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RESEARCH ARTICLE

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Exploring causal correlations between inflammatory cytokines and intervertebral disc degeneration: A Mendelian randomization

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Abstract

Background: Inflammatory cytokines have been reported to be related to intervertebral disc degeneration (IVDD) in several previous studies. However, it remains unclear about the causal relationship between inflammatory cytokines and IVDD. This study employs Mendelian randomization (MR) to analyze the causal link between inflammatory cytokines and the risk of IVDD.

Method: We used genetic variants associated with inflammatory cytokines from a meta-analysis of genome-wide association study (GWAS) in 8293 Finns as instrumental variables and IVDD data were sourced from the FinnGen consortium. The main analytical approach utilized Inverse-Variance Weighting (IVW) with random effects to assess the causal relationship. Additionally, complementary methods such as MR-Egger, weighted median, simple mode, weighted mode, and MR pleiotropy residual sum and outlier were employed to enhance the robustness of the final results.

Result: We found interferon-gamma (IFN-γ, $p = 2.14 \times 10–6$, OR = 0.870, 95% CI = 0.821–0.921), interleukin-1 beta (IL-1b, $p = 0.012$, OR = 0.951, 95% CI = 0.914–0.989), interleukin-4 (IL-4, $p = 0.034$, OR = 0.946, 95% CI = 0.899–0.996), interleukin-18 (IL-18, $p = 0.028$, OR = 0.964, 95% CI = 0.934–0.996), granulocyte colony-stimulating factor (GCSF, $p = 0.010$, OR = 0.919, 95% CI = 0.861–0.980), and Stromal cell-derived factor 1a (SDF1a, $p = 0.014$, OR = 1.072, 95% CI = 1.014–1.134) were causally associated with risk of IVDD.

Conclusion: Our MR analyses found a potential causal relationship between six inflammation cytokines (IFN-γ, IL-1b, IL-4, IL-18, SDF1a, and GCSF) and altered IVDD risk.

KEYWORDS
GCSF, IFN-γ, IL-18, IL-1b, IL-4, intervertebral disc degeneration, Mendelian randomization, SDF1a

1 | INTRODUCTION

Low back pain (LBP) is one of the prevailing musculoskeletal issues globally.[1] The incidence of low back pain is high, and it is common in people of all ages and economic conditions.[2] It affects a significant proportion of the population and has the highest health-related economic expenditure in the United States.[3] Statistics reveal the lifetime prevalence of low back pain is close to 80%.[4] In addition, the related cost of LBP treatment and rehabilitation in the United States exceeded 134.5 billion US dollars in 2016, which was higher than other 153 diseases included in the statistics, and ranked first in the disease burden list.[5] In addition to causing pain, numbness, and limited activity, LBP also seriously affects the working status and mobility of patients.[6] Although the cause of most cases of low back pain is unknown, intervertebral disc degeneration (IVDD) is widely acknowledged as a pivotal factor.[7,8]

IVDD is the key pathological basis of spinal degenerative diseases, which often causes low back pain, spinal stenosis, and ultimately leads to limited activity and loss of mobility.[9] Growing evidence suggests that abnormal production of inflammatory cytokines is strongly correlated to the progression of IVDD, and these cytokines cause a series of pathogenic responses in intervertebral disc cells that can lead to apoptosis, pyroptosis, and senescence.[10–13] Previous studies proved that tumor necrosis factor-alpha (TNF-α) and interleukin-1 beta (IL-1b) increase the expression of genes encoding matrix-degrading enzymes.[14] Compared with non-degenerative disc tissue, degenerative and herniated discs showed higher TNF-a and IL-1b expression, with higher levels of IL-1b.[10] Similarly, other cytokines, such as interleukin-4 (IL-4), interleukin-18 (IL-18), and interferon-gamma (IFN-γ), have been reported to be closely associated with IVDD.[15–17] The serum level of IL-18 significantly increased with the increase of IVDD grade.[15] Another study demonstrated that an increase in mechanical load resulted in significant elevations of inflammatory factors (IL-6, IL-1b, and IFN-γ) and extracellular matrix catabolic markers.[17] In contrast, IL-4 exhibits a significant anti-inflammatory effect on LPS-stimulated nucleus pulposus cells.[16] However, traditional epidemiological studies are prone to bias, confusion, and reverse causation, which can lead to misleading results.[18] Therefore, it is important to study the causal relationship between inflammatory cytokines and IVDD.

Mendelian randomization (MR) is a statistical model using genetic variation as an instrumental variable, which can avoid the potential confounder, reverse causality, and other bias risks brought by observational studies, and is similar to randomized controlled studies to a certain extent.[19] It serves as a robust methodology for assessing the causal relationship between exposure factors and outcome variables.[20,21] In this study, we used MR analysis to investigate the causal relationship between inflammatory cytokines and IVDD, aiming to offer evidence for alleviating and delaying the development of IVDD.

2 | METHODS

2.1 | MR assumptions

The MR analysis has three key assumptions: (1) Association hypothesis: genetic instrumental variables (IVs) are closely related to exposure factors; (2) Independence hypothesis: IVs should not exhibit associations with potential confounders; (3) Exclusivity hypothesis: IVs should impact outcome factors entirely through exposure factors.[22] The study design is depicted in Figure 1. In this study, two genome-wide association study (GWAS) datasets were employed to identify genetically significant single nucleotide polymorphisms (SNPs) for 41 inflammatory cytokines and IVDD.

2.2 | GWAS data source

Summary GWAS data for IVDD were available from the FinnGen consortium specifically form R8 release (https://www.finngen.fi/fi), including 29 508 cases and 227 388 controls. IVDD diagnosis was based on International Classification of Diseases (ICD)-10 M51, ICD-9722, and ICD-8725. The ICD-10 M51 is defined as thoracic, thoracolumbar, and lumbosacral disc disorders. ICD-9722 is defined as Intervertebral disc disorders. ICD-8725 is defined as displacement of intervertebral disc. The GWAS data for 41 inflammatory cytokines originated from the study providing genome variant associations with 41 cytokines and growth factors in 8293 Finnish individuals.[23]

2.3 | Instrumental variable selection

First, for each inflammatory cytokine, we selected SNPs with genome-wide significant threshold at $p < 1 \times 10^{-5}$. When we selected SNPs with genome-wide significant threshold at the 5×10^{-8}, there were too few SNPs with genome-wide significant in inflammatory cytokine GWAS result. In this condition, problems such as low statistical power and weak instrumental variables will occur in MR Analysis. Therefore, we chose threshold at the 1×10^{-5} to select SNPs. Second, we clumped these SNPs with thresholds at kb = 10 000, $r^2 = 0.001$ to eliminate linkage disequilibrium.[24] Third, we calculated the F statistic of instruments for each cytokine. And we set a threshold of $F > 10$ to select strong instruments for further MR analysis.[25] Palindromic SNPs were discarded by harmonizing processes. In adherence to the independence hypothesis, the selected

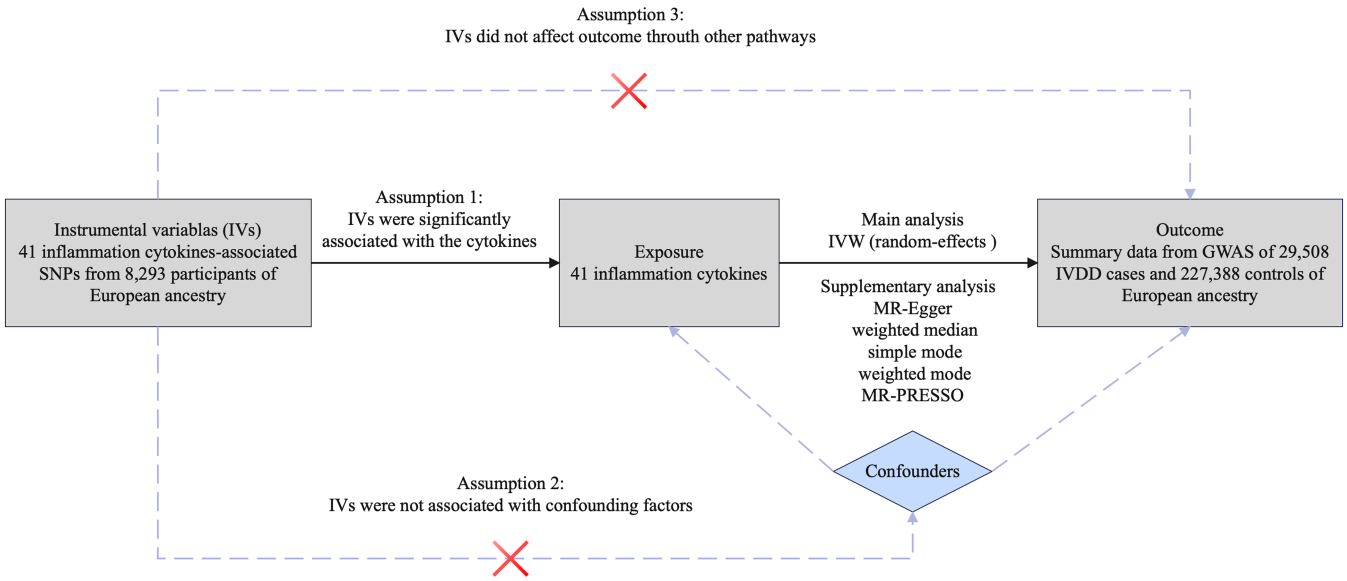

FIGURE 1 Overall flow chart of this study. IVDD: Intervertebral disc degeneration; IVW: Inverse-variance weighted.

SNPs underwent further scrutiny in the PhenoScanner database to exclude IVs associated with confounders.

2.4 | MR analysis

The primary analysis method employed in MR analysis was Inverse-Variance Weighted (IVW) with random effects.[26] Additionally, MR-Egger, weighted median, simple mode, and weighted mode were employed as supplementary analysis methods.[27-29] IVW, a meta-method for multiple site effects in the analysis of multiple SNPs, assumes that all SNPs are effective IVs and entirely independent of each other.[26] For each SNP, IVW calculates its effect estimate on exposure and outcome, assigns weights to each SNP based on the variance of the effect estimate, and then obtains the overall causal effect estimate through weighted averaging. MR-Egger method takes into account the possible heterogeneity of IVs and provides a corrected estimate of causal effect.[28] MR-Egger performs regression analysis using effect estimates of all SNPs and obtains corrected causal effect estimates through regression analysis. MR-Egger also provides an intercept term that can be used to detect and correct for certain biases. Weighted median can yield robust estimates when at least half IVs are effect. The method ranks the effect estimates of each SNP and calculates the weighted median as the estimate of the causal effect.[27] Simple mode provides robustness for pleiotropy, although it is not as powerful as IVW. Simple mode selects a strongly correlated SNP as the IV and uses the effect estimate of this SNP to estimate the causal effect of the exposure on the outcome.[29] Weighted mode is a method of weighting SNP effect estimates. Based on the variance of the SNP and the correlation between the size of the SNP effect and exposure, different weights are assigned to the effect estimates for each SNP, which are then applied to the estimation of SNP effects.[29]

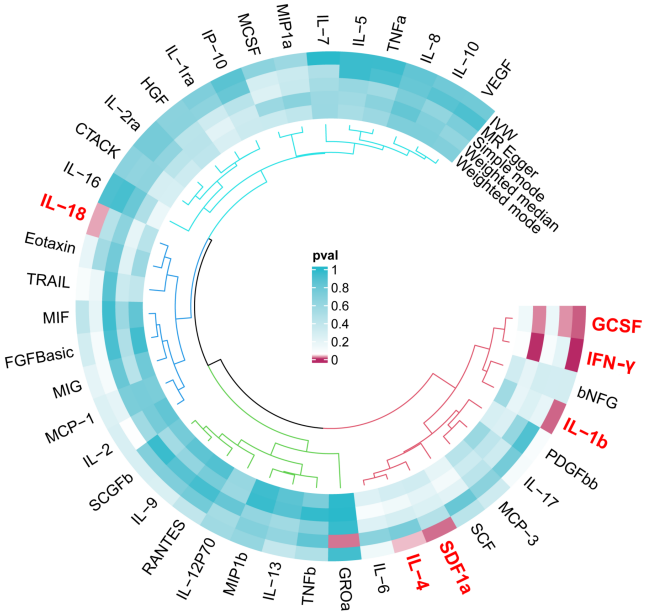

FIGURE 2 A panorama of all the 41 inflammatory cytokines features and their causal estimate on the risk of IVDD. Six cytokines were found to have causal association with IVDD using IVW, including GCSF, IFN-γ, IL-1b, SDF1a, IL-4, and IL-18. GCSF and IFN-γ were found to have causal association with IVDD using weighted median. In addition, GCSF and GROa were found to have causal association with IVDD using MR Egger. IVDD: Intervertebral disc degeneration; IVW, inverse variance weighted.

2.5 | Sensitivity analysis

Q-test for IVW and MR-Egger was used to check the heterogeneity of MR analysis results, and $p > 0.05$ signifies the absence of heterogeneity.[30] MR-Egger regression and MR pleiotropy residual

exposure	nsnp	method	pval	OR(95% CI)
GCSF	18	MR Egger	**0.023**	0.868 (0.778 to 0.969)
	18	Weighted median	**0.020**	0.918 (0.854 to 0.986)
	18	Inverse variance weighted	**0.010**	0.919 (0.861 to 0.980)
	18	Simple mode	0.159	0.900 (0.783 to 1.035)
	18	Weighted mode	0.137	0.902 (0.792 to 1.027)
IFN−γ	14	MR Egger	0.202	0.927 (0.830 to 1.035)
	14	Weighted median	**<0.001**	0.882 (0.819 to 0.950)
	14	Inverse variance weighted	**<0.001**	0.870 (0.821 to 0.921)
	14	Simple mode	0.092	0.890 (0.784 to 1.009)
	14	Weighted mode	0.066	0.886 (0.787 to 0.997)
IL−1b	9	MR Egger	0.183	0.934 (0.854 to 1.022)
	9	Weighted median	0.181	0.963 (0.911 to 1.018)
	9	Inverse variance weighted	**0.012**	0.951 (0.914 to 0.989)
	9	Simple mode	0.248	0.943 (0.860 to 1.034)
	9	Weighted mode	0.559	0.983 (0.928 to 1.040)
IL−4	18	MR Egger	0.739	1.021 (0.906 to 1.151)
	18	Weighted median	0.135	0.948 (0.883 to 1.017)
	18	Inverse variance weighted	**0.034**	0.946 (0.899 to 0.996)
	18	Simple mode	0.279	0.934 (0.829 to 1.053)
	18	Weighted mode	0.293	0.939 (0.839 to 1.052)
IL−18	18	MR Egger	0.446	0.972 (0.905 to 1.044)
	18	Weighted median	0.191	0.972 (0.930 to 1.014)
	18	Inverse variance weighted	**0.028**	0.965 (0.934 to 0.996)
	18	Simple mode	0.655	0.985 (0.922 to 1.052)
	18	Weighted mode	0.435	0.977 (0.922 to 1.035)
SDF1a	17	MR Egger	0.459	1.051 (0.925 to 1.193)
	17	Weighted median	0.199	1.054 (0.973 to 1.142)
	17	Inverse variance weighted	**0.014**	1.072 (1.014 to 1.134)
	17	Simple mode	0.248	1.078 (0.954 to 1.219)
	17	Weighted mode	0.309	1.064 (0.948 to 1.193)

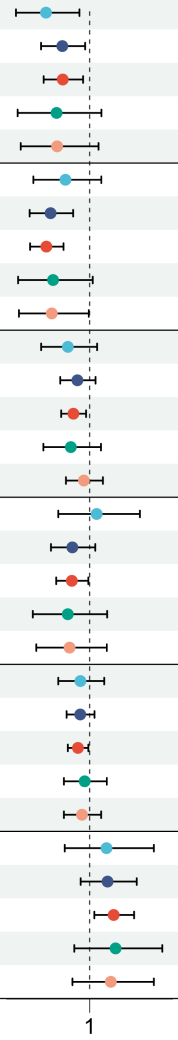

FIGURE 3 Positive MR estimates of the causal association between inflammatory cytokines and the risk of IVDD. MR: Mendelian randomization; IVDD: Intervertebral disc degeneration; nSNP: Number of single nucleotide polymorphism; OR: Odds ratio; CI: Confidence interval.

sum and outlier (MR-PRESSO) were utilized to check horizontal pleiotropy ensuring that genetic variation was independently associated with exposure and outcome, and $p > 0.05$ signifies the absence of pleiotropy.[28,31] MR-PRESSO was further applied to identify and exclude potential outliers, providing adjusted results.[31] Leave-one-out analysis was conducted to analyze the possibility that whether the causal association was driven by any single SNP.[32]

2.6 | Statistical analysis

All MR analyses were conducted using the "TwoSampleMR" and "Mendelian Randomization" package in R4.3.2 software. To solve the problem of multiple tests, Benjamini-Hochberg method was applied to adjust the p value and control the error discovery rate. $p < 0.05$ was considered as statistically significant.

3 | RESULTS

At the genome-wide significance level, 6 to 30 independent SNPs were identified as IVs for cytokines, with corresponding F-statistics ranging from 14.05 to 177.30 (Supplementary file 1). In the PhenoScanner database, two SNPs (rs10761731 and rs385076) that showed association with confounders were excluded at the early stage. The results of the above analysis methods about causal relationship between 41 cytokines and IVDD were illustrated in Figure 2 and Supplementary file 2. Six cytokines were found to have causal association with IVDD using IVW, including GCSF,

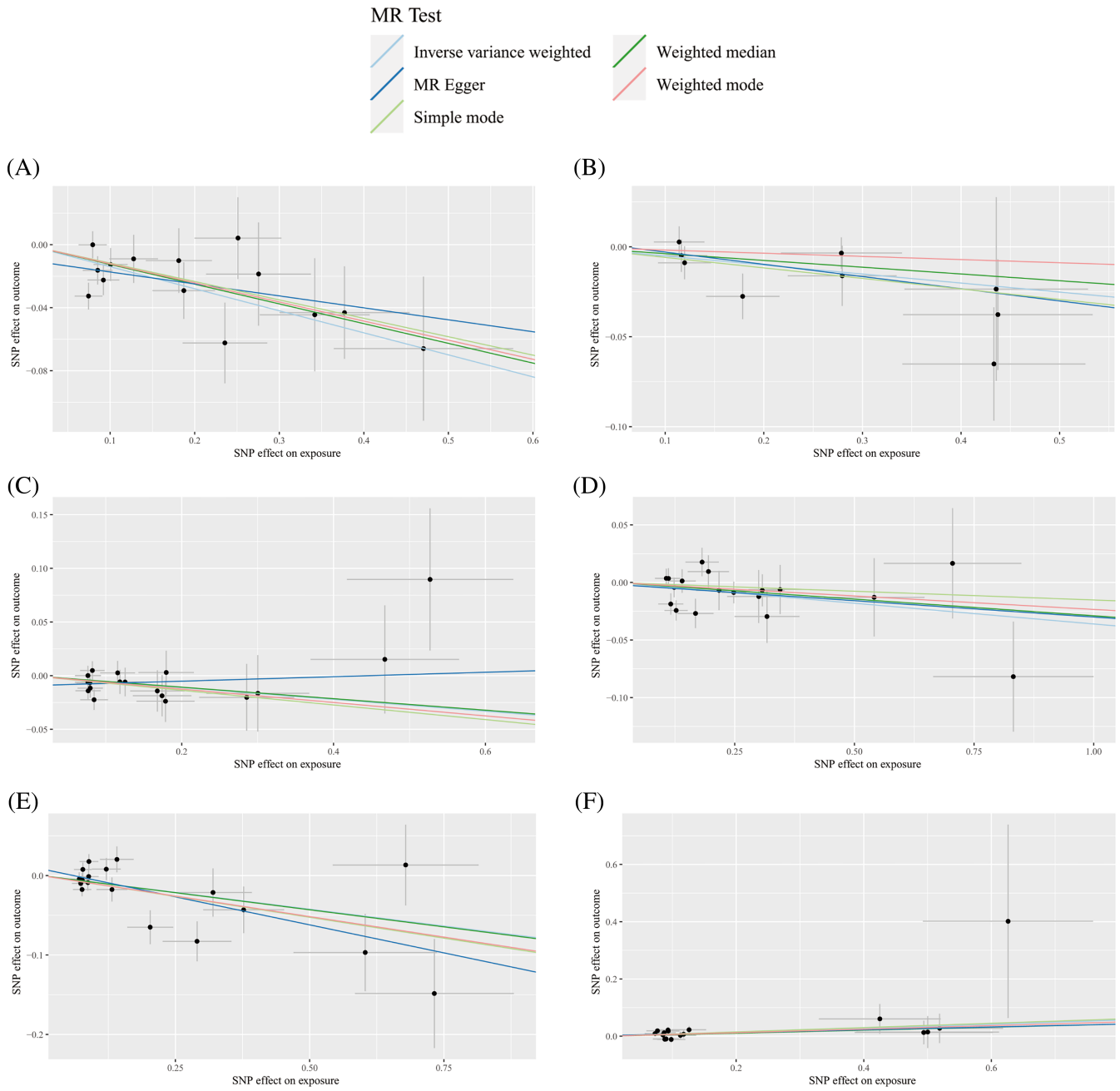

FIGURE 4 Scatter plot of genetic associations with inflammation cytokines against the risk of IVDD using different MR methods. (A). The relationship between IFN-γ and the risk of IVDD; (B). the relationship between IL-1b and the risk of IVDD; C. the relationship between IL-4 and the risk of IVDD; D. the relationship between IL-18 and the risk of IVDD; E. the relationship between GCSF and the risk of IVDD; F. the relationship between SDF1a and the risk of IVDD. MR: Mendelian randomization; IVDD: Intervertebral disc degeneration; IFN-γ: Interferon-gamma; IL-1b: Interleukin-1 beta; IL-4: Interleukin-4; IL-18: Interleukin-18; GCSF: Granulocyte colony-stimulating factor; SDF1a: Stromal cell-derived factor 1a.

IFN-γ, IL-1b, SDF1a, IL-4, and IL-18. GCSF and IFN-γ were found to have causal association with IVDD using weighted median. In addition, GCSF and GROa were found to have causal association with IVDD using MR Egger. The result heterogeneity and horizontal pleiotropy assay were presented in Supplementary file 3. The causal association of six cytokines with IVDD is shown in Figures 3 and 4.

3.1 | Causality between IFN-γ and IVDD

IVW analysis indicated that IFN-γ was linked to a decreased risk of IVDD ($p = 2.14 \times 10^{-6}$, OR = 0.870, 95% CI = 0.821–0.921). Weighted median analysis similarly demonstrated a negative causal association between IFN-γ and IVDD ($p = 0.001$, OR = 0.882, 95% CI = 0.819–0.950). In addition, no heterogeneity was

TABLE 1 Heterogeneity analysis of inflammatory cytokines.

	IVW (heterogeneity)		MR Egger (heterogeneity)	
Cytokine	*p* value	Q	*p* value	Q
IFNg	0.378	12.878	0.322	14.762
IL-1b	0.464	6.673	0.552	6.855
IL-4	0.821	10.804	0.757	12.686
IL-18	0.220	20.014	0.270	20.086
GCSF	0.016	30.346	0.010	33.246
SDF1a	0.273	17.809	0.327	17.955

Abbreviations: GCSF, Granulocyte colony-stimulating factor; IFNg, interferon-gamma; IL-18, Interleukin-18; IL-1b, Interleukin-1 beta; IL-4, Interleukin-4; IVW, inverse-variance weighted; SDF1a, Stromal cell-derived factor 1a.

TABLE 2 Pleiotropy analysis of inflammatory cytokines.

	MR Egger (pleiotropy)		MR-PRESSO Global test	
Cytokine	*p* value	Intercept	*p* value	RSSobs
IFNg	0.210	−0.010	0.404	16.831
IL-1b	0.682	0.004	0.512	10.599
IL-4	0.189	−0.009	0.761	14.143
IL-18	0.814	−0.002	0.324	22.065
GCSF	0.234	0.008	0.015	37.387
SDF1a	0.731	0.002	0.365	20.209

Abbreviations: GCSF, Granulocyte colony-stimulating factor; IFNg, interferon-gamma; IL-18, Interleukin-18; IL-1b, Interleukin-1 beta; IL-4, Interleukin-4; MR-PRESSO, MR pleiotropy residual sum and outlier; SDF1a, Stromal cell-derived factor 1a.

observed in MR-Egger and IVW test ($p = 0.322$; $p = 0.378$) (Table 1). Furthermore, no pleiotropy was observed in MR egger-intercept and MR-PRESSO global test ($p = 0.210$; $p = 0.404$) (Table 2). The result of MR-PRESSO was similar to that of IVW ($p = 3.87 \times 10^{-4}$).

3.2 | Causality between IL-1β and IVDD

IVW analysis indicated that IL-1β was linked to a decreased risk of IVDD ($p = 0.012$, OR = 0.951, 95% CI = 0.914–0.989). In addition, no heterogeneity was observed in MR-Egger and IVW test ($p = 0.552$; $p = 0.464$) (Table 1). Furthermore, no pleiotropy was observed in MR egger-intercept and MR-PRESSO global test ($p = 0.682$; $p = 0.512$) (Table 2). The result of MR-PRESSO was similar to that of IVW ($p = 0.027$).

3.3 | Causality between IL-4 and IVDD

IVW analysis indicated that IL-4 was linked to a decreased risk of IVDD ($p = 0.034$, OR = 0.946, 95% CI = 0.899–0.996). In addition, no heterogeneity was observed in MR-Egger and IVW test ($p = 0.757$; $p = 0.821$) (Table 1). Furthermore, no pleiotropy was observed in MR egger-intercept and MR-PRESSO global test ($p = 0.189$; $p = 0.761$) (Table 2). The result of MR-PRESSO was similar to that of IVW ($p = 0.025$).

3.4 | Causality between IL-18 and IVDD

IVW analysis indicated that IL-18 was linked to a decreased risk of IVDD ($p = 0.028$, OR = 0.964, 95% CI = 0.934–0.996). In addition, no heterogeneity was observed in MR-Egger and IVW test ($p = 0.270$; $p = 0.220$) (Table 1). Furthermore, no pleiotropy was observed in MR egger-intercept and MR-PRESSO global test ($p = 0.814$; $p = 0.324$) (Table 2). The result of MR-PRESSO was similar to that of IVW ($p = 0.042$).

3.5 | Causality between SDF1a and IVDD

IVW analysis indicated that SDF1a was linked to an increased risk of IVDD ($p = 0.014$, OR = 1.072, 95% CI = 1.014–1.134). In addition, no heterogeneity was observed in MR-Egger and IVW test ($p = 0.327$; $p = 0.273$) (Table 1). Furthermore, no pleiotropy was observed in MR egger-intercept and MR-PRESSO global test ($p = 0.731$; $p = 0.365$) (Table 2). The result of MR-PRESSO was similar to that of IVW ($p = 0.026$).

3.6 | Causality between GCSF and IVDD

IVW analysis revealed a significant association between GCSF and a reduced risk of IVDD ($p = 0.010$, OR = 0.919, 95% CI = 0.861–0.980). Although heterogeneity was detected in Q-test for MR-Egger and IVW ($p = 0.010$; $p = 0.016$) (Table 1), we used IVW with random effects to alleviate the problem. No directional pleiotropy was identified using MR Egger-intercept ($p = 0.731$) (Table 2); The MR-PRESSO global test indicated the presence of horizontal pleiotropy ($p = 0.015$), but no outlier was identified using MR-PRESSO outlier test. The result of MR-PRESSO was similar to that of IVW ($p = 0.020$).

The leave-one-out analysis indicated that the removal of single SNP did not substantially influence the results (Figure 5). After adjustments of *P*-values (Supplementary file 4), only IFN-γ was still associated with IVDD significantly (adjusted $p = 8.77 \times 10^{-5}$).

4 | DISCUSSION

IVDD is a prevalent health concern that significantly influences patients' quality of life and exerts a substantial economic burden on countries.[33] Up to now, despite the high morbidity rate of IVDD, a comprehensive understanding of its causal factors of IVDD has

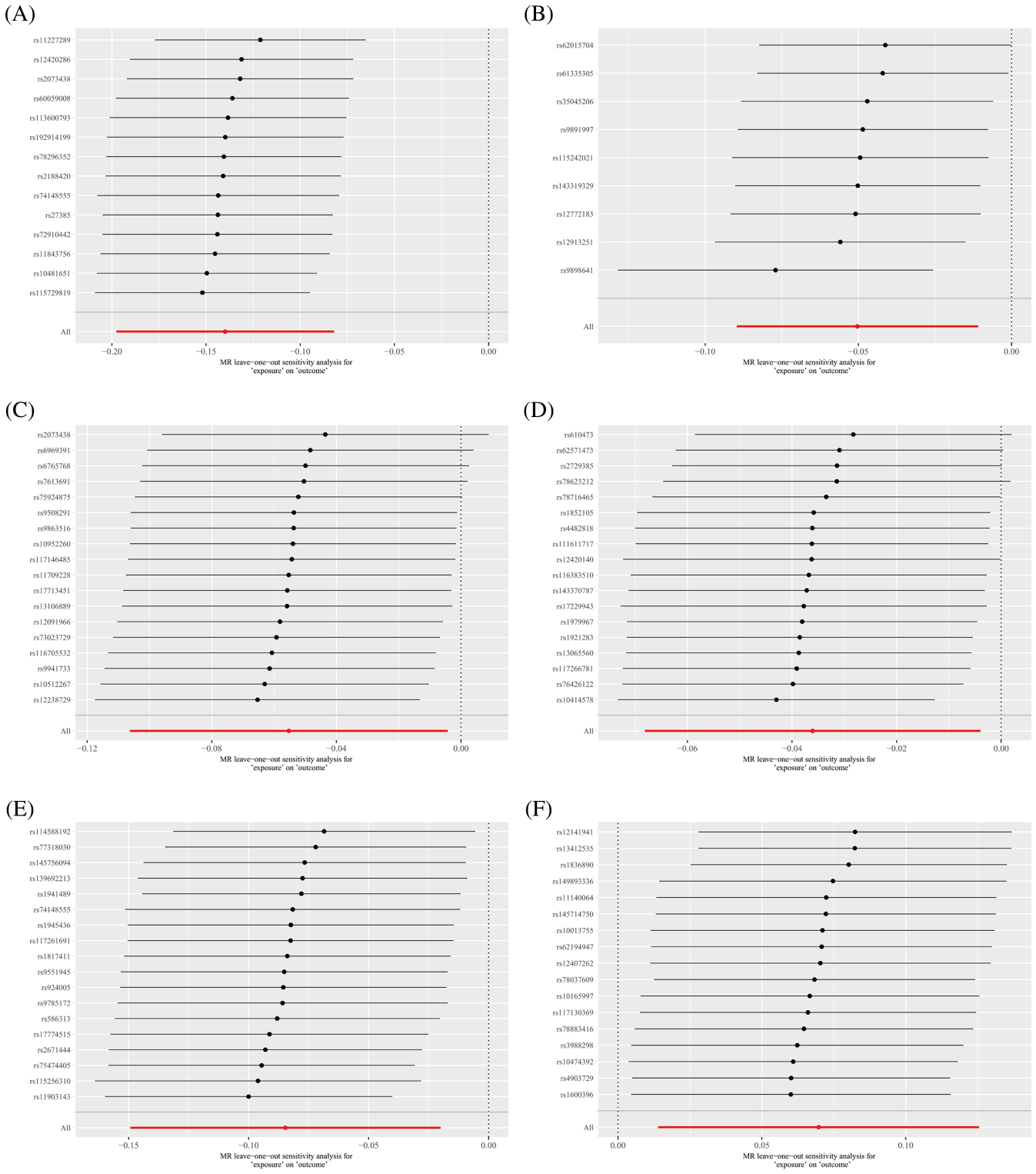

FIGURE 5 Plots of "leave-one-out" analyses for MR analyses of the causal effect of inflammatory cytokines with the risk of IVDD. (A). represents IFN-γ on the risk of IVDD; (B). represents IL-1b on the risk of IVDD; (C). represents IL-4 on the risk of IVDD; (D). represents IL-18 on the risk of IVDD; (E). represents GCSF on the risk of IVDD; (F). represents SDF1a on the risk of IVDD; MR: Mendelian randomization; IVDD: Intervertebral disc degeneration; IFN-γ: Interferon-gamma; IL-1b: Interleukin-1 beta; IL-4: Interleukin-4; IL-18: Interleukin-18; GCSF: Granulocyte colony-stimulating factor; SDF1a: Stromal cell-derived factor 1a.

remained elusive. Recent evidence has highlighted that inflammatory cytokines may be associated with the progression of IVDD, which has focused research direction on inflammation.[10] In this study, a two-sample Mendelian Randomization (MR) analysis was conducted to investigate the potential association between inflammatory cytokines and IVDD based on the genetic level.

The findings of our study provide evidence supporting a causal association between elevated levels of SDF1a and increased risk of IVDD. Furthermore, elevated levels in IFN-γ, IL-1b, IL-4, IL-18, and GCSF were suggestive of a decreased risk of IVDD.

IFN-γ, the sole type II IFN member that is primarily released by macrophages and T cells, is a vital regulator of immune function and offers a strong first line of defense against invasive infections.[34] It has been shown that the level of IFN-γ is significantly increased in degenerated discs.[17] However, IFN-γ has a paradoxical role in tissue inflammation; Although IFN-γ was known as a proinflammatory cytokine, increasing evidence suggests that IFN-γ also has immunosuppressive activity.[35,36] Notably, studies in a canine model of disc degeneration demonstrated a significant downregulation of IFN-γ expression in disc extrusion over the entire course, which challenged the traditional understanding of IFN-γ's role in IVDD.[37] Our current MR analysis revealed that high levels of circulating IFN-γ were associated with a reduced risk of IVDD. IL-1b and IL-18 were considered as pro-inflammatory cytokines that were highly involved in the development of IVDD.[14,15] However, our current MR result concluded that IL-1b and IL-18 were also negatively correlated with the risk of IVDD. This result also contradicts the current view. Further studies with more comprehensive data are needed to elucidate the causal relationship between IL-1b, IL-18, and IFN-γ with IVDD.

IL-4, a cytokine secreted by T helper cells, plays a role in promoting the proliferation of B cells.[38] Te Velde et al first demonstrated that IL-4 has anti-inflammatory effects.[39] Kedong et al. found that IL-4 significantly reduces the expression of inflammatory genes (CD68, IFN-β, IL-6, IL-8) deduced by lipopolysaccharide in disc cells.[16] Sibel et al. found higher levels of IL-4 expression in disc samples from patients with IVDD compared to normal disc samples.[40] Another study found that serum IL-4 levels are higher in patients with low back pain than pain-free healthy controls.[41] IL-4 as an anti-inflammatory cytokine may have analgesic effect.[42] This may explain the elevated levels of IL-4 in some patients with low back pain. In contrast to the above research, Capossela et al. found lower serum IL-4 levels in patients with low back pain.[43] Our analysis suggested a potential causal association between IL-4 and IVDD. High circulating levels of IL-4 may suppress the development of IVDD ($p = 0.034$, OR = 0.946, 95% CI = 0.899–0.996). Based on previous studies and the present MR analysis, IL-4 is crucial in the disease's etiology and is an important therapeutic target in IVDD.

GCSF is a hematopoietic growth factor influencing the proliferation, differentiation, and activation of hematopoietic cells.[44,45] GCSF has the capacity to inhibit apoptotic factors and promote anti-apoptotic factors and anti-inflammatory factors on host cells.[46] In general, GCSF plays its role in anti-apoptotic effect via increasing BCL-2 expression and inhibiting Bax expression and results in subsequent inhibition of caspase-3 activation. Previous study has proved that pro-inflammatory cytokines produce the expression of mRNA and protein of SDF1a, and SDF1a is also upregulated in the degenerative disc tissues.[47] This was confirmed by other studies, documenting significantly higher SDF-1α levels in IVDD group compared with the control group.[48] Fattah et al. reported that GCSF alleviates the progression IVDD and promotes its recovery. In addition, the expression of SDF1a was significantly decreased with the treatment of GCSF.[45] However, these studies have difficulty in establishing definite causal correlations. Our current MR analysis revealed that GCSF was considered as a causal protective factor for IVDD and SDF1a was causally associated with the risk of IVDD.

This study is the first to employ MR methods to investigate the causal relationship between IVDD and inflammation cytokines. However, certain limitations should be acknowledged. First, due to the study individuals included in the GWAS data were European population, the conclusions should be promoted with caution in other races. Second, we set 1×10^{-5} as the threshold value for genome-wide significance to select the IVs. This might introduce false-positive variants, potentially biasing the results. Nonetheless, the *F* statistic of all selected IVs was more than 10, which reduced weak IV bias. Third, there might not be a linear relationship between inflammatory cytokines and IVDD, and we were unable to get individual-level data for a further non-linear MR study.

5 | CONCLUSION

Our MR analyses found a potential causal relationship between six inflammation cytokines (IFN-γ, IL-1b, IL-4, IL-18, SDF-1a, and GCSF) and altered IVDD risk. Further investigations are needed to verify these results, delve into the underlying pathogenesis of IVDD, and focus on developing effective treatment strategies for IVDD.

AUTHOR CONTRIBUTIONS

Conception and design of the work: Yingchi Zhang; Acquisition of data: Tao Xu and Guangzi Chen. Analysis of data: Tao Xu, Guangzi Chen, and Jian Li; Interpretation of data: Yingchi Zhang and Tao Xu; Draft and revise the work: Tao Xu and Yingchi Zhang;

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CONFLICT OF INTEREST STATEMENT

The authors have no conflicts of interest relevant to this article.

DATA AVAILABILITY STATEMENT

All data generated or analyzed during this study are included in this published article and its supplementary information files.

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SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

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