Medicine®

SYSTEMATIC REVIEW AND META-ANALYSIS

OPEN

Single Nucleotide Polymorphisms and Osteoarthritis

An Overview and a Meta-Analysis

Ting Wang, BSci, Yuting Liang, MD, Hong Li, MD, Haibo Li, MD, Quanze He, PhD, Ying Xue, MD, Cong Shen, MD, Chunhua Zhang, MD, Jingjing Xiang, PhD, Jie Ding, MD, Longwei Qiao, MD, and Qiping Zheng, MD, PhD

Abstract: Osteoarthritis (OA) is a complex disorder characterized by degenerative articular cartilage and is largely attributed to genetic risk factors. Single nucleotide polymorphisms (SNPs) are common DNA variants that have shown promising and efficiency, compared with positional cloning, to map candidate genes of complex diseases, including OA.

In this study, we aim to provide an overview of multiple SNPs from a number of genes that have recently been linked to OA susceptibility. We also performed a comprehensive meta-analysis to evaluate the association of SNP rs7639618 of double von Willebrand factor A domains (DVWA) gene with OA susceptibility.

A systematic search of studies on the association of SNPs with susceptibility to OA was conducted in PubMed and Google scholar. Studies subjected to meta-analysis include human and case-control studies that met the Hardy–Weinberg equilibrium model and provide sufficient data to calculate an odds ratio (OR). A total of 9500 OA cases and 9365 controls in 7 case-control studies relating to SNP rs7639618 were included in this study and the ORs with 95% confidence intervals (CIs) were calculated.

Over 50 SNPs from different genes have been shown to be associated with either hip (23), or knee (20), or both (13) OA. The ORs of these SNPs for OA and the subtypes are not consistent. As to SNP rs7639618 of DVWA, increased knee OA risk was observed in all genetic models analyzed. Specifically, people from Asian with G-allele showed significantly increased risk of knee OA (A versus G: OR = 1.28, 95% CI 1.13–1.46; AA versus GG: OR = 1.60, 95% CI 1.25–2.05; GA versus GG: OR = 1.31, 95% CI 1.18–1.44; AA versus GA+GG: OR = 1.34, 95% CI 1.12–1.61; AA+GA versus GG: OR = 1.40, 95% CI 1.19–1.64), but not in Caucasians or with hip OA.

Our results suggest that multiple SNPs play different roles in the pathogenesis of OA and its subtypes; SNP rs7639618 of DVWA gene is associated with a significantly increased risk of knee OA in Asians. Given the limited sample size, further studies are needed to evaluate this observation.

(*Medicine* 95(7):e2811)

Abbreviations: CIs = confidence intervals, DVWA = double von Willebrand factor A domains, HWE = Hardy–Weinberg equilibrium, ORs = odds ratios, SNPs = single nucleotide polymorphisms.

INTRODUCTION

Osteoarthritis (OA) is the most common joint disease worldwide, affecting approximately 10% of men and 18% of women over 60 years of age.[1,2] Multiple factors, including advanced age, excess body weight, repeated trauma or surgery to the joint structures, abnormal joints at birth, gout, diabetes, and hormone-related disorders, have been demonstrated to contribute to increased risk of OA.[3,4] Previous epidemiological studies from twin-pair and family-based segregation analyses have provided clear evidence of a heritable component in susceptibility of OA. However, the specific genetic factors that lead to OA are currently largely unknown. It therefore remains a challenge to identify candidate genes or risk alleles that contribute to OA pathogenesis.

Identification of candidate genes responsible for numerous monogenic disorders has been successful over the past decades with the technology of positional cloning.[5,6] Similar strategy has been used to target many complex diseases, including asthma, heart disease, cancer, and OA.[5,7,8] However, due to lack of suitable genetic markers, not many candidate genes have been identified that show a clear OA etiology. Single nucleotide polymorphisms (SNPs) are common genomic DNA variations within a population. SNPs, in combination with genome-wide association studies (GWAS), have significantly accelerated complex disease gene localization.[5,8,9] SNPs of genes *COL11-A1*, *VEGF, GDF5*, and IL-8, etc., have been associated with OA. Given the heterogeneity and complexity of OA, it is not surprising that these gene polymorphisms showed different levels of association with increased risk of OA. Some polymorphisms may be specific to OA subtypes (hip-, knee-, or hand-OA) and ethnic groups, but the results are with a wide range of discrepancy.

Recently, multiple studies have indicated an association of double von Willebrand factor A domains (DVWA) gene with susceptibility to OA. DVWA gene variants encode 2 protein isoforms, the long (L-DVWA, 385 amino acids) and short (S-DVWA, 276 amino acids) proteins. DVWA protein is predicted to have 2 domains homologous to the VWA domain,

Editor: Aaron James.
Received: November 4, 2015; revised: January 15, 2016; accepted: January 16, 2016.
From the Center for Reproduction and Genetics (TW, HL, HL, QH, YX, CS, CZ, JX, JD, LQ), Suzhou Hospital affiliated to Nanjing Medical University, Suzhou, Jiangsu; Department of Laboratory Medicine (YL), Shanghai First People's Hospital, School of Medicine, Shanghai Jiaotong University, Shanghai; Department of Hematology and Hematological Laboratory Science (QZ), Jiangsu Key Laboratory of Medical Science and Laboratory Medicine, School of Medicine, Jiangsu University, Zhenjiang, China (QZ); and Department of Anatomy and Cell Biology (QZ), Rush University Medical Center, Chicago, IL.
Correspondence: Longwei Qiao, Center for Reproduction and Genetics, Suzhou Hospital affiliated to Nanjing Medical University, Suzhou, Jiangsu 215002, China (e-mail: qiaolongwei@aliyun.com).
Qiping Zheng, Department of Hematology and Hematological Laboratory Science, Jiangsu Key Laboratory of Medical Science and Laboratory Medicine, School of Medicine, Jiangsu University, Zhenjiang 212013, China (e-mail: qp_zheng@hotmail.com).
TW and YL contributed equally to this work.
The authors have no conflicts of interest to disclose.

ISSN: 0025-7974
DOI: 10.1097/MD.0000000000002811

which typically is involved in cell adhesion and protein–protein interactions.[10,11] Interestingly, L-DVWA and S-DVWA were mainly expressed in articular cartilage, suggesting a potential function of these isoforms in OA.[12] Indeed, DVWA has been associated with susceptibility to knee OA and linked to OA etiology, possibly by interacting with β-tubulin. Data from later replication study and meta-analyses of one of the DVWA polymorphisms further confirmed its association with OA in European and Asian populations.[13] However, no significant association between DVWA and OA was found in UK patient samples and neither independent association with OA was observed in Europeans.[14] In addition, there are multiple SNPs in DVWA gene with allelic difference. The association between DVWA and OA has been inconsistent due to allelic effect and effect size of case-controlled studies analyzed.[14]

In this study, we summarized and analyzed the SNPs that have recently been associated with OA and calculated their ORs with OA susceptibility. We also provided an updated and comprehensive meta-analysis to evaluate the association of SNP rs7639618 of DVWA with OA susceptibility with consideration of publication bias and the source of heterogeneity.

MATERIALS AND METHODS

Literature Search

A systematic search of studies on the association of SNPs with susceptibility to OA was conducted in PubMed and Google scholar. ''Single Nucleotide Polymorphism'', ''SNP'', and ''Osteoarthritis'' were used as key words for the searching. The odds ratios (ORs) with 95% confidence intervals were calculated on the basis of data provided in the literature. For meta-analysis, the PubMed was searched for eligible articles up to the end of June 2015. Following keywords were used for the searching ''polymorphism'', ''SNP'', ''rs7639618'', ''von Willebrand factor A domains'', ''DVWA'', ''Osteoarthritis'', ''OA.'' Google academic searching was also performed to obtain additional information.[15]

Study Selection

Studies subjected to meta-analysis were selected according to the following inclusion criteria: human studies; studies investigating the association between the SNP rs7639618 and osteoarthritis (OA); case-control studies providing sufficient data on genotypes to calculate an OR; the genotype distribution of the control population met the Hardy–Weinberg equilibrium (HWE) model. Two reviewers were assigned to independently assess the studies using the inclusion criteria and disagreement was subjected to discussion with a third reviewer for a consensus agreement.

Data Extraction

The following data were collected from each eligible study: name of first author, years of publication, ethnicity, OA sites, HWE, number of cases and controls, genotype frequency in cases and controls. Different ethnicity descents were classified as Caucasian and Asian. When HWE in the controls was not reported, an online program (https://ihg.gsf.de/cgi-bin/hw/hwa1.pl) was used to test the HWE by χ^2 test for goodness of fit.[16]

Evaluation of the Study Quality

The quality of the studies was evaluated by 2 reviewers according to the predefined scale for quality assessment, which was modified from previous meta-analysis.[16,17] In this scale, 5 items, including the representativeness of cases, source of controls, sample size, quality control of genotyping methods, HWE, were carefully checked. Total scores were recorded with a range from 0 to 10 and a higher score indicated a better quality of the study. Any disagreement of the score was modified based on discussion between the 2 reviewers.

Statistical Analysis

Crude ORs together with their corresponding 95% CIs were used to assess the strength of association between the rs7639618 SNP and the risk of OA. The pooled ORs were calculated for allelic comparison (A versus G), heterozygote model (GA versus GG), homozygote model (AA versus GG), dominant model (AA+GA versus GG), recessive model (AA versus GA+GG), respectively. Heterogeneity among the trials was analyzed in this study using the Q statistic (significance level of P value <0.10) and the I^2 test (greater than 50% as evidence of significant inconsistency). When significant heterogeneity ($P < 0.10$ or $I^2 > 50\%$) was achieved, the random effect model was used to combine the effect sizes of the included studies. If no significant heterogeneity was found, fixed effect was selected to pool the data. Meta-regression analysis was performed to detect the source of heterogeneity. The variance (τ^2) between studies was used to quantify the degree of heterogeneity and the percentage of τ^2 was used to describe the extent of heterogeneity.[17,18] Sensitivity analyses were performed to identify individual study effect on pooled results and test the reliability of results.[17] In addition, subgroup analyses were stratified by ethnicity. Potential publication bias was estimated using the Begg test and a forest plot was used to analyze and display the results. All calculations were measured and analyzed using the STATA (version 11.0).

Ethical Statement

No ethical approval was needed for this manuscript, as the data used in this review and the meta-analyses have all been published.

RESULTS

Multiple SNPs From a Number of Genes are Associated with OA

Based on the quality of the studies and the selection criteria as described above, we summarized most of the SNPs that have recently been linked to OA susceptibility. As illustrated in Figure 1, there are 56 SNPs from 50 genes or gene loci that have been associated with OA or OA subtypes. Specifically, 23 SNPs from 21 genes, including *COL11A1* (rs1241164, rs4907986, rs2615977),[19,20] *VEGF* (rs833058), and *IGF1* (rs2195239) etc., are associated with hip OA; 20 SNPs from 17 genes, including *GDF5* (rs143383), *ADAMTS14* (rs4747096), and *DVWA* (rs7639618, rs11713836, and rs3773472), etc., are frequently seen in knee OA; while individuals containing 13 SNPs of 12 genes, including *IL8* (rs4073, rs2227306), *TGFβ1* (2227306), and *SMAD3* (rs12901499), etc., tend to develop both hip and knee OA. These results demonstrate involvement of multiple SNPs/genes in susceptibility to OA and its subtypes.

Odds Ratios of Candidate SNPs Associated With OA

To evaluate the strength of association with OA, we have summarized the crude ORs of each of the candidate SNPs with 95% confidence intervals (CIs). Listed in Table 1 are detailed

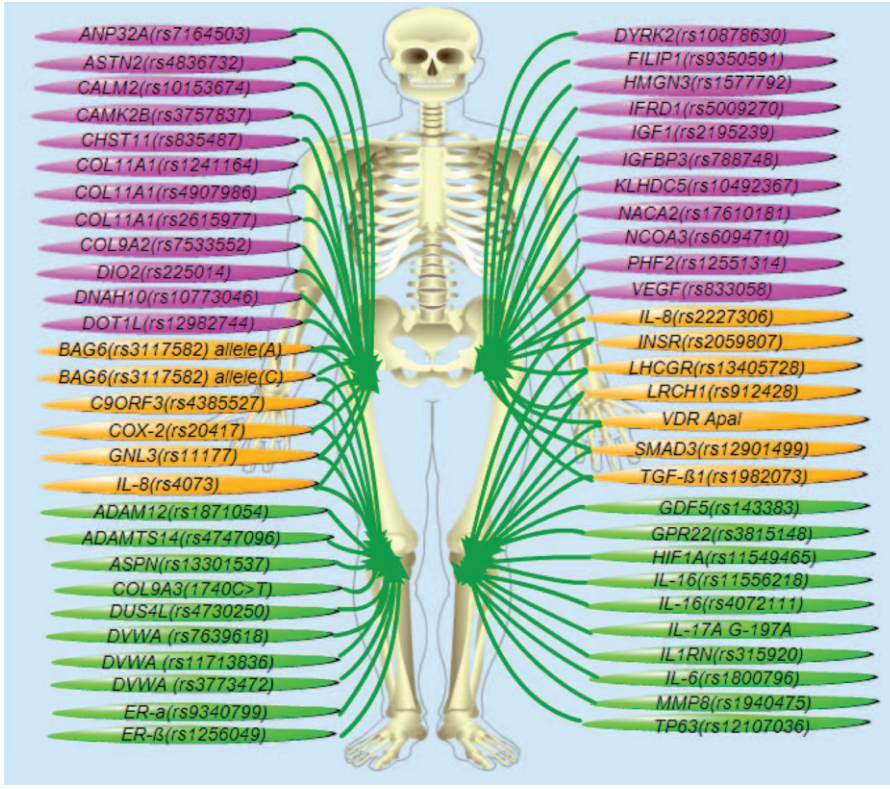

FIGURE 1. Multiple polymorphisms are associated with the risk of OA. Illustrated are 56 SNPs from 50 genes that have been linked to OA risk. Among these, 23 SNPs from 21 genes have been associated with hip OA (pink ones); 20 SNPs from 17 genes are frequently altered in knee OA (green ones); while other 13 SNPs of 12 genes are reportedly associated with both knee- and hip-OA (yellow ones).

TABLE 1. List of Genes and SNPs Associated With OA

Genes	Associated Variant	Nucleotide Change	Diseased Region	OR (95% CI)	Reference
ANP32A	rs7164503	T/C	Hip OA	0.67 (0.53–0.84)	Valdes et al[33]
ASTN2	rs4836732	C/T	Hip OA	1.19 (1.1–1.28)	Zeggini et al[34]
CALM2	rs10153674	A/G	Hip OA	1.65 (1.03–2.65)	Mototani et al[35]
CAMK2B	rs3757837	A/G	Hip OA	1.27 (1.15–1.41)	Evangelou et al[36]
CHST11	rs835487	A/G	Hip OA	1.15 (1.08–1.22)	Zeggini et al[34]
COL11A1	rs1241164	C/ T	Hip OA	0.82 (0.74–0.89)	Rodriguez et al[37]
COL11A1	rs4907986	C /T	Hip OA	1.12 (1.06–1.17)	Rodriguez et al[37]
COL11A1	rs2615977	G/T	Hip OA	1.10 (1.05–1.15)	Panoutsopoulou et al[19]
COL9A2	rs7533552	A/G	Hip OA	2.1 (1.66–2.67)	Nakki et al[38]
D *P* IO2	rs225014	C/T	Hip OA	1.79 (1.37–2.34)	Meulenbelt et al[39]
DNAH10	rs10773046	A/G	Hip OA	1.07 (1.03–1.11)	Evangelou et al[36]
DOT1L	rs12982744	C/G	Hip OA	0.88 (0.82–0.94)	Castano et al[40]
DYRK2	rs10878630	A/G	Hip OA	1.09 (1.04–1.14)	Evangelou et al[36]
FILIP1	rs9350591	C/T	Hip OA	1.2 (1.11–1.3)	Zeggini et al[34]
HMGN3	rs1577792	A/G	Hip OA	1.07 (1.04–1.11)	Evangelou et al[36]
IFRD1	rs5009270	A/G	Hip OA	1.1 (1.06–1.14)	Evangelou et al[36]
IGF1	rs2195239	C/G	Hip OA	1.18 (1.01–1.37)	Limer et al[41]
IGFBP3	rs788748	A/G	Hip OA	0.92 (0.86–0.99)	Evans et al[42]
KLHDC5	rs10492367	G/T	Hip OA	1.18 (1.11–1.27)	Zeggini et al[34]
NACA2	rs17610181	A/G	Hip OA	1.12 (1.06–1.18)	Evangelou et al[36]
NCOA3	rs6094710	A/G	Hip OA	1.28 (1.18–1.39)	Evangelou et al[36]
PHF2	rs12551314	A/C	Hip OA	1.16 (1.07–1.25)	Evangelou et al[36]

Genes	Associated Variant	Nucleotide Change	Diseased Region	OR (95% CI)	Reference
VEGF	rs833058	C/T	Hip OA	0.92 (0.88–0.97)	Rodriguez et al[37]
ADAM12	rs1871054	C/T	Knee OA	1.84 (1.57–2.23)	Wang et al[43]
ADAMTS14	rs4747096	A/G/T	Knee OA	1.25 (1.05–1.5)	Rodriguez et al[44]
ASPN	rs13301537	A/G	Knee OA	1.61 (1.32–1.97)	Liang et al[45]
COL9A3 (1740C>T)	rs61734651	C/T	Knee OA	1.31 (1.05–1.63)	Ikeda et al[46]
DUS4L	rs4730250	A/G	Knee OA	1.17 (1.11–1.24)	Evangelou et al[47]
DVWA	rs7639618	C/T	Knee OA	1.29 (1.15–1.45)	Miyamoto et al[22]
DVWA	rs11713836	A/G	Knee OA	1.37 (1.16–1.63)	Miyamoto et al[22]
DVWA	rs3773472	C/G	Knee OA	1.44 (1.22–1.7)	Miyamoto et al[22]
ER-α	rs9340799	A/G	Knee OA	1.47 (1.08–1.99)	Ren et al[48]
ER-β	rs1256049	G/A	Knee OA	1.68 (1.16–2.44)	Lee et al[49]
GDF5	rs143383	C/T	Knee OA	1.18 (1.1–1.27)	Zhang et al[50]
GPR22	rs3815148	A/C	Knee OA	1.14 (1.09–1.19)	Kerkhof et al[51]
HIF1A	rs11549465	C/T	Knee OA	5.03 (1.53–18.2)	Fernandez et al[52]
IL-16	rs11556218	G/T	Knee OA	0.77 (0.66–0.9)	Luo et al[53]
IL-16	rs4072111	A/G	Knee OA	0.69 (0.58–0.81)	Luo et al[53]
IL-17A G-197A	rs2275913	A/G	Knee OA	0.71 (0.53–0.96)	Han et al[54]
IL1RN	rs315920	C/T	Knee OA	1.18 (1–1.39)	Limer et al[41]
IL-6	rs1800796	G/C	Knee OA	0.51 (0.32–0.8)	Fernandes et al[55]
MMP8	rs1940475	C/T	Knee OA	0.72 (0.57–0.90)	Nakki et al[56]
SMAD3	rs12901499	A/G	Hip OA	1.22 (1.09–1.36)	Valdes et al[57]
SMAD3	rs12901499	A/G	Knee OA	1.22 (1.12–1.34)	Valdes et al[57]
TP63	rs12107036	A/G	Knee OA	1.23 (1.13–1.35)	Zeggini et al[34]
BAG6	rs3117582	C /A	OA	0.63 (0.045–0.89)	Etokebe et al[58]
BAG6	rs3117582	A/C	OA	1.58 (1.12–2.23)	Etokebe et al[58]
C9ORF3	rs4385527	C/T	OA	0.62 (0.52–0.73)	Cui et al[59]
COX-2	rs20417	C/G	OA	0.57 (0.43–0.75)	Schneider et al[60]
GNL3	rs11177	C/T	OA	1.16 (1.11–1.22)	Zeggini et al[34]
IL-8	rs4073	A/T	OA	1.41 (1.02–1.94)	Yu et al[61]
IL-8	rs2227306	C/T	OA	1.48 (1.02–2.14)	Yu et al[61]
INSR	rs2059807	C/T	OA	1.15 (1.01–1.31)	Cui et al[59]
LHCGR	rs13405728	A/G	OA	0.82 (0.7–0.95)	Cui et al[59]
LRCH1	rs912428	C/T	OA	1.42 (1.07–189)	Spector et al[62]
VDR Apal	NA	NA	NA	1.16 (1.02–1.32)	Zhu et al[63]
TGF-β1	rs1982073	C/G/T	OA	1.26 (1.13–1.41)	Cong et al[64]

ADAM12 = ADAM metallopeptidase domain 12; ADAMTS14 = ADAM metallopeptidase with thrombospondin type 1 motif 14; ANP32A = aci-acidic nuclear phosphoprotein 32 family member A; ASPN = aspirin; ASTN2 = astrotactin 2; BAG6 = BCL2 associated athanogene 6; C9ORF3 = -chromosome 9 open reading frame 3; CALM2 = calmodulin 2; CAMK2B = calcium/calmodulin-dependent protein kinase II beta; CHST11 = carbohydrate (chondroitin 4) sulfotransferase 11; CIs = confidence intervals; COL11A1 = collagen, type XI, alpha 1; COL9A2 = colla-collagen, type IX, alpha 2; COL9A3 = collagen, type IX, alpha 3; COX-2 = cyclooxygenase 2; DIO2 = deiodinase, iodothyronine, type II; DNAH10 = dynein, axonemal, heavy chain 10; DOT1L = DOT1-like histone H3K79 methyltransferase; DUS4L = dihydrouridine synthase 4-like; DVWA = double von Willebrand factor A domains; DYRK2 = dual specificity tyrosine-(Y)-phosphorylation regulated kinase 2; ER-α = estrogen receptor 1 (alpha); ER-β = estrogen receptor 2 (ER beta); FILIP1 = filamin A interacting protein 1; GDF5 = growth differentiation factor 5; GNL3 = guanine nucleotide binding protein-like 3; GPR22 = G protein-coupled receptor 22; HIF1A = hypoxia inducible factor 1, alpha subunit; HMGN3 = high mobility group nucleosomal binding domain 3; IFRD1 = interferon-related developmental regulator 1; IGF1 = insulin like growth factor 1; IGFBP3 = insulin like growth factor binding protein 3; IL-16 = interleukin 16; IL-17A G-197A = interleukin-17A; IL1RN = interleukin 1 receptor antagonist; IL-6 = interleukin 6; IL-8 = interleukin 8; INSR = insulin receptor; KLHDC5 = kelch domain containing 5; LHCGR = Luteiniz-Luteinizing hormone/choriogonadotropin receptor; LRCH1 = leucine-rich repeats and calponin homology (CH) domain containing 1; MMP8 = ma-matrix metallopeptidase 8; NA = not available; NACA2 = nascent polypeptide-associated complex alpha subunit 2; NCOA3 = nuclear receptor coactivator 3; OR = odds ratios; PHF2 = PHD finger protein 2; SMAD3 = SMAD family member 3; TGF-?1 = transforming growth factor, beta 1; TP63 = tumor protein p63; VDR Apal = vitamin D (1,25-dihydroxyvitamin D3) receptor Apal; VEGF = vascular endothelial growth factor.

information about the names of associated genes, the SNP numbers, locations of OA (knee- or hip-OA), OR values, and literature source. Most of the SNPs associated with hip OA showed a value of odds ratio greater than 1.0, with SNP rs225014 of *DIO2* showing the highest value of 1.79 (1.37–2.34, Figure 2A, Table 1). The odds ratios of SNPs that are associated with knee OA were also calculated and most are above the value of 1.2 with SNP rs4747096 of ADAM12 showing the highest value of 1.84 (1.57–2.23, Figure 2B, Table 1). Similar results were observed for SNPs of genes that are associated with both knee and hip OA (Figure 2C, Table 1), although some SNPs showed an OR value less than 1.0 as seen in groups of hip- or knee-OA (Figure 2, Table 1). Together, these results suggest that multiple SNPs from a variety of genes showed different levels of association with knee and/or hip OA.

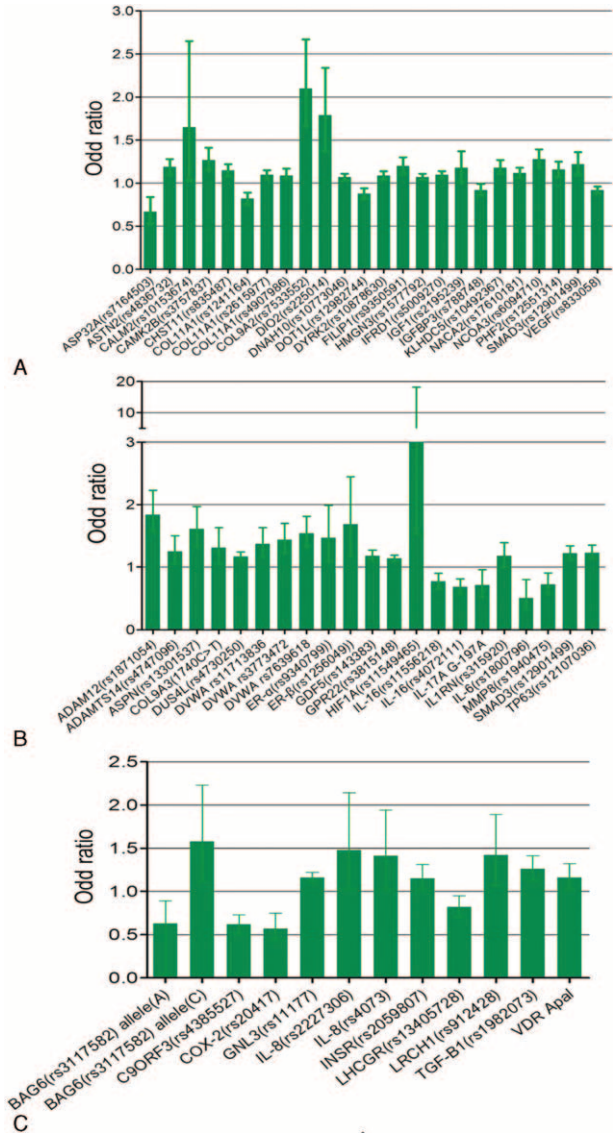

FIGURE 2. The odd ratios (OR) of the SNPs associated with OA or OA subtypes. A, The ORs of most of the SNPs associated with hip OA showed a value greater than 1.0. SNP rs225014 of *DIO2* showed the highest value of 1.79 (1.37–2.34). B, The ORs of SNPs associated with knee OA were mostly above the value of 1.2. SNP rs4747096 of ADAM12 showed the highest value of 1.84 (1.57–2.23). C, SNPs of genes associated with both knee and hip OA showed similar OR values. The OR value of SMAD3 is only available for individual knee- or hip-OA.

Eligible Studies Selected for Meta-Analysis of DVWA and OA

By searching the PubMed and Google scholar database using above key words (SNP, rs7639618, DVWA, and OA), we have identified approximately 50 relevant papers, including 38 studies that potentially show an association between DVWA and OA. After exclusion of duplicated studies and improper studies determined by reading the title and abstracts, 9 studies meet the selection criteria. Due to lack of genotype frequency in 2 of the studies, 7 studies were eventually subjected to meta-analysis of the association between SNP rs7639618 of DVWA and OA. The detailed process of literature screening is outlined in Figure 3. In these studies, 9500 OA cases and 9365 controls were identified according to the inclusion and exclusion criteria. Six knee-OA-related studies contained information from 6807 knee OA cases and 7785 controls,[13,14,21–24] while 3 studies involving hip OA include 2693 hip OA cases and 1580 controls.[13,14,25] The HWE of genotype distribution in the controls was tested in all studies and they were all in consistent with HWE. Detailed information about the OA types, the year and countries the studies conducted, allelic difference, power of HWE, and the quality score of the studies is provided in Table 2.

SNP rs7639618 is Significantly Associated with Knee OA

Through meta-analysis of SNP rs7639618 of DVWA, we observed a significantly increased risk of knee OA susceptibility in allelic comparison in Asians (A versus G: OR = 1.16, 95% CI 1.04–1.30 Figure 4). The OR values for each of the allelic models are listed in Table 2 and illustrated in Figure 5A (homozygote model, AA versus GG: OR = 1.39, 95% CI 1.10–1.76), Figure 5B (heterozygote model, GA versus GG: OR = 1.20, 95% CI 1.11–1.30), Figure 6A (recessive model, AA versus GA+GG: OR = 1.26, 95% CI 1.05–1.50), and Figure 6B (dominant model, AA+GA versus GG: OR = 1.20, 95% CI 1.04–1.39). As shown in Table 3, there was significant heterogeneity between studies, ranging from 0 to 0.029. We therefore performed subgroup analysis according to ethnicity. No heterogeneity was shown in heterozygote model and thus, a fixed model was applied for its pooled OR. For other allelic models that show significant heterogeneity, a randomized effect model was used. The results showed that there was a statistically increased knee OA risk in all allelic models (A versus G: OR = 1.28, 95% CI 1.13–1.46, Figure 4; AA versus GG: OR = 1.60, 95% CI 1.25–2.05, Figure 5A; GA versus GG: OR = 1.31, 95% CI 1.18–1.44, Figure 5B; AA versus GA+GG: OR = 1.34, 95% CI 1.12–1.61 Figure 6A; AA+GA versus GG: OR = 1.40, 95% CI 1.19–1.64, Figure 6B and Table 3). The results in Asians were similar to that of overall comparisons of pooled eligible researches (Table 3), while in Caucasians, even with increased sample size, there is no significant association in any allelic models compared as illustrated in Figure 4 (A versus G: OR = 1.00, 95% CI 0.87–1.16), Figure 5A (AA versus GG: OR = 0.96, 95% CI 0.61–1.51), Figure 5B (GA versus GG: OR = 1.04, 95% CI 0.92–1.19), Figure 6A (AA versus GA+GG: OR = 0.95, 95% CI 0.62–1.46), Figure 6B (AA+GA GA versus GG: OR = 1.00, 95% CI 0.82–1.22), and Table 3. Together, these results support that SNP rs7639618 of DVWA was only associated with an increased risk of knee OA in Asians.

No Significant Association Between SNP rs7639618 and Hip OA

The association between SNP rs7639618 of DVWA and the risk of hip OA was analyzed in 3 independent studies. Random-effects model was used in the dominant model and heterozygote model due to the presence of heterogeneity, while fixed-effects model was used in other models without significant heterogeneity. Overall, no significant association was identified in any of the allelic models analyzed (A versus G: OR = 0.98, 95% CI 0.89–1.08, AA versus GG: OR = 1.03, 95% CI 0.77–1.37, GA versus GG: OR = 0.89, 95% CI 0.74–1.07,

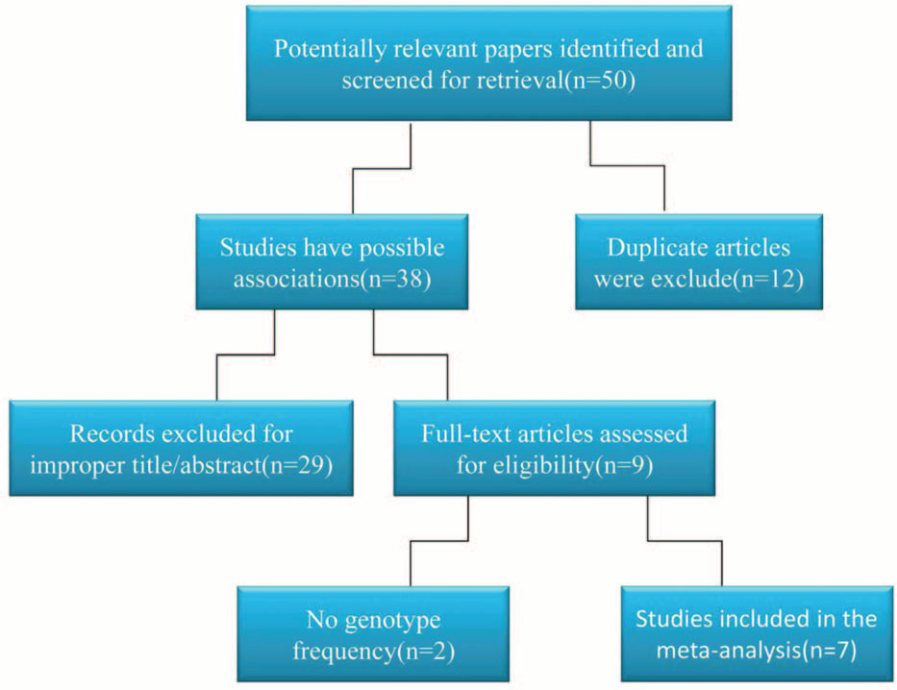

FIGURE 3. Flow chart of study selection. Studies selected for current meta-analysis are as illustrated. Thirty-eight studies from 50 relevant papers show a potential association between DVWA and OA. Only 9 studies meet the selection criteria. Seven studies were eventually subjected to meta-analysis as 2 of the studies lack genotype frequency.

AA versus GA+GG: OR = 1.10, 95% CI 0.85–1.41, and AA+GA versus GG: OR = 0.91, 95% CI 0.75–1.09, Figure 7). Five studies in 2 papers were carried out in Caucasian population. Subgroup analysis of Caucasian population was also assessed, and there is no significant association between SNP rs7639618 and hip OA in any allelic models analyzed (A versus G: OR = 1.00, 95% CI 0.89–1.12, AA versus GG: OR = 1.25, 95% CI 0.83–1.89, GA versus GG: OR = 0.91,

TABLE 2. Characteristics of Eligible Studies

Studies and Subtypes	Year	Country	Case			Control			P for HWE	Quality
Knee OA			GG	GA	AA	GG	GA	AA		
Valdes[14]	2008	Chingford	188	68	8	357	143	12	0.820	6.5
Valdes[14]	2008	Nottingham	505	201	27	475	169	11	0.350	6.5
Miyamoto[22]	2008	Japan	99	107	36	166	222	95	0.184	10
Miyamoto[22]	2008	China	145	187	85	106	192	115	0.156	10
Meulenbelt[13]	2009	UK	275	85	6	504	215	19	0.487	5
Meulenbelt[13]	2009	The Netherlands	98	36	3	538	188	13	0.459	5
Meulenbelt[13]	2009	Spain	171	72	6	189	70	12	0.103	5
Meulenbelt[13]	2009	Greece	280	80	8	291	97	11	0.401	5
Meulenbelt[13]	2009	Japan 1	253	293	95	162	327	140	0.304	5
Meulenbelt[13]	2009	Japan 2	99	107	36	166	222	95	0.184	5
Meulenbelt[13]	2009	China	145	187	85	106	192	115	0.156	5
Takahashi[23]	2010	Japan	369	428	136	397	600	228	0.960	5
Lee[24]	2013	Korea	519	857	361	212	374	139	0.835	7
Bravata[21]	2015	Sicilian	41	18	2	72	25	3	0.390	6
Hip OA										
Valdes[14]	2008	Chingford	34	15	1	357	143	12	0.820	6.5
Valdes[14]	2008	Nottingham	544	234	15	475	169	11	0.350	6.5
Meulenbelt[13]	2009	UK	808	288	20	504	215	19	0.487	5
Meulenbelt[13]	2009	The Netherlands	67	31	1	538	188	13	0.459	5
Meulenbelt[13]	2009	Spain	175	84	8	189	70	12	0.103	5
Zhu[25]	2010	China	81	185	102	106	192	115	0.160	7

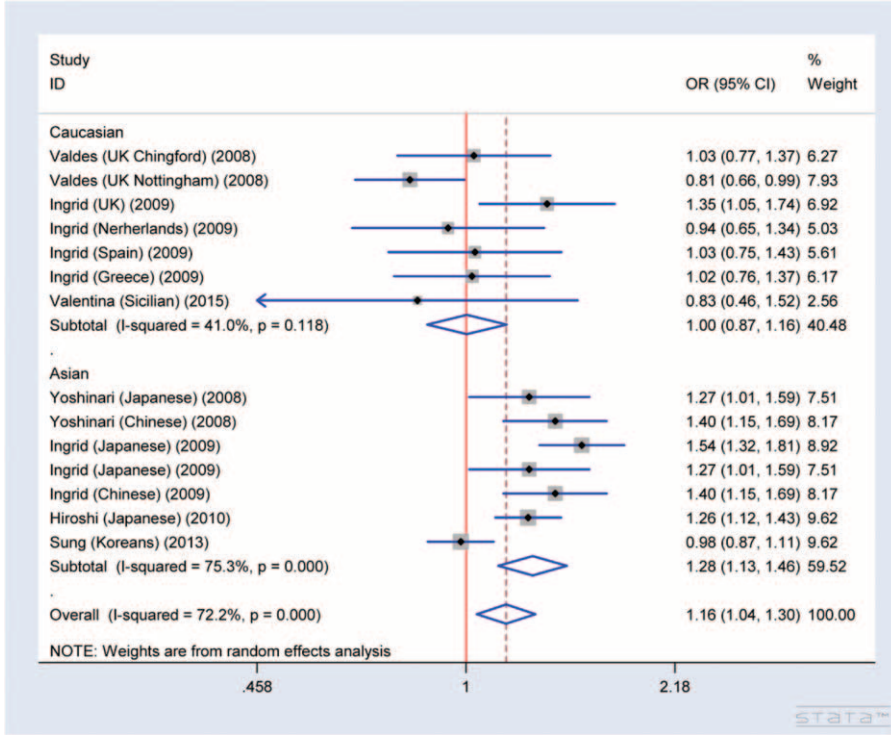

FIGURE 4. Forest plot of allele comparison of DVWA rs7639618 for overall comparison (A versus G, association of rs7639618 and knee OA). SNP rs7639618 is associated with a significantly increased risk of knee OA in allelic comparison in Asian (A versus G: OR: 1.16, 95% CI 1.04–1.30), and in all allelic models (A versus G: OR: 1.28, 95% CI 1.13–1.46), but not in Caucasians (A versus G: OR: 1.00, 95% CI 0.87–1.16).

95% CI 0.73–1.13, AA versus GA+GG: OR = 1.27, 95% CI 0.84–1.92, and AA+GA versus GG: OR = 0.92, 95% CI 0.74–1.15, Figure 7).

Test of Heterogeneity and Sensitivity

As significant heterogeneity was shown between studies in each comparison, we therefore investigated the source of heterogeneity by ethnicity, year of report, and sample size using meta-regression analysis and subjected to allelic comparison (A versus G). Studies containing more than 1000 participants were categorized as ''large,'' while studies with less than 1000 participants were categorized as ''small.'' Meanwhile, group of cases with an average age greater than 65 were categorized as ''high-risk,'' while cases younger than 65 were assigned to ''low-risk'' group. Group of ''mixed'' cases indicate that no data of age are available. The results suggested that ethnicity ($P = 0.002$), group average age ($P = 0.024$), and the year the study conducted ($P = 0.038$), but not the sample size ($P = 0.438$), contributed to the source of heterogeneity. In addition, ethnicity, the average age, and year of study could explain 37% of the variance (τ^2). We also performed sensitivity analysis to evaluate whether the present meta-analysis is stable and the results showed that no individual study affected the pooled OR (Figure 8A).

Publication Bias

A Begg funnel plot and a Begg test were used to assess for publication bias for all allelic models. The result showed no obvious asymmetry, indicating no publication bias (Figure 8B).

DISCUSSION

OA is characterized by progressive cartilage matrix degradation, subchondral bone sclerosis, and osteophyte formation.[26,27] These are nonreversible processes due to the limited repair capacity of cartilage. With the disease advances, pain becomes the most prominent symptom which is almost unbearable and eventually leads to joint replacement.[27] Thanks to the better life and medical conditions, people live longer nowadays, but it also raises the incidence and the population suffering from OA. Hence, OA is a major source of pain, disability, and a tremendous socioeconomic burden worldwide.[1] Unfortunately, given the heterogeneity of multiple subtypes and the complexity of OA pathogenesis, it remains a challenge to find unanimous biomarkers that help with the early diagnosis and targeted therapy for all OA patients. Recently, personalized medicine becomes a hot topic in health care and may be achieved due to existence of SNPs, the most common genetic variations, within a population.[28] There are many SNPs within different genes that have shown various associations with OA. Identification of these gene SNPs and their correlation with OA would further our understanding of the molecular mechanisms involved in the pathogenesis of OA, so as to develop better diagnostics and more targeted therapy at early stages of OA.

In this effort, we provided an overview of multiple SNPs in association with OA susceptibility. We identified more than 50 SNPs from a number of genes that have been linked to either hip (COL11A1, VEGF, etc.), or knee (COL9A3, ASPN, GDF5, etc.), or both (IL-8, TGF-β1, etc.) OA. Among these genes, *GDF5*, which encodes the growth differentiation factor 5, is a

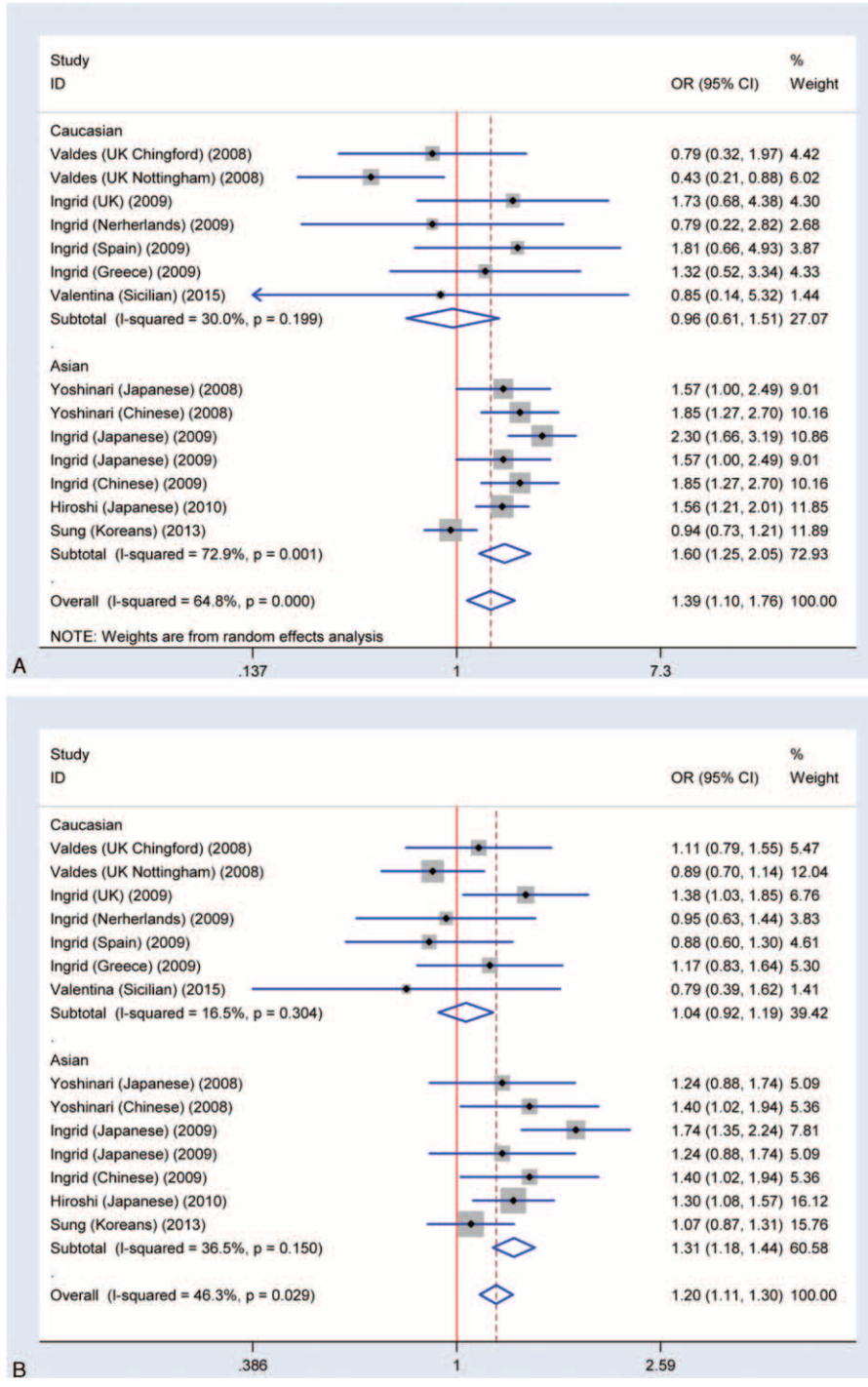

FIGURE 5. Meta-analysis of the association rs7639618 and knee OA. A, Forest plot of homozygote model for overall comparison (AA versus GG: OR: 1.39, 95% CI 1.10–1.76). B, Forest plot of heterozygote model for overall comparison (GA versus GG: OR: 1.20, 95% CI 1.11–1.30).

member of the transforming growth factor-β superfamily. GDF5 has previously been shown to play a role in development and maintenance of bone and cartilage.[14] Notably, SNP rs143383 of GDF5 is known to be associated with high risk of OA.[14] The difference of GDF5 SNPs in relationship with OA subtypes and ethnic groups was also observed.[29] In a recent GWAS associated meta-analysis of OA candidate genes, *COL11A1* and *VEGF* were significantly associated with OA. Interestingly, SNPs rs4907986 and rs1241164 of *COL11A1* and SNP rs833058 of *VEGF* all showed association with hip OA.[14] It was previously shown that mutation in *Col11a1* may cause deposition of degraded type II collagen in articular cartilage and

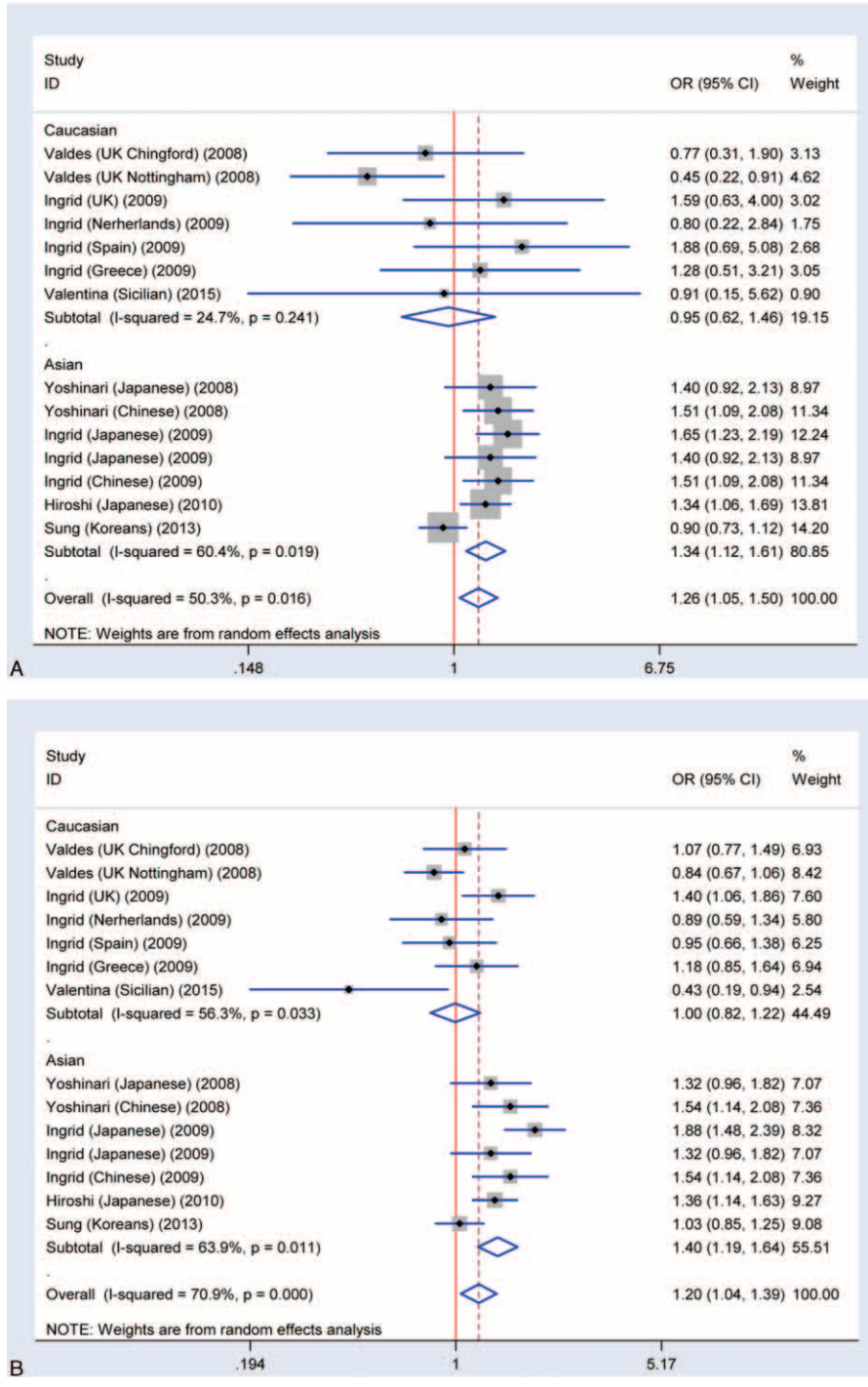

FIGURE 6. Meta-analysis of the association rs7639618 and knee OA. A, Forest plot of recessive model for overall comparison (AA versus GA+GG: OR: 1.26, 95% CI 1.05–1.50). B, Forest plot of dominant model for overall comparison (AA+GA versus GG: OR: 1.20, 95% CI 1.04–1.39).

eventually lead to OA.[30] While it is relatively established for a role of GDF5, COL11A1, and VEGF in OA pathogenesis, the association between SNPs of these genes with OA susceptibility is not always consistent. SNP rs2615977 of COL11A1 was also shown to be associated with OA. However, another COL11A1 SNP rs1676486, which has been linked to lumbar disc herniation (LDH, also a degenerative musculoskeletal disease), was found not associated with OA.[20] In our analysis, we tried to summarize most of the gene SNPs that have shown some correlation with OA or its subtypes. The OR of many of these SNPs/genes are not always consistent, further meta-analysis of multiple studies may overcome the limited sample size and

TABLE 3. Results of Meta-Analysis

	OR	P_H	OR	P_H	OR	P_H	OR	P_H	OR	P_H
Knee OA	A/G		AA/GG		GA/GG		AA/GA+GG		AA+GA/GG	
Overall	1.16 (1.04, 1.30)	0.000	1.39 (1.10, 1.76)	0.000	1.20 (1.11, 1.30)	0.029	1.26 (1.05,1.50)	0.016	1.20 (1.04,1.39)	0.000
Ethnicity										
Asian	1.28 (1.13, 1.46)	0.000	1.60 (1.25, 2.05)	0.001	1.31 (1.18, 1.44)	0.150	1.34 (1.12,1.61)	0.019	1.40 (1.19,1.64)	0.011
Caucasian	1.00 (0.87, 1.16)	0.118	0.96 (0.61, 1.51)	0.199	1.04 (0.92, 1.19)	0.304	0.95 (0.62,1.460)	0.241	1.00 (0.82,1.22)	0.033
Hip OA										
Overall	0.98 (0.89, 1.08)	0.174	1.03 (0.77, 1.37)	0.685	0.89 (0.74, 1.07)	0.105	1.10 (0.85,1.41)	0.853	0.91 (0.75,1.09)	0.086
Caucasian	1.00 (0.89, 1.12)	0.118	1.25 (0.83, 1.89)	0.833	0.91 (0.73, 1.13)	0.084	1.27 (0.84,1.92)	0.878	0.92 (0.74,1.15)	0.066

OA = osteoarthritis; OR = odds ratio; $P_H = P$ value for heterogeneity.

inadequate statistical power of single case-control studies to provide more reliable results.[31]

DVWA gene contains multiple SNPs, including rs11718863, rs7639618, rs7651842, rs7639807, rs17040821, etc. These SNPs may cause protein functional changes or diseases, as described in studies associated with OA.[21] However, as indicated above, the results of their association with OA are not consistent. In this study, we specifically analyzed the association of SNP rs7639618 of DVWA with OA through comprehensive meta-analysis. We have analyzed all available eligible studies that include 9500 OA cases and 9365 controls. Our results confirmed that SNP rs7639618 is associated with a significantly increased risk of knee OA, especially in Asian populations. Similar results were found in subgroup analysis by ethnicity. No evidence was found for the association between rs7639618 SNP and hip OA susceptibility in any genetic allelic models. We have performed heterogeneity analysis and the results showed that ethnicity, average age of case group, and the year of the study were the source of heterogeneity. When we restricted the ethnicity to Asian, there was also heterogeneity, suggesting that ethnicity was not the main source of heterogeneity. We have shown that ethnicity, age, and year of study account for 37% of the variance (τ^2) by meta-regression analysis, while the sensitivity analysis demonstrated that our meta-analysis is stable. In addition, no limitation was made in the search, and the selection bias was well controlled as demonstrated by Begg funnel plot analysis, which showed no publication bias.

In summary, it should be noted that due to limited number of studies on SNP rs7639618 and OA, the relatively small sample size may affect the power and statistics of the meta-analysis.[32] The heterogeneity in some of the genetic/allelic

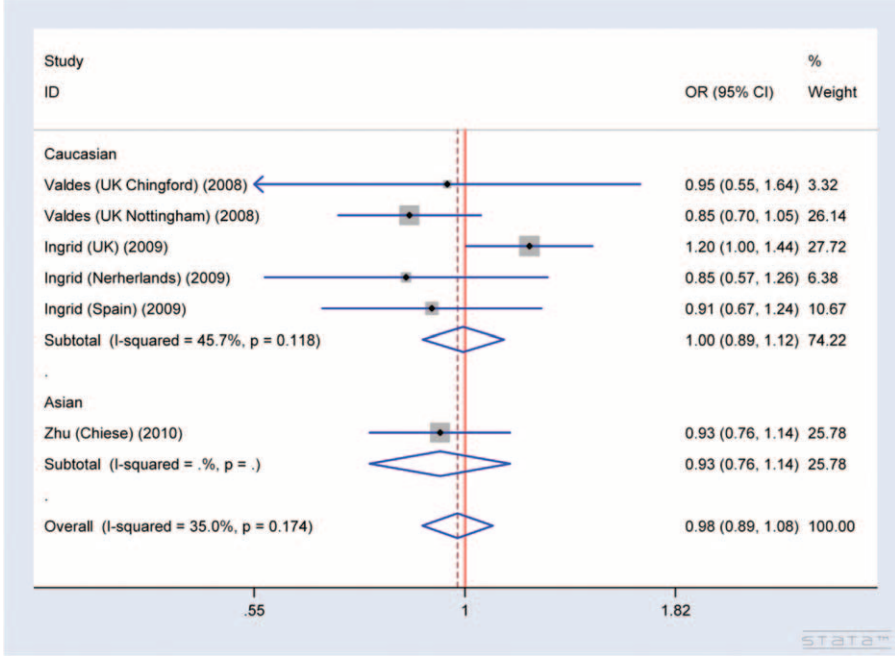

FIGURE 7. Forest plot of allele comparison of DVWA rs7639618 for overall comparison (A versus G, association of rs7639618 and hip OA). No significant association between SNP rs7639618 and hip OA in any allelic models was detected (A versus G: OR: 1.00, 95% CI 0.89–1.12, AA versus GG: OR: 1.25, 95% CI 0.83–1.89, GA versus GG: OR: 0.91, 95% CI 0.73–1.13, AA versus GA+GG: OR: 1.27, 95% CI 0.84–1.92, and AA+GA versus GG: OR: 0.92, 95% CI 0.74–1.15).

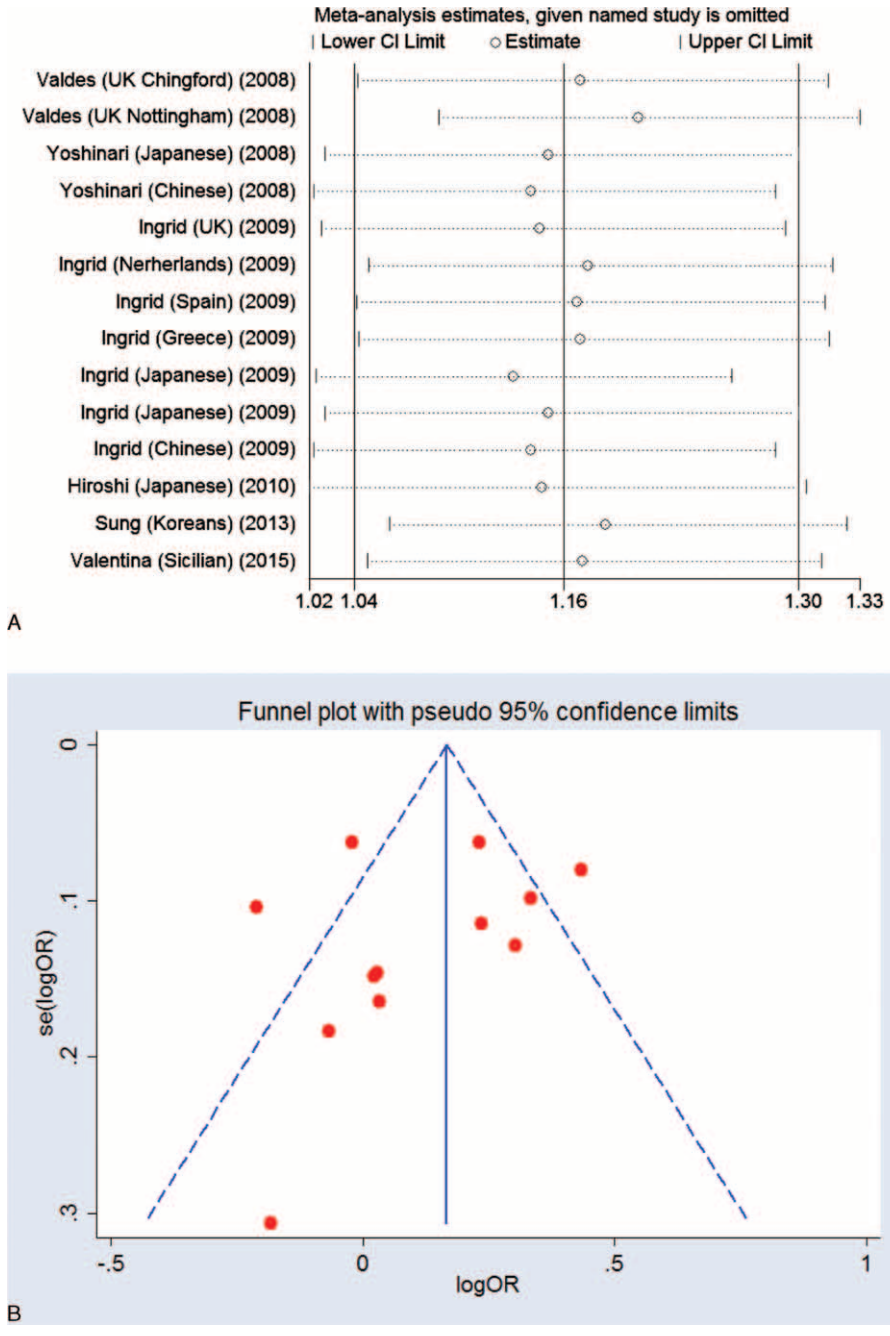

FIGURE 8. The result of sensitivity and Funnel plot analysis. A, Sensitivity analysis showed no individual study affected the pooled OR. B, Funnel plot analysis showed no potential publication bias.

models also needs to be treated with caution when interpreting the results. The insufficient sample size in single pioneer or replication studies of multiple SNPs, including DVWA, did result in a wide range of values of the ORs. However, we have provided a comprehensive overview of most of the relevant SNPs in OA or its subtypes.[33–64] We have also updated all available studies on DVWA SNPs, and our results further support an association of SNP rs7639618 with OA as recently indicated.[65,66] In addition, we have performed multiple analyses including subgroup, heterogeneity, sensitivity, and meta-regression assessment. The results showed that our meta-analysis is stable and we have analyzed the sources of the heterogeneity as indicated above. Together, our results support that rs7639618 SNP is significantly associated with increased risk of knee OA in Asians. There was insufficient data to support an association between SNP rs7639618 and the risk of hip OA or OA in Caucasians, although further studies are required to validate this genetic epidemiology and to functionally characterize this DVWA variant with OA pathophysiology.

ACKNOWLEDGMENTS

We thank the families for participating in this research project. This work is supported by clinical medicine science and technology projects of Jiangsu province (BL2013019), Jiangsu provincial health department scientific research project (Q201412), Suzhou science and technology support program (SS201429), the innovation program of Jiangsu province (QZ), and the NSFC grants, China (Nos. 31271399 and 81472047, QZ).

REFERENCES

1. Glyn-Jones S, Palmer AJ, Agricola R, et al. Osteoarthritis. *Lancet (London).* 2015;386:376–387.
2. Woolf AD, Pfleger B. Burden of major musculoskeletal conditions. *Bull World Health Organization.* 2003;81:646–656.
3. Yin YW, Sun QQ, Hu AM, et al. Association of rs9340799 polymorphism in estrogen receptor alpha gene with the risk of osteoarthritis: evidence based on 8,792 subjects. *Mol Genet Genomics.* 2015;290:513–520.
4. Brandt KD, Dieppe P, Radin E. Etiopathogenesis of osteoarthritis. *Med Clin North Am.* 2009;93:1–24.
5. Riley JH, Allan CJ, Lai E, et al. The use of single nucleotide polymorphisms in the isolation of common disease genes. *Pharmacogenomics.* 2000;1:39–47.
6. Puliti A, Caridi G, Ravazzolo R, et al. Teaching molecular genetics: chapter 4—positional cloning of genetic disorders. *Pediatr Nephrol.* 2007;22:2023–2029.
7. Abel K, Reneland R, Kammerer S, et al. Genome-wide SNP association: identification of susceptibility alleles for osteoarthritis. *Autoimmunity Rev.* 2006;5:258–263.
8. Srinivasan S, Clements JA, Batra J. Single nucleotide polymorphisms in clinics: fantasy or reality for cancer? *Crit Rev Clin Lab Sci.* 2016;53:29–39.
9. Huang Q. Genetic study of complex diseases in the post-GWAS era. *J Genet Genomics.* 2015;42:87–98.
10. Whittaker CA, Hynes RO. Distribution and evolution of von Willebrand/integrin A domains: widely dispersed domains with roles in cell adhesion and elsewhere. *Mol Biol Cell.* 2002;13:3369–3387.
11. Becker AK, Mikolajek H, Paulsson M, et al. A structure of a collagen VI VWA domain displays N and C termini at opposite sides of the protein. *Structure.* 2014;22:199–208.
12. Nakajima M, Miyamoto Y, Ikegawa S. Cloning and characterization of the osteoarthritis-associated gene DVWA. *J Bone Miner Metab.* 2011;29:300–308.
13. Meulenbelt I, Chapman K, Dieguez-Gonzalez R, et al. Large replication study and meta-analyses of DVWA as an osteoarthritis susceptibility locus in European and Asian populations. *Hum Mol Genet.* 2009;18:1518–1523.
14. Valdes AM, Spector TD, Doherty S, et al. Association of the DVWA and GDF5 polymorphisms with osteoarthritis in UK populations. *Ann Rheum Dis.* 2009;68:1916–1920.
15. Qiao L, Liang Y, Li N, et al. Endothelin-A receptor antagonists in prostate cancer treatment: a meta-analysis. *Int J Clin Exp Med.* 2015;8:3465–3473.
16. Wang F, Ma YL, Zhang P, et al. A genetic variant in microRNA-196a2 is associated with increased cancer risk: a meta-analysis. *Mol Biol Rep.* 2012;39:269–275.
17. Qiu MT, Hu JW, Ding XX, et al. Hsa-miR-499 rs3746444 polymorphism contributes to cancer risk: a meta-analysis of 12 studies. *PLoS One.* 2012;7:e50887.
18. Whitehead A, Whitehead J. A general parametric approach to the meta-analysis of randomized clinical trials. *Stat Med.* 1991;10:1665–1677.
19. Panoutsopoulou K, Southam L, Elliott KS, et al. Insights into the genetic architecture of osteoarthritis from stage 1 of the arcOGEN study. *Ann Rheum Dis.* 2011;70:864–867.
20. Raine EV, Dodd AW, Reynard LN, et al. Allelic expression analysis of the osteoarthritis susceptibility gene COL11A1 in human joint tissues. *BMC Musculoskelet Disord.* 2013;14:85.
21. Bravata V, Minafra L, Forte GI, et al. DVWA gene polymorphisms and osteoarthritis. *BMC Res Notes.* 2015;8:30.
22. Miyamoto Y, Shi D, Nakajima M, et al. Common variants in DVWA on chromosome 3p24.3 are associated with susceptibility to knee osteoarthritis. *Nat Genet.* 2008;40:994–998.
23. Takahashi H, Nakajima M, Ozaki K, et al. Prediction model for knee osteoarthritis based on genetic and clinical information. *Arthritis Res Ther.* 2010;12:R187.
24. Lee SJ, Kim MJ, Kee SJ, et al. Association study of the candidate gene for knee osteoarthritis in Koreans. *Rheumatol Int.* 2013;33: 783–786.
25. Zhu L, Shi D, Dai J, et al. Lack of evidence for association between DVWA gene polymorphisms and developmental dysplasia of the hip in Chinese Han population. *Rheumatol Int.* 2011;31:883–887.
26. Saito T, Fukai A, Mabuchi A, et al. Transcriptional regulation of endochondral ossification by HIF-2alpha during skeletal growth and osteoarthritis development. *Nat Med.* 2010;16:678–686.
27. Miller RE, Tran PB, Das R, et al. CCR2 chemokine receptor signaling mediates pain in experimental osteoarthritis. *Proc Natl Acad Sci U S A.* 2012;109:20602–20607.
28. Sawitzke AD. Personalized medicine for osteoarthritis: where are we now? *Ther Adv Musculoskelet Dis.* 2013;5:67–75.
29. Chapman K, Takahashi A, Meulenbelt I, et al. A meta-analysis of European and Asian cohorts reveals a global role of a functional SNP in the 5' UTR of GDF5 with osteoarthritis susceptibility. *Hum Mol Genet.* 2008;17:1497–1504.
30. Rodriguez RR, Seegmiller RE, Stark MR, et al. A type XI collagen mutation leads to increased degradation of type II collagen in articular cartilage. *Osteoarthritis Cartilage.* 2004;12:314–320.
31. He XF, Wei W, Liu ZZ, et al. Association between the CYP1A1 T3801C polymorphism and risk of cancer: evidence from 268 case-control studies. *Gene.* 2014;534:324–344.
32. Qiao L, Liang Y, Mira RR, et al. Mammalian target of rapamycin (mTOR) inhibitors and combined chemotherapy in breast cancer: a meta-analysis of randomized controlled trials. *Int J Clin Exp Med.* 2014;7:3333–3343.
33. Valdes AM, Lories RJ, van Meurs JB, et al. Variation at the ANP32A gene is associated with risk of hip osteoarthritis in women. *Arthritis Rheum.* 2009;60:2046–2054.
34. Zeggini E, Panoutsopoulou K, Southam L, et al. Identification of new susceptibility loci for osteoarthritis (arcOGEN): a genome-wide association study. *Lancet.* 2012;380:815–823.
35. Mototani H, Iida A, Nakamura Y, et al. Identification of sequence polymorphisms in CALM2 and analysis of association with hip osteoarthritis in a Japanese population. *J Bone Miner Metab.* 2010;28:547–553.
36. Evangelou E, Kerkhof HJ, Styrkarsdottir U, et al. A meta-analysis of genome-wide association studies identifies novel variants associated with osteoarthritis of the hip. *Ann Rheum Dis.* 2014;73:2130–2136.
37. Rodriguez-Fontenla C, Calaza M, Evangelou E, et al. Assessment of osteoarthritis candidate genes in a meta-analysis of nine genome-wide association studies. *Arthritis Rheumatol.* 2014;66:940–949.
38. Nakki A, Videman T, Kujala UM, et al. Candidate gene association study of magnetic resonance imaging-based hip osteoarthritis (OA): evidence for COL9A2 gene as a common predisposing factor for hip OA and lumbar disc degeneration. *J Rheumatol.* 2011;38:747–752.

39. Meulenbelt I, Min JL, Bos S, et al. Identification of DIO2 as a new susceptibility locus for symptomatic osteoarthritis. *Hum Mol Genet.* 2008;17:1867–1875.
40. Castano Betancourt MC, Cailotto F, Kerkhof HJ, et al. Genome-wide association and functional studies identify the DOT1L gene to be involved in cartilage thickness and hip osteoarthritis. *Proc Natl Acad Sci U S A.* 2012;109:8218–8223.
41. Limer KL, Tosh K, Bujac SR, et al. Attempt to replicate published genetic associations in a large, well-defined osteoarthritis case-control population (the GOAL study). *Osteoarthritis Cartilage.* 2009;17:782–789.
42. Evans DS, Cailotto F, Parimi N, et al. Genome-wide association and functional studies identify a role for IGFBP3 in hip osteoarthritis. *Ann Rheum Dis.* 2015;74:1861–1867.
43. Wang L, Guo L, Tian F, et al. Analysis of single nucleotide polymorphisms within ADAM12 and risk of knee osteoarthritis in a Chinese Han population. *BioMed Res Int.* 2015;2015:518643.
44. Rodriguez-Lopez J, Pombo-Suarez M, Loughlin J, et al. Association of a nsSNP in ADAMTS14 to some osteoarthritis phenotypes. *Osteoarthritis Cartilage.* 2009;17:321–327.
45. Liang W, Gao B, Xu G, et al. Association between single nucleotide polymorphisms of asporin (ASPN) and BMP5 with the risk of knee osteoarthritis in a Chinese Han population. *Cell Biochem Biophys.* 2014;70:1603–1608.
46. Ikeda T, Mabuchi A, Fukuda A, et al. Association analysis of single nucleotide polymorphisms in cartilage-specific collagen genes with knee and hip osteoarthritis in the Japanese population. *J Bone Miner Res.* 2002;17:1290–1296.
47. Evangelou E, Valdes AM, Kerkhof HJ, et al. Meta-analysis of genome-wide association studies confirms a susceptibility locus for knee osteoarthritis on chromosome 7q22. *Ann Rheum Dis.* 2011;70:349–355.
48. Ren Y, Tan B, Yan P, et al. Association between polymorphisms in the estrogen receptor alpha gene and osteoarthritis susceptibility: a meta-analysis. *BMC Musculoskelet Disord.* 2015;16:44.
49. Lee SW, Song JH, Choi WS, et al. The single nucleotide polymorphism (SNP) of the estrogen receptor-beta gene, rs1256049, is associated with knee osteoarthritis in Korean population. *Knee.* 2014;21:242–246.
50. Zhang R, Yao J, Xu P, et al. A comprehensive meta-analysis of association between genetic variants of GDF5 and osteoarthritis of the knee, hip and hand. *Inflamm Res.* 2015;64:405–414.
51. Kerkhof HJ, Lories RJ, Meulenbelt I, et al. A genome-wide association study identifies an osteoarthritis susceptibility locus on chromosome 7q22. *Arthritis Rheum.* 2010;62:499–510.
52. Fernandez-Torres J, Hernandez-Diaz C, Espinosa-Morales R, et al. Polymorphic variation of hypoxia inducible factor-1 A (HIF1A) gene might contribute to the development of knee osteoarthritis: a pilot study. *BMC Musculoskelet Disord.* 2015;16:218.
53. Luo SX, Li S, Zhang XH, et al. Genetic polymorphisms of interleukin-16 and risk of knee osteoarthritis. *PLoS One.* 2015;10: e0123442.
54. Han L, Lee HS, Yoon JH, et al. Association of IL-17A and IL-17F single nucleotide polymorphisms with susceptibility to osteoarthritis in a Korean population. *Gene.* 2014;533:119–122.
55. Fernandes MT, Fernandes KB, Marquez AS, et al. Association of interleukin-6 gene polymorphism (rs1800796) with severity and functional status of osteoarthritis in elderly individuals. *Cytokine.* 2015;75:316–320.
56. Nakki A, Rodriguez-Fontela C, Gonzalez A, et al. Association study of MMP8 gene in osteoarthritis. *Connect Tissue Res.* 2015. Nov 17: 1-9. [Epub ahead of print].
57. Valdes AM, Spector TD, Tamm A, et al. Genetic variation in the SMAD3 gene is associated with hip and knee osteoarthritis. *Arthritis Rheum.* 2010;62:2347–2352.
58. Etokebe GE, Jotanovic Z, Mihelic R, et al. Susceptibility to large-joint osteoarthritis (hip and knee) is associated with BAG6 rs3117582 SNP and the VNTR polymorphism in the second exon of the FAM46A gene on chromosome 6. *J Orthopaed Res.* 2015;33: 56–62.
59. Cui L, Li G, Zhong W, et al. Polycystic ovary syndrome susceptibility single nucleotide polymorphisms in women with a single PCOS clinical feature. *Hum Reprod.* 2015;30:732–736.
60. Schneider EM, Du W, Fiedler J, et al. The (-765 G–>C) promoter variant of the COX-2/PTGS2 gene is associated with a lower risk for end-stage hip and knee osteoarthritis. *Ann Rheum Dis.* 2011;70:1458–1460.
61. He Y, Liang X, Wu X, et al. Association between interleukin 8-251 A/T and +781 C/T polymorphisms and osteoarthritis risk. *Immunol Lett.* 2014;162:207–211.
62. Spector TD, Reneland RH, Mah S, et al. Association between a variation in LRCH1 and knee osteoarthritis: a genome-wide single-nucleotide polymorphism association study using DNA pooling. *Arthritis Rheum.* 2006;54:524–532.
63. Zhu ZH, Jin XZ, Zhang W, et al. Associations between vitamin D receptor gene polymorphisms and osteoarthritis: an updated meta-analysis. *Rheumatology (Oxford).* 2014;53:998–1008.
64. Cong Y, Ru JY, Bao NR, et al. A single nucleotide polymorphism in the TGF-beta1 gene (rs1982073 C>T) may contribute to increased risks of bone fracture, osteoporosis, and osteoarthritis: a meta-analysis. *Clin Rheumatol.* 2014.
65. Zhang R, Yao J, Xu P, et al. Association between genetic variants of DVWA and osteoarthritis of the knee and hip: a comprehensive meta-analysis. *Int J Clin Exp Med.* 2015;8:9430–9437.
66. Wang D, Zhou K, Chen Z, et al. The association between DVWA polymorphisms and osteoarthritis susceptibility: a genetic meta-analysis. *Int J Clin Exp Med.* 2015;8:12566–12574.