ELLIPSOID ZONE MAPPING AND OUTER RETINAL ASSESSMENT IN STARGARDT DISEASE

SRUTHI AREPALLI, MD,* ELIAS I. TRABOULSI, MD, MED,* JUSTIS P. EHLERS, MD*†

Purpose: To quantify and correlate ellipsoid zone and photoreceptor outer segment changes with visual acuity in Stargardt disease.

Methods: An institutional review board–approved study of 32 eyes with Stargardt disease was performed. After spectral domain optical coherence tomography, the macular cube was exported into a novel analysis tool and volumetric assessment from the ellipsoid zone to the retinal pigment epithelium was performed. Using this information, mapping was completed with en face representation of the height between the ellipsoid zone and retinal pigment epithelium. This analysis provided quantification of ellipsoid zone and photoreceptor outer segments, including atrophy (ellipsoid zone to retinal pigment epithelium thickness = 0 μm) and attenuation (ellipsoid zone to retinal pigment epithelium thickness $<$20 μm). These parameters were compared with visual acuity and controls (n = 12 eyes).

Results: Visual acuity ranged from 20/30 to 20/250. The central foveal B-scan area of ellipsoid and photoreceptor outer segments was significantly less than controls (0.13 ± 0.05 mm^2 vs. 0.17 ± 0.03 mm^2, respectively, $P = 0.0074$). The central foveal B-scan mean thickness measured 22.52 ± 9.0 μm in Stargardt versus 30.0 ± 5.08 μm ($P = 0.0096$). Atrophy and attenuation were significantly higher in Stargardt patients (22% vs. 1%, $P = 0.005$ and 43% vs. 1%, $P = 0.0002$). Visual acuity directly correlated with ellipsoid zone/outer segment volume ($R = 0.57$, $P < 0.005$) and inversely correlated with attenuation and atrophy ($R = -0.53$ and -0.57; $P < 0.005$ for all).

Conclusion: Eyes with Stargardt disease frequently have significant disruption of the ellipsoid zone and outer segments. This degenerative change was successfully quantified with a novel assessment platform and identified correlates with visual function. This software provides the opportunity for quantitative assessment and possible longitudinal surveillance.

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Karl Stargardt[1] first described Stargardt disease in 1909. Characterized by macular atrophy, deep retinal yellow flecks, and progressive vision loss, it is the most common inherited juvenile macular dystrophy, occurring in 1 of 10,000 individuals.[2,3] Most commonly, the disease follows an autosomal recessive inheritance pattern with mutations in the gene *ABCA4*, which codes for a retina-specific ATP-binding cassette transporter, ABCR. This transporting molecule is integral to the movement of all-trans-retinol produced in rod outer segments. Dysfunction of this molecule leads to the toxic accumulation of lipofuscin in retinal pigment epithelial (RPE) cells.[4]

Interestingly, visual acuity in these patients is highly variable, ranging from 20/20 to light perception.[5] A number of studies have linked the decrease in visual acuity to inner and outer retinal changes.[5–7] Investigations into the retinal structure to explain the decline in vision have revealed loss of foveal photoreceptor and RPE layers, as well as disruptions in photoreceptor quantity.[8–14] In particular, various analyses reveal a decrease in the foveal ellipsoid zone and photoreceptor outer segments in Stargardt patients, indicated by ellipsoid zone disruption on spectral domain optical coherence tomography (SD-OCT).[9,10] Although these studies have demonstrated a qualitative decrease in these retinal layers, quantitative assessment of ellipsoid zone and outer segment loss remains uninvestigated. Ellipsoid zone mapping has been previously described in other macular disorders, including macular degeneration, hydroxychloroquine retinopathy, and ocriplasmin maculopathy.[15,16] The purpose of this study was to use a novel ellipsoid zone mapping platform to quantify and characterize the ellipsoidal and photoreceptor outer segment changes noted in Stargardt disease.

Methods

This institutional review board–approved retrospective case series included 32 eyes of 17 patients diagnosed with Stargardt disease between October 2012 and November 2014. Demographic and clinical data were collected through patient charts and included sex, age at presentation, visual acuity (VA, Snellen visual acuity was converted to logMAR for the purposes of statistical analysis), and visible macular pathology, including flecks, atrophy, and pigmentary changes. Inclusion criteria included an underlying diagnosis of Stargardt disease and SD-OCT macular cube data of sufficient quality for analysis.

Spectral domain optical coherence tomography images were reviewed for qualitative features including epiretinal membrane, atrophy, and fluid. In addition to qualitative review, macular SD-OCT cubes were exported for each patient into a novel ellipsoid zone and retinal layer mapping tool, as previously described.[15,16] This mapping tool allows for en face analysis and volumetric evaluation of the ellipsoid zone and the underlying photoreceptor outer segments as defined with OCT reflectivity patterns. The inner segmentation boundary was the ellipsoid zone and the outer segmentation boundary was the RPE. The automated mapping tool provides multiple quantitative outputs. The specific parameters examined in this report included the central foveal B-scan ellipsoid zone to the RPE mean thickness (defined as the mean distance between the ellipsoid zone and RPE on the horizontal B-scan that included the foveal pit), the central foveal B-scan ellipsoid zone to RPE (defined as area occupied by the ellipsoid zone and photoreceptor outer segments on the 6 mm horizontal B-scan that included the foveal pit), and the ellipsoid and photoreceptor outer segment macular volume (defined as the volume across the entire macular scan occupied by the space from the ellipsoid zone to the RPE). In addition, the en face ellipsoid zone map provided calculations related to the percentage of ellipsoid zone atrophy (i.e., complete loss of the ellipsoid zone with the measurement value of the ellipsoid zone to the RPE height of 0 μm) and attenuation (i.e., ellipsoid zone to RPE distance of $<$20 μm). These measurements were then compared with the normal ellipsoid zone and underlying photoreceptor outer segment measurements of 12 controls, as previously described.[15]

Results

Thirty-two eyes of 17 patients were included in this analysis. Ten of the 17 patients were men (59%). The mean age at presentation was 35.8 years (median, 36 years; range 11–65 years). Twenty eyes (63%) had macular flecks and 19 (59%) had clinical macular atrophy. Visual acuity ranged from 20/30 to 20/250.

Qualitative assessment of OCT scans identified epiretinal membranes in 3 of the 32 (9%), although one of these was very slight. Four eyes had intraretinal fluid (13%) and two had lamellar macular holes (6%). All 32 patients had subfoveal ellipsoid zone disruption, whereas 8 had extrafoveal atrophy (25%). Figure 1 displays fundus photographs, autofluorescence, and SD-OCT displaying the foveal ellipsoid zone and photoreceptor outer segment loss in a representative case.

In the Stargardt disease cohort, the ellipsoid zone and the photoreceptor outer segment central foveal B-scan area measured 0.13 ± 0.05 mm^2 compared with 0.17 ± 0.03 mm^2 (P = 0.0074) in the control group. The corresponding central foveal B-scan mean ellipsoid zone to RPE thickness was 22.52 ± 9.0 μm compared with 30.0 ± 5.08 μm (P = 0.0096) in controls. En face ellipsoid zone to RPE thickness maps were generated across the macular cube. These maps facilitated visualization of overall alterations, regional location, and severity of loss (Figures 2 and 3). The mean percentage of macular ellipsoid zone and outer segment attenuation was significantly higher in Stargardt disease eyes (43%) versus normal eyes ($<$1%, P = 0.0002). In addition, atrophy was significantly higher in Stargardt eyes compared with controls (22% vs. $<$1%, P = 0.0005). Visual acuity was inversely correlated with ellipsoid zone attenuation and atrophy (R = −0.53, P = 0.00061 and R = −0.57, P = 0.00162, respectively). In addition, ellipsoid zone and photoreceptor outer segment volume were directly correlated with visual acuity (R = 0.57, P = 0.00064).

From the *Cole Eye Institute, Cleveland Clinic, Cleveland, Ohio; and †Ophthalmic Imaging Center, Cleveland Clinic, Cleveland, Ohio.

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Reprint requests: Justis P. Ehlers, MD, Cole Eye Institute, Cleveland Clinic Foundation, 9500 Euclid Avenue/i32, Cleveland, OH 44195; e-mail: ehlersj@ccf.org

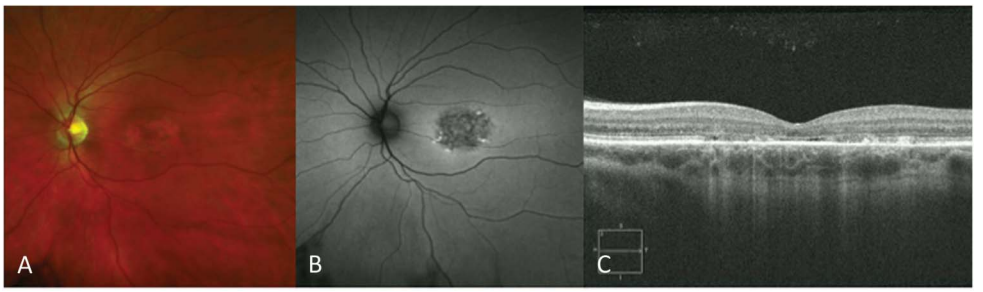

Fig. 1. Composite image of SD-OCT, fundus photograph, and fundus autofluorescence. **A.** Fundus photograph. **B.** Fundus autofluorescence with areas of macular atrophy. **C.** SD-OCT displaying foveal ellipsoid zone loss.

Discussion

Stargardt disease is most commonly characterized by juvenile onset macular degeneration linked to RPE toxicity from lipofuscin accumulation due to a defect in the gene *ABCA4*.[4] Histopathologic analysis confirms lipofuscin deposits in the foveal RPE with relative sparing of the periphery.[14] Early reports established the relationship between macular atrophy and vision loss, whereas later investigations identified the importance of ellipsoid zone preservation in maintaining visual acuity.[8–10,12,13] Confirmation of photoreceptor loss has been achieved with infrared scanning laser ophthalmoscopy and OCT.[5–10,12] Degeneration of the ellipsoid zone has been identified as the key feature of the Stargardt disease in previous reports, but quantitative assessment of these outer retinal alterations in Stargardt disease has not been assessed. We used a novel imaging analysis technique to provide both an en face representation of the ellipsoid zone and photoreceptor outer segments alterations and volumetric assessment of these layers in eyes of patients with Stargardt disease.

Greenstein et al[9] established a pattern of progressive retinal atrophy that progressed outward from

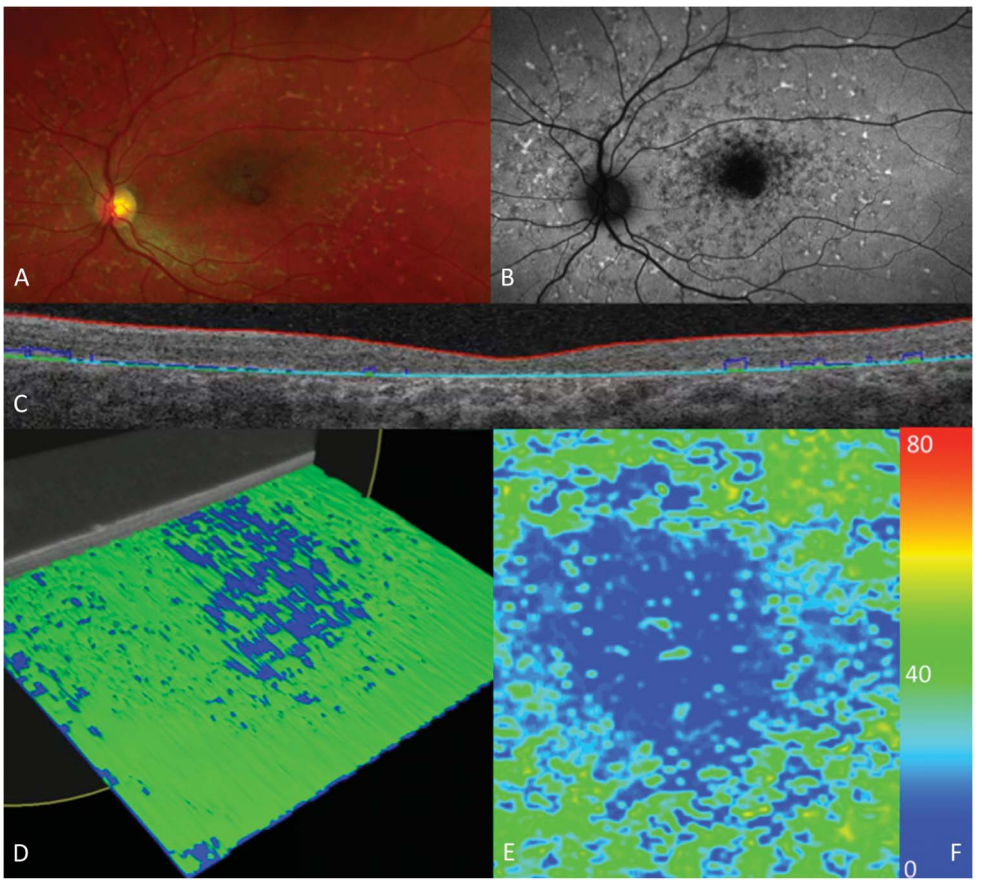

Fig. 2. Ellipsoid zone and photoreceptor outer segment mapping in a patient with moderate attenuation. **A.** Fundus photograph with macular changes. **B.** Fundus autofluorescence showing macular atrophy. **C.** Ellipsoid and photoreceptor outer segment segmentation of B-scan. **D.** Reconstruction of a three-dimensional macular cube with moderate loss. **E.** En face map with central areas of loss (blue) surrounded by normal thickness of the ellipsoid and photoreceptor outer segments (green). **F.** Color scale depicting the thickness range (in micrometers) visualized on an en face map, from atrophy (0 μm, dark blue) to 80 μm (red). Normal ellipsoid zone and photoreceptor outer segment thickness (40 μm) is green.

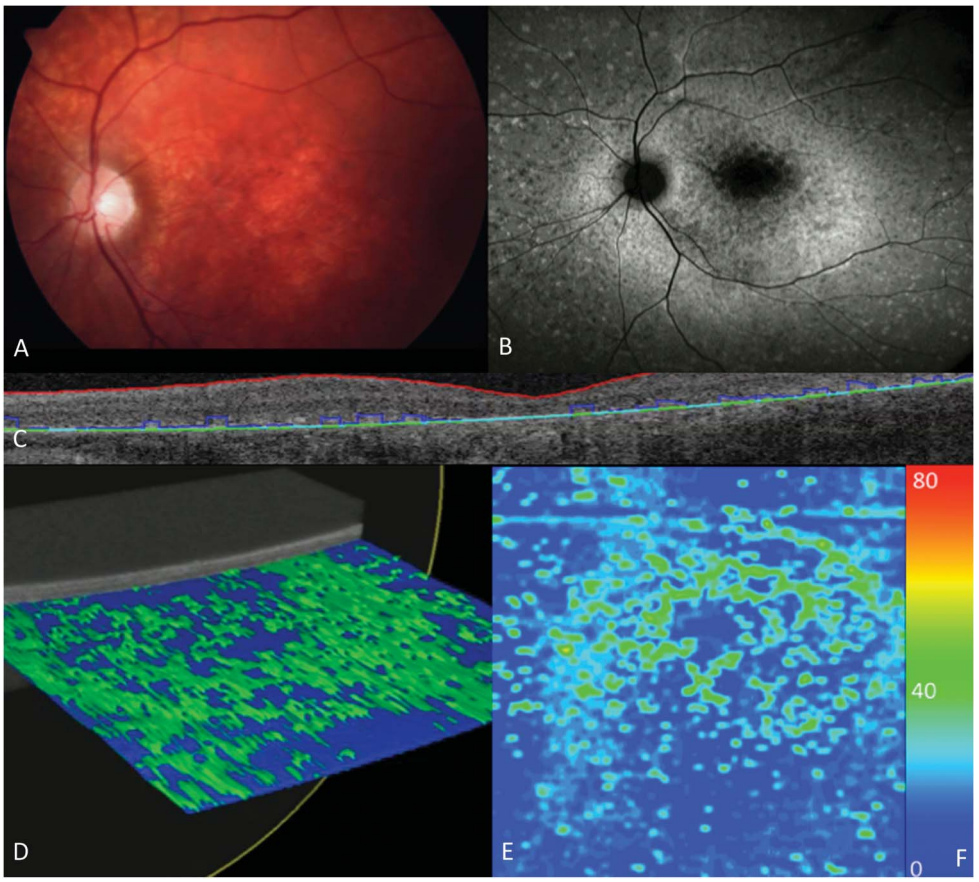

Fig. 3. Ellipsoid zone and photoreceptor outer segment mapping in a patient with severe attenuation. **A.** Fundus photograph with macular changes. **B.** Fundus autofluorescence showing macular atrophy. **C.** Ellipsoid zone and photoreceptor outer segment segmentation of B-scan. **D.** Reconstruction of a three-dimensional macular cube with severe loss. **E.** En face map with larger central areas of loss (blue), with small areas of normal (green). **F.** Color scale depicting the thickness range (in micrometers) visualized on an en face map, from atrophy (0 μm, dark blue) to 80 μm (red). Normal ellipsoid zone and photoreceptor outer segment thickness (40 μm) is green.

central foveal lesions in Stargardt disease. An additional report assessed the presence or absence of hyperreflective bands of the outer retina in Stargardt disease with OCT. The hyperreflective ellipsoid zone was uniformly present outside of the fovea, but only present in the fovea of 12% of the patients.[10] The current report not only confirms the high frequency of ellipsoid zone and photoreceptor outer segment loss in Stargardt disease but also quantifies the degeneration. Furthermore, the analysis platform provided en face mapping of the thickness between the ellipsoid zone and the RPE that includes the photoreceptor outer segments that allowed for a unique visualization of atrophy and attenuation.

The association between visual acuity and outer retinal integrity has been well documented in Stargardt disease. One previous report compared the preservation of the foveal photoreceptors in a patient with 20/20 vision and the paucity of the same layer in another patient with 20/100 vision.[5] Other studies have correlated transverse photoreceptor loss with visual acuity.[6] Similarly, the current report demonstrated the relationship between visual acuity and ellipsoid zone status. Specifically, visual acuity was directly correlated with ellipsoid zone/photoreceptor outer segment volume and inversely correlated with en face ellipsoid zone loss/atrophy and attenuation of the photoreceptor outer segments/ellipsoid zone.

This study does have important limitations that should considered, including its retrospective nature. In addition, this study does not include assessment of longitudinal outer retinal changes. Most of the patients included in this study had fairly advanced disease, increasing the probability of ellipsoid and photoreceptor outer segment loss. Observing these patients, as well as eyes with earlier disease, over a longer period with serial OCTs would allow for prospective analysis of the longitudinal dynamics that occur.

This novel imaging analysis technique may be instrumental in facilitating identification and surveillance of the outer retinal loss that occurs in Stargardt disease. Higher order quantitative assessment of these outer retinal changes provides a unique opportunity for disease monitoring and future clinical trial assessments. Future studies should include longitudinal assessment of quantitative metrics and evaluation of less severe phenotypes.

Key words: Stargardt disease, OCT, optical coherence tomography, ellipsoid zone, ellipsoid zone mapping.

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