TNFRSF10A-LOC389641 rs13278062 But Not REST-C4orf14-POLR2B-IGFBP7 rs1713985 Was Found Associated With Age-Related Macular Degeneration in a Chinese Population

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Purpose. To reassess the association between TNFRSF10-LOC389641 rs13278062 and REST-C4orf14-POLR2B-IGFBP7 rs1713985 with the risk of AMD in a Chinese case-control collection.

Methods. The primary study consisted of 1826 subjects, including 1226 controls, 300 cases with nAMD, and 300 cases with PCV. Genomic DNA was extracted from venous blood leukocytes. The allelic variants of rs13278062 and rs1713985 were determined by matrix-assisted laser desorption/ionization time-of-flight mass spectrometry. The difference in allele distribution between cases and controls was tested using a χ^2 test. We also performed a meta-analysis of case-control studies for rs13278062 and rs1713985 in Hong Kong and Singaporean late AMD collections of Chinese descent (1273 cases and 1652 controls) via an inverse-variance, fixed effects model as previously described. Subgroup analysis of CNV and PCV subtypes were also performed.

Results. We found no evidence to support a significant association of markers rs13278062 or rs1713985 with either nAMD or PCV, or total AMD in our Beijing study ($P > 0.05$ for all comparisons). Upon meta-analysis of all sample collections, we note nominally significant association between rs13278062 and increased risk of late AMD, consistent with previous findings in Japanese individuals (ORmeta = 1.17, Pmeta = 0.004). No association was detected between rs1713985 and AMD when all data were meta-analyzed.

Conclusions. SNP rs13278062, but not rs1713985 showed nominal evidence of association with AMD in a total of 1273 cases and 1652 controls of Chinese descent. The difference between different effect sizes in our study and other studies suggested that future studies with much larger sample sizes is necessary.

Keywords: neovascular age-related macular degeneration, polypoidal choroidal vasculopathy, TNFRSF10A-LOC389641, REST-C4orf14-POLR2B-IGFBP7, single nucleotide polymorphism

Age-related macular degeneration (AMD) is the most common cause of legal blindness in the elderly in developed countries. It results in irreversible central vision loss.[1] Late-stage AMD can be divided into two forms: geographic atrophy (dry AMD) and neovascular AMD (wet AMD, or AMD). The neovascular form of the disease is characterized by the development of choroidal neovascular (CNV) membranes that are the main cause of visual impairment in macular degeneration.[2] Polypoidal choroidal vasculopathy (PCV), showing choroidal vascular networks with polypoidal lesions at their borders, is associated with a reduction of vision in the elderly Asian population. PCV has been described as a distinct clinical entity from AMD and the other diseases associated with subretinal neovascularization.[3,4] Previous studies have revealed several genetic loci that are linked to the development of AMD and PCV. Nevertheless, whether PCV represents a subtype of neovascular AMD remains controversial. Evidence suggests that AMD and PCV, despite their different phenotypic manifestations, may share common genetic risk factors.[5–9]

Recently, data from Arakawa et al.[10] showed genome-wide significant evidence of association between two genetic loci (rs13278062 at TNFRSF10A-LOC389641 on chromosome 8p21 and rs1713985 at REST-C4orf14- POLR2B-IGFBP7) and risk of AMD in a Japanese AMD sample collection. The association between rs13278062 was confirmed by Nakata et al.[11] in case-control studies in a Japanese population involving 1687 cases and 3172 controls. However, the association between rs1713985 and late AMD could not be replicated. As it is well-documented that genotype-phenotype associations may vary in different populations, there is a definite place for further

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Table 1. Demographic Distribution of the Study Subjects

	nAMD-CNV, $n = 300$	PCV, $n = 300$	Controls, $n = 1866$
Females, *n* (%)	111 (37.0)	112 (37.3)	1226 (60.4)
Males, *n* (%)	189 (63.0)	188 (62.7)	486 (39.6)
Age* range, y	50–90	42–85	45–95
Mean age ± SD, y	69.4 ± 8.9	66.8 ± 9.7	60.0 ± 9.80

* Age of presentation.

investigation comprising attempts at replication of these associations in other Asian collections.

It is now increasingly recognized that individual studies could be underpowered to robustly confirm previously reported genotype-phenotype associations.[12,13] Thus, we systematically pooled the results of all available Chinese populations and carried out a meta-analysis to estimate the strength of the genetic association with AMD for a more accurate evaluation of the association.

Methods

Subjects

A total of 1826 unrelated Chinese subjects were studied in this case-control cohort; 300 patients had nAMD and 300 patients had PCV. A total of 1226 individuals without age-related maculopathy (ARM) were studied as controls. The sex and ages of the controls and cases are given in Table 1. The study participants were recruited at the Department of Ophthalmology in the Peking University People's Hospital, and the study was approved by the Ethical Committee of Peking University People's Hospital. An informed consent process was established following the guidelines of the Helsinki Declaration, and consent forms were signed by all subjects. All subjects received a comprehensive ophthalmic examination, including visual acuity measurements, slit-lamp biomicroscopy, and dilated fundus examination performed by a retinal specialist. All cases with nAMD and PCV underwent fluorescein angiography, optical coherence tomography (OCT), and indocyanine green angiograms with HRA2 (Heidelberg Engineering, Heidelberg, Germany). The diagnosis of nAMD or ARM was defined by the International Classification System for ARM.[14] The diagnosis of PCV was based on indocyanine green angiography (ICGA) results that showed a branching vascular network terminating in aneurismal enlargements. Exclusion criteria included any eye with any other macular abnormalities, such as pathologic myopia, idiopathic CNV, presumed ocular histoplasmosis, angioid streaks, and any other secondary CNV. Healthy controls were defined as having no clinical evidence of nAMD or PCV in either eye or any other eye diseases, excluding mild age-related cataracts. Subjects with severe cataracts were excluded from the study.

Genetic Analysis

Blood samples were collected from all participants and stored at −80°C before DNA extraction. Genomic DNA was extracted from venous blood leukocytes using a genomic extraction kit (Beijing eBios Biotechnology Co., Ltd., Beijing, China), and genotyping was performed by matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF-MS), as previously described.[15] Briefly, approximately 30 ng of genomic DNA was used to genotype each sample. The primer sequences for rs13278062 were 5′-ACGTTGGATGGCCTCTAAGAGGCTTTTACG-3′ and 5′-ACGTTGGATGAAAAAAGGCAGGCTGAATC-3′, and the primer sequences for rs1713985 were 5′-ACGTTGGATGCCTTCTTAGCTTCTCAGGTC-3′ and 5′-ACGTTGGATGAGTCAGTCCTCAAAAGCTCG-3′. The DNA samples were amplified, and the PCR products were used for locus-specific single-base extension reactions. The resulting products were desalted and transferred to a 384 SpectroCHIP array (Sequenom, San Diego, CA). Allele detection was performed using MALDI-TOF-MS. The mass spectrograms were analyzed using MassARRAY Typer software version 4.0 (Sequenom).

Statistical Analysis

The data were analyzed using SPSS (version 16.0; SPSS Science, Chicago, IL). All of the identified polymorphisms were assessed for Hardy-Weinberg equilibrium using χ^2 analysis. Single-marker association analyses were performed using logistic regression incorporating an additive genetic model as previously described.[11] A logistic regression model was used to calculate the odds ratio (OR) and 95% confidence interval (CI) of nAMD or PCV, comparing case groups to the control group as the reference and adjusted for age, gender, different genotypes, and various genetic models. Statistical analyses comparing baseline demographic factors were performed with Student's *t*-test. Values are expressed as means ± SD. Values of *P* less than 0.05 were considered statistically significant for the purpose of this replication study.[16]

Meta-analysis was carried out as described previously.[17] The OR with 95% CI was calculated between rs13278062 and rs1713985 and AMD risk. Both fixed-effects (the Mantel-Haenszel method) and random-effects (Der Simonian and Laird

Table 2. The rs13278062 and rs1713985 Genotypes and Allele Frequencies Distribution and the Results of Association Tests

	Genotype						Association Analysis			
	Cases			Controls			Nomina		Adjusted	
	GG	GT	TT	GG	GT	TT	*P* Value	OR (95% CI)	*P* Value	OR (95% CI)
rs13278062										
Total	256	274	70	583	504	139	0.115	1.13 (0.97–1.30)	0.108	1.14 (0.97–1.34)
nAMD	125	139	36				0.12	1.13 (0.97–1.30)	0.12	1.18 (0.96–1.44)
PCV	131	135	34				0.36	1.09 (0.90–1.32)	0.44	1.08 (0.89–1.32)
rs1713985										
Total	37	241	322	95	470	661	0.67	1.04 (0.87–1.21)	0.68	1.04 (0.87–1.23)
nAMD	18	118	164				0.54	1.07 (0.87–1.31)	0.49	1.08 (0.87–1.35)
PCV	19	123	158				0.97	1.00 (0.82–1.23)	0.90	1.01 (0.82–1.25)

† $P < 0.05$ was considered significant.

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study	case/control	MAF (case)	MAF (control)	p value	OR	LI	UI
Beijing	600/1226	0.35	0.32	0.108	1.14	0.97	1.34
Hongkong	433/275	0.31	0.27	0.110	1.22	0.96	1.56
Singapore	240/151	0.32	0.25	0.051	1.38	1.00	1.96
Summary-Chinese	1273/1652	0.33	0.31	0.004	1.17	1.05	1.32
Isao Nakata-Kyoto	1353/3029	0.39	0.34	0.040	1.19	1.01	1.41
Isao Nakata-Yamanashi	323/115	0.39	0.30	0.012	1.54	1.10	2.13
Arakawa-GWAS	827/3323	0.42	0.34	2.46E-06	1.41	1.22	1.61
Arakawa-Replication	701/15565	0.42	0.35	8.19E-08	1.35	1.22	1.52
Overall (I-Squared=11.7%,P=0.340)							

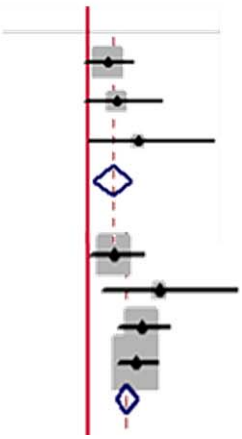

Figure 1. Meta-analysis of rs13278062 and AMD Forest plot for meta-analysis of association between R102G and AMD risk. Each study is shown by the point estimate of the OR (the size of the square is proportional to the weight of each study) and 95% CI for the OR (*extending lines*). Data from all studies included in the Chinese cohort suggested that the T allele of rs13278062 conferred risk for AMD (OR = 1.07, 95% CI: 0.86–1.33, Fig. 2). We found no evidence of heterogeneity in this evaluation ($I^2 = 0.0\%$, $P = 0.44$) and the overall (Japanese data included) ($I^2 = 11.7\%$, $P = 0.34$).

method) models were fitted. Cochran's Q statistic and the accompanying I^2 index was used to assess intercohort heterogeneity. When the effects were assumed to be homogeneous (P value for heterogeneity > 0.05), we selected the fixed-effects model, or else we chose the random-effects model. Forest plots were used to describe the results from separate studies and the summary results as well. We used Egger's tests to test for potential publication bias, and if P was less than 0.05, the publication bias was considered to be statistically significant. All statistical calculations were conducted using Stata/Se version 11.0 software (Stata Corporation, College Station, TX).

Power calculations for this replication study were performed as previously described[18] (Supplementary Table S1).

Results

A total of 1826 subjects participated in the study, including 1226 control subjects (mean age ± SD, 60.0 ± 9.8 years; ratio of female to male, 60.4:39.6), 300 cases with nAMD (mean age ± SD, 69.4 ± 8.9 years; ratio of female to male, 37.0:63.0) in one or both eyes, and 300 cases with PCV (mean age ± SD, 66.8 ± 9.7 years; ratio of female to male, 37.3:62.7) in at least one eye. The general characteristics of the study subjects are given in Table 1.

The rs13278062 and rs1713985 allele and genotype counts are given in Table 2. The minor allele frequencies (MAFs) of rs13278062 T allele are 0.32, 0.35, and 0.34 in control, nAMD, and PCV groups, respectively. The MAFs of rs1713985 G allele are 0.27, 0.26, and 0.27 in control, nAMD, and PCV groups, respectively. The two single nucleotide polymorphisms (SNPs) did not show a significant association with nAMD, PCV, or combination in the allele after correction for age and sex based on a logistic regression model (all $P > 0.05$, Table 2). The frequencies of all genotypes were in Hardy-Weinberg equilibrium in both the controls and the cases with nAMD or PCV (all $P > 0.05$, Supplementary Tables S2, S3).

We performed a meta-analysis of all three studies involving patients and controls of Chinese descent (Beijing, Hong Kong, and Singapore) into this meta-analysis (622 typical nAMD patients, 651 PCV patients, and 1652 control subjects evaluated). We note that the T allele of rs13278062 conferred increased risk for AMD (OR 1.17, 95% CI 1.04–1.32, $P = 0.004$; Fig. 1), consistent with previous observations.[10,11] For rs1713985, we observed no association between this SNP and AMD in the meta-analysis (OR 1.07, 95% CI 0.86–1.33, Fig. 2). The forest plot of the allelic models are demonstrated in Figures 1 and 2. No evidence of heterogeneity or publication bias was found for rs13278062 ($I^2 = 0\%$, $P = 0.44$; Egger's $P = 0.23$, Supplementary Fig. S1), but moderate heterogeneity was observed for rs1713985 ($I^2 = 67.0\%$, $P = 0.068$; Egger's $P = 0.37$, Supplementary Fig. S1).

We also performed subgroup analysis subdividing the late AMD cases into nAMD and PCV to assess whether rs13278062 and rs1713985 were associated with nAMD or PCV specifically in patients of Chinese descent. We found no statistical difference between nAMD or PCV in our study and the T allele. For the nAMD subgroup, there was a 1.17-fold conferred risk (95% CI 1.02–1.36) and for the PCV subgroup, the risk was 1.21-fold conferred (95% CI 1.01–1.44, Supplementary Fig. S2). No association was found in this meta-analysis with rs1713985 in either subgroup. For the nAMD subgroup, the combined OR was 1.065 (95% CI 0.81–1.40) and for the PCV subgroup, the combined OR was 1.05 (95% CI 0.88–1.25, Supplementary Fig. S3).

Discussion

Our study in a Beijing AMD case-control collection comprising 600 late-AMD cases and 1226 controls showed that rs13278062 at TNFRSF10A-LOC389641 on chromosome 8p21 polymorphism was not significantly associated with AMD risk, albeit having an effect size entirely consistent with previous observations. After formal meta-analysis of all available data (including Hong Kong and Singapore AMD collections of Chinese descent) we note nominal evidence of association between rs13278062 and increased risk of AMD, likely due to the presence of increased statistical power afforded by the increased sample size. For REST-C4orf14-POLR2B-IGFBP7 rs1713985, no association was found significantly in our study or in a meta-analysis, consistent with the replication attempt by Nakata et al.[11]

TNFRSF10A-LOC389641 on chromosome 8p21 (rs13278062) was first reported to be a new susceptibility loci for exudative (wet), late-stage AMD in a genome-wide association study in the Japanese population by Arakawa et al. in 2011.[10] This

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study	case/control	MAF (case)	MAF (control)	p value	OR	LI	UI
Beijing	600/1226	0.26	0.27	0.68	1.04	0.874	1.23
Hongkong	432/275	0.30	0.31	0.53	0.93	0.73	1.18
Singapore	240/151	0.30	0.22	0.13	1.32	0.92	1.89
Summary-Chinese	1272/1652	0.28	0.27	0.12	1.07	0.86	1.33
Isao Nakata-Kyoto	1355/3025	0.29	0.29	0.36	0.92	0.77	1.10
Isao Nakata-Yamanashi	323/115	0.30	0.23	0.06	1.39	0.99	2.05
Arakawa-GWAS	827/3323	0.33	0.29	9.03E-05	1.34	1.16	1.56
Arakawa-Replication	708/15569	0.33	0.28	5.71E-05	1.27	1.13	1.43
Overall (I-Squared=69.0%,P=0.004)							

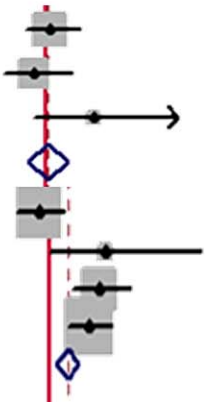

Figure 2. Meta-analysis of rs1713985 and nAMD Forest plot for meta-analysis of association between rs1713985 and nAMD. Each study is shown by the point estimate of the OR (the size of the square is proportional to the weight of each study) and 95% CI for the OR (*extending lines*). No association was found between rs1713985 and AMD (OR = 1.07, 95% CI: 0.86–1.33). We found moderate heterogeneity in this evaluation (I^2 = 67.0%, $P = 0.068$) and the overall (Japanese data included) ($I^2 = 69.0\%$, $P = 0.004$).

association was replicated by Nakata et al.[11] in two independent Japanese populations ($P = 0.040$, 95% CI 0.84 [0.71–0.99] and $P = 0.012$, 95% CI 0.65 [0.47–0.91]).[11] In our Beijing AMD study, we note a similar trend with the effect size of the risk allele (OR 1.14, $P = 0.11$) with late AMD, which is entirely in keeping with previous observations by Arakawa et al.,[10] and Nakata et al.,[11] which assessed independent Japanese AMD patient collections.

As our observed results at rs13278062 (OR 1.14, $P = 0.11$) could simply reflect lack of statistical power in our Beijing sample collection, we thus conducted a meta-analysis of all available published AMD data on individuals of Chinese descent. This revealed that the risk (T) allele of rs13278062 was significantly associated with increased risk for AMD (OR 1.17, 95% CI 1.04–1.32), consistent with previous observations without any accompanying intercohort heterogeneity or publication bias ($I^2 = 0\%$; Egger's $P = 0.23$).

In the subgroup analysis for the development of nAMD and PCV, we found no association with either nAMD ($P = 0.124$) or PCV ($P = 0.441$) subtypes, unlike the results in Hong Kong and Singapore, in which they found a significant association in the PCV subgroup ($P = 0.017$, 0.030) but not in the nAMD subgroup ($P = 0.337$, 0.330). It seems the association strength may be different in different subtypes in AMD. Even this meta-analysis result still demonstrates the T allele conferred for the risk of developing both nAMD and PCV. One explanation for the differences between our study and Hong Kong's or Singapore's is the difference of diagnosis and exclusion criteria. Another explanation is that our study comprised AMD cases and controls of Northern Chinese descent, whereas both Hong Kong and Singapore comprised mostly individuals of Southern Chinese descent, which is supported by recent genome-wide studies showing significant genetic stratification across individuals from different geographic locations across China.[19] In our power calculation[18] based on the Beijing AMD sample collection (Supplementary Table S1), we note that sufficient power under standard circumstances (≥80%) will exist only for effect sizes (OR) exceeding 1.30 per-copy of the risk allele. The per-allele OR observed by Arakawa et al.[10] for rs13278062 and rs1713985 is 1.37 at a control minor allele frequency of approximately 34%, and 1.30 at a minor allele frequency of approximately 28% in controls, respectively. Thus, our Beijing AMD study has greater than 90% power to detect both effect sizes (Supplementary Table S1). Our meta-analysis showed that the effect size of rs13278062 was significantly smaller in individuals of Chinese descent (ORmeta = 1.17, Pmeta = 0.004 for $n = 1273$ cases and 1652 controls; Fig. 1) compared with individuals of Japanese descent. We were unable to observe significant association for rs1713985 (ORmeta = 1.07; Fig. 2), suggesting this SNP does not confer a substantial, generalizable risk for late AMD. Further functional studies are necessary to make clear the mechanisms of these loci on the susceptibility to AMD.

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References

1. Lim LS, Mitchell P, Seddon JM, Holz FG, Wong TY. Age-related macular degeneration. *Lancet*. 2012;379:1728–1738.
2. Jager RD, Mieler WF, Miller JW. Age-related macular degeneration. *N Engl J Med*. 2008;358:2606–2617.
3. Yannuzzi LA, Ciardella A, Spaide RF, Rabb M, Freund KB, Orlock DA. The expanding clinical spectrum of idiopathic polypoidal choroidal vasculopathy. *Arch Ophthalmol*. 1997; 115:478–485.
4. Spaide RF, Yannuzzi LA, Slakter JS, Sorenson J, Orlach DA. Indocyanine green videoangiography of idiopathic polypoidal choroidal vasculopathy. *Retina*. 1995;15:100–110.
5. Kondo N, Honda S, Ishibashi K, Tsukahara Y, Negi A. LOC387715/HTRA1 variants in polypoidal choroidal vasculopathy and age-related macular degeneration in a Japanese population. *Am J Ophthalmol*. 2007;144:608–612.
6. Gotoh N, Yamada R, Nakanishi H, et al. Correlation between CFH Y402H and HTRA1 rs11200638 genotype to typical exudative age-related macular degeneration and polypoidal choroidal vasculopathy phenotype in the Japanese population. *Clin Experiment Ophthalmol*. 2008;36:437–442.

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7. Cheng Y, Huang L, Li X, Zhou P, Zeng W, Zhang C. Genetic and functional dissection of ARMS2 in age-related macular degeneration and polypoidal choroidal vasculopathy. *PLoS One*. 2013;8:e53665.
8. Bessho H, Kondo N, Honda S, Kuno S, Negi A. Coding variant Met72Thr in the PEDF gene and risk of neovascular age-related macular degeneration and polypoidal choroidal vasculopathy. *Mol Vis*. 2009;15:1107–1114.
9. Lima LH, Schubert C, Ferrara DC, et al. Three major loci involved in age-related macular degeneration are also associated with polypoidal choroidal vasculopathy. *Ophthalmology*. 2010;117:1567–1570.
10. Arakawa S, Takahashi A, Ashikawa K, et al. Genome-wide association study identifies two susceptibility loci for exudative age-related macular degeneration in the Japanese population. *Nat Genet*. 2011;43:1001–1004.
11. Nakata I, Yamashiro K, Akagi-Kurashige Y, et al. Association of genetic variants on 8p21 and 4q12 with age-related macular degeneration in Asian populations. *Invest Ophthalmol Vis Sci*. 2012;53:6576–6581.
12. Altshuler D, Daly MJ, Lander ES. Genetic mapping in human disease. *Science*. 2008;322:881–888.
13. Altshuler D, Hirschhorn JN, Klannemark M, et al. The common PPARgamma Pro12Ala polymorphism is associated with decreased risk of type 2 diabetes. *Nat Genet*. 2000;26:76–80.
14. Bird AC, Bressler NM, Bressler SB, et al. An international classification and grading system for age-related maculopathy and age-related macular degeneration. The International ARM Epidemiological Study Group. *Surv Ophthalmol*. 1995;39:367–374.
15. Schaeffeler E, Zanger UM, Eichelbaum M, Asante-Poku S, Shin JG, Schwab M. Highly multiplexed genotyping of thiopurine s-methyltransferase variants using MALD-TOF mass spectrometry: reliable genotyping in different ethnic groups. *Clin Chem*. 2008;54:1637–1647.
16. Consortium UIG, Barrett JC, Lee JC, et al. Genome-wide association study of ulcerative colitis identifies three new susceptibility loci, including the HNF4A region. *Nat Genet*. 2009;41:1330–1334.
17. Pan CW, Ikram MK, Cheung CY, et al. Refractive errors and age-related macular degeneration: a systematic review and meta-analysis. *Ophthalmology*. 2013;120:2058–2065.
18. Purcell S, Cherny SS, Sham PC. Genetic power calculator: design of linkage and association genetic mapping studies of complex traits. *Bioinformatics*. 2003;19:149–150.
19. Chen J, Zheng H, Bei JX, et al. Genetic structure of the Han Chinese population revealed by genome-wide SNP variation. *Am J Hum Genet*. 2009;85:775–785.

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