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Original Article

Mendelian Randomization Analysis Identifies Druggable Genes for 7 Types of Osteoarthritis

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Abstract:

Objective Osteoarthritis (OA) is a common joint disease marked by cartilage degeneration, particularly in the elderly and obese. Current treatments mainly provide symptomatic relief, emphasizing the need for new therapies targeting the disease's root causes. This study aims to identify novel druggable genes for OA treatment using drug-targeted Mendelian randomization (MR) study.

Method The study utilized a druggable genome-wide Mendelian Randomization (MR) approach, leveraging summary-level data from genome-wide association studies (GWAS) and expression quantitative trait loci (eQTL) studies. SMR analysis was conducted to evaluate the causal association of druggable genes on 7 types of OA and 4 OA-related traits. Colocalization analysis was conducted to deepen the understanding and identify potential therapeutic targets of OA. Two-sample MR and protein quantitative trait loci (pQTL) analyses were used for further validation. Identified genes were categorized based on their potential as drug targets, and a phenome-wide association study (PheWAS) assessed the safety of these targets. In addition, drug prediction, and molecular docking were performed to provide valuable guidance for the development of more effective and targeted therapeutic drugs.

Results The SMR analysis identified multiple druggable genes associated with 7 types of OA and 4 OA-related traits. Key genes such as MAPK3 and CSK were consistently associated with multiple OA traits. Colocalization analysis provided strong evidence for 35 genes, suggesting their potential as therapeutic targets. Two-sample MR and pQTL analysis further supported these findings, identifying MAPK3, CSK, IFNGR2, MVD, FES, and ITGA2 as tier 3 targets. PheWAS indicated potential side effects for these targets.

Conclusions The study identified several potential druggable genes for OA treatment, with MAPK3, CSK, IFNGR2, MVD, FES, and ITGA2 showing the most promise. These findings provide a foundation for prioritizing drug treatment for OA.

Keywords Druggable genes, Osteoarthritis, Summary-data-based Mendelian randomization

Introduction

Osteoarthritis (OA) is the most prevalent joint disease globally, characterized primarily by the degeneration of joint cartilage, particularly in the knees, hands and hip[(1)]. This condition causes significant pain and functional impairments worldwide, especially affecting the elderly and individuals with obesity[(1)]. The rapid increase in the prevalence and social and economic burden of OA has prompted an urgent need for efficient treatment for OA[(2)]. However, current

pharmacological treatments for OA are primarily limited to steroids or non-steroidal anti-inflammatory drugs that provide symptomatic relief and drugs that can alter the underlying pathophysiology of OA are still under development[3]. Consequently, identifying new drug targets are crucial for advancing innovative therapeutic strategies for OA and improving patient outcomes.

Druggable genes that encode proteins or gene expression can provide clues for the discovery of drug targets[4]. Large-scale human genetic studies have created new opportunities for drug development for many complex diseases[5]. Currently, many single nucleotide polymorphisms (SNPs) associated with OA-related traits have been identified in large scale genome-wide association studies (GWASs). However, GWAS data lack the ability to provide clear and direct evidence about drug targets for many identified SNPs are located in non-coding regions or gene intervals.

Mendelian randomization (MR) is an approach used to assess the causality between a modifiable exposure or risk factor and a clinically relevant outcome[6]. As genetic variation is randomly distributed at conception and is not modified by acquired factors and disease onset, this approach is less susceptible to confusion and reverse causality bias than observational studies[7]. These days, MR analysis has been widely used to discover novel druggable targets by aggregating summary data from disease GWASs and expression quantitative trait locus (eQTL) studies[8,9]. The expression level of genes can be considered as a lifelong exposure, while eQTLs located in genomic regions of drug-target genes are generally thought out proxy[10]. Therefore, our study utilizes Summary-data-based Mendelian Randomization (SMR) to investigate the causal impact of druggable genes on 7 types of OA and 4 OA-related traits. By integrating these genetic tools with clinical phenotypes of OA, this study aims to minimize confounding factors through an MR-based analytical framework, identify genes that can serve as potential druggable targets for multiple traits of OA, and achieve a clearer understanding of the genetic basis of OA.

Method

The conceptual diagram of the current study is shown in **Figure 1**.

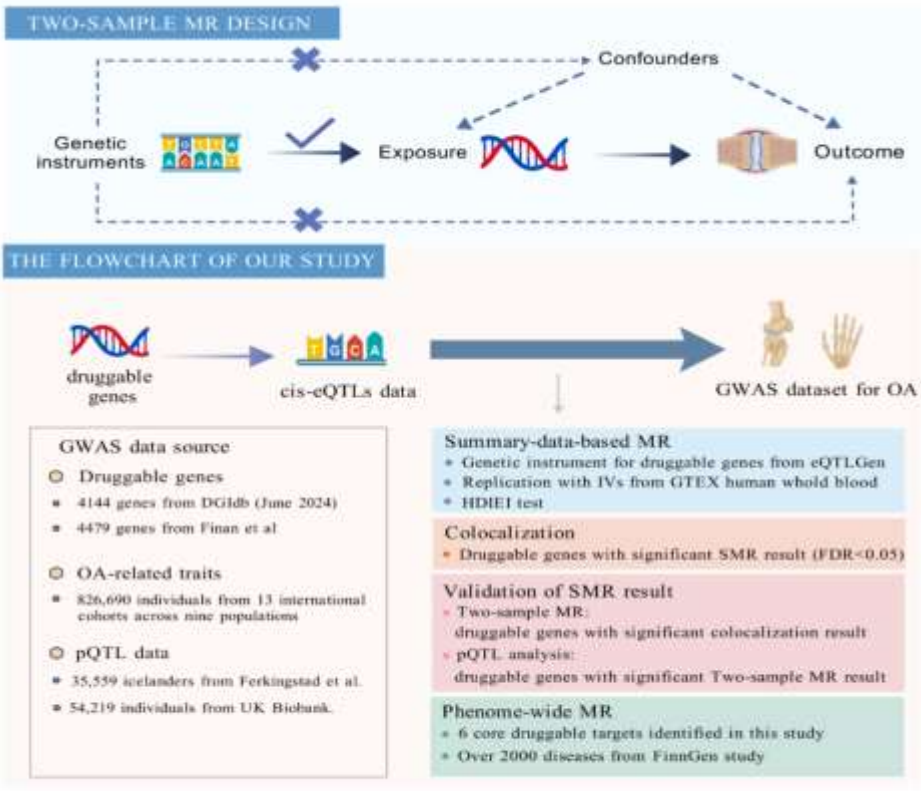

Figure 1

The datasets used were summary data; all informed consent and ethical approval were obtained in the original studies. We have adhered to the STROBE-MR guidelines and have included the STROBE-MR checklist.

Identification of druggable genes

The druggable genes in our study were derived from Drug–Gene Interaction Database (DGIdb V.5.0.7, available at https://www.dgidb.org/)[11] and the research report by Finan. C et al[4]. DGIdb is an online resource that provides information on drug-gene interactions from publications, databases, and other web-based sources[11]. We downloaded the Interactions data (released in June 2024) from DGIdb, which includes all druggable genes with drug-gene interactions mapped to Entrez genes. Additionally, a compilation of targetable genes derived from a critical review conducted by Finan et al was also incorporated into our study[4]. Ultimately, an aggregation of 6889 unique druggable genes was compile for subsequent analysis. The sources for all druggable genes used in this study could be found in Supplementary Table 1.

Selection of instruments to druggable targets

The MR method utilizes single nucleotide polymorphisms (SNPs) closely associated with exposure as instrumental variables (IVs) to assess the causal effect of exposure on outcomes. Eligible IVs must satisfy three assumptions[12]: (1) IVs are directly related to exposure; (2) IVs are independent of any confounding factors; and (3) IVs should be independent of the outcome. First, we perform 6889 potential druggable genes to obtain the eQTL dataset for druggable genes. Next, due to cis-regulatory elements have more direct and specific biological effects compared to trans-regulatory elements[13], cis-eQTL data for human whole blood were utilized (genetic variants within a 1 Mb range on either side of the coding sequence in druggable genes). This blood cis-eQTL data were obtained from the eQTLGen Consortium[14], comprising 25,482 whole blood samples collected across 37 datasets, predominantly from individuals of European descent. We used the default genome-wide (GW) significance P-value threshold of $5 \times 10{-}8$ to select the top associated eQTL as instrument variants in our SMR analyses.

GWAS datasets for 7 types of OA and 4 OA-related traits

Summary statistics from the largest genome-wide association meta-analysis of OA were used, including 826,690 individuals (177,517 patients with OA) from 13 international cohorts across nine population[2]. We obtained the following types of data from it: 7 types of OA, comprising OA at any site, knee OA, hip OA, knee and/or hip OA, finger OA, thumb OA, hand OA, spine OA and 4 OA-related traits comprising total knee replacement, total hip replacement, total joint replacement, early-onset OA[2]. The sources for all statistical summary datasets of OA used in this study could be found in **Supplementary Table 1**.

Summary-data-based Mendelian randomization and colocalization

Summary-based MR (SMR) method, version 1.31, was used to conduct a 2-sample MR analysis with OA as the outcome and druggable targets at gene expression as the exposure in the command prompt (the code is available on https://yanglab.westlake.edu.cn/software/smr/#Overview)[10]. The QTL effect size (β) of a variant reflects the direction of change in gene expression. $\beta > 0$ means positive association, and $\beta < 0$ means negative association. To adjust for multiple testing and control for genome-wide type I error rates, the FDR correction was applied to the P-values. To estimate whether the associations were influenced by the linkage disequilibrium (LD), the HEIDI test was performed. Associations demonstrating a significance level of $P < 0.01$ in the HEIDI test were excluded, as they might be attributed to LD rather than pleiotropy.

In addition, for the druggable genes exhibiting significant MR results, colocalization analysis was conducted using R package "coloc" (version 5.1.0.1) [15]. This R package provides the posterior probability for five hypotheses (0, 1, 2, 3, 4) of whether a single variant is shared between two traits, where the hypothesis 4 is that both traits are associated with a same genetic variant in the region. The commonly accepted strong threshold of colocalization is Posterior probability of the hypothesis $4 > 0.8$ (PP.H4>0.8). For each leading SNP in the outcome GWAS database, all SNPs within a 1Mb range upstream and downstream were retrieved.

Validation of Two-sample Mendelian Randomization and pQTL analysis

When multiple analytical methods yield similar results, we consider the MR results to be more robust. To further substantiate the results of the SMR analysis, the two-sample MR approach was utilized, leveraging eQTLs of druggable genes as instruments to estimate the effect on outcomes[16]. Fully significant cis-eQTL results (false discovery rate (FDR) <0.05) and allele frequency information were obtained, located within 1 Mb upstream or downstream of a druggable gene's transcription start or end points. These results were extracted from eQTLGen and clumped at $r2 < 0.01$ using European samples from the 1000 Genomes Project in each cis-QTL as the IVs for MR. Subsequently, MR analyses were performed using the "TwoSampleMR" package in R version 4.1.1. The inverse-variance weighted (IVW) MR method was applied for IVs containing more than one variant, and the Wald ratio was used for single IVs, while sensitivity analyses employed the MR-Egger and weighted median methods.

The MR analysis based on pQTL data was also used to further validate the results from SMR analysis. The pQTL data of potential drug target were obtained from Ferkingstad et al[17] and the UK Biobank[18], respectively. pQTL meets the following standards are included: (1) Genome-wide significance ($p < 5*10-8$); (2) Without linkage disequilibrium (clump: $r2 = 0.001$, $kb = 10,000$); (3) cis-acting pQTL. The threshold p-value of significance was set as 0.05.

Classification Hierarchy of genes as Potential Drug Targets

After performing MR analysis, FDR correction, colocalization analysis, Two-sample MR and eQTL analysis, we divided the identified genes into three categories (tiers) based on the following criteria: (1) SMR analysis identifies a causal relationship with osteoarthritis (OA) and shows colocalization (HH.P4 > 0.80); (2) the SMR of genes passes Two-sample MR (p-value < 0.05); (3) the SMR of genes demonstrate pQTL with OA; (4) The direction of effects in SMR analysis are consistent (both beta are greater than 0 or less than 0). Under the prerequisite of meeting the principle of directionality (criteria (4)), genes that pass 1 criteria are tier 1 targets, genes that pass 2 criteria are tier 2 targets, genes that pass 3 criteria are tier 3 targets.

Phenome-wide Mendelian randomization

To forecast drug safety and assess the risk of on-target adverse effects, a phenome-wide Mendelian Randomization (MR) analysis was executed. This approach systematically inferred the causal impacts of the expression of previously identified druggable genes on 1705 disease traits within a European ancestry cohort derived from the FinnGen study[19] (**Supplementary Table 2**). Summary statistics of disease-associated SNPs were procured from the FinnGen GWAS (https://www.finngen.fi/en/access_results).

Enrichment Analysis

To explore the functional characteristics and biological relevance of tier 3 druggable genes, the R package 'clusterProfiler' (version 4.3.2) was used for Gene Ontology (GO) and Kyoto Encyclopedia of Genes and Genomes (KEGG) enrichment studies(20). GO analysis annotates genes or proteins by mapping them to three major categories: biological processes (BP), cellular components (CC), and molecular functions (MF). KEGG analysis primarily focuses on metabolic pathways and signal transduction pathways, providing information on the role of genes in biological pathways.

Drug Prediction

Drug Signatures Database (DSigDB, http://dsigdb.tanlab. org/DSigDBv1.0/)(21) is a sizable database with 22,527 gene sets and 17,389 unique compounds spanning 19,531 genes. The online Enrichr platform provides access to the DSigDB database. Candidate drugs were ranked in the ascending order of their p-values, and p-value < 0.01 was deemed statistically significant.

Molecular Docking

The potential binding between the top candidate drug and tier 3 targets was explored using molecular docking techniques. AutoDock Tool 1.5.7 was used for the docking process and PyMOL for visualization(22). The pdb files of tier 3 genes were retrieved from PDB, followed by docking with AutoDock and visualization with PyMOL. Ligand SMILES files from PubChem were converted into pdb format using the software Open Babel GUI (https://openbabel.org/).

Result

SMR analysis of cis-eQTLs and 7 types of OA and 4 OA-related traits

We initially utilized cis-eQTLs as the genetic instruments for SMR analysis to systematically evaluate the causal relationships between druggable genes and 12 OA-related phenotypes **(Figure 2)** and results included 172 significant associations across 6889 druggable genes and 12 OA-related phenotypes. Initially, we identified 36 genes whose expression levels in whole blood were found to be correlated with Knee OA, Hip OA and OA at any sites, including LYG1, CSK and MAPK3 (corrected for P-value by Bonferroni, P_HEIDI ≥ 0.01).

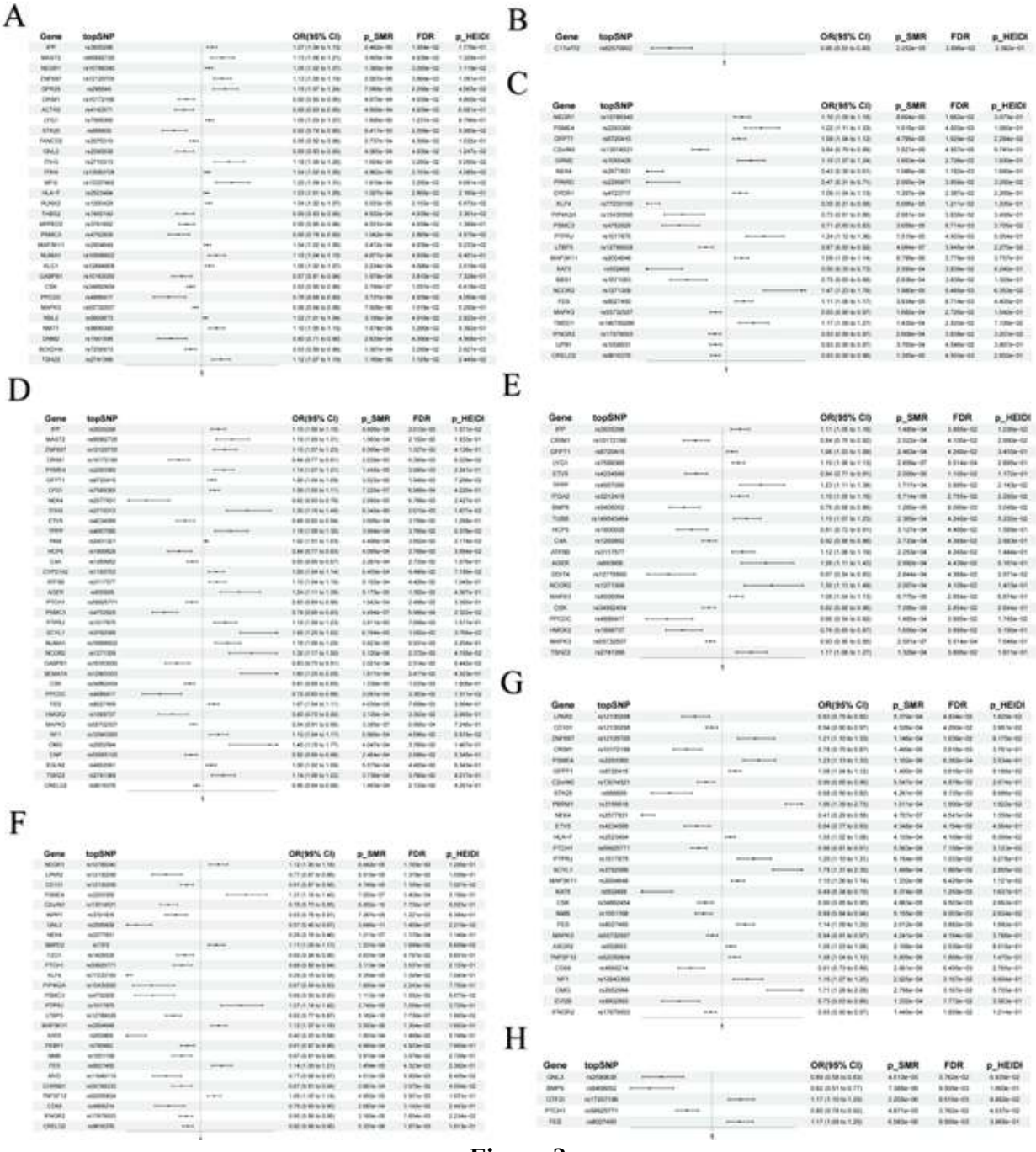

Figure 2

The expression of C2orf40 in blood is negatively correlated with Finger OA. Three types of OA that showed no causal relationships: thumb OA, hand OA, spine OA. Additionally, several genes were identified in 3 OA-related traits. IFNGR2 and MVD were identified in analyses of THR. CSK, MAPK3 and IFNGR2 were associated with TJR. FES was associated with TKR. These genes showed negative correlations with OA-related traits, suggesting that these potential genes could be used in combined treatment of these OA-related traits. All detailed data of SMR could be found in **Supplementary Table 3**.

SMR analysis of Replication cis-eQTLs and 7 types of OA and 4 OA-related traits

Results from the primary analysis dataset using GTEx whole blood and 7 types of OA and 4 OA-related traits, after excluding SNPs with P-HEIDI < 0.01 and P-values > 0.05 via SMR analysis, we identified 19 genes were identified as causally related to knee OA and hip OA including PTCH1, ZNF697, and MAPK3. The inclusion of replication cis-eQTLs in SMR analysis strengthens the validity of our findings. The replication analysis confirmed several genes initially identified, such as MAPK3, across multiple OA traits. SMR analysis of Replication could be found in **Supplementary Table 4**.

Colocalization Results

We conducted colocalization analysis of significant genes from the SMR analysis to further determine the possibility of shared causal genetic variation associated with druggable genes and OA-related traits. The results showed 35 genes with 4 types of OA and 3 OA-related traits showing strong evidence of colocalization (PP.H4>0.8) **(Figure 3)**.

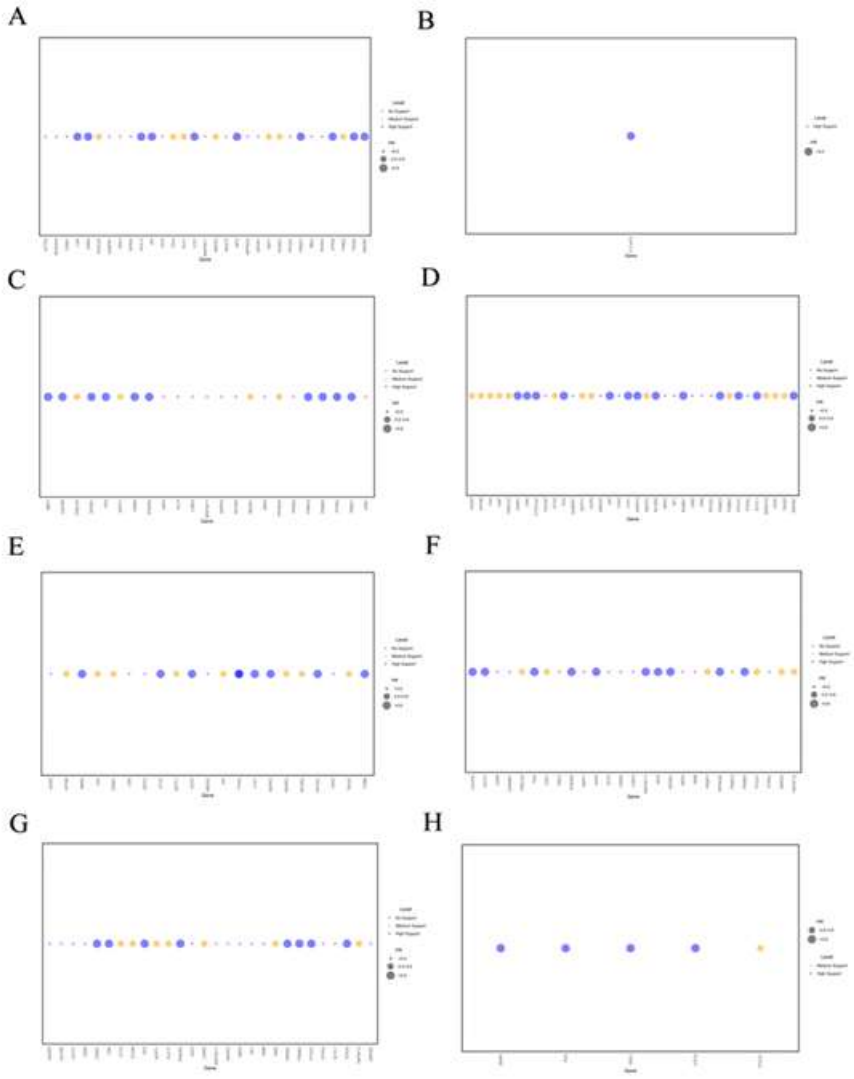

Figure 3

Notably, among those genes, CSK displayed strong degrees of colocalization evidence: for the OA at any site, PP.H4 was 93.9%, knee and/or hip OA, PP.H4 was 85.3%, and for the TJR, PP.H4 was 80.8%. MAPK3 also displayed strong degrees of colocalization evidence: for the knee OA and hip OA, PP.H4 was 81.1. This suggests that CSK and MAPK3 were potential candidate genes for further investigation into its role as a potential causal factor in these OA phenotypes. The detailed colocalization results for all genes were

presented in **Supplementary Table 5**.

Druggable targets supported by two-sample Mendelian Randomization

Two-sample MR were performed on druggable genes that had passed the colocalization analysis to assess the consistency of direction and statistical significance and 23 genes were identified. CSK, IFNGR2, MAPK3, LYG1, ZNF697, C2for40, PSME4 and PTCH1 were associated with multiple OA phenotypes. Two-sample MR results for all genes were presented in **Supplementary Table 6**.

Druggable targets supported by MR analysis of pQTLs

We further performed protein MR analysis and 6 genes were identified. Of the identified genes, CSK was associated with OA at any site, knee OA, hip OA and TJR. MAPK3 was associated with knee OA and hip OA. IFNGR2 was associated with hip OA, THR and TJR. MVD, FES and ITGA2 were associated with THR, TKR and knee OA, accordingly. These results were consistent with both cis-eQTL SMR analysis and colocalization analysis and were categorized as high priority. The results of pQTL analysis for all genes were presented in **Supplementary Table 7**.

Classification Hierarchy of genes as potential drug targets

Guided by colocalization analysis, Two-sample MR and pQTL analysis, the identified genes were divided into three categories. For knee OA, MAPK3 and ITGA2 were tier 3 targets. For knee OA and hip OA, CSK and MAPK3 were tier 3 targets. For TJR, IFNGR2 and CSK were tier 3 targets. For THR, IFNGR2 and MVD were tier 3 targets. For THR, FES was tier 3 targets. Of note, for finger OA, C17orf72 was tier 2 target. The classification hierarchy of detailed Tier 2 and Tier 3 drug targets were presented in **Supplementary Table 8**.

Phenome-wide Mendelian randomization

MR phenome-wide association studies were performed to investigate whether side effects existed in treatments targeting MAPK3, CSK, IFNGR2, MVD, FES and ITGA2 (the tier 3 targets for different OA phenotypes). Over 2000 traits of diseases in the UK Biobank were utilized for MR screening, and the Manhattan plots for the results are presented in **Figure 4**.

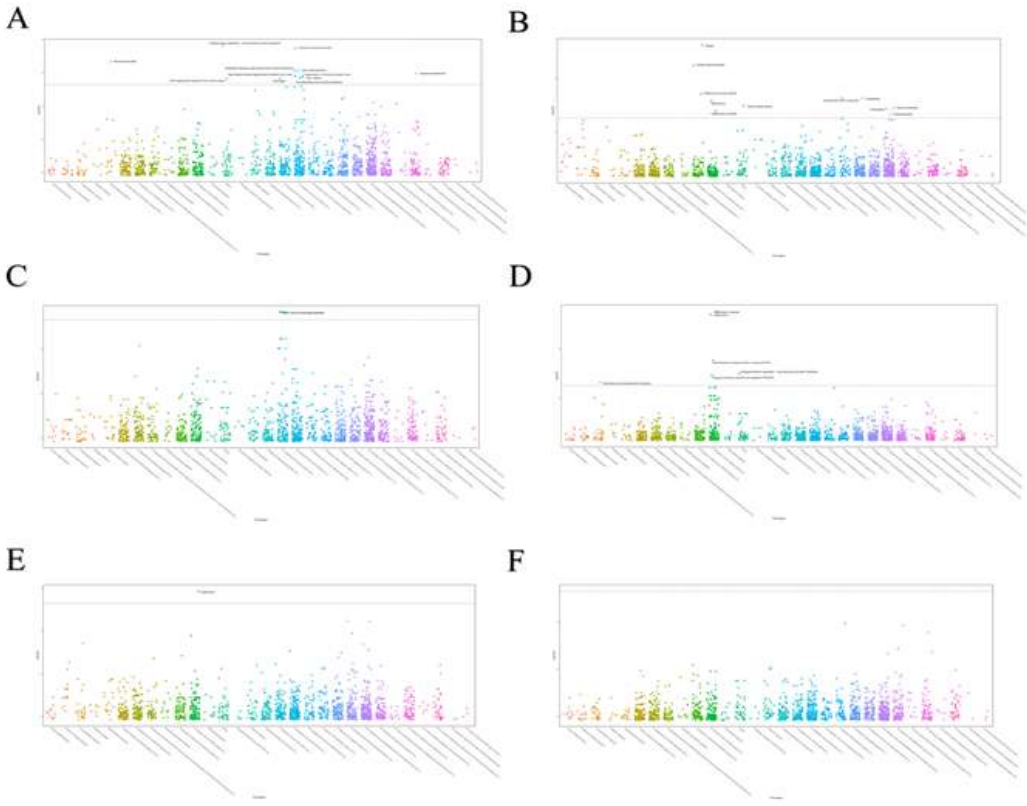

Figure 4

Using IVW method, some significant associations were identified. We discovered that higher CSK levels could increase the risk of Pregnancy hypertension and antihypertensive medication. Higher FES levels might increase the risk of malignant neoplasm of breast, while higher plasma MAPK3 levels could reduce the risk of obesity and joint diseases. Additionally, higher ITGA2 levels might increase the risk of nerve, nerve root and plexus disorders. We did not find significant causal association between MVD and other disease. The results of phenome-wide Mendelian randomization for tier 3 genes were presented in **Supplementary Table 9**.

Enrichment analysis

Through GO analysis of tier 3 targets, we found that these targets are primarily involved in BP such as protein phosphorylation (GO:0006468), phosphorylation (GO:0016310) and cellular response to mechanical stimulus (GO:0071260). The main MF include ATP binding (GO:0005524), non-membrane spanning protein tyrosine kinase activity (GO:0004715), phosphotyrosine residue binding (GO:0001784) and protein tyrosine kinase activity (GO:0004713). KEGG analysis indicates that the target genes were primarily enriched in pathways such as Pathways in cancer (hsa05200), Th1 and Th2 cell differentiation (hsa04658), Th17 cell differentiation (hsa04659) and Osteoclast differentiation (hsa04380).

Drug prediction

DSigDB was used to predict potentially effective intervention drugs and listed the top 10 potential candidate drugs based on the P-values (**Table 1**). The results indicated that mevalonic acid and Zoledronic acid were the two most significant drugs, connected respectively to FES, IPP, MVD, MAPK3 and FES, IPP, MAPK3.

Table 1 Candidate drugs predicted by DSigDB

Drug name	P-value	Adjusted P-value	Genes
mevalonic acid	1.75E-07	2.00E-04	FES; IPP; MVD; MAPK3
Zoledronic acid	2.67E-05	0.015	FES; IPP; MAPK3
phenytoin	2.58E-04	0.077	IFNGR2; ITGA2; MAPK3
VALPROIC ACID	3.79E-04	0.077	C2ORF40; NEGR1; NUMA1; PTCH1; I FNGR2; MFI2; ITGA2; IPP; EPDR1; PTPRJ; ETV5; GNL3; PSME4; CSK; TMED1; ZNF697; MAPK3; GTF2I
vorinostat	4.03E-04	0.077	FES; IFNGR2; CSK; TMED1; GTF2I
deltamethrin	4.70E-04	0.077	PSME4; MAPK3
58-64-0	4.70E-04	0.077	ITGA2; MAPK3
CADMIUM SELENIDE	6.54E-04	0.093	IFNGR2; MAPK3
monensin	0.001	0.121	MVD; ETV5
epinephrine	0.001	0.121	ITGA2; MAPK3

Molecular Docking

We used AutoDock 4.3.2 to analyze the binding sites and interactions between the top 2 candidate drugs and the proteins encoded by the corresponding genes, generating the binding energy for each interaction. We obtained 7 effective docking results between the proteins and drugs (Table 2). Docking amino acid residues and hydrogen bond lengths are shown in **Figure 5**.

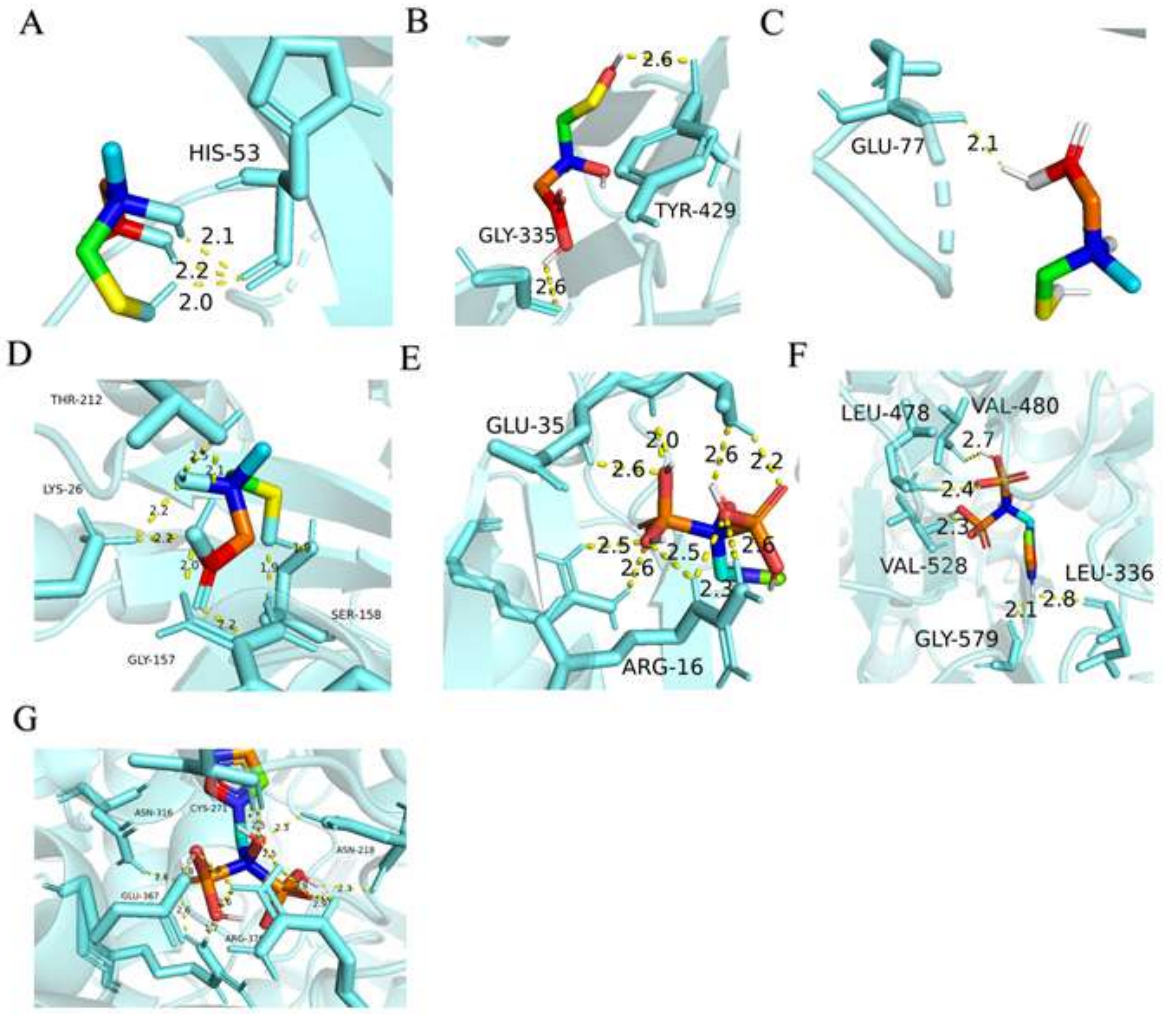

Figure 5

Table 2 Molecular docking results of available proteins and drugs

Target	PDB ID	Drug	Binding energy (kcal/ mol)
FES	7T1L	mevalonic acid	-4.89
FES	7T1L	Zoledronic acid	-6.2
MAPK3	4QTB	mevalonic acid	-4.89
MAPK3	4QTB	Zoledronic acid	-6.76
IPP	AF-Q9Y573-F1	mevalonic acid	-3.97
IPP	AF-Q9Y573-F1	Zoledronic acid	-6.93
MVD	3D4J	mevalonic acid	-6.5

Discussion

Due to the unclear pathophysiology of osteoarthritis (OA), the development of novel therapeutic agents for OA remains highly challenging. In this study, we initially identified potential gene targets through large-scale Mendelian randomization MR analysis, followed by colocalization analysis, and ultimately screened 35 genes as tier 1 targets. Subsequently, we validated tier 2 and tier 3 targets through two-sample MR and pQTL analysis. Our study provides robust evidence that six genes (MAPK3, CSK, FES, MVD, IFNGR2, and ITGA2) are tier 3 genes and can serve as potential druggable target genes with causal relationships to 3 types of OA and 3 OA-related traits.

MAPK3 gene encodes a member of the MAP kinase family. MAP kinases, also known as extracellular signal-regulated kinases (ERKs), act in a signaling cascade that regulates various cellular processes such as proliferation,

differentiation, and cell cycle progression in response to a variety of extracellular signals(23). In this study, MAPK3 (ERK1) was identified as a tier 3 target with protective effects on the knee OA and hip OA. Consistently with our study, zhou and chao et al utilized plasma pQTLs GWAS data and identified MAPK3 (ERK1) as a protective factor for Knee OA and Hip OA(24). Additionally, Previous studies have shown that MAPK3 deficient chondrocytes downregulated the expression of NRF2 and upregulated the expression of BACH1, and the lack of MAPK3 can promote cartilage degradation and accelerate the progression of OA(25). Meanwhile, researches have suggested that ERK1/2-mediated DRP1 activation is a key step in mitochondrial network fragmentation and chondrocyte apoptosis, and targeting this step may lead to the development of new therapies for OA(26). Through phenome-wide MR analysis, we did not find significant causal association between MAPK3 and other diseases, and simultaneously, MAPK3 has been found to be a protective factor for obesity and joint disorders. This suggests that MAPK3 may have relatively minor side effects as a drug target for knee OA and hip OA. Currently, Several existing drugs have now been found to exert pharmacological effects through MAPK3, among which acetylsalicylic acid and Sulindac have been used in the treatment of OA(3,27,28). However, from the perspective of clinical treatment outcomes, these drugs primarily serve to alleviate inflammation and reduce pain and have not provided significant efficacy in the complete cure of OA. Therefore, in-depth pathophysiological research on the protective effects of targeting MAPK3 in OA is needed to potentially propose new targeted drug guidance theories for the fundamental cure of OA.

C-terminal Src kinase (CSK) encodes a cytosolic tyrosine-protein kinase that primarily negatively regulates the Src family of tyrosine kinases (SFKs) through phosphorylation. It plays an important role in critical cellular decisions, such as apoptosis, survival, proliferation, and cytoskeletal organization(29). Researches have shown that in a bovine knee osteoarthritis model, excessive compression of articular cartilage causes chondrocyte death through a signaling pathway involving cell adhesion receptors mediated by SFK(30). Additionally, in vivo studies, inhibition of cSrc kinases has been found to effectively protect bone and cartilage in preclinical models of osteoarthritis(31). CSK is expected to alleviate osteoarthritis by phosphorylating Src to inhibit Src-related signaling pathways. Consistent with these results, we provide druggable genetic evidence for the directionally consistent effects of CSK in the blood tissue on OA at any sites, knee OA, hip OA and TJR. Notably, through phenome-wide MR analysis, the risk of pregnancy hypertension was found to increase with higher expression of CSK, while elevated CSK expression is positively correlated with antihypertensive medication. This probably indicates that CSK expression may influence blood pressure changes. However, whether the increased expression of CSK leads to anti-hypertension or pregnancy hypertension requires further research. Dasatinib is a multitargeted drug that inhibits several tyrosine kinases and holds potential for alleviating osteoarthritis; however, its pharmacological mechanisms remain to be studied(32). Overall, our results indicated that CSK is a promising target for treating OA by affecting Src-related signaling pathways with minimal side effects.

Fps/fes proto-oncogene (hereafter referred to as FES), encodes a subgroup IV non-receptor protein-tyrosine kinase(33). Its primary physiological functions include involvement in normal hematopoiesis and inflammatory responses through signaling via growth factor and cytokine receptors(34). Analysis of possible downstream targets of Fes revealed that Fes is a key regulator of terminal macrophage differentiation, capable of inducing macrophage development and differentiation by activating the ets family transcription factor PU.1(35). Of note, the predominant immune cells observed in the OA synovium are T lymphocytes and macrophages, which, along with activated synoviocytes, are responsible for further cytokine production and increased angiogenesis, leading to a vicious cycle that induces secretion of metalloproteinase and proteolytic enzymes, with perpetuation of cartilage degradation(36). This may suggest that in OA pathology, Fes can exacerbate the pathological damage of osteoarthritis by inducing the development and differentiation of macrophages, which triggers a vicious cycle leading to the degradation of cartilage cells. Consistent with these research conclusions, we provide druggable genetic evidence for drug target

genes and demonstrate a positive causal association between FES and TKR using SMR, MR, and pQTL methods. Through phenome-wide MR analysis, we find positively significant causal association between FES and malignant neoplasm of breast. Notably, Sangrar W et al observed that tumor onset in a mouse model of breast epithelial cancer occurred earlier in mice targeted with either null or kinase-inactivating fps/fes mutations(37). Whether increased expression of Fes could potentially elevate the risk of breast cancer remains to be further investigated. Fostamatinib is currently an investigational inhibitor of Fes/Fps; however, due to its multiple targets, its potential use in treating OA remains unclear. Therefore, FES has great in targeted therapy of OA-related traits and warrants further exploration in future research.

Integrin subunit alpha 2 (ITGA2) encodes the principal family of cell surface proteins that interact with the extracellular matrix, these interactions control the adhesion and migration of cells(38). Research showed the ITGA2 gene is important in focal adhesion, alpha6-beta4 integrin signaling, and the inflammatory response pathway in OA and result of PPI network indicated that ITGA2 is associated with various OA-related pathological mechanisms, including focal adhesion, integrin signaling, the inflammatory response pathway, and endochondral ossification, and plays a crucial role in these processes(39). Additionally, a previous report suggested that The expression level of ITGA2 in peripheral blood mononuclear cells of the patient is significantly lower than normal, and ITGA2 is a potential candidate biomarker of OA(40). However, through SMR analysis and two-sample MR validation, our results suggest that ITGA2 is a potential risk factor for OA, while pQTL analysis indicates that ITGA2 is a protective factor for OA. These conflicting results have been also reported in previous studies(40,41). It is considered to possibly be due to ITGA2 having a dual role in the inflammatory response, or it could be an error caused by the inconsistent sample sizes of the eQTL and pQTL data we used. Further mechanistic studies are needed to explore the specific causal relationship between ITGA2 and OA. Notably, phenome-wide MR analysis revealed a causal relationship between increased ITGA2 expression and neurological diseases, suggesting a potential elevated risk of developing such conditions. Overall, whether ITGA2 can serve as an effective therapeutic target for osteoarthritis still requires further pharmacological studies.

Our study has also discovered several previously unreported genes associated with OA: Interferon Gamma Receptor 2 (IFNGR2) and Mevalonate Diphosphate Decarboxylase (MVD). Specifically, we found that increased expression of IFNGR2 and MVD is likely to lead to decreased OA. IFNGR2 encodes the beta subunit of the interferon-gamma receptor complex, which is primarily responsible for mediating the signal transduction of interferon-gamma (IFN-γ)(42). MVD catalyzes the conversion of mevalonate pyrophosphate into isopentenyl pyrophosphate in one of the early steps in cholesterol biosynthesis(43). Although no previous reports have been found on the direct association of these genes with OA, they have well-defined roles in various other diseases, such as rheumatoid arthritis, multiple sclerosis and porokeratosis(44–46). Additionally, Research suggested that the upregulation of IFNGR2 may lead to rheumatoid arthritis by regulating various immune cells to induce inflammation(45), indicating that IFNGR2 may also contribute to OA through the modulation of inflammation. However, in our study, IFNGR2 and MVD were observed to be a protective factor in OA. This discrepancy may be attributed to the limited data in our study; nevertheless, it also offers a new perspective for further exploring the pathogenesis of IFNGR2 and OA. While our findings provide new insights into the potential involvement of these genes in OA development, further studies are needed to determine their exact role in the disease.

In this study, DSigDB database predicted 10 potential drugs for migraine, among which current clinical research is mainly focused on zoledronic acid. ClinicalTrials (https:// clinicaltrials.gov/) has registered multiple studies on the efficacy of zoledronic acid for osteoarthritis. Many findings differ differently and controversially. A published clinical study indicated that intravenous zoledronic acid does not protect patients with symptomatic knee osteoarthritis from total knee replacement surgery(47). However, a randomized controlled trial examining the combination of zoledronic acid and methylprednisolone treatment for OA indicated that intravenous administration

of both methylprednisolone and zoledronic acid might have a beneficial impact on the symptoms of knee OA(48). Overall, existing studies suggest that zoledronic acid tends to function more as an adjunctive treatment for OA, with its direct therapeutic effects still requiring further in-depth investigation. Meanwhile, Mevalonic Acid ranked highest among the 10 predicted drugs. However, it is regrettable that this drug has not been found to have a relevant connection with OA at present, and further research is still needed. Notably, valproic acid has been found to be linked to 18 potential drug targets, making it the predicted drug with the most connections to drug targets. Research indicated that valproic acid may improve LPS-induced chondrocyte damage by regulating the miR-302d-3p/ITGB4 pair and inactivating the PI3K-AKT pathway(49). Earlier research also proposed SCRG1 as a novel drug target for OA synovitis and identifies valproic acid as a targeted therapeutic agent for SCRG1(50). This suggests that valproic acid is a potential drug for the treatment of OA.

There are several limitations to be recognized in this study. First, the number of IVs in MR for eQTL is limited, and most are no more than three SNPs, which limits the level of confidence in the MR results. Secondly, due to the lack of individual-level statistical data from OA GWAS, our ability to exclude non-European participants is limited. Nevertheless, European participants account for most of the total sample, so the possibility of racial issues having a significant impact on our findings is relatively small. In addition, while detailed sensitivity checks indicated generally stable correlations between adjustable factors, represented by various genetic markers, with little evidence of horizontal pleiotropy, the possibility of horizontal pleiotropy cannot be entirely dismissed. Finally, this study used publicly available datasets, which may limit novelty because these datasets cannot provide new and unique resources for research. However, through careful data processing and analysis of public databases, it can provide valuable new perspectives for existing research.

Conclusions

In conclusion, we identified potential druggable genes that presented causal association with 7 types of OA and 4 OA-related traits using SMR. With the help of colocalization analysis, Two-sample MR and pQTL analysis, we identified 6 genes as tier 3 targets: MAPK3, CSK, IFNGR2, MVD, FES and ITGA2. These genes were considered to have the potential to become druggable genes for OA. However, further basic studies are needed to validate these findings and provide new insights into targeted therapy for different OA phenotypes.

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Conflicts of Interest

The authors declare no competing interests.

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