ANALYSIS OF SEGMENTATION TECHNIQUES OF RETINAL BLOOD VESSELS

A DISSERTATION REPORT

SUBMITTED IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE AWARD OF THE DEGREE OF

MASTER OF TECHNOLOGY IN SIGNAL PROCESSING AND DIGITAL DESIGN

Submitted by:

PREITY

2K18/SPD/06

Under the supervision of Dr. N JAYANTHI



ELECTRONICS AND COMMUNICATION ENGINEERING

DELHI TECHNOLOGICAL UNIVERSITY

(Formerly Delhi College of Engineering) Bawana Road, Delhi-110042 JULY, 2020

ANALYSIS OF SEGMENTATION TECHNIQUES OF RETINAL BLOOD VESSELS

A MAJOR PROJECT REPORT

SUBMITTED IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE AWARD OF THE DEGREE OF

MASTERS OF TECHNOLOGY IN SIGNAL PROCESSING AND DIGITAL DESIGN

Submitted by:

PREITY

2K18/SPD/06

Under the supervision of Dr. N JAYANTHI



ELECTRONICS AND COMMUNICATION ENGINEERING DELHI TECHNOLOGICAL UNIVERSITY

(Formerly Delhi College of Engineering) Bawana Road, Delhi-110042

JULY, 2020

ECE DEPARTMENT DELHI TECHNOLOGICAL UNIVERSITY (Formerly Delhi College of Engineering) Bawana Road, Delhi-110042

CANDIDATES DECLARATION

I, **PREITY**, **2K18/SPD/06** student of M.Tech (SPD), hereby declare that the Dissertation titled "ANALYSIS OF SEGMENTATION TECHNIQUES OF RETINAL BLOOD VESSELS" which is submitted by me to the Department of Electronics and Communication, Delhi Technological University, Delhi in partial fulfilment of the requirement for the award of the degree of Master of Technology, is original and not copied from any source without proper citation. This work has not previously formed the basis for the award of any Degree, Diploma Associateship, Fellowship or other similar title or recognition.

Place: Delhi

Date: 11th July, 2020

PREITY

Preity"

ECE DEPARTMENT DELHI TECHNOLOGICAL UNIVERSITY (Formerly Delhi College of Engineering)

Bawana Road, Delhi-110042

CERTIFICATE

I hereby certify that the Dissertation titled "ANALYSIS OF SEGMENTATION TECHNIQUES OF RETINAL BLOOD VESSELS" which is submitted by PREITY, 2K18/SPD/06 [ECE Department], Delhi Technological University, Delhi in partial fulfilment of the requirement for the award of the degree of Master of Technology, is a record of the project work carried out by the student under my supervision. To the best of my knowledge this work has not been submitted in part or full for any Degree or Diploma to this University or elsewhere.

Place: Delhi Date: 11th July, 2020 N.J. y.

Dr. N JAYANTHI SUPERVISOR ASSISTANT PROFESSOR Electronics and Communication department

DELHI TECHNOLOGICAL UNIVERSITY (Formerly Delhi College of Engineering) Bawana Road, Delhi-110042

ABSTRACT

Retinal blood vessels are one of the most significant features in the fundus image of the eye, which plays a crucial role in the early screening of different eye diseases like glaucoma, , cataract, diabetic retinopathy and hypertensive retinopathy. Also in the area of biometric systems, blood vessel structure plays a vital role as retina scan is one of the finest and reliable methods. This dissertation report proposed different segmentation technique which accurately extracts retinal blood vessels. The proposed methods are completely based on Image processing technique. The first method is a segmentation technique of retinal blood vessel using Multi-Threshold and morphological operations while the second method is the retinal vessel extraction using principle curvature. This method efficiently segments the vessels and improves the performance parameters. The accuracy of 95.3% and 96.6% achieved by the first and second proposed methods respectively. Implementation part was done in MATLAB 2015a using a DRIVE database openly available online.

ACKNOWLEDGEMENT

I would like to express my gratitude towards each and every people who helped me throughout this project.

I am overwhelmed having Dr. N JAYANTHI ma'am as my Research Supervisor, her encouragement and guidance helped me a lot during the project. Furthermore I would also like to thank HOD sir, Prof. N S RAGHAVA, who provided all the necessary equipment and format of report. This project has expanded my horizons not only in signal processing but also in biomedical area and related services.

PREITY

CONTENTS

List of tables List of Figures		vii viii	
List of Symbols and Abbreviation		ix	
CHAPTER 1 INTRODUCTION		1	
CHAPTER 2 RELATED WORK		4	
2.1 Image processing based technique		5	
CHAPTER 3 RETINAL IMAGE PROCES	SING	7	
3.1 Fundus photography		8	
3.2 Retinal database available online		9	
3.2.1 Drive database	9		
3.2.2 STARE database	10		
3.2.3 Aria database	11		
3.2.4 CHASE database	11		
3.2.5 HRF database	11		
CHAPTER 4 ABNORMALITIES AND DIS	SEASES	12	
4.1 Retinal abnormalities		12	
4.2 Ocular disease		14	
4.2.1 Diabetic Retinopathy	14		
4.2.2 Glaucoma	16		
CHAPTER 5 RETINAL VESSEL EXTRA USING PCA AND ISODATA 5.1 Methodology	ACTION	17	
		10	
5.2 Image pre-processing		18	
5.3 Image enhancement		19 20	
5.4 image segmentation			
5.5 Image Post processing		21	
5.6 Result and Discussion		21 22	

CHAPTER 6 A SEGMENTATION TECHNIQUE OF RETINAL 23 BLOOD VESSEL USING MULTI-THRESHOLD AND MORPHOLOGICAL OPERATION

6.1	.1 Proposed methodology	
6.2	Preprocessing of input image	24
6.3	Segmentation of image	25
6.4	post processing of image	26
6.5	Result and Discussion	27
6.6	Simulation Result	28
CH	APTER 7 RETINAL VESSEL EXTRACTION USIN	30
PRI	INCIPLE CURVATURE	
7.1	Proposed Methodology	30
7.2	Generation of mask	33
7.3	Gaussian Filtering	33
7.4	Principle curvature and finding lambda function	33
7.5	Contrast enhancement and ISODATA	35
7.6	Result and Discussion	26
7.7	Simulation result	36
	Simulation result	37
CO	NCLUSION	40

REFERENCES 41

LIST OF TABLES

TABLE I	Categorisation of diabetic retinopathy
TABLE II	Performance metrices
TABLE III	Accuracy, sensitivity and specificity of proposed method
TABLE IV	Comparison analysis 1
TABLE V	Performance parameters of proposed method
TABLE VI	Comparison analysis 2

LIST OF FIGURES

Abnormalities in retina Fig. 1.2 Segmentation Technique Fig. 2.1 Fig. 2.2 Comparison Graph (Image processing Technique) Fig. 3.1 Eye anatomy Fig. 3.2 fundus camera and fundus image of retina Fig. 3.3 Image from DRIVE database Fig. 3.4 Image from STARE database Fig. 4.1 Abnormal retinal image Fig. 4.2 Image containing DR Fig. 4.3 Classification of DR Fig. 4.4 Normal anatomy of retina and Glaucoma Fig. 5.1 Flow chart of the algoritm Fig. 5.2 Simulation output using ISODATA Flow chart of proposed method (Multi-Threshold) Fig. 6.1 Fig. 6.2 Results of the proposed method (Multi-Threshold) Fig. 7.1 Flow chart of proposed method (Principle curvature) Fig. 7.2 Simulation result part 1 Fig. 7.3 Simulation result part 2

Normal image of left and right eye

Fig. 1.1

LIST OF SYMBOLS, ABBREVIATIONS

DR	Diabetic Retinopathy
MA	Microaneurysms
CLAHE	Contrast limiting adaptive histogram equalisation
MD	Diabetic Maculopathy
PCA	Principle component analysis
ROC	Region of curve

CHAPTER 1

INTRODUCTION

Eye is one of the most sensitive parts of the human body through which we visualize the whole world. As it is small in size it is considered as one of the sophisticated organ in the human body. The main parts of human eye are Retinal blood vessels, Optic nerve and Macula. Ophthalmoscope is used by ophthalmologists to look into the eye and analyse the internal structure of the eye. A circular oval white area having dimension 2x1.5mm across, is present in the centre of the retina is the Optic nerve. From the centre of the optic nerve the prominent blood vessels are radiated. Near the centre of the retina of the human eye and some other animal's eyes there is a circular shaped pigmented area known as Macular or Macula lutea. An oval-shaped reddish spot which is free of blood vessels which is present approximately 17 degrees or two and half disc diameters to the left of the disc is known as fovea and is present at the centre of macula. There are certain ocular diseases like diabetic retinopathy, glaucoma, hypertension retinopathy which affect the eye and can even cause blindness, partial or full vision loss. Nowadays most of the people of age 50 and above 50 are suffering from hypertension and diabetes due unhealthy living style, so the chances of occurrence of ocular disease has also increased. Blood vessels are one of the significant features of the retina through which different diseases can be diagnosed. Not only in the field of biomedical but also in the area of biometrics, retina scan is the highest form used in security purpose. In that case also blood vessels extraction plays an important role. In image processing there are various methods through which analysis of image, object representation and visualization can be performed. Many clinical researchers conclude that blood vessels are one of the prominent features in retinal images which can be helpful to diagnose the abnormalities. The manual extraction of blood vessels is tedious and also costly so numerous automated segmentation techniques are employed to make it fast and simpler. Fig 1.1 shows the fundus image of normal retina .In case of a retina with abnormalities in that case the vessels there are some deformities present in the features of retina and for blood vessels it may be leaky or fatty also their structure will be different from normal retina. The major abnormalities in retinal images are Exudates, leaky blood vessels, Hemorrhage and Microaneurysms as shown in Fig 1.2. When blood vessels present in the eyes become blocked or weak, leaky narrow then deformation occur and it will turn into abnormality. Clotting of blood occurs and in the back of eyes some abnormal blood vessels start growing due to some sickle cell diseases. These weak blood vessels bleed which is one of the major cause of abnormality and hence cause hemorrhage (Vitreous hemorrhage). Basically it is the loss of blood from vasculature. Deposition of extravasated plasma proteins, especially lipoproteins, are the exudates, often described as hard exudates. The yellow-white dots appeared within the retina representing the exudates. They may be seen as either individual spots or clusters near the optic disk. Also soft exudates are a kind of abnormality present in the retina. Microaneurysms appear as small red dots the ballooning of capillaries. They are saclike, out-pouching in the small vessels. Analysis of retinal vessels is a non-invasive method to examine the veins and small arteries in the retina. This allows us to conclude the information about morphology and the function of small vessels. So basically in this project we deal with the extraction technique of retinal blood vessels. In this project different segmentation methods based on image processing techniques like contrast enhancement, segmentation, binarisation, thresholding and at the end morphological operation which are easier and efficient are employed. So this approach will be more efficient and accurate and also quite simple. I have organise this research dissertation report into 6 different sections. Each of the sections deals with the specific content related to the project. I have already given the overview about the report. Chapter 2 depicts all about retinal image processing. The work which has been done previously is described in chapter3, chapter 4 contains detailed descriptions of the methodology employed in this research work that is our proposed method. Chapter 5 consists of results obtained from our proposed methodology followed by the discussion related to the results obtained, at the end conclusion of the method and some details about future work.

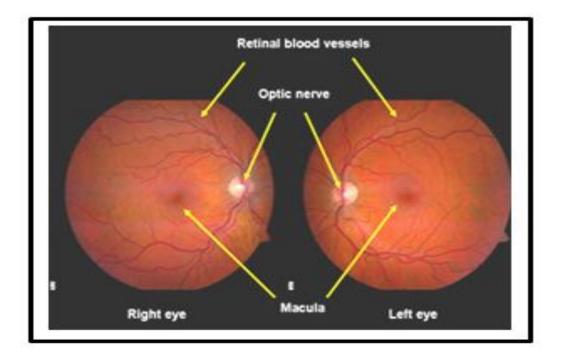


Fig 1.1 Normal image of left and right eye.

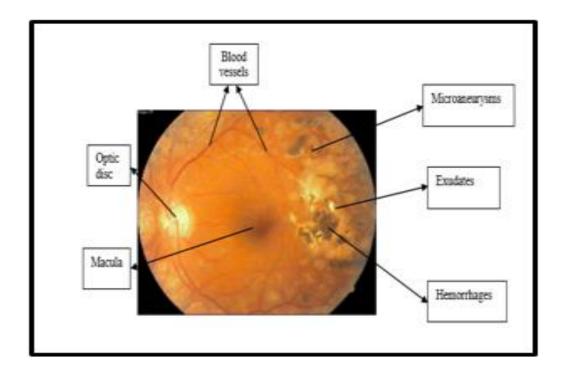


Fig 1.2 Retinal image with abnormalities.

CHAPTER 2

RELATED WORK

Many researchers have published their work in this area in recent years. Over the previous era study and analysis of retinal blood vessels became one of the basic mediums in the diagnosis of several ocular diseases. Different approaches for retinal blood extraction in fundus image were suggested which assist physicians to diagnose the disease and also in early detection of different ocular diseases. Different types of methods have been used by the researchers which incorporates basically image processing technique and machine learning based techniques.

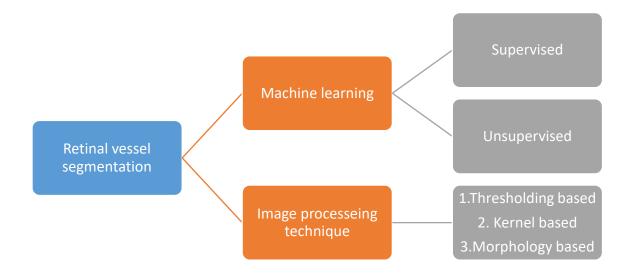


Fig 2.1 Segmentation Techniques.

2.1 IMAGE PROCESSING BASED TECHNIQUES

A vessel segmentation technique based on unsupervised fuzzy c mean clustering based thresholding in pathological digital fundus images proposed by Kande *et al.* [1] STARE and DRIVE are used for performance evaluation and the method is 93.85% accurate. ROC curves of this method were also obtained. To enhance the contrast of image matched filtering was used.

An image processing based method is proposed by Bantan *et al* [2] to auto-segment the blood vessels from the retinal image. Performance evaluation on HRF database with accuracy of 94%, specificity of 97% and sensitivity of 69%. The error calculation was done between the resulting image and ground truth image, mean squared error was calculated.

A thresholding based blood vessels extraction technique from retinal fundus images proposed by Dash *et al.* [3]. Two different databases namely STARE and DRIVE were used for implementation, the method was 95% accurate. Median filtering and mean c-threshold based method was used.

Another research work on retinal image enhancement techniques to segment the blood vessel, an algorithm was proposed by Bandara *et al.* [4]. SUACE algorithm were SUACE stands for speeded up adaptive contrast enhancement and improved form of Tyler Coye algorithm with hough line transform for blood vessel reconstruction. Performance Evaluation on DRIVE database, 94% of accuracy achieved by the author. An efficient method for the extraction of retinal blood vessels using morphological operations and ISODATA clustering technique proposed by Pilai *et al.* [5]. PCA was used for RGB to gray conversion and then ISODATA for clustering. Here ISODATA is a refinement of K-means clustering used for segmentation. One of the effective methods for segmentation.

For the screening of hypertensive retinopathy a method for color image segmentation of fundus blood vessels proposed by Latha M *et al.* [6]. Performance evaluation on data from ten patients suffered from hypertensive retinopathy. Here morphological operations were used.

Mapayi *et al.* [7] proposed an automated technique to segment the retinal vessels. A combination of difference image (DI) and fuzzy c means approaches are being used.

DRIVE database is used for performance evaluation, the method was 94.4% accurate and a high sensitivity rate of 73.02% was achieved.

NC Santosh *et al.* [10] proposed a segmentation method based on Optimised maximum principle curvature. Particle swarm Optimisation algorithm used for pre-processing of image and the maximum curvature is calculated. Hence the accuracy of 96% was achieved. The proposed algorithm implemented on DRIVE & STARE database.

There are some challenges that confronts the existing techniques for the extraction of blood vessels are presence of lesions in retinal image which can be misdetected as blood vessels, difficulties occurred in the extraction of thinner blood vessels. Since the contrast near the thinner vessels is quite low so it will be more difficult to segment the thinner vessels. Also the performance parameters evaluation as some authors did not do the comparison between the segmented images and ground truth images. Fig 2.1 shows the graphical analysis of these methods used by different authors.

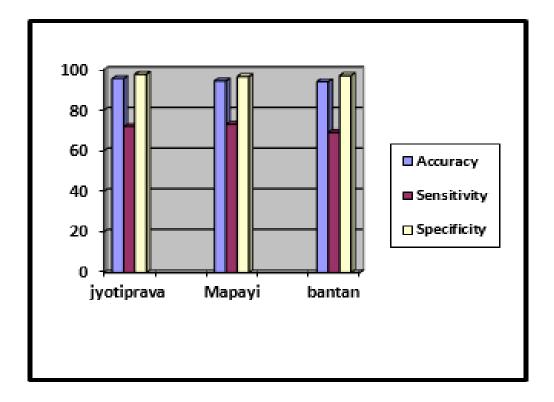


Fig 2.1 comparison graph (image processing technique)

CHAPTER 3

RETINAL IMAGE PROCESSING

Retina present at the back of human eye which is an important part of human vision. Human eye anatomy incorporates Retina, Lens Pupil, Blood vessels Cornea as shown in Fig3.1.

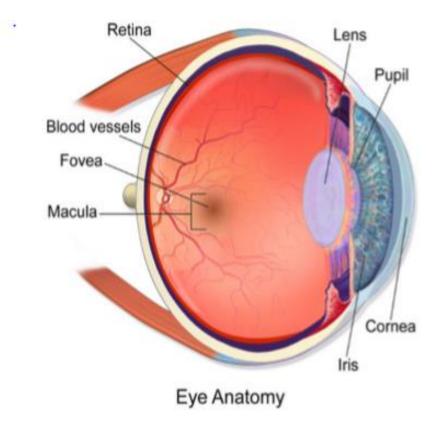


Fig 3.1 Eye Anatomy.

<u>3.1 FUNDUS PHOTOGRAPHY</u>

Retinal Imaging is typically done via an optical instrument called Fundus Camera. It is complex optical system can be viewed as a microscope of low power with a camera attached it which illuminates and image the retina.

It is well designed to capture the image of interior surface of eye, which incorporates the major parts like retina, optic disk, posterior pole, macula and blood vessels. In various research works some sort of documentation process for interior and posterior parts of retina, Fundus photography is used. Basically there are three modes in which fundus photography is done and the modes are Colour photography, Red free photography and Fluorescent angiogram.

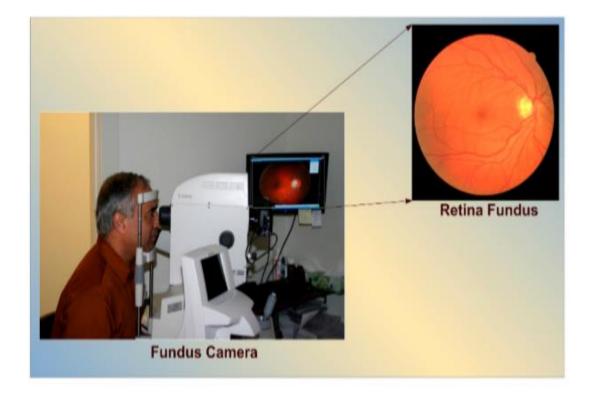


Fig 3.2 Fundus camera and fundus image of Retina.

3.2 RETINAL DATABASES AVAILABLE ONLINE

There are so many datasets publicly available. Many researchers used these databases to complete their research work successfully. DRIVE and STARE databases are used by numbers of researchers and most of the works published using these two.

3.2.1 DRIVE database

DRIVE is abbreviated as Digital Retinal Images for Vessel Extraction) is a publicly available online, it consist of 40 color fundus images. The photographs were obtained from a screening program of diabetic retinopathy held in the Netherlands. The screening population consisted of 453 subjects between 31 and 86 years of age. Images are in JPEG format. Out of 40 images, 7 contain abnormalities which is exudates, hemorrhages and pigment epithelium changes. Using a Canon CR5 non-mydriatic 3-CCD camera having field of view (FOV) of 45 degree the images were captured. The images being captured using 8 bits per color plane at 768×584 pixels. Fig 3.3 shows the sample image of DRIVE database.

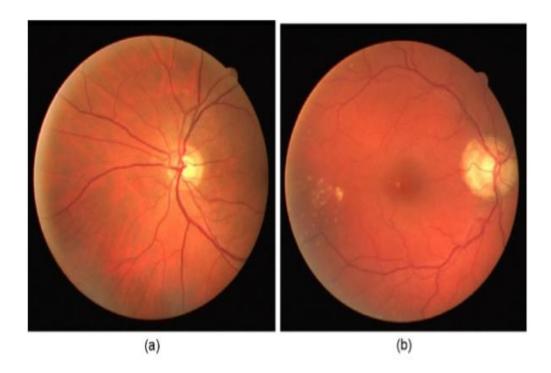


Fig 3.3 Images from DRIVE (a) normal image, (b) image contain pathology

3.2.2 STARE database

TheSTARE database is another database available online and it contains 20images for blood vessel segmentation out of these 10 contains pathology. TopConTRV-50 fundus camera is used to capture at field of view of 35 degree . Digitization of slides to 605×700 pixels, per color channel 8 bits are selected. 650×500pixels is the approx. diameter of FOV. Two observer manually segments all images present over there. Thefirstobserver segmented10.4% of pixels as vessel is segmented by first observer, second observer segmented14.9%.the second observer segment the thinner vessels more that is the reason behind segmenting more vessels than the first observer. Fig 3.4 represents the STARE images

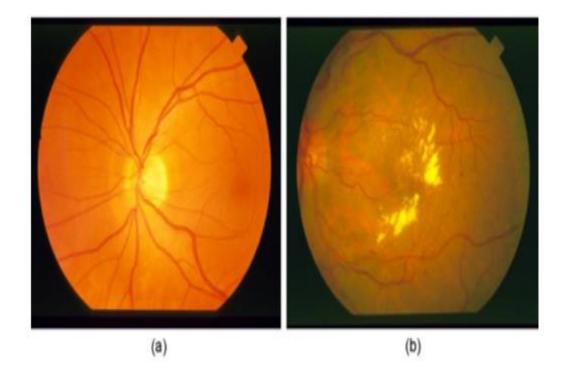


Fig 3.4 images from STARE (a) normal image, (b) image contain pathology

3.2.3 Aria database

ARIA stands for Automatic Retinal Image. Joint collaboration of the University of Liverpool and the Royal Liverpool University Hospital Trust, the UK in 2006 created this database. There are three image subsets are present namely .diabetes images (59), control group images (61), and age-related muscular degeneration images(92). Zeiss fundus camera FF450 used to capture the images at 500 FOV, 8 bit per colour plane, pixel resolution is 768*576.the storage format used in this database is TIFF. Two observers marked ground truth details of the blood vessel location, fovea location, and the optic disc.

3.2.4 CHASE database

CHASE is abbreviated as Child Health and Heart Studies in England. It includes images of different kind of diseases The first database entitled CHASE_DB1contains retinal images and its ground truth image. NM-200D fundus camera with 35° FOV, 1280 by 960pixels resolution is used to capture the images in TIF format. Under full-field illumination the images were captured. There are 28pictures altogether incorporated in the above database , these 28 images were gathered from 14 kids i.e images of left and right eyes of all kids. Two experts sectioned the ground truth image manually. All 28 retinal pictures are healthy and have no record of any type of diseases, however, they have good quality.

3.2.5 HRF database

HRF database is of high-resolution fundus images. Jan Odstrcilik created this database to study and analyse automatic segmentation algorithms and compare. There are 45 photographs in which 15 images are of normal people, 15 of patient having Gluacoma 15 of patient suffering from DR. This database is known for its high resolution it has 3504*2336 resolutions, which is comparatively higher. Binary gold standard images are available for each image, binary gold standard images are available and the experts of retinal image analysis manually segment these ground truth image. The images were captured by CanonCR-1 with FOV of 45°.

CHAPTER 4

ABNORMALITIES IN RETINA AND OCCULAR DISEASE

4.1 RETINAL ABNORMALITIES

Due to different cause abnormality occurs in the retina so there are direct and indirect causes. A normal retina incorporates retinal blood vessels and optic disk with proper structure. The Fig 4.1 shows the abnormalities occurred in the retina.

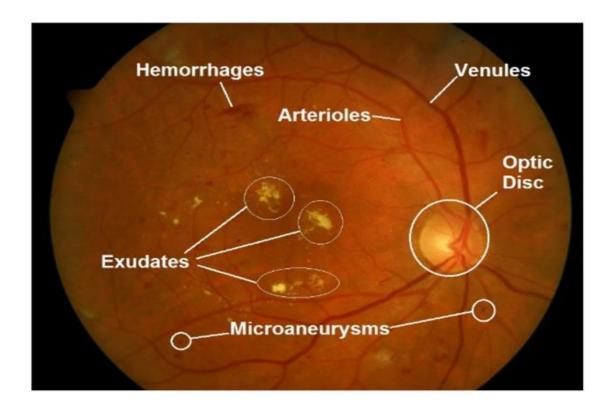


Fig 4.1 Abnormalities in retina.

All the abnormalities are somehow related to blood vessels itself. If we consider exudates or microaneurysms all are related to abnormalities due to presence of leaky blood vessels.

Leaky Blood Vessels:

Blood vessels are the prime part on human body as it is the component responsible for the flow of blood in the different parts. Similarly in human eye the arterioles and venules are the retinal vasculature responsible for the flow of blood. Due to some cardiovascular diseases the vessels present in eye got affected and becomes leaky blood vessels. These leaky vessels are responsible for the other abnormalities occurring in the retina. Also sometimes clotting of blood occurs in the vessel which restricts the proper flow of blood through the vessel which in turn becomes severe sometimes and causes vision loss also.

Exudates:

Deposition of extravasated plasma proteins, especially lipoproteins, are the exudates, often described as hard exudates. The yellow-white dots appeared within the retina representing the exudates. They may be seen as either individual spots or clusters near the optic disk. Also soft exudates are a kind of abnormality present in the retina.

Micro-aneurysm:

Micro-aneurysms appear as small red dots the ballooning of capillaries. They are saclike, out-pouching in the small vessels. Analysis of retinal vessels is a non-invasive method to examine the veins and small arteries in the retina.

Haemorrhage:

Clotting of blood occurs and in the back of eyes some abnormal blood vessels start growing due to some sickle cell diseases. These blood vessels are not strong and can bleed. These abnormal blood vessels cause haemorrhage (Vitreous haemorrhage).

4.2 OCULAR DISEASE

There are different ocular diseases related to power of eye but in this research we are looking onto those disease which affect the parts of retina and causes partial and full blindness. Those ocular disease occurred due to some other diseases like Diabetes and hypertension. These diseases affects eyes too in human body and creates severe disease like Diabetic Retinopathy, Hypertensive Retinopathy and Glaucoma etc.

4.2.1 Diabetic Retinopathy

Diabetic Retinopathy is an eye disease which is mainly occurred due to Diabetes mellitus. Maximum of population are suffering from diabetes and those who are suffering from long time they are dealing with some other abnormalities which occurs due the after effects of diabetes. As discussed earlier blood vessels are mainly responsible for the occurrence of any abnormalities in the retina. Due to weak and leaky blood vessels present in retina, the fluid discharged in the eye creates some abnormal features in retina like spots , patches which is turn creates hard and soft exudates, microaneurysms(MA), haemorrhage. This damage occurs due to diabetes and this cause vision impairment. The rate of occurrence of DR is increasing day by day and the risk of blindness is high.

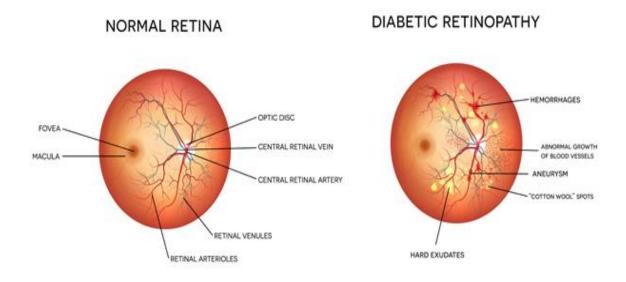


Fig 4.2 Normal image and image containing DR

The fig 4.2 represents the image which contain DR features. DR is classified into NDPR and PDR. NDPR stands for Non proliferated DR in which the condition of DR is from mild to advance stage and PDR stands for proliferated DR in which the condition is severe. The Fig 4.3 shown below represents the normal retinal image, image having NDPR and image having PDR.

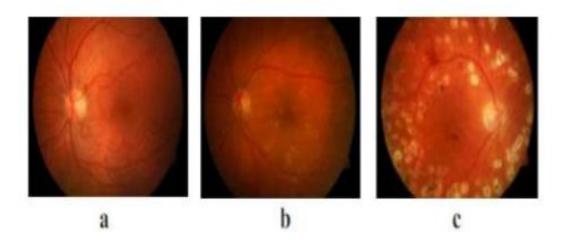


Fig 4.3 a. normal image b. NDPR c. PDR

Further the DR is characterised in 3 parts that are normal, mild and severe. These categorisation of diabetic retinopathy is based of abnormal features present in fundus image. In the detection of DR number of abnormal features like microaneurysms(MA) is counted and based on that the level of disease is decided. The TABLE I shown below the level of diabetic retinopathy based on number of microaneurysms present in the image.

TABLE I. CATEGORISATION OF DR

1	MA<5	Normal
2	5 <ma<15< td=""><td>Mild</td></ma<15<>	Mild
3	MA>15	Severe

4.2.2 Glaucoma

Glaucoma can occur in any age but mostly common in adults over the age of 60. Though it has many cause of occurrence but the main cause behind this disease is excessive pressure in the eye. Due to occurrence of excessive pressure inside the eye, the optic nerve of eye got damaged hence it leads to blur sightedness, eye pain, severe headache.

Usually Glaucoma cannot be noticed at the early age as the symptoms will occur when the condition get advanced. By measuring the pressure and examining the features of retina Glaucoma can be detected and the treatment of this disease will continue for life time once one got this disease. So early detection can prevent a person from vision loss.

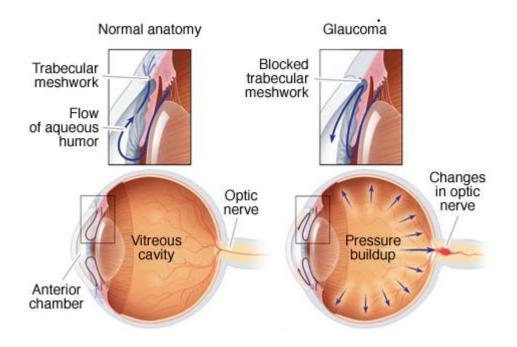


Fig 4.4 Normal anatomy of retina & Glaucoma.

The above Fig 4.4 represents the structural anatomy of a normal retina and the image of glaucoma. The trabecular meshwork of retinal is blocked due to glaucoma and due to excessive high pressure optic nerve changes. So the pressure inside the eye should be balanced.

CHAPTER 5

<u>Retinal vessel extraction using PCA, ISODATA</u> <u>&Morphological operations</u>

5.1 METHODOLOGY

Study and analysis of retinal vessel extraction techniques are used by various medical researchers as it is helpful in the screening of different diseases. There are some ocular disease like diabetic retinopathy, hypertensive retinopathy, Glaucoma etc are responsible for partial and complete blindness in human. Blood vessel extraction will be the basic step which is required in number of screening method done by oculist.

In this technique some image processing tools are used to segment the retinal blood vessels. This system is divided into three sections namely image pre-processing, image enhancement, image segmentation and at last image post processing.

The above system is performed on DRIVE database which is publicly available online and the simulation tool is MATLAB 2015a.

In this system the main focus is on PCA and ISODATA .these two makes the system robust and efficient in segmentation. Here PCA is used for RGB to gray conversion and ISODATA refers to Iterative Self-Organizing Data Analysis Technique which is an iterative algorithm used for selecting the threshold for discrimination of object-background. This has been proposed by Ridler and Calvard This is an effecting algorithm used for thresholding which requires some step which will be discussed in further in detail. Localisation of image is done by CLAHE. It is also an important step in segmentation process. At last morphological operations are employed for post processing of image. The fig 5.1 shows the flow chart of the algorithm which is to be employed step by step.

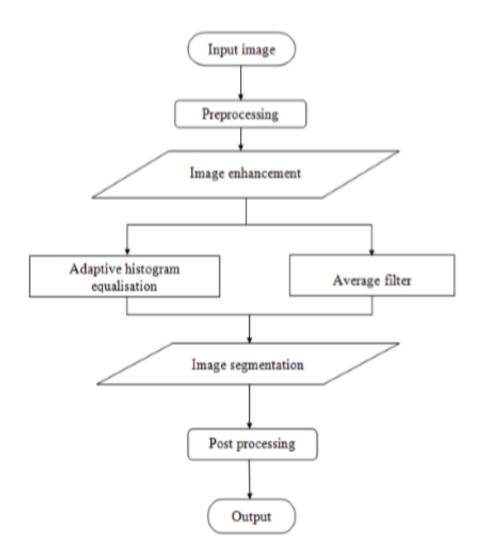


Fig 5.1 flow chart of the algorithm.

5.2 IMAGE PRE-PROCESSING

The quality of image is enhanced by performing image pre-processing technique so that the calculation and implementation will become easier and productive. Resizing, doubling, unsharp masking, contrast enhancement and calculation of gradient magnitude of the image are some renowned steps which is to be done in image preprocessing. The visual appearance of images can be improved by employing image preprocessing. An optical inspection can become significantly more reliable after image preprocessing. Some common pre-processing techniques used here are: resizing, doubling and type change. Conversion of RGB to GRAY is done by using PCA. There are three primary color intensities called red, green and blue other than that all are the combination of thses only that is the reason we represent image in RGB format. So conversion of RGB to gray is requited as the image is characterized by integers and its luminance is in the range of 0 to 255 in the language of computer. An RGB image is represented by M*N*3 array of color pixel and a gray image is represented by M*N array and to signify image intensities, the values are scaled in MATLAB. To convert RGB image to gray image, there is a rgb2gray() function which is used. We require 8(bits)*3(colors)=24bits to preserve the RGB image to gray the sum of weighted RGB color components are taken and so at the location (i, j) the RGB values of each image pixel is converted to gray scale values and the same is allocation is done to the corresponding location (i, j) in new matrix and a single value pixel for every location in the output image is obtained.

5.3 IMAGE ENHANCEMENT

Adaptive Histogram Equalization for contrast improvement. It slightly differs from histogram equalization as in AHE different histograms are computed on different sections of image. AHE over amplifies the noise hence a better variant of AHE is used to limit amplification so the problem of noise is being reduced by using this approach and that variant is Contrast Limited Adaptive Histogram Equalization which is termed as CLAHE. After this equalization process Average filter is used. Average filter is basically a mean filter in which the mean of all pixel values is replaced by a centre value and is used to reduce the noise. Equal weight is supplied by this filter and it is linear in nature. This is a special type of filter used for averaging. It smoothen the picture but it is not employed for giant noise. The default size is [3, 3] but here in this project we used [9,9].

5.4 IMAGE SEGMENTATION

The method of partitioning a digital image into multiple segments is termed as image segmentation. Segmentation is done to simplify or change the alighnment of an image into a meaningful representation and make it easier to understand. Its main purpose is to locate objects and boundaries in the images. More precisely, assignment of label to each and every pixel which share some unique characteristics is the process of The entire image is collectively covered by the output of image segmentation, or a set of contours is being extracted from the image. In a region each and every pixels are somehow similar with respect to some characteristic or such as color, intensity, or texture. And the regions adjacent to that are significantly different with respect to the same characteristic.

Here image segmentation is done by thresholding method and ISODATA thresholding is used here ISODATA refers to Iterative Self-Organizing Data Analysis Technique which is an iterative algorithm used for selecting the threshold for discrimination of objectbackground.

ISODATA ALGORITHM:

It is iterative algorithm used for thresholding. This is used for segmentation by separating the foreground and background image.

The algorithm is as follows:

It is an algorithm to classify a set of vectors $x = x_1^{+} + x_2^{+} + \dots + x_n^{+}$ into distinct classes.

1. For the mean value $c = \mu_1 + \mu_2 + \dots + \mu_n$ select some initial values.

Loop 2. The class which has closest mean, classify the samples by assigning them to that

3. Again compute the mean in each class.

4. If any value of mean has changed go to Loop, else stop.

These are the 4 basic steps needed to compute isodata threshold.

Global threshold is computed by ISODATA thresholding. The intensity image is converted into binary image or simply we can say that binarisation is done. There is a fixed range of (0, 1) in which the Level of intensity lies. Overall it is an efficient way of thresholding based segmentation.

5.5 IMAGE POST PROCESSING

There may be numerous imperfections in binary images. Performing some morphological operations to the image is the meaning of postprocessing. A structuring element to an input image is applied and then an output image of the same size is created and this is the main process of morphological operation. The value of each pixel in the output image is based on a comparison of the corresponding pixel in the input image with its neighbours in a morphological operation. By selecting the size and shape of the neighbourhood, one can construct a morphological operation that is sensitive to specific shapes in the input image. Clear boundaries of the segmented output image is defined by these morphological operations.

5.6 RESULTS AND DISCUSSION

In this method a novel method of converting RGB image to gray image is used which is done by using PCA. The implementation of this method was done on DRIVE dataset and the MATLAB 2015a is used for simulation. Here CLAHE has been used so the contrast of the output image obtained is also excellent. Final segmentation is based on ISODATA thresholding which is an efficient method to separate foreground and background image and by iteration it used to find the threshold. Overall it is simple and useful technique used for segmentation of blood vessel from fundus image.

5.7 SIMULATION RESULT

The Fig 5.2 shown below is the output obtained by implementing the method in MATLAB In the figure shown below a. represents the input image from the database used b. represents the image after converting it into gray and after contrast enhancement c. represents ISODATA thresholding and at last d. represents the final segmented vessels in output image.

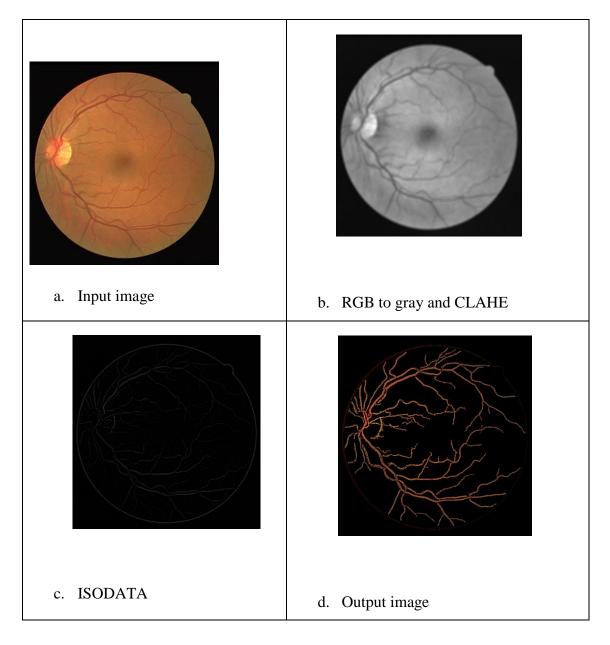


Fig 5.2 Simulation Output

<u>CHAPTER 6</u>

A Segmentation technique of blood vessels in retinal image using Multi-Threshold and morphological operations.

6.1 PROPOSED METHODOLOGY

Many research works have been done in this field and to enhance the existing method numbers or research work is still going as this area has a great aspect in biomedical science. Some of ocular diseases like DR, Glaucoma are evolving dangerously so the automated method of extraction of retinal vessels could help oculist up-to great extent. There are some challenges that confronts the existing methods for the extraction of blood vessels and those challenges are presence of lesions in retina which can be falsely selected as blood vessels, it is quite difficult to extract thinner blood vessels also. So these issues are being solved in this proposed method in which contrast limited adaptive histogram equalisation is used to enhance the contrast in a better way. Also using multi-threshold techniques the vessels are segmented so the results we achieved are better. We have calculated all the basic measuring parameters and so the comparison of results are done in our method. The details about the parameters were not mentioned in some research work so the analysis of results cannot be done properly. Hence the proposed method will be an efficient method as the exact values obtained by the technique employed in this work is clearly mentioned.

In this work, the database which is employed here is a well-known dataset used for retinal research work that is DRIVE database. All the basic parameters which are essential to analyse the method is calculated in this work. This method incorporates different steps and these steps are shown flow chart given below which clearly describes the approach which is being used here to segment the blood vessels from fundus image. Basically there are 3 partitions of the whole system that includes the pre-processing of image,

segmentation of image and post processing of the image. The Fig 5.1 shown below depicts the flow chart of proposed system. With the help of this flow chart algorithm can be understood easily

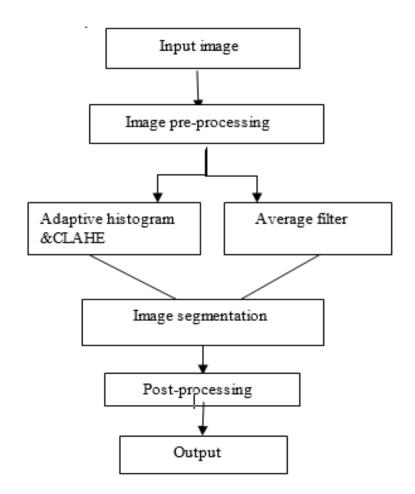


Fig 6.1 Flow chart of proposed system

6.2 PRE-PROCESSING OF INPUT IMAGE

Pre-processing of input image has to be done in first step so that the results would be better. Resizing of the input image is done and then conversion of RGB to gray is done after that histogram is performed. In this proposed method Adaptive Histogram Equalization is used for contrast improvement. It slightly differs from histogram equalization as in AHE different histograms are computed on different sections of image. AHE over amplifies the noise so to limit amplification a better variant of AHE is used hence the problem of noise is being reduced by using this approach and that variant is Contrast Limited Adaptive Histogram Equalization which is termed as CLAHE. After this equalization process Average filter is used. This is a special type of filter used for averaging. It smoothen and blur the picture, the disadvantage is that it is not employed for giant noise. Here in the proposed method we have used [9, 9] but the default size is [3, 3].

6.3 SEGMENTATION OF IMAGE

It is a technique which is used for to partitioning of the image or to represent the image in a simpler form which is easier to analyse. For edge detection, curve detection or set of contour extraction segmentation of image is done. Here in this proposed method for segmentation of retinal blood vessels Multi-threshold technique is employed which is a variant of Otsu's method. Otsu's method is itself a thresholding technique which widely used in image processing. After that conversion of segmented image into binary is done and the process is called binarization. So overall this method is based on thresholding and simple to implement

MULTI-THRESHOLD:

It is a method used for multi-level image thresholding which is based on Otsu's criteria. For an image a single threshold is computed by Otsu's method. Using this conversion of an image into two level image can be done.

If we apply multithresh on an array A which can be of any dimension then it computes thresholds which is based on overall histogram of the entire array.

There are 3 color planes in an RGB image the thresholds are calculated for the combined data of all these 3 planes.

OTSU'S METHOD:

For automatic image thresholding OTSU'S method is used. It is used to separate the image pixels into foreground and background.

To calculate this threshold minimisation of intra-class intensity variance or maximisation of inter-class intensity variance is evaluated and in extension to that threshold is obtained.

6.4 POSTPROCESSING OF IMAGE

The last step of the proposed method is postprocessing of image which is to be done for the final output. Basically morphological operations are performed to improve imperfection. Also we have compared the pixels with respect to input image to remove the imperfection occurred. In general morphological operations are used on binary images so the conversion of image to binary is done in segmentation section only. Various morphological operations are present but the major morphological operators are Dilation, Erosion, Closing Opening etc. To the boundary of an image, the addition of pixels is done by using Dilation whereas for the removal of pixels from the boundary of an image is done by using Erosion. Here in this method to remove the isolated pixels, clean operation is employed as morphological operation. By employing this the output image becomes noise free also.

6.5 RESULTS AND DISCUSSION

The proposed method is tested on the sample of images present in the DRIVE database. The performance parameters calculated by comparing the segmented image which we have obtained with the ground truth image. The simulation result obtained by the proposed method is shown in section 6.6. On the basis of pixel classification performance is evaluated. Outcome is either pixel classified as a vessel or the surrounding. In general there are 4 cases: two misclassification and two classification namely True Positive (TP), False Positive (FP), False Negative (FN) and true negative (TN). On the basis of these classified and misclassified vessels, we have calculated the Accuracy, Specificity and Sensitivity. The overall pixels correctly classified in the field of view of an image is abbreviated as FOV. TABLE II describes the mathematical formulas of performance parameters which have been used in the proposed method.

Measures	Descriptions
Accuracy(ACU)	TP+TN/ FOV
Sensitivity(SN)	TP/(FN+TP)
Specificity(SP)	TN/(FP+TN)

TABLE II. PERFORMANCE METRICS

TABLE III.ACCURACY, SENSITIVITY, SPECIFICITY OF PROPOSED METHOD

Images	Accuracy	Sensitivity	Specificity
Img1	95.75	72.02	98.57
Img2	95.36	70.16	99.45
Img3	94.70	71.18	97.66
Img4	95.11	66.38	99.50
Img5	95.28	67.91	99.38

Table III depicts the experimental results obtained by proposed algorithm. From the database sample of images taken and the algorithm is applied hence calculated the different parameters. Table IV depicts the comparison analysis of different author's approaches and proposed method. Firstly the true classified pixels and misclassified pixels are calculated and then accuracy, sensitivity and specificity is calculated for different samples of images. The average accuracy, specificity and sensitivity of 95.3%, 98.91%, 69.53% respectively is achieved. Proposed method attains high accuracy. Specificity is also improved. A simple image processing approach is employed in a better way.

Authors	Accuracy	Sensitivity	Specificity
Kande[1]	93.85%	-	-
Dash[3]	95%	70.4%	97.10%
Bantan[2]	94.70	69%	97%
Bandara[4]	94%	-	-
Proposed method 1	95.3%	69.53%	98.91%

TABLE IV.COMPARISON ANALYSIS 1

Fig 6 shown in section 6.6 is the result obtained by the proposed algorithm. The detection of ocular diseases is somehow based on vessel extraction. Even the thinner vessels are also extracted from the retinal images using this method. Also the parameters are obtained by doing comparison between ground truth image and the segmented image. Furthermore a better result with an average accuracy of 95.3% and specificity of 98.915 is achieved. Hence the method is comparatively better and simpler than existing methods.

6.6 SIMULATION RESULT

The figure shown below depicts the result obtained by proposed algorithm. The first column represents sample of input images, second column represents ground truth image and last column is the final output i.e segmented images obtained by applying the proposed algorithm,

Input image	Ground truth image	Segmented image
		A A A A A A A A A A A A A A A A A A A
	A A A A A A A A A A A A A A A A A A A	A A A A A A A A A A A A A A A A A A A
	A CCC	No. of the second secon
	A.	No.
	A A A A A A A A A A A A A A A A A A A	A CARE

Fig 6 Results of the proposed method

CHAPTER 7

RETINAL BLOOD VESSEL EXTRACTION USING MAXIMUM PRINCIPLE CURVATURE

7.1 PROPOSED METHODOLOGY

The rate of vision impairment has been increased drastically and the reason behind this is not only the ocular diseases but also due to some cardiovascular diseases. Eye is that part of human body which helps in visualisation of everything and vision loss will bring various types of other abnormalities physically and mentally also.

If we see the statistics of people suffering from ocular disease due to some other cardiovascular disease we get to know that above 80% of the people suffering from diabetes and hypertension. Both the diseases evolving drastically in human beings which becomes the cause of Diabetic Retinopathy and Hypertensive Retinopathy and these ocular abnormalities leads to partial and full vision loss in human being which is one of the important matter of concern.

Early detection of these kind of eye disease can prevent one from losing his vision also reduce the chances of getting the condition severe. Ophthalmologist uses computer aided methods to diagnose these problems. So for the screening of these eye disease, retinal features extraction plays a vital role and retinal blood vessels are one of the important features which is responsible for the further diagnosing process. By segmenting the vessels it can be used at initial stage of diagnosis. The retinal vessels can also be abnormal feature of retinal as there may be clotting in the vessel or any leaky blood vessel might be present. Hence retinal vessels extraction is a compulsory step that should be taken in diagnosis of various ocular disease like DR and MD. This novel proposed method is based on principle curvature technique. There are many different research works already done in this area using different image processing methods also some of authors presented their

work using principle curvature also but still the results they obtained were not up to the mark.

The proposed method is a new technique to extract blood vessels from the fundus image. It is completely based on image processing technique which is simple to implement. The implementation of proposed method is done in MATLAB 2015a on DRIVE database which is publicly available online.

There are basic 7 steps required to implement this methodology and the steps are :

Step 1: Read the input image from the database

Step 2: Generate the mask using morphological operation

Step 3: Apply Gaussian filtering on the image for smoothing and denoising.

Step 4: Compute Hessian matrix for each pixels which will compute second order derivative.

Step 5: Find eigen values and eigen vector to find the lambda function and principle curvature.

Step 6: Contrast enhancement is done by using adaptive histogram equalisation and ISODATA thresholding is obtained

Step 7: Final segmented image is obtained by removing falsely segmented image using morphological operation.

The algorithm follows the above 7 steps to compute the results. It is an easy image processing based method which will help oculist in screening of various diseases in an efficient way. Fig 7.1 shown below represents the flow chart of proposed method, through this flow chart the algorithm applied in the proposed method can be well understood. Also all steps are included.

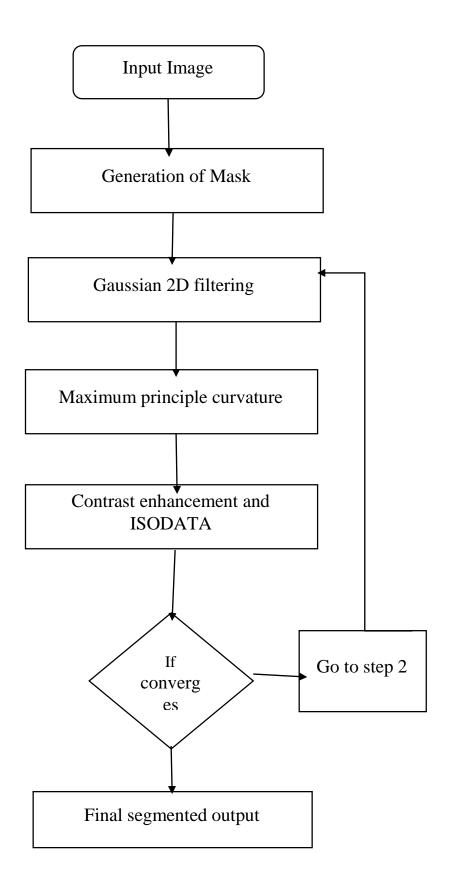


Fig 7.1 flow chart

7.2 GENERATION OF MASK

Mask is generated of the input image we have taken. To convert the image into binary image and creates diamond structured element and the size of that element is taken 20. Morphological operation is employed for this. On the basis of dimension of structuring element the neighbourhood pixels were identified. Basically there are mainly four types of operations are used which is Opening, Closing, Dilation and Erosion. Here in this method we have used Erosion to erode the mask.

The Erosion is employed to shrink the image with respect to the structuring element $A \ominus B$ represents erosion of image A to the structuring element B which has origin at (x, y). And the new pixel value obtained by the equation given below.

$$g(x, y) = \begin{cases} x & B \text{ hits } A \\ y & else \end{cases}$$
....(i)

So in this way the mask is generated by applying erosion operation on the image.

7.3 GAUSSIAN FILTERING

In this proposed method we have used Gaussian filter. As denoising is one of the important step which is to be done and here we have used Gaussian filter as it is more efficient in comparison to others. We are familiar with Gaussian expression as given below

$$F = \frac{1}{\sqrt{2\pi\sigma}} e^{-x^2/2\sigma^2}$$

Where σ stands for Standard Variance.

The value of standard deviation is responsible for smoothening of image. If the value of standard deviation is kept large then it require large convolution kernel for accurate representation. It is a linear filter and has an advantage over median filter is that it removes noise while keeping the edge sharp.

In this work we have taken the value of standard deviation is 1.45 also it is known that convolution of Gaussian to Gaussian is always a Gaussian function so, it is an efficient filtering method.

7.4 PRINCIPLE CURVATURE AND FINDING LAMBDA FUNCTION

In the proposed method principle curvature is used fetch the blood vessels which is one of the significant features in retinal fundus image. To calculate the principle curvatures and direction Hessian matrix is needed.

HESSIAN MATRIX:

Hessian matrix is basically used for partial derivatives of second order. It is a symmetric matrix of second order partial derivatives of scalar field. The differential geometry for the analysis of image and that represents the derivatives of image in which gradient is termed as first derivative.

Local curvature of a function of different variable is described by Hessian Matrix and it is represented as follows

$$\mathbf{H} = \begin{bmatrix} Ixx & Ixy\\ Iyx & Iyy \end{bmatrix}$$

Hessian matrix is a square matrix also a symmetric matrix therefore I x y = I y x. Now from this matrix eigen values and their corresponding eigen vector will be calculated. This eigen values are represented by Lambda function [λ]. The maximum value of lambda will decide the maximum principle curvature. The principle orientation is given by the eigen values. From this hessian matrix two eigen values are calculated and these are λ_1 and λ_2 . Where $\lambda_1 \leq \lambda_2$, convexity is measured by λ_1 and concavity is measured by λ_2 . Basically in any surface principle curvature and principle direction is obtained by the eigen values and eigen vector of Hessian matrix. So the eigen values is calculated and then corresponding eigen vector is calculated.

 λ_2 has the maximum value corresponds to the maximum principle curvature through which vessel segmented in the direction of eigen vector of that eigen value. Some important facts related to image topology is obtained from the spatial derivatives of the image's intensity. The variation in the in the intensity of the blood vessel corresponding to the neighbouring pixels is described by the principle curvature. In this work the maximum principle curvature is taken as the prime part for the extraction of retinal vessel from the fundus image.

In an image the highest and lowest curve bending of a surface at each point is measured by the principle curvature. After the calculation of eigen values and eigen vectors at each pixels is completed then weights are assigned to the pixels of the image as Blood Vessels also the condition of $|\lambda 2.| > 1$ is to be satisfied. In the direction of eigen vector the region is growing and this weights assigned pixels if satisfy the condition as mentioned above then it states that pixel belongs to the Vessel region otherwise the pixel is considered to be non-vessel part of the image. By doing multiplication of a rescaled mask with the rescaled maximum principal curvatures of the Hessian the region of growing of blood vessels is obtained. This mask takes two parameters namely (1) the binary form of input image and (2) diamond structuring element that takes into consideration only those pixels which will reach to the radius given as parameter and proceeds accordingly by convoluting to the maximum principal curvatures of the Hessian. By calculating maximum principal curvatures function leading to effective extraction of vessels vasculature from the given input image

Hence in this way maximum principal curvature technique is used in this proposed method for the segmentation of the retinal vessel.

7.5 CONTRAST ENHANCEMENT AND ISODATA

After finding the lambda function i.e principle curvature of the image the image is gone through the contrast enhancement and thresholding process. For contrast enhancement adaptive histogram equalisation is used. It slightly differs from histogram equalization as in AHE different histograms are computed on different sections of image. For better version of output contrast enhancement is a crucial step. After contrast enhancement isodata thresholding technique is employed. It is an iterative method use to find threshold by separating the background and foreground of an image. The detailed algorithm of isodata thresholding is already mentioned in chapter 5 and the working is already explained. After this contrast enhancement and thresholding step final filtering of image and processed output data is obtained.

The final output data is obtained after final filtering and applying some morphological operations. Here in this proposed method morphological opening is employed and then final segmented image is obtained.

7.6 RESULTS AND DISCUSSION

The proposed method implemented using DRIVE database which consist of 40 images. The proposed method is an efficient method to extract the retinal vessel using principle curvature. The performance parameter calculated in this method are Accuracy, Sensitivity and Specificity. An excellent result is obtained by this method. An average accuracy of 96.6%, 75.43 % of Sensitivity and 98.9 % of specificity is achieved by the proposed method.

 $ACCURACY = \frac{TP + TN}{TP + TN + FP + FN}$ $SENSITIVITY = \frac{TP}{TP + FN}$ $SPECIFICITY = \frac{TN}{TN + FP}$

TABLE IV represents the experimental value obtained by the proposed method on different images.

Images	Accuracy	Sensitivity	Specificity
Image 1	0.96285	0.75856	0.98792
Image 2	0.962	0.75493	0.98927
Image 3	0.96304	0.76253	0.98622
Image 4	0.96672	0.7547	0.99122
Image 5	0.9677	0.74128	0.99314

TABLE V.PERFORMANCE PARAMETERS OF PROPOSED METHOD

If we compare or results with the existing method related to principle curvature we conclude that the results obtained from the proposed method is better in terms or performance measure. Also the complexity of the method is comparatively low so this method can be easily used by the oculist for the automated diagnosis of different retinal diseases.

AUTHORS ACCURACY SENSITIVITY SPECIFICITY Dash[3] 95% 70.4% 97.10% 97% Bantan^[2] 94.70% 69% Roychaudhary[10] 75.2% 95.2% 98.3% Proposed method 96.7 % 75.4% 98.9%

TABLE VI. COMPARISON ANALYSIS II

TABLE VI represent the comparison between the existing methods and the proposed method.

7.7 SIMULATION RESULTS

PART 1:

Fig 7.4 shown below depicts half portion the simulation results by the proposed method. In this figure a. represents raw input image which from DRIVE database, b. is the real ground truth image which is manually generated by the observer, c. is the mask generated by using morphological operation and d. is the image obtained by applying Gaussian filter.

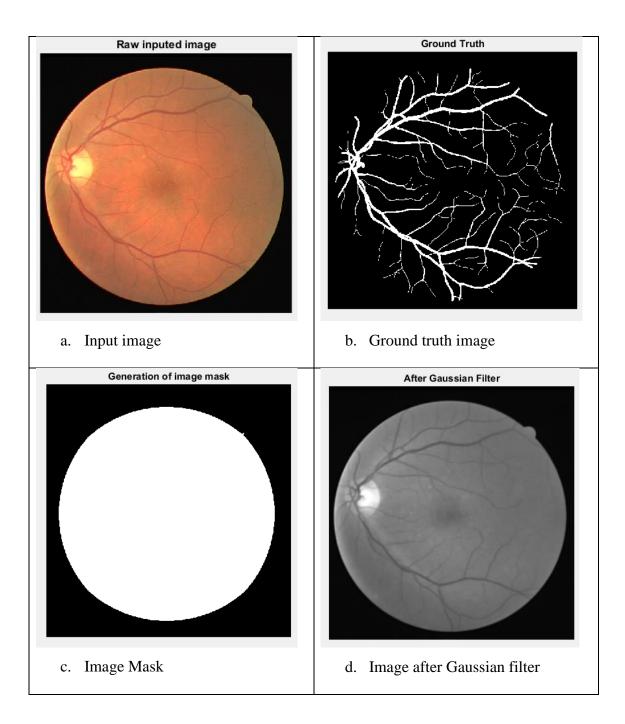


Fig 7.2 Simulation results part 1.

PART 2:

The Fig 7.5 shown below depicts the next part simulation of simulation result. In this fig e. represents the principle curvature f. represents the enhanced image g. is the image in

which small segments are filtered out and finally h. represents the output image which is the processed image.

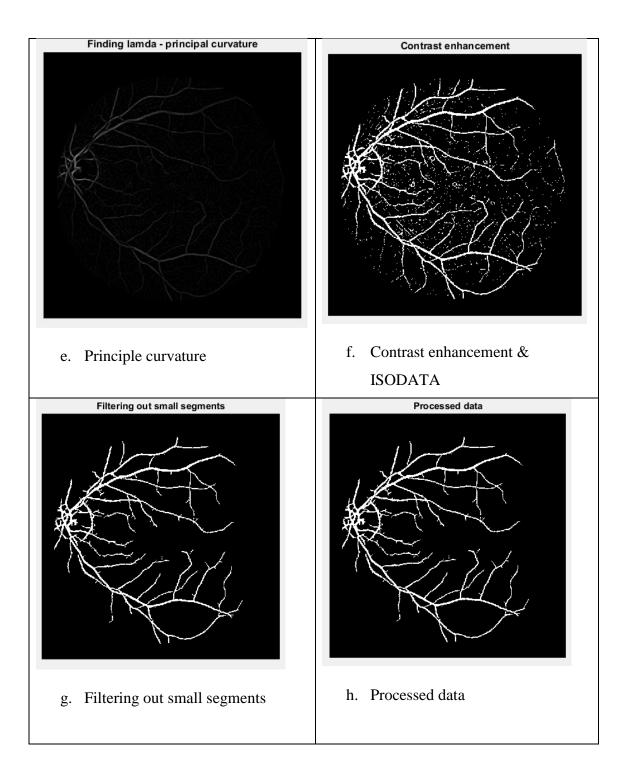


Fig 7.3 Simulation results Part 2.

CONCLUSION AND FUTURE WORK

Around 80% of the people having a history of diabetes and hypertension for the past 20 years or more are more likely to suffer from ocular diseases like diabetic retinopathy, glaucoma and hypertensive retinopathy. Hence the detection of retinal diseases are somehow based on vessel extraction. In this dissertation a segmentation technique of retinal blood vessel using multi-threshold and morphological operation is proposed and efficiently extracts the even the thinner vessels from the retinal images. The parameters are obtained by doing comparison between ground truth image and the segmented image. By this method accuracy of 95.3 % is achieved. Another method of retinal vessel extraction is proposed which is based on principle curvature. The advantage with this method is that the accuracy and specificity obtained by this method. If we compare the results of existing method and the proposed methods it is clearly seen that the proposed methods are simple and efficient.

So the proposed methods in this dissertation are truly based on image processing technique which is quite simple to implement with effective results. If we talk about the future aspects of this topic we can conclude that apart from image processing techniques, machine learning based approach can be a better option as machine learning is diverse topics and classification can be done using this.

REFERENCES

[1] Kande, Giri Babu, P. Venkata Subbaiah, and T. Satya Savithri. "Unsupervised fuzzy based vessel segmentation in pathological digital fundus images." *Journal of medical systems* 34.5 (2010): 849-858.

[2] M. T. Bantan, "Auto-segmentation of retinal blood vessels using image processing," 2016 4th Saudi International Conference on Information Technology (Big Data Analysis) (KACSTIT), Riyadh, 2016, pp. 1-6.

[3] Dash, Jyotiprava, and Nilamani Bhoi. "A thresholding based technique to extract retinal blood vessels from fundus images." *Future Computing* and Informatics Journal 2.2 (2017): 103-109.

[4] A. M. R. R. Bandara and P. W. G. R. M. P. B. Giragama, "A retinal image enhancement technique for blood vessel segmentation algorithm," 2017 IEEE International Conference on Industrial and Information Systems (ICIIS), Peradeniya, 2017, pp. 1-5.

[5] M. Anna Latha, N. Christy Evangeline and S. SankaraNarayanan, "Colour Image Segmentation of Fundus Blood Vessels for the Detection of Hypertensive Retinopathy," 2018 Fourth International Conference on Biosignals, Images and Instrumentation (ICBSII), Chennai, 2018, pp. 206-212.

[6] T. Mapayi and J. Tapamo, "Difference image and fuzzy c-means for detection of retinal vessels," 2016 IEEE Southwest Symposium on Image Analysis and Interpretation (SSIAI), Santa Fe, NM, 2016, pp. 169-172.

[7] Pillai, Sonal Wilson, L. T. Herlin, and Ashwin G. Singerji. "Retinal Blood Vessel Extraction using ISODATA Clustering and Morphological Operations.

[8] Akbar, Shahzad, Muhammad Sharif, Muhammad Usman Akram, Tanzila Saba, Toqeer Mahmood, and Mahyar Kolivand. "Automated techniques for blood vessels segmentation through fundus retinal images: A review." *Microscopy research and technique* 82, no. 2 (2019): 153-170.

[9] Roychowdhury S, Koozekanani DD, Parhi KK. Blood vessel segmentation of fundus images by major vessel extraction and subimage classification" IEEE J Biomed Health Inf 2015; 19(3):1118e28.

[10] Santosh Kumar, N. C., and Y. Radhika. "Optimized maximum principal curvatures based segmentation of blood vessels from retinal images." (2019).

LIST OF PUBLICATIONS

Publication accepted and presented.

[1] A segmentation technique of retinal blood vessels using multi-threshold and morphological operations. International Conference on Computational Performance Evaluation (ComPE-2020).

Publication communicated.

[1] Retinal vessels extraction using Maximum Principle Curvature. 4th International Conference on Recent trends in Communication and Electronics (ICCE 2020).