The Relationship Between Hepatic Lipase Gene Variant and Advanced Age-Related Macular Degeneration
A Meta-analysis

Li-Xia Lou, MD; Kai-Min Hu, MD; Kai Jin, MD; Su-Zhan Zhang, MD, PhD; Juan Ye, MD, PhD

IMPORTANCE To date, no consistency exists across studies that have evaluated the relationship between hepatic lipase gene (LIPC) rs10468017 variant and advanced age-related macular degeneration (AMD).

OBJECTIVE To summarize all relevant evidence for a relationship between LIPC variant and advanced AMD.

DATA SOURCES The PubMed and Embase databases were searched for studies potentially eligible in any language published up to September 15, 2013.

STUDY SELECTION Case-control studies of 2 or more comparison groups that included patients with advanced AMD (choroidal neovascularization or geographic atrophy).

DATA EXTRACTION AND SYNTHESIS Allele frequencies and genotype distributions of rs10468017 variant.

MAIN OUTCOMES AND MEASURES Summary odds ratios (ORs) and 95% CIs were estimated under different genetic models using meta-analytic methods. A stratified analysis by advanced AMD subtypes and race/ethnicity was performed, as well as a sensitivity analysis.

RESULTS Data from 10 case-control studies were included in the meta-analysis. The rs10468017 variant (C→T) showed significant summary ORs of 0.81 (95% CI, 0.75-0.88), 0.83 (95% CI, 0.70-0.98), and 0.60 (95% CI, 0.44-0.81) under the allelic (T vs C), heterozygous (TC vs CC), and homozygous (TT vs CC) models, respectively. Carrying at least 1 copy of the T allele decreased the risk of choroidal neovascularization and geographic atrophy by 20% (OR, 0.80; 95% CI, 0.74-0.87) and 29% (OR, 0.71; 95% CI, 0.59-0.86), respectively. The pooled OR for white race/ethnicity under an allelic model was 0.80 (95% CI, 0.74-0.87). The sensitivity analysis indicated the robustness of our findings, and no evidence of publication bias was observed in our meta-analysis.

CONCLUSIONS AND RELEVANCE Our meta-analysis indicates that LIPC rs10468017 variant is associated with a reduced risk of advanced AMD. This finding may lead to insights regarding the pathogenesis, prevention, and treatment of AMD.
Age-related macular degeneration (AMD) is a progressive disease of the central retina and a leading cause of irreversible blindness for older individuals worldwide. Most vision loss occurs when AMD progresses to 1 of 2 advanced forms, namely, choroidal neovascularization (CNV [wet AMD]) and geographic atrophy (GA [dry AMD]). It has been estimated that cases of advanced AMD will increase from 1.7 million in 2010 to 3.8 million in 2050 based on the findings of a 2009 study. Age-related macular degeneration is a multifactorial disease that involves complex interactions between environmental and genetic factors. Several environmental risk factors have been identified, including cigarette smoking, higher body mass index, and insufficient dietary antioxidants. Compelling evidence also exists for the contribution of genetic factors to the incidence and progression of AMD. Multiple genetic variants have been reported to be linked to AMD, including CFH Y402H, CFB L9H, C2 E318D, ARMS2 A69S, and HTRA1 G625A. These genetic loci account for only approximately half of the heritability of AMD.

In an attempt to identify other susceptibility loci, a genome-wide association study (GWAS) revealed a significant relationship between advanced AMD and the hepatic lipase gene (LIPC) (OMIM 011465), located on chromosome 15q21. LIPC encodes hepatic triglyceride lipase in the liver and affects high-density lipoprotein cholesterol (HDL-C) levels. Several studies have investigated the relationship between single-nucleotide polymorphisms (SNPs) in the LIPC gene and advanced AMD. The SNP rs10468017 variant (C→T), which is located 514 kilobases upstream of the LIPC gene, is the focus of research interest. However, the results from previous studies are inconsistent, with reports of a significant protective effect of the T allele and reports of no significant relationship. Therefore, we performed a meta-analysis to summarize all relevant evidence for a relationship between LIPC rs10468017 and advanced AMD (CNV or GA) to help us better understand the effect of LIPC on advanced AMD.

Methods

Search Strategy

A comprehensive literature search of PubMed and Embase databases (up to September 15, 2013) was conducted to identify relevant studies. The search strategy comprised the terms macular degeneration, retinal degeneration, retinal neovascularization, choroidal neovascularization, retinal drusen, geographic atrophy, hepatic lipase, and LIPC (more details on the search strategy are available in the eAppendix in the Supplement). No language restrictions were imposed. The titles and abstracts were scanned to exclude any clearly irrelevant studies. The full texts of the remaining articles were read to determine whether they contained information on the topic of interest. To find additional references, we also checked the reference lists of the retrieved publications.

Inclusion Criteria and Data Extraction

Two of us (L.-X.L. and K.-M.H.) independently assessed the retrieved studies and extracted data from each included study. Any inconsistencies were resolved through discussion. Studies included in the meta-analysis had to fulfill the following criteria: (1) they used a case-control design, (2) advanced AMD was diagnosed using at least 1 eligible method, (3) they reported a measure of the relationship between LIPC and advanced AMD as an odds ratio (OR) with a 95% CI or allowed for calculation of it from raw data contained in the article, and (4) they contained at least 2 comparison groups (a control group and a case group that included patients with advanced AMD [CNV or GA]).

In addition to the first author name and the year of publication, the following information was extracted from each study: study design, sample size, phenotype of the cases evaluated, the mean age and race/ethnicity of the participants, the methods for genotyping and the genotype distributions in cases and controls, and P values for Hardy-Weinberg equilibrium in controls. We contacted the authors of retrieved articles if additional data were needed.

Statistical Analysis

The strength of the relationship between LIPC rs10468017 and advanced AMD was estimated as ORs (95% CIs) under 3 genetic models, namely, an allelic model, a heterozygous model, and a homozygous model. Study-specific ORs were pooled using fixed-effects models with the Mantel-Haenszel method when heterogeneity was negligible or using random-effects models with the DerSimonian-Laird method when heterogeneity was significant. Between-study heterogeneity was assessed using the Cochran Q test and the I² statistic. P < .10 was considered statistically significant for the Cochran Q test. I² ranges between 0% and 100% (0% represents no heterogeneity), and higher values represent increasing heterogeneity. We conducted a stratified analysis by advanced AMD subtypes and race/ethnicity. A sensitivity analysis, removing each study one at a time, was performed to confirm the stability of our findings. Publication bias was assessed by the Begg test and the Egger test. The extent of publication bias was also shown with a funnel plot. All statistical analyses were performed using commercially available software (STATA 12.0; StataCorp LP). Except for heterogeneity, P < .05 was considered statistically significant, and all tests were 2-sided.

Results

Search Findings and Study Characteristics

The initial search strategy identified 43 studies from the databases (21 from PubMed and 22 from Embase). After the removal of 19 duplicate publications, 24 studies were considered of potential relevance (Figure 1). In total, 18 articles were retrieved for full-text review, 10 of which met our inclusion criteria. Characteristics of studies included in the meta-analysis are summarized in the Table. Genotype distributions for rs10468017 variant from individual studies are listed in eTable 1 in the Supplement. All studies had a case-control design; 4 of them were a GWAS. Eight studies were performed in participants of white race/ethnicity, and 2 studies were conducted among Asians.
Results of the Meta-analysis

We initially performed a meta-analysis of the relationship between LIPC variant and advanced AMD. The SNP rs10468017 showed a significant summary OR under an allelic model of T vs C (OR, 0.81; 95% CI 0.75-0.88) (Figure 2). The Cochran Q test indicated moderate but significant between-study heterogeneity across the studies ($I^2 = 49.8\%$, $P = .05$). We also found a significant relationship under a heterozygous model of TC vs CC (OR, 0.83; 95% CI, 0.70-0.98) and under a homozygous model of TT vs CC (OR, 0.60; 95% CI, 0.44-0.81), with no evidence of heterogeneity (eFigure 1 and eFigure 2 in the Supplement, respectively).

Stratified Analysis by Advanced AMD Subtypes and Race/Ethnicity

In the stratified analysis by advanced AMD subtypes, the summary ORs of rs10468017 under an allelic model were 0.80 (95% CI, 0.74-0.87) and 0.71 (95% CI, 0.59-0.86) for CNV and GA, respectively (Figure 3). Heterogeneity was not significant for either phenotype ($P = 2.2\%$, $P = .40$ for CNV and $P = 44.9\%$, $P = .12$ for GA). Next, we performed a stratified

<table>
<thead>
<tr>
<th>Source</th>
<th>Race/Ethnicity</th>
<th>Study Design</th>
<th>Phenotype of Cases</th>
<th>Odds Ratio (95% CI)</th>
<th>Case/Control Sample Size</th>
<th>Case/Control Mean Age, y</th>
<th>Genotyping Method</th>
<th>Minor Allele Frequency</th>
<th>Hardy-Weinberg Equilibrium</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lee et al, 2013</td>
<td>White</td>
<td>GWAS</td>
<td>Advanced AMD</td>
<td>0.87 (0.80-0.95)</td>
<td>3776/2009</td>
<td>79.31/72.57</td>
<td>ShAphot Multiplex Kit; Life Technologies</td>
<td>0.291 Yes</td>
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<tr>
<td>Peter et al, 2011</td>
<td>White</td>
<td>Case-control</td>
<td>Advanced AMD</td>
<td>0.57 (0.33-0.96)</td>
<td>48/1234</td>
<td>74.5/73.6</td>
<td>TaqMan; Applied Biosystems</td>
<td>0.276 Yes</td>
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<tr>
<td>Cipriani et al, 2012</td>
<td>White</td>
<td>GWAS</td>
<td>Advanced AMD</td>
<td>0.91 (0.80-1.03)</td>
<td>893/2199</td>
<td>78.6/44-45</td>
<td>BeadArray; Illumina</td>
<td>0.270 Yes</td>
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<td>Neale et al, 2010</td>
<td>White</td>
<td>GWAS</td>
<td>Advanced AMD</td>
<td>0.82 (0.77-0.88)</td>
<td>6768/5943</td>
<td>80-81/76</td>
<td>iPLEX; Sequenom and SNP GeneChip; Affymetrix</td>
<td>0.300 Yes</td>
<td></td>
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<tr>
<td>Zhang et al, 2013</td>
<td>Asian</td>
<td>Case-control</td>
<td>CNV</td>
<td>0.75 (0.51-1.10)</td>
<td>156/212</td>
<td>67/69</td>
<td>ShAphot Multiplex Kit; Life Technologies</td>
<td>0.191 Yes</td>
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<td>Reynolds et al, 2010</td>
<td>White</td>
<td>Case-control</td>
<td>Advanced AMD</td>
<td>0.61 (0.45-0.81)</td>
<td>318/140</td>
<td>81/76</td>
<td>MALDI-TOF MS</td>
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<tr>
<td>Yu et al, 2011</td>
<td>White</td>
<td>Case-control</td>
<td>Advanced AMD</td>
<td>0.68 (0.55-0.85)</td>
<td>1067/218</td>
<td>79.5-80.7/77</td>
<td>TaqMan; Applied Biosystems</td>
<td>0.337 Yes</td>
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<td>Tian et al, 2012</td>
<td>Asian</td>
<td>Case-control</td>
<td>Advanced AMD</td>
<td>0.93 (0.73-1.19)</td>
<td>467/462</td>
<td>&gt;50/50</td>
<td>MassArray; Sequenom</td>
<td>0.169 Yes</td>
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<tr>
<td>Sobrin et al, 2012</td>
<td>White</td>
<td>GWAS</td>
<td>CNV GA</td>
<td>0.83 (0.75-0.91)</td>
<td>7977/19374</td>
<td>&gt;60/60</td>
<td>iPLEX; Sequenom and SNP GeneChip; Affymetrix</td>
<td>Not available</td>
<td></td>
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<tr>
<td>Seddon et al, 2010</td>
<td>White</td>
<td>Case-control</td>
<td>Advanced AMD</td>
<td>0.75 (0.60-0.94)</td>
<td>545/275</td>
<td>Not available/50</td>
<td>MALDI-TOF MS</td>
<td>0.315 Not available</td>
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</table>

Abbreviations: AMD, age-related macular degeneration; CNV, choroidal neovascularization; GA, geographic atrophy; GWAS, genome-wide association study; MALDI-TOF MS, matrix-assisted laser desorption ionization time-of-flight mass spectrometry; NA, not applicable; SNP, single-nucleotide polymorphisms.

* Minor allele frequency in controls.

* Hardy-Weinberg equilibrium in controls.

* Data from discovery sample of GWAS.

* Main results under an allelic model.
analysis by race/ethnicity. This analysis was limited to individuals of white race/ethnicity because of the few studies available for Asians. The pooled OR for white race/ethnicity under an allelic model was 0.80 (95% CI, 0.74-0.87), with moderate heterogeneity ($I^2 = 54.2\%$, $P = .04$).

**Sensitivity Analysis**

To evaluate the robustness of the relationship between rs10468017 and advanced AMD, we performed a sensitivity analysis by removing each study one at a time and recalculating the summary OR. The pooled OR remained stable under an allelic model, indicating that our results were not influenced by any single study (eTable 2 in the Supplement). This analysis also revealed that 2 studies, by Reynolds et al and by Yu et al, were the main source of heterogeneity. The $I^2$ statistics for the ORs decreased from 49.8% to 39.7% ($P = .13$) after removing the study by Reynolds et al and to 43.1% ($P = .10$) after removing the study by Yu et al. Low heterogeneity was achieved after removing both studies ($I^2 = 20.7\%$, $P = .28$), yielding a summary OR of 0.85 (95% CI, 0.80-0.90). The sensitivity analysis also suggested that the significant relationships between rs10468017 and advanced AMD were robust under a heterozygous model and a homozygous model (data not shown).

**Publication Bias**

We assessed possible publication bias with a funnel plot. Except for the study by Peter et al, all studies were found near the apex of the funnel plot because of the much smaller SEs for ORs under an allelic model (eFigure 3 in the Supplement). Neither the Begg test ($P = .27$) nor the Egger test ($P = .17$) suggested publication bias. Also, no publication bias was detected under a heterozygous model ($P = .13$ for the Begg test and $P = .10$ for the Egger test) or under a homozygous model ($P = .71$ for the Begg test and $P = .24$ for the Egger test).

**Discussion**

Our meta-analysis showed an allelic summary OR of 0.81 (95% CI, 0.75-0.88) and strongly supported the notion that rs10468017 variant is a protective factor for advanced AMD.
In individuals carrying at least 1 copy of the protective T allele, the risk of advanced AMD decreased by approximately 20%. The pooled ORs were 0.83 and 0.60 under the heterozygous and homozygous models, respectively, suggesting that increased copy number of the T allele lowered the risk of advanced AMD.

Heterogeneity should be considered in meta-analysis interpretation.26 Our analysis based on the allelic model indicated significant between-study heterogeneity, with an $I^2$ statistic of 49.8% ($P = .05$). Compared with noncarriers, the stratified analysis revealed that carriers having at least 1 copy of the protective T allele had approximately 20% (OR, 0.80; 95% CI, 0.74-0.87) and 29% (OR, 0.71; 95% CI, 0.59-0.86) decreased risks of CNV and GA, respectively. A large proportion of the studies were conducted in participants of white race/ethnicity. After exclusion of a study77 conducted among Asians, the summary OR remained stable. The sensitivity analysis indicated that the studies by Reynolds et al23 and by Yu et al27 were the main source of heterogeneity. After excluding these 2 studies, heterogeneity was significantly decreased ($I^2 = 20.7\%$). The summary OR was essentially unchanged in the sensitivity analysis, supporting the reliability of the pooled results.

A strength of our meta-analysis was the large sample size in most of the included studies, especially inclusion of 4 studies of large-scale GWAS design. A sufficient sample size is critical in genetic association studies to achieve adequate statistical power.27 Several limitations of our meta-analysis should be noted. The analysis was based primarily on unadjusted ORs, without controlling for confounding factors. We cannot address the gene–gene and gene–environment interactions because the original literature provided insufficient information. In our meta-analysis, 5 studies22,23,25 included participants drawn from the same populations (eg, the Age-Related Eye Disease Study3), which might have affected the pooled results. However, the sensitivity analysis revealed that these studies did not substantially alter the summary OR or underestimate heterogeneity (data not shown).

Until 2010, all genetic variants associated with AMD were identified from candidate gene testing based on the complement factor pathway.28 Since 2010, several large-scale investigations of GWAS design have identified genes that had not been previously known.12,29,30 LIPC was first reported to be associated with advanced AMD by Neale et al.12 This finding was replicated in a GWAS15 but was contradicted by another GWAS.14 The GWAS by Yu et al15 was not included in our meta-analysis because the OR under an allelic model was unavailable; however, the OR under an additive genetic model (0, 1, or 2 minor alleles) was 0.84 ($P = 2.7 \times 10^{-12}$) for rs10468017. Studies focusing on the effects of LIPC on progression to advanced AMD revealed that the T allele of rs10468017 was associated with decreased risk of progression to CNV or GA in the same eye22,33 but had no effects on progression to advanced AMD in the second eye.34

The LIPC gene, which encodes hepatic triglyceride lipase, has an important role in HDL-C metabolism.35 High-density lipoprotein cholesterol transports most of the cholesterol, lutein, and zeaxanthin within the retinal pigment epithelium and Bruch membrane.12 Inefficiency of cholesterol and carotenoid delivery could lead to deposits, drusen, and stress on the retinal pigment epithelium, which may lead to AMD.36,37 However, HDL-C–decreasing alleles of ABCA1 and CETP,22 as well as HDL-C–increasing alleles of LIPC,14 are associated with a reduced risk of advanced AMD. The study by Reynolds et al23 also revealed that the HDL-C level did not seem to modify the effect of LIPC on AMD, suggesting that, although LIPC regulates the HDL-C level, this may not be the direct mechanism by which LIPC reduces the risk of AMD, and indicating that alternative pathways may have a role. Hepatic lipase has been shown to have an effect on atherogenesis,38 and cardiovascular risk factors are related to AMD.39 The vascular intimate in atherosclerosis and Bruch membrane in macular degeneration may represent parallel responses to tissue injury induced by genetic variation, impaired immune responses, and oxidative stress.39 Further research into the mechanisms of LIPC and lipoproteins in the pathogenesis of AMD is needed.

Conclusions

To date, this is the first systematic meta-analysis evaluating the relationship between the LIPC gene variant and advanced AMD. Our analysis provides substantial evidence that LIPC rs10468017 is significantly associated with reduced risk of advanced AMD. This finding expands the number of confirmed AMD susceptibility loci. The LIPC locus may represent another pathway in the pathogenesis of AMD and could lead to insights regarding disease progression, ways to modify the risk of AMD, and novel strategies for AMD treatment.
A randomized, placebo-controlled, clinical trial of high-dose supplementation with vitamins C and E, 

dietary lutein, and zeaxanthin on plasma carotenoids and their 

transport in lipoproteins in age-related macular 


27. Hong EP, Park JW. Sample size and statistical 

calculations in gene association studies. 


28. Gorin MB. Genetic insights into age-related 

degeneration: controversies addressing 

risk, causality, and therapeutics. Mol Aspects Med. 


Complications of Age-Related Macular 

Degeneration Prevention Trial Research Group. 

Genetic variants near TIP39 and high-density 

lipoprotein-associated low density lipoprotein 

susceptibility to age-related macular degeneration. 


Consorium. Seven new loci associated with 

advanced age-related macular degeneration. 


doi:10.1038/ng.2758.


Common variants in the 

region of chromosome 6p21.3 associated with age-related macular 

degeneration: AREDS report No. 8. 


32. Seddon JM, Reynolds R, Rosner B. 

Dietary 

omega-3 fatty acids, other fat intake, genetic 

susceptibility, and progression to incident 

geographic atrophy. Ophthalmology. 2013;120(5): 

1020-1028.

33. Lechanteur YT, van de Ven JP, Smallhodzic D, 

et al. Genetic, behavioral, and sociodemographic 

risk factors for second eye progression in 

age-related macular degeneration. Invest 


34. Santamarina-fojo S, Gonzalez-navarro H, 

Freeman L, Wagner E, Nong Z. Hepatic lipase, 

lipoprotein metabolism, and atherogenesis. 


35. Hageman GS, Luthert PJ, Victor Chong NH, 

Johnson LV, Anderson DH, Mullins RF. An 

integrated hypothesis that considers drusen as 

biomarkers of immune-mediated processes at the 

RPE-Bruch’s membrane interface in aging and 

age-related macular degeneration. Prog Retin Eye Res. 


36. Wang W, Connor SL, Johnson EJ, Klein ML, 

Hughes S, Connor WE. Effect of dietary lutein and 

zeaxanthin on plasma carotenoids and their 

transport in lipoproteins in age-related macular 


Cardiovascular risk factors and the long-term 

incidence of age-related macular degeneration: the 

Blue Mountains Eye Study. Ophthalmology. 2007; 

114(6):1143-1150.

38. Sivaprasad S, Bailey TA, Chong VN. Bruch’s 

membrane and the vascular intima: is there a 

common basis for age-related changes and disease? 