Heterogeneity of the Genetic Risk in Age-Related Macular Disease

A Population-Based Familial Risk Study

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Objective: To assess the extent of heterogeneity of the genetic risk of age-related macular disease (AMD) among families.

Design: Case-controlled population-based familial aggregation study.

Participants: Participants comprised 190 first-degree relatives of 65 case probands and 347 relatives of 100 control probands. All probands had been identified from the baseline phase of the Rotterdam Study in The Netherlands.

Methods: A family score was computed for each family based on the presence and type of macular disease, the expected risk of disease, and the number, extent of kinship, and age of all family members.

Main Outcome Measures: Presence and stage of AMD as diagnosed on fundus photographs, family score, and logistic regression coefficient.

Results: The family score of case families showed a peak of approximately 0 with a skewed tail (14% of families) of higher than expected risks of disease toward a maximum of 2.9. The family score of control families centered on 0, apart from 1 outlier. The risk of AMD increased significantly with higher family scores ($\beta = 1.34$; $P<0.001$).

Conclusions: The heterogeneity of genetic risk among AMD families is considerable, and the proportion of high-risk families is relatively small. The family score method is relevant for genetic counseling as well as for implementation in studies of genetic dissection of AMD. *Ophthalmology 2005;112:482–487 © 2005 by the American Academy of Ophthalmology.*

Age-related macular disease (AMD) is a common complex eye disorder presumably caused by a variety of molecular defects. Environmental factors such as smoking, oxidation, and vascular disease are well-established risk factors, but genetic factors also contribute considerably to the occurrence of AMD.[1] In a previous familial aggregation, study we estimated that approximately one fourth of all AMD is the result of genetic defects and that first-degree relatives of affected probands have a 4 times increased risk of developing a similar phenotype.[2]

As yet, molecular research has provided little information on the genes involved. Candidate gene analysis has revealed an association with the genes ABCA4,[3] APOE,[4–7] ACE,[8] manganese superoxide,[9] paraoxenase,[10] and the fibulin 5 gene.[11] Although important for the identification of potential pathogenic pathways, these genes seem to be involved in only a small proportion of AMD cases. Genome-wide screenings and linkage analyses in cohorts of siblings and small families have provided a variety of susceptibility loci with mostly low log out of odds scores.[12–17] The latter is likely to be the result of the large range of genetic heterogeneity of minor disease genes. A genome screen of a large American family with atrophic macular degeneration has elicited the Hemicentin-1 gene as the disease-causing gene that determined the association with locus 1q25 to 31,13, but this finding was not replicated in other populations.[16]

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To increase the chance of locating genes, a key question that remains is how to discriminate those with AMD caused by genetic factors from those in whom AMD develops because of environmental exposures. The aim of this study was to assess the extent of heterogeneity of the genetic risk of AMD among families. For each family of our former aggregation study, we calculated a risk score based on the observed and expected number of affected relatives using demographic and epidemiologic data. This methodology allowed us to discriminate between high- and low-risk families, to investigate their frequencies, and to assess the risk of AMD.

Patients and Methods

Collection of Families

Design of the familial aggregation study and methods of data collection were described previously.[2] In brief, all probands were identified from the baseline phase of the Rotterdam Study; case probands (n = 101) were all subjects with atrophic or neovascular AMD, and control probands (n = 154) were a randomly selected sample of study subjects who did not have any soft drusen (≥63 μm), nor any atrophic or neovascular AMD. All probands were white, of Dutch descent, and similar in gender distribution (63% women among cases vs. 56% women among controls; age-adjusted $P = 0.64$), but cases differed in age from controls (case mean age of 81.9 years vs. control mean age of 76.7 years; $P<0.001$). Genealogical data of the last 5 generations were obtained from each proband; no genealogical link was detected. All siblings, including half siblings, and children were subsequently invited for a similar ophthalmic examination as the probands at the research center or at their home. The study was approved by the Medical Ethics Committee of Erasmus University, Rotterdam, and written informed consent was obtained from all participants.

Diagnosis of Age-Related Macular Disease

The ophthalmologic examination included stereoscopic photography of both a 20° macular field (Topcon TRC-SS2 stereoscopic fundus camera; Topcon Optical Co., Tokyo, Japan) and 35° macular field (Topcon TRV-50VT fundus camera; Topcon Optical Co.). Persons who were examined at home were photographed with a portable camera (35° field; Kowa RC-2 fundus camera; Kowa Corporation Ltd., Tokyo, Japan). Fundus transparencies were graded for the presence of AMD features in a masked fashion according to the International Classification System, identical to the protocol that was used for probands.[18] Age-related macular disease was stratified in 2 stages of disease. Early AMD was defined as either the presence of soft distinct drusen with pigmentary changes or the presence of soft indistinct or reticular drusen. Late AMD was defined as the presence of atrophic (geographic atrophy) or neovascular AMD.[2]

Familial Risk Assessment

Baseline characteristics of case and control probands were compared using Student's *t* test for continuous variables and the chi-square test for categorical variables. Age-specific prevalences of early and late AMD were determined from the baseline phase of the Rotterdam Study,[19,20] and they served as the expected outcome of AMD for each relative (Table 1). A family score (FS) was computed for each family based on the disease status and the number and age of all family members available for analyses. Because a few half-siblings were included, a correction in kinship coefficient was made. The FS was calculated according to the formula described by Houwing-Duistermaat and Van Houwelingen[21]:

$$FS = \sum (\Psi_j(O_j - E_j)),$$

where j indicates a family member, Ψ_j is twice the kinship coefficient between the family member j and the proband (0.5 for siblings and children of the proband and 0.25 for half-siblings), O_j is the observed disease status (1 affected, 0 nonaffected), E_j is the expected value for each family member j stratified for age, and Σ indicates the sum of all individual scores. The risk of late AMD was estimated for the FS using logistic regression analysis, adjusting for the possible confounding effect of age and gender. No increased risk for family score ($\beta = 0$) was considered as the null hypothesis, which would indicate that the distribution of the FS in cases equals the distribution in controls. An additional analysis was performed to adjust for the potential confounding effect of smoking and atherosclerosis.

Table 1. Age-Specific Prevalence (%) of Age-Related Macular Disease in the Baseline Phase of the Rotterdam Study, 1989–1993

Age (yrs)	Early Age-Related Macular Degeneration*	Late Age-Related Macular Degeneration†	Total Age-Related Macular Degeneration
55–59	2.2	0.2	2.4
60–64	3.2	0.1	3.3
65–69	5.1	0.5	5.6
70–74	10.6	0.8	11.4
75–79	12.6	1.7	14.3
80–84	14.8	6.5	21.3
85–89	18.5	9.2	27.7
90+	21.2	17.7	38.9

*Defined as the presence of either soft distinct drusen with pigmentary changes or the presence of soft indistinct or reticular drusen.
†Defined as the presence of geographic atrophy, neovascular macular degeneration, or both.

Description of Phenotype in High-Risk Families

To assess whether high-risk families present a specific phenotype, all photographs of high-risk families were evaluated in an additional analysis. We considered high risk to be an FS of 1.3 or more, the level below which 99% of FSs of controls ranged. Features of

Table 2. General Characteristics of the Study Population

Variable	Case Families (n = 65)	Control Families (n = 100)
Total no. of relatives	190	347
No. of siblings	77	145
Mean age of siblings (yrs ± SD)	75.8 ± 8.7	75.4 ± 9.4
% women among siblings	55	59
No. of offspring	113	201
Mean age of offspring (yrs ± SD)	53.7 ± 10.4*	48.8 ± 8.9
% women among offspring	43	46

SD = standard deviation.
*$P < 0.001$ for the difference with relatives of controls.