

Novel Semi-Automated System for Multi-Dimensional Analysis of Macular Neovascularization: A Comparative Study of Quantitative Biomarkers and Morphological-Pathophysiological Classification

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Abstract

Purpose:

To introduce and validate a novel semi-automated ImageJ-based macro for comprehensive analysis of macular neovascularization (MNV) in OCTA images. The system is benchmarked against quantitative parameters from recent literature and provides novel biomarkers plus an automated morphological–pathophysiological classification that objectively distinguishes active, mature quiescent, transitional, and arteriolarized states.

Methods:

We developed an advanced image-processing pipeline incorporating hybrid multiscale vessel enhancement (Laplacian of Gaussian and tubeness filters), dynamic region-of-interest refinement, and boundary-branch exclusion. The system generates a Vascular Complexity Score and Vascular Stability Score (0–100), as well as automated classification into Medusa, Seafan, Glomerular, Tree-in-bud, and Dead-tree patterns mapped to pathophysiological states using quantitative thresholds and published clinical criteria. A total of 112 MNV lesions imaged on three OCTA platforms (Zeiss PlexElite 6×6 mm, Zeiss HD 3×3 mm, Optovue Solix 6×6 mm) were analyzed.

Results:

Raw topological metrics differed substantially by device, precluding direct numerical comparison. Device-specific principal component analysis and piecewise-linear normalization yielded convergent Standardized Complexity, Caliber Uniformity, and Maturity scores (all medians ≈50, Kruskal–Wallis $p \geq 0.276$), indicating effective removal of device- and field-of-view–dependent scaling while preserving biological information. Automated morphological classification identified the full spectrum of MNV subtypes across all platforms, supporting cross-device robustness.

Conclusion:

This system provides a robust platform for standardized, device-independent quantification of MNV architecture. By integrating morphological patterns with quantitative biomarkers and automated pathophysiological classification, it enables objective differentiation of active angiogenesis from mature remodeling and may improve clinical decision-making and patient stratification in neovascular AMD.

Introduction

Optical Coherence Tomography Angiography (OCT-A) has revolutionized the visualization of macular neovascularization (MNV) ¹. Nevertheless, translating qualitative observations into objective quantitative metrics remains a considerable challenge. Although general-purpose tools such as AngioTool perform well with high-contrast immunofluorescence images ², they often struggle when applied to OCT-A data, which are typically low in contrast and prone to imaging artifacts ^{3–5}. Furthermore, variations in image

quality across acquisition protocols and devices lead to difficulties in accurately delineating fine capillaries within dense MNV networks and result in inconsistent quantitative parameters across machines ⁶.

Another critical hurdle in MNV analysis is the characterization of vascular remodeling, particularly the distinction between active neovascularization and mature quiescent states. While patterns like "Medusa" and "Seafan" are widely recognized, their quantitative definition and clinical interpretation often vary ^{7,8}. Furthermore, the term "mature" is used inconsistently in the literature, leading to significant ambiguity. It sometimes describes a well-organized but active lesion (morphological maturity), and other times a quiescent, pruned vascular tree (pathophysiological maturity) ^{1,7,8}. This terminological ambiguity hinders standardized assessment and cross-study comparisons.

This paper introduces a novel, semi-automated system designed to resolve these ambiguities. We present a systematic comparison of our parameters against the latest literature and detail an automated morphological-pathophysiological classification logic that maps specific morphological patterns - Medusa, Seafan, Glomerular, Tree-in-bud, and Dead-tree - to their underlying pathophysiological states (Active, Mature Quiescent, Transitional, Arteriolarized). This logic leverages quantitative biomarkers like the Euler number, Loop count, and Vascular Stability Score, integrating them with established clinical criteria proposed previously ⁹.

Methods

Study Population and Acquisition Protocols

This study was approved by the Ethics Committee of the Yokohama City University Medical Center. The study protocol adhered to the tenets of the Declaration of Helsinki and informed consent was obtained from all eligible patients. This study included 112 MNV lesions imaged with three distinct OCTA platforms and field-of-view configurations to establish a multi-device validation framework. The large group (n=49) comprised 6×6mm acquisitions on the Zeiss PlexElite 9000 (swept-source OCTA, 1060nm). The small_3mm group (n=30) comprised 3×3mm acquisitions on the Zeiss CIRRUS HD-OCT with AngioPlex (spectral-domain OCTA, 840nm). The small group (n=33) comprised 6×6mm acquisitions on the Optovue Solix module (swept-source OCTA, 1060nm). All lesions were diagnosed as Type 1 or Type 2 MNV secondary to neovascular AMD by retinal specialists based on structural OCT and multimodal imaging criteria. The use of three independent hardware platforms - differing in manufacturer, OCTA modality, central wavelength, scanning protocol, and field size - was intentional: it provides an inherent multi-device validation framework for the proposed standardization approach.

Analytical Framework

The analytical framework was initially created as a comprehensive macro for ImageJ and later implemented in Python.

The methodology comprised two parallel components: mapping the current landscape of quantitative biomarkers and developing a novel analytical pipeline to compute and further refine these metrics. A focused literature search was conducted to identify studies reporting quantitative OCT-A analyses of MNV. From this review, key quantitative parameters were extracted, and several new metrics were introduced to achieve a more comprehensive characterization of MNV morphology and flow dynamics, as described below.

The core of our framework is a semi-automated pipeline that operates following an initial freehand delineation of the MNV lesion by the user. This input defines the region of interest (ROI) for subsequent automated processing. The pipeline comprises three primary stages.

Artifact Mitigation and Spatial Organization

This stage implements novel solutions to the most critical flaws in existing analysis software.

1 ROI Refinement: To prevent boundary artifacts, an iterative algorithm automatically adjusts the user-drawn ROI. For 5 iterations, each vertex of the ROI polygon is evaluated. A search is conducted within a 3-pixel radius, and the vertex is moved to the pixel with the minimum intensity (darkest pixel), effectively pushing the boundary into non-perfused tissue. This ensures the analytical boundary conforms to the true lesion edge.

2. Hybrid Multi-Scale Vessel Enhancement: A combination of Frangi and Laplacian of Gaussian (LoG) filters is applied to accentuate vessels of varying calibers. This hybrid approach is based on recent evidence demonstrating that the fusion of multiple, complementary filters significantly improves vessel detection accuracy over single-filter methods¹⁰⁻¹². The Frangi filter (scales: 0.8-4.0) excels at identifying tubular structures and the LoG filter detects edges. Their combined use provides a more robust and sensitive enhancement, crucial for the complex and multi-scale nature of MNV networks, particularly in low-contrast OCT-A images.

3. Binarization: The Phansalkar local adaptive thresholding method is used to create a binary representation of the vascular network, preserving fine vessel details. The radius is dynamically calculated based on image resolution to correspond to 24 μ m, with $k=0.1$ and $R=0$.

4. Post-processing and Skeletonization: The binary image is refined through morphological reconstitution. The final, cleaned image is then skeletonized to produce a one-pixel-wide centerline representation for topological analysis.

5. Boundary Branch Exclusion: To prevent skewed measurements from incomplete vessels, a spatial classification protocol is employed. The refined ROI is logically divided into a "Center" zone and a "Periphery" zone. Branches existing solely within the Periphery or spanning both zones are flagged as "Boundary Branches" and are excluded from calculations of average branch length, junction density, and tortuosity.

Quantitative Analysis

This stage computes a comprehensive set of parameters, including all major metrics identified in our literature review, and introduces several novel, integrated scores.

Table1 provides the metrics obtained in this study. To objectively identify pathologically dilated segments, the system uses a statistical threshold of mean + 2.0 * SD of all vessel diameters. Contiguous segments exceeding this threshold are flagged, and their count, length, and density are reported. This provides an adaptive, objective measure of vascular remodeling.

Standardized Score Computation

Standardized Vascular Complexity Score (0–100)

The score was derived using principal component analysis (PCA) of four topological metrics computed from the skeletonized MNV network: inverse Euler number ($-\text{Euler}_{\text{total}}$), total loop count, junction density, and global fractal dimension. Reference distributions were constructed independently for each acquisition stratum (large, small_3mm, small) using all lesions within that stratum. Features were standardized using stratum-specific means and standard deviations prior to PCA. The dominant first principal component (PC1), carrying uniformly positive loadings across all four metrics, explained 63.7%, 73.9%, and 70.2% of total variance for the large, small_3mm, and small strata, respectively, confirming that a single latent axis captures overall network architectural complexity in each acquisition context. A secondary component (PC2, loading primarily on junction density) captured residual variance (24.8%, 21.9%, and 22.3%). PC1 and PC2 scores were combined with a trunk distribution metric (TrunkDist; fixed at 50 in the absence of device-specific calibration data) using weights of 0.7, 0.2, and 0.1, respectively. To ensure a common 0–100 scale across platforms, both PC1 and PC2 scores were independently subjected to piecewise-linear normalization in which the within-stratum median maps to 50, the stratum minimum maps to 0, and the stratum maximum maps to 100.

Standardized Caliber Uniformity Score (0–100)

Vascular caliber uniformity was quantified using four radial profile metrics derived from the diameter-versus-distance profile: (1) coefficient of variation of vessel diameter (CV); (2) mean adjacent inter-bin change; (3) residual CV after linear detrending; and (4) diameter range normalized to mean diameter. Stratum-specific PCA was applied to these four metrics. The dominant PC1 carried uniformly positive loadings across all four instability indicators, explaining 89.6%, 81.1%, and 87.3% of variance for the large, small_3mm, and small strata, respectively, confirming that a single latent instability axis dominates caliber variability. PC1 was sign-inverted ($-\text{PC1}$) to obtain a stability-direction score, and piecewise-linear normalization (median $\rightarrow$ 50) was applied independently to $-\text{PC1}$ and PC2. The final Standardized Caliber Uniformity Score was computed as $0.7 \times (-\text{PC1 score}) + 0.2 \times \text{PC2 score} + 0.1 \times 50$.

Standardized Maturity Index (0–100)

The Standardized Maturity Index was defined as: $\text{Maturity Index} = \text{clip50} + (\text{Caliber Uniformity Score} - \text{Complexity Score}) / 2$, 0, 100. This formulation reflects the pathophysiological concept that vascular maturity is characterized by increasing caliber uniformity (stability) relative to network architectural complexity: values above 50 indicate a maturity-dominant state consistent with vascular remodeling or quiescence, while values below 50 indicate a complexity-dominant state consistent with active angiogenesis.

Statistical Analysis

Cross-device comparability of standardized scores was assessed using the Kruskal–Wallis test across the three acquisition strata. All analyses were performed in Python (version 3.11) using the scikit-learn library for PCA and the SciPy library for statistical testing. A p-value of < 0.05 was considered statistically significant.

Results

Robust Segmentation and Artifact Mitigation

Representative OCT-A images indicate that the system provides consistent segmentation of MNV lesions, with the semi-automatically delineated lesion area highlighted in yellow and the dilated neovascular segment shown in red (Figure). In these examples, the segmented neovascular network follows the lesion architecture rather than spurious high-contrast boundaries. The Intelligent ROI Refinement algorithm reduces boundary artifacts by adjusting user-drawn ROIs to the non-perfused tissue border, thereby limiting the influence of artificial edges on subsequent quantitative analyses. The Boundary Branch Exclusion protocol further limits the contribution of incomplete vessel segments at the ROI periphery to topological and morphometric metrics.

Cross-Device Raw Metrics and the Necessity of Standardization

Table 2 presents the raw topological and morphometric parameters across the three acquisition protocols. As expected from the differences in imaged vascular territory and device-specific resolution characteristics, raw metrics differed substantially across platforms. Mean total loop count ranged from 300.3 ± 199.2 in the large (PlexElite 6×6mm) group to 165.2 ± 118.0 in the small_3mm (HD series 3×3mm) group and 89.5 ± 53.4 in the small (Solix 6×6mm) group - a 3.4-fold difference between the highest and lowest values. Similarly, mean Euler number ranged from -168.4 ± 146.0 to -61.6 ± 48.2 , and mean junction density from 25.85 ± 1.77 to $15.89 \pm 2.88 \text{ mm}^{-2}$. Mean vessel diameter showed the reverse ordering (16.0, 23.7, and 32.3 μm for large, small_3mm, and small, respectively), reflecting the inverse relationship between field size and measured vessel caliber. These systematic differences confirm that direct cross-device comparison of raw parameters is not feasible and motivate the PCA-based standardization framework described in the Methods.

Convergence of Standardized Scores Across Devices

Table 3 presents the standardized scores following device-specific PCA and piecewise-linear normalization. Despite the substantial differences in raw metrics, the Standardized Complexity Score converged to statistically indistinguishable median values of 48.0 (IQR 42.4–55.4), 49.2 (45.7–57.8), and 49.9 (41.8–57.9) for the large, small_3mm, and small groups, respectively (Kruskal–Wallis, $p=0.276$). The high PC1 explained variance ratios (63.7–73.9%) across all three strata confirm that a single dominant latent axis captures the principal structure of MNV architectural complexity in each acquisition context, and that this axis is biologically consistent across devices. Analogous convergence was observed for the Standardized Caliber Uniformity Score (medians 50.6, 50.3, 49.9; $p=0.636$), for which PC1 explained 81.1–89.6% of variance, and for the Standardized Maturity Index (medians 51.3, 51.4, 49.6; $p=0.764$). The convergence of all three scores to values within one unit of 50 - achieved through within-device normalization without any cross-device calibration - provides empirical evidence that the proposed framework effectively removes device- and field-of-view-dependent scaling while preserving the underlying biological information.

Automated Morphological Classification

Table 4 presents the distribution of morphological subtypes across acquisition platforms. The full spectrum of MNV subtypes was identified across all three devices. Glomerular pattern was the most prevalent subtype in the large group ($n=29$, 59.2%) and was also well-represented in the small_3mm and small groups (30.0% and 36.4%, respectively). Seafan pattern was more common in the small_3mm group ($n=11$, 36.7%) than in the other groups. Dead-tree pattern, reflecting mature quiescent MNV, was identified in all three groups (8.2%, 20.0%, and 15.2%). Tree-in-bud pattern was present in all groups (20.4%, 13.3%, 42.4%). The qualitative consistency of subtype representation across acquisition platforms provides initial evidence for the cross-device robustness of the classification logic.

Discussion

This work introduces a significant advancement in the quantitative analysis of MNV by systematically addressing the limitations of current methodologies and introducing a new dimension of integrated, multi-parameter scoring. Our comparative analysis (Table 1) clearly illustrates that while existing tools and recent studies provide a foundational set of metrics, they fail to capture the holistic nature of the neovascular complex and are vulnerable to significant methodological artifacts.

A defining feature of the present system is its capacity to generate comparable morphological scores across fundamentally different acquisition conditions. In contrast to prior OCTA analysis tools that report raw, device-specific metrics unsuitable for cross-platform comparison, our PCA-based normalization framework produces three standardized scores - the Standardized Complexity Score, Standardized Caliber Uniformity Score, and Standardized Maturity Index - that are anchored to a common scale irrespective of the acquiring device or field-of-view. The empirical demonstration of score convergence across the Zeiss PlexElite 9000 (6×6mm), Zeiss HD series (3×3mm), and Optovue Solix

(6×6mm) platforms - three devices from two manufacturers spanning two field-of-view configurations - provides evidence that this convergence is not a mathematical artifact but reflects the extraction of a consistent biological signal. The high and consistent PC1 explained variance ratios (63.7–73.9% for Complexity; 81.1–89.6% for Caliber Uniformity) across strata further indicate that a single dominant latent axis underlies MNV morphological complexity and caliber stability regardless of acquisition platform, lending biological plausibility to the standardization approach. To our knowledge, this is the first quantitative MNV scoring framework explicitly validated across commercially available OCTA devices from different manufacturers and with different field-of-view configurations.

Several key aspects of our study merit emphasis. First, the Intelligent ROI Refinement algorithm represents a novel solution to a long-standing challenge in quantitative OCT-A analysis^{7,13–15}. By algorithmically ensuring that the ROI boundary is confined to non-perfused tissue, the algorithm provides a more accurate and reproducible foundation for all subsequent calculations.

Recent studies have underscored the utility of the Euler number as a reliable measure of network connectivity^{16–18}. The Euler number quantifies loop formation within a network, where increasingly negative values denote higher connectivity and structural complexity. We adopted the Euler number and loop count as core elements of the Vascular Complexity Score because these metrics offer a robust framework to quantify the connectivity and structural organization of MNV. For example, immature neovascularization, driven by active angiogenesis, manifests as a dense, highly interconnected, and often chaotic plexus of fine capillaries forming a reticulated or honeycomb-like pattern¹⁹. Euler number from the skeletonized MNV provides a direct, quantitative biomarker of its mesh-like state - a highly negative Euler number, in conjunction with an elevated loop count, strongly indicates an immature, actively proliferating, and potentially treatment-responsive lesion. Active MNV is also characterized by a highly complex and densely interconnected vascular architecture consistent with ongoing angiogenesis. As such a markedly negative Euler number reflects extensive vascular connectivity and numerous loop formations. Clinically, this configuration corresponds to active disease requiring treatment and is typically associated with a favorable response to anti-VEGF therapy.

Terms such as “dead tree,” “tree in bud,” “medusa,” “sea fan,” and “glomerular” have previously been applied in a primarily descriptive and subjective manner^{20–22}. In contrast, the proposed framework provides operational definitions for these lesion phenotypes. Lesions corresponding to the “dead tree” pattern are characterized in this system by large-caliber trunk vessels with limited fine branching, high Vascular Stability Scores, low junction density, and an Euler number approaching zero, indicating loop loss and network simplification consistent with nonexudative, low-angiogenic MNV. Medusa and glomerular patterns, both reflecting dense, high-flow vascular networks, are represented as highly complex, loop-rich configurations occupying the upper range of the Vascular Complexity Score, while remaining distinguishable based on differences in trunk distribution and junction density. By expressing both stabilization and arteriolarization in quantitative terms and demonstrating that the corresponding scores converge across acquisition protocols, this framework recasts qualitative pattern labels as

reproducible, cross-device measurable states that separate benign remodeling from treatment-resistant vascular transformation and refine prognostic interpretation.

Lastly, our work addresses a long-standing terminological ambiguity in the MNV literature. Terms such as “maturation,” “arterialization,” and “abnormalization” have been used inconsistently and at times interchangeably^{23–25}. This lack of standardization has impeded objective assessment of treatment response. Our framework contributes to resolving this issue by directly mapping these biological processes to quantitative metrics. In addition, Arterialization, defined as the formation of large-caliber, potentially treatment-resistant vessels, is captured by our Arteriolarization Segment Detection algorithm. The unstable vascular state described by Spaide as abnormalization²⁶ is quantified by the Vascular Stability Score, with low values indicating an unstable, treatment-responsive phenotype and high values reflecting maturation and stabilization.

Conclusion

We have developed and validated a novel, semi-automated ImageJ-based system that represents a paradigm shift in the quantitative analysis of macular neovascularization. By systematically addressing the methodological flaws of existing tools, incorporating all major established biomarkers from the most recent literature, and introducing a suite of innovative, integrated scores for complexity, stability, and spatial organization, our system provides the most comprehensive and accurate characterization of MNV architecture to date. Crucially, it is the first system to provide quantitative tools to differentiate the clinically critical pathways of arterialization and abnormalization, and to offer an automated morphological-pathophysiological classification logic that resolves long-standing terminological ambiguities. This powerful new platform standardizes research, accelerates the discovery of prognostic imaging biomarkers, and ultimately enhances the clinical management of neovascular AMD.

Declarations

Conflict of Interest

Y. Yanagi Consultant/Speaker for Astellas Pharmaceutical, Bayer Yakuhin Ltd, Roche/Chugai Pharmaceutical Co., Ltd., Novartis Pharma K.K., Boehringer Ingelheim Co., Ltd., Santen Pharmaceutical Co., Ltd., Senju Pharmaceutical Co.

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Y.Y. wrote the main manuscript text and T.I. and M. I. prepared figure. All authors reviewed the manuscript.

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Tables

Table 1. Quantitative parameters of analysis platforms

Category	Parameter	Description & Definition
Morphometry	Vessel Density (VD)	Proportion of perfused lesion area: (binarized vessel pixels inside refined ROI / total ROI pixels) × 100%.
	Vessel Area (VA)	Absolute perfused area: binarized vessel pixels inside refined ROI × pixel area (mm ²).
	Total Vessel Length	Sum of all skeletonized centerline lengths: skeleton pixels × pixel size (mm).
	Mean / Max Diameter	Local vessel diameter (2 × Euclidean distance from skeleton to background). Mean and max computed over all skeleton pixels in ROI.
Topology	Junction / Node Density	Total node count (branch/endpoints) divided by physical ROI area (nodes/mm ²).
	Loop Count	Total number of independent closed vascular loops (graph-based cycle detection after Boundary Branch Exclusion).
	Euler Number	Global connectivity index: (number of connected components) – (number of loops). More negative = higher connectivity.
Complexity	Fractal Dimension (Df)	Box-counting dimension of the skeletonized MNV, derived from the slope of the log–log plot.
	Tortuosity Index	Mean segment tortuosity: (path length along skeleton / straight-line distance) averaged over eligible branches.
	Standardized Complexity Score (0–100)	PCA-based latent score: $0.7 \times \text{PC1} + 0.2 \times \text{PC2} + 0.1 \times \text{TrunkDist}$. Normalized so min, median, max map to 0, 50, 100.
Stability	Standardized Caliber Uniformity Score (0–100)	PCA-based latent stability from 4 radial metrics: $0.7 \times (-\text{PC1}) + 0.2 \times \text{PC2} + 0.1 \times 50$. Median fixed at 50.
	Diameter CV	Coefficient of variation: (SD of local diameters / mean diameter) × 100%.
Advanced	Intelligent ROI Refinement	5-iteration vertex optimization (3-pixel search radius) to align ROI boundary with non-perfused tissue.
	Arteriolarization Detection	Identification of branches where local diameter > mean + 2.0 × SD. Reports count, length, and area density.
	Automated Morphological Classification	Rule-based classifier assigning patterns (Medusa, Seafan, etc.) to pathophysiological classes (Active, Mature, etc.).
Multi-device	Device-Agnostic Score Normalization	Within-device standardization and stratum-specific PCA to align all devices to a common 0–100 axis.

Field-of-View Correction	Physical unit conversion using device-specific scaling to ensure comparability across 3 × 3mm and 6 × 6mm scans.
Cross-Platform Score Validation	Statistical equivalence confirmation using non-parametric tests (e.g., Kruskal–Wallis) ensuring medians converge at 50.
Standardized Maturity Index (0–100)	$\text{clip}(50 + (\text{CaliberUniformityScore} - \text{ComplexityScore}) / 2, 0, 100)$. Scores > 50 indicate stability-dominance; scores < 50 indicate complexity-dominance.

Table 2. Raw topological and morphometric parameters by acquisition protocol

Parameter	Large PlexElite 6×6mm (n = 49)	Small_3mm HD series 3×3mm (n = 30)	Small Solix 6×6mm (n = 33)
Topological metrics			
Total Loop Count	300.3 ± 199.2	165.2 ± 118.0	89.5 ± 53.4
Euler Number (total)	−168.4 ± 146.0	−135.0 ± 98.4	−61.6 ± 48.2
Junction Density (mm ^{−2})	25.85 ± 1.77	21.99 ± 3.51	15.89 ± 2.88
Morphometric metrics			
Fractal Dimension	1.391 ± 0.081	1.378 ± 0.076	1.310 ± 0.110
Mean Vessel Diameter (μm)	16.0 ± 0.3	23.7 ± 2.0	32.3 ± 3.8
Caliber stability metrics (pre-standardization)			
Diameter CV (%)	14.7 ± 6.8	16.8 ± 10.9	19.4 ± 10.3
Diameter Range / Mean (%)	50.4 ± 25.5	55.3 ± 35.1	65.9 ± 37.1

Values are mean ± SD. Three acquisition protocols were employed: 6×6mm field on the Zeiss PlexElite 9000 (large); 3×3mm field on the Zeiss HD series (small_3mm); 6×6mm field on the Heidelberg Solix (small). Raw metrics differ substantially across protocols owing to field-of-view and device-dependent characteristics. Total loop count and Euler number reflect the complete network including both center and periphery zones. Mean vessel diameter shows the inverse relationship expected with field size. These systematic differences preclude direct cross-device comparison of raw parameters and motivate the PCA-based standardization described in the Methods.

Table 3. Standardized scores following PCA-based normalization, by acquisition protocol

	Large PlexElite 6×6mm (n = 49)	Small_3mm HD series 3×3mm (n = 30)	Small Solix 6×6mm (n = 33)	p- value†
Standardized Complexity Score				
PC1 explained variance	63.7%	73.9%	70.2%	-
Median (IQR)	48.0 (42.4–55.4)	49.2 (45.7–57.8)	49.9 (41.8–57.9)	0.276
Mean ± SD	48.4 ± 12.2	54.0 ± 12.8	51.4 ± 13.5	NS
Standardized Caliber Uniformity Score				
PC1 explained variance	89.6%	81.1%	87.3%	-
Median (IQR)	50.6 (41.3–60.1)	50.3 (48.2–56.2)	49.9 (43.2–64.5)	0.636
Mean ± SD	50.8 ± 14.4	52.8 ± 12.8	52.9 ± 19.1	NS
Standardized Maturity Index				
Median (IQR)	51.3 (48.6–54.1)	51.4 (46.8–54.3)	49.6 (43.6–56.2)	0.764
Mean ± SD	51.2 ± 5.0	49.4 ± 7.6	50.7 ± 8.6	NS

Values are median (IQR) and mean ± SD. PC1 explained variance ratios reflect stratum-specific PCA of four input features (Complexity: inverse Euler number, loop count, junction density, fractal dimension; Caliber Uniformity: diameter CV, mean adjacent change, residual CV, diameter range/mean). Piecewise-linear normalization was applied independently to PC1 and PC2 scores within each stratum, anchoring the within-stratum median at 50. The Standardized Maturity Index = clip[50 + (Caliber Uniformity Score – Complexity Score) / 2, 0, 100]. †Kruskal–Wallis test across three acquisition groups. IQR, interquartile range; NS, not significant.

Table 4. Morphological subtype distribution by acquisition protocol

Morphological Pattern	Large PlexElite 6×6mm (n = 49)	Small_3mm HD series 3×3mm (n = 30)	Small Solix 6×6mm (n = 33)	Pathophysiological State
Glomerular	29 (59.2%)	9 (30.0%)	12 (36.4%)	Active
Medusa	6 (12.2%)	0 (0%)	0 (0%)	Active
Seafan	0 (0%)	11 (36.7%)	2 (6.1%)	Active
Tree-in-bud	10 (20.4%)	4 (13.3%)	14 (42.4%)	Active / Transitional
Dead tree	4 (8.2%)	6 (20.0%)	5 (15.2%)	Mature Quiescent

Values are n (%). Five morphological subtypes were identified: Glomerular, Medusa, Seafan, Tree-in-bud, and Dead-tree. The full spectrum of subtypes was represented across all three acquisition platforms, providing evidence for the cross-device robustness of the automated classification logic. Subtype prevalence differences across platforms may partly reflect differences in cohort composition (treatment-naïve vs. post-treatment proportion) in addition to genuine platform effects.

Table 5. Morphological-Pathophysiological Correlation Matrix

Morphological Pattern	Primary State	Secondary State	Key Discriminating Metrics
Medusa	Active	Transitional	Junction Density, Loop Count, Trunk Distribution
Seafan	Active	Transitional	Peripheral Arcade, Trunk eccentricity
Glomerular	Active	-	High Complexity Score, Low Caliber Uniformity Score
Tree-in-bud	Active	Transitional	High branching density, Euler number
Dead tree	Mature Quiescent	Arteriolarized	Mean Diameter, Caliber Uniformity Score
Pruned tree	Mature Quiescent	Transitional	Caliber Uniformity Score, Vessel Density
Large vessels	Arteriolarized	Abnormalized	Diameter CV, Arteriolarization Index

The correlation matrix maps each morphological pattern to its most likely pathophysiological state and the key discriminating metrics used by the automated classification logic. Primary state indicates the dominant pathophysiological interpretation; secondary state indicates an alternative state that may be assigned when threshold criteria are not fully met. Key discriminating metrics are those with the highest weight in the automated classification decision.

Figures

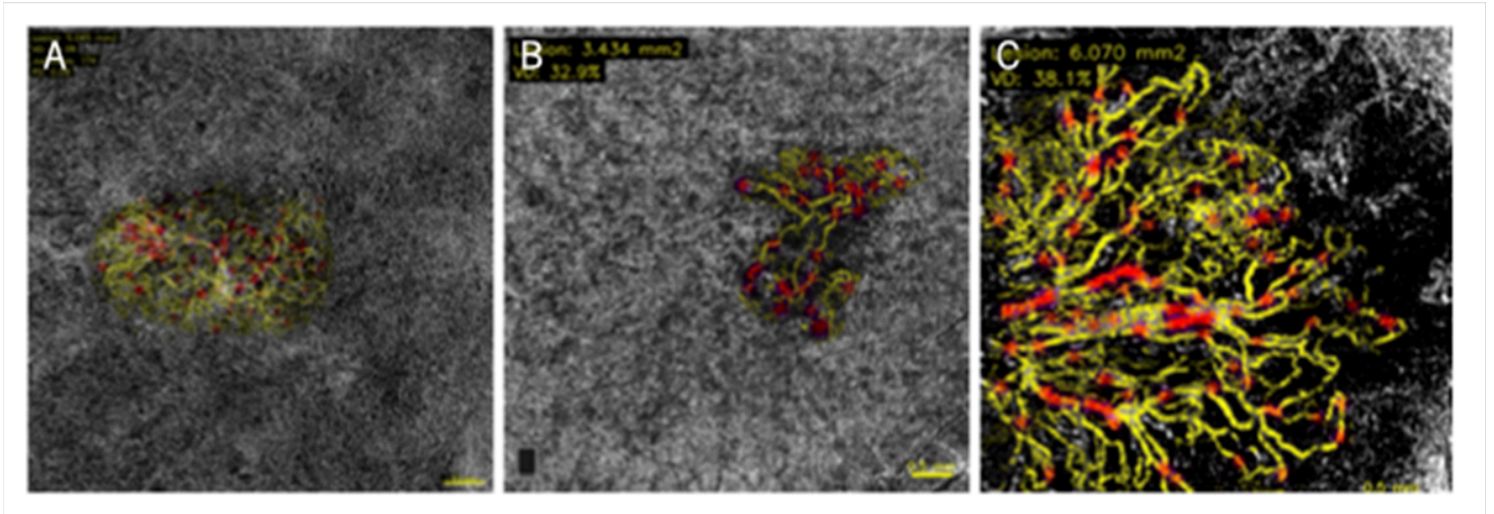


Figure 1

Representative OCT-A images demonstrating consistent MNV segmentation across multiple platforms.

Images were acquired using: **(A)** Zeiss PlexElite 9000 (6×6 mm), **(B)** Optovue Solix (6×6 mm), and **(C)** Zeiss HD series (3×3 mm). The system provides consistent segmentation of MNV lesions across these devices. The semi-automatically delineated lesion areas (yellow) and dilated neovascular segments (red) accurately follow the internal lesion architecture. Note that the region of interest (ROI) is refined to the non-perfused tissue border to minimize boundary artifacts, and incomplete peripheral vessel segments are excluded from the quantitative analysis to ensure morphometric accuracy.