Meta-analysis of the pharmacogenetics of ARMS2 A69S polymorphism and the response to advanced age-related macular degeneration

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Abstract

Background

Age-related macular degeneration (AMD) causes irreversible vision loss, and targeted anti-vascular endothelial growth factor (VEGF) therapy is now the most common and effective treatment. The aim of this meta-analysis is to discuss whether genetic polymorphism of ARMS2 A69S could confer susceptibility to advanced AMD with the response to anti-VEGF treatment.

Methods

We performed a meta-analysis of relevant published studies selected through electronic databases. A total of 21 preferred studies regarding the association between ARMS2 gene and anti-VEGF treatment response in advanced AMD were generally included in the meta-analysis.

Results

The pooled results demonstrated that the carriage of G allele for ARMS2 A69S presented a better clinical prognosis for advanced AMD treated with anti-VEGF drugs (OR=1.38, 95% CI=1.13-1.69, P=0.002). Additionally, in the subgroup analysis based on ethnicity, ARMS2 polymorphisms were more likely to be a positive responder for East Asian patients (OR=1.67, 95% CI=1.29-2.16, P<0.001).

Conclusion

This meta-analysis through a series of rigorous methodology data demonstrated a significant association between ARMS2 A69S polymorphism and the anti-VEGF treatment response in advanced AMD, especially among East Asian population. Numerous well-designed, randomized, multicenter clinical trials with large sample size are required to validate the association.

Background

Age-related macular degeneration (AMD) is a chronic, progressive condition presenting untreatable in up to 90% of patients, and is a major cause of irreversible blindness in the elderly individuals [1]. AMD primarily affects people aged over 50, and implicates a population of 30–50 million worldwide [2]. About two thirds of people aged over 80 years are afflicted by AMD to some degree in USA [3], while the prevalence of AMD among Asian population aged 40 to 79 is estimated at 6.8% [4]. AMD is classified into two types, dry and wet, based on the presence or absence of choroidal neovascularization and bleeding, which further differentiated into exudative AMD (advanced AMD) and geographic atrophy (late “dry” AMD). In advanced AMD, choroidal neovascularization breaks through to the neural retina, causing the leakage of fluid, lipids, and blood, thus leading to fibrous scarring. Although the etiology of AMD is multifactorial, some common risk factors including ageing, smoking, gender, hypertension, obesity and lifetime light exposure have certain epidemiologic associations with its progression [5].

It has been demonstrated that vascular endothelial growth factor (VEGF) could be extracted in choroidal neovascular membranes obtained from AMD patients [6]. Besides, the VEGF levels in the aqueous humor of AMD patients were significantly increased as compared with control eyes without ocular or systemic diseases [7]. In animal models, the expression of VEGF has been identified in experimental choroidal neovascularization, and it presented tendency to induce the neovascularization [8]. Intravitreal treatment with anti-VEGF drugs like ranibizumab, aflibercept, bevacizumab, remains a valid approach to tackle the progression of advanced AMD [9], which dramatically intensifies or stabilizes visual acuity in a large number of patients. Recently, a long time-span prospective study has corroborated that legal blindness attributable to AMD could be reduced by half in some countries when introducing VEGF antagonists [10].

Current targeted therapies for advanced AMD are based on VEGF inhibition. However, we should take into account the potential risk of long-term consequences of intraocular VEGF suppression on the retina due to inhibition on physiological VEGF function. A large community of AMD patients receiving anti-VEGF treatment could not have their lost vision properly improved or recovered, even with persistent macular edema, despite continuous injection of bevacizumab or ranibizumab [11, 12]. Other etiologies like genetic backgrounds may illustrate the likelihood. Just as Andreoli et al. pointed out that each individual would have a

characteristic genome phenotype towards intravitreal injection [13], we have had an eye on gene polymorphism as to this tough problem.

Though the most significant loci associated with advanced AMD is focused on the gene for complement factor H located on chromosome 1q31, increasing evidence from the latest large case-control genome-wide association studies confirmed the association with other genes involved as candidate risk factors. The polymorphisms of age-related maculopathy susceptibility 2 (ARMS2, also called LOC387715) gene at 10q26 locus have been reported to be strongly associated with the inverse effect of hormone replacement therapy on AMD. Investigators have demonstrated that the encoded mRNA and corresponding peptide are expressed in the retina, denoting that ARMS2 transcript takes the responsibility for the relationship with AMD [14]. Besides, the ARMS2 protein has been described as a component of the extracellular matrix and its mRNA owns a unique splice form in the retina but commonly no in other tissues [15, 16]. Polymorphic site A69S (rs10490924) within the ARMS2 locus was mainly investigated in several studies in regard to AMD, and the results showed strong correlation of poor visual acuity in advanced AMD with the response of ant-VEGF intervention [13].

Although several studies have been reasonably substantiated that ARMS2 A69S polymorphism conferred susceptibility to advanced AMD, others have declared no such associations. We postulated that these disparities are probably triggered by small sample size, ethnic difference, low statistical power, or clinical heterogeneity. In order to better understand the natural association of ARMS2 genetic polymorphism with the anti-VEGF treatment response in advanced AMD, we conducted a systematic meta-analysis. The present study aimed to determine whether ARMS2 A69S polymorphism plays a crucial role in regulating the risk of advanced AMD.

Materials And Methods

Search strategy

A comprehensive online search for relevant studies in the common electronic databases including PubMed, Web of Science, EMBASE, and Cochrane Library was performed by two independent researchers for the meta-analysis. Search terms in this study were “gene”, “polymorphism”, “SNP”, “variant”, “macular degeneration”, “age-related macular degeneration”, “AMD”, “age-related maculopathy susceptibility 2”, “ARMS2”, “LOC387715”, “anti-vascular endothelial growth factor” “anti-VEGF” and “anti-angiogenesis”. Search syntaxes like “OR” and “AND” were used between those search terms. The studies were searched without any limitation on language, ethnicity, and publication year. In addition, the reference lists from retrieved studies were also checked to identify the source of any relevant ones that were not yet retrieved by our computerized databases. The final search was up-to-date as of November 8, 2019.

Literature screening

The available studies retrieved from electronic databases were inspected in terms of the title, keywords, abstract, and full-text in our meta-analysis. Any disputes were resolved by group consensus through discussion in collaboration with a third researcher. Studies were considered eligible for inclusion if they met the following criteria: a) evaluation on the relationship between ARMS2 A69S polymorphism and the anti-VEGF treatment response in advanced AMD; b) independent retrospective or prospective study; c) detailed numbers or percentages in genotype distribution of ARMS2 A69S could be obtained in patients; d) the study subjects were human beings. Results from previous article, conference, case report, review, opinion, letter, and insufficient raw data after contacting with the corresponding author were extracted and excluded.

Data extraction and quality assessment

Detailed data were carefully extracted from our eligible studies independently by two researchers (J.Z. and Z.L.). The collected data from each study contained first author, publication year, nationality and racial make-up of the study population, study size, characteristics of included cases, information on study design, frequency or percentage in genotype distribution of ARMS2 A69S,

treatment modalities, and study endpoint. The quality evaluation of the selected studies was also performed by two researchers (J.Z. and Z.L.) using the Newcastle-Ottawa scale [17]. A study with more than six stars was regarded as high quality, and one with nine stars was regarded as the highest quality. The listed information was rigorously inspected by two researchers (J.Z. and Z.L.), and an absolute consensus on all respects of an included paper was reached without any disputes. Any disagreement would be validated and resolved by a third researcher (S.H.).

Statistical analysis

A series of quantitative analyses were performed by the statistical software STATA V.12.0 (StataCorp LP, College Station, Texas, USA). The odds ratio (OR) and 95% confidence interval (CI) were calculated, and pooled ORs were evaluated for the comparison on the frequency of ARMS2 A69S polymorphism in advanced AMD patients. Cochran's Q test was used to identify potential heterogeneity ($P < 0.1$, treated as significant level across all studies). Moreover, the quantitative I^2 statistic was utilized for evaluation of inconsistency in our meta-analysis, representing the percentage of the observed variability due to heterogeneity rather than chance. The quantification of heterogeneity was assigned of low, moderate, high, and extremely high to I^2 values of 0–25%, 25–50%, 50–75%, and 75–100%, respectively. Either the fixed-effect model ($I^2 < 50\%$ and $P > 0.1$) or random-effect model ($I^2 \geq 50\%$ and $P < 0.1$) was applied for the pooled ORs and 95%CIs according to the heterogeneity. We also performed sensitivity analysis to assess the effect of each study on the pooled ORs by omitting one study in each turn. Moreover, subgroup analysis was executed to apprehend the strength of association between different ethnicities. Series of meta-regression analysis were applied for the discussion of main heterogeneity. Publication bias was estimated by Begg's test with the funnel plot and Egger's regression test. P value less than 0.05 was considered statistically significant in these comparisons.

Results

General characteristics of selected studies and quality assessment

A flow diagram of study selection process is presented in Figure 1. The search strategy resulted in 223 relevant articles from the four databases. Of which, 175 studies were excluded for duplication (156 articles) and ineligible categories like abstract, review, conference, and case report (19 articles). The rest 48 full-text articles were left in our meta-analysis. Among them, 11 studies did not provide detailed genotype results after perusing the documents and additional files, 16 articles did not discuss the association between *ARMS2* and anti-VEGF intervention. Finally, a total of 21 preferred studies regarding the association between *ARMS2* gene and the anti-VEGF treatment response in advanced AMD were generally included in the current meta-analysis [18-38]. The main characteristics and data of the enrolled studies are listed in Table 1. Regarding ethnicity, nine of these studies were performed in Caucasian, eleven in East Asian, and one in Middle East people. Four studies used aflibercept, three used bevacizumab, and eight used ranibizumab, while four studies used either ranibizumab or bevacizumab, one used bevacizumab or photodynamic therapy, and another one used VEGF inhibition. The follow-up duration ranged from three months to two years. We classified the definition of positive or negative response based on the improvement on visual acuity or macular morphological changes. Moreover, the quality assessment of all included studies is presented in the Table 1 and all studies were of high quality with more than six stars.

Bias assessment of the included studies

The evaluation of potential bias was primarily expounded in **Table 2**. Thirteen studies were prospective, and eight was retrospective. Overall, the quality of the included studies was consistently robust. Of the studies, there were no obvious biases in the selection of advanced AMD patients, confounding, or selective outcome reports. The result with definition of a positive responder could be validated in our meta-analysis.

Association between *ARMS2* A69S polymorphism and advanced AMD susceptibility with anti-VEGF treatment

We meta-analyzed the 21 included studies for the pooled associations between treatment response in advanced AMD and *ARMS2* A69S genotypes in **Table 3**. We summarized the statistical differences involving five genotype comparisons (19 studies for G *vs* T, GG *vs* TT, and GT *vs* TT, 20 studies for GG+GT *vs* TT, and GG *vs* GT+TT) of the study population (allelic model: OR=1.38, 95% CI=1.13-1.69, P=0.002, random model; homozygote model: OR=1.88, 95% CI=1.32-2.67, P<0.001, random model; heterozygote model: OR=1.36, 95% CI=1.14-1.62, P=0.001, fixed model; dominant model: OR=1.62, 95% CI=1.27-2.06, P<0.001, random model; recessive model: OR=1.51, 95% CI=1.15-2.00, P=0.004, random model) (**Figure 2-6**). Besides, subgroup analysis based on three different racial groups revealed that there were no significant differences between any Caucasian or Middle East genotypic comparison models, but a significant association was indicated in the pooled analyses between A69S and anti-VEGF effects in all genotypic comparisons for East Asian participants (allelic model: OR=1.67, 95% CI=1.29-2.16, P<0.001, random model; homozygote model: OR=2.38, 95% CI=1.44-3.93, P=0.001, random model; heterozygote model: OR=1.50, 95% CI=1.19-1.62, P=0.001, fixed model; dominant model: OR=1.62, 95% CI=1.27-2.06, P<0.001, random model; recessive model: OR=1.51, 95% CI=1.15-2.00, P=0.004, random model) (**Table 3**).

Heterogeneity test and sensitivity analysis

Significant heterogeneity between the A69S polymorphism in *ARMS2* gene and the anti-VEGF treatment response in advanced AMD was observed among four genotype comparisons (all P<0.1) (**Figure 2, 3, 5, 6**). Our subgroup analysis in the homozygote and recessive model suggested the primary heterogeneity owed to racial origin (**Table 3**). The pooled ORs for sensitivity analysis remained substantial when omitting one study at each time. Our results demonstrated that no study absolutely changed the relationship between *ARMS2* A69S and treatment response in advanced AMD, signifying that the pooled ORs of the present meta-analysis were relatively robust (**Figure 7,** others in supplementary Figure 1-4).

Meta-regression

We tried to apply meta-regression for analyzing the heterogeneity of G *vs* T genotype to the greatest extent. The study design, ethnicity, follow up duration, intervention, mean age, and study size were thoroughly treated as independent covariate factors. As delineated in **Table 4**, our meta-regression analysis showed no significant correlations between the above variables except ethnicity and number of cases (for ethnicity: P=0.006, 95% CI=1.35-4.71; number of cases: P=0.011, 95% CI=1.24-4.41)**.**

Publication bias

The Begg's test with the funnel plot and Egger's regression test were performed to assess the source of publication bias between *ARMS2* A69S and the treatment response in advanced AMD. There were no conspicuous asymmetries of funnel plots in any genotypic comparisons (**Figure 8,** others in supplementary Figure 5-8); moreover, no statistically significant relationship was found in Egger's regression test, suggesting no apparent evidence of publication bias (P>0.05, **Table 5**).

Discussion

AMD is the predominant cause for irreversible blindness in senile population worldwide, especially the advanced type. Anti-VEGF treatment is the primary therapy for patients with vision loss caused by advanced AMD [39, 40]. A total of 13 prospective and 8 retrospective studies involving 4,008 cases with the association between anti-VEGF and A69S was included in our current meta-analysis. Our results revealed that ARMS2 A69S allele might be a better clinical prognosis for advanced AMD in the involved population among the five genetic models (allelic model: OR = 1.38, 95% CI = 1.13–1.69; homozygote model: OR = 1.88, 95% CI = 1.32–2.67; heterozygote model: OR = 1.36, 95% CI = 1.14–1.62; dominant model: OR = 1.62, 95% CI = 1.27–2.06; recessive model: OR = 1.51, 95% CI = 1.15-2.00). Specifically, the pooled results based on ethnicity indicated that there was a significant association between ARMS2 A69S and anti-VEGF treatment response in exudative AMD in the East Asian group, but not in Caucasian or Middle East group. Thus, these findings may enable clinicians to make the best of these effective medications, at least for East Asian patients.

In our meta-analysis, a group of patients investigated by Park et al. [28] had partial polypoidal choroidal vasculopathy and received combined treatment consisting of photodynamic therapy with intravitreal bevacizumab, another group of patients in the same condition explored by Hata et al. [32] were treated with ranibizumab, and the rest studies focused on advanced AMD subtype. Being consistent with the positive summarized effect, removing the genotype of the cases with polypoidal choroidal vasculopathy exerted no influence on the overall comparison due to its small sample size (n = 51, n = 77, respectively).

Obviously, considerable genetic heterogeneity existed in our current study, though the results of sensitivity analysis by removing one study in each turn showed that the present meta-analysis were relatively robust. Our subgroup analysis based on involved races in the study population suggested that heterogeneity is particularly pronounced among East Asian and Middle East population. Meta-regression was carried out to explore the heterogeneity of G vs T allele, and the results indicated that ethnicity and number of cases were the origin of heterogeneous factors. Additionally, heterogeneity may also have arisen from the genetic background, sex difference, lifestyle, the level of socio-economic development, genotyping method, and definition of response.

It has been verified that ARMS2 lies in the AMD susceptibility locus identified on chromosome 10q26 and is commonly expressed in the retina. Evidence has suggested that ARMS2 variants could bring about dysfunction of retinal pigment epithelium due to mitochondrial DNA damage that accumulates in the retina and its pigment epithelium. Sobrin et al. [41] suggested that genetic mutation at this locus has conferred a differential risk for choroidal neovascularization versus geographic atrophy. With respect to the possible biological mechanism, the ARMS2 gene product has been localized to the mitochondrial outer membrane; it has been proposed that the A69S polymorphism could alter ARMS2 function and increase the susceptibility of photoreceptor cells to oxidative damage and ageing [14]. Furthermore, the mitochondria are known to be a major source of superoxide anion in the cells, implying that damages to the mitochondria would bring about oxidative stress in neovascular AMD possibly affecting the response to anti-VEGF inhibitors. Besides, Kortvely et al. [15] reported that ARMS2 protein is mostly confined to choroid pillars in human eyes, corresponding to the principal sites of drusen deposition. Accordingly, abnormal expression of ARMS2 would exhibit higher susceptibility to the development of drusen and AMD. As such, it is understandable to increase the risk of developing AMD, but the role of non-synonymous A69S single nucleotide polymorphism in the pathogenesis of influencing response to anti-VEGF treatment remains unknown. Abedi et al. [22] implied that the loss of structural integrity of Bruch's membrane, driven by oxidative pathways mediated by ARMS2 A69S variants, may largely contribute to the underlying biological mechanisms for a weak response to intravitreal anti-VEGF treatment.

Yamashiro et al. [36] argued that ARMS2 A69S was not significantly associated with the changes in visual acuity or anatomic outcomes during the one-year anti-VEGF treatment in treatment-naïve advanced AMD patients, while Kitchens et al. [24] confirmed that A69S was positively correlated with the achievement of dry maculae after three monthly injections of ranibizumab or bevacizumab, suggesting better response of ARMS2 A69S with advanced AMD. Moreover, Teper et al. [42] indicated a significant influence of the risk homozygosis (TT) for ARMS2 A69S on the visual response after ranibizumab treatment. The differences could be owed to the small sample size and relatively short-term follow-up period in those studies. We have a large enough sample size containing 4008 cases in the current meta-analysis, the results strongly demonstrated that G allele carriage or GG homozygosis might be useful for better predicting visual outcomes after anti-VEGF treatment for advance AMD.

A previous meta-analysis conducted by Hu et al. [43] has revealed a significant association between ARMS2 A69S and the response to anti-angiogenesis treatment in advanced AMD patients among three genotypic models (homozygote model: OR = 1.34, 95% CI 1.01 to 1.77, P = 0.039; heterozygote model: OR = 1.58, 95% CI 1.08 to 2.31, P = 0.018; dominant model: OR = 1.74, 95% CI 1.19 to 2.52, P = 0.004). Consistently, our meta-analysis also found the significance of the difference in the comparison of G vs T (OR = 1.38, 95% CI = 1.13–1.69, P = 0.002). Moreover, our meta-analysis focused on three ethnicities including East Asian, Caucasian, and Middle East population, though no significant association was validated in last two races. Furthermore, the pooled results in the current study did not delineate any inconsistency in the exploration of sensitivity test and publication bias, which differs from the conclusions from Hu et al. [43]. The explanation for this difference could be drawn on the increasing number of studies and larger sample size, thus guaranteeing our cumulative ORs higher definiteness, stability, and accuracy than before.

In order to accurately interpret our combined results, several potential limitations of the current study should not be ignored. First, heterogeneity based on meta-regression was found in covariate factors including ethnicity and number of cases. Second, the

criteria for positive and negative therapeutic responses based on visual acuity come in great variety among studies, though the included ones were given adequate definition of positive and negative responses. Third, the number of patients was relatively small in each study; thus, a research with larger sample size involving different ethnic regions is necessary for further analysis. Fourth, the definition of response and the study endpoint mostly came from changes in visual acuity or optical coherence tomography. Changes in central macular thickness and maximum lesion thickness from baseline were also meaningful outcomes owing to anti-VEGF treatment in advanced AMD community. Unfortunately, the included studies failed to point out any relationship between these two important treatment outcomes and the genetic variants. Fifth limitation is that only three ethnic backgrounds with relatively small studies were included in this meta-analysis, thus further efforts to lower the incidence of ethnic bias will be taken into account. Finally, all included studies were of high quality, but presenting a challenge that the non-randomized study design used was not considered in this meta-analysis.

Conclusion

The current meta-analysis demonstrated a series of rigorous methodology data for a significant association between ARMS2 A69S and the anti-VEGF treatment response in advanced AMD, especially in East Asian. More studies, well-designed work with randomized, multicenter clinical trials or intervention studies incorporating different ethnicities together with gene-gene or gene-environment interaction, are needed to further prove the bio-functional role of this common ARMS2 polymorphism.

Declarations

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Availability of data and materials

All data has been shared in the Figures and Tables.

Authors' contributions

J.Q. conceived the study. J.Z. and Z.L. performed and checked the available information from eligible articles in this meta-analysis. S.H. analyzed the data. Z.L. prepared the Figure 1-8, supplementary Figure 1-8 and Table 1-5. J.Z. wrote the main manuscript text. J.Q. reviewed and revised the manuscript. All authors read and approved the final manuscript.

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Declaration of competing interests

The authors declare that they have no conflicts of interest to this work.

References

1. Ambati J, Fowler BJ: **Mechanisms of age-related macular degeneration**. *Neuron* 2012, **75**(1):26-39.
2. Javitt JC, Zhou Z, Maguire MG, Fine SL, Willke RJ: **Incidence of exudative age-related macular degeneration among elderly Americans**. *Ophthalmology* 2003, **110**(8):1534-1539.
3. Friedman DS, O'Colmain BJ, Munoz B, Tomany SC, McCarty C, de Jong PT, Nemesure B, Mitchell P, Kempen J, Eye Diseases Prevalence Research G: **Prevalence of age-related macular degeneration in the United States**. *Archives of ophthalmology* 2004, **122**(4):564-572.
4. Kawasaki R, Yasuda M, Song SJ, Chen SJ, Jonas JB, Wang JJ, Mitchell P, Wong TY: **The prevalence of age-related macular degeneration in Asians: a systematic review and meta-analysis**. *Ophthalmology* 2010, **117**(5):921-927.
5. Wong CW, Yanagi Y, Lee WK, Ogura Y, Yeo I, Wong TY, Cheung CMG: **Age-related macular degeneration and polypoidal choroidal vasculopathy in Asians**. *Progress in retinal and eye research* 2016, **53**:107-139.
6. Lopez PF, Sippy BD, Lambert HM, Thach AB, Hinton DR: **Transdifferentiated retinal pigment epithelial cells are immunoreactive for vascular endothelial growth factor in surgically excised age-related macular degeneration-related choroidal neovascular membranes**. *Investigative ophthalmology & visual science* 1996, **37**(5):855-868.
7. Tong JP, Chan WM, Liu DT, Lai TY, Choy KW, Pang CP, Lam DS: **Aqueous humor levels of vascular endothelial growth factor and pigment epithelium-derived factor in polypoidal choroidal vasculopathy and choroidal neovascularization**. *American journal of ophthalmology* 2006, **141**(3):456-462.
8. Lim LS, Mitchell P, Seddon JM, Holz FG, Wong TY: **Age-related macular degeneration**. *Lancet* 2012, **379**(9827):1728-1738.
9. Chakravarthy U, Harding SP, Rogers CA, Downes SM, Lotery AJ, Culliford LA, Reeves BC, investigators Is: **Alternative treatments to inhibit VEGF in age-related choroidal neovascularisation: 2-year findings of the IVAN randomised controlled trial**. *Lancet* 2013, **382**(9900):1258-1267.
10. Bloch SB, Larsen M, Munch IC: **Incidence of legal blindness from age-related macular degeneration in denmark: year 2000 to 2010**. *American journal of ophthalmology* 2012, **153**(2):209-213 e202.
11. Kanoff J, Miller J: **Pharmacogenetics of the treatment response of age-related macular degeneration with ranibizumab and bevacizumab**. *Seminars in ophthalmology* 2013, **28**(5-6):355-360.
12. Fauser S, Lambrou GN: **Genetic predictive biomarkers of anti-VEGF treatment response in patients with neovascular age-related macular degeneration**. *Survey of ophthalmology* 2015, **60**(2):138-152.
13. Andreoli MT, Morrison MA, Kim BJ, Chen L, Adams SM, Miller JW, DeAngelis MM, Kim IK: **Comprehensive analysis of complement factor H and LOC387715/ARMS2/HTRA1 variants with respect to phenotype in advanced age-related macular degeneration**. *American journal of ophthalmology* 2009, **148**(6):869-874.
14. Kanda A, Chen W, Othman M, Branham KE, Brooks M, Khanna R, He S, Lyons R, Abecasis GR, Swaroop A: **A variant of mitochondrial protein LOC387715/ARMS2, not HTRA1, is strongly associated with age-related macular degeneration**. *Proceedings of the National Academy of Sciences of the United States of America* 2007, **104**(41):16227-16232.
15. Kortvely E, Hauck SM, Duetsch G, Gloeckner CJ, Kremmer E, Alge-Priglinger CS, Deeg CA, Ueffing M: **ARMS2 is a constituent of the extracellular matrix providing a link between familial and sporadic age-related macular degenerations**. *Investigative ophthalmology & visual science* 2010, **51**(1):79-88.
16. Wang G, Scott WK, Whitehead P, Court BL, Kovach JL, Schwartz SG, Agarwal A, Dubovy S, Haines JL, Pericak-Vance MA: **A novel ARMS2 splice variant is identified in human retina**. *Experimental eye research* 2012, **94**(1):187-191.
17. Stang A: **Critical evaluation of the Newcastle-Ottawa scale for the assessment of the quality of nonrandomized studies in meta-analyses**. *European journal of epidemiology* 2010, **25**(9):603-605.
18. Brantley MA, Jr., Fang AM, King JM, Tewari A, Kymes SM, Shiels A: **Association of complement factor H and LOC387715 genotypes with response of exudative age-related macular degeneration to intravitreal bevacizumab**. *Ophthalmology* 2007, **114**(12):2168-2173.
19. Orlin A, Hadley D, Chang W, Ho AC, Brown G, Kaiser RS, Regillo CD, Godshalk AN, Lier A, Kaderli B *et al*: **Association between high-risk disease loci and response to anti-vascular endothelial growth factor treatment for wet age-related macular degeneration**. *Retina* 2012, **32**(1):4-9.

20. Yamashiro K, Tomita K, Tsujikawa A, Nakata I, Akagi-Kurashige Y, Miyake M, Ooto S, Tamura H, Yoshimura N: **Factors associated with the response of age-related macular degeneration to intravitreal ranibizumab treatment**. *American journal of ophthalmology* 2012, **154**(1):125-136.
21. Park DH, Kim IT: **LOC387715/HTRA1 variants and the response to combined photodynamic therapy with intravitreal bevacizumab for polypoidal choroidal vasculopathy**. *Retina* 2012, **32**(2):299-307.
22. Abedi F, Wickremasinghe S, Richardson AJ, Islam AF, Guymer RH, Baird PN: **Genetic influences on the outcome of anti-vascular endothelial growth factor treatment in neovascular age-related macular degeneration**. *Ophthalmology* 2013, **120**(8):1641-1648.
23. Lotery AJ, Gibson J, Cree AJ, Downes SM, Harding SP, Rogers CA, Reeves BC, Ennis S, Chakravarthy U, Alternative Treatments to Inhibit ViPwA-RCNSG: **Pharmacogenetic associations with vascular endothelial growth factor inhibition in participants with neovascular age-related macular degeneration in the IVAN Study**. *Ophthalmology* 2013, **120**(12):2637-2643.
24. Kitchens JW, Kassem N, Wood W, Stone TW, Isernhagen R, Wood E, Hancock BA, Radovich M, Waymire J, Li L *et al*: **A pharmacogenetics study to predict outcome in patients receiving anti-VEGF therapy in age related macular degeneration**. *Clinical ophthalmology* 2013, **7**:1987-1993.
25. Hautamaki A, Kivioja J, Vavuli S, Kakko S, Savolainen ER, Savolainen MJ, Liinamaa MJ, Seitsonen S, Onkamo P, Jarvela I *et al*: **Interleukin 8 promoter polymorphism predicts the initial response to bevacizumab treatment for exudative age-related macular degeneration**. *Retina* 2013, **33**(9):1815-1827.
26. Hagstrom SA, Ying GS, Pauer GJT, Sturgill-Short GM, Huang J, Callanan DG, Kim IK, Klein ML, Maguire MG, Martin DF *et al*: **Pharmacogenetics for genes associated with age-related macular degeneration in the Comparison of AMD Treatments Trials (CATT)**. *Ophthalmology* 2013, **120**(3):593-599.
27. Fang K, Tian J, Qing X, Li S, Hou J, Li J, Yu W, Chen D, Hu Y, Li X: **Predictors of visual response to intravitreal bevacizumab for treatment of neovascular age-related macular degeneration**. *Journal of ophthalmology* 2013, **2013**:676049.
28. Park UC, Shin JY, Kim SJ, Shin ES, Lee JE, McCarthy LC, Newcombe PJ, Xu CF, Chung H, Yu HG: **Genetic factors associated with response to intravitreal ranibizumab in Korean patients with neovascular age-related macular degeneration**. *Retina* 2014, **34**(2):288-297.
29. Matsumiya W, Honda S, Yanagisawa S, Miki A, Nagai T, Tsukahara Y: **Evaluation of clinical and genetic indicators for the early response to intravitreal ranibizumab in exudative age-related macular degeneration**. *Pharmacogenomics* 2014, **15**(6):833-843.
30. van Asten F, Rovers MM, Lechanteur YT, Smailhodzic D, Muether PS, Chen J, den Hollander AI, Fauser S, Hoyng CB, van der Wilt GJ *et al*: **Predicting non-response to ranibizumab in patients with neovascular age-related macular degeneration**. *Ophthalmic epidemiology* 2014, **21**(6):347-355.
31. Kuroda Y, Yamashiro K, Miyake M, Yoshikawa M, Nakanishi H, Oishi A, Tamura H, Ooto S, Tsujikawa A, Yoshimura N: **Factors Associated with Recurrence of Age-Related Macular Degeneration after Anti-Vascular Endothelial Growth Factor Treatment: A Retrospective Cohort Study**. *Ophthalmology* 2015, **122**(11):2303-2310.
32. Hata M, Tsujikawa A, Miyake M, Yamashiro K, Ooto S, Oishi A, Nakanishi H, Takahashi A, Yoshimura N: **Two-year visual outcome of ranibizumab in typical neovascular age-related macular degeneration and polypoidal choroidal vasculopathy**. *Graefe's archive for clinical and experimental ophthalmology = Albrecht von Graefes Archiv fur klinische und experimentelle Ophthalmologie* 2015, **253**(2):221-227.
33. Chaudhary V, Brent M, Lam WC, Devenyi R, Teichman J, Mak M, Barbosa J, Kaur H, Carter R, Farrokhyar F: **Genetic Risk Evaluation in Wet Age-Related Macular Degeneration Treatment Response**. *Ophthalmologica Journal international d'ophtalmologie International journal of ophthalmology Zeitschrift fur Augenheilkunde* 2016, **236**(2):88-94.
34. Bardak H, Bardak Y, Ercalik Y, Turkseven Kumral E, Imamoglu S, Gunay M, Ozbas H, Bagci O: **Effect of ARMS2 gene polymorphism on intravitreal ranibizumab treatment for neovascular age-related macular degeneration**. *Genetics and molecular research : GMR* 2016, **15**(4).
35. Kikushima W, Sakurada Y, Yoneyama S, Sugiyama A, Tanabe N, Kume A, Mabuchi F, Iijima H: **Incidence and risk factors of retreatment after three-monthly aflibercept therapy for exudative age-related macular degeneration**. *Scientific reports* 2017, **7**:44020.

36. Yamashiro K, Mori K, Honda S, Kano M, Yanagi Y, Obana A, Sakurada Y, Sato T, Nagai Y, Hikichi T *et al*: **A prospective multicenter study on genome wide associations to ranibizumab treatment outcome for age-related macular degeneration**. *Scientific reports* 2017, **7**(1):9196.
37. Park UC, Shin JY, Chung H, Yu HG: **Association of ARMS2 genotype with response to anti-vascular endothelial growth factor treatment in polypoidal choroidal vasculopathy**. *BMC ophthalmology* 2017, **17**(1):241.
38. Mohamad NA, Ramachandran V, Mohd Isa H, Chan YM, Ngah NF, Ching SM, Hoo FK, Wan Sulaiman WA, Inche Mat LN, Mohamed MH: **Association of HTRA1 and ARMS2 gene polymorphisms with response to intravitreal ranibizumab among neovascular age-related macular degenerative subjects**. *Human genomics* 2019, **13**(1):13.
39. Group CR, Martin DF, Maguire MG, Ying GS, Grunwald JE, Fine SL, Jaffe GJ: **Ranibizumab and bevacizumab for neovascular age-related macular degeneration**. *The New England journal of medicine* 2011, **364**(20):1897-1908.
40. Liew G, Mitchell P: **Ranibizumab for neovascular age-related macular degeneration**. *The New England journal of medicine* 2007, **356**(7):747-748; author reply 749-750.
41. Sobrin L, Reynolds R, Yu Y, Fagerness J, Leveziel N, Bernstein PS, Souied EH, Daly MJ, Seddon JM: **ARMS2/HTRA1 locus can confer differential susceptibility to the advanced subtypes of age-related macular degeneration**. *American journal of ophthalmology* 2011, **151**(2):345-352 e343.
42. Teper SJ, Nowinska A, Pilat J, Palucha A, Wylegala E: **Involvement of genetic factors in the response to a variable-dosing ranibizumab treatment regimen for age-related macular degeneration**. *Molecular vision* 2010, **16**:2598-2604.
43. Hu Z, Xie P, Ding Y, Yuan D, Liu Q: **Association between variants A69S in ARMS2 gene and response to treatment of exudative AMD: a meta-analysis**. *The British journal of ophthalmology* 2015, **99**(5):593-598.

Tables

Table 1. The general characteristics of all studies included in our meta-analysis.

Refs	Year	Country	Ethnicity	Number of cases	Intervention	Mean age (years)	Follow up (months)	Endpoint	Quality score
Brantley et al.	2007	America	Caucasian	86	IVB	79.8	6	VA(Snellen)	8
Orlin et al.	2012	America	Caucasian	149	IVR / IVB	80.6	24	VA(Snellen)	7
Yamashiro et al.	2012	Japan	East Asian	77	IVR	75.1	12	OCT	6
Park et al.	2012	Korea	East Asian	51	IVB +PDT	70.1	12	ICGA	7
Abedi et al.	2013	Australia	Caucasian	206	IVR / IVB	78	12	VA(ETDRS)	6
Lotery et al.	2013	UK	Caucasian	254	VEGF inhibition	77.7	12	TRT	6
Kitchens et al.	2013	America	Caucasian	100	IVR / IVB	80	4	OCT	7
Hautamaki et al.	2013	Finland	Caucasian	96	IVB	78	3.5	OCT	6
Hagstrom et al.	2013	America	Caucasian	834	IVR / IVB	78.5	12	VA(ETDRS)	7
Fang et al.	2013	China	East Asian	143	IVB	68.8	6	VA(ETDRS)	6
Park et al.	2014	Korea	East Asian	273	IVR	69.5	5	VA(ETDRS)	7
Matsumiya et al.	2014	Japan	East Asian	120	IVR	76	3	OCT	8
van Asten et al.	2014	Netherland*	Caucasian	391	IVR	80	3	VA(ETDRS)	6
Kuroda et al.	2015	Japan	East Asian	236	IVR	74.3	12	OCT	8
Hata et al.	2015	Japan	East Asian	77	IVR	75.8	24	VA(ETDRS)	7
Chaudhary et al.	2016	Canada	Caucasian	70	IVR	80.8	6	VA(ETDRS)	7
Bardak et al.	2016	Turkey	Middle East	39	IVR	NA	3	OCT	8
Kikushima et al.	2017	Japan	East Asian	140	IVA	75	12	OCT	6
Yamashiro et al.	2017	Japan	East Asian	426	IVA	73.6	12	OCT	8
Park et al.	2017	Korea	East Asian	95	IVA	67.6	12	VA(ETDRS)	7
Mohamad et al.	2019	Malaysia	East Asian	145	IVA	69.1	6	VA(Snellen)	8

Table 2. The main results of all studies included in our meta-analysis.

Table 3. The main results of pooled ORs and analysis of *ARMS2* gene polymorphism with advanced AMD in our meta-analysis.

Refs(year).	Design	Definition of a positive response	Number of cases with good response					Total number of cases				
			G	GG	GT	TT	T	G	GG	GT	TT	T
Brantley et al (2007)	Retrospective	Improved visual acuity	47	15	17	6	29	104	33	38	15	68
Orlin et al (2012)	Retrospective	Improved/unchanged visual acuity	107	35	37	21	79	173	55	63	31	125
Yamashiro et al (2012)	Retrospective	Retinal exudate resolved by OCT	38	9	20	18	56	61	16	29	32	93
Park et al (2012)	Retrospective	Complete polyp regression	39	12	15	11	37	42	12	18	21	60
Abedi et al (2013)	Prospective	<15 letters of vision loss	NA	146		33	NA	NA	162		44	NA
Lotery et al (2013)	Prospective	≥75th percentile in TRT change	126	NA	NA	NA	126	280	NA	NA	NA	228
Kitchens et al (2013)	Retrospective	No retinal fluid at the fourth month	110	38	34	7	48	129	45	39	16	71
Hautamaki et al (2013)	Prospective	Retinal exudate resolved by OCT	47	10	27	7	41	98	21	56	19	94
Hagstrom et al (2013)	Prospective	Gain of 15 or more letters	280	77	126	48	222	930	266	398	170	738
Fang et al (2013)	Prospective	Gain of 15 or more letters	33	6	21	19	59	76	16	44	83	210
Park et al (2014)	Prospective	Gain of 8 or more letters	96	19	58	59	176	181	36	109	128	365
Matsumiya et al (2014)	Retrospective	No retinal fluid or edema	84	23	38	35	108	103	27	49	44	137
van Asten et al (2014)	Prospective	<30% letters of visual acuity loss	374	115	144	85	314	420	127	166	98	362
Kuroda et al (2015)	Retrospective	No retinal hemorrhage or fluid	47	11	25	27	79	132	33	66	76	218
Hata et al (2015)	Prospective	Improved/unchanged visual acuity	39	10	19	25	69	52	10	32	35	102
Chaudhary et al (2016)	Prospective	Gain of 15 or more letters	30	11	8	4	16	84	27	30	13	56
Bardak et al (2016)	Prospective	Absence of retinal fluid	9	3	3	5	13	34	10	14	15	44
Kikushima et al (2017)	Retrospective	Retinal fluid resolved by OCT	50	15	20	11	42	110	26	58	56	170
Yamashiro et al(2017)	Prospective	absence of intra- or sub-retinal fluid	136	42	52	36	124	361	90	181	155	491
Park et al(2017)	Prospective	<5 letter of visual acuity loss	21	4	13	8	29	57	11	35	34	103
Mohamad et al(2019)	Prospective	improvement of three lines or greater	54	16	22	16	54	99	21	57	67	191

Polymorphism (rs10490924)	Ethnicity	Number of studies	Test of heterogeneity			Test of association		
			I^2(%)	P_H	Model	OR	95%CI	P for test
G vs T(allelic)	Overall	20	69.1	<0.001	R	1.38	(1.13, 1.69)	0.002
	Caucasian	8	55.5	0.028	R	1.10	(0.86, 1.40)	0.455
	East Asian	11	61.1	0.004	R	1.67	(1.29, 2.16)	<0.001
	Middle East	1	NA	NA	NA	0.86	(0.32, 2.33)	0.765
GG vs TT(homozygote)	Overall	19	53.6	0.003	R	1.88	(1.32, 2.67)	<0.001
	Caucasian	7	31.9	0.184	F	1.25	(0.92, 1.70)	0.147
	East Asian	11	55.1	0.014	R	2.38	(1.44, 3.93)	0.001
	Middle East	1	NA	NA	NA	0.86	(0.15, 4.82)	0.861
GT vs TT(heterozygote)	Overall	19	29.2	0.114	F	1.36	(1.14, 1.62)	0.001
	Caucasian	7	42.8	0.106	F	1.20	(0.91, 1.60)	0.201
	East Asian	11	19.2	0.260	F	1.50	(1.19, 1.88)	<0.001
	Middle East	1	NA	NA	NA	0.55	(0.10, 2.89)	0.476
GG+ GT vs TT(dominant)	Overall	20	46.7	0.012	R	1.62	(1.27, 2.06)	<0.001
	Caucasian	8	54.5	0.031	R	1.52	(0.98, 2.35)	0.063
	East Asian	11	40.6	0.078	R	1.76	(1.31, 2.36)	<0.001
	Middle East	1	NA	NA	NA	0.67	(0.16, 2.75)	0.575
GG vs GT+TT(recessive)	Overall	19	47.3	0.012	R	1.51	(1.15, 2.00)	0.004
	Caucasian	7	0.0	0.718	F	1.09	(0.87, 1.38)	0.460
	East Asian	11	53.3	0.018	R	1.90	(1.21, 2.98)	0.005
	Middle East	1	NA	NA	NA	1.13	(0.23, 5.45)	0.884

Table 4. Meta-regression for the heterogeneity of G vs T comparison in our meta-analysis.

Covariate factors	Exp(b)	Std. Err.	P value	95%CI
design	1.33	0.25	0.052	0.99-1.78
ethnicity	2.52	0.75	0.006	1.35-4.71
follow up (months)	1.74	0.60	0.130	0.84-3.60
intervention	1.02	0.33	0.944	0.52-2.01
mean age (years)	4.14	5.75	0.319	0.22-76.4
number of case	2.34	0.71	0.011	1.24-4.41

Table 5. Bias between *ARMS2* genetic polymorphism with advanced AMD in our meta-analysis

Polymorphism (rs10490924)	Number of publication	Publication bias (P value)	
		Begg's test	Egger's test
G vs T	20	0.206	0.092
GG vs TT	19	0.142	0.125
GT vs TT	19	0.889	0.455
GG+ GT vs TT	20	0.456	0.240
GG vs GT+TT	19	0.142	0.075

Figures

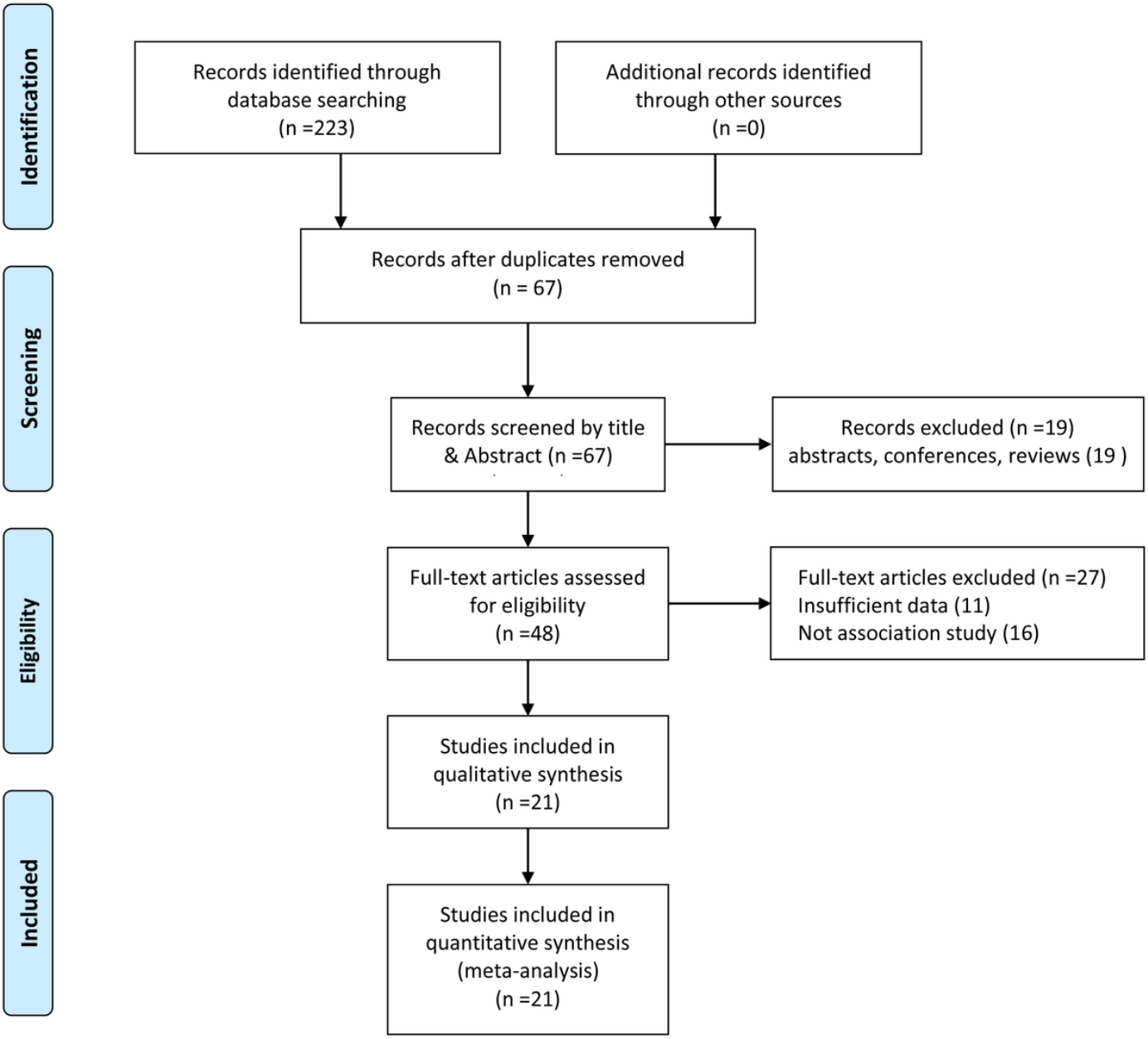

Figure 1

Flow diagram presenting the result of literature searching process in meta-analysis.

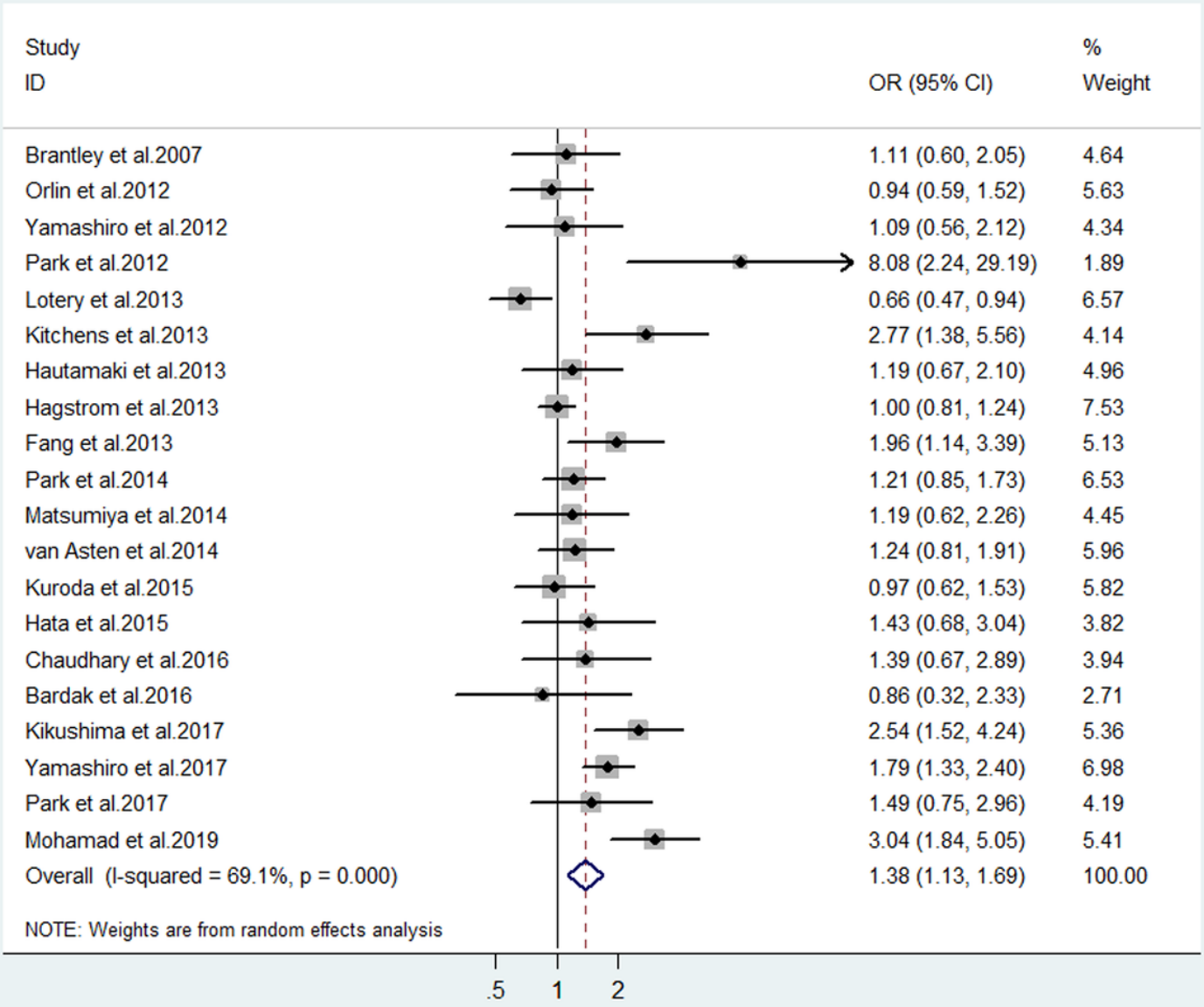

Figure 2

Evaluation of the association between ARMS2 gene polymorphism (G VS T) with advanced AMD.

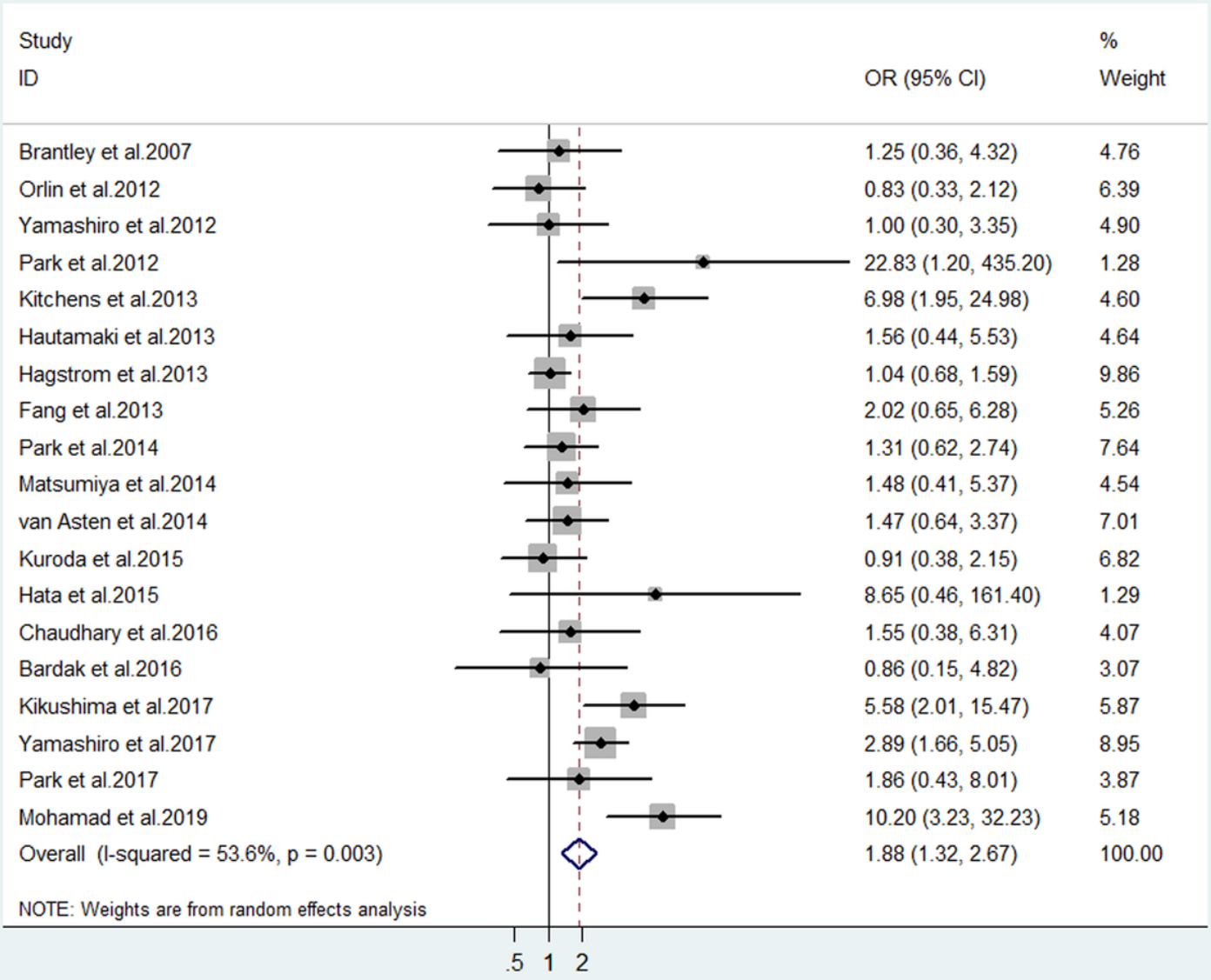

Figure 3

Assessment of the association between ARMS2 gene polymorphism (GG VS TT) with advanced AMD.

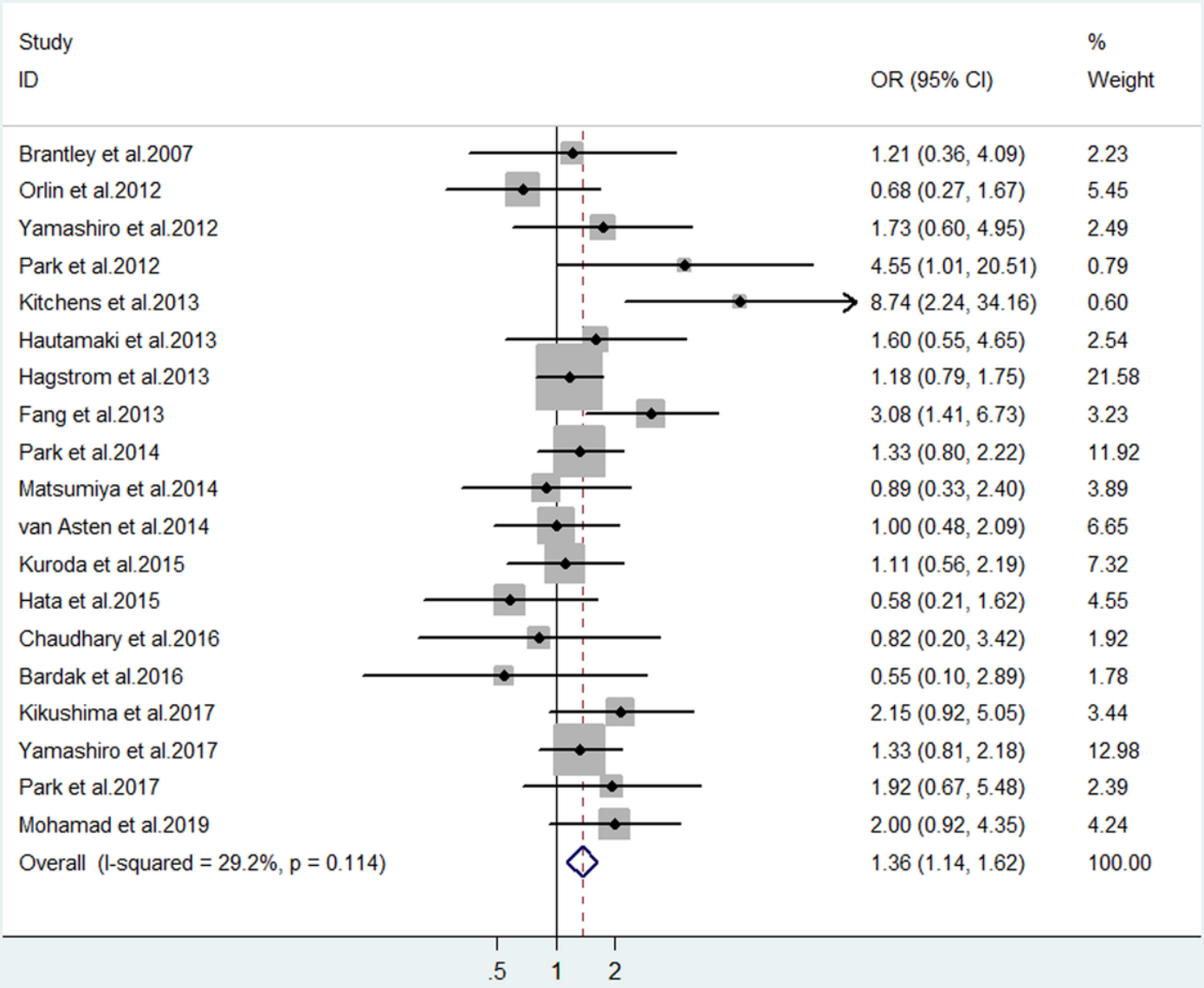

Figure 4

Estimation of the association between ARMS2 gene polymorphism (GT VS TT) with advanced AMD.

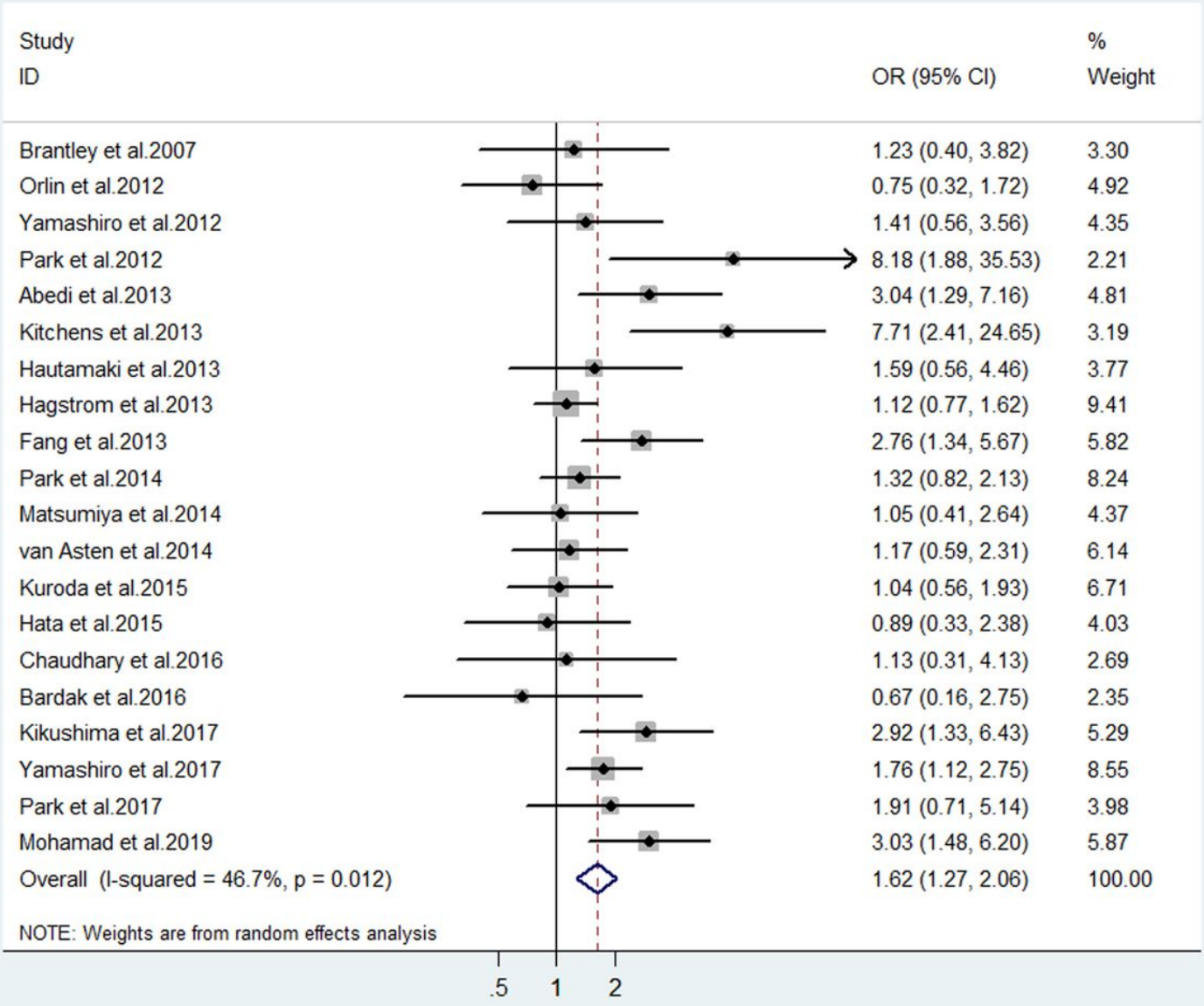

Figure 5

Evaluation of the association between ARMS2 gene polymorphism (GG+GT VS TT) with advanced AMD.

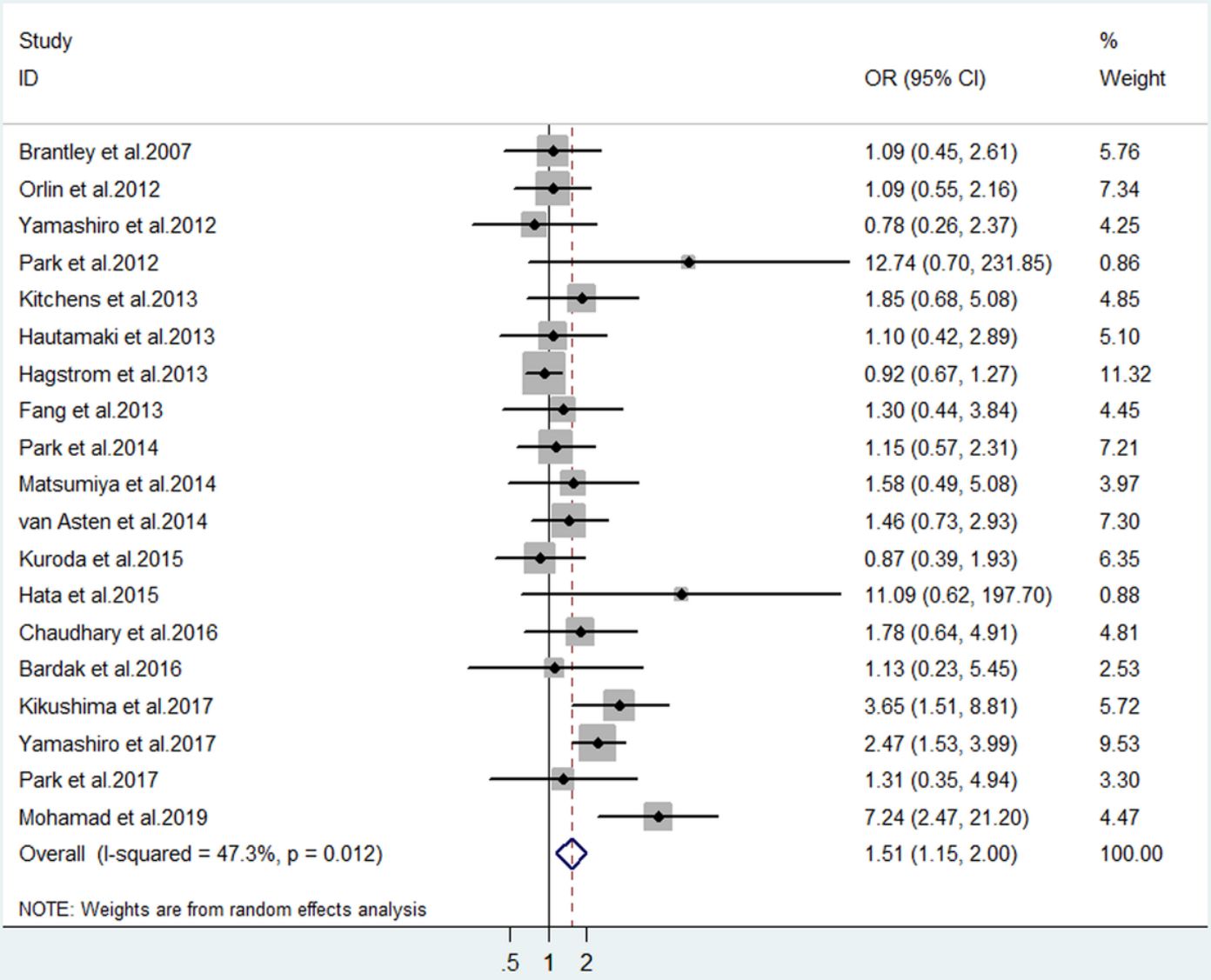

Figure 6

Assessment of the association between ARMS2 gene polymorphism (GG VS GT+TT) with advanced AMD.

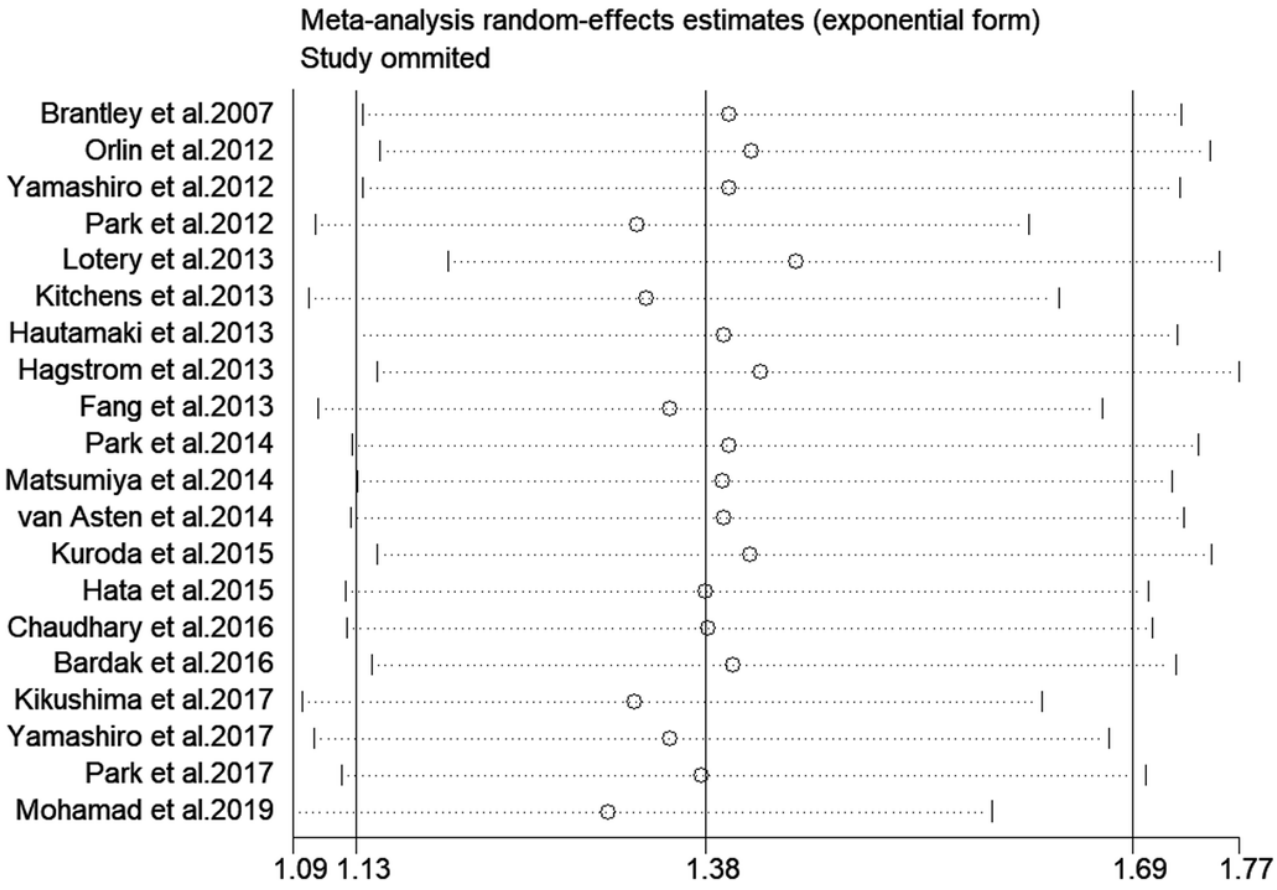

Figure 7

Assessment of the sensitivity analysis between ARMS2 gene polymorphism (G VS T) with advanced AMD.

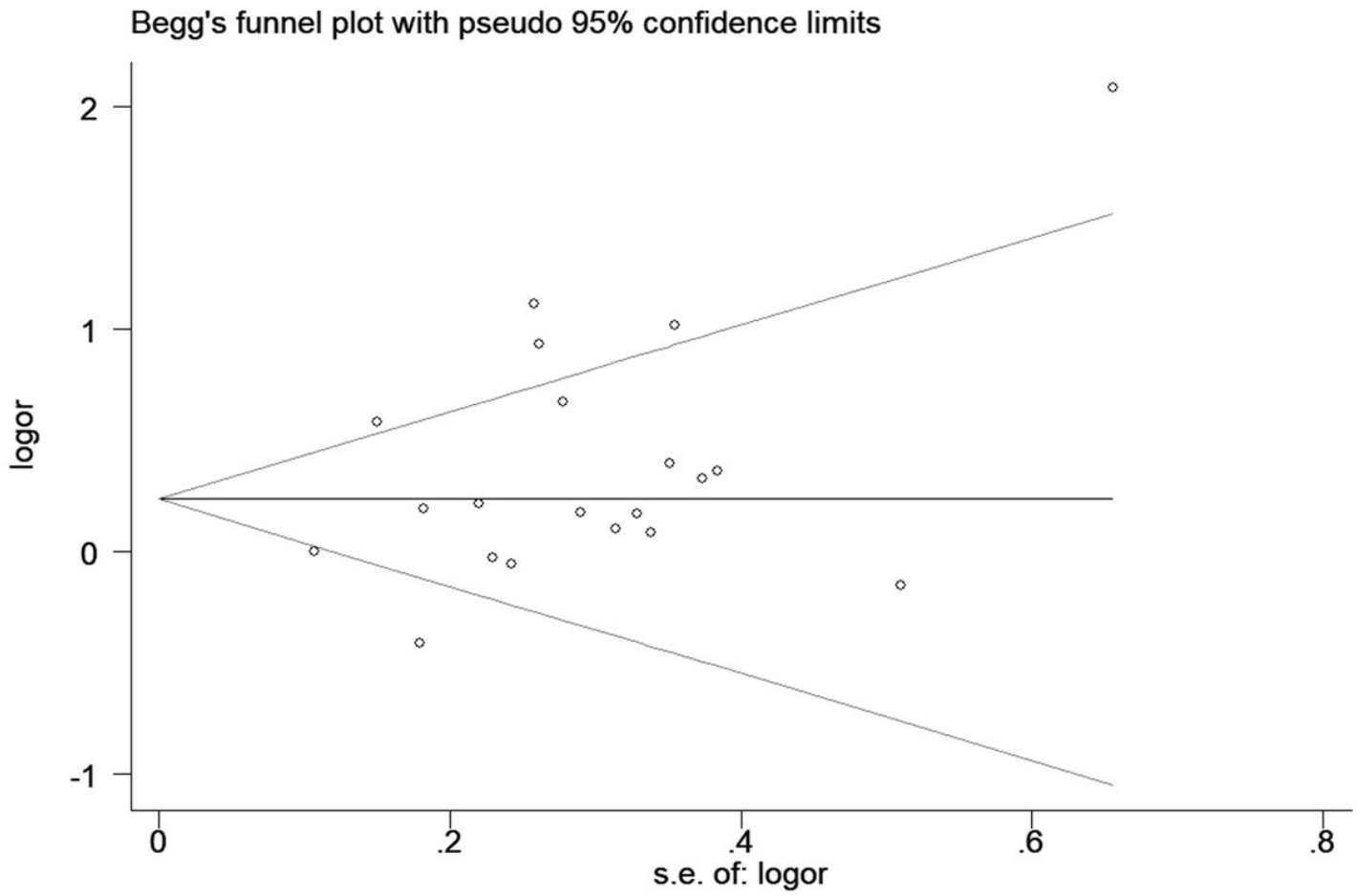

Figure 8

Estimation of the publication bias between ARMS2 gene polymorphism (G VS T) with advanced AMD.