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Retinal Vascular Segmentation Using Superpixel-based Line Operator and Its Application to Vascular Topology Estimation

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Abstract

Purpose: Automatic methods of analyzing of retinal vascular networks, such as retinal blood vessel detection, vascular network topology estimation, and arteries / veins classification are of great assistance to the ophthalmologist in terms of diagnosis and treatment of a wide spectrum of diseases.

Methods: We propose a new framework for precisely segmenting retinal vasculatures, constructing retinal vascular network topology, and separating the arteries and veins. A non-local total variation inspired Retinex model is employed to remove the image intensity inhomogeneities and relatively poor contrast. For better generalizability and segmentation performance, a superpixel based line operator is proposed as to distinguish between lines and the edges, thus allowing more tolerance in the position of the respective contours. The concept of *dominant sets clustering* is adopted to estimate retinal vessel topology and classify the vessel network into arteries and veins.

Results: The proposed segmentation method yields competitive results on three public datasets (STARE, DRIVE, and IOSTAR), and it has superior performance when compared with unsupervised segmentation methods, with accuracy of 0.954, 0.957, and 0.964, respectively. The topology estimation approach has been applied to five public databases

(DRIVE,STARE, INSPIRE, IOSTAR, and VICAVR) and achieved high accuracy of 0.830, 0.910, 0.915, 0.928, and 0.889, respectively. The accuracies of arteries / veins classification based on the estimated vascular topology on three public databases (INSPIRE, DRIVE and VICAVR) are 0.90.9, 0.910, and 0.907, respectively.

Conclusions: The experimental results show that the proposed framework has effectively addressed crossover problem, a bottleneck issue in segmentation and vascular topology reconstruction. The vascular topology information significantly improves the accuracy on arteries / veins classification.

Keywords: retinal vascular, segmentation, topology, superpixel, line operator, dominant sets

1 Introduction

Analysis of retinal vascular structure is imperative for clinical applications to support examination, early detection, diagnosis and optimal treatment of eye disease. This has the potential to perform automated screening for pathological conditions, and to provide crucial hints on various diseases [1, 2, 3, 4, 5, 6], in particular diabetic retinopathy (DR), malaria retinopathy (MR), glaucoma, and hypertensive retinopathy.

The above-mentioned diseases often cause vascular abnormalities, amongst which changes in vascular caliber and tortuosity are the most common ones. It is crucial to be able to identify and characterise the structure of individual vessels from the entire retinal vessel network. This calls for precise description of vascular structure in terms of geometrical and topological properties from retinal images.

Manual annotation of vascular structure is an exhausting task for graders, and computer-aided automatic/semi automatic vascular detection methods can significantly reduce the time consumption. Over the past two decades, a tremendous amount of vessel segmentation methods have been developed for different types of medical images.

Numerous fully automated, and semi-automated methods have been proposed, as evidenced by extensive reviews [7, 8, 9]. In general, all established automated segmentation methods may be categorized as either supervised segmentation [10, 11, 12, 13, 14, 15] or unsupervised segmentation [16, 17, 18, 19, 9, 20, 21] regarding the overall system design and architecture. Unsupervised segmentation refers to methods that achieve the segmentation of blood vessels without using train-

ing data or explicitly using any supervised classification techniques [22]. This category includes most segmentation techniques in the literature, such as active contour models [19, 23], wavelets [20], line operator[16] and our new framework, as described in this paper. In contrast, supervised methods [10, 11, 12, 13, 14, 24, 25] require a manually annotated set of training images for classifying a pixel either as vessel or non-vessel. Most of these methods in supervised catergory use Support Vector Machine (SVM), AdaBoost, Neural Networks, Conditional Random Field (CRF), etc.

By contrast, automated estimation of retinal vascular topology is still understudied despite its significance in understanding the structure of vessels. To the best of our knowledge, only a small number of studies have addressed this subject directly.

A semi-automatic method of measuring and quantifying the topological properties of retinal vessels was proposed by Martinez-Perez et al. [26], which is considered to be the first work on retinal vascular topology estimation. Measurements of length, area, angles and connectivity between branches were taken from the labeled segmented vessel trees. Qureshi et al. [27] used a Bayesian approach to address the configuration of vascular junctions, and utilized a probabilistic model and Maximum A Posterior (MAP) to construct the vascular trees. Estrada et al. [28] regularized the topology estimation problem with a generative, parametric tree-growth model. A combination of greedy approximation and heuristic search algorithm was proposed to explore the space of possible trees. This method has not only applied on retinal vessels, but also on plant roots, and synthetic tree data. De et al. [3, 29] proposed a graph-theoretical method to trace tree structures in neuronal and retinal images. The topology estimation problem was reformulated as label propagation over directed graphs: in this way the graph is decomposed into sub-graphs, and each vessel tree may be separated from the vessel network. Another graph-based approach for retinal vessel topology estimation was introduced by Dashtbozorg et al. [30]. They classified the entire vessel networks depending on the type of graph node and assigned one of two labels to each vessel segment.

However, numerous factors cause inaccuracy in vascular structure analysis (vessel segmentation and topology estimation), such as the high degree of anatomical variation across the population, the complexity of the surrounding tissue, varying scales of vessels within an image, and pathologies (e.g., micro-aneurysms, hemorrhages, and exudate). Moreover, during image acquisition, such as noise, poor contrast and low resolution, exacerbate this problem.

In this paper we propose a novel vascular structure analysis framework, which furthermore

extends the approach proposed in [31] from the case of vessel segmentation, and thus making it applicable to vascular topology estimation and arteries / veins classification. The contributions of this work may be summarized as three folds: **1)** The sensitivity for the detection of vessels is significantly improved after the applying the Retinex-based inhomogeneity correction and superpixel-based line operator. It achieves competitive performance in the comparison studies on four publically available retinal image datasets with different imaging modalities. **2)** The concept of dominant sets clustering [32, 33] was introduced to tackle the problem of vessel topology estimation and proved to be an efficient way of addressing problems in tracing crossovers. In addition, the underlying vessel topology is able to better distinguish arteries from veins. This has been validated quantitatively using three publicly accessible datasets with promising results. **3)** We have established manual annotations of vessel topologies of three publicly available datasets, and these annotations will be released for public access to facilitate other researchers in the community to do research and development on the same and related topics after the paper has been accepted.

2 Method

In this section, we describe the proposed framework for the extraction of vessels and topology estimation. It comprises the following main phases. **1)** A non-local total variation regularized intensity inhomogeneity correction is adopted to correct the imbalanced illumination of the retinal image. **2)** A superpixel enabled line operator is used for vessel segmentation. **3)** A skeletonization method is then used to generate the vessel centerline map from the segmented results, based on which significant points, such as bifurcation, crossing, intersection points will be identified. **4)** The significant points are then utilized to create a graph. **5)** The dominant sets concept is used to classify the significant nodes, in order to estimate the vessel topology. The main steps of our approach are illustrated in Figure 1.

2.1 Inhomogeneity Correction

Intensity inhomogeneity poses a significant challenge to image processing tasks, for instance, retinal images acquired with a fundus camera sometimes have poor contrast due to too strong or too low illumination conditions inherited from image acquisition. To this end, an inhomogeneity correction

method is proposed to handle these problems.

In this paper, a non-local total variation (TV) regularized model supporting the Retinex theory is employed. The TV regularizer is very effective in recovering edges of images [34]. Such phenomenon coincides with the partial differential equation based Retinex method: the gradient of the reflectance corresponds to the sharp details in the image, and the illumination is spatially smooth. Hence, the regularization can be formulated as a minimization problem: the regularization terms is able to find the sharp details, and the L_2 norm smooth the illumination. For more details, we refer readers to the paper [23]. Note, the parameter t of Eq.(1) in [23] balances the regularization and L_2 norm, and is set as 0.6 in this work. Figure 2 shows two enhanced results produced by applying the non-local TV based Retinex model. It has successfully corrected the contrast between vessels and background, as well as the region of the optic disc. In consequence, the vessels are more easily identifiable.

2.2 Superpixel-based Line Operator for Vessel Segmentation

The line operator is a common choice to generate the vesselness map. Note, the *vesselness* map represents the probability of a given pixel being part of a vessel. The basic line operator considers 12 angles with angular resolution of 15 degrees. The largest average grey level $\mathbf{L}$ is found, which the pixel lies on a line passing through the target pixel. Then the line strength of the pixel is calculated by

$$\mathbf{S}(i) = \mathbf{L}(i) - \mathbf{N}(i), \tag{1}$$

where $\mathbf{N}(i)$, is the average grey-level of a square window, centered on the target pixel i, with edge length equal to μ. The winning line is aligned within a vessel if the line strength is large, while the line strength is lower if the line is partially overlapped. In general, the length μ is empirically chosen, such as 15 in [16], and 5 in [35].

However, there usually are varying scales of vessels within an image, and a single value of μ tends to yield imbalance responses on the vessels. Therefore, in order to achieve better segmentation performance, in this work we applied a modified line operator on the superpixel generated patches rather than on the entire image, in particular in regions with low signal noise ratio. The length μ was set to be half of the minimum object length of corresponding superpixel. (*Minimum object*

length: length in pixels of the minor axis of the ellipse that has the same normalized second central moments as the region, returned as a scalar.)

To achieve this, we first generate the superpixel upon the vesselness map. The SLIC superpixel algorithm [36] is adapted to replace the rigid structure of the pixel grid. The SLIC is a k-means clustering based method, and is able to assign each pixel to a superpixel according to their intensities and spatial locations. The superpixel clustering procedure starts with the generation of initial cluster centers. Then a distance measure D to cluster centers for all pixels is defined, aims to associate to their nearest cluster centers. The Euclidean distance (d_c) and spatial distance (d_s) are used to define this measure:

$$D = \sqrt{d_c^2 + (\frac{d_s}{S})^2 m^2}, \tag{2}$$

where $S = \sqrt{N/k}$ is the grid interval. k is the desired superpixel number and N is the total number of pixels. m indicates a parameter to balance the weighting of intensity and coordinates. Figure 3 shows an example of superpixel representation, with 400, 800, and 1200 superpixels, respectively.

Let $\mathcal{P}_t \in T$ be a viable local representation as a superpixel t ($t = 1, 2, \cdots, T$), and let I indicate the input image. The line strength of the pixel in superpixel $\mathcal{P}$ is defined as $\mathbf{S}_{\mathcal{P}_t}(i) = \mathbf{L}_{\mathcal{P}_t}(i) - \mathbf{N}_{\mathcal{P}_t}(i)$. In practice, the line path rarely matches the pixel grid, hereby, the line and region averages at arbitrary orientations are obtained by using nearest neighbour interpolation instead of bi-linear interpolation.

Multiscale analysis is also performed in this framework. The line strength of the pixel under multi-level superpixel is defined as

$$\mathbf{S}(i) = \frac{1}{P} \sum_{p=1}^{P} \mathbf{S}(i)(\mathcal{P}_t^p | i \in \mathcal{P}_t^p). \tag{3}$$

where P indicates the levels of superpixels that the input image is segmented to. Parameter tuning for optimal numbers of superpixels and levels (P and M) will be discussed in Section 3.1. The second column of Figure 4 demonstrate the final vessel responses of the proposed method. In order to extract the vessel from the response map, our previous proposed infinite perimeter active contour with hybrid region (IPACHR) method [37] is employed for its good performance. The IPACHR uses an infinite perimeter active contour model for its effectiveness in detecting vessels with irregular and oscillatory boundaries. For more details, we refer readers to the original paper [37]. The third

column of Figure 4 depict the segmentation results.

2.3 Vascular Graph Generation

An iterative morphology thinning operation [20] is performed on the vessel segmentation results to obtain a single-pixel-wide skeleton map. The generated skeleton map is shown in Figure 5 (B). The vascular bifurcation/crossover points, and vessel ends (terminal points) can be extracted from the skeleton map by locating intersection points (pixels with more than two neighbors) and terminal points (pixels with just one neighbor). All the intersection points and their neighbors may then be removed from the skeleton map, in order to obtain an image with clearly separated vessel segments. Finally, a vessel graph can be generated by linking first and last nodes in the same vessel segment, as shown as Figure 5 (C).

The generated graph usually includes misrepresentations of the vessels, and so it is important to modify this incorrect graph in order to avoid false classification of nodes. As summarized in [30], typical errors are *node splitting* and *false link*. The representation and modification of these two errors are:

(1) *False link* is demonstrated in Figure 6 (A): an incorrect link c between two nodes n_1 and n_2 is created. This happens when two vessels are close to but do not cross each other. To resolve this case, the angles α and β between the edges connected to each node are computed. If the angles satisfy $\alpha_1, \alpha_2 \in (180^\circ \pm 10^\circ)$ and $\beta_1, \beta_2 \in (90^\circ \pm 10^\circ)$, then we consider link c to be a false link, which should be removed. Figure 6 (c) demonstrates the corrected graph.

(2) *Node splitting* is illustrated in Figure 6 (D): false nodes n_1 and n_2 are created. This happens when two vessels are close enough to cross each other. To address this problem, we define two angles α and β as shown in Figure 6 (E). If the measured angles satisfy $\alpha_1, \alpha_2 < 60^\circ$ and $\beta_1, \beta_2 > 90^\circ$, this situation can be considered as an instance of node splitting, and edge c should be removed and the two neighborhood intersect point n_1 and n_2 merged as one node n. Figure 6 (F) reveals the misrepresented graph after this correction.

2.4 Vascular Topology Estimation via Dominant Set Clustering

Node analysis is broken down into four categories (node degrees 2-5), based on four different types of nodes: connecting points (2), bifurcation points (3, 4), and crossing/meeting points (3, 4, 5).

The number in the bracket indicates the possible number of links connected to each node (node degree). The method proposed by Dashtbozorg et al. [30] is used to handle the cases of nodes of degree 2-3. For the more complicated categories, nodes of degree 4 and 5, a classification method based on dominant sets clustering is proposed. In this work, for each centerline pixel, its intensities in R, G, B channels, orientations, curvatures, and vessel diameters are used as the input of the dominant sets clustering based classifier.

The nodes to be classified are represented as an undirected edge-weighted graph with $G = (V, E, \omega)$, where the node set $V = \{1, \cdots, n\}$, and usually $n \leq 5$. The edge set $E \subseteq V \times V$ indicates all the possible connections. $\omega : E \rightarrow R_+^*$ is the positive weight function. Nodes in G correspond to vessel node ends: edges represent node relationships, and edge weight reveals similarity between pairs of linked nodes. The symmetric matrix $A = (a_{ij})$ is used to represent the graph G with weighted adjacency matrix. This non-negative adjacency matrix is defined as:

$$a_{ij} = \begin{cases} \omega(i,j) & \text{if } (i,j) \in E \\ 0 & \text{otherwise.} \end{cases} \tag{4}$$

Note: all elements on the main diagonal of A are zero, since G is self-loops free.

In general, the weights of edges within a vessel segment should be large, representing high internal homogeneity or similarity. By contrast, the weights of edges will be small for two or more different vessel segments because those on the edges connecting the vessel ends represent high inhomogeneities [32]. The assignment of the edge-weights can be analyzed based on the above perspectives. Let $S \subseteq V$ be a nonempty subset of nodes, $i \in S$, and $j \notin S$. Intuitively, the similarity between nodes j and i can be defined as:

$$\phi_S(i,j) = a_{ij} - \frac{1}{|S|} \sum_{j \in S} a_{ij} \tag{5}$$

This measure is with respect to the mean similarity between i and its surroundings in S, and $\phi_S(i,j)$ can be either positive or negative. $\frac{1}{|S|} \sum_{j \in S} a_{ij}$ is the average weighted degree of i with regard to S. It can be observed that $\frac{1}{|S|} \sum_{j \in S} a_{ij} = 0$ for any $i \in V$, and $\phi_{\{i\}}(i,j) = a_{ij}$. For each

node $i \in S$, the weight of i with regard to S is assigned as:

$$\omega(i) = \begin{cases} 1 & \text{if } |S| = 1 \\ \sum_{j \in S \setminus \{i\}} \phi_{S \setminus \{i\}}(j,i)\omega_{S \setminus \{i\}}(j) & \text{otherwise.} \end{cases} \tag{6}$$

where $S \setminus \{i\}$ indicates the the nodes set S excluding the node i, and $\omega_S(i)$ demonstrates the overall similarity between node i and the nodes of $S \setminus \{i\}$ with respect to the overall similarity among the nodes in $S \setminus \{i\}$.

Finally, the total weight of S can be calculated by summing $\omega_S(i)$: $W(S) = \sum_{i \in S} \omega_S(i)$. For example, Figure 7 demonstrates an edge-weighted graph, and we have:

$$\omega_{1,2,3}(1) = \phi_{2,3}(3,1)\omega_{2,3}(2) + \phi_{2,3}(2,1)\omega_{2,3}(3) = 12. \tag{7}$$

Similarly, $\omega_{1,2,3}(2) = 0$ and $\omega_{1,2,3}(3) = 12$ are obtained, which yield $W(1,2,3) = 12 + 0 + 12 = 24$.

We define set as a *dominant set* if the set satisfies the following two conditions: 1. $\omega_S(i) > 0$, for all $i \in S$; 2. $\omega_{S \cup \{i\}}(i) < 0$, for all $i \notin S$. It is evident from the above properties that the first condition defining a dominant set is internal homogeneity, whereas the second concerns external incoherence. We can find a dominant set by first localizing a solution of program:

$$\begin{aligned} &\text{maximize} \quad f(\mathbf{x}) = \mathbf{x}' A \mathbf{x} \\ &\text{subject to} \quad \mathbf{x} \in \Delta \end{aligned} \tag{8}$$

where a prime denotes transposition and

$$\Delta = \left\{ \mathbf{x} \in R^n : \sum_{i=1}^{n} x_i = 1, \text{and } x_i \leq 0 \text{ for all } i = 1 \cdots n \right\} \tag{9}$$

As suggested in [38, 39], the effective optimization approach to extract a dominant set from a graph is given by the so-called *replicator dynamics*. Figure 5 (E) represents the estimate vascular network with topological information.

3 Experimental Results

In this section, the validations of the proposed vessel segmentation method and its application - vessel topology estimation are given individually. All the experiments were carried out in MATLAB 2015a on a PC with an Intel Core i7-4790K CPU, 4.00GHz, and 16GB RAM.

3.1 Vessel Segmentation

Datasets and metrics: Three publically available retinal datasets are used in this work to evaluate the proposed segmentation framework: STARE[1], DRIVE[2], and a newly released dataset IOSTAR[3]. The image resolutions of these datasets are 565×584, 700×605, and 1024×1024, respectively. These datasets are chosen primarily because of the availability of reference standard from manual annotations of the retinal vessels by experts. The segmentation performance is measured by sensitivity se, specificity sp, and accuracy acc. They are defined as $se = \frac{tp}{tp+fn}$, $sp = \frac{tn}{fp+tn}$, $acc = \frac{tp+tn}{tp+fp+tn+fn}$, respectively. Here, true positive tp is the count of pixels marked as vessel pixels in both the segmented image and its ground truth. Similarly, false positive fp identifies the number of incorrectly identified vessel pixels; true negative tn is the number of correctly identified non-vessel pixels; false negative fn indicates the number of incorrectly identified non-vessel pixels. In general, reporting the se and sp obtained at highest acc is a common way in the retinal image segmentation. However, it is possible to produce imbalanced results where a higher sp is favored since the amount of vessel pixels is significantly lower than background pixels. In such a case, acc will be skewed by the dominant classes. Consequently, in order to evaluate the performance of the proposed vessel segmentation method, the receiving operator characteristics (ROC) curve is computed with true positive ratio versus the false positive ratio. The area under the ROC curve (AUC) is calculated to quantify the performance of the segmentation, since it has the ability to reflect the trade-offs between the sensitivity and specificity.

In this experiment, the green channel of the color fundus images were used for vessel segmentation. Figure 4 illustrates examples of vessel detection performance on three datasets, and manual annotation from observer 2 of the DRIVE and STARE dataset were used as groundtruth.

[1] `http://www.ces.clemson.edu/~ahoover/stare/`
[2] `http://www.isi.uu.nl/Research/Databases/DRIVE/`
[3] `http://www.retinacheck.org`

To reveal the relative performance of our proposed method, we compared it with several existing state-of-the-art vessel detection methods on the most popular datasets: DRIVE and STARE. The results are shown in Table 1, and the chosen methods have been ordered by the category the methods belonging to: the most recent seven supervised methods [10, 11, 12, 13, 14, 24, 25], and nine unsupervised segmentation methods [16, 40, 9, 20, 37, 41, 42, 21, 43]. Note, a different AUC calculation was used in [37]: $AUC = (se + sp)/2$.

Overall, our framework yields competitive performances in most of the quality metrics used, as it took into account the global features through the Retinex analysis and the local features through the superpixel-based line operator, therefore, more fine vessels may be detected. It is worth noting that our previous saliency driven vessel segmentation model (SAD) [23] has higher performance than the proposed method. However, the computational complexity has been significantly deceased: SAD has a computational complexity of $O(N^2)$ in the estimation of compactness based saliency detection, and another $O(N^2)$ in the estimation of intensity based saliency detection, and requires $2 \times O(N^2)$ in total. By contrast, the proposed method has a computational complexity of $O(N)$ to obtain the response of line operator. N indicates the number of superpixel. Overall, the average computation time of a single image from DRIVE and STARE datasets by SAD method is 43 ± 4.5 seconds, while it has dramatically reduced to 2.3 ± 0.7 seconds by the proposed method.

Note, to the best knowledge of the authors, only Zhang et al. [43] has tested their segmentation method on IOSTAR dataset. In consequence, we only compared with the performance obtained by [43] in the bottom of Table 1, and is by no means exhaustive. In contrast, our method has better performance in terms of all metrics.

Furthermore, three state-of-the-art vessel enhancement methods were employed for comparison purposes. These methods were: isotropic undecimated wavelet filter [20], local phase filter [44] and Combination Of Shifted Filter Responses (BCOSFIRE) [21]. In the interests of reproducibility, the recommended parameters in the literature were used in the experiments. In Figure 8, we show examples of applying different enhancement methods on a representative patch with multiple vascular bifurcations, curvature changes, intensity inhomogeneity on large vessel and low intensities on tiny vessels. Overall, the proposed method is not only able to detect the vessel regions, but also has the ability to suppress noise and artifacts. In other words, the results obtained by the proposed method seem more pleasing: stronger enhancement results on tiny vessels, better responses on

bifurcations / crossovers, and higher uniformity on intensity inhomogeneity.

The Effectiveness of Superpixel and Retinex

In this section, the effectiveness of superpixel enabled line operator and Retinex based image enhancement are validated individually.

Figure 9 demonstrates the segmentation results obtained by the proposed models with and without superpixel enabled. It can be observed from Figure 9 (C) that superpixel contributes significantly to the final performance - more tiny vessels have been detected, and the sensitivity of the vessel segmentation has been improved.This observation is also confirmed by the ROC curves over three different datasets (DRIVE: se=0.781 and sp=0.977; STARE: se=0.781 and sp=0.977; IOSTAR: se=0.761 and sp=0.975), as illustrated at Figure 10 (red line). Most existing line operator based segmentation approaches have a certain edge length μ, such as 15 pixels in [16], and 5 pixels in [35]. In this work, the edge length is self-adapting, and it is more sensitive to capture the varying scales of vessels within an image, and this leads to higher se, acc, and AUC.

In addition, the ROC curves of the proposed method with and without Retinex enhancement applied are illustrated at Figure 10(green line). Overall, Retinex affects the final performance significantly, since the optic disk and foveal area tend to have inhomogeneous intensities, which inhomogeneities were corrected after applying Retinex. In contrast, the segmentation performances were relatively poor in datasets STARE and IOSTAR than DRIVE when without applying Retinex. That is because STARE and IOSTAR datasets contain some images with pathologies, e.g. presents bright lesions or exudates, blurring vessel, and features that can cause more false detections (lower sp). While the proposed Retinex method is capable of normalizing these regions to a similar level with the background, and increase the contast between the vessels and background, thus avoiding false detection (higher sp), and raising the sensitivity score.

Parameters Tuning

In this section, we experimentally investigate the optimal numbers of superpixels M and levels of superpixel partition P. It is known that too large number of superpixel leads to false detection, and on the contrary, too few superpixels result in a loss of the edge information of the vessel[45]. To this end, in this experiment, the numbers of superpixels were set to be successively 400, 800, 1200,

1600, and 2000. The left column of Figure 11 shows the ROC curve of the proposed method under these numbers, and the proposed method achieves the best result when the superpixel number is 1200. As aforementioned at Section 2.2, multiscale analysis was used to detect vessel more precisely when an image contains varying scales of vessels. The right column of Figure 11 shows the segmentation performance under different superpixel levels when the number of superpixels was set to 1200, and it can clearly be seen that the proposed method yields the best performance when the number of levels is 3. In consequence, the number of superpixels at the other two levels are $\frac{1}{3} \times 1200 = 400$, and $\frac{2}{3} \times 1200 = 800$.

3.2 Vascular Topology Estimation

We evaluated the proposed topology estimation method on five publicly available retinal image datasets: DRIVE, STARE, INSPIRE [46], VICAVR [47], and IOSTAR datasets.

The gold standards of vessel topology of DRIVE and STARE were manually annotated by De et al. [3, 29]. For the later three datasets, INSPIRE has 40 high resolution images, each of 2392×2048 pixels, VICAVR includes 58 images with a resolution of 784×584 pixels each, and IOSTAR contains 24 images taken with a scanning laser camera (SLO) each of 1024×1024 pixels. All of these three datasets have manual annotations on arteries/veins classification [48, 30, 49], and the IOSTAR dataset also has annotations on vessel bifurcation/crossing. However, none of these three datasets has annotations on vessel topology. Therefore, we asked a clinician expert to manually label the topological information of the vascular structure on all the images from these datasets. Each single vessel tree is marked with a distinct color, as shown in second column of Figure 12.

The third column of Figure 12 illustrates the results of our DOminant Sets based topology estimation method (DOS) on datasets INSPIRE, VICAVR, and IOSTAR, respectively. Compared with the manual annotations, it reveals from visual inspection that our method is able to trace most vascular structures correctly: only a few crossing points were incorrectly traced, as shown in the right column of Figure 12 - the pink squares indicate the incorrectly traced significant points. To facilitate better observation of the performance of the proposed method, the accuracy results with regard to different significant points (connecting, bifurcation, and crossing points) are presented in Table 2. It can be seen that the method achieved accuracy of 0.915, 0.889, and 0.928

in INSPIRE, VICAVR, and IOSTAR, respectively. The accuracy indicate the percentage of the relevant significant points that were correctly identified.

The results obtained by the proposed DOS vascular topology estimation method in DRIVE and STARE datasets, were also compared with those obtained by five state-of-the-art topology estimation and label propagation methods in Table 3: Class Distribution Relational Neighbor classifier (CDRN) [50], Weighted Vote Relational Neighbor classifier (WVRN) [50], Digraph variant of the Commute Time Kernel classifier (CTK) [51] and Symmetrized Graph Laplacian (SGL) [52], and Matrix-Forest Theorem of Directed graphs (MFTD) method [29]. Note, all the results of topology estimation methods are reported in [29], and the recommended parameters from the original source code or literature were used.

For the purposes of a fair comparison, the vessel segmentation results obtained by [29] were used for DOS based topology estimation. The evaluation metric, accuracy score, is calculated by the sum of all true positive segments counts divided by the total number of instances. It is worth noting that the vessel segments' centerline pixels are utilized for accuracy calculation.

The results show that our method achieves the best performance, with an accuracy score of 0.83 and 0.91 in DRIVE and STARE dataset, respectively. Figure 13 illustrates the visual comparison with the results obtained by De et al. [29], and it can be revealed our method has better performance in branch points and crossovers.

3.3 Arteries / Veins Classification

In addition, the estimated vascular topology will also benefit to the classification of arteries/veins (A/V). After estimating the vessel topology, the complete vessel network is separated into several *subgraphs* with individual labels. The final goal is to assign these labels to one of two classes: artery and vein. The features listed in Table 4 and the DOS classifier are utilized to classify these individual labels into two clusters, A and B. For each subgraph v, the probability of its being A is computed by the number of vessel pixels classified by DOS as A: $P_A^v = n_A^v/(n_A^v + n_B^v)$, where n_A^v is the number of pixels classified as A, and n_B^v is the number of pixels classified as B. For each subgraph, the higher probability is used to define whether the subgraph is assignable to category A or B. Cluster A and B are then assigned as artery and vein, respectively, based on their average intensity in the green channel: higher average intensity is classified as artery and lower as vein.

Figure 14 shows the A/V classification performances of the DOS classifier on sample images based on their topological information. Overall, our proposed method correctly distinguished most of the A/V labels on all three datasets, when compared with the corresponding manual annotations. In order to better demonstrate the superiority of the proposed method, Table 5 reports the comparison of our method with the state-of-the-art methods over three datasets in terms of pixel-wise sensitivity (Se), specificity (Sp), and accuracy (Acc). It is clear that our method outperforms all the compared methods on all datasets, except that the sp score on DRIVE dataset is 0.3% lower than [28].

4 Discussions and Conclusion

In this paper, we have presented a new framework for retinal vessel analysis that exploits the advantages of non-local total variation based Retinex model for intensity inhomogeneity correction, superpixel-based line operator for vessel segmentation, dominant set clustering based topology estimation, and arteries /veins classification.

In general, bifurcation and crossover of vessels, small vessels and highly curved vessels are the most challenging ones in retinal vessel segmentation. The traditional line operator-based segmentation model usually applied on the entire image and yields imbalance response on the vessels, as evidenced of the results achieved by [16]. In this work, we applied the line operator on the SLIC generated patches rather than entire image. The results of the proposed method showed that all the evaluation metrics have much improved when compared to those scores by the traditional line operator. In addition, distinguish from the method we proposed in [23], the computational complexity has been significantly reduced by the proposed method. The quantitative evaluations on publically-available datasets showed that, compared to established methods, the proposed method achieves competitive vessel segmentation performance. In particular, it shows better performance in handling small, bifurcation, and crossover vessels, even in the case of poor contrast.

The problem of estimating the topology of vascular trees was formalized as a pairwise clustering problem. It is demonstrated that our method achieves competitive results when compared with existing state-of-the-art methods. In addition, the vascular topological information was utilized to distinguish the arteries from veins, and it reaches high accuracies when validated quantitatively

using three publicly accessible datasets with promising results.

In contrast, our previous work [23] has higher sensitivities in segmenting vessels, however, it yields many disconnected vessels (fragments) by close visual inspection. The disconnected vessel problems will in turn affect the performance of the reconstruction of vessel topology: one single vessel branch will form as two or more subgraphs due to the disconnections after topology estimation. By contrast, the line operator extracts the vessels more complete with reduced disconnections of vessels. For example, Figure 15D and Figure 15E have equal sensitivity scores on topology estimation, however, Figure 15 has lower specificity as the topological information of red arrow indicated vessels are incorrectly assigned, and leads to a lower accuracy when compared with Figure 15E.

In addition, a further major advantage of the proposed work is that manual annotations of vessel topologies of three publicly accessible datasets (IOSTAR, INSPIRE, and VICAVR) were established as the groundtruth. Topological properties of retinal blood vessels in fundus images can provide valuable clinical information in diagnosing diseases.

In our future work, a widely used metric in biological tracing community, the DIADEM score [53], will be used for topology estimation validation. In addition, we will test our vessel topology estimation method on other retinal datasets (e.g., RITE [54]) and neuronal datasets (e.g., DIADEM [53]), and the validations will be taken under both significant points-wise and centreline pixel-wise. The proposed method has the prospect of becoming a powerful tool for analyzing vasculature for better management of a wide spectrum of vascular-related diseases.

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6 Conflict of Interest

The authors have no relevant conflicts of interest todisclose.

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7 Figure Legends

Figure 1: Overview of the main steps of our method: (A) A random selected color fundus image; (B) The green channel of (A); (C) Results after applying Retinex on (B); (D) Superpixelized results of (C); (E) Vessel response of the proposed method; (F) Segmentation result by the proposed method. (G) Generated vessel graph; (H) Estimated vascular network topology where each tree is denoted with different colors.

Figure 2: Illustrative results of image enhancement by using non-local total variation based Retinex approach. (A) and (D): Two randomly selected color fundus images; (B) and (E): The green channels of (A) and (D). (C) and (F): Results after applying Retinex on (B) and (E).

Figure 3: Illustration of different superpixel numbers generated on an example image: (A) The green channel of a random selected color fundus image; (B) 400 superpixels; (C) 800 superpixels; (D) 1200 superpixels.

Figure 4: Examples of vessel segmentation by the proposed method on 3 datasets. From left to right: green channel of random selected color fundus images, results after superpixel enabled line operator, automated segmentation results, and manual annotations.

Figure 5: Reconstruction of graph and topology of retinal vasculature. (A) Original color fundus image. (B) Map of skeletonized vessels. (C) Generated vessel graph. (D) Graph generated with significant nodes overlaid. Red dots indicate terminal points, green triangle bifurcations, and blue squares intersection or crossover points. (E) Estimated vascular network topology where each tree is denoted with different colors.

Figure 6: Two types of graph correction. (A)(B) illustrate a false link, (C) shows the corrected two separate vessels, while (D)(E) illustrate a node splitting, (F) shows the corrected single node at an intersection.

Figure 7: Edge-weighted graph.

Figure 8: A comparative study with other enhancement techniques on a selected region with tiny vessel (yellow arrow), bifurcation (green arrow), and crossover (red arrow). (A) The green channel of a selected region of a color fundus image. (B) isotropic undecimated wavelet filter. (C) Local phase. (D) BCOSFIRE. (E) Proposed method.

Figure 9: Segmentation results of the proposed method, and the snapshot of selected region with small vessels. (A) Original image. (B) Segmentation result without superpixel applied. (C) Segmentation result with superpixel applied. (D) Groundtruth.

Figure 10: The ROC curves of the proposed framework with and without the Retinex enhancement applied and superpixel-based the line operator applied over three different datasets respectively. (The reader is referred to the color version of this figure.)

Figure 11: The ROC curves of the proposed method with (left) different numbers of superpixels: 400,800, 1200, 1600, and 2000; (right) different numbers of levels, after setting the optimal number of the superpixels to 1200.(The reader is referred to the color version of this figure.)

Figure 12: Examples of vascular topology estimation on retinal datasets INSPIRE, VICAVR, and IOSTAR. From left to right column: original image, manual annotations, the automatic topology estimation results. Correct and incorrect connections are highlighted by green disks and pink squares, respectively.

Figure 13: Comparisons of vascular topology estimation on retinal datasets DRIVE and STARE. From left to right column: original image, manual annotations, the automatic topology estimation results by method proposed in [29], and results by proposed method. Correct and incorrect connections are highlighted by grey squares and pink disks, respectively.

Figure 14: A/V classification results on three different datasets. From left to right column: original image; vessel topology; A/V classification results of the proposed method; and corresponding manual annotations.

Figure 15: Performances of vascular topology estimation based on different segmentation methods. (A) Original image. (A1) Selected region with weak vessels for demonstration. (A2) Topology ground truth of (A1). (B) Segmentation results based on [23]. (C) The proposed segmentation results. (D) Reconstructed topology of (B). (E) Reconstructed topology of (C).