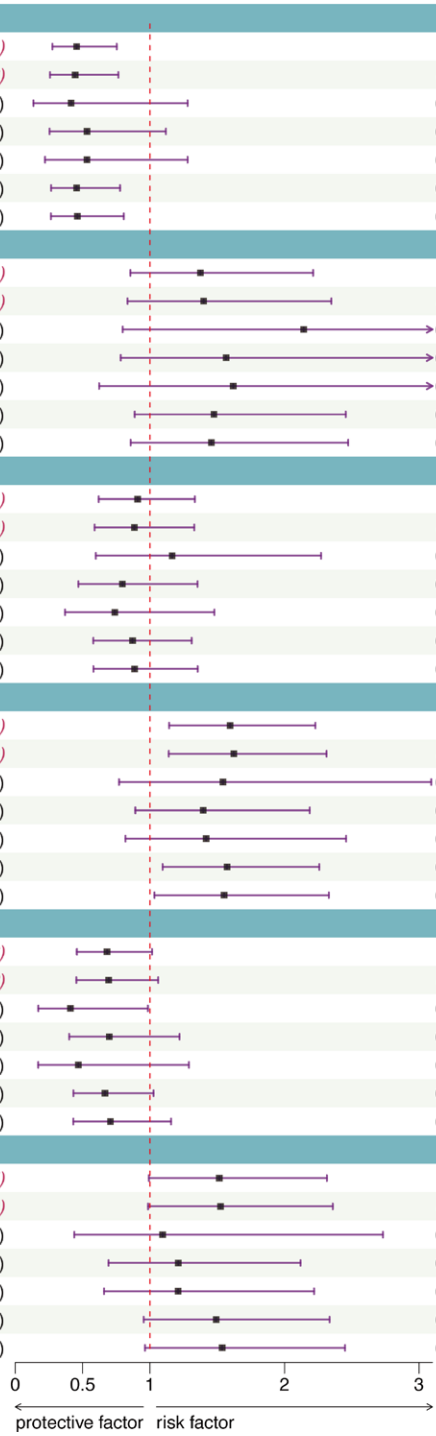
3.1.1. Selection of IVs. After rigorous selection and harmonization of IVs, 2330 SNPs linked to 91 inflammatory cytokines and 561 SNPs linked to 41 inflammatory cytokines were used as instruments for subsequent analysis, with all computed F-statistics above 10. Each selected IV had an F-statistic > 10, indicating strong IVs and absence of weak instrumental bias in the study (Tables S4 and S5, Supplemental Digital Content, https://links.lww.com/MD/O1000).

3.1.2. MR analysis and BWMR analysis. MR analysis of 132 inflammatory cytokines as exposure variables in GBM revealed 2 inflammatory proteins with a causal relationship with GBM (Tables S6 and S7, Supplemental Digital Content, https://links.lww.com/MD/O1000). The primary results of the main MR analyses are shown in Figs. 2–4.

According to the IVW results, elevated levels of fibroblast growth factor 21 (odds ratio [OR] = 0.456, 95% CI: 0.276–0.754, $P = .002$) were associated with a reduced risk of GBM. In addition, increased levels of tumor necrosis factor β (OR = 1.597, 95% CI: 1.143–2.230, $P = .006$) may be linked to an increased risk of GBM. This result was corroborated by the BWMR analysis results for fibroblast growth factor 21 (FGF21) (OR = 0.444, 95% CI: 0.258–0.766, $P = .004$) and tumor necrosis factor beta (TNF-β) (OR = 1.624, 95% CI: 1.140–2.313, $P = .007$). Other supplementary methods for MR analysis corroborated similar trends and findings related to the impact of these inflammatory proteins on GBM.

3.1.3. Sensitivity analysis. In this study, a series of sensitivity analyses was conducted to evaluate the pleiotropy and heterogeneity of the MR results. Initially, Cochran Q test revealed no significant heterogeneity among the effect sizes of the included SNPs across the different studies, indicating good consistency in our analytical outcomes (Table 2). This implies that our results are uniform, thus enhancing the credibility of our findings. Subsequently, the MR-Egger intercept test was used to assess the presence of directional gene pleiotropy. The results did not indicate significant pleiotropy, reinforcing the notion that our IVs were generally unrelated to potential confounders, thereby supporting the trustworthiness of our study's results (Tables S8 and S9, Supplemental Digital Content, https://links.lww.com/MD/O1000). Furthermore, utilizing the MR-Pleiotropy Residual Sum and Outlier method and Radial MR analysis, we identified 142 outliers within our dataset (Tables S10 and S11, Supplemental Digital Content, https://links.lww.com/MD/O1000). To ensure the robustness and reliability of our MR analysis, we excluded these outliers

GWAS ID	Exposuer/Method	nSNP	b	se	OR(95% CI)	pval
GCST90274788	FGF21					
	IVW	*25*	*−0.786*	*0.257*	*0.456 (0.276 to 0.754)*	*0.002*
	BWMR	*25*	*−0.811*	*0.278*	*0.444 (0.258 to 0.766)*	*0.004*
	MR Egger	25	−0.88	0.575	0.415 (0.134 to 1.280)	0.140
	Weighted median	25	−0.629	0.378	0.533 (0.254 to 1.119)	0.096
	Weighted mode	25	−0.63	0.447	0.533 (0.222 to 1.280)	0.172
	RAPS	25	−0.787	0.274	0.455 (0.266 to 0.778)	0.004
	CML MR	25	−0.773	0.284	0.462 (0.264 to 0.806)	0.007
GCST90274795	IL−10					
	IVW	*30*	*0.319*	*0.242*	*1.376 (0.856 to 2.213)*	*0.188*
	BWMR	*30*	*0.336*	*0.264*	*1.400 (0.834 to 2.349)*	*0.203*
	MR Egger	30	0.762	0.504	2.143 (0.797 to 5.760)	0.142
	Weighted median	30	0.449	0.354	1.566 (0.783 to 3.131)	0.205
	Weighted mode	30	0.482	0.487	1.619 (0.623 to 4.210)	0.331
	RAPS	30	0.389	0.26	1.476 (0.887 to 2.455)	0.134
	CML MR	30	0.376	0.27	1.457 (0.858 to 2.475)	0.164
GCST90274821	MIP 1a					
	IVW	*29*	*−0.095*	*0.195*	*0.909 (0.620 to 1.334)*	*0.626*
	BWMR	*29*	*−0.122*	*0.208*	*0.885 (0.589 to 1.330)*	*0.556*
	MR Egger	29	0.154	0.341	1.166 (0.598 to 2.273)	0.656
	Weighted median	29	−0.228	0.271	0.796 (0.468 to 1.354)	0.400
	Weighted mode	29	−0.302	0.354	0.739 (0.369 to 1.480)	0.401
	RAPS	29	−0.138	0.208	0.871 (0.579 to 1.311)	0.509
	CML MR	29	−0.121	0.217	0.886 (0.580 to 1.355)	0.577
ebi−a−GCST004425	TGF beta					
	IVW	*8*	*0.468*	*0.17*	*1.597 (1.143 to 2.230)*	*0.006*
	BWMR	*8*	*0.485*	*0.18*	*1.624 (1.140 to 2.313)*	*0.007*
	MR Egger	8	0.434	0.354	1.544 (0.771 to 3.091)	0.266
	Weighted median	8	0.334	0.229	1.397 (0.892 to 2.187)	0.144
	Weighted mode	8	0.35	0.281	1.419 (0.819 to 2.459)	0.253
	RAPS	8	0.453	0.185	1.573 (1.095 to 2.259)	0.014
	CML MR	8	0.44	0.207	1.552 (1.034 to 2.329)	0.034
ebi−a−GCST004434	MIP 1a					
	IVW	*15*	*−0.383*	*0.203*	*0.682 (0.458 to 1.016)*	*0.060*
	BWMR	*15*	*−0.365*	*0.217*	*0.694 (0.454 to 1.062)*	*0.092*
	MR Egger	15	−0.89	0.447	0.410 (0.171 to 0.985)	0.068
	Weighted median	15	−0.358	0.284	0.699 (0.400 to 1.220)	0.208
	Weighted mode	15	−0.758	0.517	0.469 (0.170 to 1.290)	0.165
	RAPS	15	−0.406	0.221	0.666 (0.432 to 1.027)	0.066
	CML MR	15	−0.347	0.252	0.707 (0.431 to 1.158)	0.168
ebi−a−GCST004444	IL−10					
	IVW	*15*	*0.416*	*0.216*	*1.516 (0.992 to 2.315)*	*0.054*
	BWMR	*15*	*0.422*	*0.222*	*1.526 (0.987 to 2.359)*	*0.057*
	MR Egger	15	0.09	0.467	1.095 (0.439 to 2.732)	0.850
	Weighted median	15	0.192	0.285	1.211 (0.692 to 2.120)	0.502
	Weighted mode	15	0.191	0.31	1.210 (0.660 to 2.220)	0.547
	RAPS	15	0.401	0.228	1.493 (0.954 to 2.335)	0.079
	CML MR	15	0.43	0.238	1.537 (0.964 to 2.450)	0.071



pval<0.05 was considered statistically significant

Figure 2. Forest plot for the significant associations. We mainly used the IVW method and BWMR. BWMR = Bayesian weighted Mendelian randomization; cML-MR = constrained maximum likelihood-based Mendelian randomization; FGF21 = fibroblast growth factor 21; IL10 = interleukin-10; IVW = inverse variance weighted; MIP 1a = macrophage inflammatory protein 1a; MR = Mendelian randomization; MRPresso = MR-pleiotropy residual sum and outliers; RAPS = Robust adjusted profile score; TGF beta = tumor necrosis factor β.

and subsequently reevaluated the genetic IVs in relation to the exposure and outcome variables. For example, when we investigated the causal link between interleukin-10 (IL-10) (from the EBI database) and GBM, the radial MR method successfully identified an outlier (SNP: rs4655953) (Fig. 5). The advantage of radial MR analysis lies in its ability to adapt to complex patterns in data and provide a flexible way to address pleiotropy issues, thereby enhancing the reliability and accuracy of our results. In addition, an approximately symmetric funnel plot corroborated our findings, suggesting the robustness of our results. Finally, we performed a leave-one-out analysis of the data, and no specific SNPs were found to drive the association between inflammatory cytokines and GBM. The results of the analysis are presented as a forest plot (Fig. 6).

3.1.4. Meta-analysis. To further minimize the bias of the results due to different data sources, we performed a meta-analysis of the results of the same inflammatory cytokines

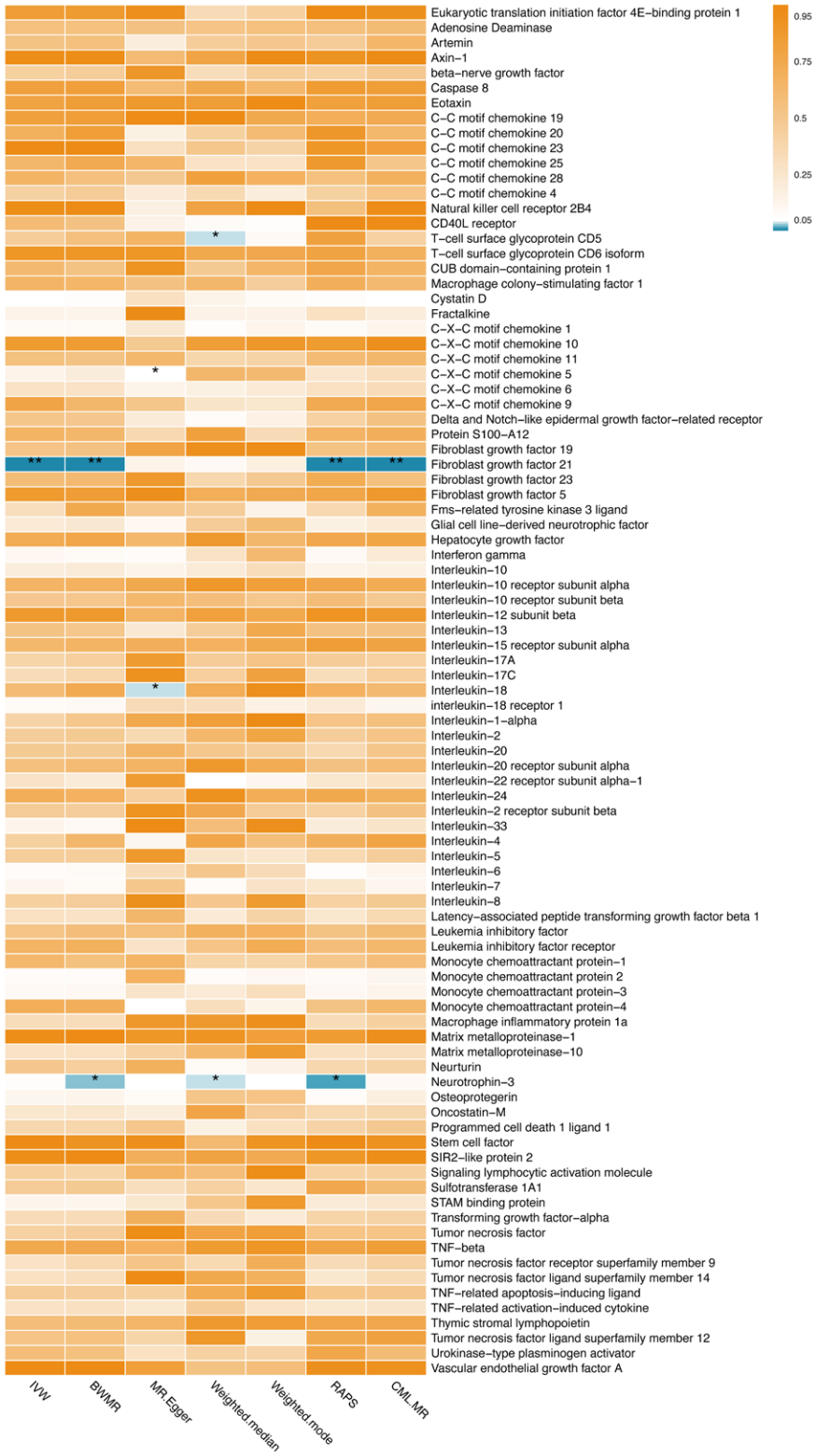


Figure 3. Heatmap of Mendelian randomization analysis of the causal effect of 91 inflammatory cytokines on glioblastoma. ***$P < .001$; **$0.001 \le P < .01$; *$0.01 \le P < .05$. BWMR = Bayesian weighted Mendelian randomization; cML-MR = constrained maximum likelihood-based Mendelian randomization; IVW = inverse variance weighted; RAPS = robust adjusted profile score.

from different database sources. Finally, 2 inflammatory cytokines, IL-10 (OR = 1.452, 95% CI: 1.059–1.992, $P = .021$) and MIP-1a (OR = 0.743, 95% CI: 0.558–0.990, $P = .042$), were screened out. The results are shown as forest plots (Fig. 7).

3.2. The causal effect of GBM on inflammatory proteins

To further investigate the reverse causal effects of GBM on 132 inflammatory cytokines, we performed reverse MR analysis conducted primarily using the IVW method. In this analysis, we identified only 8 SNPs that demonstrated a robust association

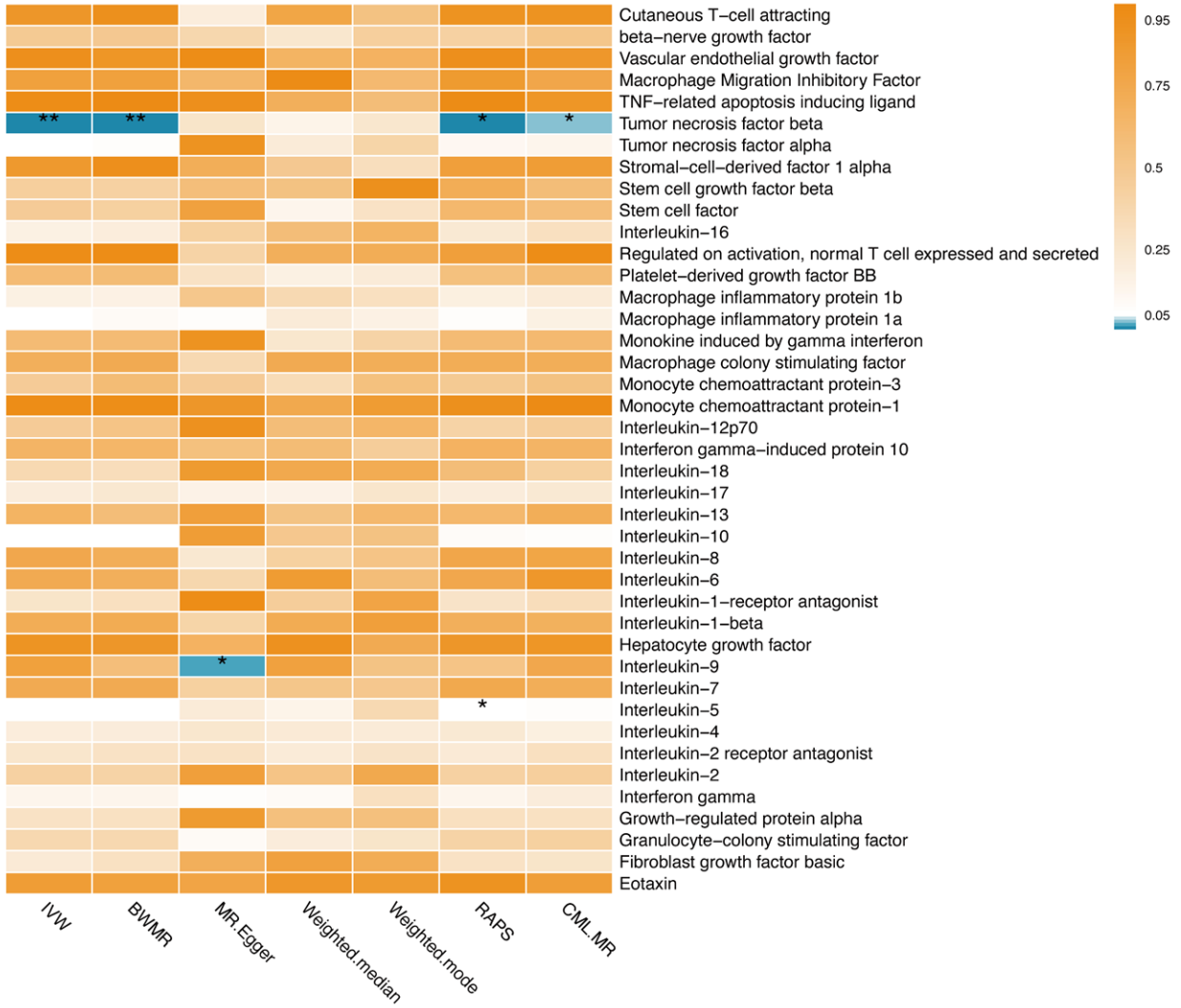


Figure 4. Heatmap of Mendelian randomization analysis of the causal effect of 41 inflammatory cytokines on glioblastoma. ***$P < .001$; **$0.001 \le P < .01$; *$0.01 \le P < .05$. BWMR = Bayesian weighted Mendelian randomization; cML-MR = constrained maximum likelihood-based Mendelian randomization; IVW = inverse variance weighted; RAPS = robust adjusted profile score.

Table 2

Heterogeneity and pleiotropy analyses.

	Heterogeneity test				Horizontal pleiotropy test				MRPresso	
	Cochrane Q				MR-Egger intercept		MR-PRESSO global		MR analysis	
Exposure	method	Q	df	*P*	Intercept	*P*	RSSobs	*P*	Causal Estimate	*P*
FGF21	MR Egger	21.424	23	.555	0.010	.856	23.073	.646	-0.786	.004
GCST90274788	IVW	21.457	24	.612						
IL10	MR Egger	24.892	28	.634	-0.052	.325	27.479	.655	0.319	.174
GCST90274795	IVW	25.896	29	.631						
MIP 1a	MR Egger	18.715	27	.880	-0.040	.380	21.172	.876	-0.095	.564
GCST90274821	IVW	19.510	28	.882						
TGFβ	MR Egger	6.971	6	.324	0.010	.915	8.635	.514	0.468	.029
ebi-a-GCST004425	IVW	6.986	7	.430						
MIP 1a	MR Egger	10.929	13	.617	0.092	.224	14.432	.582	-0.383	.067
ebi-a-GCST004434	IVW	12.557	14	.562						
IL10	MR Egger	11.365	13	.580	0.044	.445	14.319	.615	0.416	.056
ebi-a-GCST004444	IVW	11.984	14	.608						

FGF21 = fibroblast growth factor 21, IL10 = interleukin-10, IVW = inverse variance weighted, MIP 1a = macrophage inflammatory protein 1a, MR = Mendelian randomization, MRPresso = MR-Pleiotropy Residual Sum and Outliers, TGFβ = tumor necrosis factor β.

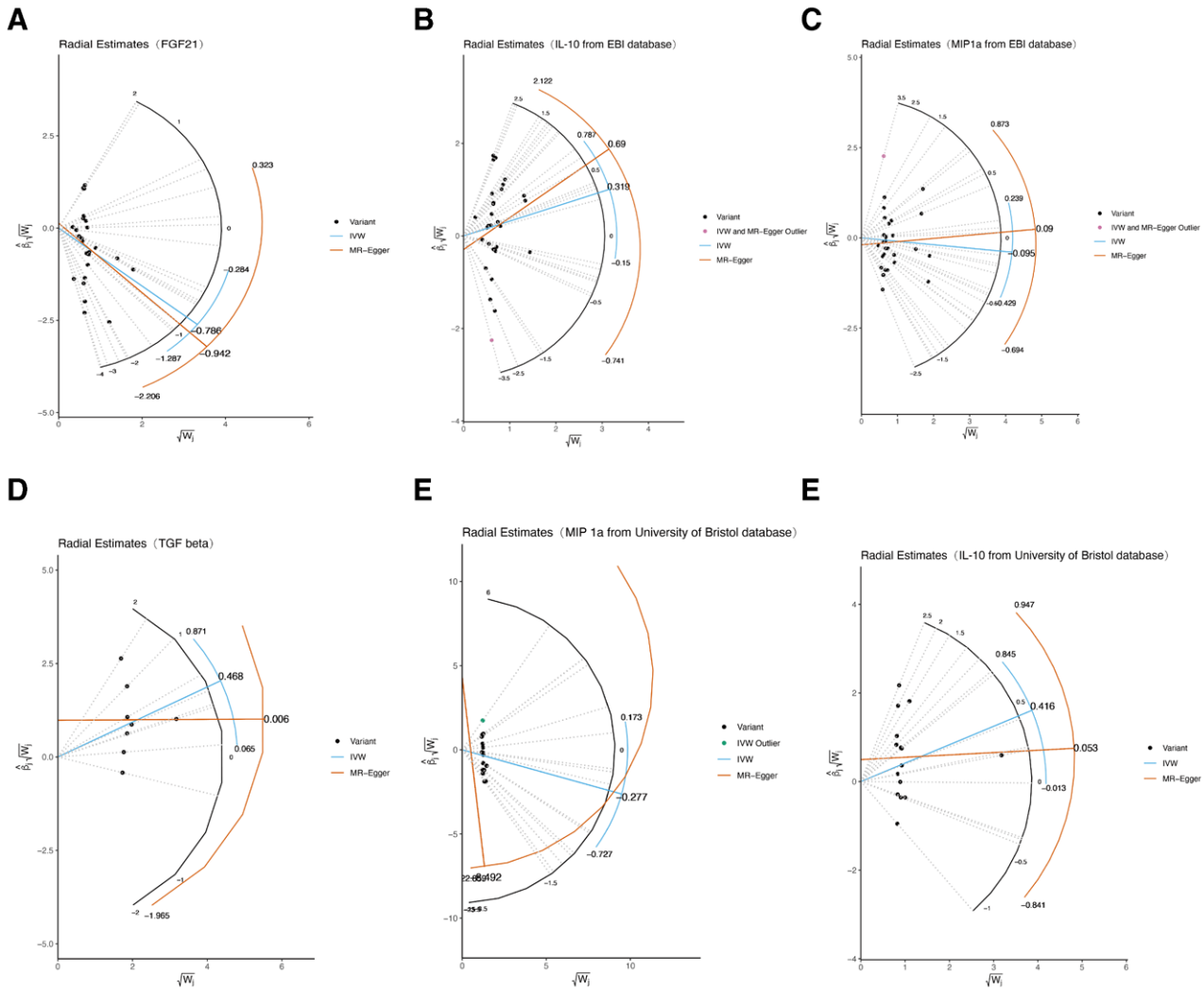


Figure 5. Outliers identified by the radial MR analysis. (A) Fibroblast growth factor 21 on glioblastoma; (B) interleukin-10 on glioblastoma (91 cytokines); (C) macrophage inflammatory protein 1a on glioblastoma (91 cytokines); (D) tumor necrosis factor β on glioblastoma; (E) macrophage inflammatory protein 1a on glioblastoma (41 cytokines); (F) interleukin-10 on glioblastoma (41 cytokines). The purple portions signify the identified outliers by IVW and MR-Egger. The green portions signify the identified outliers by IVW.

with GBM. Only 90 inflammatory cytokines (from the EBI database) and 28 inflammatory cytokines (from the University of Bristol database) were successfully detected via MR analysis using the IVW method (Tables S12 and S13, Supplemental Digital Content, https://links.lww.com/MD/O1000). Ultimately, our findings did not provide evidence that GBM significantly influences the levels of these cytokines (Figs. 8 and 9).

4. Discussion

In this study, we explored the relationship between inflammatory cytokines and GBM based on genetic data from publicly available databases using MR analysis. Understanding the causal link between the levels of inflammatory cytokines and GBM is crucial for determining the underlying causes of this disease and for devising effective preventative and curative strategies. Further analysis revealed that, among the 132 inflammatory cytokines, low levels of FGF21 and high levels of TNF-β were associated with a greater risk of GBM. In addition, the results of the meta-analysis indicated that 2 additional inflammatory cytokines were associated with the risk of GBM. Low levels of MIP-1a and high levels of IL-10 correlated with a greater likelihood of developing GBM. In the reverse analysis, no causal relationship between GBM and inflammatory cytokines was found.

The tumor microenvironment plays a crucial role in cancer initiation and progression.[33] Inflammatory cytokines, which are key components of the tumor microenvironment, are produced by various cell types, including tumor cells, immune cells, and stromal cells.[34] Dysregulation of these genes can lead to chronic inflammation, which can either promote[35] or inhibit tumor growth. Understanding the complex interplay between inflammatory cytokines and the tumor microenvironment is essential for developing targeted therapies that modulate the microenvironment and improve patient outcomes.[36] MR studies have been applied to investigate the causal relationships between inflammatory cytokines and various cancers, including prostate cancer, breast cancer, colorectal cancer, lung cancer, and diffuse large B-cell lymphoma.[37–40]

FGF21 is a multifaceted hormone with a complex role in the regulation of metabolism, energy expenditure, and glucose homeostasis.[41] However, its role in cancer remains complex and context dependent.[42] Several recent studies have shown that FGF21 is associated with the development and progression of cancers, such as liver cancer,[43–46] lung cancer,[47] thyroid cancer,[48] and ovarian cancers.[49] For example, in prostate cancer,

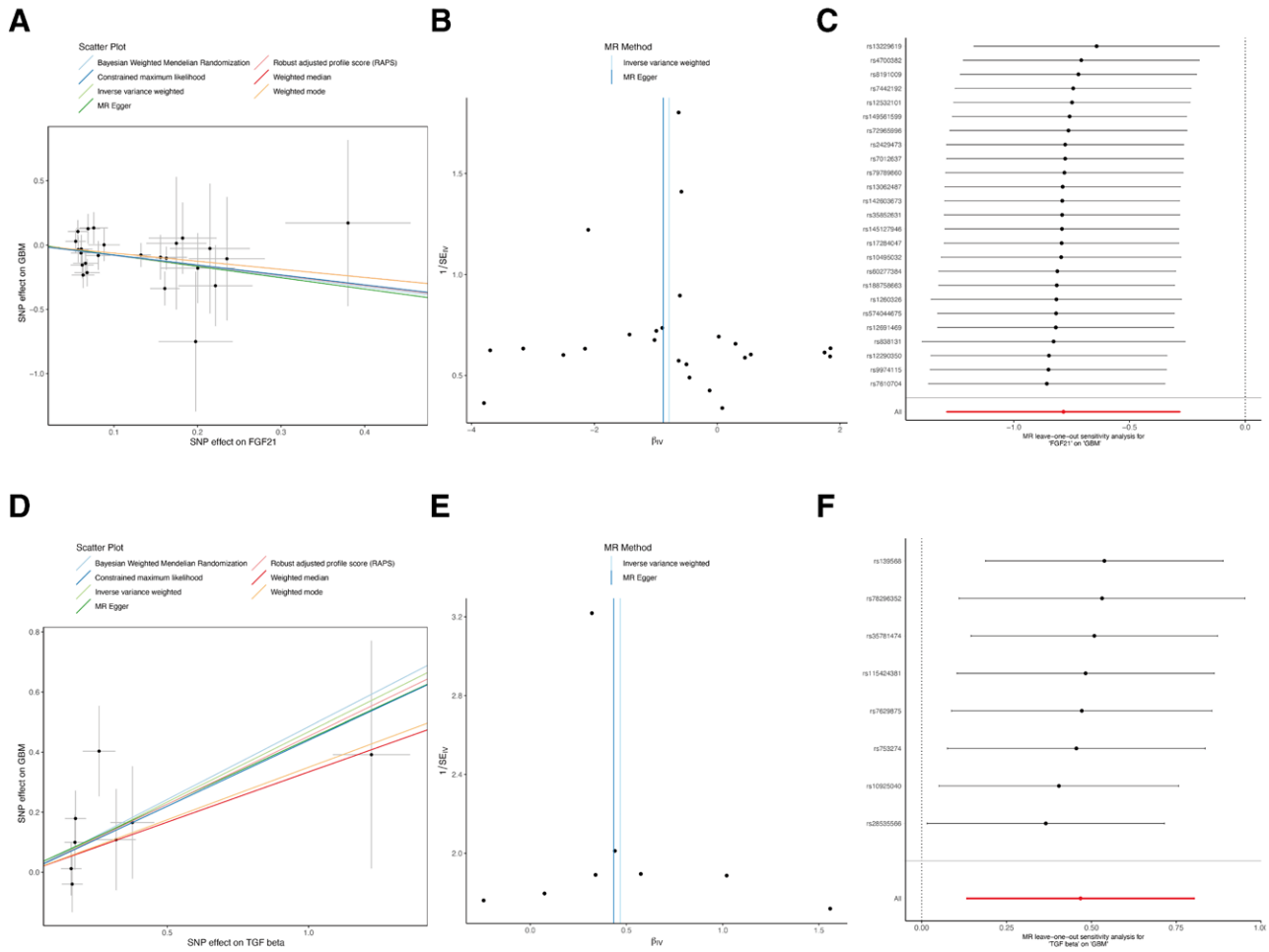


Figure 6. Results of the causal relationship between fibroblast growth factor 21, tumor necrosis factor β and glioblastoma. (A) Scatter plots of the causal relationship between fibroblast growth factor 21 and glioblastoma at using 7 different MR methods. (B) Funnel plots of fibroblast growth factor 21 on glioblastoma; (C) leave-one-out sensitivity analysis of fibroblast growth factor 21 on glioblastoma; (D) scatter plots of the causal relationship between tumor necrosis factor β and glioblastoma at using 7 different MR methods; (E) funnel plots of tumor necrosis factor β on glioblastoma; (F) leave-one-out sensitivity analysis of tumor necrosis factor β on glioblastoma.

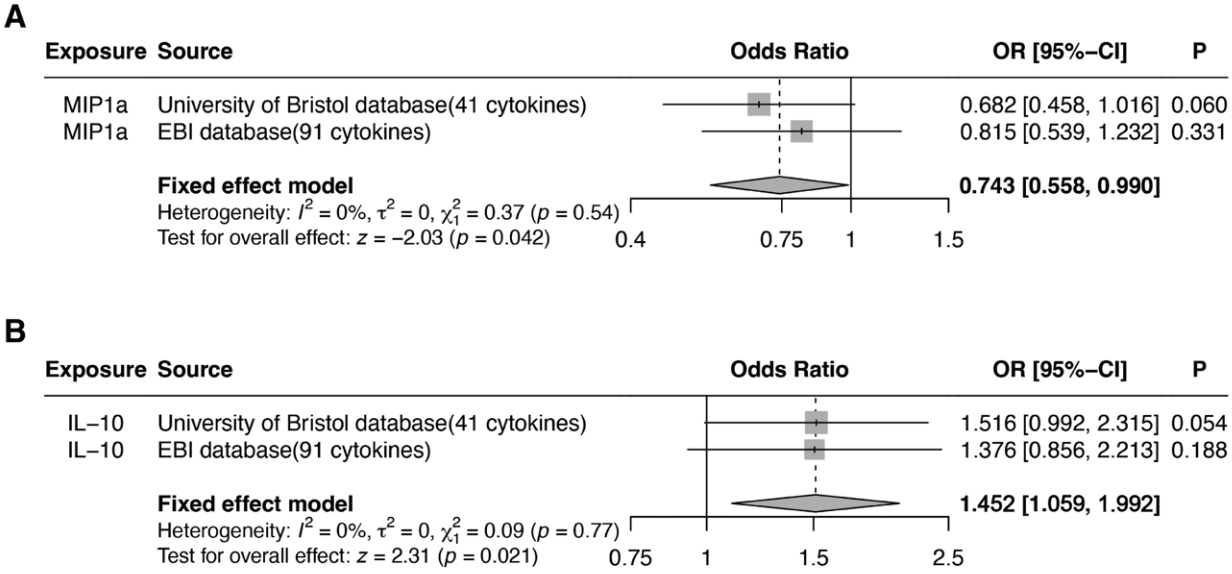
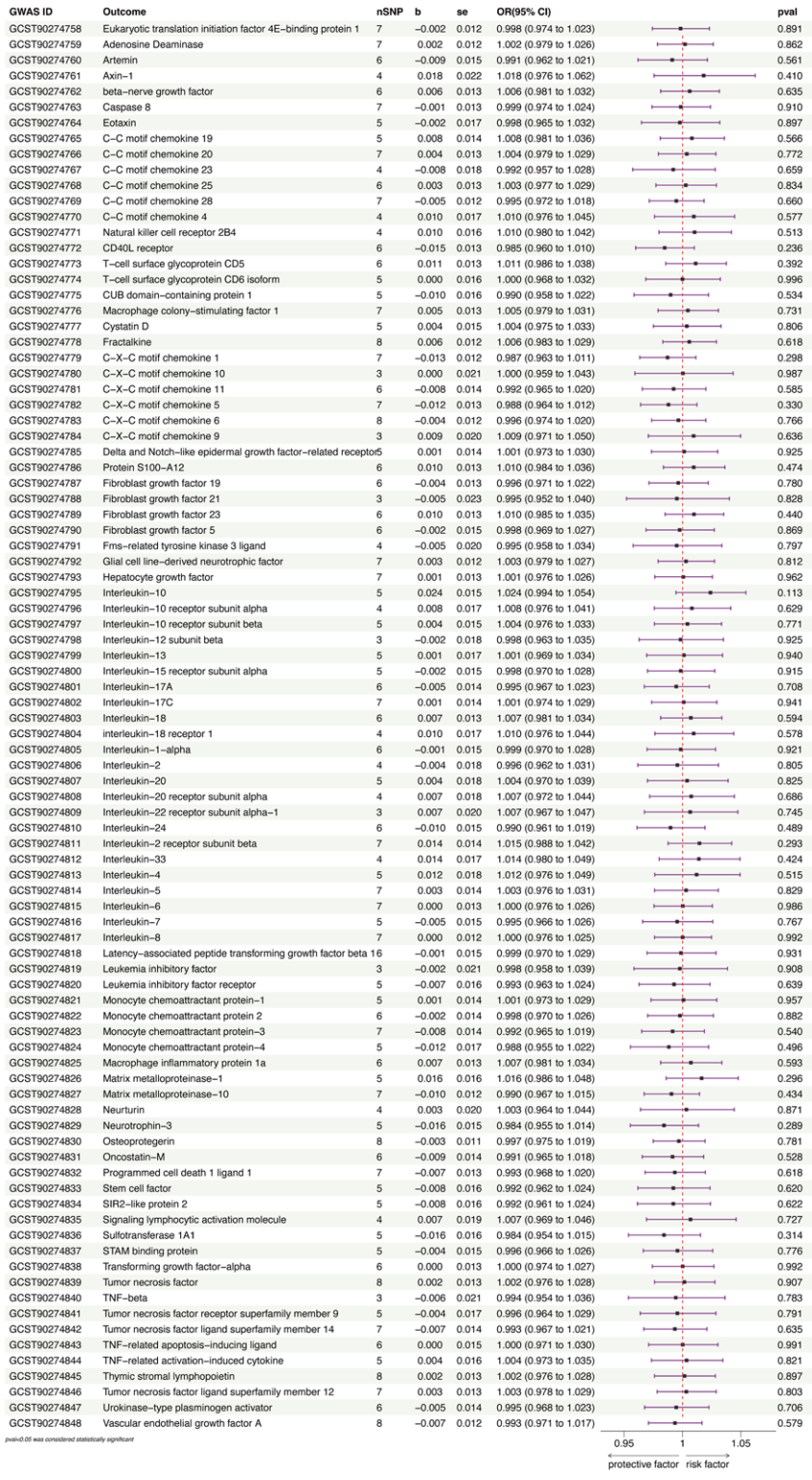


Figure 7. The forest plot shows the 2 sets of results from the analysis of inflammatory cytokines on glioblastoma using meta-analysis methods combined to assess the reliability of positive or potentially positive results. (A) Macrophage inflammatory protein 1a on glioblastoma. (B) Interleukin-10 on glioblastoma.



GWAS ID	Outcome	nSNP	b	se	OR(95% CI)	pval
GCST90274758	Eukaryotic translation initiation factor 4E–binding protein 1	7	−0.002	0.012	0.998 (0.974 to 1.023)	0.891
GCST90274759	Adenosine Deaminase	7	0.002	0.012	1.002 (0.979 to 1.026)	0.862
GCST90274760	Artemin	6	−0.009	0.015	0.991 (0.962 to 1.021)	0.561
GCST90274761	Axin–1	4	0.018	0.022	1.018 (0.976 to 1.062)	0.410
GCST90274762	beta–nerve growth factor	6	0.006	0.013	1.006 (0.981 to 1.032)	0.635
GCST90274763	Caspase 8	7	−0.001	0.013	0.999 (0.974 to 1.024)	0.910
GCST90274764	Eotaxin	5	−0.002	0.017	0.998 (0.965 to 1.032)	0.897
GCST90274765	C–C motif chemokine 19	5	0.008	0.014	1.008 (0.981 to 1.036)	0.566
GCST90274766	C–C motif chemokine 20	7	0.004	0.013	1.004 (0.979 to 1.029)	0.772
GCST90274767	C–C motif chemokine 23	4	−0.008	0.018	0.992 (0.957 to 1.028)	0.659
GCST90274768	C–C motif chemokine 25	6	0.003	0.013	1.003 (0.977 to 1.029)	0.834
GCST90274769	C–C motif chemokine 28	7	−0.005	0.012	0.995 (0.972 to 1.018)	0.660
GCST90274770	C–C motif chemokine 4	4	0.010	0.017	1.010 (0.976 to 1.045)	0.577
GCST90274771	Natural killer cell receptor 2B4	4	0.010	0.016	1.010 (0.980 to 1.042)	0.513
GCST90274772	CD40L receptor	6	−0.015	0.013	0.985 (0.960 to 1.010)	0.236
GCST90274773	T–cell surface glycoprotein CD5	6	0.011	0.013	1.011 (0.986 to 1.038)	0.392
GCST90274774	T–cell surface glycoprotein CD6 isoform	5	0.000	0.016	1.000 (0.968 to 1.032)	0.996
GCST90274775	CUB domain–containing protein 1	5	−0.010	0.016	0.990 (0.958 to 1.022)	0.534
GCST90274776	Macrophage colony–stimulating factor 1	7	0.005	0.013	1.005 (0.979 to 1.031)	0.731
GCST90274777	Cystatin D	5	0.004	0.015	1.004 (0.975 to 1.033)	0.806
GCST90274778	Fractalkine	8	0.006	0.012	1.006 (0.983 to 1.029)	0.618
GCST90274779	C–X–C motif chemokine 1	7	−0.013	0.012	0.987 (0.963 to 1.011)	0.298
GCST90274780	C–X–C motif chemokine 10	3	0.000	0.021	1.000 (0.959 to 1.043)	0.987
GCST90274781	C–X–C motif chemokine 11	6	−0.008	0.014	0.992 (0.965 to 1.020)	0.585
GCST90274782	C–X–C motif chemokine 5	7	−0.012	0.013	0.988 (0.964 to 1.012)	0.330
GCST90274783	C–X–C motif chemokine 6	8	−0.004	0.012	0.996 (0.974 to 1.020)	0.766
GCST90274784	C–X–C motif chemokine 9	3	0.009	0.020	1.009 (0.971 to 1.050)	0.636
GCST90274785	Delta and Notch–like epidermal growth factor–related receptor	5	0.001	0.014	1.001 (0.973 to 1.030)	0.925
GCST90274786	Protein S100–A12	6	0.010	0.013	1.010 (0.984 to 1.036)	0.474
GCST90274787	Fibroblast growth factor 19	6	−0.004	0.013	0.996 (0.971 to 1.022)	0.780
GCST90274788	Fibroblast growth factor 21	3	−0.005	0.023	0.995 (0.952 to 1.040)	0.828
GCST90274789	Fibroblast growth factor 23	6	0.010	0.013	1.010 (0.985 to 1.035)	0.440
GCST90274790	Fibroblast growth factor 5	6	−0.002	0.015	0.998 (0.969 to 1.027)	0.869
GCST90274791	Fms–related tyrosine kinase 3 ligand	4	−0.005	0.020	0.995 (0.958 to 1.034)	0.797
GCST90274792	Glial cell line–derived neurotrophic factor	7	0.003	0.012	1.003 (0.979 to 1.027)	0.812
GCST90274793	Hepatocyte growth factor	7	0.001	0.013	1.001 (0.976 to 1.026)	0.962
GCST90274795	Interleukin–10	5	0.024	0.015	1.024 (0.994 to 1.054)	0.113
GCST90274796	Interleukin–10 receptor subunit alpha	4	0.008	0.017	1.008 (0.976 to 1.041)	0.629
GCST90274797	Interleukin–10 receptor subunit beta	5	0.004	0.015	1.004 (0.976 to 1.033)	0.771
GCST90274798	Interleukin–12 subunit beta	3	−0.002	0.018	0.998 (0.963 to 1.035)	0.925
GCST90274799	Interleukin–13	5	0.001	0.017	1.001 (0.969 to 1.034)	0.940
GCST90274800	Interleukin–15 receptor subunit alpha	5	−0.002	0.015	0.998 (0.970 to 1.028)	0.915
GCST90274801	Interleukin–17A	6	−0.005	0.014	0.995 (0.967 to 1.023)	0.708
GCST90274802	Interleukin–17C	7	0.001	0.014	1.001 (0.974 to 1.029)	0.941
GCST90274803	Interleukin–18	6	0.007	0.013	1.007 (0.981 to 1.034)	0.594
GCST90274804	interleukin–18 receptor 1	4	0.010	0.017	1.010 (0.976 to 1.044)	0.578
GCST90274805	Interleukin–1–alpha	6	−0.001	0.015	0.999 (0.970 to 1.028)	0.921
GCST90274806	Interleukin–2	4	−0.004	0.018	0.996 (0.962 to 1.031)	0.805
GCST90274807	Interleukin–20	5	0.004	0.018	1.004 (0.970 to 1.039)	0.825
GCST90274808	Interleukin–20 receptor subunit alpha	4	0.007	0.018	1.007 (0.972 to 1.044)	0.686
GCST90274809	Interleukin–22 receptor subunit alpha–1	3	0.007	0.020	1.007 (0.967 to 1.047)	0.745
GCST90274810	Interleukin–24	6	−0.010	0.015	0.990 (0.961 to 1.019)	0.489
GCST90274811	Interleukin–2 receptor subunit beta	7	0.014	0.014	1.015 (0.988 to 1.042)	0.293
GCST90274812	Interleukin–33	4	0.014	0.017	1.014 (0.980 to 1.049)	0.424
GCST90274813	Interleukin–4	5	0.012	0.018	1.012 (0.976 to 1.049)	0.515
GCST90274814	Interleukin–5	7	0.003	0.014	1.003 (0.976 to 1.031)	0.829
GCST90274815	Interleukin–6	7	0.000	0.013	1.000 (0.976 to 1.026)	0.986
GCST90274816	Interleukin–7	5	−0.005	0.015	0.995 (0.966 to 1.026)	0.767
GCST90274817	Interleukin–8	7	0.000	0.012	1.000 (0.976 to 1.025)	0.992
GCST90274818	Latency–associated peptide transforming growth factor beta 1	6	−0.001	0.015	0.999 (0.970 to 1.029)	0.931
GCST90274819	Leukemia inhibitory factor	3	−0.002	0.021	0.998 (0.958 to 1.039)	0.908
GCST90274820	Leukemia inhibitory factor receptor	5	−0.007	0.016	0.993 (0.963 to 1.024)	0.639
GCST90274821	Monocyte chemoattractant protein–1	5	0.001	0.014	1.001 (0.973 to 1.029)	0.957
GCST90274822	Monocyte chemoattractant protein 2	6	−0.002	0.014	0.998 (0.970 to 1.026)	0.882
GCST90274823	Monocyte chemoattractant protein–3	7	−0.008	0.014	0.992 (0.965 to 1.019)	0.540
GCST90274824	Monocyte chemoattractant protein–4	5	−0.012	0.017	0.988 (0.955 to 1.022)	0.496
GCST90274825	Macrophage inflammatory protein 1a	6	0.007	0.013	1.007 (0.981 to 1.034)	0.593
GCST90274826	Matrix metalloproteinase–1	5	0.016	0.016	1.016 (0.986 to 1.048)	0.296
GCST90274827	Matrix metalloproteinase–10	7	−0.010	0.012	0.990 (0.967 to 1.015)	0.434
GCST90274828	Neurturin	4	0.003	0.020	1.003 (0.964 to 1.044)	0.871
GCST90274829	Neurotrophin–3	5	−0.016	0.015	0.984 (0.955 to 1.014)	0.289
GCST90274830	Osteoprotegerin	8	−0.003	0.011	0.997 (0.975 to 1.019)	0.781
GCST90274831	Oncostatin–M	6	−0.009	0.014	0.991 (0.965 to 1.018)	0.528
GCST90274832	Programmed cell death 1 ligand 1	7	−0.007	0.013	0.993 (0.968 to 1.020)	0.618
GCST90274833	Stem cell factor	5	−0.008	0.016	0.992 (0.962 to 1.024)	0.620
GCST90274834	SIR2–like protein 2	5	−0.008	0.016	0.992 (0.961 to 1.024)	0.622
GCST90274835	Signaling lymphocytic activation molecule	4	0.007	0.019	1.007 (0.969 to 1.046)	0.727
GCST90274836	Sulfotransferase 1A1	5	−0.016	0.016	0.984 (0.954 to 1.015)	0.314
GCST90274837	STAM binding protein	5	−0.004	0.015	0.996 (0.966 to 1.026)	0.776
GCST90274838	Transforming growth factor–alpha	6	0.000	0.013	1.000 (0.974 to 1.027)	0.992
GCST90274839	Tumor necrosis factor	8	0.002	0.013	1.002 (0.976 to 1.028)	0.907
GCST90274840	TNF–beta	3	−0.006	0.021	0.994 (0.954 to 1.036)	0.783
GCST90274841	Tumor necrosis factor receptor superfamily member 9	5	−0.004	0.017	0.996 (0.964 to 1.029)	0.791
GCST90274842	Tumor necrosis factor ligand superfamily member 14	7	−0.007	0.014	0.993 (0.967 to 1.021)	0.635
GCST90274843	TNF–related apoptosis–inducing ligand	6	0.000	0.015	1.000 (0.971 to 1.030)	0.991
GCST90274844	TNF–related activation–induced cytokine	5	0.004	0.016	1.004 (0.973 to 1.035)	0.821
GCST90274845	Thymic stromal lymphopoietin	8	0.002	0.013	1.002 (0.976 to 1.028)	0.897
GCST90274846	Tumor necrosis factor ligand superfamily member 12	7	0.003	0.013	1.003 (0.978 to 1.029)	0.803
GCST90274847	Urokinase–type plasminogen activator	6	−0.005	0.014	0.995 (0.968 to 1.023)	0.706
GCST90274848	Vascular endothelial growth factor A	8	−0.007	0.012	0.993 (0.971 to 1.017)	0.579

pval<0.05 was considered statistically significant

Figure 8. Forest plot of MR analysis of causal effects of glioblastoma on 90 inflammatory cytokines from EBI database (91 cytokines). MR = Mendelian randomization.

FGF21 promotes autophagy in LNCaP cells by inhibiting the phosphatidylinositol 3-kinase–Akt kinase–mammalian target of rapamycin (PI3K–Akt–mTOR) pathway. It also inhibits the migration and invasion of prostate cancer cells.[50] However, its role in GBM has not been reported to date, and our study reveals for the first time the importance of FGF21 as a key factor in GBM. This finding provides a new outlook for the potential clinical application of FGF21 drugs for the treatment of GBM, which should be investigated in future studies.

TNF-β, also called lymphotoxin-α, a member of the tumor necrosis factor superfamily, is a cytokine produced by lymphocytes.[51,52] Although much is known about the importance of

GWAS ID	Outcome	nSNP	b	se	OR(95% CI)	pval
ebi–a–GCST004420	CTACK	3	0.010	0.044	1.010 (0.927 to 1.101)	0.816
ebi–a–GCST004421	beta–nerve growth factor	2	–0.006	0.043	0.994 (0.914 to 1.081)	0.892
ebi–a–GCST004423	Macrophage Migration Inhibitory Factor	3	–0.009	0.037	0.991 (0.921 to 1.066)	0.813
ebi–a–GCST004426	Tumor necrosis factor alpha	2	0.014	0.054	1.014 (0.912 to 1.127)	0.793
ebi–a–GCST004427	Stromal–cell–derived factor 1 alpha	2	0.014	0.038	1.014 (0.942 to 1.092)	0.707
ebi–a–GCST004429	Stem cell factor	2	0.016	0.028	1.017 (0.963 to 1.073)	0.553
ebi–a–GCST004430	Interleukin–16	2	0.001	0.043	1.001 (0.920 to 1.088)	0.987
ebi–a–GCST004432	Platelet–derived growth factor BB	4	–0.012	0.022	0.988 (0.946 to 1.031)	0.571
ebi–a–GCST004433	Macrophage inflammatory protein 1b	2	–0.022	0.035	0.978 (0.914 to 1.048)	0.533
ebi–a–GCST004435	Monokine induced by gamma interferon	4	0.004	0.033	1.004 (0.942 to 1.070)	0.903
ebi–a–GCST004436	Macrophage colony stimulating factor	2	–0.007	0.065	0.993 (0.874 to 1.129)	0.920
ebi–a–GCST004438	Monocyte chemoattractant protein–1	2	–0.002	0.035	0.998 (0.932 to 1.069)	0.960
ebi–a–GCST004439	Interleukin–12p70	2	–0.003	0.035	0.997 (0.931 to 1.067)	0.925
ebi–a–GCST004441	Interleukin–18	2	–0.027	0.042	0.973 (0.896 to 1.057)	0.521
ebi–a–GCST004442	Interleukin–17	2	–0.010	0.037	0.990 (0.920 to 1.065)	0.786
ebi–a–GCST004446	Interleukin–6	3	–0.005	0.029	0.995 (0.939 to 1.054)	0.869
ebi–a–GCST004447	Interleukin–1–receptor antagonist	2	–0.013	0.054	0.987 (0.887 to 1.098)	0.805
ebi–a–GCST004449	Hepatocyte growth factor	3	–0.002	0.029	0.998 (0.943 to 1.057)	0.956
ebi–a–GCST004450	Interleukin–9	2	–0.015	0.053	0.986 (0.888 to 1.094)	0.784
ebi–a–GCST004451	Interleukin–7	2	0.012	0.055	1.012 (0.909 to 1.127)	0.826
ebi–a–GCST004452	Interleukin–5	2	0.002	0.055	1.002 (0.900 to 1.115)	0.974
ebi–a–GCST004453	Interleukin–4	4	0.000	0.026	1.000 (0.951 to 1.052)	0.994
ebi–a–GCST004454	Interleukin–2 receptor antagonist	4	0.013	0.033	1.013 (0.950 to 1.080)	0.697
ebi–a–GCST004455	Interleukin–2	2	–0.001	0.043	0.999 (0.918 to 1.087)	0.980
ebi–a–GCST004456	Interferon gamma	2	0.003	0.036	1.003 (0.934 to 1.076)	0.944
ebi–a–GCST004458	Granulocyte–colony stimulating factor	2	–0.024	0.036	0.976 (0.911 to 1.047)	0.503
ebi–a–GCST004459	Fibroblast growth factor basic	2	–0.032	0.036	0.969 (0.902 to 1.040)	0.384
ebi–a–GCST004460	Eotaxin	2	0.004	0.028	1.004 (0.950 to 1.061)	0.885

pval<0.05 was considered statistically significant

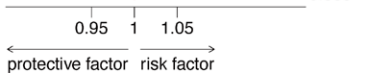


Figure 9. Forest plot of MR analysis of causal effects of glioblastoma on 28 inflammatory cytokines from University of Bristol database (41 cytokines). MR = Mendelian randomization.

tumor necrosis factor alpha (TNF-α) in carcinogenesis, data on TNF-β are very limited. Similar to TNF-α, several studies have demonstrated that TNF-β can regulate proliferation, survival, invasion, migration, and colony formation in colorectal and ovarian cancers.[53,54] The potential mechanism is that TNF-β activates the NF-κB signaling pathway to induce cancer cell proliferation and invasion.[55–57] Several reports have shown that resveratrol blocks NF-κB activation, inhibits the cell cycle, and induces apoptosis in various tumors and colorectal cancer cells.[58,59] Furthermore, Buhrmann demonstrated that Calebin A component of Curcuma longa, could inhibit TNF-β-induced NF-κB-mediated malignancy in CRC cells in vitro.[60] Our findings reveal a causal relationship between TNF-β and GBM, and the use of TNF-β blockade strategies may be a potential treatment for GBM.

Chemokines, as cardinal regulators of immune cell trafficking, play a pivotal role in inflammation and orchestrate the intricate immune landscape within the tumor microenvironment.[61] Analogous to their role in inflammation, chemokines mediate the recruitment of immune cells to the tumor niche, exerting both direct and indirect effects on tumor cells.[62] MIP-1a (Chemokine (C-C motif) ligand 3 [CCL3])[63] is a small secreted protein belonging to the C-C chemokine subfamily.[64,65] Its impact on tumorigenesis is complex, exhibiting both protumorigenic and antitumorigenic effects, depending on the context. They can promote angiogenesis, tumor growth, and metastasis by attracting immunosuppressive cells or by stimulating the release of growth factors. Conversely, they can also recruit immune effector cells to the tumor microenvironment, potentially enhancing antitumor immunity. The chemotactic effect of CCL3 enhances antitumor immunity by promoting dendritic cell homing to the tumor microenvironment.[66] Additionally, CCL3-mediated NK cell recruitment is crucial for further bolstering the CD8-positive antitumor response.[67] This study revealed the genetic correlation between MIP-1a and GBM, providing valuable insights for future research on the pathogenesis and pharmacological intervention of GBM.

The number of studies on the role of IL-10 in tumor diseases has gradually increased; however, its role in tumorigenesis and progression remains highly controversial.[68] Some studies have shown that IL-10 may also exert antitumor effects through certain mechanisms, such as promoting CD8+ T-cell proliferation and activation, and enhancing their antitumor ability.[69] Li Tang et al demonstrated that CAR-T cells expressing IL-10 resist dysfunction and mediate durable clearance of solid and metastatic tumors, including colon, breast, melanoma, and pancreatic cancers.[70] However, other studies have shown that elevated IL-10 levels suppress T cell-mediated killing of tumor cells and that blocking IL-10 in animal models improves the ability of the immune system to eliminate tumor cells.[71,72] Additionally, research indicates that IL-10 can create an immunosuppressive environment by inhibiting the activation of antigen-presenting cells (APCs), leading to inhibitory effects on T cells and ultimately inducing tumor immune escape.[73] Our study demonstrated that higher IL-10 levels promote GBM development. However, the role of IL-10 in the tumor microenvironment is complex and multifaceted. However, further studies are required to elucidate the mechanism by which IL-10 promotes GBM growth.

In this study, we investigated the causal relationship between inflammatory cytokines and GBM using an integrated approach involving MR analysis and BWMR. MR analysis is favored for its superior ability to circumvent confounding factors and mitigate the impact of ethical biases compared with conventional epidemiological statistical methods, thereby enhancing the reliability of our findings over those of prior research. Furthermore,

to bolster the robustness of our study outcomes, we selected the most recent and extensive GWAS data from Open GWAS databases, which feature the largest sample cohorts available.

Although this investigation offers valuable insights into the potential causal associations between inflammatory cytokines and GBM, it is essential to recognize several limitations. First, the sample size for the GBM GWAS data in our analysis was relatively modest, which may have compromised the statistical power of our MR approach. A smaller sample size can lead to wider confidence intervals and a reduced capacity to detect modest causal effects, potentially resulting in over- or underestimation of the effect sizes. To address the strength of the IVs, we employed the F-statistic, including only IVs with values exceeding 10 in subsequent analyses. Although this step enhances the reliability of our results by mitigating weak instrument bias, it does not fully eliminate the limitations imposed by the modest sample size. Consequently, these findings should be cautiously interpreted. Second, the broader threshold applied in our analysis could have increased the risk of false positives. Third, our findings should be interpreted with caution because of the complexity of cytokine signaling pathways and the limitations of our sample size. Validation in a larger cohort is essential to confirm these results. Fourth, all the GWAS data in this study were derived from European populations. Although this approach has minimized the confounding effects of population stratification, it also underscores the necessity of validating causal conclusions in other non-European cohorts, such as those of Asian descent. Finally, larger-scale GWAS and more comprehensive MR studies are required to enhance the robustness and applicability of our findings. Incorporating multi-omics data, such as proteomics and transcriptomics, could further clarify the mechanistic contributions of inflammatory cytokines in GBM, providing a stronger foundation for these preliminary observations.

5. Conclusion

In conclusion, the MR analysis revealed a causal relationship between specific inflammatory cytokines and GBM. Specifically, TNF-β and IL-10 have been identified as risk factors for GBM, whereas FGF21 and MIP-1a are recognized as protective factors against this disease. Notably, a reverse investigation revealed that GBM is not associated with elevated levels of inflammatory cytokines. These targeted inflammatory cytokines may provide a promising strategy for the treatment and prevention of GBM. Further research is needed to confirm these findings and elucidate the underlying biological mechanisms involved.

Acknowledgments

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Author contributions

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