

Image Analysis for Nail-fold Capillaroscopy

VLADIMIR VUCIC



KTH Electrical Engineering

Master's Degree Project
Stockholm, Sweden 2015

TRITA-EE 2015:58

Kungliga Tekniska Högskola
Electrical Engineering

August 2015

Master's Thesis Project
Image Analysis for Nail-fold Capillaroscopy

VLADIMIR VUCIC

Company supervisor: Alexander Fagrell
Examiner: Markus Flierl

TRITA-EE 2015:58

Abstract

Detection of diseases in an early stage is very important since it can make the treatment of patients easier, safer and more efficient. For the detection of rheumatic diseases, and even prediction of tendencies towards such diseases, capillaroscopy is becoming an increasingly recognized method. Nail-fold capillaroscopy is a non-invasive imaging technique that is used for analysis of microcirculation abnormalities that may lead to disease like systematic sclerosis, Reynauds phenomenon and others.

The main goal of this master thesis project is to provide new tools and techniques for the analysis of capillaroscopy images from the nail-fold area. Image processing and machine learning techniques are applied to images obtained by digital microscopes, like Mediscope as produced by Optilia Instruments AB, Sollentuna. This thesis offers a novel way for segmentation of capillaries from images as well as (semi)automatic capillary width calculation and automatic annotation of capillaries. These tools provide new insights into the structure of capillaries and also reduce the time required for measurement/annotation of capillaries.

Acknowledgements

I would like to express my sincere gratitude to everyone from Optilia Instruments AB, especially Alexander Fagrell, Hazar Mutgan and Sasan Esmaceli, for providing me with the topic and guidance throughout the whole master thesis project. They provided me an opportunity to work on this challenging topic and together with other company employees made a pleasant working atmosphere for me.

My sincere thanks also goes to my coordinator and examiner, Markus Flierl for giving valuable guidance and suggestions at each stage of the project. I would like to thank Chen Wang for friendship and support that helped me to carry out this project.

I would also like to thank my family for constant emotional support throughout writing this thesis and life in general.

Contents

1	Introduction	1
1.1	Background	1
1.2	Problem definition	2
1.3	Purpose	3
1.4	Goals	3
1.5	Structure of the thesis	4
2	Related work	5
2.1	Image enhancement and annotation	5
2.1.1	Image filtering	5
2.1.2	Optimal color space selection	8
2.1.3	Automatic annotation	9
2.2	Theoretical background	13
2.2.1	Contrast Limited Adaptive Histogram Equalization	13
2.2.2	Hough transform	14
2.2.3	Histogram of oriented gradients	15
2.2.4	Support vector machine	17
3	Image analysis system	20
3.1	Filtering of capillaries	20
3.2	Rotation of image	23
3.3	Width calculation	25
3.4	Annotation of capillaries	27
4	Experimental setup and results	29
4.1	Data	29
4.2	Experimental setup	31
4.3	Results	31
4.3.1	Filtering, Rotation and Width calculation	31
4.3.2	Annotation of capillaries	36
5	Discussion and future work	41
6	References	43

List of Figures

1	Optilia Mediscope	2
2	Examination process	3
3	Examination images	3
4	Algorithm proposed by Goffredo et al[18]	10
5	Sensitivity-specifity of different image channels[18]	10
6	CLAHE[25]	14
7	Histogram of oriented gradients[29]	15
8	Support Vector Machine [wikipedia]	17
9	Original image	20
10	Preprocessed images	21
11	Second derivatives	23
12	Filtered image	23
13	Image with marked lines	24
14	Rotated image	25
15	Edges added to filtered image	25
16	Length calculation	26
17	Capillary width calculation histogram	26
18	Capillary width marked image	26
19	Training set — positive samples	27
20	Marked positions	28
21	Annotated capillaries	28
22	Doctor A dataset sample	30
23	Doctor B dataset sample	30
24	User interface	31
25	Original images	32
26	Filter from paper [18]	32
27	Proposed filter	33
28	Different sigma values	33
29	Example of wrong rotation	34
30	Unrotated images	35
31	Width calculation with added edges	35
32	Width calculation	35
33	Enlarged capillaries issue	36
34	Successfully annotated capillaries	38
35	Successfully annotated capillaries	39
36	Line algorithm	40

List of Tables

1	Orientation pattern	8
2	Test dataset	31
3	Processing time	34
4	Doctor A dataset	37
5	Doctor B dataset	37
6	Manually marked normal capillaries dataset	37
7	Manually marked enlarged capillaries dataset	38
8	Whole dataset	39

1 Introduction

This master thesis project addresses the problem of processing and analysis of images obtained during capillaroscopy examination. During the past years hardware used for these examinations has improved a lot, and currently there are many devices that offer high quality digital images. This opens up possibilities for many new software features that would provide new insights and information from images. Goal of the thesis is to use image processing techniques in order to extract more information from images, improve and speed up capillaroscopy examination. This thesis will focus on processing of capillary images obtained with Optilia “Mediscope” digital microscope produced by Optilia Instruments AB. In this chapter focus will be on the background of capillaroscopy examination in order to better understand current issues and needs of doctors. Furthermore, problem definition, goals of this project, as well as structure of the thesis will be presented.

1.1 Background

Capillaroscopy is a non-invasive in-vivo method for observation of capillaries and microcirculation that can be performed on various parts of skin. Most common one is nail-fold capillaroscopy because fingers are easily accessible for this type of examination and observation site can be accurately defined[1]. Furthermore, fingers are involved in pathological process of some autoimmune disorders so they can point out to certain diseases. Microcirculation abnormalities can point to certain autoimmune rheumatic diseases like Reynauds phenomenon, systemic sclerosis, digital ulcer and others. It is believed that in the future analysis of capillaries from nailfold area might be used for prediction and early detection of microvascular heart problems. In general, there are three types of devices that are used to perform nail-fold capillaroscopy examinations [2]:

- *Stereomicroscope*: Device is often used today because of its ease of use and low cost. On the other hand, it has a low magnification capacity and therefore obtained images are not high-quality
- *Ophthalmoscope and dermatoscope*: Main purpose of these devices is not capillaroscopy examination but still they are used in the lack of more adequate equipment. These devices have low magnification capacity and images which are not of a high-quality
- *Videocapillaroscope*: They are specifically built for the purpose of capillaroscopy examinations, and consist of high magnification lens coupled with digital video camera. They provide high-quality images and since images are digital they can be further processed with the aid of software tools

This thesis project was done in cooperation with company Optilia Instruments AB which produces digital microscopes for both industrial and medical purposes. The product that was used for the project is “Optilia Mediscope” – digital medical microscope (videocapillaroscope) that is used for nail-fold capillaroscopy examinations. Optilia’s “Mediscope” is a hand-held videocapillaroscope that provides 200 and 300 times magnification lenses and captures images of 3 megapixels. Besides image quality, other advantages of this device are that

it provides examinations that are non-invasive, fast and efficient. Device enables examinations in all circumstances (with certain patients traditional microscope examination cannot be performed) and technique is easy to learn.

High quality images open up many possibilities for further processing and from the academic point of view, there are many research papers that tackled the topic of vessel segmentation. Some of the research tackled specifically the nailfold capillaroscopy examination. This will be discussed in more details in Chapter 2.



Figure 1: Optilia Mediscope

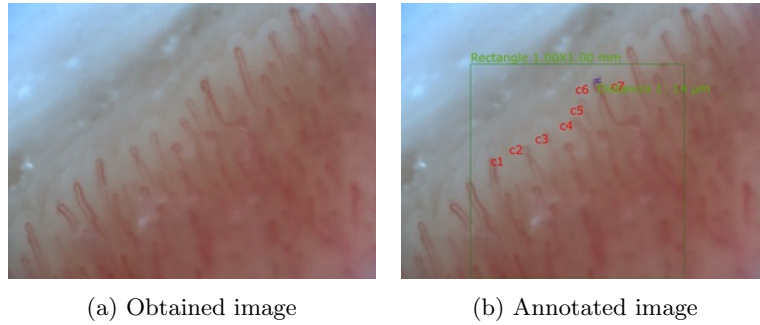
1.2 Problem definition

In order to better understand problems that doctors currently have when performing capillaroscopy examination, it is necessary to know some basics about the examination procedure. When a patient comes for the examination, he must first rest for a 20 minutes so that his capillaries come to normal condition, otherwise false results may appear. Before taking the images, oil is put on the finger in order to create smooth surface on the nail-fold (this does increase the quality of images but it also introduces the glare artifact). After that video-capillaroscope is placed on the nail-fold and images are taken. Doctors must make sure that there is no pressure to finger since that can influence the results. Each finger except for the thumbs is examined and 4 images of each finger are recorded and that makes 32 images per patient. After acquiring images, doctors have to examine each image separately. First important thing is to count the number of capillaries per square millimeter. Doctors currently do that manually by clicking on each capillary with the mouse pointer. Next thing is to measure the size of certain capillaries in order to find the giant capillaries, this is especially important for the early phase of disease. Doctors have to now zoom in the image and manually place the points between which the distance will be measured. Furthermore, doctors also have to look for micro bleeding and ramifications (branchy, bushy or coiled capillaries). Based on all this information doctors can conclude the current state of the patient [3]. As there are 32 images per patient, all this takes a lot of time and energy from the doctors that could be used for some other activities. Looking at all this images that are often quite similar can in some cases lead to errors because eyes get tired and lose focus.

This examination procedure can certainly be speeded up and simplified. Many researches tackled the topic of vessels enhancement that would make easier for doctors to see more clearly the structure of capillaries. Furthermore, some studies included automatic annotation of capillaries as well as classification of capillaries.



Figure 2: Examination process



(a) Obtained image

(b) Annotated image

Figure 3: Examination images

1.3 Purpose

Purpose of this degree project is to study the current methods that are used for filtering the blood vessels and develop a method that is most suitable for filtering of nail-fold capillaries from provided digital images. Good filtering will open up possibility for development of other features that can improve the capillaroscopy examination. Main benefit from this work should have physicians who would be able to offer better and faster capillaroscopy examinations. However, findings from this degree project may even be interesting for academic purposes and further research in this field since this is becoming big area for research.

1.4 Goals

Goal of this project is to provide physicians with a software tool that would allow them to improve and speed up capillaroscopy examination. After a discussion with microscope manufacturer and their customers following goals are chosen:

1. Filtering of capillaries from image
2. Rotation of image so that capillaries point upwards
3. Counting of capillaries
4. Calculation of capillaries width

It is important to note that the goal of this thesis work is not to replace physicians in any sense but only to provide tools that would help them in their work.

1.5 Structure of the thesis

Chapter 2 presents relevant academic research that is done in this field, as well as theoretical background required for understanding the developed system. In Chapter 3 each part of system is presented and implementation is described. Chapter 4 focuses on testing of the system where dataset, experimental setup and results are presented and discussed. Conclusion and short discussion about future steps are in Chapter 5.

2 Related work

2.1 Image enhancement and annotation

During the early years of capillaroscopy examinations technology was not very advanced so the acquired images were not of a high quality and it was important to find good image enhancement techniques. There has been many research in this field, most of them were focused on the enhancement of blood vessels in general but some research were focused in particular field such as blood vessels from eye retina, or capillaroscopy images as topic of this master thesis project.

2.1.1 Image filtering

One of the basic things for image enhancement is applying the well known filtering techniques and comparing their results. In the paper “An Evaluation of Image Enhancement Techniques for Capillary Imaging” [4] authors did exactly that on nail-fold capillaroscopy images. The main goal of this research was to find the optimal filter that would preserve the edges of capillaries without taking into account the processing time required. Images were first filtered with one of the ten selected filters and afterwards image was filtered with Sobel edge detector [5]. Authors claim that this particular edge detector is the best solution because of its simplicity and optimization. However, there are also many other edge detection algorithms like Canny algorithm, and it would be also good to see their results as well. It is important to point out that images for this study were obtained using stereomicroscope coupled with external light source so the image quality was not too high. Results of this study show that Gaussian filter, α -trimmed filter, ϵ filter and wavelet filter have rather poor results when it comes to enhancement of provided images. Filters that provide better results are median and adaptive damped wave equation, while best results are achieved with anisotropic diffusion, non local means filtering, bilateral filter and bilateral enhancer. In the end, only the edge preservation was taken as a performance measure and it proves that bilateral enhancer has the best edge preserving capabilities among the tested filters.

Extracting the edges of capillaries can provide us with a lot of valuable information about the capillary so it is not surprising that various methods were tested in order to properly do this task. Authors of the paper “Markov Chain Based Edge Detection Algorithm for Evaluation of Capillary Microscopic Images” [6] propose a novel way for capillary edge detection using the Markov Chain. Previous attempts were based on usage of different filters in order to detect edges of capillaries, and most of these approaches did not provide very good performance. In this paper authors focus on a different approach in which not only local features are taken into account but also relations among pixels. Biggest problem in capillaroscopy images is that quite often they can be low resolution or have other factors that affect the overall image quality. Capillaries do not have sharp edges because images are often blurred and with certain noise that comes from the fact that imaging is done through the skin. Without going into all details, algorithm is based on calculation of second derivatives of the pixels and uses the walking algorithm to process whole image. In order to get good results it is necessary to run the algorithm more than once and in

this paper authors ran up to 100 iterations. With small number of iterations, edges of capillaries are extracted but also a lot of noise is extracted as well, as the number of iteration increase noise reduces but also some of the capillaries become less connected and even disappear. Another issue with this algorithm is speed, using the images that are much lower resolution than the ones we have one but also using the lower performance computer, it took almost a minute for 100 iterations on a single image. Authors mostly focused on the precision and they concluded that their results are better than the ones with other edge-detection filters. Measure for precision is simply human eye since there are no ground truth images. However it is noticeable that all the images have noise in the background so probably some further post-processing is required for better final result.

Doshi et al [7] point out the importance of good capillary segmentation from the image for a precise diagnosis. This task is difficult due to many disturbing factors, such as noise, glare from immersion oil, small dust particles, inability of holding hand completely steady and this causes some blur on the images with high magnification. There were some previous research in this area done by Wen et al [8][9] and Lo et al[10] but Doshi et al [7] claim that previous results are unsatisfactory so they propose an improved approach. Previous research would use rather simple binarization algorithms and therefore effectiveness of these methods is limited. Authors focus on developing an algorithm that will provide precise skeletonization of capillaries in image. First step is to apply Difference of Gaussian filter that should remove the variations in the background and make illumination uniform while preserving the capillaries shape. After that, images are binarised using Otsu algorithm and post-processed in order to remove some artifacts. In comparison with other skeletonization algorithms, authors claim that their results are better than the ones presented. However, this algorithm is not very flexible and only parameters that could be adjusted are the ones concerning the capillary radius in the DoG filtering.

Another paper that focuses on skeleton extraction from capillaries is the “Robust naïvefold capillary skeleton extraction” [11]. Authors state that provided results can be used for automatic measurements such as capillary density, count of capillaries or some other features. They also state that they solution clearly outperforms other methods for skeleton extraction. Algorithm has 3 steps:

- *Preprocessing*: based on the previous research [4] bilateral filter in combination with histogram equalization is used in this step
- *Binarisation*: this step provides us with black and white image, and the most important thing is to select the right threshold that would leave us only with capillaries. It is important to have uniform background for this step. Illumination across images is not constant therefore authors have decided to use DoG (with specified parameters). After that Otsu method for threshold calculation is used, followed with two passes of median filter in order to remove the small objects that may appear
- *Skeletonization*: thinning algorithm is applied to binary image in order to extract skeletons of capillaries. Authors have decided to use 8-pixel connectivity approach in order to preserve the capillary curvature. This

part has a two-step algorithm, where different conditions are checked in each pass for each pixel, and algorithm stops when there are no more pixels that could be removed

Results are compared with Wen et al [8][9] and Lo et al [10], and this algorithm produces better images with less noise and less fake vessels (artifacts). Proposed algorithm provides quite clearly defined capillaries when it comes to early and active stage images. For late stage images results are not that good and there is still room for improvement in that area, authors conclude. One of the things that are very important for practical implementations is speed; however authors did not include any numbers about that.

Another approach to this problem is presented in paper “Multiscale vessel enhancement filtering” [12] by Frangi et al. This approach is based on the Hessian filtering in order to enhance vessels from the image. Vesselness is calculated by the value of eigenvalues of Hessian matrix where this value presents the likelihood of vessel presence. The basic idea behind this approach is to get the principal directions from the local second order structure, and this directly provides us with the direction of the smallest curvature. This approach is adapted for both 2D and 3D, but since this master thesis project is based on images, focus will be only on the 2D implementation. Local behavior of an image is considered as Taylor expansion in the neighborhood of a point x_0 :

$$L(x_0 + \delta x_{0,s}) \approx L(x_{0,s}) + \delta x_0^T \nabla_{o,s} + \delta x_0^T \mathcal{H}_{o,s} \delta x_0$$

where $\nabla_{o,s}$ is the gradient vector and $\mathcal{H}_{o,s}$ is hessian matrix computed in x_0 at scale s . Differential operators of L are calculated as a convolution with derivatives of Gaussians.

$$\frac{\partial}{\partial x} L(x, s) = s^\gamma L(x) * \frac{\partial}{\partial x} G(x, s)$$

$$G(x, s) = \frac{1}{\sqrt{(2\pi s^2)^D}} e^{-\frac{x^2}{2s^2}}$$

where parameter γ is used for normalization that is necessary when it comes to differential operators at multi scales. Hessian matrix presents a second order derivation and it is intuitive to use it for discovering of vessels areas. Result of applying the second derivative Gaussian kernel is a probe kernel which measures contrast in the range $(-s, s)$ in the direction of derivative. From the eigenvectors of the Hessian matrix, eigenvalues are calculated and in the case of image there are two eigenvalues λ_1 and λ_2 . Based on their values it is possible to distinguish what kind of vessel is detected.

λ_1	λ_2	Orientation pattern
N	N	Noisy, no preferred direction
L	H-	Tubular structure (bright)
L	H+	Tubular structure (dark)
H-	H-	Blob-like structure (bright)
H+	H+	Blob-like structure (dark)

Table 1: Orientation pattern

where H = high value, L = low value, N = noisy, +/- indicate the sign of eigenvalue.

Based on all this information, vesselness measure $\mathcal{V}_0(s)$ is constructed for all scales s . After that all the scales are combined into one image so that the highest value for each pixel position is attained.

$$\mathcal{V}_0(s) = \begin{cases} 0 & \text{if } \lambda_2 > 0 \\ \exp\left(-\frac{\lambda_1^2}{2\beta^2\lambda_2^2}\right) \left(1 - \exp\left(-\frac{s^2}{2c^2}\right)\right) & \text{if } \lambda_2 < 0 \end{cases}$$

$$\mathcal{V}_0(\gamma) = \max_{s_{\min} \leq s \leq s_{\max}} \mathcal{V}_0(s, \gamma)$$

Other research [13][14] also tried to segment vessels using the Hessian matrix but they did not use all the eigenvalues of the Hessian. This approach can be seen as generalization and improvement of those earlier approaches. Results show that vessel segmentation is quite good with excellent noise and background suppression. This shows a great potential of this approach to vessel segmentation problem.

A big problem when it comes to processing capillaroscopy images is the low contrast between capillaries and the background. Paper “Segmentation and Extraction of Morphologic Features from Capillary Images” [15] studies exactly that characteristic. Authors propose three steps to overcome this issue: illumination improvement, enhancement of contrast and image smoothing. In order to achieve this, different color spaces are used, Y component from YIQ color space is used for illumination improvement and S component from HSV color space for contrast improvement. After that wiener filter is applied for smoothing because of its capabilities to increase signal to noise ration and keep the edges preserved. Finally authors also take components M, A and Cr from color spaces CMY, Lab and YCbCr respectively. The end result is quite good; however complexity of the algorithm may influence its performance making it unusable for real-world applications.

2.1.2 Optimal color space selection

There are many research papers that tackled the topic of optimal color combination when it comes to enhancement of blood vessels. One of the research

is presented in paper “Optimal Combinations of Color Space Components for Detection of Blood Vessels in Eye Fundus Images” [16] that is focused on detection of blood vessels from the eye fundus. Goal of this research was to find optimal combination of RGB components that would provide best classification of eye fundus vessels. Many researchers have shown that green channel offers very good results so the goal was to beat those results. Authors have developed three combinations that are useful in different situations, local, global and super-global. Many of the methods are developed to work only with green channel but authors believe that their solution could be used instead of green channel without having a negative effect on overall results. Results show that combination of color channels could have its advantages and that it should be further researched. Paper “Ranking of color space components for detection of blood vessels in eye fundus images” [17] also takes into account eye fundus blood vessels but this time authors take into consideration channels of different color spaces. So besides RGB, color spaces like YIQ, HSV, HSL and XYZ were examined as well. In the end results confirm that green channel carries a lot of information and that wide usage of that channel is justified. However, green-blue channel should also be taken into account as well as hue component in HSV. Also, saturation value from HSV can be useful for reflect detection problems.

Paper “Quantitative color analysis for capillaroscopy image segmentation used nailfoldcapillaroscopy image” [18] proposes an algorithm (Figure 4) in order to discover what color components carry the most information with the main goal to annotate the capillaries in the image. First step in the algorithm is selection of color model and after that correction of non uniform illumination is done since in most images central part has higher illumination than other parts. This is done by subtracting the estimated global differences of light from original image. Global light differences are estimated with a morphological operation dilation using disk with the 25pixel radius. In the end gamma correction is applied and image is binarized where white areas present capillaries while black is background. Results show that average accuracy of the proposed algorithm is higher than 80% but this included only capillaries with the specified shape and granularity. As for the color model, different linear combinations of RGB are tested and the best results are proved to be with linear combination of G and R components – $1.5G-0.5R$ (Figure 5). This combination is chosen by using different values and then comparing results with the already annotated data. However research shows that green channel offers only slightly less information than proposed linear combination of RGB components.

2.1.3 Automatic annotation

More sophisticated approach to automatic annotation of capillaries would be to use machine learning techniques. Sainthillier et al [19] focused their research on creating a system for automatic annotation of capillaries in the images obtained

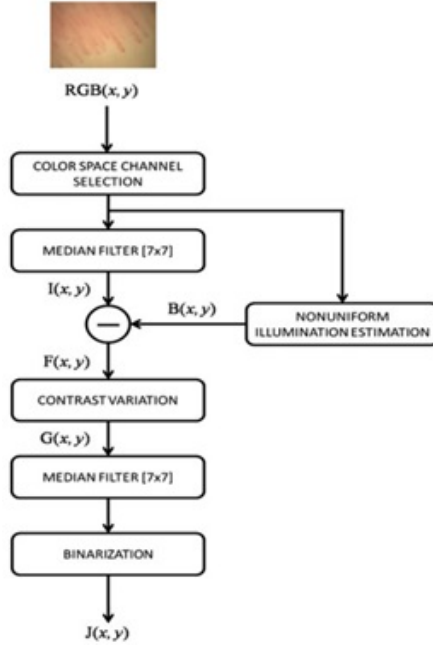


Figure 4: Algorithm proposed by Goffredo et al[18]

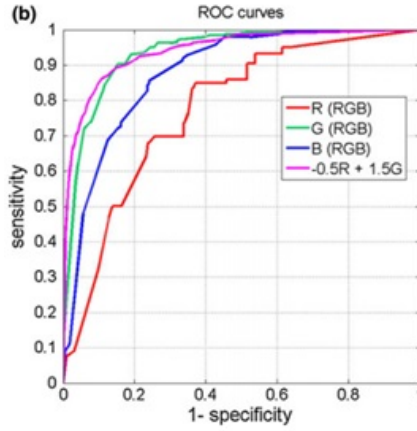


Figure 5: Sensitivity-specificity of different image channels[18]

from the skin (not nailfold area). For this purpose authors have decided to use autoassociative neural network because it is able to store and learn many patterns and generalize them. This type of neural network is not generally used in recognition task, but in this case problem is observed as linear separable categorization task so this neural network was good enough, according to the authors. Autoassociative memory can be viewed as a filter that can reconstruct learned shapes so it can work well even with some degree of noise in the images. These patterns in the memory are actually equivalent to performing principal component analysis to the set of provided patterns. Input in neural network is

the window (part of the image), therefore the sliding window approach is used for scanning the whole image. If the output of neural network is higher than certain threshold then that position is marked as area with capillary presence. This threshold is the important parameter because based on its value ratio between positive and false positive detections varies. If the value is too low, most of the capillaries are marked but there are also many wrong marked positions, on the other hand if the value is too high only few of the capillaries will be marked. Authors focused a lot on finding the optimal value and ratio between good and bad marked areas. Results show that on average system was able to correctly annotate 82% of capillaries in the image, while false detection rate was 10%. For these tests authors were only using the gray-scale image as the input, and they suggest that certain pre-processing techniques could improve the results. In the paper they have tried out the method with using the color information. Idea is based on that artifacts and capillaries looks similar in gray-scale image while they have different colors. Their approach was to detect a region of interest based on the color and then run the sliding window only in that area. Results show that number of false positives reduced, but not significantly. In the end they also proposed that post-processing could also improve results. For example, region that is marked as capillary could be further examined in order to see if it really is a capillary or not.

Research presented in the paper Capillaroscopy Image Analysis as an Automatic Image Annotation Problem [21] focuses on the same topic as this master thesis project, annotation of capillaries from nail-fold capillaroscopy images. Proposed auto-annotation process has two steps training and processing. For the training phase a set of already annotated images is used, where these annotated images are transferred into feature vectors. These feature vectors are later used for training. Most difficult part is getting the right features from the image, and many would think that because medial images have limited range of colors and textures that this task should not be that difficult but on the contrary this makes things more difficult and complicated. There are 4 steps for feature extraction filtering, image pre-processing, segmentation and calculation of feature vectors.

First, multi-scale TopHat filter is applied to the green channel of the RGB image, this filter proves to have good results for blood vessel and vascular extraction problems. Next step is to apply hessian matrix calculation that enhances blood vessels even more. All this is done in multi scale so the important part is that in the next step thinning is done in the correspondence to the scale. In order to perform this correctly, images are divided with rectangle grid. This method is very robust and stable but it has a disadvantage because real objects cannot be represented in the segmented image. Final step is the feature vector calculation, and for each segment in the image a feature vector is created. Feature vectors contain information from RGB color space, HSV color space, Tophat filtered image, Hessian matrix and thinned image, which makes in total 76 features for each segment. These features are calculated from mean standard deviation for each channel, size of distinct regions in the segment, region information, average location of segment in the image, mean values, features based on thinned image and zoom level. For the auto annotation three algorithms are used:

- Multiple class machine learning
- Multiple class machine learning with balanced averaging
- Continuous relevance model

Authors did not only focus on the annotation of capillaries but also on the calculation of capillary morphology, microvascular architecture, etc. However, most interesting part for this thesis project is the annotation itself so the focus will be on it. Results show the accuracy of 77% and that is rather good result. But the parameters were manually adjusted for this particular set of images and big disadvantage is long time required for finding the optimal parameters. Authors have also proposed a method for automatic parameter selection but final results of that approach are slightly worse. Thing that is not mentioned is the time required for annotation, and that is a very important parameter when it comes to implementation of this particular method into real world applications.

Further work in this filed goes in the direction of automatic detection of the state of disease. In the paper “Preliminary Clinical Evaluation of Semi-automated Nailfold Capillaroscopy in the Assessment of Patients with Raynauds Phenomenon” [20], besides the enhancement of capillaries from the image, authors want to create a semi-automated system that is able to classify the capillaries based on the disease group. This approach is based on geometric calculations of the extracted capillaries. After the calculation of capillary features, mathematical morphology was used for measuring the width. Tortuosity is calculated using orientation histograms, and based of its dispersion the dominant orientation of capillary is calculated. Based on the dispersion of dominant orientations of capillaries derangement is calculated. Finally, distance between capillaries is calculated as well and that is done by detection of central lines of capillaries and its intersection with a line parallel to the locus of the apices. Performance of the proposed system is compared with manual measurements and the results show that there is moderate correlation for the distance, width and tortuosity results while for derangement that correlation was weaker. Biggest advantage of semi-automated system is the increase in speed, and the results also show that when it comes to recognition of current stage (phase) of disease (three different groups existed in test) semi-automatic system is comparable to the manual classification. This system can save 64 minutes to doctors according to the authors who state that manual measurements take around 10 minutes per image while their system allows that to be done in 2 minutes. Limitations of the provided approach is that preprocessing and enhancement steps are optimized towards the normal capillaries and therefore some uncharacteristic capillaries can be lost during this procedure and in the end this leads to false results.

Similar work is done by Wen et al [22] during the development method that is able to distinguish between normal and abnormal capillary images. This approach uses skeletons of vessels and inputs them into the neural network. Output of the network is the group in which capillary belongs according to Cutolo taxonomy [3]:

1. Enlarged/giant vessel
2. Hemorrhages point

3. Loss of capillaries
4. Disorganization vessel
5. Ramified/bushy capillaries

Results of the system show that for normal (or usual types of disease) cases it provides good results but it still requires modification for some more complex situations. Another approach in classification of current disease stage is to use the texture from images. This approach was used by Doshi et al [23] who used texture analysis in order to discover underlying patterns. Algorithm is based on local binary pattern approach which contains both local patterns and local contrast information. These features are inputted into one-against-one SVM which is trained based on ground truth images. Tests show that this approach can provide good classification results and this may be the area to be discussed in the future.

2.2 Theoretical background

2.2.1 Contrast Limited Adaptive Histogram Equalization – CLAHE

Histogram equalization is a well known procedure for contrast adjustment in images by making the histogram of original image flatter. This method can be applied either globally on the whole image or locally to small segments of the image. When it is applied locally it is known as adaptive histogram equalization (AHE). In this case histogram equalization is applied for each segment of the image independently and this provides better contrast adjustments locally. This provides better results when there are some areas in image with more or less light than other areas. Resulting image has enhanced contrast but often noise can be too amplified especially in the areas with somewhat uniform color.

Contrast Limited Adaptive Histogram Equalization (CLAHE) [25] was developed in order to reduce the noise amplification that occurred sometimes with AHE algorithm. CLAHE is of our interest because it is an important part of this master thesis work. CLAHE limits the range of output image so that there are no high peaks in the histogram. Threshold that is used for clipping the histogram is called clip limit and it can be defined by user, as well as the size of window that is used. Another important thing is that all the parts that are above the clip limit are not discarded but are evenly distributed over all histogram bins. This step can be seen as renormalization and this makes real clip limit actually bit higher than the set one. This can be solved by multiple use of the same algorithm until that difference between real and desired clip limit becomes negligible.

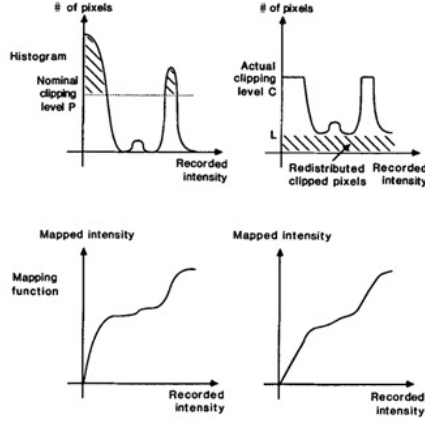


Figure 6: CLAHE[25]

2.2.2 Hough transform

The Hough Transform is well known feature extraction technique that is used in image processing. Main idea is to detect the imperfect shapes of particular class in image. The basic implementation of Hough transform was developed with the purpose of line recognition in image. Later this was generalized to recognition of circles, ellipses or even more complex shapes [26]. Since this master thesis project only uses the line detection, we will focus on that part and shortly explain the underlying algorithm. As mentioned, Hough Transform should be able to detect imperfect shapes, or in this particular case – lines. That means that lines in images are quite often noisy, not perfectly straight or certain pixels may be missing and they should be detected with this approach. First important part is to use the edge detector in order to extract the imperfect edges, and afterwards use Hough Transform to group those edges. When we talk about the lines, we know that following representation is often used:

$$y = kx + a$$

where k is the slope or the gradient of the line and a determines the point at which line crosses the y-axis. However this representation of line has problems with vertical lines since the slope parameter can rise to unbounded values. Duda et al [26] proposed the usage of Hesse nominal form for line representation:

$$r = x \cos \theta + y \sin \theta$$

where r is the closest distance of line from origin and θ is angle between that shortest line between origin and line and x-axis. So every line can be represented in the (r, θ) plane that is also known as Hough space.

For the purpose of line detection Hough algorithm uses two-dimensional array that is called accumulator. For each pixel in the image, algorithm will look at the surrounding pixels and calculate the possibility of having a straight line. If there is possibility, parameters r and θ are calculated and corresponding bins

in accumulator are increased. In the end, the highest values in the accumulator will point out to the most likely lines. Each peak actually presents a line and extraction of the peaks can be done in different manners. For example, we can introduce the threshold and take all peaks that have value higher than threshold, or we can take certain amount of highest peaks. However length of the line is not saved in the accumulator and it can be calculated by finding the extreme points of the line. However because of imperfections in lines these calculations are often not easy. OpenCV offers an implementation that is based on the progressive probabilistic Hough transform [27], an approach that optimizes the number of computations making it very useful for real-time applications. This implementation also calculates the line length and its minimum length can be specified in the algorithm.

2.2.3 Histogram of oriented gradients – HoG

Histogram of oriented gradients (HoG) is a feature detector that can be used for object detection tasks where gradient orientation in segments of image is important. It was first introduced in paper “Histogram of Oriented Gradients for Human Detection” by Dalal et al. [28] where it showed very good results for pedestrian detection. After that, same approach was used for other object recognition tasks and it proved to provide good performance. The main idea of HOG is that shapes of objects in the image can be described by the distribution of intensity gradients which point out to the dominant edge directions.

Image is first divided into small segments of the same size that are called cells, afterwards histogram of gradient directions is calculated within each cell, and in the end all these histograms are combined. The way they are combined depends on the implementation, but often 16 or 32 cells are joined into blocks and those blocks overlap (see Figure 7). Local contrast normalization can be applied across the larger segment of image called block in order to deal with varying illumination and shadowing. HOG features are invariant to geometric and photometric transformations because they operate on localized cells. In the paper [28] these features proved to be tolerant to body movement and that means that object does not have to be in the exact position in order to be recognized. This makes usage of these features perfect for object recognition because in many cases objects are not in the same position.

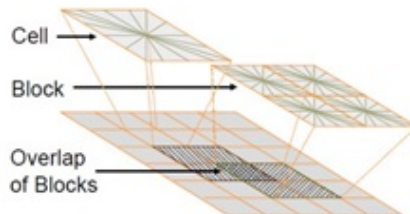


Figure 7: Histogram of oriented gradients[29]

Gradient computation:

First step in HoG feature extraction is the calculation of image gradients and this is done by applying a one dimensional centered, point discrete derivative mask in vertical and horizontal directions. Dadal states that some more complex masks such as Sobel or Canny can be used, but in the end they result in the poorer performance. After that magnitude and orientation of each pixel is calculated using the following formulas:

$$G_{mag}(x, y) = \sqrt{G_x^2(x, y) + G_y^2(x, y)}$$
$$\theta(x, y) = \arctan\left(\frac{G_y(x, y)}{G_x(x, y)}\right) + \pi/2$$

where $G_x(x, y)$ and $G_y(x, y)$ are gradient values at position (x, y) in horizontal and vertical direction, respectively.

Orientation binning:

Cells are segments of images that are rectangular in most cases and bins of the histograms are in the range between 0 and 180 degrees. Every histogram bin has a spread of 20 degrees so it means that every pixel in the cell will be distributed into one of 9 histogram bins based on its direction. Gradient magnitude is most often used for weight of votes but some other functions can also be used.

Descriptor blocks:

Gradient strengths must be locally normalized in order to obtain invariance to illumination and contrast changes. This is the reason why cells are grouped into the larger regions called blocks which overlap so that each cell can influence more than one block. Most often used parameter is 2x2 cell blocks of 8x8 pixel cells. Sometimes we are not much interested in the pixels that are on the edge of block so we can apply Gaussian spatial window before histogram voting in order to suppress those pixels.

Block normalization:

There are different ways to normalize the blocks, if we denote v the non-normalized feature vector of a block, $\|v\|_k$ its k norm and ϵ as a small positive constant then normalization schemes are following:

- L2-norm: $\hat{v} = \frac{v}{\sqrt{\|v\|_2^2 + \epsilon}}$
- L1-norm: $\hat{v} = \frac{v}{\|v\|_1 + \epsilon}$
- L1-sqrt: $\hat{v} = \sqrt{\frac{v}{\|v\|_1 + \epsilon}}$
- L2-Hys is re-normalized clipped L2-norm.

All these normalization schemes provide better results compared to the non-normalized.

HOG feature descriptor is a vector that contains elements of normalized cell histograms from all blocks from the image.

2.2.4 Support vector machine – SVM

In the pattern recognition tasks quite often we have a set of patterns (objects) that we want to recognize and based on that set we are constructing our separator of classes. There are some relatively simple methods like linear separation, or methods based on least mean squares but often they do not perform well enough in bit more complex situations. In the search of the best possible classifier, researchers have come up with many different methods like neural networks, support vector machines or many others.

Support vector machine (SVM) is a computationally powerful class of supervised learning networks for solving pattern recognition tasks. Idea of SVMs dates back to 1960s but the rapid development and usage of SVMs starts from the mid-90s with Cortes and Vapnik [30]. Aim of the SVM is to provide good separation between the classes in the training data. When it comes to separation of classes, critical points are the ones that are closest to other classes. Points that are closest to the boundary are called support vectors. If the classes are marked as $+1$, then decision boundary estimate is:

$$y = \sum_{i=1}^N w_i x_i + b = x_i w + b$$

x_i is the input pattern, w is the weight vector and b is offset. In this particular case our separation boundary would be $y = 0$, so when $y > 0$ that means that x_i belongs to class 1 and vice versa.

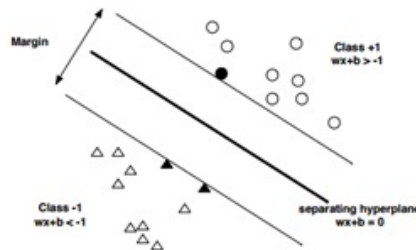


Figure 8: Support Vector Machine [wikipedia]

In order to have the good separation between classes it is necessary to take into consideration the distance of hyper plane to the origin. That distance is

called margin M and it is defined:

$$M = \frac{1-b}{\|w\|} = \frac{2}{\|w\|}$$

where w is the norm of w . We can see from this equation that in order to maximize M we need to minimize w . However, classes are often not linearly separable but still the task is to minimize M even though some points will be on the other side. This is a special case of SVM that produces linear boundaries between classes and it is called linear support vector machine LSVM.

More general case is when the classes are not linearly separable and therefore non-linear classifier is necessary. Only difference to the linear case is the kernel that is applied to input pattern so the formula now looks the following:

$$F(x) = \sum_{i=1}^N w_i k(z_i, x) + b$$

Kernel allows mapping the input data into higher dimensional space where it can be linearly separated. Classifier is the hyper plane in space with higher dimensionality than the input pattern. Various kernels can be used for transformation of input space. Some of them are the following:

Linear kernel:

This is the simplest kernel function, and it is calculated using the inner product with an offset c . It does not increase dimensionality so it is simple linear classifier.

$$k(x, y) = x^T y + c$$

Polynomial kernel:

This is non-stationary kernel and it works well when all data is normalized.

$$k(x, y) = (ax^T y + c)^d$$

Gaussian kernel:

It is a radial basis function kernel and it is good to use when we do not have much information about the data that we are modeling.

$$k(x, y) = \exp(-\gamma y^2)$$

$$\gamma = \frac{1}{2\sigma^2}$$

It is important to note that when transforming space to higher dimensions can

result in over fitting (Platt et al [31], Lewis [32]). Soft margin is one more parameter of SVM that has been introduced in order to get the better classifier when it is difficult to clearly separate classes. This method adds a new variable ξ_i which measures (penalizes) the degree of data misclassification.

$$y_i (w \bullet x_i - b) \geq 1 - \xi_i$$

This makes the optimization problem a tradeoff between large margin and small error penalty. In the linear case optimization function would be:

$$\arg \min_{w, \xi, b} \left\{ \frac{1}{2} \|w\|^2 + C \sum_{i=1}^n \xi_i \right\}$$

When it comes to selection of parameters it is necessary to choose the right γ and the Gaussian kernel C which is soft margin approach. Best combination is found with the grid search method with exponentially growing sequences C and γ . Every combination is checked with cross validation and parameters with highest accuracy are selected. SVM is good for two class problem while the generalization to multiple classes have several issues that may be difficult to overcome in some situations. Output of the SVM is uncalibrated class membership probability, and also parameters of the model are often not easy to interpret.

3 Image analysis system

This section will describe in detail the work of this master thesis project image analysis system for images obtained during nail-fold capillaroscopy examination. There are 4 goals that are set for this work and in each of the following sections solution to each goal will be described. First step is the filtering of capillaries from the image and this is very important step because all following steps are based on using the filtered image. After that image is rotated so that capillaries always point upwards and this has two advantages – one is for doctors to always have consistently rotated images and the other is for our automatic annotation system since it provides better results with correctly rotated images. Third step is calculation of width of capillaries and the final one is the automatic annotation of capillaries. During development of the whole system two performance measures were always considered – precision and speed. Speed is necessary because this software should be usable in practice.

3.1 Filtering of capillaries

Very important part of the whole system is the capillary filtering algorithm; therefore a lot of attention was focused on this particular part. First, let us look at the example image in the Figure 9. This particular image is of a very good quality without many disturbing factors like glare or blur. As we can see edges of the capillaries are not sharp so that makes their filtering challenging. According to previous studies (Chapter 2), green channel of RGB image carries most of the information therefore its usage is justified and also when it comes to performance, usage of just one channel reduces computational time.



Figure 9: Original image

After the extraction, green channel is filtered with median filter with the mask size of 7x7 pixels. Size of the kernel is determined after testing various kernel sizes on different images. This step is performed in order to remove some

of the noise that is always present in the image. In this particular image the noise removal may not be really visible but in certain cases it can provide big improvements.

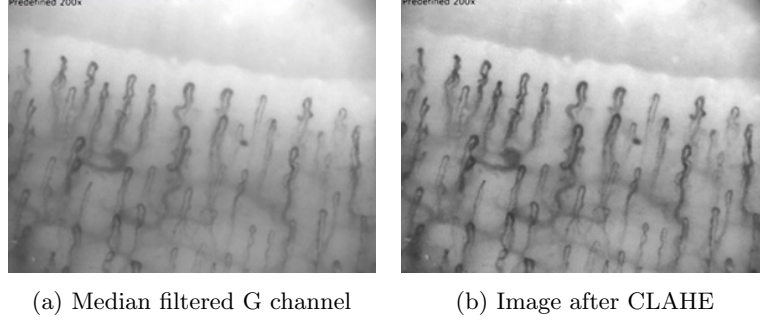


Figure 10: Preprocessed images

Next step is to apply CLAHE filtering in order to improve the contrast of image, making capillaries more distinguishable from background. As mentioned before, this also increases the noise and also the background contrast so the threshold is selected to be quite low. Based on the previous work in this field, filtering of capillary areas is based on values of Hessian matrix. Hessian matrix is a square matrix of second order partial derivatives and the basic form is following:

$$H = \begin{bmatrix} \frac{\partial^2 f}{\partial x_1^2} & \cdots & \frac{\partial^2 f}{\partial x_1 \partial x_n} \\ \vdots & \ddots & \vdots \\ \frac{\partial^2 f}{\partial x_n \partial x_1} & \cdots & \frac{\partial^2 f}{\partial x_n^2} \end{bmatrix}$$

In the case of image this would mean that we need to calculate for each pixel its second derivative, and this value would present local image intensity variations around the selected pixel. So we need to calculate second order partial derivatives Dxx , Dyy and Dxy . In order to calculate this computationally efficiently we can apply the derivative directly to the smoothing function that is used to filter the image. Simple Gaussian function is good enough for this purpose. General function is:

$$G(\sigma) = \frac{1}{2\pi\sigma^2} e^{-\frac{X^2+Y^2}{2\sigma^2}}$$

where X and Y are masks with following values and x, y are positions inside the masks:

$$X(x, y) = \begin{pmatrix} -3\sigma & -3\sigma & \cdots & -3\sigma & -3\sigma \\ -3\sigma + 1 & -3\sigma + 1 & \cdots & -3\sigma + 1 & -3\sigma + 1 \\ \vdots & \vdots & \ddots & \vdots & \vdots \\ 3\sigma - 1 & 3\sigma - 1 & \cdots & 3\sigma - 1 & 3\sigma - 1 \\ 3\sigma & 3\sigma & \cdots & 3\sigma & 3\sigma \end{pmatrix}$$

$$Y(x, y) = \begin{pmatrix} -3\sigma & -3\sigma + 1 & \cdots & 3\sigma - 1 & 3\sigma \\ -3\sigma & -3\sigma + 1 & \cdots & 3\sigma - 1 & 3\sigma \\ \vdots & \vdots & \ddots & \vdots & \vdots \\ -3\sigma & -3\sigma + 1 & \cdots & 3\sigma - 1 & 3\sigma \\ -3\sigma & -3\sigma + 1 & \cdots & 3\sigma - 1 & 3\sigma \end{pmatrix}$$

and σ is a parameter that determines the amount of smoothing that will be performed on the image. This also determines the scale at which this operation will be performed and this parameter seems to be quite important for us since it depends on the width of capillaries. The value that provides best results on provided images is $\sigma = 7$ and results obtained with different values will be discussed in Chapter 4. Second order Gaussians are calculated using the following formulas:

$$Gauss_{xx}(x, y) = \frac{1}{2\pi\sigma^4} \left(\frac{X(x, y)^2}{\sigma^2} - 1 \right) e^{-\frac{X(x, y)^2 + Y(x, y)^2}{2\sigma^2}}$$

$$Gauss_{yy} = Gauss_{xx}$$

$$Gauss_{xy}(x, y) = \frac{1}{2\pi\sigma^6} X(x, y) Y(x, y) e^{-\frac{X(x, y)^2 + Y(x, y)^2}{2\sigma^2}}$$

These masks should now be applied to the image, but since these filters are linearly separable we will first separate them and then apply in separated form. This provides exactly the same results as without separation but reduces the number of computations. This means that processing time is reduced and that is very important for practical usage of this filtering technique (see Figure 11). The output image is then calculated using the following equation:

$$Output = \sigma^2 \left(\sqrt{\sigma^2(D_{xx} - D_{yy})^2 + 4D_{xy}} + \sigma(D_{xx} + D_{yy}) \right)$$

After the 7x7 pixels median filter is applied the output image we get the final image (see Figure 12)

This example image that is used here is of very good quality without many disturbing factors so the resulting filtered image is also very good and capillaries are well extracted from the image. In some other examples we can see that also some glare and other artifacts will be filtered with this algorithm.

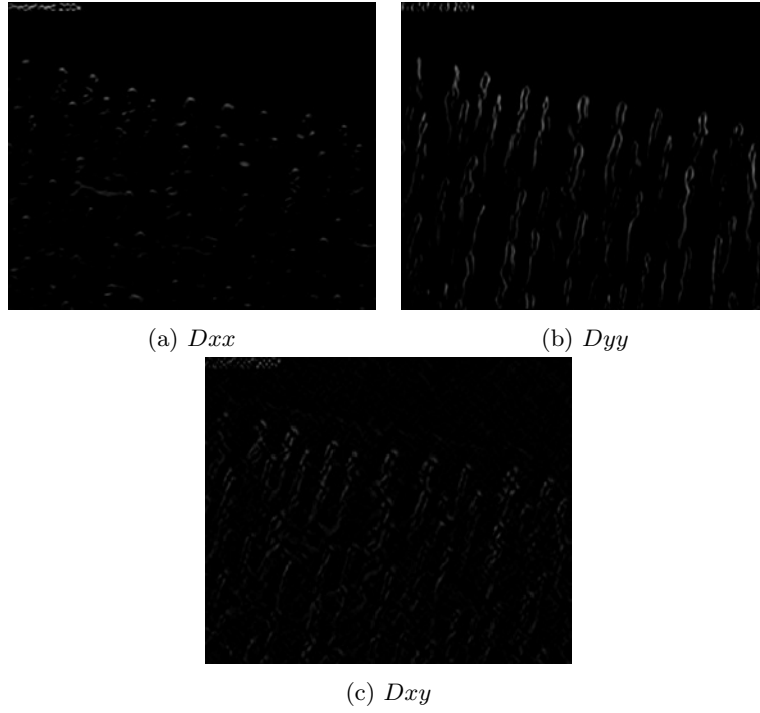


Figure 11: Second derivatives



Figure 12: Filtered image

3.2 Rotation of image

Next step in the system is to rotate the image so that capillaries point upwards. Filtered image is used in order to determine the orientation of capillaries and based on them an angle of rotation. Algorithm for this purpose is based on Hough Line Transform that is used to detect straight lines in the image. First step is to transfer the filtered image into the binary image using the Otsu threshold [24]. It is advisable to use some edge detector before applying the Hough Line Transform but considering the characteristics of filtered image, binariza-

tion seems to be a good solution and it would allow us to use the probabilistic Hough transform for lines detection [27]. Parameters that can be set are the distance resolution, angle resolution, threshold parameter, minimum line length and maximum gap between points on the same line but what is most important for us is to take the line length and limit it to minimum of 100 pixels. After the lines with the specified parameters are found, based on their direction lines that are vertical, horizontal and have angle larger than defined threshold (angle between -40 and $+40$) are filtered and this is the result where detected lines are marked with red. This angle is selected because often in that selected range straight lines may occur from the glare and we do not want to use that for our calculations.

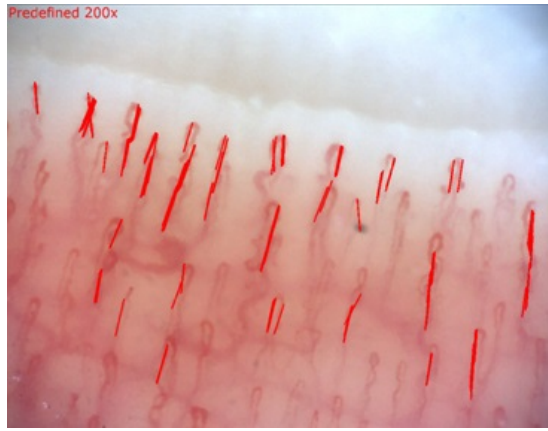


Figure 13: Image with marked lines

Next step is to determine the dominant direction of capillaries based on the direction of each line and for that we need to sort them first. It is important to be careful in this step since some of the angles are actually negative so we need to add or subtract 90 degrees in order to get the angle relative to vertical line. Once we have that, we take the middle value from that sorted array that is actually median value. In order to provide more precise results we also take two values around median, and then average them in order to get the final angle. In certain cases when we do not have enough of lines, or when number of positive and negative is similar that may mean that rotation of extracted lines is not consistent so rotation angle is outputted as 0. Based on the calculated angle rotated image is calculated and output is shown in Figure 14.

This function should provide doctors with consistently rotated images because it is difficult to always obtain images with good rotation. Another very important use of rotated image is during the automatic annotation of capillaries and that will be discussed later in this Chapter.

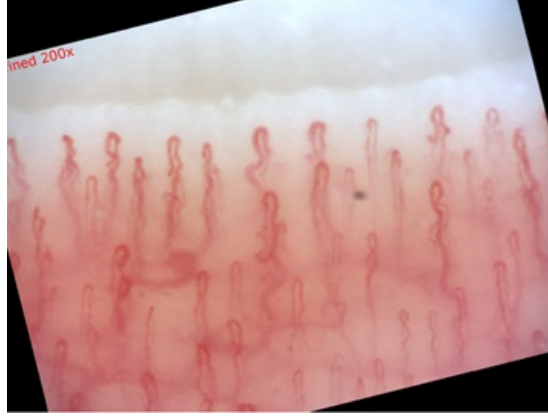


Figure 14: Rotated image

3.3 Width calculation

As it is already mentioned in Section 1, normal procedure during capillaroscopy examination is to measure the width of capillaries at certain positions. Main idea is to use the filtered image that we have for calculation of capillaries width. Filtered image provides a very good approximation of capillary boundaries so what now needs to be done is calculation of width on the selected line. However on certain location, especially around the curvatures, certain artifacts appear, so in order to compensate for those artifacts, image is first preprocessed. Same as in rotation algorithm, image is binarized with the Otsu threshold. Since the artifacts are usually darker than central part of capillaries during this thresholding they will be discarded. After that Canny edge detector is used to extract edges from the binarized image, and finally these edges are added as black pixels to the filtered image. This is necessary because the edges themselves are often not connected so the calculations cannot be directly done on them, and once we add the images we can use all values from filtered image and we can recognize the places where the added edges are.

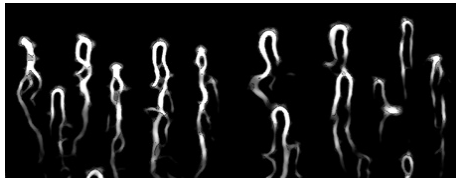


Figure 15: Edges added to filtered image

The line on which the measurement will be done is defined by two points that are manually inputted by clicking at desired locations. After the points are defined, we extract the values of pixels that lay on the line between the points. Another important aspect is the orientation of line and how are the pixels sampled. It is checked where is larger coordinate difference — on x or y axis, and on the one with larger difference pixels are sampled. This increases precision of

our calculations. We also calculate the angle of rotation of defined line so that we can use cosines or sinus to measure the real width. Basically that means that the length that we want to calculate is the hypotenuse of triangle and depending if we have x or y (Figure 16) axis length we can calculate it using cosine or sinus of line slope.

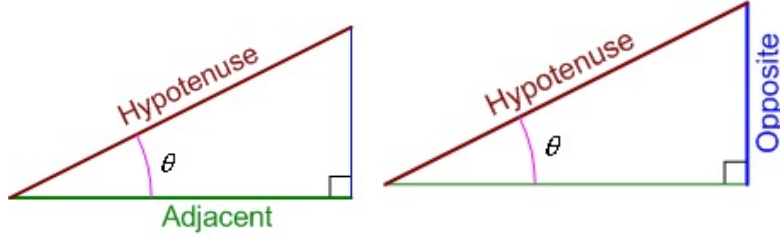


Figure 16: Length calculation

From the pixel values (Figure 17) we can clearly see that many values are 0 and they represent background and these peaks are actually capillaries. We can also see where are our edges from preprocessing (points where peaks go to 0). Based on this we can measure the width of peak based on the number of pixels. Adding the edge into the filtered image has its advantages but also its flaws. As mentioned advantage is that we can remove artifacts but the flaw is that we limit the peaks in histogram with these zero values. For example, if we want to calculate the whole peak width we would have to somehow skip this zero values or before extraction of edges we could dilate the binarized image.

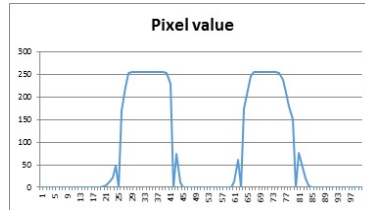


Figure 17: Capillary width calculation histogram



Figure 18: Capillary width marked image

3.4 Annotation of capillaries

The final and most challenging part of the system is the automatic annotation of the capillaries and for this purpose window sliding technique is used together with HoG features and support vector machine. For this part we use rotated filtered image. It is important to have rotated image since our SVM is trained to recognize capillaries that point upwards.

Since there is a need to go through the whole image, or at least specified part of image, sliding window technique is chosen because this is most often used method for this purpose and it provides good performance. Size of the window is 64x64 pixels and this size is chosen by taking few parameters into consideration. First one is the size of window itself because it is necessary to cover the whole tip of the capillary, and the second is about HoG parameters. There are certain limitations with the current implementation of HoG features in OpenCV so the only supported cell size is 8x8, therefore we have selected window size to be 64x64. This way we end up with 576 parameters per window. It is also possible to change these properties and that would lower the number of parameters and required time but the efficiency would also be affected. This will be more discussed in next section.



Figure 19: Training set – positive samples

Support vector machine is used for classification of windows if they are from a capillary or not. Very important part for the good results is to have a good training data. For this purpose data is collected manually from the filtered images and there are 150 positive (Figure 19) and 150 negative samples in the training data. Each sample is 64x64 pixels as our window, and for each sample HoG feature vector is calculated. These features vectors are used as the input in the SVM, and the output is distance from the border line. So if the output is a positive number the window is considered to be a capillary area and if it is negative, area is background. We have included also a threshold, so the number has to be larger than specified threshold so that it can be considered as capillary, this way we put more strict condition. As this threshold changes we get different results in the end, depending what is more important to mark only capillaries for which system is sure that are capillaries and have a small number of false positives or mark most of the capillaries but also get higher positive rate.

When the window gets output from SVM that is higher than specified threshold, that area is marked with white circle on black background. So we get a

binary image with marked locations (see Figure 20).

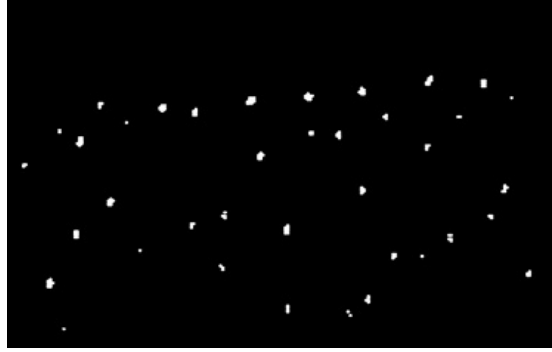


Figure 20: Marked positions

This image is then further processed using morphological operation dilation in order to better connect the circles. After that contours are extracted together with their area sizes. If their area is larger than specified threshold that means that enough windows at that location were above the SVM threshold and we can mark that position as capillary. This threshold can also be changed and it affects the final result in similar manner as the SVM threshold. In the final image, capillaries are marked with blue circles (see Figure 21).

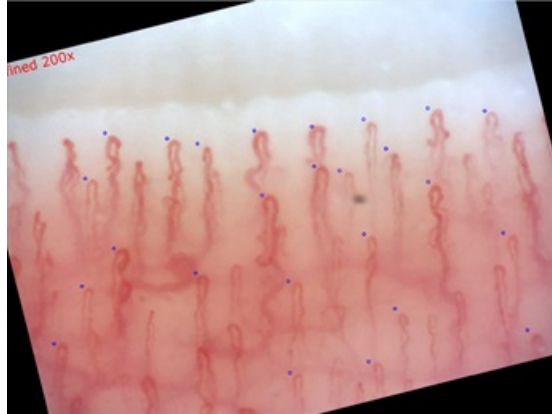


Figure 21: Annotated capillaries

In this example image and with the selected thresholds, most of the capillaries in the image are marked, but still some of them remain unmarked. For the demonstration purposes only one fixed setting is used but in the next section different parameters will be used and results will be discussed.

4 Experimental setup and results

This section will present the data that is used during the development and testing of the system. Experimental setup will be presented, system will be tested with different parameters and results will be discussed and compared with other approaches.

4.1 Data

Images in our dataset are obtained using Optilia “Mediscope” digital microscope with 200x magnifying lenses. Images have resolution 2000x1500 pixels, and in our testing dataset there are 55 images divided into 4 groups that are provided for the purpose of the thesis project by Optilia Instruments AB. Some of the images are obtained by employees at Optilia Instruments AB during the testing of the microscope while most of the images come from doctors. As for the images that come from the doctors, identity of both doctors and patients is anonymous even for the author this project. Images in dataset contain mostly normal capillaries or the ones from the early stage of Reynolds disease. Abnormal capillaries can have very different shapes and sizes and that would make the task of filtering and annotation much more difficult, and system is also developed with the main focus on normal capillaries. Furthermore, goal of this tool is to help doctors to speed up and improve examination procedure so this tool will help in the part that always has to be done and often takes a lot of time from doctors.

One of the issues for this project was getting the ground truth images with which we can compare our results. This is especially interesting in the case of annotation of capillaries since not all of the capillaries in the image should be annotated. Even though there are certain rules when it comes to annotation, those rules differ from doctor to doctor. In our dataset we have 20 images that are annotated by doctors using two different techniques therefore we have divided them into two groups of 10 images. First group we will name Doctor A group, and images in that set are annotated in the following way:

- A line that represents the first distal row is drawn
- All the capillaries that are on or above the line are annotated. This includes even the capillaries that are barely visible

Doctors always calculate the capillary density and it is expressed as number of capillaries per square millimeter. Therefore in the Doctor B approach there are following steps:

- Region of 1mm x 1mm is selected and marked. This region is usually selected by taking a part of image where there are no disturbing factors
- Imaginary distal row line is drawn
- Capillaries above the line are counted, however not all of them. In this dataset few different doctors with the same technique marked images but they had different criteria on which capillaries should be marked

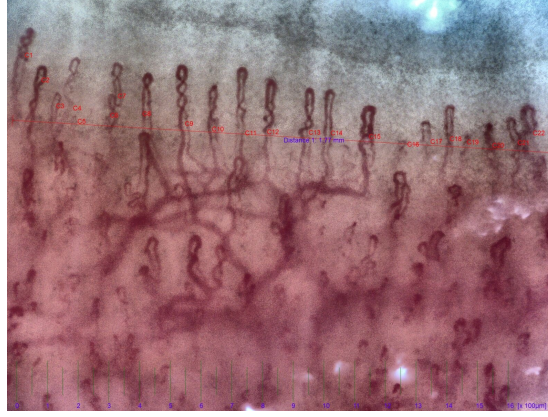


Figure 22: Doctor A dataset sample

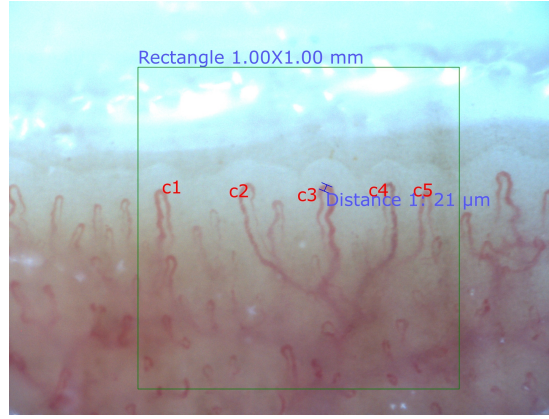


Figure 23: Doctor B dataset sample

It is noticeable that these two methods provide us with different results; therefore in order to obtain ground truth images manually from the rest of the data set following rules are used:

- Region of 1mm x 1mm is chosen so that it takes part of the image with the least disturbing factors (blur, out of focus, glare, etc.)
- Distal row line is taken so that it is around the second row of capillaries (second from up to down)
- Capillaries on and above the line are annotated but only the ones that are visible enough, so the capillaries that are deeper in the skin are excluded

This method is more similar to the ones used by Doctor B, and it is based on examination method described by Cutolo et al [3].

In our data set there are 30 images with normal capillaries (normal refers to their size and width, however some of them have high level tortuosity) and

	Doctor A	Doctor B	Man. normal	Man. enlarged
Number of images	10	10	30	5

Table 2: Test dataset

5 images of enlarged capillaries so we can consider that we have two distinct groups in our manually marked dataset normal and enlarged capillaries.

4.2 Experimental setup

For the purpose of testing, simple interface is created using Qt in order to provide us with a good, simple and fast testing environment. All important input parameters can be easily changed in the interface and it also provides us with useful output parameters like processing time.

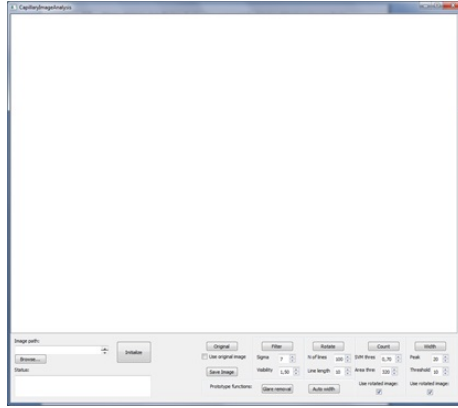


Figure 24: User interface

Computer that is used for the testing has following specifications:

- Intel®Xeon®CPU E3-1225 v3 with 4 CPUs at 3.20 GHz
- 4 GB of RAM

Graphics card is not used in the implementation because software will be used in commercial purposes and run on customers computers, so there may be difficulties with certain configurations. However, using the GPU for image processing operations should speed them up, and that may be interesting to implement in future.

4.3 Results

4.3.1 Filtering, Rotation and Width calculation

Most of the algorithms proposed in research papers were focused on enhancement of capillaries or vessels regions in general, and not in the complete extraction of these regions. Even in the papers that proposed edge extraction

techniques, there were often some other parts that were extracted so further processing was necessary. In order to compare our proposed method we will use method from paper [18] as a reference since the final result of that approach is a black and white image that should clearly extract capillaries from the background. In the following figure we can see the example of two images and results obtained with both approaches. Image of the left is of a very good quality and with small amount of disturbing factors while the image on the right has a glare from the immersion oil. When the image is of a good quality method from paper [18] can filter out the capillaries in the first distal row, but all the other ones are only partially filtered. However first distal row is often the only region that is of the interest for doctors, so this can be good enough in certain situations. When we look at the image which has some amount of glare, we can see that this method extracted and amplified the glare while capillaries are not filtered well. If we compare this to our proposed method, we can clearly see the difference in the quality of obtained images, where glare is extracted but not amplified and what is more important most of the capillaries are extracted together with their original shape. This result provides us with other possibilities and further processing. For example, if the capillary regions are precisely extracted we can precisely measure the width of capillaries (see next section), furthermore we can calculate the percentage of capillary are in the image and this may hold certain information about the disease stage but this requires further research.

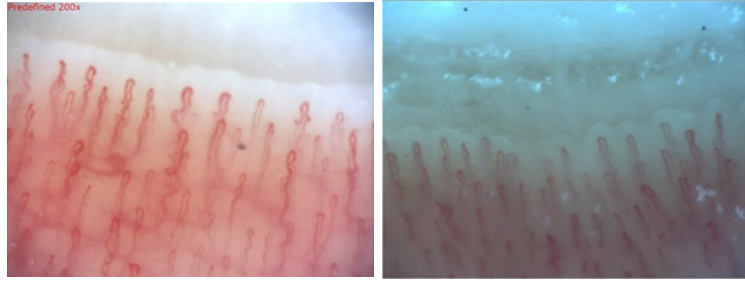


Figure 25: Original images

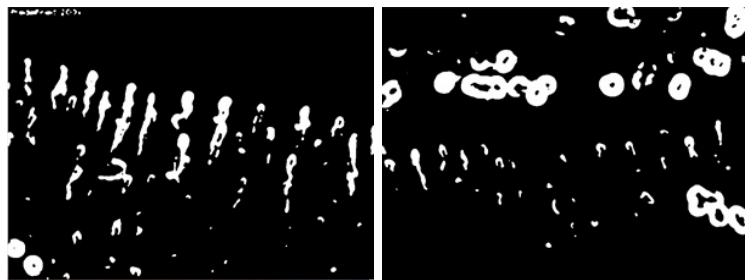


Figure 26: Filter from paper [18]

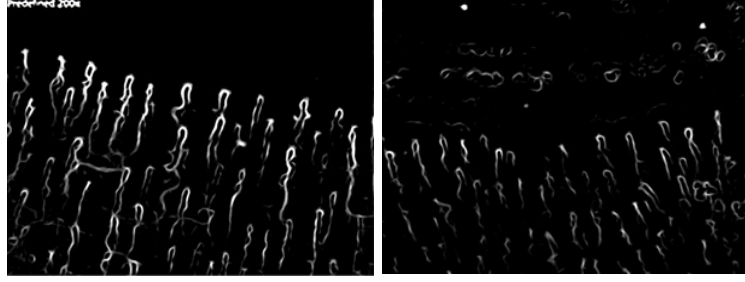


Figure 27: Proposed filter

One important parameter of our proposed filtering algorithm is the sigma parameter. As it is already mentioned this parameter affects the scale of filtering. In practice this means that sigma should be associated with the width of capillaries. Since we are working with fixed magnification and normal capillaries we can set up the parameter sigma to be fixed and by doing this we speed up the processing time. As we increase the sigma so we increase the size of matrix that is used for filtering, also number of computation and computational time increase. In Table 2 we can see numbers about the filtering time with different sigma values.

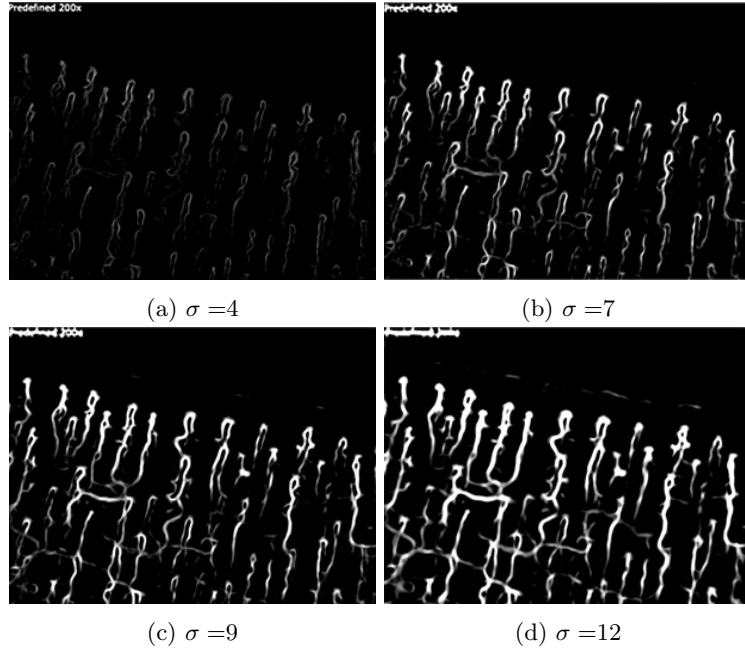


Figure 28: Different sigma values

As we can see from Table 2, multiscale filtering would take a lot more time since it would be necessary to calculate filtered image for each sigma and then put all that into the final image. Speed is an important aspect of the software since it is not practical if doctors have to wait very long time in order to get

	$\sigma = 4$	$\sigma = 6$	$\sigma = 7$	$\sigma = 9$	$\sigma = 12$	$\sigma = 15$
Average time (sec):	0.67	0.82	0.89	1.11	1.60	2.38

Table 3: Processing time

filtered image. In the following figure we can see the resulting images for different sigma values and after testing with different images we have concluded that sigma should have value 7 since it provides the best filtering results in most cases. Time of 0.89 seconds on average is good enough for this filter to be used in practice.

Next part of our algorithm is the automatic rotation of images. If we take a look at the images from our dataset, out of 35 different images (in the datasets we have sometimes same image in two different sets) we got none of them rotated in a wrong way and 4 of them were not rotated. The rate of not rotated images is 11,5%, and most of the images did not really need rotation, so we cannot consider that as an error.

However, rotation of images has some limitations since it takes in consideration only certain angles, so if the capillaries are rotated too much this algorithm will not detect them. As we could see from images in our database that occurs very rarely in practice but it still is a limitation of this approach. This limitation was necessary because of glare and foreign objects can often provide wrong directions and image would not be rotated properly. And this is example of what would happen without that condition (see Figure 29).

If we look at the Figure 30 we can see two images that were not rotated because algorithm could not determine the dominant rotation of capillaries. Image on the left is a bit noisy and with glare so capillaries are not that distinct from background. Therefore our algorithm was not able to determine the angle of rotation so image was not rotated. The reason for this is that there was similar number of positive and negative line angles and this is one of the conditions that points out that dominant rotation of capillaries cannot be determined. Image on the right has some enlarged capillaries and also many of them already point upwards. Once the vertical lines are removed the ones that remained were not sufficient to determine the dominant rotation, so again no rotation is performed. In both of these cases not rotating the image was a good decision and we cannot consider this as an error.

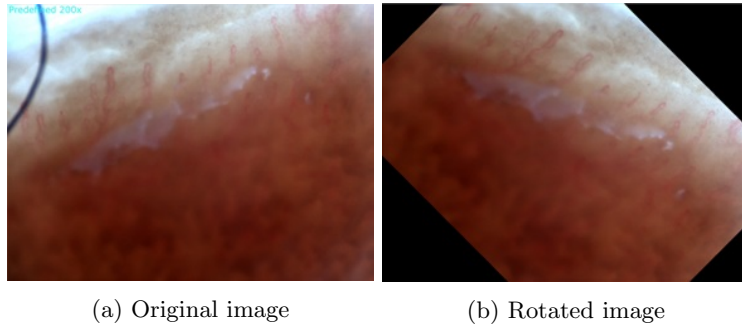


Figure 29: Example of wrong rotation



Figure 30: Unrotated images

Next part of the algorithm is the width calculation, and the only way to compare these results is by simple observation. When capillaries are clearly filtered out and without any artifacts, algorithm provides very good and precise results. However, artifacts can often occur so there are added edges to the image, and this can sometimes provide good results but sometimes can also provide false results. Examples:

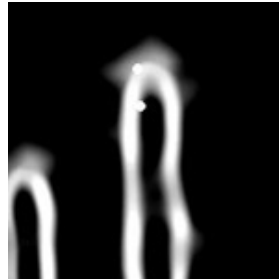


Figure 31: Width calculation with added edges



Figure 32: Width calculation

Sometimes the edge is detected inside the capillary so we get the black line inside the capillary and this affects the results, making one of the widths not

calculated. This is one of the flaws that addition of edge to filtered image causes. Another issue is that we cannot calculate the complete width of peaks in line histogram because those inserted zeros cut the peaks however this could be solved by either modifying the algorithm so that it skips the zeros or with different approach in edge extraction.

When capillaries larger than normal appear, they often do not have good enough boundaries or they have a black area inside. In this case algorithm can make false calculation, or make two calculations instead of one. This is definitely an issue that should be considered in the future, since usually the enlarged capillaries are the ones that are measured. However this is also the limitation of using only one sigma value during filtering, so the multiscale filtering may provide us with better filtered image and better width calculation of enlarged capillaries.

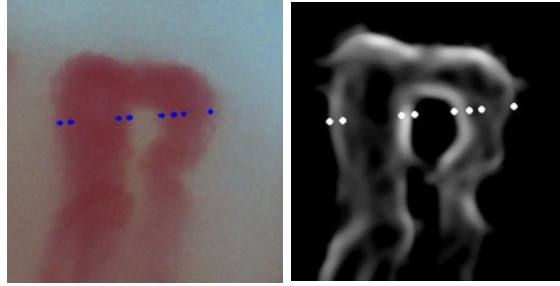


Figure 33: Enlarged capillaries issue

4.3.2 Annotation of capillaries

For the automatic annotation part, we have set up 3 sets of ground truth images, 10 from Doctor A, 10 from Doctor B and 30 manually marked. In addition to that, there are also 5 manually marked images with enlarged capillaries in order to see how the system will perform in that situation. For the testing purposes we will compare each group separately with different SVM settings. Different settings should provide us with different number of marked capillaries as the threshold goes down number of annotated capillaries is expected to increase but also number of false positives should also increase. Tests will be conducted with 7 different thresholds in the SVM 0, 0.2, 0.5, 0.7, 0.9, 1.1 and 1.3. As for the number of successfully marked capillaries, only the ones that are marked in ground truth images are considered. However, it happens that when the threshold goes down that some of the capillaries that are not marked in the ground truth images become marked during test. However, we cannot consider this as an error because the real capillary is actually marked. For false positives only situations when glare (or some other disturbing factor) is marked, or a capillary is marked twice are considered as error while false detection rate is calculated as number of false detection among all the discoveries. In the Doctor A dataset, as we have seen, even the barely visible capillaries are marked so this set is expected to be most difficult to match.

Threshold:	0	0.2	0.5	0.7	0.9	1.1	1.3
Number of marked (168):	142	130	113	93	75	48	14
Percentage of marked:	84.5	77.4	67.3	55.4	44.6	26.6	8.3
False marked:	10	5	0	0	0	0	0
False detection rate:	6.6%	3.7%	0%	0%	0%	0%	0%

Table 4: Doctor A dataset

Results show that our algorithm is capable to mark almost 85% of all capillaries, however with this threshold there is a certain number of false positives. In order not to have false positives it is possible to mark 67% and this is quite good result considering the characteristics of this dataset. As for the processing time, it depends on the image itself because black regions in images are not processed therefore if there are not many black regions it will take more to process the whole image. Time varied from 0,48 to 2,12 seconds per image with an average of 1,3 sec for whole image. It is important to note that this is the time for whole image, while in most cases it would be necessary to scan only a part of image.

Threshold:	0	0.2	0.5	0.7	0.9	1.1	1.3
Number of marked (85):	81	75	61	49	33	19	6
Percentage of marked:	95.3	88.2	71.8	57.6	38.8	22.3	7.1
False marked:	9	4	0	0	0	0	0
False detection rate:	10%	5%	0%	0%	0%	0%	0%

Table 5: Doctor B dataset

Doctor B dataset contains images that are marked by couple of different doctors that use different criteria on which capillaries should be marked and which ones should not. So we can consider this data set to be made of mixed images since there is no one specific rule that is used. Overall image quality in this dataset is lower in than in the Doctor A dataset. Results show that it is possible to annotate up to 95% of capillaries but with the false detection rate of 10%. With the zero false detection rate, it is possible to annotate 72% of capillaries. Processing time varied from 1.05 to 1.84 sec with the average time for whole image of 1.42 seconds.

Threshold:	0	0.2	0.5	0.7	0.9	1.1	1.3
Number of marked (229):	227	224	204	171	120	68	26
Percentage of marked:	99.1	97.8	89	75.7	52.4	29.7	11.4
False marked:	37	13	1	0	0	0	0
False detection rate:	14%	5.5%	0.5%	0%	0%	0%	0%

Table 6: Manually marked normal capillaries dataset

Results from the images that are manually marked show that it is possible to annotate almost all capillaries in images (99%) but the false detection rate in this case is 14%. It is also important to point out that with threshold 0 almost every capillary in the image is annotated and often doctors do not want

to annotate all of them. So this means that apart from the false positives, some doctors would also have to remove markings from certain capillaries. When false detection rate falls to negligible level of 0.5%, algorithm successfully annotated 89% of capillaries in the images. Average time required for the annotation of whole image was on average 1.35 seconds.

Threshold:	0	0.2	0.5	0.7	0.9	1.1	1.3
Number of marked (25):	23	19	14	6	4	1	1
Percentage of marked:	92	76	56	24	16	4	4
False marked:	11	6	2	0	0	0	0
False detection rate:	32.3%	24%	12.5%	0%	0%	0%	0%

Table 7: Manually marked enlarged capillaries dataset

There are only 5 images with enlarged capillaries but this set is important because it shows us how the algorithm behaves in abnormal situations. As we can see from the results it is possible to achieve quite high result of 92% of successfully annotated capillaries but that comes with price of very high false detection rate of 32%, which means that every third point is actually the false positive. Even with setting the SVM threshold to 0.5 we retain relatively high false detection rate of 12.5% and 56% of successfully annotated capillaries. Average time for processing the whole image is 1.38 seconds. These results are not that bad, but they prove that our algorithm is not very good in abnormal situations and for this part further research and development is required. Since these abnormal cases occur we will include them as well in our final results in order to get more realistic figures.

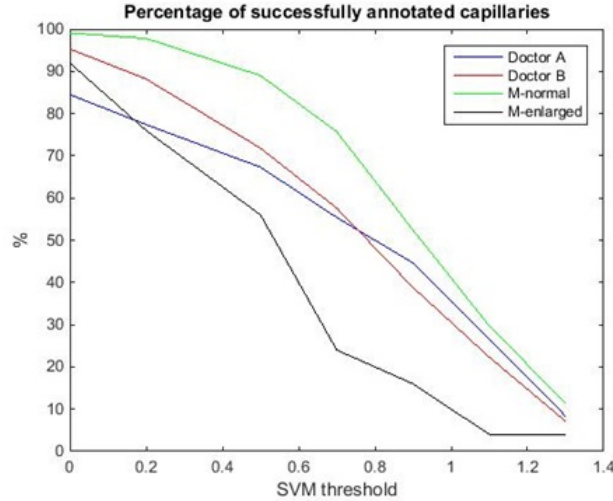


Figure 34: Successfully annotated capillaries

Finally, all the results are combined in order to get the performance of the proposed annotation system. Setting the unique threshold is quite difficult

Threshold:	0	0.2	0.5	0.7	0.9	1.1	1.3
Number of marked (507):	473	448	392	319	232	136	47
Percentage of marked:	93.3	88.4	77.3	62.9	45.8	26.8	9.3
False marked:	67	28	3	0	0	0	0
False detection rate:	12.4%	5.9%	0.8%	0%	0%	0%	0%

Table 8: Whole dataset

since it is already explained that selection of capillaries to annotate is quite subjective and differs from doctor to doctor. When the threshold is low, number of marked capillaries is very high but also false positives appear and it may happen that some of the marked capillaries should not be marked. Also, during discussions with Optilia Instruments AB, we concluded that it would be easier for doctors to add few more capillaries manually than to remove the falsely marked ones. Taking all this into consideration, threshold of 0.5 may seem the most appropriate one since the false detection rate is very small – 0.8%, while the number of successfully marked capillaries is 77.3%.

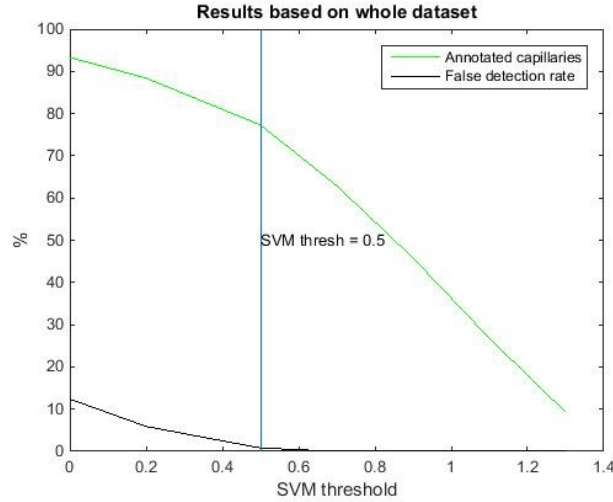


Figure 35: Successfully annotated capillaries

Sainthillier et al [19] achieved results of 82% detection rate which is a bit better than our algorithm with SVM threshold of 0.5, however their false detection rate was 10% and that is significantly higher than proposed algorithm. In order to match the same false detection rate, our system would have around 90% of detection rate. Murray et al [21] reported accuracy of 77% which is almost the same as in our algorithm. However, they did not mention the false rate detection and the result they achieved is with manual parameter setting which takes quite some time.

When it comes to processing time, all the numbers that are presented here are based on annotation of the whole image. However, as we see from the dataset quite often only capillaries inside the square millimeter should be annotated, and

even not all of them but only the ones that are above the imaginary line. On average it took 1.38 seconds to annotate one image so that means that if we limit the area this can be significantly reduced. Area of the whole image is around 2.3 square millimeters and that would mean that for one square millimeter processing time would be around 0.6 seconds. This leads to a conclusion that this algorithm is fast enough to be used in practical purposes, and this time can be even further improved by reducing the scanning area even further.

When we look at the Doctor A dataset there is a logical thought that this annotation problem can be solved with a very simple algorithm where user would define line and based on number of intersections of line and capillaries we can annotate them. Therefore, we can use our width calculation tool to manually set up the points and based on the extracted widths we can annotate the capillaries. This annotation algorithm is much simpler than the proposed one and it is also much faster. Results show that with this approach we get 69.4% of successfully marked images with only 2.5% of false detection rate. These results are not that bad but it should be pointed out that grouping of points was done by human. It means that some of the capillaries are cut twice by the line and some are cut only once so it is necessary to develop an algorithm that could cluster those points successfully. Another possibly big flaw of this approach is that some of the capillaries can be completely above the line so they will not be marked. In many cases there are capillaries that are completely above the line. For example we can see in Figure 36 where the line is drawn at the same position as in Doctor A dataset but many capillaries remain above the line. Maybe the solution could be to draw multiple lines but again there is a problem of clustering the points from different lines and deciding if they come from same or different capillaries. False positives occur because there can be certain artifacts in the filtered image, and then they are also marked as capillaries.

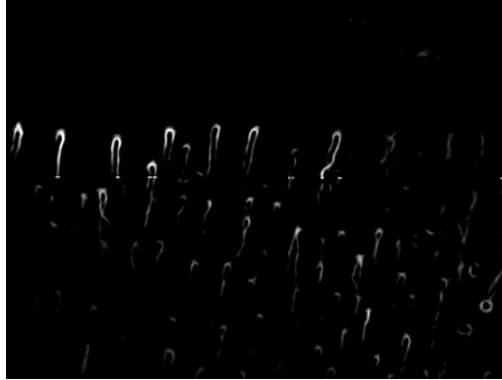


Figure 36: Line algorithm

Algorithm proposed in this thesis proves to have better results and that is more robust than the simple line algorithm. Another big advantage of proposed algorithm is that it recognizes the tip of the capillary and therefore we can get the exact location of each capillary. This not only looks better but it is also important for future work where it may be necessary to extract capillary in order to determine its tortuosity or other characteristics.

5 Discussion and future work

This thesis work was focused on a development of system that has a main goal to improve the capillaroscopy examination with the usage image processing and machine learning techniques. The goals were set up based on the current needs of the doctors and the current research areas in this field. As we can see from the results, this system shows quite good results and performance that allows it to be used in practice. All of the features, except for width calculation, will be implemented in next version of OptiPix software developed by Optilia Instruments AB.

From the practical point of view, results show that this system can efficiently accomplish tasks such as width measure and annotation of capillaries that was previously done manually. This can save time and energy for doctors and allow them to focus on other, more important things. However, this system is currently optimized to work with normal capillaries so the logical step for future research would be to work with abnormal, enlarged capillaries. One of the ways that this could be accomplished is by using the multiscale filtering and also multiscale windows for scanning the images during annotation. This all leads to more computations and more processing time, therefore simplification of current algorithm and optimization should be taken into account as well. Many of the research papers presented in Section 2 focused mostly on precision and final result, while processing time was not on priority list. However, when it comes to implementation of software for practical purposes, processing time is very important because if the same thing can be done faster (and often better) manually then there is no use of this type of software tool.

From the academic point of view, this thesis project presents a novel way of solving the problem of automatic annotation of capillaries in digital image. In this specific field, there are not that much research and that makes this work even more valuable. Obtained results show that this system can outperform other systems [19][21] that are also designed for capillary annotation. However, it is quite difficult to directly compare these systems because different datasets are used. Images are not even obtained in the same manner, and having a device like Mediscope that provided us with a high quality images certainly had an effect on final results.

For the future work there are two directions that should be pointed out. One is of course the further development of this kind of algorithms that would become more robust and be able to successfully annotate different types of capillaries. As already mentioned in the Results section, proposed algorithm could be improved with multi-scale filtering and different window sizes when scanning for capillaries to annotate. However, as we could see that quality of image is very important for good filtering and that leads to better overall results. Further hardware development could provide us with even better images but it may be interesting to also research on software tools that could remove glare from images. In this field there are many researchers [33][34] that are dealing with removal of glare that occurs during medical imaging. Even though they are dealing with glare that occurs from reflection of light from tissue and not from immersion oil that we have in capillaroscopy, these findings may still be useful and probably adapted for capillaroscopy as well.

Another direction for future research and development would be to classify the capillaries based on their class. This would mean that we already know the

position of capillaries and that we can extract them in order to perform further analysis. Based on the width, tortuosity and some other parameters it should be able to classify capillaries based on classification proposed by Cutolo et al[3]. This specific topic has already attracted attention of researchers [20] [22] and there have promising results. Based on those results it would be possible to automatically detect current stage of disease.

6 References

- [1] Cutolo, M., et al. "Nailfoldcapillaroscopy is useful for the diagnosis and follow-up of autoimmune rheumatic diseases. A future tool for the analysis of microvascular heart involvement?." *Rheumatology* 45.suppl 4 (2006): iv43-iv46
- [2] Souza, Eduardo Jos do Rosrio, and Cristiane Kayser. "Nailfoldcapillaroscopy: relevance to the practice of rheumatology." *Revista Brasileira de Reumatologia (English Edition)* (2014).
- [3] Cutolo, Maurizio, ed. *Atlas of capillaroscopy in rheumatic diseases*. Elsevier srl, 2011.
- [4] Doshi, Niraj P., Gerald Schaefer, and Arcangelo Merla. "An evaluation of image enhancement techniques for capillary imaging." *Systems, Man, and Cybernetics (SMC), 2012 IEEE International Conference on*. IEEE, 2012.
- [5] Gonzalez, Rafael C. *Digital image processing*. Pearson Education India, 2009.
- [6] Hamar, Gabor, et al. "Markov chain based edge detection algorithm for evaluation of capillary microscopic images." *11th Mediterranean Conference on Medical and Biomedical Engineering and Computing 2007*. Springer Berlin Heidelberg, 2007.
- [7] Doshi, Niraj P., Gerald Schaefer, and Shao Ying Zhu. "An Improved Binarisation Algorithm for Nailfold Capillary Skeleton Extraction." *Systems, Man, and Cybernetics (SMC), 2013 IEEE International Conference on*. IEEE, 2013.
- [8] Wen, Chia-Hsien, Wei-Duen Liao, and Kuan-Ching Li. "Classification framework for nailfold capillary microscopy images." *TENCON 2007-2007 IEEE Region 10 Conference*. IEEE, 2007.
- [9] Wen, Chia-Hsien, et al. "A novel method for classification of high-resolution nailfold capillary microscopy images." *Ubi-Media Computing, 2008 First IEEE International Conference on*. IEEE, 2008.
- [10] Lo, Lun-chien, John Y. Chiang, and Yu-shan Cai. "Three-dimensional vision-based nail-fold morphological and hemodynamic analysis." *Bioinformatics and Bioengineering (BIBE), 2011 IEEE 11th International Conference on*. IEEE, 2011.
- [11] Doshi, Niraj P., Gerald Schaefer, and Arcangelo Merla. "Robust Nailfold Capillary Skeleton Extraction." *Emerging Trends in Engineering and Technology (ICETET), 2012 Fifth International Conference on*. IEEE, 2012.
- [12] Frangi, Alejandro F., et al. "Multiscale vessel enhancement filtering." *Medical Image Computing and Computer-Assisted Intervention MICCAI 98*. Springer Berlin Heidelberg, 1998. 130-137.

- [13] Lorenz, Cristian, et al. "Multi-scale line segmentation with automatic estimation of width, contrast and tangential direction in 2D and 3D medical images." CVRMed-MRCAS'97. Springer Berlin Heidelberg, 1997.
- [14] Sato, Yoshinobu, et al. "3D multi-scale line filter for segmentation and visualization of curvilinear structures in medical images." CVRMed-MRCAS'97. Springer Berlin Heidelberg, 1997.
- [15] Riao-Rojas, J. C., et al. "Segmentation and extraction of morphologic features from capillary images." Artificial Intelligence-Special Session, 2007. MICAI 2007. Sixth Mexican International Conference on. IEEE, 2007.
- [16] Pataius, Martynas, et al. "Optimal Combinations of Color Space Components for Detection of Blood Vessels in Eye Fundus Images." Elektronikairelektrotechnika 3.91 (2009): 53-56.
- [17] Patasius, Martynas, et al. "Ranking of color space components for detection of blood vessels in eye fundus images." 4th European Conference of the International Federation for Medical and Biological Engineering. Springer Berlin Heidelberg, 2009.
- [18] Goffredo, Michela, et al. "Quantitative color analysis for capillaroscopy image segmentation." Medical & biological engineering & computing 50.6 (2012): 567-574.
- [19] Sainthillier, JeanMarie, et al. "Skin capillary network recognition and analysis by means of neural algorithms." Skin Research and Technology 11.1 (2005): 9-16.
- [20] Murray, Andrea K., et al. "Preliminary Clinical Evaluation of Semiautomated Nailfold Capillaroscopy in the Assessment of Patients with Raynauds Phenomenon." Microcirculation 18.6 (2011): 440-447.
- [21] Kwasnicka, Halina, and Mariusz Paradowski. "Capillaroscopy image analysis as an automatic image annotation problem." Computer Information Systems and Industrial Management Applications, 2007. CISIM'07. 6th International Conference on. IEEE, 2007.
- [22] Wen, Chia-Hsien, et al. "A novel method for classification of high-resolution nailfold capillary microscopy images." Ubi-Media Computing, 2008 First IEEE International Conference on. IEEE, 2008.
- [23] Doshi, Niraj P., Gerald Schaefer, and ArcangeloMerla. "Nailfoldcapillaroscopy pattern recognition using texture analysis." Biomedical and Health Informatics (BHI), 2012 IEEE-EMBS International Conference on. IEEE, 2012.
- [24] Otsu, Nobuyuki. "A threshold selection method from gray-level histograms." Automatica 11.285-296 (1975): 23-27.
- [25] Pizer, Stephen M., et al. "Adaptive histogram equalization and its varia-

tions." Computer vision, graphics, and image processing 39.3 (1987): 355-368.

[26] Duda, Richard O., and Peter E. Hart. "Use of the Hough transformation to detect lines and curves in pictures." Communications of the ACM 15.1 (1972): 11-15.

[27] Matas, Jiri, Charles Galambos, and Josef Kittler. "Robust detection of lines using the progressive probabilistic Hough transform." Computer Vision and Image Understanding 78.1 (2000): 119-137.

[28] Dadal, N. and Triggs, B.: Histogram of Oriented Gradients for Human Detection, In Proceedings of IEEE Conference Computer Vision and Pattern Recognition, San Diego, USA, pages 886-893, June 2005

[29] Dadal, N. and Triggs, B.: Finding People in Images and Videos, PhD thesis, French National Institute for Research in Computer Science and Control (INRIA), July 2006

[30] Cortes, Corinna, and Vladimir Vapnik. "Support-vector networks." Machine learning 20.3 (1995): 273-297.

[31] Platt, John. "Sequential minimal optimization: A fast algorithm for training support vector machines." (1998).

[32] Lewis, J. P. "A short SVM (support vector machine) tutorial." CGIT Lab/IMSC, University Southern California (2004).

[33] Lange, Holger. "Automatic glare removal in reflectance imagery of the uterine cervix." Medical Imaging. International Society for Optics and Photonics, 2005.

[34] Blair, Kerry L. "Method and apparatus for reflective glare removal in digital photography useful in cervical cancer detection." U.S. Patent No. 6,088,612. 11 Jul. 2000.