Lipid Transporter Activity-Related Genetic Polymorphisms Are Associated With Steroid-Induced Osteonecrosis of the Femoral Head: An Updated Meta-Analysis Based on the GRADE Guidelines

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Aims: The purpose of this study was to assess the relationship between genetic variants and steroid-induced osteonecrosis of the femoral head (SONFH) in steroid use populations.

Methods: We searched the public databases up to April 15, 2018. This study analyzed only the single-nucleotide polymorphisms (SNPs) that have appeared in more than three studies and assessed the level of evidence by classifying the outcomes according to the Grading of Recommendations Assessment, Development, and Evaluation (GRADE) approach.

Results: The ABCB1 rs1045642 C>T mutation had a protective effect against SONFH in the allelic model ($I^2 = 50.2\%$; OR: 0.74; 95% CI: 0.55–1.00; $p = 0.046$). The rs2032582 mutation in the ABCB1 gene showed no relationship to SONFH (allelic model: $I^2 = 50.3\%$; OR: 0.85; 95% CI: 0.58–1.23; $p = 0.382$). In ApoB rs693, four models showed that mutations can increase SONFH risk, but the allelic model did not. The ApoB rs1042031 mutation increased SONFH risk in the dominant model ($I^2 = 50.3\%$; OR: 2.90; 95% CI: 1.49–5.66; $p = 0.002$).

Conclusion: An allelic model of ABCB1 rs1045642 showed that mutations have a protective effect against SONFH at a very low level of evidence. The mutations in ApoB rs693 and rs1042031 increase the SONFH risk with moderate levels of evidence.

Keywords: osteonecrosis, steroids, glucocorticoid, genetic polymorphism, meta-analysis

INTRODUCTION

Osteonecrosis of the femoral head (ONFH) is caused by the obstruction or interruption of local blood supply, resulting in tissue ischemia, necrosis, and finally bone collapse (Kim et al., 2011). The core decompression method only relieved the pressure of the pulp cavity and delayed the progression of necrosis (Yu et al., 2018). At present, long-term and/or large-dose use of steroids have become the most important risk factors for non-traumatic ONFH (Shigemura et al., 2011).
The pathogenesis of steroid-induced ONFH (SONFH) is not yet clear and may be related to an imbalance in lipid metabolism and abnormal microcirculation (Johnson et al., 2004). The abnormal blood supply leads to apoptosis of osteocytes and osteoblasts, which causes bone loss and reduces bone mineral density (Luo et al., 2018). In addition, disordered lipid metabolism is another crucial pathogenesis that leads to increases in circulating lipid levels, microvascular fat embolisms, and lipid accumulation in the pulp cavity and ultimately causes osteocyte death.

Genetic polymorphisms have been found to be related to SONFH due to individual differences. Studies exploring the association between single-nucleotide polymorphisms (SNPs) and SONFH will help to identify the high SONFH risk population who use steroids. For these patients, it is necessary to avoid steroid application or to change the application strategy, increase the frequency of detection of SONFH, and enable early intervention. Furthermore, genetic variant-related studies can help to understand the pathogenesis of SONFH. Presently, several meta-analyses have been published that indicate that PAI-I rs1799768 (Gong et al., 2013), ABCB1 rs1045642 (Gong et al., 2013; Li et al., 2014; Zhou et al., 2015), and CYP3A variants (Guo and Deng, 2017) are related to the occurrence of SONFH. However, there is still a dispute over ABCB1 rs2032582 (Gong et al., 2013; Li et al., 2014; Zhou et al., 2015). This study will explicitly exclude studies in which the control group includes a healthy population or other types of ONFH; only comparison studies between ONFH and non-ONFH in the steroid use population are included. Because all studies are based on case-control and cohort designs with low evidence levels, we also used the Grading of Recommendations Assessment, Development, and Evaluation (GRADE) guidelines to assess the outcomes (Guyatt et al., 2008). Overall, the relationship between genetic variants and steroid-induced osteonecrosis of the femoral head in steroid use populations will be assessed in this study.

METHODS

This meta-analysis was performed in accordance with the Preferred Reporting Items for Systematic Reviews (PRISMA) guidelines (Moher et al., 2009).

Data Source and Search Strategy

Two authors independently performed a literature search using PubMed, Embase, the Cochrane Library, and the Chinese public databases, including the China National Knowledge Infrastructure (CNKI), the China Biology Medicine (CBM) Database, the China Science Periodical Database (CSPD), Wanfang Database, and the VIP Journal Integration Platform (VJIP). Searches were performed for studies published up to April 15, 2018. The following terms were used in the search strategy: hormone, glucocorticoid, steroid, corticosteroid, osteonecrosis, necrosis, femoral, femur, femoris, whirlbone, polymorphism, SNP, genetic, mutation, genotype, allele, allelic, and variation. We also conducted manual searches of the reference lists of relevant reviews to avoid omissions.

Selection Criteria

The following studies were included in our meta-analysis if they fulfilled the inclusion criteria: (1) study has a case-control or cohort study design; (2), steroid-using patients are included; (3), study compares ONFH and non-ONFH patients after steroid use; (4), study assesses the association between SNPs and SONFH; and (5), study indicates the frequencies of specific alleles or the effect sizes of individual genotypes between cases and controls.

The exclusion criteria included the following: (1), study compares patients with SONFH to healthy populations or populations with other types of ONFH; (2), study is about family heredity; (3), study is not SNP-related; and (4), study does not report data pertaining to allelic frequencies or calculable effect size. In addition, conference reports, editor comments, reviews, and academic dissertations were also excluded from the analysis.

Data Extraction and Quality Assessment

Two authors independently extracted the following information from each eligible study: the first author’s name, publication year, research location, sample size, average age, primary disease, diagnostic mode, and genes of interest. The methodological quality of the included studies was evaluated using the Newcastle-Ottawa Scale (NOS), a validated tool for evaluating the quality of observation studies that includes the following 3 subscales: selection, comparability, and exposure (Stang, 2010). We assessed all results for the level of evidence using the GRADE approach (Andrews et al., 2013), the average NOS score, and the number of patients included and then presented the data in a three-dimensional Manhattan plot.

Statistical Analysis

The Hardy-Weinberg equilibrium (HWE) of the included populations was calculated to assess the consistencies of allele frequencies between generations (Chen and Chatterjee, 2007). Association analysis was performed using five genetic models (Areeshi et al., 2013). The effect size was estimated by calculating the summary odds ratio (OR) and its 95% confidence intervals (CIs). The 2 statistics was used to estimate the degree of heterogeneity among the studies. If the 2 ≥ 50% (Q test, p < 0.1), a random-effect model was used; otherwise, a fixed-effect model was used. Subgroup analysis was also performed according to the ethnicity and location of the population. We assessed publication bias using the Begg and Egger tests. All tests were two-tailed, and a p-value of less than 0.05 was deemed statistically significant. We analyzed the data using the R program (Version 3.3.1) and STATA (Version 14.0).

We used GRADE to assess the level of evidence with four levels graded from high (best) to very low (worst). All objective studies used an observational study design, which downgrades the quality of evidence. Inconsistency was evaluated using the 2 and p-values of the Q test, and the outcome was imprecise if the standard error (SE) was larger than 0.2. Publication bias (reporting bias) also reduced the level of evidence. Finally, a large (OR > 2 or <0.5) or very large (OR > 5 or <0.2) effect upgraded the quality of evidence.
# TABLE 1 | Characteristics of the included studies.

| References          | Local          | Sample size         | Average age | Type of steroids | Primary disease                                      | Genes           | NOS score |
|---------------------|----------------|---------------------|-------------|------------------|-----------------------------------------------------|-----------------|-----------|
| Zhao et al., 2017   | China          | 193 (78/115)        | 40 (18–48)  | Prednisone       | SLE; ALL; RT; LT; NS; RA; AS                         | GRG             | 9         |
| Plesa et al., 2017  | Caucasian      | 304 (32/272)        | NA          | Prednisone       | ALL                                                 | BCL2L11         | 9         |
| Karol et al., 2015  | Multinational  | 2955 (400/2555)     | NA          | Prednisone;      | ALL etc.                                            | GWAS            | 7         |
| Wei et al., 2015    | China          | 75 (45/30)          | 39 ± 10     | Prednisone       | SLE; NS; ENT; dermatologic disease                  | ApoA1; ApoB;    | 8         |
| Zhang et al., 2014  | China          | 200 (94/106)        | 44.5 (18–82)| Prednisolone     | Hematologic; dermatologic; renal; ophthalmopathy;  | ABCB1           | 7         |
| Xue et al., 2014    | China          | 322 (105/217)       | 39 (18–48)  | Prednisone       | SLE; ALL; OT                                        | ABCB1           | 9         |
| Cui et al., 2014    | China          | 424 (223/201)       | 42.27 ± 15.7| Prednisolone     | NA                                                   | ApoA5           | 7         |
| Zeng et al., 2014   | China          | 206 (108/98)        | 40 ± 10     | Prednisone       | SLE; NS; RT; Purpura; ENT; dermatologic disease     | ApoB            | 8         |
| Zhang et al., 2013  | China          | 200 (94/106)        | 44.5 (18–82)| Prednisolone     | Hematologic; dermatologic; renal; ophthalmopathy;  | PAI-1           | 7         |
| Wang et al., 2013   | China          | 200 (94/106)        | 44.5 (18–82)| Prednisolone     | Hematologic; dermatologic; renal; ophthalmopathy;  | PON-1           | 6         |
| Li et al., 2012     | China          | 123 (70/53)         | 29 (18–73)  | Prednisone       | NA                                                   | ABCB1           | 6         |
| He, 2011a           | China          | 134 (63/71)         | 35.17 ± 11.73| Prednisone       | NA                                                   | ApoB; CYP1A2    | 8         |
| He, 2011b           | China          | 134 (63/71)         | 35.17 ± 11.73| Prednisone       | NA                                                   | Factor V; GR;   | 8         |
| Bond et al., 2011   | UK             | 110 (43/67)         | NA          | Dexamethasone    | ALL                                                  | CYP3A4; ABCB1;  | 7         |
| He and Li, 2009a    | China          | 48 (31/17)          | 32 (12–59)  | Prednisone       | NA                                                   | CYP1A2          | 8         |
| He and Li, 2009b    | China          | 48 (31/17)          | 18–60       | Prednisone       | SLE; RA; psoriasis; nephropathy; desmosis, etc.     | ABCB1; ApoB;    | 7         |
| Kuribayashi et al., 2008 | Japan       | 157 (34/123)      | 35 (9–64)   | Methylprednisolone; prednisolone                  | RT             | 9         |
| French et al., 2008 | USA            | 361 (51/310)        | NA (10–20)  | Prednisone;      | ALL                                                  | MDR1(ABCB1) et  | 7         |
| Wang et al., 2008   | China          | 53 (16/37)          | 35 (16–78)  | Methylprednisolone; prednisolone                  | RT             | 8         |
| Tamura et al., 2007 | Japan          | 157 (34/123)       | 35 (9–64)   | Methylprednisolone; prednisolone                  | RT             | 9         |
| Yang and Xu, 2007   | China          | 127 (21/106)        | 34 (11–67)  | Methylprednisolone; prednisolone                  | RT             | 8         |
| Hirata et al., 2007a| Japan          | 112 (20/92)         | NA          | Methylprednisolone; prednisolone                  | RT             | 9         |
| Hirata et al., 2007b| Japan          | 158 (34/124)       | 36.1 (9–64) | Methylprednisolone; prednisolone                  | RT             | 7         |
| Ekmecki et al., 2006| Turkey         | 57 (19/38)          | 34.2 ± 9.3  | NA          | RT                                                   | ApoB            | 8         |
| Celik et al., 2006  | Turkey         | 50 (11/39)          | 41 ± 11.79  | Prednisolone   | RT                                                   | Factor V; Prothrombin | 7 |
| Relling et al., 2004| USA            | 64 (25/39)          | 8.6 (2.7–18.8)| Prednisone     | ALL                                                  | MTHFR           | 8         |
| Asano et al., 2004  | Japan          | 137 (31/106)        | 36 (9–63)   | Methylprednisolone; prednisolone                  | RT             | 8         |
| Asano et al., 2003a | Japan          | 80 (26/54)          | NA          | Prednisolone   | RT                                                   | CYP3A4; CYP2D6; | 7 |

(Continued)
TABLE 1 | Continued

| References               | Local   | Sample size | Average age | Type of steroids        | Primary disease | Genes | NOS score |
|--------------------------|---------|-------------|-------------|-------------------------|-----------------|-------|-----------|
| Asano et al., 2003b      | Japan   | 136 (30/106) | 35.5 (9–63) | Methylprednisolone; prednisolone | RT              | ABCB1 | 8         |
| Ferrari et al., 2002     | Switzerland | 228 (26/202) | 50 ± 12     | Prednisone               | RT              | PAI-1 | 8         |

ALL, Acute lymphoblastic leukemia; AS, Ankylosing spondylitis; ENT, Ear, nose, and throat; GWAS, Genome-wide association study; LT, Liver transplant; NA, Not available; NOS, Newcastle-Ottawa Scale; NS, Nephrotic syndrome; OT, Organ transplant; RA, Rheumatic arthritis; RT, Renal transplant; SLE, Systemic lupus erythematosus.

#: Mean ± Standard deviation; Mean/Median (Minimum-Maximum).

##: TYMS; VDR; BGLAP; ESR1; LRP5; MTHFR; PAI-1; ABCB1(MDR1); PTH; PTHrP; AGPS.

###: CYP3A4; CYP3A5; TPMT; UGT1A1; TYMS; GSTT1; GSTM1; RFC; MTHFR; GRG(NR3C1); MDR1(ABCB1); VDR; GSTP1.

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FIGURE 1 | PRISMA flow chart illustrating the selection process of the studies included in our analysis. SNP, single-nucleotide polymorphism; SONFH, steroid-induced osteonecrosis of the femoral head.
RESULTS

Our research returned 278 English articles and 285 Chinese articles after removing duplications. After screening the titles and abstracts, 482 of these articles were excluded. The full texts of 81 articles were assessed, among which the control group did not receive steroid therapy (25); the article was a review (7); the study was non-SNP-related (5); the study was basic (3); the study did not report frequencies or effect size results (2); the study did not include steroid-related ONFH (2); the study was a case report (2); the research was microRNA related (2); the report was a duplicate (2); and the research was of heredity SONFH (1). Finally, we collected 30 trials assessing 7,553 patients for our meta-analysis (Table 1, Figure 1).

All studies used a case-control design except one that used a cohort design (Karol et al., 2015). In addition, Weibao Fang’s two studies (Wei, 2011a,b), and Kuribayashi and Tamura’s studies (Tamura et al., 2007; Kuribayashi et al., 2008) included the same patient population but did not assess the same SNPs. We analyzed different SNP results and excluded duplicated results. The NOS scores were six to nine points, and the overall quality was ideal in observational studies (Table 1).

Six SNPs in four genes were included in the meta-analysis. In the ABCB1 gene, rs1045642 is also known as C3435T, is located in the coding region, and is a synonymous mutation. In this research, the pooled results for rs1045642 showed that the C > T mutation protected against SONFH in the allelic model ($I^2 = 50.2\%$; OR: 0.74; 95% CI: 0.55–1.00; $p = 0.046$) (Figure 2, Table 2). In the ApoB gene, rs693 is located in the coding region. The results showed that there was no significant relationship between this mutation and SONFH in the allelic model ($I^2 = 58\%$; OR: 2.63; 95% CI: 0.92–7.54; $p = 0.072$).

However, other models showed that this mutation could increase SONFH risk (heterozygous model: $I^2 = 54.5\%$; OR: 2.46; 95%
## TABLE 2 | Meta-analysis of the associations between SNPs and SONFH in the SNP results in more than three studies.

| Gene | Location | SNP/HWE test | Model | No. of study | Sample size | OR  | LCI  | UCI  | P-value | \( P^2 \) | P for \( P^2 \) | Begg's test | Egger's test |
|------|----------|--------------|-------|-------------|-------------|-----|------|------|---------|--------|-------------|-------------|--------------|
| ABCB1 | Multinational | rs1045642 | Allelic model | 8 | 1141 | 0.74 | 0.55 | 1.00 | 0.046 | 50.20% | 0.02 | 0.902 | 0.797 |
|       |          |             |         | 8           | 1141        | 0.80 | 0.61 | 1.06 | 0.117 | 0%     | 0.758        | 1           | 0.891       |
|       |          |             | Homozygous model | 8 | 1141 | 0.48 | 0.20 | 1.15 | 0.101 | 67.10% | 0.06 | 0.548 | 0.458 |
|       |          |             | Control: 0.0423 | 9 | 1420 | 0.78 | 0.61 | 1.00 | 0.051 | 3.80% | 0.403        | 0.602        | 0.838       |
|       |          |             | Total: 0.0209 | 8 | 1141 | 0.57 | 0.24 | 1.37 | 0.21  | 71.80% | 0.002        | 0.548        | 0.427       |
| ABCB1 | China     | rs1045642 | Allelic model | 5 | 790  | 0.84 | 0.54 | 1.31 | 0.434 | 67.00% | 0.017        | 0.806        | 0.664       |
|       |          |             | Heterozygous model | 5 | 790  | 0.83 | 0.60 | 1.15 | 0.263 | 0.00%  | 0.803        | 0.806        | 0.427       |
|       |          |             | Case: 0.4946 | 5 | 790  | 0.64 | 0.18 | 2.27 | 0.493 | 79.00% | 0.003        | 1           | 0.934       |
|       |          |             | Control: 0.106 | 5 | 790  | 0.78 | 0.57 | 1.06 | 0.111 | 11.90% | 0.338        | 0.462        | 0.518       |
| ABCB1 | Multinational | rs2032582 | Allelic model | 7 | 964  | 0.85 | 0.58 | 1.23 | 0.382 | 63.40% | 0.012        | 1           | 0.517       |
|       |          |             | Heterozygous model | 7 | 964  | 0.78 | 0.56 | 1.08 | 0.138 | 11.70% | 0.34         | 0.042        |             |
|       |          |             | Case: 0.3666 | 7 | 964  | 0.68 | 0.32 | 1.42 | 0.3   | 60.10% | 0.028        | 0.707        | 0.731       |
|       |          |             | Control: 0.2588 | 7 | 964  | 0.81 | 0.59 | 1.10 | 0.167 | 39.30% | 0.13         | 1           | 0.227       |
|       |          |             | Total:0.0047 | 7 | 964  | 0.72 | 0.21 | 2.47 | 0.601 | 80.40% | 0.002        | 1           | 0.889       |
| ABCB1 | China     | rs2032582 | Allelic model | 5 | 764  | 0.97 | 0.60 | 1.59 | 0.914 | 70.60% | 0.009        | 0.806        | 0.408       |
|       |          |             | Heterozygous model | 5 | 764  | 0.80 | 0.55 | 1.15 | 0.231 | 41.00% | 0.148        | 1           | 0.077       |
|       |          |             | Case:0.0021 | 5 | 764  | 0.85 | 0.32 | 2.24 | 0.742 | 69.70% | 0.019        | 1           | 0.99        |
|       |          |             | Control:0.4073 | 5 | 764  | 0.82 | 0.45 | 1.51 | 0.528 | 57.70% | 0.051        | 0.806        | 0.238       |
|       |          |             | Total:0.009 | 5 | 764  | 1.10 | 0.49 | 2.44 | 0.819 | 68.30% | 0.024        | 1           | 0.87        |
| ApoB  | Multinational | rs693     | Allelic model | 4 | 570  | 2.63 | 0.92 | 7.54 | 0.072 | 58.00% | 0.068        | 1           | 0.67        |
|       |          |             | Heterozygous model | 4 | 570  | 2.46 | 1.27 | 4.77 | 0.008 | 54.50% | 0.111        | 0.461        |             |
|       |          |             | Case:0.0027 | 4 | 570  | 7.70 | 1.23 | 48.16 | 0.029 | 24.40% | 0.25         | 1           | NA          |
|       |          |             | Control:0.2173 | 5 | 725  | 2.99 | 1.71 | 5.21  | -0.001 | 31.40% | 0.212        | 0.624        | 0.82        |
| ApoB  | China     | rs693     | Allelic model | 3 | 415  | 2.82 | 0.51 | 15.57 | 0.235 | 72.20% | 0.028        | 1           | 0.77        |
|       |          |             | Heterozygous model | 3 | 415  | 1.59 | 0.32 | 7.86  | 0.57  | 70.20% | 0.067        | 1           | NA          |
|       |          |             | Case:0.0008 | 3 | 415  | 16.17 | 0.89 | 293.00 | 0.06  |        |             |             |             |
|       |          |             | Control: 1 | 3 | 415  | 2.41 | 0.54 | 10.82 | 0.252 | 62.40% | 0.07         | 1           | 0.881       |
| ApoB  | Multinational | rs1042031 | Allelic model | 4 | 572  | 2.90 | 1.49 | 5.66  | 0.002 | 50.30% | 0.11         | 0.308        | 0.146       |
|       |          |             | Case:0.0039 | 3 | 415  | 4.81 | 2.05 | 11.31 | <0.001 | 0.00%  | 0.886        | 1           | 0.769       |
| ApoB  | China     | rs1042031 | Allelic model | 3 | 251  | 0.92 | 0.59 | 1.44  | 0.71  | 12.90% | 0.317        | 1           | 0.401       |
|       |          |             | Heterozygous model | 3 | 251  | 0.62 | 0.33 | 1.18  | 0.144 | 22.10% | 0.277        | 1           | 0.549       |
| MTHFR | Multinational | rs1801133 | Allelic model | 3 | 251  | 1.24 | 0.48 | 3.22  | 0.653 | 0%     | 0.744        | 1           | NA          |
CI: 1.27–4.77; OR: 1.71–5.21; 95% CI: 0.002–0.296; p = 0.107).

DISCUSSION

Our study analyzed the association between genetic polymorphisms and SONFH risk by comparing ONFH and non-ONFH in steroid use populations. In the SNP results of the included studies, more than three allelic models of ABCB1 rs1045642 showed that mutations protect against SONFH. The ApoB rs693 and rs1042031 mutations increase SONFH risk. According to the GRADE guidelines, the evidence levels for ApoB rs693 and rs1042031 were moderate, and ABCB1 rs1045642 was very low.

In a previous meta-analysis, Gong et al analyzed 23 studies and 35 genes and indicated that PAI-1 rs1799768 and ABCB1 rs1045642, but not MTHFR rs1801133 or ABCB1 rs2032582, were related to the risk of osteonecrosis (Gong et al., 2013). However, in this study, the control population included a healthy group and a population with other types of ONFH, which might affect the accuracy. In our study, there was no relationship between PAI-1 rs1799768 after adjusting for the control population. Guo et al assessed the correlation between SONFH and hepatic CYP3A activity and high found that CYP3A activity could reduce SONFH risk. However, the conclusion was based on animal model results, and it is unclear whether the risk is reduced in humans (Guo and Deng, 2017).

ABCB1 is a member of a superfamily of ATP-binding cassette transporters that transport variant molecules across cellular membranes. Zhou et al. researched the association between ABCB1 polymorphisms and SONFH and found that rs1045642 and rs2032582 are associated with SONFH (Zhou et al., 2015). However, this study also included a healthy population in the control population. In contrast, a Chinese review showed that rs1045642 could reduce SONFH risk, while rs2032582 is not related to SONFH (Yang J. et al., 2016). For rs1045642, our results are the same as those of a previous study, but with a very low level of evidence. The mutation does not cause changes in the amino acid sequence; thus, the specific mechanism of SONFH risk change caused by this mutation is still unknown. However, this mutation has been shown to be related to the metabolism of some
drugs and to the prognosis of some cancers. This relationship may be related to differences in the characteristics of non-expressed proteins, such as codon bias, expression efficiency, and differences in mRNA characteristics. Therefore, this mutation is more suitable for clinical use as a predictor of SONFH risk for steroid users to guide individual drug application. People with the rs1045642 T allele have a low risk of SONFH with steroid application, while individuals with C allele have a high SONFH risk and should avoid long-term high-dose steroid application, with increased duration of follow-up and frequency of diagnosis. For rs2032582, our results found that the mutation is not related to SONFH (allelic model: \( p = 0.382 \)) with statistical heterogeneity (\( I^2 = 63.4\% \), \( p = 0.012 \)).

Additionally, we performed a post hoc analysis of Seth E. Karol’s research that excluded SNP results with very low-level evidence (Karol et al., 2015). Then, we matched potential SNPs with GO/KEGG annotations (Table 3). According to a gene set enrichment analysis based on overrepresentation enrichment analysis, only the molecular functions of ABCB1 and ApoB showed obvious relationships with lipid transporter activity (GO: 0005319) (Zhang et al., 2005). This finding indicates that at present, SNPs involved in lipid transporter activity are more related to SONFH risk than other SNPs, which supports the lipid metabolism disorder theory. ApoB is an important factor in normal lipid metabolism, the mutation of which can also be used as a predictor of SONFH occurrence to assess the risk of SONFH and to guide steroid clinical application. Although rs693 is a synonymous variant, rs693 is related to the circulating concentration of LDL cholesterol. T allele carriers have high levels of TG, TC, and LDL-C and low HDL-C (Sandhu et al., 2008). In addition, rs1042031 is a coding sequence variant that causes glutamate to lysine mutation (C > T), or the introduction of a stop codon in the amino acid sequence (C > A). The rs1042031 mutation is important for regulating the binding of apolipoprotein B to the LDL cholesterol receptor (Liu et al., 2013). In this work, these two mutations were found to be related to SONFH. Therefore, we hypothesized that the increase of circulating LDL induced by mutation increased the risk of SONFH, supporting the hypothesis that lipid metabolism disorders are associated with SONFH. Abnormal lipid metabolism leads to bone marrow adipogenesis of the femoral head that is mainly due to hyperlipidemia and inhibition of mitochondrial dehydrogenation by steroid application. The abnormal lipid metabolism then leads to fat embolism, which blocks local blood perfusion and exacerbates local inflammation. Therefore, the question remains: does reducing circulating LDL effectively reduce the risk of SONFH with steroid application? In animal models, it has been suggested that reduced circulating lipid levels can reduce the risk of SONFH (Yang Z. et al., 2016), but the clinical effect remains to be confirmed. Additionally, in the location subgroup analysis of rs1042031 results, the heterogeneity decreased significantly from 50 to 0%. The pool results of the three Chinese studies had low heterogeneity, and the population was concentrated in Guangxi and Shandong in China (Wei, 2011a; Zeng et al., 2014; Wei et al., 2015). Another study analyzed the Japanese population (Hirata et al., 2007b). Therefore, the centralization of the included population may be the main reason for the reduction in heterogeneity.

**Limitations**

The present study still has several limitations. First, our study was performed at the study level instead of the individual level. Second, our study did not consider the impact of primary disease or steroid therapy strategies on the results. Third, there is a large amount of heterogeneity in the results of this study. Although random effect models are used for heterogeneous results, these
heterogeneities can still have a potential impact on the results. Fourth, our study only analyzed the SNPs that were examined in more than three studies, but with new research and evidence, more SNPs may be studied. Therefore, this study was limited by the research available at the time.

CONCLUSION

An allelic model of ABCB1 rs1045642 showed that mutations had a protective effect against SONFH at a very low level of evidence. The mutations in ApoB rs693 and rs1042031 increased the SONFH risk with moderate levels of evidence.

**AUTHOR CONTRIBUTIONS**

XC performed the conception and design of the work. XC and LZ drafted and revised the work critically. DL and JL analyzed data for work. FL and HM acquired data for this work. All authors provide approval for publication of the content.

**FUNDING**

This study was supported by 2017 Special of Scientific Research of Traditional Chinese Medicine of Henan Province (No. 2017ZY2032) and 2016 Luoyang medical and health plan (No. 1603004A-8).
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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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