Iris Pigmentation and Extent of Disease in Patients With Neovascular Age-Related Macular Degeneration

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Purpose. To determine whether the extent of disease in age-related macular degeneration (AMD) varies with iris pigmentation.

Methods. The authors assessed visual function and macular appearance in the fellow eye or both eyes of 132 white patients with unilateral neovascular AMD. All patients had a visual acuity of 20/60 or better in the fellow eye. Eighty-nine of the patients were coded as having light irides (blue, green, or hazel) and 43 were coded as having dark irides (brown); the two groups of patients had comparable mean ages.

Results. By the Mann-Whitney test for differences in mean rank, fellow eyes with light irides showed a marginally worse visual acuity \((P = .156)\) but significantly more visual field impairment by letter recognition perimetry \((P = .011)\) and the macular threshold test of the Humphrey Field Analyzer \((P = .043)\), and more retinal pigment epithelial atrophy \((P = .009)\) than fellow eyes with dark irides. For eyes with a choroidal neovascular membrane (CNVM), those with light irides had significantly lower visual acuities \((P = .006)\) and larger scars \((P = .006)\) than eyes with dark irides. In addition, extent of disease in the eye with a CNVM was positively correlated with extent of disease in the fellow eye for most comparisons.

Conclusions. These observations suggest that light iris pigmentation is associated with more extensive retinal disease in patients with unilateral neovascular AMD. Furthermore, in such patients, those with worse disease in the eye with a CNVM may tend to have more extensive atrophic disease in the fellow eye. Invest Ophthalmol Vis Sci. 1994;35:2734-2740.

There is a continuing interest in identifying risk factors for age-related macular degeneration (AMD) that might provide insight into its cause and, possibly, treatment. Iris pigmentation has been considered by several studies. There have been reports of an increased prevalence of AMD among patients with light irides in the United States\(^1^,\) and of a higher incidence in white patients in England than in black patients in Africa.\(^3\) AMD resulting in blindness was found to be limited to white patients in East Baltimore.\(^4\) In contrast, other studies have found no difference based on iris pigmentation in the frequency of affected Scan-
Except where noted, findings are from the baseline visit. Patients were referred primarily from retinal specialists in New England; a few patients were self-referred. Eligibility criteria were the presence of a choroidal neovascular membrane (CNVM) in the affected eye, a corrected Snellen visual acuity ≥ 20/60 and sufficiently clear media to allow adequate evaluation of the fundus in the fellow eye, macular drusen in both eyes, and no sign of other retinal disease. Eligible patients also had to be able to fixate within the central 2° with their fellow eye as determined by ophthalmoscopy. To our knowledge, iris color was not considered in the referral of any patient. This report includes all white patients meeting the eligibility criteria who were enrolled in our prospective study; to rule out a possible confounding effect of racial background on iris pigmentation, the black patient who was enrolled was not included in this presentation.

The diagnosis of unilateral neovascular AMD was confirmed by ocular examination, fundus photographs, fluorescein angiograms, and previous records. Based on fundus photographs of fellow eyes, 83% of the patients had at least some soft, confluent drusen or pigmentary changes in the macula. Extent of involvement of the macula by drusen was estimated to have a median value of 4% and a mean value of 7%; these are similar to the mean value (4.8%) reported recently for the fellow eyes of patients with unilateral geographic atrophy or exudative macular degeneration in a population study. All patients gave informed consent after the nature and possible consequences of the study had been explained. The procedures followed the tenets of the Declaration of Helsinki and were approved by the internal review boards of the Massachusetts Eye and Ear Infirmary and Harvard Medical School.

Iris color was determined both by asking the patient and by examiner observation with the aid of a penlight, before evaluation of visual function, pupillary dilation, and assessment of retinal appearance. In all but one case, the two judgments agreed; for the exception, the judgment of the examiner was coded. Iris pigmentation was then categorized dichotomously as light (blue, green, or hazel) or dark (brown). Table 1 compares demographic and other characteristics for these two groups of patients, including some that have been considered as risk factors for AMD. Eighty-nine patients were coded as having light irides, and 43 patients were coded as having dark irides. The patients with light irides were not significantly different from those with dark irides with respect to mean age, percentage by sex, mean refractive error (equivalent sphere), percentage with a positive family history of AMD, percentage with cardiovascular disease, or percentage with high blood pressure. The two groups were also not significantly different with respect to dietary intake of nutrients (including supplements) based on a standardized food-frequency questionnaire that converts the weekly intake of 61 food items into 55 nutrients; this questionnaire was completed after the baseline visit by 98% of available patients. No median nutrient value differed significantly between the two groups of patients (see Table 1 for examples); this was true even when analyses were done by sex (not shown).

### TABLE 1. Demographic and Other Characteristics Versus Iris Pigmentation

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Light Iris</th>
<th>Dark Iris</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number</td>
<td>89 Patients</td>
<td>43 Patients</td>
<td>—</td>
</tr>
<tr>
<td>Age (mean ± SE)</td>
<td>74.7 ± 0.7 yrs</td>
<td>73.8 ± 1.1 yrs</td>
<td>0.485*</td>
</tr>
<tr>
<td>Males</td>
<td>40%</td>
<td>53%</td>
<td>0.158†</td>
</tr>
<tr>
<td>Refractive error (mean ± SE)</td>
<td>+0.82 ± 0.21 D</td>
<td>+1.33 ± 0.31 D</td>
<td>0.170*</td>
</tr>
<tr>
<td>+ Family Hx of AMD</td>
<td>25%</td>
<td>19%</td>
<td>0.434†</td>
</tr>
<tr>
<td>Cardiovascular disease</td>
<td>27%</td>
<td>21%</td>
<td>0.453†</td>
</tr>
<tr>
<td>High blood pressure</td>
<td>44%</td>
<td>37%</td>
<td>0.470†</td>
</tr>
<tr>
<td>Dietary protein (median)</td>
<td>74.68 g</td>
<td>74.18 g</td>
<td>0.635‡</td>
</tr>
<tr>
<td>Dietary animal fat (median)</td>
<td>31.20 g</td>
<td>28.51 g</td>
<td>0.680‡</td>
</tr>
<tr>
<td>Dietary vegetable fat (median)</td>
<td>30.61 g</td>
<td>29.48 g</td>
<td>0.410‡</td>
</tr>
<tr>
<td>Dietary cholesterol (median)</td>
<td>252.4 g</td>
<td>248.7 g</td>
<td>0.984‡</td>
</tr>
<tr>
<td>Dietary carotenoids (median)</td>
<td>12570 IU</td>
<td>9524 IU</td>
<td>0.430‡</td>
</tr>
<tr>
<td>Dietary vitamin A (median)</td>
<td>16800 IU</td>
<td>15816 IU</td>
<td>0.531‡</td>
</tr>
<tr>
<td>Dietary vitamin E (median)</td>
<td>11.17 mg</td>
<td>9.0 mg</td>
<td>0.934‡</td>
</tr>
<tr>
<td>Dietary calcium (median)</td>
<td>790.0 mg</td>
<td>682.4 mg</td>
<td>0.201‡</td>
</tr>
<tr>
<td>Dietary zinc (median)</td>
<td>13.35 mg</td>
<td>12.41 mg</td>
<td>0.324‡</td>
</tr>
</tbody>
</table>

* Student's t-test.
† Chi-square test.
‡ Mann-Whitney test.
§ Alpha-tocopherol equivalents.
Assessment of Visual Function

Best-corrected visual acuity was measured at 18 feet on a projected Snellen chart with a background luminance of ~120 cd/m² or, for many eyes with a CNVM, by finger counting. The latter was converted to decimal acuity based on distance from the patient. For fellow eyes, we remeasured visual acuity with a chart used by the Early Treatment Diabetic Retinopathy Study (ETDRS) projected onto the screen at the same background luminance. This chart contains five letters of comparable difficulty on each line; letters on adjacent lines differ in size by 0.1 log unit. Visual acuity was quantified as the number of letters identified (45 letters = 20/20), which provided a more continuous distribution than that for Snellen acuity. For eight patients whose ETDRS chart acuities were not available, we estimated them from the linear regression of ETDRS chart acuity on Snellen acuity based on the remaining patients (R = .87, P < .001).

We measured visual field impairment of fellow eyes by a method of letter recognition perimetry and by the macular threshold test of the Humphrey Field Analyzer. The number of field defects on each of these tests was previously reported to be strongly correlated with the extent of retinal pigment epithelial (RPE) atrophy among patients with AMD. Letter recognition perimetry, formerly referred to as computerized acuity mapping of the macula or CAMM, involved identification of upper-case black letters flashed consecutively on a computer screen (Macintosh, Apple Computer, Cupertino, CA) in random alphabetical order and location within the central 10° X 10°. Each letter was flashed to one of 100 locations distributed in a grid pattern of 1° intervals and to fixation provided a continuous distribution than that for Snellen acuity. For eight patients whose ETDRS chart acuities were not available, we estimated them from the linear regression of ETDRS chart acuity on Snellen acuity based on the remaining patients (R = .87, P < .001). We measured visual field impairment of fellow eyes by a method of letter recognition perimetry and by the macular threshold test of the Humphrey Field Analyzer. The number of field defects on each of these tests was previously reported to be strongly correlated with the extent of retinal pigment epithelial (RPE) atrophy among patients with AMD. Letter recognition perimetry, formerly referred to as computerized acuity mapping of the macula or CAMM, involved identification of upper-case black letters flashed consecutively on a computer screen (Macintosh, Apple Computer, Cupertino, CA) in random alphabetical order and location within the central 10° X 10°. Each letter was flashed to one of 100 locations distributed in a grid pattern of 1° intervals and to fixation provided a cross in the center. The screen had a luminance of 100 cd/m² and was viewed at a distance of 1 foot with the appropriate optical correction; the fellow eye was occluded. While fixating on the cross, patients clicked the “mouse” to cause the cross to disappear and to flash a letter for 20 msec. Patients attempted to name the letter, and their answers were then entered at the keyboard by the examiner. This returned the fixation cross to the screen, and the patient repeated the procedure. Letter size increased with increasing distance from the cross according to a formula that compensated for the normal decline in recognition of flashed letters of fixed size with increasing eccentricity. Locations where letters were missed were retested after the initial run to help eliminate errors due to lapses of attention or fixation or due to misidentification of ambiguous letters. The computer then generated an error count. Because the upper (95%) limit of normal based on 31 volunteers (age range, 63 to 84 years) was four errors, this value was subtracted from each patient’s score and negative values were converted to zero. The number of resulting errors was then considered as the number of field defects.

The macular threshold test of the Humphrey Field Analyzer (model 611, Allergan Humphrey, San Leandro, CA) was administered to a consecutive subset of patients (n = 97) on a visit after baseline with the standard stimulus (Goldmann size III, 200 msec duration) and background (10 cd/m²) and with the pupil monitor; the fellow eye was patched, and the room lights were dimmed. Sensitivities in decibels were determined for the 16 locations within a 4° X 6° rectangular matrix. Locations with reduced sensitivity were identified based on estimated normal age-corrected 95% confidence limits provided by the manufacturer and were considered as field defects.

Assessment of Retinal Appearance

In addition to the ophthalmoscopic examination, macular appearance was evaluated from color slides projected onto a screen. Photographs were of sufficient quality to allow detailed evaluation in 129 (98%) of the fellow eyes and in 121 (92%) of the eyes with a CNVM. For the fellow eyes, we estimated the percentage of macula with RPE atrophy within a circle drawn on a plastic overlay that subtended 15°. The atrophy ranged from patchy hypopigmentation to large, geographic, sharply demarcated, almost totally white areas of RPE absence. We also counted the number of focal areas of RPE hyperpigmentation within the circle. These fellow eyes were also rated as low risk or high risk for neovascularization based on their clinical appearance with respect to the extent of soft, confluent drusen and RPE pigmented changes, in accordance with criteria set forth in previous studies. This dichotomous variable, like iris pigmentation, was tested as a risk factor for predicting macular loss in the eyes with a CNVM. For the eyes with a CNVM, rated in sessions separate from those used to rate fellow eyes, we estimated the equivalent diameter of macular area in disc diameters affected by scarring, hemorrhage, and exudation (hereafter referred to as scar diameter). Macular appearance of a given eye was rated by observers masked to that patient’s iris pigmentation, visual findings, and retinal appearance of the other eye.

Statistical Analyses

Because the frequency distributions were skewed for most outcome measures, we used the distribution-free Mann-Whitney test for differences in mean rank to assess the relationship of each outcome measure to iris pigmentation. We also used this test to relate scar diameter of eyes with a CNVM to risk for neovascularization of fellow eyes. A multiple regression model was then used to determine the relationship of scar diameter, converted to logarithms to approximate a normal distribution, to both iris pigmentation and risk for neo-
vascularization of the fellow eye. With this model, the effect of each factor on scar diameter was evaluated while controlling for the effect of the other factor. The association of these two factors, iris pigmentation and risk for neovascularization of the fellow eye, was determined by chi-square analysis. Lastly, we assessed the interocular correlation between each pair of outcome measures by the Spearman rank correlation. Analyses were done with JMP, version 2.0.4 (SAS Institute, Cary, NC) on a Macintosh computer.

RESULTS

Fellow Eye Correlations With Iris Pigmentation

Figure 1 illustrates plots of the variation by iris pigmentation for the 10th, 25th, 50th (median), 75th, and 90th percentiles of the cumulative frequency distribution of scores for each outcome measure in fellow eyes. For each measure, the median and other percentiles for eyes with light irides tend to correspond to worse scores compared to those for eyes with dark irides. Eyes with light irides had marginally lower visual acuities ($P = .156$) but significantly more visual field defects by letter recognition perimetry ($P = .011$) and by the Humphrey Field Analyzer ($P = .043$), more RPE atrophy ($P = .017$), and more focal areas of RPE hyperpigmentation ($P = .002$) than eyes with dark irides.

CNVM Eye Correlations With Iris Pigmentation and the Fellow Eye's Risk for Neovascularization

For the eyes with a CNVM, those with light irides had significantly lower visual acuities ($P = .006$) and larger scars ($P = .006$) than those with dark irides (Fig 2). Irrespective of iris pigmentation, scar diameter was also found to be significantly larger for patients whose fellow eye was rated at high risk for neovascularization ($n = 63$) than for patients whose fellow eye was rated at

**FIGURE 1.** Percentile plots of decimal visual acuity, the number of visual field defects based on letter recognition perimetry (maximum = 97) and on the macular threshold test of the Humphrey Field Analyzer (maximum = 16), the percentage area of RPE atrophy, and the number of focal areas of RPE hyperpigmentation in fellow eyes versus iris pigmentation. $P$ values are for the Mann-Whitney test. Visual acuities are decimal equivalents based on the number of letters read on a projected ETDRS chart.
low risk (n = 58) (P < .001). Scar diameter remained significantly higher in eyes with light irides (P < .001) and in patients whose fellow eye was considered at high risk for neovascularization (P = .002) when the effect of each factor on scar diameter was determined while controlling for the effect of the other factor (Fig 3). Because iris pigmentation and the risk for neovascularization were independent of each other (P = 0.962) and of comparable strength in predicting scar diameter (R² = 0.085 and 0.071, respectively), their combination provided twice the predictive power (R² = 0.154) of either alone. On average, patients with light irides and a fellow eye judged to be at high risk for neovascularization had a scar diameter in the opposite eye that was 44% larger than that of patients with dark irides and a fellow eye judged to be at low risk for neovascularization.

**Interocular Correlations**

Table 2 presents the interocular correlation for each pair of outcome measures (dependent variables). The correlations were significant and in the expected direction for visual acuity of the eye with a CNVM versus number of errors on letter recognition perimetry and on the macular threshold test by testing the fellow eye. The correlations were significant and in the expected direction also for scar diameter in the eye with a CNVM versus visual acuity, number of errors on letter recognition perimetry, number of errors on the macular threshold test, and percent area of atrophy determined for the fellow eye.

**DISCUSSION**

This study shows that light iris pigmentation was associated with more extensive macular disease than dark iris pigmentation in both eyes of patients with unilat-

teral neovascular AMD. Fellow eyes of patients with lighter pigmented irides tended to have more visual field impairment by letter recognition perimetry and by the macular threshold test of the Humphrey Field Analyzer and more RPE atrophy and focal areas of RPE hyperpigmentation. On the other hand, visual acuity did not vary significantly with iris pigmentation; this could reflect, in part, our eligibility constraint that patients have acuities ≥ 20/60 in their fellow eyes. For eyes with a CNVM, those with lighter irides tended to have worse visual acuities and larger scars than those with darker irides.

These findings may help to explain the inconsistent results of studies that compared AMD prevalence to iris pigmentation. Because referrals tend to be biased toward patients with symptoms, the increased prevalence of light irides among patients with AMD in some case–control studies1,2 may be due, in part, to the tendency for patients with light irides to have more extensive disease (and symptoms) than patients with dark irides. Similarly, the finding that recruited patients with light irides were generally younger than those with dark irides2 might be explained if the former became symptomatic at an earlier age. In contrast, referrals based on the presence of neovascular disease, as in a study that failed to find an association between prevalence of AMD and iris pigmentation,3 would probably not depend upon symptoms deriving from milder, atrophic changes. Although our results do not bear upon the prevalence of AMD, they raise the possibility that severity of disease, as it relates to iris pigmentation, may have been a confounding factor in some previous studies.

**FIGURE 2.** Percentile plots of decimal visual acuity and scar diameter in disc diameters in eyes with a CNVM versus iris pigmentation. P values are for the Mann-Whitney test.

**FIGURE 3.** Mean ± SE for scar diameter in log₅ disc diameters in eyes with a CNVM versus iris pigmentation and versus the risk for neovascularization based on macular appearance in the fellow eye. Plotted values for the effect of iris pigmentation on scar diameter have been adjusted for the effect of risk for neovascularization in the fellow eye on scar diameter, and vice versa. P values are for Student’s t-tests.
TABLE 2. Interocular Correlations for Extent of Disease

<table>
<thead>
<tr>
<th>Neovascular Eye</th>
<th>Fellow Eye</th>
<th>R*</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Visual acuity (decimal)</td>
<td>Visual acuity (letters)</td>
<td>.11</td>
<td>.221</td>
</tr>
<tr>
<td>Letter recognition perimetry (errors)</td>
<td>-.22</td>
<td>.015</td>
<td></td>
</tr>
<tr>
<td>Macular threshold test (errors)</td>
<td>-.31</td>
<td>.003</td>
<td></td>
</tr>
<tr>
<td>RPE atrophy (% area)</td>
<td>.03</td>
<td>.758</td>
<td></td>
</tr>
<tr>
<td>Focal hyperpigmentation (areas)</td>
<td>-.09</td>
<td>.361</td>
<td></td>
</tr>
<tr>
<td>Scar diameter (DD)</td>
<td>Visual acuity (letters)</td>
<td>-.22</td>
<td>.016</td>
</tr>
<tr>
<td>Letter recognition perimetry (errors)</td>
<td>.29</td>
<td>.001</td>
<td></td>
</tr>
<tr>
<td>Macular threshold test (errors)</td>
<td>.30</td>
<td>.004</td>
<td></td>
</tr>
<tr>
<td>RPE atrophy (% area)</td>
<td>.21</td>
<td>.021</td>
<td></td>
</tr>
<tr>
<td>Focal hyperpigmentation (areas)</td>
<td>.11</td>
<td>.236</td>
<td></td>
</tr>
</tbody>
</table>

* Spearman rank correlation coefficient.

We also observed a positive relationship, independent of iris pigmentation, between scar diameter in the eye with a CNVM and the presence of more extensive soft, confluent drusen and pigmentary changes in the fellow eye, factors thought to increase the risk for neovascularization. Because iris pigmentation and the fundus risk for neovascularization of the fellow eye were uncorrelated and, therefore, independent predictors, their combination in our regression model provided greater power than did either alone. Several studies have shown that drusen have little, if any, direct bearing upon visual loss in eyes with dry AMD, for which reason the present study did not consider the amount of drusen as an outcome measure. Instead, the clinical significance of extensive soft, confluent drusen may be to increase not only the likelihood, but also the consequence, of the neovascular process. When this risk factor is combined with light iris pigmentation in a given patient, the prognosis for neovascularization may be particularly grave.

The link between iris pigmentation as an independent variable and extent of disease in both eyes of our patients as dependent variables naturally raised the probability of a correlation between the dependent variables as well, which was confirmed for most outcome measures. Thus, among our patients with unilateral neovascular AMD, those with worse disease in the eye with a CNVM tended to have worse atrophic disease in the fellow eye. This interocular correlation may reflect a cause and effect relationship. For example, scar size due to a CNVM may depend upon the degree of preexisting atrophy, as inferred from the retinal appearance of the fellow eye. Alternatively, the extent of disease in both eyes may be governed, in part, by some common factor (systemic, environmental, genetic). Whichever was true, it would in no way affect the validity of the finding that iris pigmentation helps to predict visual and retinal outcome in both eyes.

A physiological basis for this proposed association remains speculative. It is thought by some that increased ocular melanin, which is positively correlated with increased iris pigmentation, may protect the retina through better absorption of light and scavenging of free radicals produced by photo-oxidation. Perhaps corroborative is the epidemiologic evidence that long-term outdoor light exposure promotes the occurrence of severe macular degeneration and that wearing eyeglasses, sunglasses, or a hat may protect against the appearance of retinal signs of AMD. However, another study found that the prevalence of neovascular disease was unrelated to history of sunlight exposure. Alternatively, iris pigmentation in our sample of patients may be a surrogate for some ocular, medical, or ethnic risk factor for AMD that modifies disease expression. Review of our patients' refractive errors, family histories, medical histories, and diets, however, revealed no significant variation based upon iris pigmentation. Nevertheless, the possibility that disease severity is related to another factor correlated with iris pigmentation needs to be further addressed.

Key Words
macular degeneration, neovascularization, iris pigmentation, visual acuity, visual field

References