High-Resolution Fourier-Domain Optical Coherence Tomography of Choroidal Neovascular Membranes Associated with Age-Related Macular Degeneration

Susanna S. Park, Steven N. Truong, Robert J. Zawadzki, Subail Alam, Stacey S. Choi, David G. Telander, John S. Werner, and Lawrence S. Morse

Purpose. To investigate the use of high-resolution Fourier-domain optical coherence tomography (Fd-OCT) to image choroidal neovascular membranes (CNVMs) associated with exudative age-related macular degeneration (eAMD).

Methods. An Fd-OCT instrument with axial resolution of 4 to 4.5 μm and transverse resolution of 10 to 15 μm was used to image 21 eyes (19 subjects) with newly diagnosed eAMD. A raster series of 100 B-scans separated by 60 μm was used to study the growth pattern of CNVM and associated morphologic changes. CNVM size was determined using 250 to 300 serial virtual C-scans of reconstructed three-dimensional macular volume.

Results. A highly reflective subretinal and/or subretinal pigment epithelial (RPE) lesion that co-localized with the CNVM seen on fluorescein angiography was detected in all eyes by Fd-OCT. Although a combined subretinal and sub-RPE growth pattern of various degrees was noted in 15 (71%) eyes, a statistically significant difference in the distribution of growth pattern was noted when classic CNVM was compared with occult CNVM (χ² = 10.4, df = 2, P < 0.005). Classic lesions had >90% subretinal growth pattern, whereas occult lesions had a more variable growth pattern. Angiographic CNVM size correlated with size on Fd-OCT but correlation was better for classic CNVM (classic, r = 0.99, P < 0.0001; nonclassic, r = 0.78, P < 0.001).

Conclusions. Fd-OCT is a promising potential alternative modality to visualize CNVM with AMD. Angiographic lesion size and type correlated with growth pattern and size of CNVM on Fd-OCT, with correlation being stronger for classic lesions.

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Exudative age-related macular degeneration (eAMD) is a leading cause of irreversible blindness in the elderly in the United States. The anatomy and growth patterns of choroidal neovascular membranes (CNVM) associated with eAMD have been studied in the past using either enucleated eyes or excised tissue from submacular surgery, in an attempt to understand the pathogenesis of this condition. Several studies have demonstrated that CNVM comprises a dynamic proliferation of fibrovascular tissue through Bruch’s membrane. Gass used enucleated eyes and classified the neovascular growth pattern as subretinal pigment epithelial (RPE) (type 1), subretinal (type 2), or combined.

Several studies attempted to demonstrate a correlation between the angiographic classification of CNVM (classic versus occult) and the anatomic classification (type 1 vs. type 2). However, data on postmortem eyes are limited. Several groups have reported on clinicopathologic correlation using surgically removed CNVMs. These reports showed that classic CNVMs tend to exhibit either a combined or subretinal growth pattern while occult CNVMs tend to be composed of tissue that appeared to be from the sub-RPE space. However, tissue classification of surgically removed CNVMs can have limited accuracy since anatomy is disrupted, and incomplete excision of CNVM during surgery cannot be ruled out.

With the emergence of optical coherence tomography (OCT), a new method for studying CNVM growth patterns in living subjects became available. OCT provides a noninvasive, nondestructive method of obtaining detailed anatomic data in vivo. A study using the commercially available time-domain OCT system Stratus OCT (Carl Zeiss Meditec, Inc., Dublin, CA) suggested that classic CNVM tends to be subretinal whereas occult CNVM tends to be sub-RPE. However, the Stratus OCT system that was used provides only six radial scans of the macula and has limited axial and transverse resolution. Thus, it is not possible to visualize the whole extent of the CNVM with this instrument.

Fourier-domain OCT (Fd-OCT) systems are newer generation instruments that allow a reduction in image acquisition time by an additional factor of 20 to 40 when compared with the Stratus OCT. These modifications allow acquisition of rapid serial fine-cut B-scans of the macula in a single scanning sweep, so that the entire macula can be imaged and analyzed. The new Fd-OCT system has allowed detailed imaging of macular lesions that may be missed with the time-domain OCT system. There are several recent reports using Fd-OCT to image eyes with eAMD. These reports have concentrated on improved visualization of the retinal layers and drusen in eyes with eAMD before and after treatment with inhibitors of vascular endothelial growth factor when compared to the traditional time-domain OCT. One report describes volume measurements of CNVM before and after treatment but did not mention the growth pattern of CNVM.
In this report, a high-resolution Fd-OCT system developed at our institution was used to image eyes with newly diagnosed eAMD to evaluate the usefulness of this instrument in visualizing CNVM and associated morphologic changes.

**Materials and Methods**

This prospective observational case series enrolled 21 eyes of 19 patients (8 men, 11 women; 48–92 years of age) with newly diagnosed eAMD seen in the Retina Clinic at the University of California Davis Eye Center between September 2005 and June 2006. For this study, eyes diagnosed with retinal angiomatosus proliferation or concurrent macular hemorrhage that may obscure part of the CNVM on fluorescein angiography (FA) were excluded. Informed written consent was obtained from all patients before enrollment. This study was approved by the Institutional Review Board of the University of California, Davis, and was conducted in compliance with the Declaration of Helsinki.

All patients underwent a dilated fundus examination, fundus photography, and standard FA. Each angiogram was independently evaluated by five retinal specialists (SNT, SA, DGT, LSM, SSP) and classified according to the Macular Photocoagulation Study protocol as classic, minimally classic, occult with late leakage of undetermined source, or occult with pigment epithelial detachment (PED); i.e., fibrovascular PED.25 For the minimally classic lesions, the classic and occult components were quantified. In rare cases when angiogram interpretation varied among reviewers, the angiogram was reviewed as a group to reach consensus. The CNVMs were then outlined and the GLD was measured (IMAGEnet 2000 ver. 2.55; Topcon America Corp., Paramus, NJ). For fibrovascular PED, the entire lesion (including area of PED) was included.

A state-of-the-art Fd-OCT system similar to that described by Wojtkowski et al.26 and further improved by Nassif et al.27 was used to image all 21 eyes on the same day as FA. This system was constructed at the University of California, Davis Medical Center.18 The instrument used a superluminescent diode as a light source (855 nm at 7 nm bandwidth; model D855; Superlum Diodes Ltd., Moscow, Russia), and created images with an axial resolution of 4 to 4.5 μm, and calculated transverse resolution between 10 and 15 μm. A raster series of 100 B-scans (1000 A-scans/frame, nine frames/s) imaged over a 6 × 6 × 2-mm volume of retina and underlying RPE (lateral × lateral × depth) centered over the macula was obtained. The total acquisition time for a single macular sweep was 11 seconds. Each consecutive B-scan image was laterally separated by 60 μm on the retina. After the 100 B-scans were acquired, the images were registered by using custom software to minimize fine axial motion artifacts. After careful inspection and manual correction of registration, the B-scans were shifted and rotated to reduce axial shift.

Each single B-scan image was analyzed to carefully identify the CNVM. CNVM appeared on the gray-scale B-scan Fd-OCT images as a highly reflective lesion in the subretinal space, sub-RPE space, or both.5,7,22-24,26 A CNVM that was localized predominantly (>90% of the lesion) in the subretinal space on Fd-OCT was classified as type 2. A CNVM that was identified predominantly (>90% of the lesion) in the sub-RPE space was classified as type 1. Any CNVM that was not found predominantly (>90%) in the subretinal or sub-RPE space was classified as combined. The serial B-scan Fd-OCT images of each subject were independently analyzed by five retinal specialists (SNT, SA, DGT, LSM, SSP) who were blinded to the angiographic classification of the lesion at the time of review. In rare cases of disagreement among the retinal specialists, the Fd-OCT images were reviewed as a group until consensus was reached. The serial B-scan Fd-OCT images were reviewed also for the presence of cystoid macular edema (CME), subretinal fluid (SRF) or PED. PED on Fd-OCT B-scans was identified by elevation of the RPE layer over a hyperreflective space and did not include elevation of the RPE layer over hyperreflective lesions suggestive of CNVM or drusen.

**Results**

Among 21 eyes of 19 patients with newly diagnosed eAMD that met the inclusion and exclusion criteria of this study and were imaged with Fd-OCT, a highly reflective subretinal and/or sub-RPE lesion was visualized in the macula of all 21 eyes by Fd-OCT. These lesions co-localized to the area of CNVM seen on FA. Table 1 lists the demographic information, the angiographic classification of CNVM, and the Fd-OCT findings. Of the 21 eyes imaged, FA showed classic CNVM in 7 (33%) eyes, minimally classic CNVM in 3 (14%) eyes and an occult CNVM in 11 (53%) eyes. Among eyes with occult CNVM, seven (35%) had late leakage of undetermined source and four (20%) had fibrovascular PED (i.e., occult CNVM with late irregular pooling into the PED).

By analyzing the 100 serial Fd-OCT B-scans of the macula, we determined the anatomic growth pattern of the CNVM in all but one eye (95%). A combined sub-RPE and subretinal growth pattern of varying degrees was noted in 15 (71%) eyes. However, based on the classification of growth pattern as defined by this study, 7 (33%) eyes had >90% sub-RPE (i.e., type 1) growth patterns, 10 (48%) had >90% subretinal (i.e., type 2) growth pattern, and 3 (14%) had a combined growth pattern (Table 1).

Table 2 summarizes the anatomic growth pattern of the CNVM on Fd-OCT for the various angiographic lesion types. Of the seven eyes that had classic CNVM on FA, >90% subretinal (type 2) growth pattern was observed in six (86%) eyes, and one eye was indeterminant. Among those seven eyes, four had a small sub-RPE component that was <10% of the lesion.

Among 11 eyes with occult CNVM, 7 (64%) had >90% sub-RPE (type 1) growth pattern, 2 had >90% subretinal growth pattern, and 2 had a combined growth pattern. The two eyes with occult CNVMs with >90% subretinal (type 2) growth pattern had fibrovascular PED (Fig. 1).

A statistically significant difference in the distribution of type 1 and 2 growth patterns was noted among eyes with classic CNVM when compared with eyes with occult CNVM.
(χ² = 10.4, df = 2, P < 0.005)—that is, a type 1, or >90% sub-RPE growth pattern, was more likely to be associated with angiographically occult lesions, whereas type 2, or >90% subretinal growth pattern was more likely associated with classic lesions.

Among the three eyes with minimally classic pattern of leakage on FA, two had a >90% subretinal (type 2) growth pattern (Fig. 2) and one had a combined pattern of growth on Fd-OCT.

Tables 1 and 3 summarize the retinal morphologic changes associated with CNVM as seen on Fd-OCT. Among the seven eyes with classic CNVM, three had associated CME and six had associated SRF visible on Fd-OCT. None of the eyes had PED. Among the seven eyes with occult CNVM without PED, all had CME, five had SRF, and five had PED. On Fd-OCT serial B-scans, all eyes with occult CNVM associated with PED (i.e., fibrovascular PED) on FA had PED on Fd-OCT, two eyes had CME, and three eyes had SRF. There was no statistically significant difference in the distribution of these morphologic changes between eyes with classic CNVM and eyes with occult CNVM (χ² = 5.272, df = 4, P > 0.05) in this small series. Nonetheless, none of the eyes with classic CNVM had an associated PED on Fd-OCT, whereas 9 of 11 eyes with occult CNVM on FA had PED on Fd-OCT. Of note, 71% of eyes with occult CNVM without PED on FA had PED on Fd-OCT.

Further analysis of possible association of these morphologic changes in the retina with specific growth pattern of CNVM seen on Fd-OCT showed no statistically significant difference in the distribution of CME, SRF, and PED with type 1 or 2 growth pattern of CNVM seen on Fd-OCT (χ² = 2.282, df = 4, P > 0.05), but the numbers in each group were small.

Virtual C-scan images of the macula were analyzed serially to determine the GLD of the highly reflective subretinal/sub-RPE lesion seen on serial B-scan Fd-OCT images. In most cases, the GLD could be determined by using a single reconstructed C-scan, but in a few cases (Fig. 1D), serial C-scans were overlaid to obtain a two-dimensional image of the CNVM to obtain the GLD. In all eyes, the highly reflective lesion co-localized with the CNVM seen on FA when the C-scan images were overlaid. GLD. In all eyes, the highly reflective lesion co-localized with the CNVM seen on FA when the C-scan images were overlaid. GLD. In all eyes, the highly reflective lesion co-localized with the CNVM seen on FA when the C-scan images were overlaid.

DISCUSSION

In this study, a high-resolution Fd-OCT instrument developed in our institution was used to image eyes with newly diagnosed
In our study of 21 eyes, 54% of eyes had occult CNVM and 33% of eyes had classic CNVM. When compared with previous reports of 60% to 73% incidence of occult CNVM and 20% to 21% incidence of classic CNVM, our study population had slightly higher proportion of eyes with classic CNVM,31–33 perhaps because our study excluded eyes with nonclassic lesions associated with RAP or macular hemorrhage. Among the 21 eyes studied, 71% had some degree of combined subretinal and sub-RPE growth pattern of CNVM detected on Fd-OCT. Nonetheless, a statistically significant difference in the distribution of growth pattern was noted when eyes with classic CNVM were compared to eyes with occult CNVM. We found that 86% of eyes with classic CNVM on FA had a >90% subretinal (type 2) growth pattern on Fd-OCT, although many also had a small sub-RPE component that made up less than 10% of the CNVM. These findings are similar to those of the SST study, which described classic CNVM as having a subretinal growth pattern or a combined growth pattern.14 For eyes with occult CNVM, a more variable growth pattern was noted, although 64% had >90% sub-RPE (type 1) growth pattern. These observations are consistent with the limited histologic data in postmortem eyes that suggest that occult CNVMs tend to exhibit a sub-RPE or combined pattern of growth.10–14 Most of the eyes with occult CNVM in our study also had a PED visualized on Fd-OCT, although not always appreciated on FA.

Table 1. Summary of Demographics, Angiographic, and High-Resolution Fd-OCT Findings of CNVM Characteristics Associated with eAMD

<table>
<thead>
<tr>
<th>Case</th>
<th>Age</th>
<th>Sex</th>
<th>Eye</th>
<th>Lesion Type on FA</th>
<th>CNVM Growth Pattern On Fd-OCT</th>
<th>Morphologic Findings on Fd-OCT</th>
<th>Angiographic CNVM Size, GLD (mm)</th>
<th>Fd-OCT CNVM Size, GLD (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>79</td>
<td>M</td>
<td>OS</td>
<td>100% Classic</td>
<td>Type 2*</td>
<td>SRF</td>
<td>0.99</td>
<td>0.94</td>
</tr>
<tr>
<td>2</td>
<td>82</td>
<td>M</td>
<td>OD</td>
<td>100% Classic</td>
<td>Indeterminate</td>
<td>CME</td>
<td>1.83</td>
<td>1.73</td>
</tr>
<tr>
<td>3</td>
<td>65</td>
<td>M</td>
<td>OD</td>
<td>100% Classic</td>
<td>Type 2*</td>
<td>CME, SRF</td>
<td>2.79</td>
<td>2.70</td>
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<tr>
<td>4</td>
<td>88</td>
<td>F</td>
<td>OS</td>
<td>100% Classic</td>
<td>Type 2</td>
<td>CME, SRF</td>
<td>1.91</td>
<td>2.06</td>
</tr>
<tr>
<td>5</td>
<td>91</td>
<td>F</td>
<td>OD</td>
<td>100% Classic</td>
<td>Type 2*</td>
<td>SRF</td>
<td>1.24</td>
<td>1.19</td>
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<tr>
<td>6</td>
<td>48</td>
<td>F</td>
<td>OD</td>
<td>100% Classic</td>
<td>Type 2</td>
<td>SRF</td>
<td>1.19</td>
<td>1.23</td>
</tr>
<tr>
<td>7</td>
<td>48</td>
<td>F</td>
<td>OD</td>
<td>100% Classic</td>
<td>Type 2*</td>
<td>SRF</td>
<td>2.00</td>
<td>1.89</td>
</tr>
<tr>
<td>8</td>
<td>82</td>
<td>M</td>
<td>OS</td>
<td>40% Classic, 60% occult</td>
<td>Type 2*</td>
<td>SRF, PED</td>
<td>1.91</td>
<td>1.87</td>
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<tr>
<td>9</td>
<td>92</td>
<td>F</td>
<td>OD</td>
<td>60% Occult, 40% classic</td>
<td>Type 2</td>
<td>CME</td>
<td>2.46</td>
<td>1.32</td>
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<tr>
<td>10</td>
<td>77</td>
<td>M</td>
<td>OD</td>
<td>70% Occult, 30% classic</td>
<td>Combined</td>
<td>CME, PED</td>
<td>1.21</td>
<td>1.29</td>
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<tr>
<td>11</td>
<td>72</td>
<td>M</td>
<td>OD</td>
<td>Occult with PED†</td>
<td>Type 2*</td>
<td>SRF, PED</td>
<td>4.11</td>
<td>3.68</td>
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<tr>
<td>12</td>
<td>82</td>
<td>F</td>
<td>OS</td>
<td>Occult with PED†</td>
<td>Type 1</td>
<td>CME, SRF, PED</td>
<td>3.41</td>
<td>1.54</td>
</tr>
<tr>
<td>13</td>
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<td>F</td>
<td>OD</td>
<td>Occult with PED†</td>
<td>Type 1</td>
<td>PED</td>
<td>1.34</td>
<td>0.72</td>
</tr>
<tr>
<td>14</td>
<td>83</td>
<td>F</td>
<td>OS</td>
<td>Occult with PED†</td>
<td>Type 2</td>
<td>SRF, PED</td>
<td>5.58</td>
<td>2.07</td>
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<tr>
<td>15</td>
<td>74</td>
<td>M</td>
<td>OD</td>
<td>Occult</td>
<td>Type 1†</td>
<td>CME, SRF, PED</td>
<td>3.60</td>
<td>3.34</td>
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<tr>
<td>16</td>
<td>74</td>
<td>M</td>
<td>OD</td>
<td>Occult</td>
<td>Type 1†</td>
<td>CME, SRF, PED</td>
<td>4.44</td>
<td>4.40</td>
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<tr>
<td>17</td>
<td>84</td>
<td>M</td>
<td>OS</td>
<td>Occult</td>
<td>Type 1†</td>
<td>CME, SRF, PED</td>
<td>2.30</td>
<td>2.34</td>
</tr>
<tr>
<td>18</td>
<td>84</td>
<td>F</td>
<td>OD</td>
<td>Occult</td>
<td>Type 1†</td>
<td>CME, PED</td>
<td>6.79</td>
<td>4.31</td>
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<tr>
<td>19</td>
<td>84</td>
<td>F</td>
<td>OD</td>
<td>Occult</td>
<td>Combined</td>
<td>CME, SRF</td>
<td>5.41</td>
<td>3.98</td>
</tr>
<tr>
<td>20</td>
<td>76</td>
<td>F</td>
<td>OS</td>
<td>Occult</td>
<td>Type 1</td>
<td>CME, SRF</td>
<td>2.84</td>
<td>2.16</td>
</tr>
<tr>
<td>21</td>
<td>92</td>
<td>F</td>
<td>OS</td>
<td>Occult</td>
<td>Combined</td>
<td>CME, PED</td>
<td>1.84</td>
<td>1.61</td>
</tr>
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</table>

Table 2. Growth Pattern of Classic and Occult CNVM Due to eAMD as Determined by High-Resolution Fd-OCT

<table>
<thead>
<tr>
<th>Case</th>
<th>Age</th>
<th>Sex</th>
<th>Eye</th>
<th>Lesion Type on FA</th>
<th>Classic CNVM (n = 7)</th>
<th>Minimally Classic CNVM (n = 3)</th>
<th>All Occult CNVM (n = 11)</th>
<th>Occult CNVM with PED* (n = 4)</th>
<th>Occult CNVM without PED (n = 7)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>79</td>
<td>M</td>
<td>OS</td>
<td>100% Classic</td>
<td>0</td>
<td>0</td>
<td>7</td>
<td>2</td>
<td>5</td>
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<tr>
<td>2</td>
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<td>M</td>
<td>OD</td>
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<td>6</td>
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</tr>
<tr>
<td>3</td>
<td>65</td>
<td>M</td>
<td>OD</td>
<td>100% Classic</td>
<td>0</td>
<td>1</td>
<td>2</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>4</td>
<td>88</td>
<td>F</td>
<td>OS</td>
<td>100% Classic</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

* Fibrovascular PED.
When occult CNVM was associated with a PED on FA (i.e., fibrovascular PED), the growth pattern of CNVM as determined by Fd-OCT was variable. Two of four eyes with occult CNVM associated with PED on FA demonstrated >90% subretinal (type 2) growth pattern on Fd-OCT although no classic lesion was noted on FA. These cases suggest that, in the presence of significant pooling of fluorescein dye into the PED, the angiographic characteristic of the CNVM may be misleading and may not correlate with the anatomic growth pattern of the CNVM.

**TABLE 3.** Associated Retinal Findings on High-Resolution Fd-OCT among Eyes with Classic or Occult CNVM due to Degeneration in eAMD

<table>
<thead>
<tr>
<th></th>
<th>Classic CNVM</th>
<th>Minimally Classic CNVM</th>
<th>All Occult CNVM</th>
<th>Occult CNVM with PED*</th>
<th>Occult CNVM without PED</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>(n = 7)</td>
<td>(n = 3)</td>
<td>(n = 11)</td>
<td>(n = 4)</td>
<td>(n = 7)</td>
</tr>
<tr>
<td>CME</td>
<td>3 (43)</td>
<td>2 (67)</td>
<td>9 (82)</td>
<td>2 (50)</td>
<td>7 (100)</td>
</tr>
<tr>
<td>Subretinal Fluid</td>
<td>6 (86)</td>
<td>1 (33)</td>
<td>8 (73)</td>
<td>3 (75)</td>
<td>5 (71)</td>
</tr>
<tr>
<td>PED</td>
<td>0</td>
<td>2 (67)</td>
<td>9 (82)</td>
<td>4 (100)</td>
<td>5 (71)</td>
</tr>
</tbody>
</table>

Data are number of eyes (% of total in group).

* Fibrovascular PED.
Fd-OCT images of the CNVM also allowed us to visualize associated retinal morphologic changes such as CME, SRF, and PED, which may not be well appreciated with FA. Although we hypothesized that occult lesions may be more likely to have an associated PED due to the sub-RPE location of the CNVM and that classic lesions may be more likely to have subretinal fluid due to the subretinal location of the lesion, statistical analysis did not reveal an association. Nonetheless, PED was noted on Fd-OCT in 9 (82%) of 11 eyes with occult CNVM and in none of the eyes with classic CNVM. CME and SRF were frequent findings associated with either growth pattern and most likely result from increased levels of vascular endothelial growth factor (VEGF) associated with both classic and occult CNVMs, which permeates the RPE and affects all the retinal layers.34,35

The finding was similar to report using Stratus OCT, with which subretinal fluid was noted in all eyes with predominantly classic CNVM and in most eyes with occult CNVM; PED was noted in all eyes with occult CNVM.16

There are several limitations to our study that are worth noting. First, although 21 eyes were imaged in total, the number of eyes in each category was small due to the various manifestations of eAMD. Second, since Fd-OCT is a noninvasive technique, there is no histologic correlate to confirm that the highly reflective lesion visualized with Fd-OCT is CNVM. Nonetheless, the highly reflective subretinal and/or sub-RPE lesion seen on Fd-OCT in this and prior studies co-localized to the CNVM seen on FA.17,20,54,56

Other structural changes associated with CNVM such as subretinal hemorrhage or exudate may appear as highly reflective on Fd-OCT. To minimize this confusion, we limited this study to eyes without concurrent macular hemorrhage. Finally, the classification of the growth pattern of the CNVM on Fd-OCT relies on correctly identifying the GLD by FA. (A) Analysis of eyes with classic CNVM shows a close linear correlation between GLD of the CNVM obtained by Fd-OCT and that obtained by FA (r = 0.99, F1,5 = 214.10, P < 0.0001, n = 7). (B) Analysis of eyes with nonclassic CNVM showing a less strong but significant correlation between GLD of the CNVM by Fd-OCT with GLD by FA (r = 0.78, F1,12 = 18.44, P < 0.001, n = 14). The FA GLD was measured, and the Fd-OCT GLD was determined by examining serial reconstructed C-scan images.

References

13. Hermans P, Lommatzsch A, Bomfeld N, Pauleikhoff D. Angiographic-histological correlation of late exudative age-related mac-


