Association between polymorphisms in the estrogen receptor alpha gene and osteoarthritis susceptibility: a meta-analysis

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Abstract

Background: Osteoarthritis (OA) is a common chronic disease of the joints. Genetic factors may play a role in its development, and polymorphisms in the estrogen receptor alpha gene (ERα) have been associated with OA. However, previous studies into this relationship have reported inconsistent results, so we aimed to systematically review the association between ERα polymorphisms and OA susceptibility.

Methods: We conducted a comprehensive literature search of Ovid MEDLINE, EMBASE, CBM, and PubMed databases, and Google scholar, and identified 11 eligible studies that examined the association between ERα polymorphisms and OA susceptibility. We carried out a meta-analysis of these studies based on ERα XbaI (rs9340799) and Pvul (rs2234693) genotypes.

Results: Seventeen comparisons involving 10 European and seven Asian populations of 5,325 OA patients and 10,834 controls were included in the study. The ERα XbaI polymorphism were significantly associated with OA in Europeans (AA vs. AG + GG: OR = 1.17, 95% confidence interval (CI) = 1.02–1.34, P = 0.03; AG vs. AA + GG: OR = 0.86, 95% CI = 0.75–0.99, P = 0.04) but not in Asian populations. No association was found between OA and the ERα Pvul polymorphism in any population (C vs. T, OR = 0.98, 95% CI = 0.93–1.03, P = 0.37; CC vs. TT + CT, OR = 0.97, 95% CI = 0.89–1.06, P = 0.55; CT vs. CC + TT, OR = 0.99, 95% CI = 0.92–1.06, P = 0.75; TT vs. CC + CT, OR = 1.01, 95% CI = 0.92–1.12, P = 0.79).

Conclusions: This study suggested that there may be a weak relationship between the ERα XbaI polymorphism and OA in Europeans but not Asians, and that the ERα Pvul polymorphism was not associated with OA in either population. However, large well-designed studies are necessary to confirm these results in more homogeneous populations.

Keywords: Estrogen receptor, Osteoarthritis, Polymorphism, Meta-analysis

Background

Osteoarthritis (OA) is the most common joint disease worldwide, and primarily affects the knees, hips, hands, and spine. It is a leading cause of disability among older individuals and also affects their quality of life [1]. It is characterized by the progressive degeneration of articular cartilage, and by subchondral sclerosis resulting in pain and joint stiffness [2].

The etiology of OA is multifactorial, including genetic and environmental risk factors. Associated genes include GDF5 [3], ASPN [4], FRZB [5], and COL2A1 [6], while environmental factors may include obesity [7-9], history of knee injury [10], occupational activities [11,12], sex hormones and structural changes [13], meniscectomy [14], gender, and age [15]. Twin-pair and family genetic data show that more than 50% of OA can be attributed to genetic factors [16]. A gender difference is also apparent, with females having a greater prevalence of OA after the age of 50 years [17]. Additionally, the disease is more common among European populations [18]. The observation that the estrogen receptor (ER) is expressed in human articular chondrocytes and bone cells suggests that it may be involved in the etiology of OA [19].
The ER has two isoforms: ERα and ERβ. ERα expression affects the growth of bone cells, while ERβ participates in the formation and resorption of bone [20]. ERα is located on chromosome 6q25.1 and contains eight exons and seven introns [21], as well as two common restriction fragment length polymorphisms (RFLPs): XbaI and PvuII. The XbaI RFLP detects an A–G substitution at position 351 (−351int A/G; rs9340799), while PvuII detects a T–C substitution at position 397 (−397int T/C; rs2234693). A previous meta-analysis confirmed the association between bone mineral density and ERα [22].

A number of studies have investigated the association between ERα polymorphisms and the risk of OA in different populations, but the results are inconsistent. Some discovered that ERα polymorphisms were associated with an increased risk of OA [23-28], while others found no association with OA risk [28,29], or an association with a reduced risk of OA [30-34]. To our knowledge, no systematic review has examined the evidence for a relationship between ERα polymorphisms and OA. Therefore, we conducted a meta-analysis to analyze the association between ERα polymorphisms and OA susceptibility.

Methods
This systematic review was conducted according to 2009 PRISMA guidelines [35].

Search strategy
We performed a systematic research of available studies that assessed the association between ERα polymorphisms and OA. We carried out a comprehensive literature search for published studies in OVID MEDLINE, EMBASE, CBM, and PubMed databases, and Google Scholar. Primary key search terms included estrogen receptor, polymorphism, osteoarthritis, and OA. Index terms for OVID MEDLINE were: “estrogen receptor”, “polymorphism”, and “osteoarthritis” or “OA”. The last query was updated on 30 November 2014. There were no language or other limitations on the search. Reference lists in the retrieved articles or relevant reviews were also screened to identify other eligible studies. We also searched unpublished studies by contacting clinical experts and the Arthritis Foundation National Office. A flow diagram of our literature identification strategy is shown in Figure 1.

Figure 1 Flow diagram of study selection according to the PRISMA statement.
Inclusion and exclusion criteria
Eligible studies were required to satisfy the following criteria: (1) the study was a cohort or a case–control study; (2) OA was diagnosed based on clinical criteria defined by the American College of Rheumatology; (3) the original study assessed the association between ERα polymorphisms (XbaI or PvuII) and OA susceptibility; and (4) the study provided sufficient genetic frequency or sufficient data for extraction. If overlapping study populations were identified between studies, only the most complete one was included in the meta-analysis. Animal studies and literature reviews were excluded.

Quality assessment of included studies
Study quality was independently assessed by two authors, based on the Newcastle–Ottawa scale (NOS) quality score systems [36]. The NOS contains eight items divided into three categories: selection, comparability, and outcome (for cohort studies) or exposure (for case–control studies). Quality scores ranged from 0 to 9. When there was disagreement on the quality scores between the two authors, discrepancies were resolved through discussion and consultation with a third author.

The quality of included studies was also assessed by the Hardy–Weinberg equilibrium (HWE) for the control genotype distribution. Studies consistent with HWE were defined as high-quality, while those inconsistent with HWE were defined as low-quality studies.

Data extraction
The following data were extracted from each full-text study using a standardized data extraction form: the name of the first author, year of publication, country in which the study was performed, study design, number of cases and controls, gender, age, genotyping, OA site, OA definition, polymorphism, and numbers of cases and controls for each of the PvuII (rs2234693), and XbaI (rs9340799) genotypes. When the information extracted from studies was inconsistent, disagreement was resolved through discussion and consultation with a third author until consensus was achieved on every item.

Statistical analysis
STATA 12.0 and Review Manager 5.2 software were used for data analysis. The pooled odds ratio (OR) and its 95% confidence interval (95% CI) were calculated to assess the association between ERα polymorphisms and the risk of OA for the following contrasts: G vs. A, AG vs. AA + GG, GG vs. AG + AA, AA vs. AG + GG, C vs. T, CC vs. TT + CT, CT vs. CC + TT, and TT vs. CC + CT. Subgroup analysis based on ethnicity was also performed. The Chi-square test was used to determine if the identified study was consistent with HWE for the control genotype distribution. Heterogeneity between studies was evaluated with the $I^2$ test and the $Q$ statistic. We used the Cochrane system for heterogeneity grading: $I^2$ 0–40%, might not be important; 30–60%, moderate heterogeneity; 50–90%, substantial heterogeneity; 75–100%, considerable heterogeneity. Heterogeneity was assessed to be significant when $I^2 > 30\%$ or when $P < 0.1$ for $Q$ statistics.

The pooled effects were estimated using the DerSimonian and Laird method for random effects and the Mantel–Haenszel method for fixed effects [37]. If the studies were significantly heterogeneous, we used the random effects model. Otherwise, we used the fixed effects model to calculate the pooled OR and 95%CI. The random effects model assumes that different studies have substantial diversity and assesses both within-study sampling error and between-study variation [38]. The fixed effects model assumes that genetic factors have similar effects on OA susceptibility across all studies, and that observed variations between studies are caused by chance alone [39]. Sensitivity analyses were performed for the effect size omitting the trial for which data were imputed, and were used to evaluate the stability of the results. Publication bias was graphically represented by funnel plots and further evaluated with the Begg’s test and Egger’s test [40,41].

Results
Search results and studies included in the meta-analysis
Seventy-seven relevant studies were preliminarily identified in the database search, of which 11 [24–34] eventually satisfied the eligibility criteria for our meta-analysis. All included studies investigated the relationship between ERα polymorphisms and OA susceptibility. Of these, one study [33] contained data on three different OA sites and four different geographical locations, so these seven comparisons were treated independently. Therefore, a total of 17 separate comparisons were included in the present meta-analysis. Ten studies with a total of 8,502 participants (2,181 OA patients and 6,321 controls), which involved three European and seven Asian populations, evaluated the association between the ERα XbaI polymorphism and OA susceptibility, while 17 with 16,159 total participants (5,325 OA patients and 10,834 controls), involving 10 European and seven Asian populations, evaluated the association between the ERα PvuII polymorphism and OA susceptibility. Study characteristics are summarized in Table 1.

Allele and genotype counts
Allelic counts of the ERα XbaI polymorphism were evaluated for G and A alleles. In general, the frequency of the A allele was higher in OA cases than in controls. Genotype counts of the ERα XbaI polymorphism were evaluated for GG, AG, and AA genotypes, and the
| Study [Ref.]            | Year | Country (City)    | Study design | Genotyping | Numbers | Gender (M/F) | Age     | Polymorphism (s) | Quality score |
|------------------------|------|-------------------|--------------|------------|---------|--------------|---------|------------------|---------------|
| Toshio Ushiyama et al. | 1998 | Japan             | Case-control | PCR        | 65      | 318          | 0/65    | XbaI, PvuII      | 7 (2/2/3)     |
| John Loughlin et al.   | 2000 | UK (Oxford)       | Case-control | PCR        | 371     | 369          | 155/216 | XbaI, PvuII      | 8 (3/2/3)     |
| Barton L. Wise et al.  | 2009 | USA               | Cohort       | PCR        | 307     | 214          | 258/263 | XbaI, PvuII      | 8 (4/2/2)     |
| Barton L. Wise et al.  | 2009 | USA               | Cohort       | PCR        | 304     | 211          | 253/262 | XbaI, PvuII      | 9 (4/2/3)     |
| V. M. Borgonio-Cuadra et al. | 2012 | Mexico           | Case-control | PCR        | 115     | 117          | 23/92   | XbaI, PvuII      | 8 (3/2/3)     |
| J. A. Riancho et al.   | 2010 | Spain (Santander) | Case-control | PCR        | 272     | 802          | 95/177  | XbaI, PvuII      | 8 (3/2/3)     |
| J. A. Riancho et al.   | 2010 | Spain (Santander) | Case-control | PCR        | 254     | 473          | 47/207  | XbaI, PvuII      | 8 (3/2/3)     |
| J. A. Riancho et al.   | 2010 | UK (Oxford)       | Case-control | PCR        | 445     | 862          | 176/269 | XbaI, PvuII      | 8 (3/2/3)     |
| J. A. Riancho et al.   | 2010 | Spain (Coruña)    | Case-control | PCR        | 252     | 244          | 90/162  | XbaI, PvuII      | 8 (3/2/3)     |
| J. A. Riancho et al.   | 2010 | Spain (Santiago)  | Case-control | PCR        | 287     | 473          | 110/177 | XbaI, PvuII      | 8 (3/2/3)     |
| J. A. Riancho et al.   | 2010 | UK (Oxford)       | Case-control | PCR        | 1278    | 862          | 503/775 | XbaI, PvuII      | 8 (3/2/3)     |
| K. Lian M.D. et al.    | 2007 | USA               | Cohort       | PCR        | 569     | 4134         | 0/569   | XbaI, PvuII      | 8 (4/2/2)     |
| Sheng-Yu Jin et al.    | 2004 | Korea             | Case-control | PCR        | 151     | 397          | 53/98   | XbaI, PvuII      | 8 (3/2/3)     |
| Zhi Tian et al.        | 2009 | China             | Case-control | PCR        | 38      | 40           | 0/38    | XbaI, PvuII      | 7 (2/2/3)     |
| Jiexiang Yang et al.   | 2009 | China             | Case-control | PCR        | 41      | 40           | 31/50   | XbaI, PvuII      | 6 (2/1/3)     |
| Yan Xue et al.         | 2004 | China             | Case-control | PCR        | 55      | 176          | 0/55    | XbaI, PvuII      | 7 (3/1/3)     |
| Xiaoyu Dai et al.      | 2014 | China             | Case-control | PCR        | 469     | 522          | 113/356 | XbaI, PvuII      | 7 (3/1/3)     |
frequency of the AA genotype was higher in OA cases than in the control group in all but one study [24]. The frequency of the AG genotype was lower in OA cases than in the control group in all but the same study [24]. There was no obvious difference in the frequency of the GG genotype between OA cases and controls. Allele and genotype counts for the \( \text{Era XbaI} \) polymorphism in cases and controls are shown in Table 2.

Allelic counts of the \( \text{Era PvuII} \) polymorphism were evaluated for C and T alleles. In general the T allele frequency

| Group       | Study                     | Country (City) | OA site | P (C) OA Control | p (T) OA Control | pp (TT) OA Control | Pp (CT) OA Control | PP (CC) OA Control |
|-------------|---------------------------|----------------|---------|------------------|------------------|--------------------|--------------------|--------------------|
| European    | Barton L. Wise et al.     | USA            | Hand    | 261              | 192              | 347                | 230                | 101                | 65                 | 145               | 100               | 58                | 46                |
|             | John Loughlin et al.      | UK             | Hip,Knee| 213              | 217              | 291                | 271                | 89                 | 76                 | 113               | 119               | 50                | 49                |
|             | K. Lian M.D. et al.       | USA            | Hip     | 399              | 776              | 491                | 948                | 426                | 253                | 595               | 442               | 257               | 167               |
|             | Sheng-Yu Jin et al.       | Korea          | Knee    | 44               | 200              | 66                 | 162                | 21                | 40                 | 24                | 82                | 10                | 54                |
|             | Toshio Ushiyama et al.    | Japan          | Hand    | 57               | 260              | 73                 | 376                | 19                | 115               | 35                | 146               | 11                | 57                |
|             | V. M. Borgonio-Cuadra et al. | Mexico       | Knee    | 77               | 82                | 153                | 152                | 52                | 51                 | 49                | 50                | 14                | 16                |
|             | Jianxiang Yang et al.     | China          | Knee    | 29               | 34                | 47                 | 46                 | 16                | 15                 | 15                | 16                | 7                 | 9                 |
|             | Yan Xue et al.            | China          | Knee    | 37               | 33                | 45                 | 47                 | 14                | 12                 | 17                | 23                | 10                | 5                 |
|             | Xiaoyu Dai et al.         | China          | Knee    | 387              | 390              | 551                | 638                | 167               | 198               | 217               | 242               | 85                | 74                |
| Asian Total | Sheng-Yu Jin et al.       | Korea          | Knee    | 112              | 307              | 190                | 487                | 61                | 152               | 68                | 183               | 22                | 62                |
|             | Toshio Ushiyama et al.    | Japan          | Hand    | 112              | 307              | 190                | 487                | 61                | 152               | 68                | 183               | 22                | 62                |
|             | V. M. Borgonio-Cuadra et al. | Mexico       | Knee    | 77               | 82                | 153                | 152                | 52                | 51                 | 49                | 50                | 14                | 16                |
|             | Zhi Tian et al.           | China          | Knee    | 29               | 34                | 47                 | 46                 | 16                | 15                 | 15                | 16                | 7                 | 9                 |
|             | Jianxiang Yang et al.     | China          | Knee    | 37               | 33                | 45                 | 47                 | 14                | 12                 | 17                | 23                | 10                | 5                 |
|             | Yan Xue et al.            | China          | Knee    | 53               | 151              | 57                 | 201                | 17                | 57                 | 23                | 87                | 15                | 32                |
|             | Xiaoyu Dai et al.         | China          | Knee    | 387              | 390              | 551                | 638                | 167               | 198               | 217               | 242               | 85                | 74                |
| Asian Total | Vanderlinde et al.        | 752             | Knee    | 1257             | 1116             | 1947               | 346               | 600               | 424               | 747               | 164               | 379               |
| Total       | Source                   | 4600            | Knee    | 9642             | 6046             | 11984              | 1735              | 3329              | 2576              | 5326              | 1012              | 2158              |
was higher in OA cases than in the control group. Genotype counts of the \(ErbA\) \(Pvu\) II polymorphisms were evaluated for TT, CT, and CC genotypes, and the TT genotype frequency was generally higher in OA cases than in controls. The CC genotype frequency was generally lower in OA cases than controls, although there was no obvious difference in the frequency of the CT genotype between the two groups. Allele and genotype counts for the \(ErbA\) \(Pvu\) II polymorphism in cases and controls are shown in Table 3.

### Quality assessment of included studies

All 11 studies had a satisfactory NOS quality score as shown in Table 1. The distribution of genotypes in the controls was in accordance with HWE (\(P > 0.05\)) in all studies, so all were classed as high-quality.

### Meta-analysis findings

A summary of the meta-analysis findings are shown in Table 4. The \(ErbA\) \(Xba\) I polymorphism was shown not to be associated with OA risk in all populations (G vs. A: OR = 0.87, 95% CI = 0.73–1.04, \(P = 0.13\); AA vs. AG + GG: OR = 1.16, 95% CI = 0.94–1.44, \(P = 0.17\); AG vs. AA + GG: OR = 0.93, 95% CI = 0.84–1.04, \(P = 0.22\); GG vs. AG + AA: OR = 0.88, 95% CI = 0.67–1.17, \(P = 0.38\)). However, subgroup analysis by ethnicity showed that the AA and AG genotypes of the \(ErbA\) \(Xba\) I polymorphism were associated with OA risk among Europeans (AA vs. AG + GG: OR = 1.17, 95% CI = 1.02–1.34, \(P = 0.03\); AG vs. AA + GG: OR = 0.86, 95% CI = 0.75–0.99, \(P = 0.04\)), but not among Asian populations (Figure 2).

There was no significant association between the \(ErbA\) \(Pvu\) II polymorphism and susceptibility to OA in all populations (C vs. T, OR = 0.98, 95% CI = 0.93–1.03, \(P = 0.37\); CC vs. TT + CT, OR = 0.97, 95% CI = 0.89–1.06, \(P = 0.55\); CT vs. CC + TT, OR = 0.99, 95% CI = 0.92–1.06, \(P = 0.75\); TT vs. CC + CT, OR = 1.01, 95% CI = 0.92–1.12, \(P = 0.79\)). In the subgroup analysis based on ethnicity, no significant association was found for the \(ErbA\) \(Pvu\) II polymorphism in either European or Asian populations (Figure 3).

### Table 4 Meta-analysis of \(ErbA\) \(Xba\) I and \(Pvu\) II polymorphisms and OA susceptibility

| Polymorphism comparison | Population OA site | No. of studies | Test of association | Test of heterogeneity | Test of publication bias |
|-------------------------|--------------------|----------------|--------------------|-----------------------|-------------------------|
|                         |                    |                | OR 95% CI p-value  | Model Q test p-value  | I² Z test p-value Begg’s test Egger’s test |
|                         |                    |                |                    |                       |                         |
| Xba (G vs. A)           | Overall            | 10             | 0.87 0.73–1.04 0.13| Random 29.71 0.0005 70%| –0.80 0.42 –1.29 0.23  |
|                         | European           | 3              | 0.91 0.82–1.01 0.08| Fixed 1.67 0.43 0%   |                         |
|                         | Asian              | 7              | 0.80 0.57–1.13 0.21| Random 27.74 0.0001 78%|                         |
| AA vs. AG + GG          | Overall            | 10             | 1.16 0.94–1.44 0.17| Random 25.55 0.002 65%| 1.16 0.25 0.94 0.38  |
|                         | European           | 3              | 1.17 1.02–1.34 0.03| Fixed 1.91 0.39 0%   |                         |
|                         | Asian              | 7              | 1.22 0.84–1.79 0.30| Random 21.69 0.001 72%|                         |
| AG vs. AA + GG          | Overall            | 10             | 0.93 0.84–1.04 0.22| Fixed 10.61 0.30 15% | 0.09 0.03 0.47 0.65  |
|                         | European           | 3              | 0.86 0.75–0.99 0.04| Fixed 1.52 0.47 0%   |                         |
|                         | Asian              | 7              | 1.06 0.89–1.26 0.52| Fixed 5.87 0.44 0%   |                         |
| GG vs. AG + AA          | Overall            | 10             | 0.88 0.67–1.17 0.38| Random 14.23 0.11 37%| –0.80 0.42 1.89 0.10  |
|                         | European           | 3              | 0.97 0.79–1.20 0.81| Fixed 0.85 0.65 0%   |                         |
|                         | Asian              | 7              | 0.65 0.36–1.19 0.17| Random 12.58 0.05 52%|                         |
| Pvu (C vs. T)           | Overall            | 17             | 0.98 0.93–1.03 0.37| Fixed 19.58 0.24 18% | 0.66 0.51 0.87 0.40  |
|                         | European           | 10             | 0.97 0.90–1.04 0.14| Random 13.61 0.14 34%|                         |
|                         | Asian              | 7              | 1.07 0.95–1.21 0.25| Fixed 3.19 0.78 0%   |                         |
| CC vs. TT + CT          | Overall            | 17             | 0.97 0.89–1.06 0.55| Fixed 13.09 0.67 0%  | 0.08 0.03 0.26 0.80  |
|                         | European           | 10             | 0.94 0.85–1.04 0.21| Fixed 5.05 0.83 0%   |                         |
|                         | Asian              | 7              | 1.17 0.94–1.47 0.16| Fixed 4.94 0.55 0%   |                         |
| CT vs. CC + TT          | Overall            | 17             | 0.99 0.92–1.06 0.75| Fixed 19.85 0.23 19% | 0.41 0.68 0.73 0.48  |
|                         | European           | 10             | 1.01 0.91–1.13 0.82| Random 15.43 0.08 42%|                         |
|                         | Asian              | 7              | 0.96 0.81–1.14 0.64| Fixed 4.29 0.64 0%   |                         |
| TT vs. CC + CT          | Overall            | 17             | 1.01 0.92–1.12 0.79| Random 23.83 0.09 33%| –0.25 0.81 –1.15 0.27  |
|                         | European           | 10             | 1.02 0.90–1.17 0.74| Random 20.33 0.02 50%|                         |
|                         | Asian              | 7              | 0.95 0.80–1.13 0.57| Fixed 2.35 0.88 0%   |                         |
Sensitivity analysis and publication bias

As shown in Table 4, heterogeneity was observed among studies in all populations and also in subgroup analyses. To explore the sources of heterogeneity across studies we performed a sensitivity analysis, which revealed that none of the studies significantly affected the pooled ORs and CIs. Sequential removal of each study had little effect on the pooled ORs.

The funnel plot revealed no obvious publication bias (Figure 4), and this was confirmed by Begg’s test and Egger’s test.

Discussion

Although the pathogenesis of OA is considered to be the result of many factors, genetics are thought to be one of the most important determinants [42]. Despite the fact that \( E{R_a} \) is one of the most studied genes in OA [43], to the best of our knowledge this is the first meta-analysis of the relationship between \( E{R_a} \) polymorphisms \( Xba{I} \) and \( Pu{v_II} \) and OA risk.

Our meta-analysis included 11 published studies (with 17 comparisons) of 16,159 participants (5,325 OA patients and 10,834 controls). Ten studies with a total of 8,502 participants evaluated the association between the \( E{R_a} Xba{I} \) polymorphism and OA susceptibility, and our meta-analysis suggested that it was significantly associated with OA in European but not Asian populations. The pooled OR for homozygote AA carriers showed that they were associated with a 17% increased risk for OA compared with AG and GG carriers, and that European AG carriers had a decreased OA risk. The heterogeneity of genetic effects between European and Asian populations suggests the existence of gene–environment or gene–gene interactions. No heterogeneity was detected in European populations with respect to the \( E{R_a} Xba{I} \) polymorphism and OA, suggesting that the genetic effect of this polymorphism is stronger in European than Asian populations.

Seventeen studies with a total of 16,159 participants evaluated the association between the \( E{R_a} Pu{v_II} \) polymorphism and OA susceptibility. Our meta-analysis suggested that there was no association between the polymorphism and susceptibility to OA in any population. The same result was obtained for the subgroup analysis based on ethnicity.

Gender differences are known to affect the development of OA; for example, the prevalence of knee OA is greater in women than men [15]. Only two of the studies included in our meta-analysis were stratified according to participant gender [25,32], and both reported no significant
| Study or Subgroup | CC Events | Total Events | TT + CT Events | Total Weight | Odds Ratio M-H, Fixed, 95% CI | Odds Ratio M-H, Fixed, 95% CI |
|------------------|-----------|--------------|----------------|-------------|------------------------------|------------------------------|
| 17.2.1 European  |           |              |                |             |                              |                              |
| Barton L.Wise 2009 | 58        | 104          | 246            | 411         | 4.6%                         | 0.85 [0.55, 1.31]             |
| J.A. Riancho-1 hip 2010 | 50       | 99           | 202            | 397         | 4.1%                         | 0.99 [0.63, 1.53]             |
| J.A. Riancho-2 hip 2010 | 257      | 424          | 1021           | 1716        | 16.5%                        | 1.05 [0.84, 1.30]             |
| J.A. Riancho-2 knee 2010 | 77       | 244          | 368            | 1063        | 9.8%                         | 0.87 [0.65, 1.17]             |
| J.A. Riancho-3 hip 2010 | 80       | 259          | 279            | 902         | 8.9%                         | 1.00 [0.74, 1.35]             |
| J.A. Riancho-3 knee 2010 | 53       | 232          | 219            | 842         | 7.6%                         | 0.84 [0.60, 1.19]             |
| J.A. Riancho-4 knee 2010 | 46       | 126          | 208            | 601         | 4.7%                         | 1.09 [0.73, 1.62]             |
| J.A. Riancho-4hip 2010 | 51       | 131          | 236            | 629         | 5.2%                         | 1.06 [0.72, 1.56]             |
| John Loughlin 2000 | 74       | 146          | 297            | 594         | 6.0%                         | 1.03 [0.72, 1.48]             |
| K. Lian M.D 2007 | 102      | 986          | 465            | 3694        | 18.2%                        | 0.90 [0.64, 1.21]             |
| Subtotal (95% CI) | 2751     | 10849        | 85.6%          | 0.94 [0.85, 1.04] |
| Total events     | 848      |              | 3541           |             |                              |                              |

Heterogeneity: Chi² = 5.05, df = 9 (P = 0.83); I² = 0%
Test for overall effect: Z = 1.25 (P = 0.21)

17.2.2 Asian

| Study or Subgroup | CC Events | Total Events | TT + CT Events | Total Weight | Odds Ratio M-H, Fixed, 95% CI | Odds Ratio M-H, Fixed, 95% CI |
|------------------|-----------|--------------|----------------|-------------|-------------------------------|-------------------------------|
| Jiexiang Yang 2009 | 10        | 15           | 31             | 66          | 0.4%                          | 2.26 [0.70, 7.33]             |
| Sheng-Yu Jin 2004 | 22        | 84           | 129            | 464         | 3.0%                          | 0.92 [0.54, 1.56]             |
| Toshio Ushiyama 1998 | 11        | 68           | 54             | 315         | 1.7%                          | 0.93 [0.46, 1.90]             |
| V.M. Bortonio-Cuadra 2012 | 14      | 30           | 101            | 202         | 1.4%                          | 0.88 [0.41, 1.93]             |
| Xiaoyu Dai 2014 | 85        | 159          | 384            | 824         | 6.0%                          | 1.32 [0.94, 1.86]             |
| Yan Xue 2004 | 15        | 47           | 40             | 184         | 1.1%                          | 1.69 [0.83, 3.42]             |
| Zhi Tian 2009 | 7         | 16           | 31             | 62          | 0.7%                          | 0.78 [0.26, 2.35]             |
| Subtotal (95% CI) | 419       |              | 2117           | 14.4%       | 1.17                          | [0.94, 1.47]                  |
| Total events     | 164       |              | 770            |             |                               |                               |

Heterogeneity: Chi² = 4.94, df = 6 (P = 0.55); I² = 0%
Test for overall effect: Z = 1.39 (P = 0.16)

Total (95% CI) | 3170 | 12966 | 100.0% | 0.97 [0.89, 1.06] |

Total events | 1012 | 4311 |

Heterogeneity: Chi² = 13.09, df = 16 (P = 0.67); I² = 0%
Test for overall effect: Z = 0.59 (P = 0.55)
Test for subgroup differences: Chi² = 3.14, df = 1 (P = 0.08); I² = 68.1%

Favours [CC] Favours [TT + CT]

0.01 0.1 1 10 100

Figure 3 Meta-analysis of the association between the ERα PvuII polymorphism and OA (CC vs. TT + CT).

Figure 4 Funnel plot of the meta-analysis of the ERα PvuII polymorphism with susceptibility to OA (C vs. T).
differences in the ERα polymorphisms between OA patients and controls of the same sex. However, because of the small number of this type of study and the limited raw data based on gender differences in genotype distributions and allele frequencies, we were unable to perform a subgroup analysis according to gender.

Several limitations should be taken into consideration in the current meta-analysis. First, it was based on unadjusted OR estimates because not all studies presented adjusted ORs, or the ORs were not adjusted by the same potential confounders, such as age and gender. This lack of information could have caused serious confounding bias. Second, OA is influenced by both genetic and environmental risk factors such as obesity, injury, occupational activities, and meniscectomy. However, the studies included in the meta-analysis did not control for these environmental risk factors. Third, some studies included individuals with OA in different sites, but we were unable to perform subgroup analysis of this within the same ethnic population because of the limited available data. For instance, hand OA is known to be more influenced by genetic and hormonal influences than other types of OA, but the relationship between the ERα XbaI polymorphism and hand OA was only reviewed in one study of Europeans [30] and one of Asians [24]. Other studies of the ERα XbaI polymorphism and OA susceptibility in Europeans focused on three different OA sites. Finally, although our current findings suggest that the ERα XbaI polymorphism is associated with OA in Europeans, it was not possible to determine whether this polymorphism is in linkage disequilibrium with any other potentially functional polymorphisms. However, our meta-analysis also had some advantages, including a satisfactory quality of all included studies, and a well-designed method.

Conclusions
The present results suggest that there may be a weak relationship between the ERα XbaI polymorphism and OA in European but not Asian populations, while the ERα PvuII polymorphism did not appear to be associated with OA in either Europeans or Asians. Because the studies included in the meta-analysis reviewed the relationship between the ERα XbaI polymorphism and OA susceptibility at three different sites in Europeans, large well-designed studies are necessary to confirm our findings in more homogeneous populations.

Competing interests
The authors declare that they have no competing interests.

Authors’ contributions
YR, BT, Y Wu, and Y Wang participated in the conception and design of the study. YR and BT carried out the literature search. YR, BT, and YY carried out the data collection. YR, BT, and PY performed the statistical analysis. Y Wu and Y Wang assessed the quality of the studies. YR and BT wrote the manuscript. Y Wu and Y Yang revised the manuscript. All authors read and approved the final manuscript.

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