

# The Effect of Age on the Area of Complete Spatial Summation for Chromatic and Achromatic Stimuli

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**PURPOSE.** Previously, an association between the area of complete spatial summation (Ricco's area) and age under scotopic conditions had been found. The authors sought to determine whether Ricco's area is similarly associated with age under photopic achromatic and selective S-cone conditions in peripheral vision and whether any association relates to a loss of ganglion cell density as determined by measurements of peripheral grating resolution acuity.

**METHODS.** Achromatic spatial summation functions were plotted for 68 healthy subjects (aged 20–77 years) in four oblique meridians on a gray background field of 10 cd/m<sup>2</sup>. Similar functions were generated for the S-cone pathway (isolated using Stiles' two-color threshold method) for the same locations. Ricco's area was determined using two-phase regression analysis. Achromatic peripheral grating resolution acuity was measured at the same locations using high-contrast Gabor stimuli, as an estimate of localized functional ganglion cell density.

**RESULTS.** There was a notable decrease in overall contrast sensitivity with age for all stimulus sizes. However, there was no evidence of age-related change in Ricco's area for either achromatic (superior field,  $r^2 = 0.05$ ; inferior field,  $r^2 = 0.0007$ ; all  $P > 0.05$ ) or chromatic (superior field,  $r^2 = 0.01$ ; inferior field,  $r^2 = 0.006$ ; all  $P > 0.05$ ) stimuli, despite a significant decrease in peripheral grating resolution acuity with age (superior field,  $r^2 = 0.15$ ; inferior field,  $r^2 = 0.17$ ; both  $P < 0.05$ ).

**CONCLUSIONS.** An age-related decline in functional ganglion cell density is not accompanied by a significant change in Ricco's area for achromatic or chromatic stimuli. (*Invest Ophthalmol Vis Sci.* 2010;51:6533–6539) DOI:10.1167/iovs.10-5717

Ricco's law states that for a range of small stimuli projected on the retina, the total energy of the stimulus is constant at threshold.<sup>1</sup> Thus, at the limit of visual detection of such stimuli, the area ( $A$ ) and intensity ( $I$ ) of the stimulus are inversely

proportional ( $A \times I = k$ , where  $k = -1$ ). However, for larger stimuli, the law of complete summation does not hold, and only partial summation of signals occurs.<sup>2,3</sup> The largest stimulus size for which Ricco's law holds true is known as the area of complete spatial summation or, more commonly, Ricco's area.

Spatial summation has long been considered a mechanism for improving signal detection in the presence of noise; however, greater summation usually comes at the expense of reduced spatial resolution. Despite decades of classic studies on spatial summation, the exact physiological mechanism that dictates the size of Ricco's area is still a topic of intense debate. Various attempts to explain this phenomenon have resulted in different and often conflicting hypotheses.<sup>4–13</sup> When one considers that spatial summation becomes incomplete beyond a critical stimulus area, it is reasonable to speculate that some sort of spatial inhibitory mechanism might be initiated and that the limit of complete spatial summation might represent some physiological or anatomic limit. Indeed, a relationship has been shown between Ricco's area and changing adaptation levels that are known to be associated with lateral inhibition.<sup>6,14,15</sup> The size of Ricco's area is also influenced by visual field eccentricity,<sup>9,11,16–19</sup> stimulus duration,<sup>17,20</sup> and wavelength.<sup>5,9,21</sup> Significant previous work has led to the popular hypothesis that the initial transition between complete summation and partial summation results from inhibition at the retinal level,<sup>6,9,11,12,14,17</sup>; however, other works have concluded that changes in spatial summation can largely be accounted for by optical factors, at least in the fovea,<sup>7,22</sup> or by second-stage spatial filters at a higher processing site.<sup>10,13</sup> Other studies remain undecided about the physiological basis of Ricco's area but offer intriguing arguments for and against various stages in the visual pathway, from the preneural ocular structures to the visual cortex.<sup>8</sup>

In the past 20 years, many different retinal ganglion cell types have been described, and their physiological roles have been investigated at length (Dacey DM, et al. *IOVS* 2002;43:ARVO E-Abstract 2983).<sup>23–29</sup> Consequently, many studies have attempted to compare the size of Ricco's area to what we now know about the size and distribution of various cells in the retina in an effort to accept or reject some of the aforementioned hypotheses and to explore any neural contribution to the phenomenon. Volbrecht et al.<sup>9</sup> measured Ricco's area as a function of eccentricity under both L-cone and S-cone isolation conditions and found a stronger association between Ricco's area and ganglion cell density across the retina than between Ricco's area and the respective cone density for either pathway. These authors proposed that Ricco's area increased with eccentricity to maintain a constant number of underlying retinal ganglion cells. Vassilev et al.<sup>11</sup> found that, under selective S-cone conditions, Ricco's area was closely associated with the size of the dendritic field of the small bistratified cell with increasing retinal eccentricity. These studies lend attractive suggestions toward an explanation of the finding of Wilson<sup>17</sup>

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Presented in part at the annual meeting of the Association for Research in Vision and Ophthalmology, Fort Lauderdale, Florida, May 2008.

Supported by a PhD studentship from the Department of Employment and Learning, Northern Ireland (TR) and by the NIHR Biomedical Research Centre for Ophthalmology, Moorfields Eye Hospital NHS Foundation Trust and UCL Institute of Ophthalmology (TR).

Submitted for publication April 15, 2010; revised June 14, 2010; accepted July 7, 2010.

Disclosure: **T. Redmond**, None; **M.B. Zlatkova**, None; **D.F. Garway-Heath**, None; **R.S. Anderson**, None

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that, though Ricco's area increased monotonically with retinal eccentricity, the increment threshold for a stimulus of the same size as Ricco's area remained constant. Although it is tempting to conclude that the change in Ricco's area with eccentricity is related to the changing density or distribution of retinal cells, it must be borne in mind that the size of Ricco's area can be altered by modifying the temporal profile of the stimulus and the background adaptation level. Thus, Ricco's area may be determined not only by the density of the underlying cells but also by the adaptation features of the responding cell type, the higher-level neurons to which they project, or both.

Using imaging and psychophysical techniques as well as anatomic counts, several previous cross-sectional studies<sup>30-34</sup> have indicated that the number of ganglion cells and their axons in the human retina is associated with age. This has also been observed in two longitudinal imaging studies.<sup>35,36</sup> Since the number of responding ganglion cells underlying Ricco's area appears to be constant under constant adaptive conditions, at least with eccentricity, one might assume that the critical summation area would be larger in older subjects, in parallel with the age-related reduced ganglion cell number to encompass a constant number of remaining cells. One might also presume a change in Ricco's area with pathologic ganglion cell loss (see accompanying paper<sup>37</sup>).

Dannheim and Drance<sup>38</sup> measured detection thresholds for a range of differently sized achromatic stimuli and constructed spatial summation functions. Although they concluded that thresholds for all stimuli were certainly elevated in older subjects and that the shape of the summation function did not change, they did not specifically determine a parameter for Ricco's area. Brown et al.<sup>39</sup> noted no significant change in critical summation areas with age. However, this study investigated spatial summation in only a small number of subjects (9 young and 10 elderly observers), and it is now known that sizable interindividual differences occur in the size of Ricco's area.<sup>8</sup> Latham et al.<sup>40</sup> demonstrated that, except for a reduced sensitivity to the largest stimuli compared with the smallest stimuli in older observers, summation curves for younger and older observers could be superimposed neatly. The only study to date that has purposefully investigated age-related changes in Ricco's area, under consideration of Ricco's law and in a relatively large number of subjects, is that of Scheffrin et al.<sup>8</sup> This study was performed under scotopic conditions, in which the signal response is driven by rod photoreceptors. The authors found that Ricco's area increased linearly over their range of subject ages and that, even though statistical significance was reached, the relationship between Ricco's area and age was weak. By considering optical, retinal, and cortical factors that might affect their spatial summation functions, various mechanisms were proposed by the authors for such a change in Ricco's area. The authors concluded that the most likely explanation for the changes they found in Ricco's area was retinal rewiring (i.e., a greater convergence of photoreceptor signals on the remaining ganglion cells) in response to age-related decline in ganglion cell density.

The purpose of the present study was threefold. First, we sought to establish the association between Ricco's area and age under photopic and S-cone conditions. Second, we wanted to compare the values of achromatic Ricco's area with the underlying ganglion cell density estimated by achromatic peripheral grating resolution acuity (see Ref. 41 for a major review) for each subject.<sup>42</sup> If the size of Ricco's area is indeed determined, in part, by the underlying retinal ganglion cell density, one might expect both estimates to change together with increasing age. Third, knowledge of the nature of any change with age in Ricco's area will aid in the interpretation of

pathologic findings, as has been suggested may occur in early glaucoma<sup>12</sup> and as investigated in the accompanying paper.<sup>37</sup>

In light of the available literature about the way in which Ricco's area changes under various adaptive and physiological conditions, we might have expected, in this study, 1 of 3 possible outcomes. The first possibility was that an entirely upward shift of the spatial summation curve, with no accompanying change in Ricco's area, might be observed with a uniform loss of sensitivity across all stimulus sizes. The second possibility was that we might observe an entirely rightward shift of the spatial summation curve with no upward shift, indicating a change in Ricco's area but no overall loss of sensitivity. Such a finding may accompany age-related ganglion cell loss. The third possibility was a mixed upward *and* rightward shift of the spatial summation curve. In this instance, an enlargement in Ricco's area would be observed along with an elevated threshold for Ricco's area.

## SUBJECTS AND METHODS

### Subjects

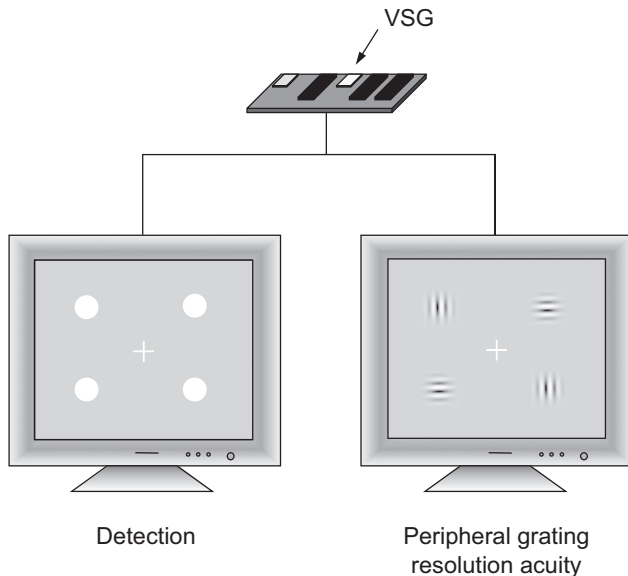
Sixty-eight healthy white European subjects (mean age, 43 years; range, 20-77 years), were tested. Of these subjects, 47 were tested at the University of Ulster (Coleraine, Northern Ireland, UK), and 21 were tested at Moorfields Eye Hospital (London, UK). Each had a refractive error  $< \pm 6.00$  D in any meridian with astigmatism  $< 1.25$  D, and each achieved a best-corrected visual acuity of 6/9 or better. All had clear media. Optic nerve head rim area measurements were classified as within normal limits for all observers by Moorfields Regression Analysis (Heidelberg Retina Tomograph II; Heidelberg Engineering, Heidelberg, Germany). No subject had any other abnormal ocular or systemic condition that was considered to affect visual performance. A conventional visual field test (SITA 24-2 strategy) was performed twice for each subject (Humphrey HFAII; Zeiss Meditec, Dublin, CA). All subjects demonstrated reliable results on both occasions, and none had any visual field defects.

For the experimental tests, subjects underwent achromatic and chromatic contrast detection tests using six circular test stimuli and a peripheral grating resolution acuity test as described. Recruitment of patients and subjects adhered to the tenets of the Declaration of Helsinki.

### Apparatus and Stimuli

All stimuli were generated on either a  $\gamma$ -corrected 21-inch RGB monitor (GDM-500PST; Sony Corp., Tokyo, Japan; pixel resolution, 1280  $\times$  965; frame rate, 73 Hz) or a  $\gamma$ -corrected 21-inch grayscale monitor (Phillips Fimi MGD-403; Ampronix, Irvine, CA; pixel resolution, 1280  $\times$  965; frame rate, 73 Hz) using a visual stimulus generator card (Cambridge Research Systems, Rochester, UK) and software (Psycho v2.0; Cambridge Research Systems). For achromatic tests, stimuli were superimposed on a uniform gray background with a luminance of 10 cd/m<sup>2</sup>. The maximum available luminance of test stimuli was 273 cd/m<sup>2</sup>. Chromaticity coordinates of the background central-fixation cross and stimuli were  $x = 0.218$  and  $y = 0.328$  measured with a spectrophotometer (Spectrascan PR-650 Spectra Colorimeter; Photo Research Inc., Chatsworth, CA). The viewing distance for the achromatic tests was 102 cm. To plot achromatic spatial summation functions, thresholds were measured for six circular incremental stimuli in four oblique retinal locations at 10° eccentricity (as shown in Fig. 1), ranging in size between 0.01 and 2.67 deg<sup>2</sup> (achromatic) and 0.03 and 4.74 deg<sup>2</sup> (chromatic). Stimulus duration was 200 ms with a square temporal profile. The meridians chosen for both contrast sensitivity tests were 36°, 144°, 216°, and 324°.

For the chromatic tests, the S-cone pathway was isolated using Stiles' two-color threshold technique.<sup>43,44</sup> The yellow light was generated using a slide projector fitted with an OG530 long-wavelength pass filter. Light was projected from below through a polymethyl methac-



**FIGURE 1.** Schematic diagram of apparatus for achromatic detection tasks (*left monitor*) and resolution acuity task (*right monitor*). All four stimuli are shown simultaneously on each monitor for the purposes of illustration. For the chromatic detection task, stimuli were arranged as in the *left monitor* but with a black background; the monitor was viewed through a semi-silvered mirror, and the yellow adapting field was directed into the eye from a projector below.

rylate (Perspex; Lucite, Southampton, UK) diffusing screen and was reflected into the eye with a large 30% reflection semi-silvered mirror inclined at 45°. The luminance of the resultant yellow light was 600 cd/m<sup>2</sup>, with chromaticity coordinates of  $x = 0.521$ ,  $y = 0.474$ . Subjects viewed the computer monitor, which was placed behind the mirror. Blue circular test stimuli of different sizes were presented to the same four retinal locations as the achromatic stimuli, and thresholds were determined accordingly. Stimuli were presented on a uniform black background on the Sony monitor. The maximum luminance of blue stimuli was 6 cd/m<sup>2</sup>, with chromaticity coordinates  $x = 0.151$  and  $y = 0.070$ . The OG530 yellow filter was chosen to isolate the S-cones because of its efficacy at maximally stimulating, and thus adapting, the L- and M-cones. However, it still causes some slight S-cone excitation; thus, increment contrast thresholds ( $\Delta I/I$ ) were calculated. The fixation target consisted of two blue squares ( $0.4^\circ \times 0.4^\circ$ ) separated vertically by  $0.2^\circ$  in the center of the monitor. Subjects were asked to fixate the gap between the two squares during the experiment. The viewing distance for chromatic tests was 60 cm.

Achromatic peripheral grating resolution acuity was investigated under the same viewing conditions as the achromatic contrast detection task. Static sinusoidal grating stimuli (diameter,  $3^\circ$ ; contrast, 90%) that had the same mean luminance as the background were presented within a Gaussian window (SD, 1.5) at the same four retinal locations as the detection tasks. The gratings appeared either vertical or horizontal at random. The achromatic background was identical with that used in the achromatic detection experiment. Stimuli were presented for 1 second, including a 300 ms stimulus onset and 300 ms decay.

The actual sizes of all stimuli were measured by hand from the screen using a graticule. Nominal contrast levels were also regularly checked by direct measurements from the monitor (the stimulus and background were arranged as a bipartite scene for calibration) using a photometer (OptiCal; Cambridge Research Systems, Rochester, UK). Results were corrected accordingly.

### Correction of Refractive Error

Objective (retinoscopy) and subjective refraction were initially performed foveally at 6 m for all subjects. Refractive correction was worn

for each psychophysical test where necessary, corrected according to the particular experimental conditions. For the preliminary standard automated perimetry (SAP) test (Humphrey HFAII; Zeiss Meditec), the appropriate refractive correction was determined by the perimeter software and was incorporated accordingly. For both the chromatic and the achromatic tests, correction was subjectively refined for the appropriate working distance and for the peripheral test locations using a high spatial frequency grating target in the plane of the test stimulus. Full-aperture trial lenses were introduced to find the optimum correction for the task.<sup>45</sup> Chromatic refraction typically resulted in a correction approximately 1 D more myopic than achromatic refraction because of longitudinal chromatic aberration.

### Psychophysical Procedure

Chromatic and achromatic experiments were performed in a random order and on separate days; however, the achromatic peripheral grating resolution test was carried out on the same day as the achromatic detection test, but these were conducted in random order. One drop of tropicamide hydrochloride (1%) was instilled in the test eye before the experiments. When mydriasis (pupil  $\geq 8$  mm) was achieved, each subject was asked to place his or her chin on a chinrest and head against a headrest while looking straight ahead at the central fixation target. Subjects adapted to the achromatic background for 1 minute before commencement of the achromatic tests. Detection thresholds were measured (in random order) for six different spot stimulus sizes, each in a separate run. Within each run, all four retinal locations were tested in an interleaved fashion. A Yes/No procedure and a best-PEST adaptive thresholding algorithm<sup>46</sup> were used to determine threshold for each stimulus. Stimuli with 0% contrast were occasionally presented at the same loci to estimate the false-positive rate. The false-negative rate was assessed by presenting stimuli with a contrast level fixed sufficiently above the average threshold expected for their age group, with consideration given to the stimulus size. Subjects were asked to press one button if they were aware of a stimulus alongside the simultaneous audio signal or a second button if they were unaware of a stimulus. Thresholds were recorded by the software once the confidence level exceeded 50%. An identical psychophysical procedure was used for the chromatic tests; however, before these tests began, subjects underwent dark adaptation for 10 minutes and 3 minutes of adaptation to the yellow S-cone isolation field.

A spatial two-alternative forced choice strategy was used for the achromatic peripheral grating resolution acuity test. Again, subjects were asked to fixate a central target while gratings were presented to the same four locations. The test was administered as an orientation discrimination task in which subjects were required to press 1 of 2 buttons to indicate whether they perceived the peripherally viewed grating to be oriented vertically or horizontally. The spatial frequency increased by 10% after three correct responses and decreased by 10% after one incorrect response. Threshold values were determined as the average of four reversals. Checks for false-negative responses were also used in this test. Fixation was monitored visually during all tests. All subjects underwent a practice run on each test before the commencement of each test. Tests were performed only after subjects indicated that they fully understood the procedure.

### Statistical Analysis

Thresholds for each stimulus size were initially averaged per hemifield (i.e., superior and inferior) for each observer, and spatial summation functions were constructed (two averaged functions per person). Two-phase regression analysis<sup>47</sup> (Levenberg-Marquardt estimation) was performed on each function using SPSS (v15.0; SPSS Inc., Chicago, IL) to determine a value for Ricco's area. For the purposes of this analysis, the slope of the first line was constrained to a value of  $-1$  in accordance with Ricco's law, whereas the slope of the second line and the breakpoint were allowed to vary. The estimated breakpoint was taken to represent Ricco's area. Estimates of Ricco's area were determined for each hemifield (superior and inferior) in each subject. Estimates were

excluded from further analysis if the bilinear model did not fit the data well ( $r^2 < 0.9$ , for the purposes of this study) or if the statistical program could not perform a bilinear fit of the data because of an atypical spread of data points. Of the 136 summation curves generated across all subjects (68 superior and 68 inferior), six functions (four superior, two inferior) were excluded from the achromatic data and 20 functions (10 superior, 10 inferior) were excluded from the chromatic data. Subjects were also divided into five groups, based on age. Thresholds for each stimulus size were averaged for each hemifield across observers in each group, and spatial summation functions were constructed accordingly. Detection of the smallest chromatic stimulus proved difficult for the older subjects, and several of them could not detect it at maximum contrast (i.e., they demonstrated a *ceiling* effect). Consequently, for these particular observers, data for the smallest stimulus were considered unrepresentative of true contrast sensitivity and were removed from the spatial summation curves. These data were also removed from the averaged curve for the oldest group.

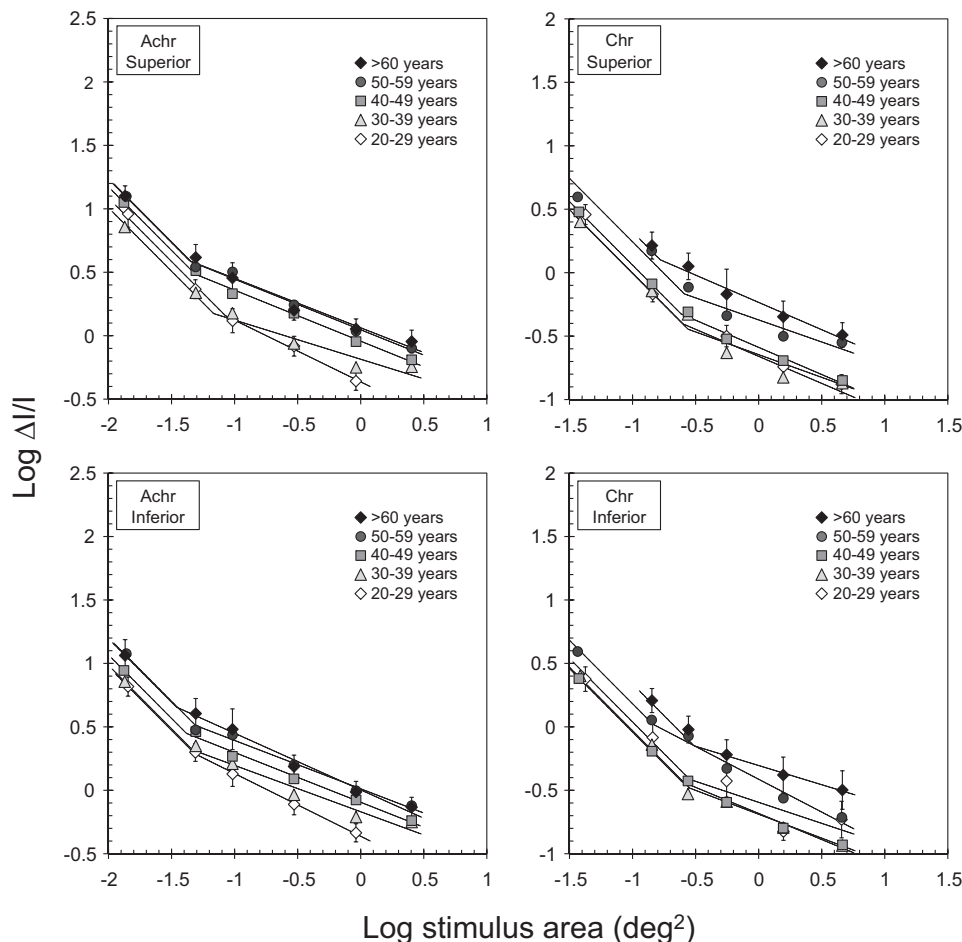
## RESULTS

Average summation curves for each group and under each stimulus condition for the superior and inferior hemifields are plotted along with two-phase regression lines in Figure 2. Our results show an age-related decline in overall sensitivity for each stimulus size for both the achromatic and the chromatic pathways. For achromatic stimuli, the mean threshold for the smallest stimulus for older observers (older than 60 years; mean age, 67 years; range, 60–77 years) was 0.19 log units higher than the mean threshold for our youngest observers (mean age, 22 years; range, 20–29 years). For the largest

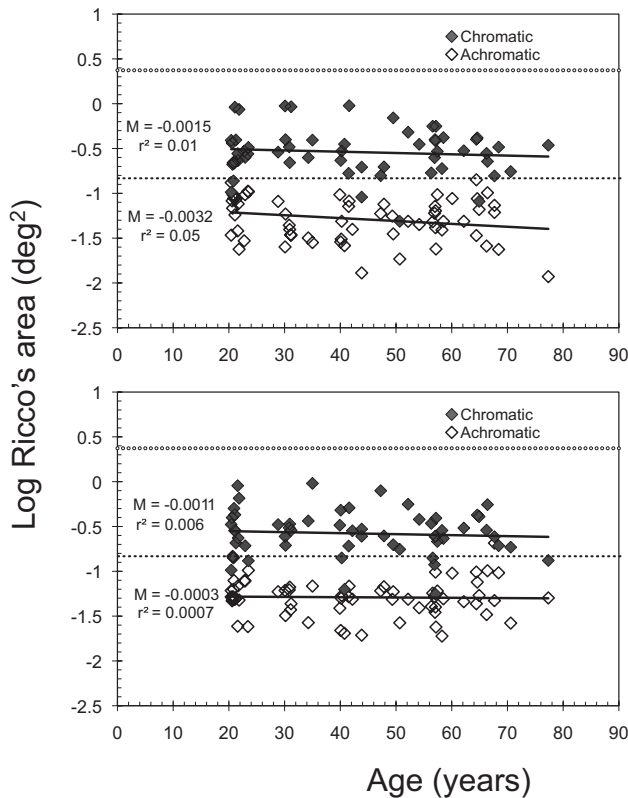
stimulus, this difference increased to 0.37 log units. For the S-cone pathway, the difference in threshold between the oldest and youngest groups was 0.25 log units, and between the smallest and largest stimulus it was 0.30 log units, with the older group again demonstrating higher threshold values than the younger group. An independent-samples *t*-test performed on data for the largest and smallest stimuli revealed significant differences between mean thresholds for the youngest and oldest observers under both achromatic and selective S-cone conditions (all  $P < 0.01$ ). It is also apparent from this initial analysis that there is no notable change in Ricco's area as a function of age for either stimulus type.

Ricco's area for each observer is plotted as a function of age in Figure 3. In agreement with previous studies,<sup>5,9,48</sup> Ricco's area estimates were overall larger for chromatic than for achromatic stimuli. We found no significant association between the size of Ricco's area with age for either achromatic stimuli (superior field,  $r^2 = 0.05$ ; inferior field,  $r^2 = 0.0007$ ; both  $P > 0.05$  for  $r^2$  values) or chromatic stimuli (superior field,  $r^2 = 0.01$ ; inferior field,  $r^2 = 0.006$ ; both  $P > 0.05$  for  $r^2$  values). A paired *t*-test on all Ricco's area data for achromatic and chromatic stimuli showed no significant hemifield difference in Ricco's area for our subjects (achromatic,  $P = 0.52$ ; chromatic,  $P = 0.58$ ). Separate paired *t*-tests on estimates of Ricco's area for the youngest observers (20–29 years) and for the oldest observers (older than 60 years) revealed no significant hemifield difference in either group.

The effect of age on achromatic peripheral grating resolution acuity is shown in Figure 4. Linear regression revealed a significant age-related decline in peripheral grating resolution



**FIGURE 2.** Average spatial summation curves for each age group. *Left:* results from the achromatic tests. *Right:* results from the chromatic tests. *Top:* superior field. *Bottom:* inferior field. Error bars are shown for the youngest and oldest group and represent the 95% confidence intervals for each averaged point.



**FIGURE 3.** The effect of age on Ricco's area for chromatic and achromatic stimuli in both the superior hemifield (*top*) and the inferior hemifield (*bottom*). The broken lines at  $y = -0.83$  and  $y = 0.37$  represent the size of a Goldmann III target (used in SAP) and a Goldmann V target (used in SWAP), respectively.

acuity in both hemifields (superior,  $r^2 = 0.15$ ; inferior,  $r^2 = 0.17$ ; both  $P < 0.05$ ), with the slope indicating a change of 0.24 and 0.28 cyc/deg per decade for the superior field and the inferior field, respectively. However, these hemifield differences were not significant ( $P = 0.5$ , paired  $t$ -test).

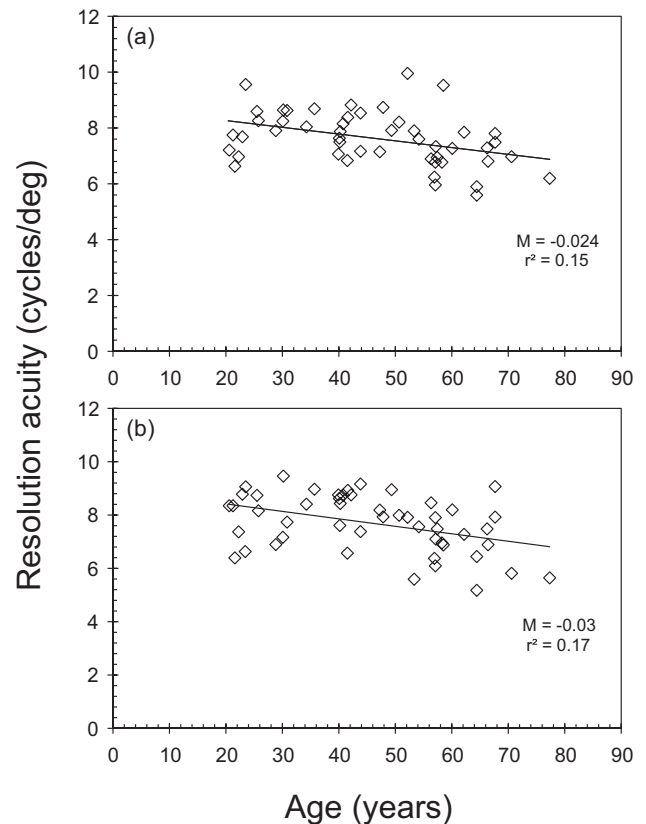
## DISCUSSION

In accordance with previous studies,<sup>30,33,34</sup> we found an age-related decline in achromatic peripheral grating resolution acuity and, hence, ganglion cell sampling density at the different retinal locations tested. However, despite this decline in resolution acuity, we found no evidence for an accompanying change in Ricco's area for either stimulus type. Since the exact physiological basis for Ricco's area has yet to be determined, an explanation of these findings requires consideration of age-related changes at various sites along the visual pathway (both optical and neural). Normal aging in the eye is accompanied by both a reduction in clarity of the crystalline lens<sup>49–51</sup> and a reduction in the population of retinal ganglion cells and their axons.<sup>31,32,52–55</sup>

It is interesting that, given the numerous well-known age-related changes in the structure and function of the visual system, the fundamental function of spatial summation should remain unchanged into advancing years. It is tempting to report that our findings are similar to those of other studies that measured spatial summation as a function of age<sup>38,40</sup>; however, it is important to bear in mind that differences in experimental conditions (e.g., adaptation level and the statistical models used to describe the data) somewhat prohibit direct comparison between studies. It is reasonable to suggest that either (or

both) of these factors could partially explain differences in findings between studies of spatial summation. Indeed, though these differences limit comparison, they may aid in the interpretation of different results for different visual pathways. Although a numerical difference between thresholds for large and small stimuli allows us to make broad assumptions about the amount of spatial summation that has occurred between these stimuli, this analysis affords little information about the *nature* of summation and the point at which summation becomes incomplete (Ricco's area). Furthermore, though previously published polynomial models satisfactorily describe spatial summation data,<sup>38,40</sup> they do not take into account the classic notion of complete spatial summation for small stimuli. Here we have used a two-phase regression model for each data set, similar to that used by Scheffrin et al.<sup>8</sup> This model might also be of limited use if one were to describe spatial summation across larger stimulus sizes, for which the probability of summation may approach zero; however, for the range of stimulus sizes used in the present study, the model is sufficient while taking into account Ricco's law.

It is clear that Ricco's area has a very strong neural component because it is highly affected by factors such as stimulus duration and background adaptation. However, it is also apparent that optical factors may make a contribution, primarily because of widening of the point-spread-function (PSF) and forward light scatter. By comparing the performance of live subjects and an ideal observer model, Davila and Geisler<sup>7</sup> demonstrated that optical factors can account for most, if not all, of the phenomenon of Ricco's area in the fovea. However, in a recent adaptive optics study in young subjects, Dalimier and Dainty<sup>22</sup> showed that though the foveal Ricco's area estimate is diminished in size, it is not eliminated by the correction of optical aberrations; an area of complete spatial summation is



**FIGURE 4.** The effect of age on achromatic peripheral grating resolution acuity in (a) the superior hemifield and (b) the inferior hemifield.

still observed, demonstrating that Ricco's area has a neural component. The low-pass effect of young optics on the smallest stimuli would result in a steepening of the left limb of the spatial summation function. When a subsequent two-phase regression model is fitted to these data and the slope of the first line is constrained to  $-1$ , this will result in an *apparently* larger Ricco's area without adaptive optics. In the peripheral retina, as tested in the experiments described here, however, in which Ricco's area and the stimuli to measure it are much larger, any low-pass effect of optics would be much smaller. Artal et al.<sup>56</sup> measured the change in the optical modulation transfer function (MTF) with age and found the greatest deterioration at low spatial frequencies. It would follow, therefore, that, in the present study, thresholds for our larger stimuli should be disproportionately affected by aging optics, resulting in an effect opposite to that shown by Dalimer and Dainty, with an apparent decrease in Ricco's area. As a control experiment, Scheffrin et al.<sup>8</sup> modeled the effects of increased intraocular straylight and reduced MTF on their spatial summation data. They concluded that though these factors caused an overall elevation in stimulus detection threshold, neither had a significant effect on the Ricco's area estimate in their model. Although this kind of modeling at the *retinal* level might lead one to conclude that there is no effect of reduced optical quality on Ricco's area, the same cannot be concluded from our data, for which spatial summation curves were plotted using measurements of stimulus size from the monitor. Considering an enlarged PSF resulting from increased wide-angle scatter, one might appreciate that because the spatial extent is larger, Ricco's area is "filled" sooner than if it were not affected. The amount of luminous flux within this area would govern threshold. If this is correct, one might expect an *apparent* reduction in Ricco's area under such conditions. In the present study, it is reasonable to suggest therefore that an increased Ricco's area is masked by an artifactual decrease as a result of age-related optical change.

Another attribute of reduced optical quality in older persons is increased lens brunescence. The effect of lens brunescence on the detection of achromatic Goldman III spot stimuli (area,  $-0.83 \log \text{deg}^2$ ) used in conventional perimetry has been reported as negligible (McDowell DR, et al. *IOVS* 2005; 46:ARVO E-Abstract 710).<sup>57</sup> However, many reports detail the effects of lens yellowing on thresholds for larger (Goldmann V stimulus; area,  $0.37 \log \text{deg}^2$ ) chromatic stimuli used in short-wavelength automated perimetry<sup>58,59</sup> and peripheral grating resolution acuity.<sup>60</sup> One might, therefore, expect a greater age-related change in threshold (upward shift of the curves) for chromatic stimuli than for achromatic stimuli because of selective absorption of blue light by the yellow crystalline lens.

If changes in Ricco's area occur to maintain a constant number of underlying responsive ganglion cells at any one time,<sup>9,11</sup> one would expect an enlargement of Ricco's area because of age-related ganglion cell loss. Estimates of underlying ganglion cell density (per  $\text{mm}^2$ ) were obtained from individual resolution acuity values in each hemifield of each subject using the conversion algorithm of Thibos et al.<sup>61</sup> and assuming a hexagonal array of ganglion cells. Linear least squares regression indicates that our indirect estimate of ganglion cell density declines at a rate of 0.02 and 0.03 log units per decade for the superior and inferior fields, respectively. One may initially assume that Ricco's area enlarges by the same amount in the absence of optical influence. Given these small numbers, it is reasonable to speculate that even small reductions in the optical MTF might confound any measurable change in Ricco's area with age.

Regardless of the effect of neural loss or declining optics on Ricco's area, in a clinical setting, an age-related change in Ricco's area may not be evident. Other factors may also mask

any observable change in Ricco's area with age. The data presented here are cross-sectional; therefore, high between-subject variability (as seen in Fig. 3) could potentially mask any subtle changes in Ricco's area that might occur. Another factor that could potentially add to the observed variability is measurement noise. Two-phase regression analysis was used to determine estimates of Ricco's area based on six stimuli. Although up to 50 iterations were used to determine breakpoint values (representing Ricco's area), these are, nonetheless, estimates based on a small number of test points. We are confident that our Ricco's area values approximate the true value given the regression coefficients of the nonlinear functions.

Ocular pathology, particularly if characterized by a greater loss of retinal ganglion cells than that observable as a function of normal aging, may afford a larger dynamic range over which Ricco's area may change. The accompanying paper investigates changes in Ricco's area in glaucoma.<sup>37</sup>

### Acknowledgments

The authors thank David P. Crabb for statistical advice.

### References

1. Ricco A. Relazione fra il minimo angolo visuale e l'intensità luminosa. *Memorie della Regia Accademia di Scienze, lettere ed arti in Modena*. 1877;17:47-160.
2. Piper H. Über die Abhängigkeit des Reizwertes leuchtender Objekte von ihre Flächen-bezw. Winkelgrasse. *Zeitschrift für Psychologie und Physiologie der Sinnesorgane*. 1903;32:98-112.
3. Kleitman N, Pieron H. Contribution à l'étude des facteurs régissant le taux de summation des impressions lumineuses de surface inégale. *L'année psychologique*. 1928;29:57-91.
4. Hartline HK. The effects of spatial summation in the retina on excitation of the fibers of the optic nerve. *Am J Physiol*. 1940;130:700-711.
5. Brindley GS. The summation areas of human colour-receptive mechanisms at increment threshold. *J Physiol*. 1954;124:400-408.
6. Glezer VD. The receptive fields of the retina. *Vision Res*. 1965;5:497-525.
7. Davila KD, Geisler WS. The relative contributions of pre-neural and neural factors to areal summation in the fovea. *Vision Res*. 1991; 31:1369-1380.
8. Scheffrin BE, Bieber ML, McLean R, Werner JS. The area of complete scotopic spatial summation enlarges with age. *J Opt Soc Am A Opt Image Sci Vis*. 1998;15:340-348.
9. Volbrecht VJ, Shrago EE, Scheffrin BE, Werner JS. Spatial summation in human cone mechanisms from 0 degrees to 20 degrees in the superior retina. *J Opt Soc Am A Opt Image Sci Vis*. 2000;17: 641-650.
10. Swanson WH, Feliuss J, Pan F. Perimetric defects and ganglion cell damage: interpreting linear relations using a two-stage neural model. *Invest Ophthalmol Vis Sci*. 2004;45:466-472.
11. Vassilev A, Ivanov I, Zlatkova MB, Anderson RS. Human S-cone vision: relationship between perceptible field and ganglion cell dendritic field. *J Vis*. 2005;5:823-833.
12. Anderson RS. The psychophysics of glaucoma: improving the structure/function relationship. *Prog Retin Eye Res*. 2006;25: 79-97.
13. Pan F, Swanson WH. A cortical pooling model of spatial summation for perimetric stimuli. *J Vis*. 2006;6:1159-1171.
14. Barlow HB. Temporal and spatial summation in human vision at different background intensities. *J Physiol*. 1958;141:337-350.
15. Lelkens AM, Zuidema P. Increment thresholds with various low background intensities at different locations in the peripheral retina. *J Opt Soc Am*. 1983;73:1372-1378.
16. Graham CH, Bartlett NR. The relation of size of stimulus and intensity in the human eye, II: intensity thresholds for red and violet light. *J Exptl Psychol*. 1939;24:574-587.
17. Wilson ME. Invariant features of spatial summation with changing locus in the visual field. *J Physiol*. 1970;207:611-622.

18. Scholtes AM, Bouman MA. Psychophysical experiments on spatial summation at threshold level of the human peripheral retina. *Vision Res.* 1977;17:867-873.
19. Vassilev A, Mihaylova MS, Racheva K, Zlatkova M, Anderson RS. Spatial summation of S-cone ON and OFF signals: effects of retinal eccentricity. *Vision Res.* 2003;43:2875-2884.
20. Hood DC, Finkelstein MA. Sensitivity to light. In: Boff KR, Kaufman L, Thomas JP, eds. *Handbook of Perception and Human Performance, I: Sensory Processes and Perception*. New York: John Wiley; 1986:5.1-5.66.
21. King-Smith PE, Carden D. Luminance and opponent-color contributions to visual detection and adaptation and to temporal and spatial integration. *J Opt Soc Am.* 1976;66:709-717.
22. Dalimier E, Dainty C. Role of ocular aberrations in photopic spatial summation in the fovea. *Opt Lett.* 2010;35:589-591.
23. Curcio CA, Allen KA. Topography of ganglion cells in human retina. *J Comp Neurol.* 1990;300:5-25.
24. Dacey DM. The mosaic of midget ganglion cells in the human retina. *J Neurosci.* 1993;13:5334-5355.
25. Dacey DM. Morphology of a small-field bistratified ganglion cell type in the macaque and human retina. *Vis Neurosci.* 1993;10:1081-1098.
26. Dacey DM, Lee BB. The 'blue-on' opponent pathway in primate retina originates from a distinct bistratified ganglion cell type. *Nature.* 1994;367:731-735.
27. Masland RH. The fundamental plan of the retina. *Nat Neurosci.* 2001;4:877-886.
28. Dacey DM, Packer OS. Colour coding in the primate retina: diverse cell types and cone-specific circuitry. *Curr Opin Neurobiol.* 2003;13:421-427.
29. Lee BB. Paths to colour in the retina. *Clin Exp Optom.* 2004;87:239-248.
30. Anderson RS, McDowell DR. Peripheral resolution using stationary and flickering gratings: the effects of age. *Curr Eye Res.* 1997;16:1209-1214.
31. Kerrigan-Baumrind LA, Quigley HA, Pease ME, Kerrigan DF, Mitchell RS. Number of ganglion cells in glaucoma eyes compared with threshold visual field tests in the same persons. *Invest Ophthalmol Vis Sci.* 2000;41:741-748.
32. Alamouti B, Funk J. Retinal thickness decreases with age: an OCT study. *Br J Ophthalmol.* 2003;87:899-901.
33. Zlatkova MB, Coulter E, Anderson RS. Short-wavelength acuity: blue-yellow and achromatic resolution loss with age. *Vision Res.* 2003;43:109-115.
34. Beirne RO, Zlatkova MB, Chang CK, Chakravarthy U, Anderson RS. How does the short-wavelength-sensitive contrast sensitivity function for detection and resolution change with age in the periphery? *Vision Res.* 2008;48:1894-1901.
35. Airaksinen PJ, Tuulonen A, Alanko HI. Rate and pattern of neuroretinal rim area decrease in ocular hypertension and glaucoma. *Arch Ophthalmol.* 1992;110:206-210.
36. See JL, Nicolela MT, Chauhan BC. Rates of neuroretinal rim and peripapillary atrophy area change: a comparative study of glaucoma patients and normal controls. *Ophthalmology.* 2009;116:840-847.
37. Redmond T, Garway-Heath DF, Zlatkova MB, Anderson RS. Sensitivity loss in early glaucoma can be mapped to an enlargement of the area of complete spatial summation. *Invest Ophthalmol Vis Sci.* 2010;51:6540-6548.
38. Dannheim F, Drance SM. Studies of spatial summation of central retinal areas in normal people of all ages. *Can J Ophthalmol.* 1971;6:311-319.
39. Brown B, Peterken C, Bowman KJ, Crassini B. Spatial summation in young and elderly observers. *Ophthalmic Physiol Opt.* 1989;9:310-313.
40. Latham K, Whitaker D, Wild JM. Spatial summation of the differential light threshold as a function of visual field location and age. *Ophthalmic Physiol Opt.* 1994;14:71-78.
41. Thibos LN. Acuity perimetry and the sampling theory of visual resolution. *Optom Vis Sci.* 1998;75:399-406.
42. Dacey DM. Primate retina: cell types, circuits and color opponency. *Prog Retin Eye Res.* 1999;18:737-763.
43. Stiles WS. Investigations of the scotopic and trichromatic mechanisms of vision by the two-colour threshold technique. *Arch Ophthalmol Rev Gen Ophthalmol.* 1949;28:215-237.
44. Stiles WS. Further studies of visual mechanisms by the two-colour threshold technique. *Coloquio sobre Problemas Opticas de la Vision.* 1953;1:65-103.
45. Wang YZ, Thibos LN, Lopez N, Salmon T, Bradley A. Subjective refraction of the peripheral field using contrast detection acuity. *J Am Optom Assoc.* 1996;67:584-589.
46. Pentland A. Maximum likelihood estimation: the best PEST. *Percept Psychophys.* 1980;28:377-379.
47. Seber GAF, Wild CJ. *Nonlinear Regression*. New York: John Wiley & Sons; 1989.
48. Felius J, Swanson WH, Fellman RL, Lynn JR, Starita RJ. Spatial summation for selected ganglion cell mosaics in patients with glaucoma. In: Wall M, Heijl A, eds. *Perimetry Update 1996/1997 Proceedings of the XIIIth International Perimetric Society Meeting*. New York: Kugler; 1997:213-221.
49. Norren DV, Vos JJ. Spectral transmission of the human ocular media. *Vision Res.* 1974;14:1237-1244.
50. Werner JS. Development of scotopic sensitivity and the absorption spectrum of the human ocular media. *J Opt Soc Am.* 1982;72:247-258.
51. Pokorny J, Smith VC, Lutze M. Aging of the human lens. *Appl Opt.* 1987;26:1437-1440.
52. Vrabcic F. Senile changes in the ganglion cells of the human retina. *Br J Ophthalmol.* 1965;49:561-572.
53. Balazsi AG, Rootman J, Drance SM, Schulzer M, Douglas GR. The effect of age on the nerve fiber population of the human optic nerve. *Am J Ophthalmol.* 1984;97:760-766.
54. Jonas JB, Muller-Bergh JA, Schlotzer-Schrehardt UM, Naumann GO. Histomorphometry of the human optic nerve. *Invest Ophthalmol Vis Sci.* 1990;31:736-744.
55. Parikh RS, Parikh SR, Sekhar GC, Prabakaran S, Babu JG, Thomas R. Normal age-related decay of retinal nerve fiber layer thickness. *Ophthalmology.* 2007;114:921-926.
56. Artal P, Ferro M, Miranda I, Navarro R. Effects of aging in retinal image quality. *J Opt Soc Am A.* 1993;10:1656-1662.
57. Johnson CA, Adams AJ, Adams CW, Lewis RA. Evidence for a neural basis of age-related visual field changes. In: *Digest of Topical Meeting on Noninvasive Assessment of the Visual System*. Washington, DC: Optical Society of America. 1988;44-47.
58. Johnson CA, Adams AJ, Twelker JD, Quigg JM. Age-related changes in the central visual field for short-wavelength-sensitive pathways. *J Opt Soc Am A.* 1988;5:2131-2139.
59. Sample PA, Martinez GA, Weinreb RN. Short-wavelength automated perimetry without lens density testing. *Am J Ophthalmol.* 1994;118:632-641.
60. Zlatkova MB, Coulter EE, Anderson RS. The effect of simulated lens yellowing and opacification on blue-on-yellow acuity and contrast sensitivity. *Vision Res.* 2006;46:2432-2442.
61. Thibos LN, Cheney FE, Walsh DJ. Retinal limits to the detection and resolution of gratings. *J Opt Soc Am A.* 1987;4:1524-1529.