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LETTER

Common variant rs10033900 near the complement factor I gene is associated with age-related macular degeneration risk in Han Chinese population

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Age-related macular degeneration (AMD) is a late-onset, neurodegenerative disease that causes visual impairment and blindness in the elderly population.[1] Multiple genetic loci have been identified as contributing to AMD, including complement pathway-related genes (*CFH*,[2–4] *C2-CFB*,[5] *C3*,[6,7] *CFI*[8]), lipoprotein metabolism-related genes (*APOE*,[9–11] *LIPC*,[12] *TIMP3*[13]) and additional loci such as *VEGFA*[14] and *ARMS2/HTRA1*.[15,16] Fagerness *et al*[8] initially reported the complement factor I (*CFI*) gene region associated with AMD in a Caucasian cohort (1228 cases and 825 controls), with the rs10033900 variant located in a downstream region near the *CFI* gene showing the most significant signal ($P = 6.46 \times 10^{-8}$). This association signal was also confirmed by meta-analysis in a genome-wide association study (2594 cases and 4134 controls) with follow-up replication (5640 cases and 52174 controls) in the largest sample of individuals with European ancestry ($P = 4.1 \times 10^{-10}$).[14] In addition, the C allele of rs10033900 has been commonly identified as a protective allele against AMD in genetic studies.[8,14,17,18] However, there remains debate as to whether rs10033900 contributes to AMD susceptibility, due to contradicting evidence published in this journal.[18–20] For example, Ennis *et al*[20] confirmed the association between the *CFI* region and AMD susceptibility in a UK cohort, although rs10033900 showed no association with AMD. Kondo *et al*[18] demonstrated the association of rs10033900 with the C allele in a Japanese cohort, showing the same association direction as the results of Fagerness *et al*.[8] In contrast, Cipriani *et al*[19] observed no association between rs10033900 and AMD in a study of two independent cohorts from England and Scotland. To investigate the reported association of rs10033900 in the Han Chinese population, we studied this locus in our cohort of 288 unrelated AMD patients and 384 healthy controls.

Standard informed consent was obtained from all participants. A demographic summary of the phenotypic information is shown in Table 1. High-molecular-weight genomic DNA was prepared from venous blood using standard phenol-chloroform extraction. The variant of interest was genotyped using a TaqMan SNP genotyping assay (Life Technologies, Carlsbad, CA, USA) by laboratory personnel blinded to the sample status. This variant was in Hardy–Weinberg equilibrium among controls ($P = 0.08$). We also evaluated the association between alleles and AMD in terms of odds ratios (ORs), 95% confidence intervals (CIs) and corresponding *P*-values based on Fisher's exact test. We further evaluated the association between genotypes and AMD using logistic regression applied in three different statistic models: an additive model, dominant model and recessive model. In the additive model, homozygotes with two risk alleles were coded as 2, while heterozygotes with one risk allele were coded as 1 and homozygotes with non-risk alleles were coded as 0. In the dominant model, either homozygotes or heterozygotes with risk alleles were coded as 1, while homozygotes with non-risk alleles were coded as 0. In the recessive model, only homozygotes with two risk alleles were coded as 1, so both homozygotes and heterozygotes with non-risk alleles were coded as 0. Akaike's information criterion (AIC) values were calculated for each model, and the model with lowest AIC was considered the best-fitting model. All of the statistical tests were carried out using the R software package (http://www.r-project.org/), and a *P*-value < 0.05 was defined as statistical significance. Statistic power was calculated using the G*Power program.[21]

Table 1 Characteristics of the participants

Characteristics	*Case*	*Control*
N	288	384
GA subtype (%)[a]	53.5	—
CNV subtype (%)[a]	19.1	—
Female (%)	50.6	55.9
Age (years)	75.1 ± 7.0 (53–90)	67.6 ± 6.6 (52–86)

Abbreviations: CNV, choroidal neovascularization; GA, geographic atrophy.
Data are presented as the mean ± s.d. with range (minimum to maximum).
[a]The remainder of the cases (27.4%) were diagnosed with age-related macular degeneration, but we could not acquire specific subtype information.

Our results replicated the association between rs10033900 and AMD (OR, 1.33; 95% CI, 1.04–1.69; $P = 0.021$) in the Han Chinese population. We found genotypic associations both in the additive model (OR, 1.30; 95% CI, 1.03–1.63; $P = 0.025$) and dominant model (OR, 1.42; 95% CI, 1.03–1.97; $P = 0.033$). The results of a previous Japanese study[18] were in favor of the recessive model ($P = 0.0035$ compared with $P = 0.036$ in the additive model), whereas in our study, the additive model was found to be the best-fitting model of inheritance with the lowest AIC among the three models (Table 2). Furthermore, our study was well-powered because the current sample size demonstrated >90% statistical power to detect association significance (OR, 1.33). Interestingly, we observed that the minor allele, that is, the C allele, conferred an increased risk for the development of AMD, whereas a decreased risk of such an association with AMD was reported in previous studies.[8,17,18,20] In particular, a recent Chinese study (119 exudative AMD patients and 120 controls) also reported a protective effect of rs10033900 against AMD.[22] However, we believe our result is reliable because the frequency of the C allele in our controls was 0.324, which is close to the frequency of 0.328 reported for CHB (Han Chinese in Beijing, China) from the HapMap project (http://hapmap.ncbi.nlm.nih.gov/). Moreover, the use of controls from this Chinese study with a C allele frequency of 0.293[22] indicated a higher risk effect for AMD (OR, 1.53; 95% CI, 1.11–2.13; $P = 0.008$). Although Wu *et al*[22] focused on exudative AMD samples and we used overall AMD samples, we do not believe that the subtypes of AMD in our study caused the observed difference in the effective direction, as the frequencies of the C allele were very close between different subtypes (0.387 in geographic atrophy, 0.381 in choroidal neovascularization and 0.395 in cases diagnosed with AMD but without further subtype information) and were all higher

Table 2 Allelic and genotypic associations between rs10033900 and age-related macular degeneration

		Genotype distributions				*Genotype*	
	C allele frequency	*CC/CT/TT*	*Associations*	*Allele*	*Additive*	*Dominant*	*Recessive*
Case	0.388	0.168/0.440/0.392	*P*-value[a]	**0.021**	**0.025**	**0.033**	0.14
Control	0.324	0.126/0.396/0.479	OR (95% CI)	1.33 (1.04–1.69)	1.30 (1.03–1.63)	1.42 (1.03–1.97)	1.40 (0.89–2.21)
CHB	0.328	0.109/0.453/0.438	AIC		839.23	839.67	842.07

Abbreviations: AIC, Akaike's information criterion; CHB, Han Chinese in Beijing, China from the International HapMap Project; CI, confidence interval; OR, odds ratio.
[a]Statistical significance with *P*-values <0.05 are indicated in bold type.

than the frequencies observed in control groups (0.293–0.328). In the European population, the C allele is a major allele with a frequency of 0.549, according to the CEU (Utah residents with Northern and Western European ancestry from the CEPH collection), whereas it is a minor allele among CHB. Also, the prevalence of AMD seems to be lower among Chinese than Caucasian populations.[23–26] Therefore, the inconsistent association direction may be due to genetic heterogeneity and differences in population prevalence.

In addition to rs10033900, other common SNPs located within the *CFI* region have also been associated with AMD in several genome-wide association studies, including rs4698775,[27] rs2285714[13] and rs7690921,[12] although none of these SNPs occurs in high LD with rs10033900 (r^2 ranges from 0.107 to 0.138 according to HapMap for CHB and JPT(Japanese in Tokyo, Japan)). Additionally, rs10033900 also seems to be independent of other common SNPs ($r^2<0.445$) located in a LD block spanning 40 kb downstream to rs10033900 near the genes *PLA2G12A* and *CASP6*, whereas SNPs within the *PLA2G12A* locus have been correlated with rs10033900 in European populations.[8,20] Thus, it is unlikely that this variant represents a superior marker for causal variants in this AMD-susceptible region in Asians. Notably, the roles that rare *CFI* mutations have in the development of AMD were demonstrated in recent studies.[28,29] For example, van de Ven *et al*[29] identified a rare, highly penetrant missense mutation c.355G>A (p.(Gly119Arg); RefSeq NM_000204.3) that confers a high risk of AMD (OR, 22.20; $P=3.79\times10^{-6}$) via the regulation of C3b degradation. Although this likely causal mutation was also identified by Seddon *et al*,[28] the individual mutation showed no associations with AMD ($P=0.24$);[28] instead, the burden of rare functional *CFI* variant enrichment in this gene was significantly increased in AMD cases (OR, 3.6; $P=2.0\times10^{-8}$), indicating the combined effects of multiple rare mutations on the modulation of AMD risk.[28] Therefore, these rare variant studies have provided important implications for causal SNPs or mutations related to AMD in the Chinese population. However, no mutated alleles for c.355G>A (p.(Gly119Arg))[29] were detected in 192 of our AMD cases (data were not shown). The sample sizes of such studies must be scaled up because these low-frequency variants are not easily detected. In addition, due to discrepancies in rare allelic architectures between Chinese and Caucasian populations, the same rare mutations found in Caucasians may not be replicated in Chinese cohorts. Thus, resequencing of the functional regions (eg, exons) of the *CFI* gene as well as the downstream region (eg, *PLA2G12A*) would help to better identify causal variants contributing to AMD susceptibility.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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DATA SUBMISSION

The data in this manuscript have been submitted to public database GWAS Central (https://www.gwascentral.org/) and the study ID is HGVST1714.

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