



# Integrity of the Hyperreflective Layer in the Inner Choroid in Eyes with Drusen

Young Ho Kim · Cheolmin Yun · Jaeryung Oh

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## ABSTRACT

**Introduction:** This study aimed to compare the integrity of the hyperreflective layer of the inner choroid in eyes with and without drusen.

**Methods:** Swept-source optical coherence tomography images of patients with drusen and normal controls were reviewed. Using a line plot of ImageJ, choroidal reflectivity was measured at the subfovea, and the integrity of the hyperreflective layer of the inner choroid was determined.

**Results:** In total, 63 eyes with drusen and 30 control eyes without drusen were included. The integrity of the hyperreflective layer of the inner choroid was preserved in 81.0% of eyes with drusen and 93.3% of normal controls. The proportion of eyes with the hyperreflective layer

did not differ between eyes with and without drusen. Of the 63 subjects with drusen, this hyperreflective layer was observed in all 28 eyes (100%) with pachydrusen but only in 68.6% of the 35 eyes with soft drusen, and its prevalence was significantly different ( $P = 0.001$ ).

**Conclusion:** The prevalence of the hyperreflective layer between the choriocapillaris and medium or large choroidal vessels in eyes with soft drusen differed from that in eyes with pachydrusen. These findings support the suggestion that changes within the choroidal stroma may be involved in the pathogenesis of age-related macular degeneration.

**Keywords:** Age-related macular degeneration; Choroid; Drusen; Hyperreflective layer of the inner choroid; Optical coherence tomography; Pachydrusen

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### Key Summary Points

#### *Why carry out this study?*

The complex composition of the choroid, which includes blood vessels and various stromal components, contributes to the broad spectrum of irregular reflectivity observed in optical coherence tomography images.

This study aimed to investigate the variation in choroidal reflectivity representing the choroidal stroma in eyes with and without drusen.

#### *What was learned from the study?*

The hyperreflective layer in the inner choroid was identified between the choriocapillaris and the medium or large choroidal vessels.

The hyperreflective layer was less frequently observed in eyes with soft drusen than in eyes with pachydrusen.

Measurement of the inner choroidal hyperreflective layer may provide new insights into the study of the pathophysiology of age-related macular degeneration.

## INTRODUCTION

Drusen are characteristic features of age-related macular degeneration (AMD) and their pathogenesis has been widely studied [1–4]. In addition to the degeneration of Bruch's membrane (BM) and the retinal pigment epithelium (RPE), recent studies using optical coherence tomography (OCT) have suggested a role for the choroid in the development of different types of drusen, such as soft drusen and pachydrusen [2–5]. The choroid is a highly vascularized tissue that helps maintain homeostasis of the overlying neuroretina [6]. Choroidal vascular flow is purportedly essential for maintaining the

supply of oxygen and nutrients to the retina and RPE. In addition to classical techniques for the kinetic measurement of choroidal vascular flow, such as indocyanine green angiography and Doppler flowmetry, a newly proposed static measurement of the absolute and relative proportion of the choroidal vascular lumen using OCT has revealed choroidal vascular variation in eyes with and without various types of drusen [7, 8]. Baek et al. [8] suggested that the large choroidal vessels in Haller's layer were associated with pachydrusen. The choroidal vascularity index has been used to characterize pathological changes in the choroid in various chorioretinal diseases [7].

Various progressive changes in the choroid owing to aging have been observed [6]. Age-related overall thinning of the choroid has been presented in several studies with histological samples and with OCT [6, 9, 10]. A linear reduction in choroidal blood flow with aging has been reported in many studies with volunteers [6, 11]. While early studies on age-related choroidal changes have focused on the choriocapillaris and choroidal flow [12–15], in addition to vessels, the choroid is composed of stroma, such as various cells and neurons. Aging changes in choroidal stroma have also been reported in various studies [16, 17]. A recent study showed that the choroidal stromal volume decreased with age [16]. A decrease in hyaluronic acid in the choroid stroma was reported in a previous study [18], and a trend of decreasing content with aging in choroidal melanocytes was observed in another study [19]. However, whether these age-related changes in the choroidal stromal component are associated with the development of drusen remains unclear.

Therefore, in this study, using swept-source (SS)-OCT, we investigated the variation in choroidal reflectivity representing the choroidal stroma in eyes with and without drusen.

## METHODS

The Institutional Review Board (IRB) of the Korea University Hospital approved the study design (IRB number: 2023AN0096). Informed

consent was waived for this study, as it was a retrospective review of anonymized image data. This study adhered to the tenets of the Declaration of Helsinki. After reviewing the medical records of subjects who visited retina clinics, we included the eyes of subjects with drusen ( $\geq 125 \mu\text{m}$  in the smallest diameter) [20, 21]. We also excluded eyes with only reticular pseudodrusen, as well as those with geographic atrophy, disciform scarring, choroidal neovascularization (CNV), or pigmented epithelial detachment in AMD-affected eyes. Other exclusion criteria were as follows: (1) those with other chorioretinal diseases including central serous chorioretinopathy, retinal vascular occlusion, diabetic retinopathy, macular hole, and uveitis; (2) those with inherited retinal dystrophy; (3) those with a history of intravitreal injection, laser treatment, vitrectomy, and other ocular surgery except cataract surgery; and (4) patients with high myopia (axial length  $\geq 28.0$  mm or mean spherical equivalent  $< -6.50$  D). For comparison, we also included normal eyes of subjects who had undergone OCT examination but had no apparent chorioretinal disease in either eye, except for the idiopathic epiretinal membrane in one eye. We also excluded images with poor quality to prevent measurement of imaging parameters. For subgroup analysis, drusen were classified into two subgroups: soft drusen and pachydrusen. We determined soft drusen and pachydrusen using color fundus photography and OCT, according to previously described criteria [2, 4]. Several eyes with soft drusen also had reticular pseudodrusen (subretinal drusen deposits), and these were classified into the soft drusen group.

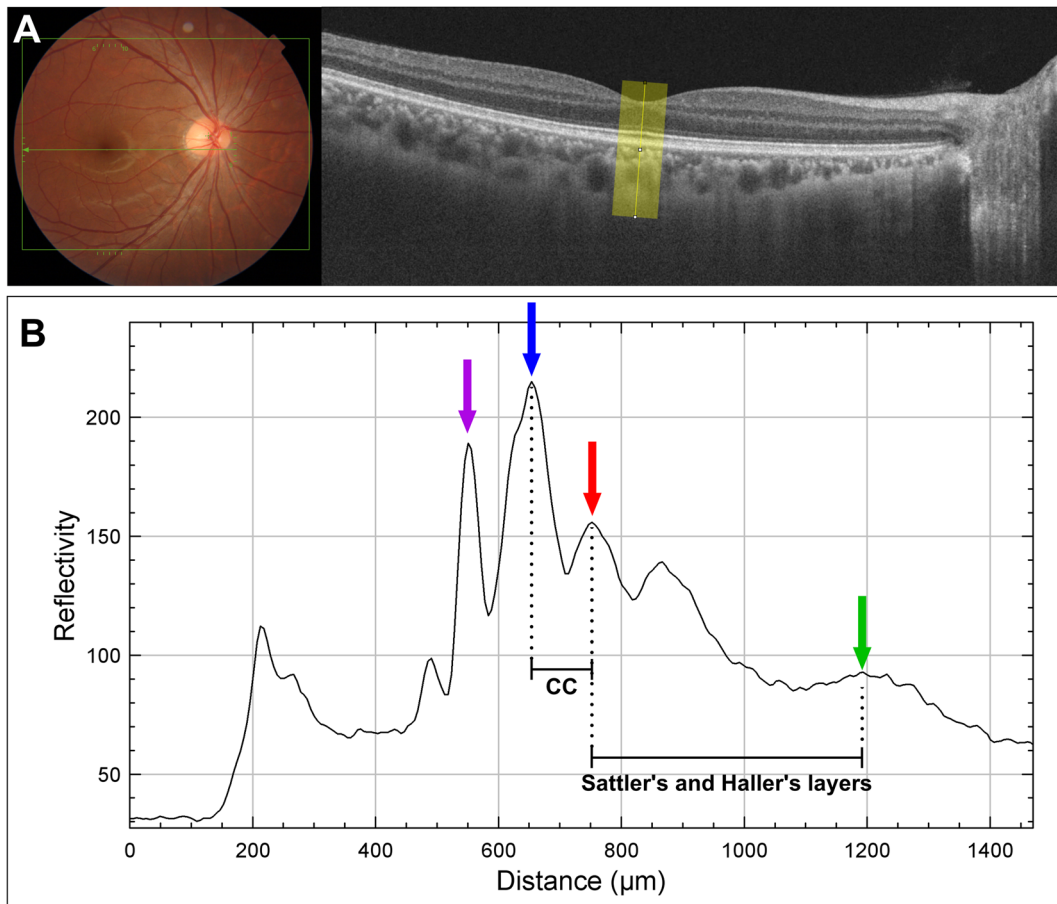
### OCT Measurements

The SS-OCT device (DRI OCT Triton, Topcon Corp., Tokyo, Japan) uses a central wavelength of 1050 nm, and it operates at a speed of 100,000 A-scans per second with a horizontal resolution of 20  $\mu\text{m}$  and an axial resolution of 8  $\mu\text{m}$ . High-resolution B-scan images averaged

from eight B-scans obtained using a five-line cross-scan protocol centered on the fovea were used. A horizontal line scan crossing the macular center was selected for the measurement. On the horizontal B-scan images from SS-OCT, we measured choroidal thickness in the subfoveal area using a previously presented method [5]. Two independent observers (Y.K. and J.O.) determined subfoveal choroidal thickness. The average of the measurements of the subfoveal choroidal thickness of the two observers was used for analysis. Using built-in software, a retinal thickness map was automatically generated from a 12-mm  $\times$  9-mm macular volume scan centered between the optic disc and fovea. The central macular thickness was defined as the mean retinal thickness on the 1000- $\mu\text{m}$ -diameter inner central circle in the Early Treatment Diabetic Retinopathy Study (ETDRS) grid. The segmentation line and the position of the center of the ETDRS grid were reviewed by a retinal specialist (Y.K.) and adjusted, if necessary.

### Measurement of Vitreous, RPE-BM Complex, and Choroidal Reflectivity

The reflectivity of the vitreous, RPE-BM complex, and choroid was measured at the foveal center. The reflectivity was determined on the horizontal B-scan images of the SS-OCT using the “line plot” tool of ImageJ software (National Institutes of Health, [imagej.nih.gov/ij](http://imagej.nih.gov/ij)) (Fig. 1). After exporting the B-scan image from SS-OCT, the TIFF image was uploaded to ImageJ. After changing to an 8-bit image, the scale of the images was converted from pixels to micrometers. A linear “line plot” tool was applied to measure the reflectivity, and the profile of the line plot was determined using a “plot profile” function in ImageJ. The width of the line plot was set at 500  $\mu\text{m}$ . To measure the reflectivity of the vitreous, RPE, and choroid, the linear line plot was set perpendicular to the BM. The mean vitreous reflectivity was determined using a linear line plot in a 500- $\mu\text{m}$   $\times$  500- $\mu\text{m}$  area of the vitreous at the foveal center. The peak



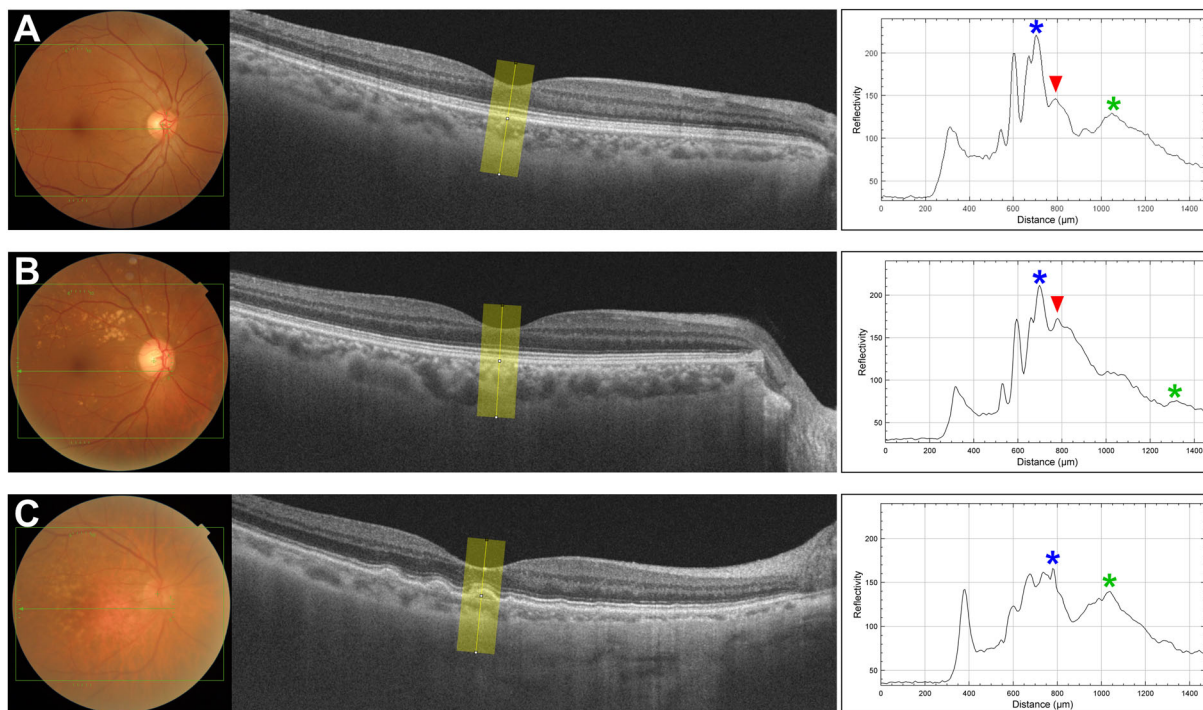
**Fig. 1** Determining choroidal reflectivity. **A** Choroidal reflectivity was determined on horizontal B-scan images of the swept-source OCT using a line plot of ImageJ with a width of 500  $\mu\text{m}$ . The 500- $\mu\text{m}$ -wide line plot was set perpendicular to Bruch's membrane (blue arrow). **B** Using the "plot profile" function of ImageJ, the reflectivity of the choroid was determined. The reflectivity profile plot shows several inflection points, with peak reflectance corresponding to the ellipsoid zone (purple arrow), retinal pigment

epithelium (blue arrow), and the choroidal–scleral junction (green arrow). The hyperreflective layer of the inner choroid (red arrow), which had the highest reflectivity within areas with inverted choroidal reflectivity, was observed between the choriocapillaris (CC) and medium-sized and large choroid vessel in Sattler's and Haller's layers. CC choriocapillaris, OCT optical coherence tomography

reflectivity of the RPE complex was determined as the outermost highest reflectance of the RPE-BM complex layer. The reflectivity of the choroid generally decreases with distance from the RPE-BM complex. The inverted reflectivity of the choroid was determined when there was a zone of increasing reflectance. Within areas with inverted choroidal reflectivity, the highest reflectivity was defined as the peak reflectance of the choroid.

### Determination of the Hyperreflective Layer of the Inner Choroid

The hyperreflective layer of the inner choroid was identified when the peak reflectivity of the choroid was located between the choriocapillaris layer and the medium or large choroidal vessels (Figs. 1 and 2). The optical intensity ratio (OIR) of the hyperreflective layer of the inner choroid was defined as the mean OCT reflectivity of the peak reflectivity of the inner



**Fig. 2** Representative cases of the hyperreflective layer of the inner choroid. In most of the eyes with normal fundus (**A**) or pachydrusen (**B**), the peak choroidal reflectance (red arrowheads) was located between the choriocapillaris and a large choroid vessel layer. However, the peak choroidal

reflectance was not observed in some eyes with soft drusen (**C**). The blue and green asterisks indicate the retinal pigment epithelium and choroidal–scleral junction, respectively

choroid, normalized to the mean reflectivity of the vitreous. To obtain the OIR [22] of the hyperreflective layer of the inner choroid, we calculated the peak choroid OIR by dividing the average intensity of the hyperreflective layer of the inner choroid by the mean vitreous intensity. The distance between the RPE–BM complex and the hyperreflective layer was measured. In eyes with extracellular deposits between RPE and BM, the distance between the BM and the hyperreflective layer was measured.

### Statistical Analysis

SPSS Statistics (version 20.0; IBM Corp., Armonk, NY, USA) was used for statistical analysis. Continuous variables were compared using the independent *t*-test, and categorical variables were compared using the chi-squared test or Fisher’s exact test. Linear correlations

between the parameters were analyzed using Pearson’s correlation test. A *P* value < 0.05 was considered statistically significant.

## RESULTS

### General Characteristics

We included 93 eyes from 93 subjects. The mean age of the 93 subjects was  $66.5 \pm 15.0$ . The study included 46 women and 47 men. The mean subfoveal choroidal thickness was  $224.6 \pm 121.7$  (66.5–619.0)  $\mu\text{m}$ . Of the 93 eyes, the integrity of the hyperreflective layer of the inner choroid was preserved in 80 (86.0%). In 80 eyes with the hyperreflective layer of the inner choroid, the mean peak reflectance of the hyperreflective layer was  $145.3 \pm 15.1$ . The peak reflectance of the hyperreflective layer did

**Table 1** Comparison of choroidal characteristics between eyes with and without drusen

Variables	Eyes with drusen ( <i>n</i> = 63)	Eyes without drusen ( <i>n</i> = 30)	<i>P</i> value
Age, years, mean ± SD	72.7 ± 9.5	53.6 ± 16.4	< 0.001
Sex, <i>n</i> (%)			
Male	33 (52.4)	14 (46.7)	0.606
Female	30 (47.6)	16 (53.3)	
Eye, <i>n</i> (%)			
Right	31 (49.2)	17 (56.7)	0.501
Left	32 (50.8)	13 (43.3)	
Central macular thickness (μm), mean ± SD	230.0 ± 29.2	237.2 ± 22.3	0.240
Subfoveal choroidal thickness (μm), mean ± SD	243.4 ± 131.2	247.0 ± 101.2	0.896
Hyperreflective layer of the inner choroid, <i>n</i> (%)			
Present	51 (81.0)	28 (93.3)	0.213
Absent	12 (19.0)	2 (6.7)	
OIR of peak reflectance of hyperreflective layer of the inner choroid, mean ± SD	4.56 ± 0.62	4.47 ± 0.45	0.396
Peak reflectance of hyperreflective layer of the inner choroid, mean ± SD	143.4 ± 16.3	141.2 ± 16.0	0.533
Distance between hyperreflective layer and the RPE-BM complex <sup>a</sup>	48.7 ± 13.0	49.0 ± 17.1	0.931

OIR optical intensity ratio, RPE-BM retinal pigment epithelium-Bruch's membrane, SD standard deviation

not correlate with age ( $r = 0.135$ ,  $P = 0.231$ ). The OIR of the hyperreflective layer did not correlate with age ( $r = 0.165$ ,  $P = 0.145$ ). The mean distance between the hyperreflective layer and the RPE-BM complex was  $45.6 \pm 10.1$  μm.

#### Comparison of the Hyperreflective Layer of the Inner Choroid Between Eyes With and Without Drusen

Of the 93 eyes, 63 eyes of 63 subjects had drusen. Thirty eyes of 30 subjects in the control group had no fundus lesions or history of chorioretinal diseases. The mean age was different between eyes with and without drusen ( $P < 0.001$ ) (Table 1). The central macular and

subfoveal choroidal thickness did not differ between the groups. The proportion of eyes with the hyperreflective layer of the inner choroid was different between those with (81.2%) and without drusen (93.3%) (Table 1). However, it was not statistically significant ( $P = 0.213$ ). No differences between groups were observed in the peak reflectance of the hyperreflective layer in the inner choroid or the OIR of the peak reflectance, or in the distance between the hyperreflective layer and the RPE-BM complex.

**Table 2** Comparison of the hyperreflective layer of the inner choroid between eyes with soft drusen and pachydrusen

Variables	Eyes with soft drusen ( <i>n</i> = 35)	Eyes with pachydrusen ( <i>n</i> = 28)	<i>P</i> value
Age, years, mean ± SD	77.5 ± 7.6	66.8 ± 8.6	< 0.001
Sex, <i>n</i> (%)			
Male	15 (42.9)	18 (64.3)	0.091
Female	20 (57.1)	10 (35.7)	
Eye, <i>n</i> (%)			
Right	17 (48.6)	14 (50.0)	0.910
Left	18 (51.4)	14 (50.0)	
Central macular thickness (μm), mean ± SD	229.9 ± 36.5	230.3 ± 16.9	0.958
Subfoveal choroidal thickness (μm), mean ± SD	158.7 ± 70.4	349.3 ± 111.0	< 0.001
Drusen within 500 μm of the foveal center, <i>n</i> (%)			
Present	18 (51.4)	1 (3.6)	< 0.001
Absent	17 (48.6)	27 (96.4)	
Hyperreflective layer of the inner choroid, <i>n</i> (%)			
Present	24 (68.6)	28 (100)	0.001
Absent	11 (31.4)	0 (0)	
OIR of peak reflectance of hyperreflective layer of the inner choroid <sup>a</sup> , mean ± SD	4.60 ± 0.50	4.77 ± 0.59	0.266
Peak reflectance of hyperreflective layer of the inner choroid <sup>a</sup> , mean ± SD	145.3 ± 15.9	148.0 ± 14.4	0.514
Distance between hyperreflective layer and the RPE-BM complex <sup>a</sup>	49.5 ± 13.7	47.7 ± 12.2	0.588

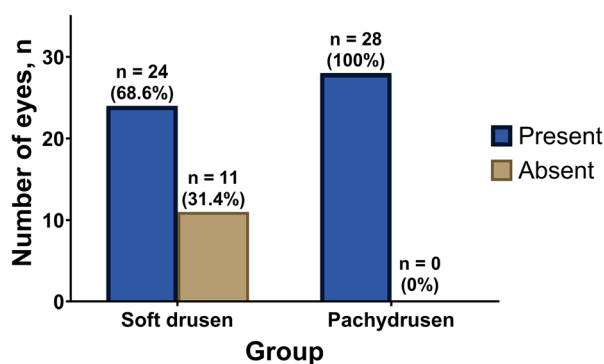
<sup>a</sup>Data and *P* values were obtained only from the eyes with the hyperreflective layer of the inner choroid  
*OIR* optical intensity ratio, *RPE-BM* retinal pigment epithelium-Bruch's membrane, *SD* standard deviation

### Subgroup Analysis for Comparison of the Hyperreflective Layer of the Inner Choroid Between Eyes with Pachydrusen and Soft Drusen

Of the 63 subjects with drusen, 35 had soft drusen, and 28 had pachydrusen. The mean age was lower in the eyes with pachydrusen ( $P < 0.001$ ) (Table 2). The central macular thickness did not differ between the groups ( $P = 0.958$ ), but the subfoveal choroidal thickness was thicker in the pachydrusen group

( $P < 0.001$ ). The proportion of eyes with the hyperreflective layer of the inner choroid differed between eyes with soft drusen and pachydrusen ( $P = 0.001$ ). The hyperreflective layer was observed in all eyes (100%) with pachydrusen but only in 68.6% of eyes ( $n = 24$ ) with soft drusen (Fig. 3). Drusen were more frequently observed within 500 μm of the foveal center in eyes with drusen than those with pachydrusen ( $P < 0.001$ ).

When comparing 28 eyes with pachydrusen and 24 eyes with soft drusen that had a hyperreflective layer of the inner choroid, the OIR of



**Fig. 3** Distribution of the hyperreflective layer of the inner choroid in eyes with soft drusen and pachydrusen

the peak reflectance of the hyperreflective layer did not differ between the groups. The distance between the hyperreflective layer and the RPE-BM complex also did not differ.

#### Comparison of Variables in Eyes with Soft Drusen According to the Presence or Absence of the Hyperreflective Layer of the Inner Choroid

Of the 35 eyes with soft drusen, the mean subfoveal choroidal thickness was  $167.5 \pm 70.2$  and  $139.5 \pm 70.2$   $\mu\text{m}$  in eyes with and without the hyperreflective layer, respectively ( $P = 0.281$ ) (Table 2). Among them, 18 showed drusen within 500  $\mu\text{m}$  of the foveal center (Table 3). The proportion of eyes with or without the hyperreflective layer of the inner choroid did not differ between eyes with and

without drusen within 500  $\mu\text{m}$  of the foveal center ( $P = 0.803$ ).

## DISCUSSION

The variation in reflectivity in the choroid depends mainly on the distribution of the vascular lumen [17, 23]. Choroidal vessels have been classified into three layers according to vessel dimensions: the choriocapillaris, Sattler's layer, and Haller's layer. Although Sattler's layer and Haller's layer are not easily distinguishable in OCT images of all eyes, the morphology of medium or large choroidal vessels in Sattler's or Haller's layer can be determined as a hyporeflective lumen surrounded by the hyperreflective vascular wall or stromal tissues in OCT images [23]. Aside from medium or large vessels in Sattler's and Haller's layers, vessels in the choriocapillaris layer cannot be separated as each vessel in OCT B-scan images. In addition, en face OCT angiography images have shown blood flow in the choriocapillaris [24], but it is difficult to discriminate each vessel in the choriocapillaris slab. Instead, the choriocapillaris layer can be easily identified as a linear hyporeflective area between the RPE layer and the hyperreflective layer beneath the choriocapillaris layer [24]. In this study, we obtained the reflectivity of the hyperreflective layer comprising the outer margin of the choriocapillaris layer. The hyperreflective layer in the inner choroid was present in almost all eyes with a normal fundus in this study. The origin of the hyperreflective layer in the inner choroid

**Table 3** Presence of the hyperreflective layer of the inner choroid in 35 eyes with soft drusen according to location of drusen

Variables	Hyperreflective layer of the upper choroid				Total	<i>P</i> value <sup>a</sup>
	Present		Absent			
	<i>n</i>	%	<i>n</i>	%		
Drusen within 500 $\mu\text{m}$ of the foveal center						
Present	12	66.7	6	33.3	18	0.803
Absent	12	70.6	5	29.4	17	

<sup>a</sup>*P* values were obtained by chi-squared test



remains unclear. Its location corresponds to the area of the choroidal stroma and the medium-sized vessel walls of the inner choroid (Fig. 1). Recent studies using en face OCT have suggested that melanin pigmentation contributes to hyperreflectivity in this area [25–27]. These results suggest that the hyperreflective layer in the inner choroid may be a characteristic finding representing the pigmentation of the inner choroid.

In this study, we focused on choroidal reflectivity and hypothesized that the hyperreflective layer in the inner choroid in OCT B-scans may represent variation in the stromal component of the choroid [25, 28]. We compared the proportion of eyes with the hyperreflective layer in the inner choroid between those with soft drusen and pachydrusen. The prevalence of the hyperreflective choroid layer differed between the eyes with soft drusen and pachydrusen. It is unclear how the difference in the prevalence of the hyperreflective layer contributes to the difference in the phenotype of drusen, such as soft drusen or pachydrusen. One possibility is that the variation in choroidal vascular and stromal components associated with soft drusen or pachydrusen may be related to the difference in the prevalence of the hyperreflective layer in the inner choroid. Another possibility is that the change within choroidal stromal components, such as melanin pigment, in eyes with soft drusen could be related to the difference in the prevalence of the hyperreflective layer in the inner choroid. In a previous study [27], changes in choroidal reflectivity on OCT B-scan images were reported in eyes with a sunset glow fundus due to Vogt–Koyanagi–Harada disease. In another study [26], the hyperreflective choroidal foci on OCT decreased in the inner choroid of eyes with sunset glow fundus. These findings may support the notion that depigmentation in the choroid could be associated with the development of soft drusen, which is a hallmark of classic AMD. However, this requires further research. Melanin in the choroid has been suggested to protect the choriocapillaris, choroidal vessels, and the neuroretina [29, 30]. Melanin depletion in the inner choroid may be related to the development of soft drusen. In a previous study with

histological samples, Weiter et al. [19] showed that the total choroidal melanin density did not change with age. In our study, the peak reflectance of the hyperreflective layer did not correlate with age. However, the hyperreflective layer was more frequently missing in the eyes with soft drusen. Further studies with more cases are needed to evaluate the quantitative relationship between the peak reflectance of the hyperreflective layer and the development of soft drusen.

In an earlier study, drusen in AMD were classified as hard drusen, soft drusen, and compound drusen [31], and their pathogenesis has been widely studied [1, 4]. Furthermore, in aging eyes, the association of the aging of the RPE and BM with the development of drusen has been thoroughly described [4, 31]. After the introduction of OCT in AMD studies, choroidal changes were suggested to contribute to the pathogenesis of drusen, which were classified as soft drusen, pachydrusen, and reticular pseudodrusen [2, 3]. Early studies have suggested that choroidal vascular changes with aging contribute to the development of AMD. Choroidal thinning and vascular depletion are suspected to provide an environment conducive to the development of soft drusen or reticular pseudodrusen in eyes with AMD. Zhou et al. [16] showed that the stromal or vascular volume of the choroid decreased with age; however, they also reported that the choroidal vascularity index or choroidal stroma-to-volume ratio did not [16]. The role of the relative depletion of vascular or stromal components in the development of AMD has been questioned. In other studies using OCT angiography [12, 13], a generalized flow deficit in the choriocapillaris of the macular area was correlated with the development of AMD. Furthermore, a focal defect in the choriocapillaris overlying pachyvessels was also suggested to be related to pachydrusen [32]. While previous studies on the relationship between flow deficit in the choriocapillaris/choroid and the phenotype of drusen have been presented [14, 15], it is still not clear why soft drusen are uncommon in Asians with darker pigmentation [3]. The finding of the current study that depigmentation in the inner choroid is more frequent in eyes with soft

drusen may provide a new perspective on the role of stroma in the development of AMD.

This study faced some limitations associated with its retrospective nature. Patients with drusen were older than those without. As several previous studies have shown that subfoveal choroidal thickness decreases with age [33, 34], this difference might have an effect on the choroidal reflectivity. However, the small number of cases in this study made it difficult to perform further subgroup analyses after considering the effect of age on subfoveal choroidal thickness. Because there was no difference in the subfoveal choroidal thickness between eyes with and without drusen, we believe that the bias had a limited effect on the results. However, further studies with a large number of cases are needed to investigate the relationship between different variables. Another limitation is that we quantified the reflectivity of the hyperreflective layer of the inner choroid only at the foveal center. We used the SS-OCT device, which has less signal reduction than the spectral domain OCT device, but due to OCT technology principles, the reflectivity of the inner choroid can be affected by the retina overlying it. Retinal thickness can be affected by age [35, 36], which may affect choroidal reflectance. To reduce the influence of the retina, we excluded eyes with diseases within the retina. There was no difference in central macular thickness between the groups. The properties of the RPE-BM complex could also affect our measurements. In this study, eyes with soft drusen more frequently had extracellular deposits within 500  $\mu\text{m}$  of the fovea than eyes with pachydrusen, and the extracellular deposits may have affected the measurement of the choroidal reflectance. However, the peak reflectance of the hyperreflective layer and its OIR did not differ between eyes with soft drusen and pachydrusen. In addition, the proportion of eyes with the hyperreflective layer of the inner choroid did not differ between eyes with and without soft drusen within 500  $\mu\text{m}$  of the foveal center. Further research at various locations of the choroid may provide more information for eyes with and without drusen. Another limitation is that we could only include Asian subjects with pigmented eyes. For the generalization of

our results to other races, further studies with different ethnic populations are required.

## CONCLUSION

The prevalence of the hyperreflective layer between the choriocapillaris and the medium or large choroidal vessels in eyes with soft drusen was different from that in eyes with pachydrusen. These findings support the notion that changes in the choroidal stroma, such as changes in reflectance, may have different effects on the development of soft drusen and pachydrusen.

**Authorship.** All named authors meet the International Committee of Medical Journal Editors (ICMJE) criteria for authorship for this article, take responsibility for the integrity of the work as a whole, and have given their approval for this version to be published.

**Author Contributions.** Jaeryung Oh contributed to the study conception and design, data collection, analysis and/or interpretation, original draft preparation, and manuscript review. Young Ho Kim contributed to the study design, data collection, analysis and/or interpretation, drafting and editing of the article, and manuscript review. Cheolmin Yun contributed to the study conception, data interpretation, and manuscript review. All authors read and approved the final manuscript.

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**Data Availability.** The datasets generated during and/or analyzed during the current

study are available from the corresponding author on reasonable request.

### Declarations

**Conflict of Interest.** Young Ho Kim, Cheolmin Yun, and Jaeryung Oh declare that they have no financial or nonfinancial competing interests in any of the products described in this study.

**Ethical Approval.** This study was conducted in accordance with the Declaration of Helsinki (as revised in 2013). The study was approved by the Institutional Review Board of Korea University Hospital (IRB number: 2023AN0096), and the requirement for individual consent for this retrospective analysis was waived.

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