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Original Article

Polymorphism of the low-density lipoprotein receptor-related protein 5 gene and fracture risk

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Abstract: Several molecular epidemiological studies have been conducted to examine the association between low-density lipoprotein receptor-related proteins (LRP5) Ala1330Val polymorphism and fracture; however, the conclusions remained controversial. We therefore performed an extensive meta-analysis on 10 published studies with 184479 subjects. Electronic databases, including PubMed, Excerpta Medica Database (EMBASE), Cochrane, Elsevier Science Direct and China National Knowledge Infrastructure (CNKI) databases were searched. Summary odds ratios (ORs) and corresponding 95% confidence intervals (CIs) were estimated using random-effects models. LRP5 Ala1330Val polymorphism was associated with a significantly increased risk of fracture (OR = 1.10; 95% CI, 1.06-1.14; I^2 = 29%). We also found that this polymorphism increased fracture risk in Caucasians. In the subgroup analysis according to gender, women was significantly associated with risk of fracture. In the subgroup analysis by type of fracture, LRP5 Ala1330Val polymorphism showed increased osteoporotic fracture risk. In conclusion, this meta-analysis suggested that an increased risk of fracture was associated with the LRP5 Ala1330Val polymorphism.

Keywords: Fracture, low-density lipoprotein receptor-related protein, meta-analysis, polymorphism

Introduction

Osteoporotic fractures are a major and increasing cause of morbidity, and they have caused a serious burden to health services in the world [1]. With the increasing ageing population and the high prevalence of osteoporosis, hip fracture is causing more serious damage to the public health [2]. Recently, numerous studies have attempted to explore the pathogenesis of this disease [3]. Bone mineral density (BMD) has been found to be an important clinical predictor of fracture risk. Most variance in BMD could be due to genetic factors, with as much as 65-92% of the difference in BMD attributable to genetic influences [4].

The role of Wnt signaling in bone formation gained significant recognition in 2001 when Gong and colleagues reported loss-of-function mutations in the low-density lipoprotein receptor-related proteins (LRP5) co-receptor cause the autosomal recessive disorder osteoporosis-pseudoglioma syndrome (OPPG), which is characterized by low bone mass, ocular defects, and a predisposition to fractures [5]. These findings were recapitulated in germline LRP5 knockout mice, which developed a low bone mass phenotype similar to patients with OPPG due to decreased osteoblast proliferation [6]. Thus, LRP5 might play an important role in the development of fracture. Many studies investigated the association of LRP5 polymorphism with fracture [7-16]. However, the result was still inconsistent. The aim of this study was to investigate whether there is an association between the Ala1330Val (rs3736228) in exon 18 and fracture risk.

Methods

Publication search

Electronic databases, including PubMed, Excerpta Medica Database (EMBASE), Cochrane, Elsevier Science Direct and China National Knowledge Infrastructure (CNKI) databases, were searched for identification of the studies on LRP5 Ala1330Val polymorphism and fracture published up to October 10, 2014. Search

Table 1. Characteristics of the studies included in this meta-analysis

First author/Year	Ethnicity	Gender	Age	No. of patients	Adjusted
Bollerslev/2005	Caucasian	Female	75.2	1301	BMD
Meurs 1/2006	Caucasian	Male	68	2592	Age, height, weight, BMD, femoral neck width
Meurs 2/2006	Caucasian	Female	70	3781	Age, height, weight, BMD, femoral neck width
Grundberg/2008	Caucasian	Male	75.4	3014	Age, body weight, height, study location
Joyce 1/2008	Caucasian	Male	> 18	11035	Age, weight, height, menopausal status, use of hormone therapy
Joyce 2/2008	Caucasian	Female	> 18	20164	Age, weight, height, menopausal status, use of hormone therapy
Richards 1/2008	Caucasian	Mixed	68.5	5921	BMD
Richards 2/2008	Caucasian	Mixed	62.1	718	BMD
Furuya/2009	Asian	Female	57	563	Age, BMI, J-HAQ score, daily prednisolone dose
Saarinen/2010	Caucasian	Mixed	10	301	NA
Korvala/2010	Caucasian	Male	20	192	NA
Riancho/2011	Caucasian	Female	80	1437	Age
Estrada 1/2012	Caucasian	Mixed	NA	27320	NA
Estrada 2/2012	Caucasian	Mixed	NA	54244	NA
Estrada 3/2012	Caucasian	Mixed	NA	51896	NA

BMD, bone mineral density; BMI, body mass index; J-HAQ, Japanese Health Assessment Questionnaire; NA, not available.

Table 2. Results of this meta-analysis

	Test of association			Heterogeneity		
Characteristics	OR (95% CI)	*Z*	*P* Value	χ^2	*P* Value	I^2 (%)
All studies	1.10 (1.06-1.14)	4.84	< 0.00001	19.81	0.14	29
Caucasian	1.10 (1.06-1.14)	4.71	< 0.00001	19.04	0.11	33
Female	1.07 (1.01-1.14)	2.06	0.04	4.13	0.39	3
Male	1.08 (0.98-1.19)	1.48	0.14	3.30	0.35	9
Osteoporotic	1.24 (1.03-1.49)	2.23	0.03	12.12	0.02	64
Vertebral	1.06 (0.99-1.12)	1.79	0.07	2.26	0.69	0

OR, odds ratio; CI, confidence intervals.

terms included 'fracture' and 'LRP5 OR low-density lipoprotein receptor-related protein'. There was no language restriction. Review articles and original papers were searched by hand for additional eligible studies.

Inclusion criteria

The following criteria were used to select the eligible studies: (a) evaluation of the association between LRP5 Ala1330Val polymorphism and fracture risk; (b) an unrelated case-control study in which family members were excluded; (c) sufficient published data for estimating an odds ratio (OR) with 95% confidence interval (CI). When authors reported two or more publications on the same patient population, only the largest study was selected. Additionally, when a study reported the results on different subpopulations, we treated them as a separate study.

Data extraction

Information was carefully extracted from all eligible publications independently by two investigators. Disagreement was resolved by discussion between the two investigators. The following data were collected from each study: the first author's name, publication year, ethnicity, age, gender, sample size (numbers of cases and controls), and adjustment. Authors were contacted for further information when necessary.

Statistical analysis

The strength of association between the LRP5 Ala1330Val polymorphism and fracture risk was assessed by calculating OR with 95% CI. The pooled ORs were performed in recessive model. A statistical test for heterogeneity was performed based on the Q statistic. The $P > 0.10$ of the Q-test indicated a lack of heterogeneity among studies. The summary OR estimate of each study was calculated by the random-effects model (the DerSimonian and Laird method). Stratified analysis was performed by race, gender and type of fracture. Cumulative meta-analysis and sensitivity analysis were also conducted. Potential publication bias was examined by Egger's test [17]. All statistical tests were performed with the software STATA

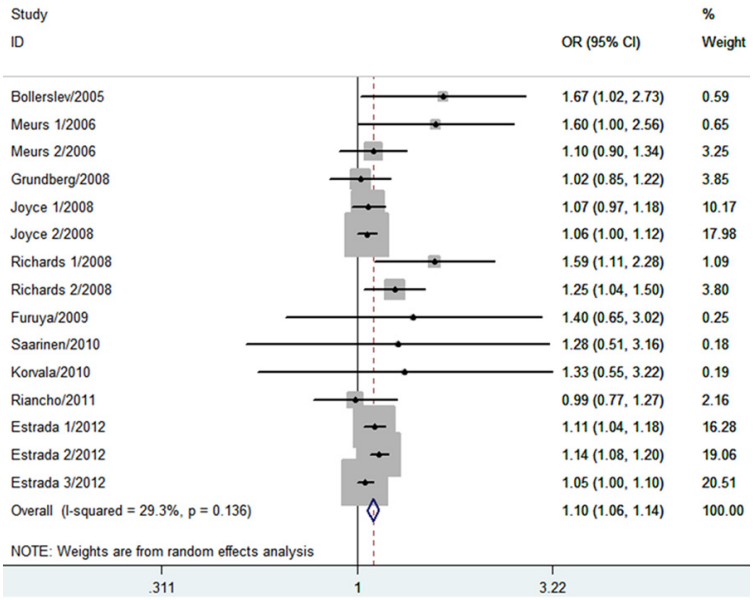

Figure 1. Meta-analysis for the association between LRP5 Ala1330Val polymorphism and fracture risk.

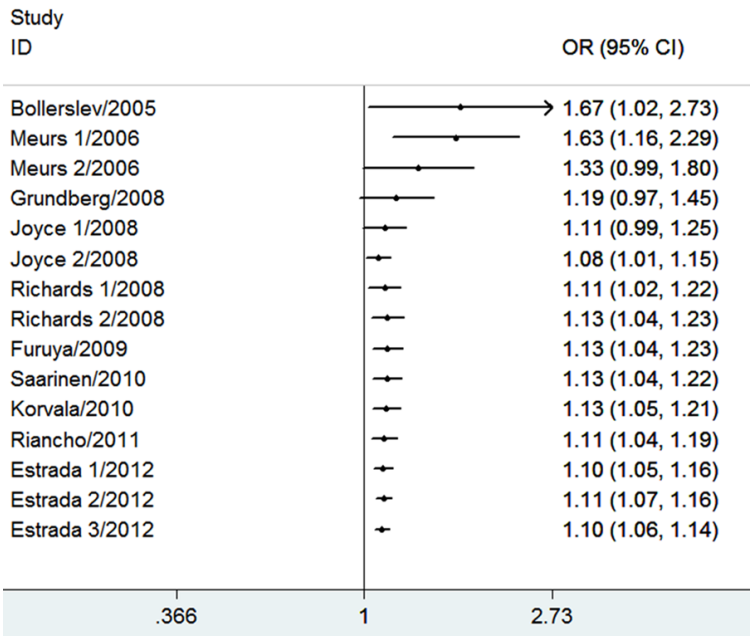

Figure 2. Cumulative meta-analysis for the association between LRP5 Ala1330Val polymorphism and fracture risk.

version 11.0 (Stata Corporation, College station, TX, USA). A *P* value < 0.05 was considered statistically significant.

Results

Study characteristics

Ten articles on LRP5 Ala1330Val polymorphism and fracture risk met the study inclusion criteria, and were included in the meta-analysis [7-16]. Three studies reported two case-control studies and one study reported three case-control study. Therefore, a total of 15 case-control studies with 184479 subjects were included in this meta-analysis. Only one study was performed in Asians, while other studies were conducted in Caucasians. Characteristics of studies investigating the association of LRP5 Ala1330Val polymorphism and fracture risk are presented in **Table 1**.

Results of meta-analysis

The evaluations of the association between LRP5 Ala1330Val polymorphism and fracture risk are listed in **Table 2**. LRP5 Ala1330Val polymorphism was associated with a significantly increased risk of fracture (OR = 1.10; 95% CI, 1.06-1.14; I^2 = 29%; **Figure 1**). Ten studies reported adjusted ORs. The combination of adjusted ORs for fracture was 1.12 (95% CI, 1.04-1.21). We also found that this polymorphism increased fracture risk in Caucasians (OR = 1.10; 95% CI, 1.06-1.14; I^2 = 33%). In the subgroup analysis according to gender, women was significantly associated with risk of fracture (RR = 1.07; 95% CI, 1.01-1.14; I^2 = 3%). However, no significant association was found in men (RR = 1.08; 95% CI, 0.98-1.19; I^2 = 9%). In the subgroup analysis by type of fracture, LRP5 Ala1330Val polymorphism showed increased osteoporotic fracture risk (RR = 1.24; 95% CI, 1.03-1.49; I^2 = 64%). However, this polymorphism did not influence vertebral fracture risk (RR = 1.06; 95% CI, 0.99-1.12; I^2 = 0%). As shown in **Figure 2**, significant associations were evident with each addition of more data over time. The results showed that the pooled ORs tended to be stable. A single study involved in the meta-analysis was deleted each time to reflect the influence

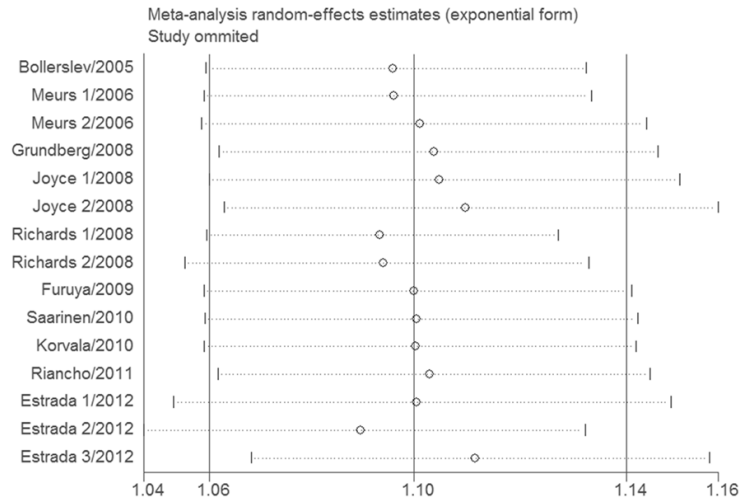

Figure 3. Sensitivity meta-analysis for the association between LRP5 Ala1330Val polymorphism and fracture risk.

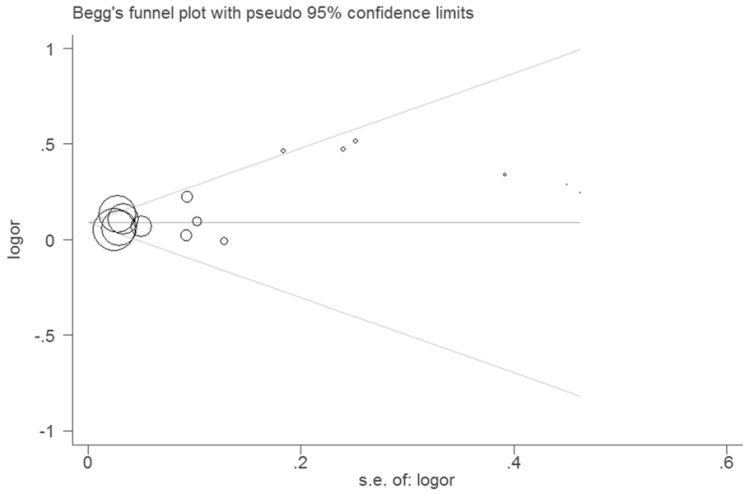

Figure 4. Funnel plot for the association between LRP5 Ala1330Val polymorphism and fracture risk.

of the individual data set to the pooled ORs, and the corresponding pooled ORs were not materially altered (**Figure 3**).

The shape of the funnel plots did not reveal any evidence of obvious asymmetry (**Figure 4**). The Egger test also did not displayed any evidence of publication bias ($P = 0.275$).

Discussion

The present meta-analysis, including 184479 subjects from 15 case-control studies, explored the association of LRP5 Ala1330Val polymorphism and fracture risk. We demonstrated that the LRP5 Ala1330Val polymorphism was associated with a significant increased fracture risk. Furthermore, in the stratified analysis by race, we found that Caucasians with LRP5 Ala1330Val polymorphism had increased fracture risk. However, it should be noted that only one study was performed in Asians. Thus, more studies with Asians are still needed to assess the association between LRP5 Ala1330Val polymorphism and fracture risk. We also found that female patients with LRP5 Ala1330Val polymorphism had increased fracture risk. However, we failed to find a significant relationship between LRP5 Ala1330Val polymorphism and fracture risk in men. In addition, in the stratified analysis by type of fracture, we found that patients with LRP5 Ala1330Val polymorphism showed increased osteoporotic fracture risk but not vertebral fracture risk. This result suggested that LRP5 Ala1330Val polymorphism may play an important role in the development of osteoporotic fracture.

LRP5 is a member of the low-density lipoprotein (LDL) receptor family [18]. Koay et al. [19] reported that the LRP5 gene and the Wnt signaling pathway are key players in bone formation and the risk of osteoporosis, and that LRP5 signaling is essential for normal morphology, developmental processes and bone health. Previous studies have reported positive associations between the LRP5 Ala1330Val polymorphism and low BMD [20, 21]. The Ala1330Val polymorphism lies within a second low density lipoprotein (LDL) receptor domain of LRP5. The function of this region in LRP5 is unknown, but similar domains in the LDL receptor domain interact with the propeller domain [22].Therefore, variations in the LDL receptor domains, such as Ala1330Val, may still alter protein function. Indeed, a recent report showed in vitro that Wnt-signaling capacity of the LRP5 Val1330 variant was decreased compared to the Ala1330 variant [23].

Our study had some advantages. First, the methodological issues for meta-analysis, such as, one-way sensitivity analysis and cumulative meta-analysis were well investigated. Second, the main result remained statistically significant when the adjusted ORs were combined. Results from one-way sensitivity analysis and cumulative meta-analysis suggested high stability and reliability of our results. Besides, significant heterogeneity was not observed in this meta-analysis. Moreover, funnel plots and Egger's tests were used to find potential publication bias. The results indicated that there was no significant publication bias.

Some limitations should be taken into account. First, there was no study investigated the association of LRP5 Ala1330Val polymorphism and fracture risk in Africans. Therefore, more studies are needed to further identify the association among Africans. Second, lack of the original data of the reviewed studies limited our further evaluation of potential interactions, because the interactions between gene-to-gene and gene-to-environment may modulate lung cancer risk. These gene-to-gene and gene-to-environment interactions should be further evaluated. Third, due to the lack of sufficient and uniform information in original studies, data were not stratified by other factors.

In conclusion, this meta-analysis suggested that an increased risk of fracture was associated with the LRP5 Ala1330Val polymorphism.

Disclosure of conflict of interest

None.

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