A MAJOR PROJECT REPORT ON

RETINAL BLOOD VESSEL SEGMENTATION

A dissertation submitted in partial fulfillment of the

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in

INFORMATION TECHNOLOGY

Submitted by

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ACCREDITED BY NBA, AICTE & Affiliated to JNTU-H

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CERTIFICATE

This is to certify that the Project Report entitled **"RETINAL BLOOD VESSEL SEGMENTATION"** is a bonafide work done and submitted by P.**Sai Saket pradyumna (15B81A1275)**, V.**Sai Shanthan (15B81A1277)** and **K.T.Snehith Kishore Reddy(15B81A12A8)** during the academic year 2018-2019, in partial fulfillment of requirement for the award of Bachelor of Technology degree in Information Technology from Jawaharlal Nehru Technological University Hyderabad, is a bonafide record of work carried out by them under my guidance and supervision.

Certified further that to the best of my knowledge, the work in this dissertation has not been submitted to any other institution for the award of any degree or diploma.

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ABSTRACT

Digital images are obtained from the retina and graded by trained professionals. Progression of diabetic retinopathy is assessed by its severity, which in turn determines the frequency of examinations. However, a significant shortage of professional observers has prompted computer assisted monitoring. Assessment of blood vessels network plays an important role in a variety of medical disorders. Manifestations of several vascular disorders, such as diabetic retinopathy, depend on detection of the blood vessels network. In this work green channel of the fundus RGB image was used for obtaining the traces of blood vessels. The algorithm developed used morphological operation to smoothen the background, allowing veins, to be seen clearly. Disc structuring elements were used in this work. The algorithm implemented has employed modules such as contrast enhancement, background exclusion and thresholding. The techniques described in the paper are based on morphological operation and apply on publicly available DRIVE, diaretdb0, diaretdb1 databases and images from eye hospital. Experimental results obtained by using gray-scale/green-channel images have been presented. The implemented algorithm has been shown to be a highly effective method for classifying retinal blood vessels. The implemented algorithm being simple and easy to implement, is best suited for fast processing applications.

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1. INTRODUCTION

EYE

The human eye is an organ which reacts to light and pressure. As a sense organ, the mammalian eye allows vision. Human eyes help to provide a three dimensional, moving image, normally coloured in daylight. Rod and cone cells in the retina allow conscious light perception and vision including color differentiation and the perception of depth. The human eye can differentiate between about 10 million colors and is possibly capable of detecting a single photon.

Similar to the eyes of other mammals, the human eye's non-image-forming photosensitive ganglion cells in the retina receive light signals which affect adjustment of the size of the pupil, regulation and suppression of the hormone melatonin and entrainment of the body clock.

Structure of Eye

The eye is not shaped like a perfect sphere, rather it is a fused two-piece unit, composed of the anterior segment and the posterior segment. The anterior segment is made up of the cornea, iris and lens. The cornea is transparent and more curved, and is linked to the larger posterior segment, composed of the vitreous, retina, choroid and the outer white shell called the sclera. The cornea is typically about 11.5 mm (0.3 in) in diameter, and 1/2 mm (500 μm) in thickness near its center. The posterior chamber constitutes the remaining five-sixths; its diameter is typically about 24 mm. The cornea and sclera are connected by an area termed the limbus. The iris is the pigmented circular structure concentrically surrounding the center of the eye, the pupil, which appears to be black. The size of the pupil, which controls the amount of light entering the eye, is adjusted by the iris' dilator and sphincter muscles.

Light energy enters the eye through the cornea, through the pupil and then through the lens. The lens shape is changed for near focus (accommodation) and is controlled by the ciliary muscle. Photons of light falling on the light-sensitive cells of the retina (photoreceptor cones and rods) are converted into electrical signals that are transmitted to the brain by the optic nerve and interpreted as sight and vision.

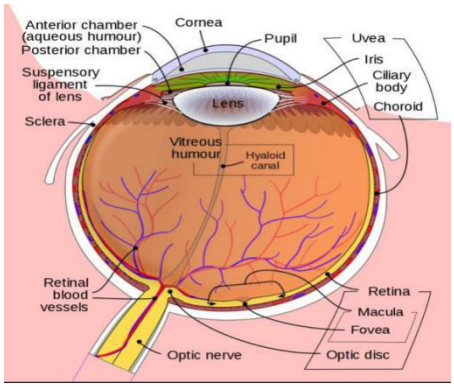

Figure1: Structure of eye

Retina

The **retina** is the innermost, light-sensitive layer of tissue of the eye of most vertebrates and some molluscs. The optics of the eye create a focused two-dimensional image of the visual world on the retina, which translates that image into electrical neural impulses to the brain to create visual perception, the retina serving a function analogous to that of the film or image sensor in a camera.

The neural retina consists of several layers of neurons interconnected by synapses, and is supported by an outer layer of pigmented epithelial cells. The primary light-sensing cells in the retina are the photoreceptor cells, which are of two types: rods and cones. Rods function mainly in dim light and provide black-and-white vision. Cones function in well-lit conditions and are responsible for the perception of colour, as well as high-acuity vision used for tasks such as reading. A third type of light-sensing cell, the photosensitive ganglion cell, is important for entrainment of circadian rhythms and reflexive responses such as the pupillary light reflex.

Light striking the retina initiates a cascade of chemical and electrical events that ultimately trigger nerve impulses that are sent to various visual centres of the brain through the fibres of the optic nerve. Neural signals from the rods and cones undergo processing by other neurons, whose output takes the form of action potentials in retinal ganglion cells whose axons form the optic nerve. Several important features of visual perception can be traced to the retinal encoding and processing of light.

In vertebrate embryonic development, the retina and the optic nerve originate as outgrowths of the developing brain, specifically the embryonic diencephalon; thus, the retina is

considered part of the central nervous system (CNS) and is actually brain tissue. It is the only part of the CNS that can be visualized non-invasively.

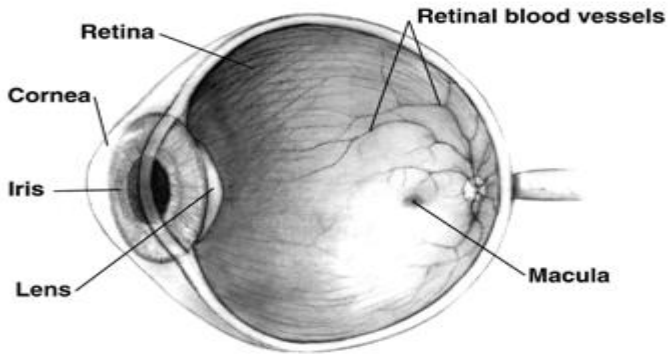

Figure 2:Structure of retina

Retinal layer

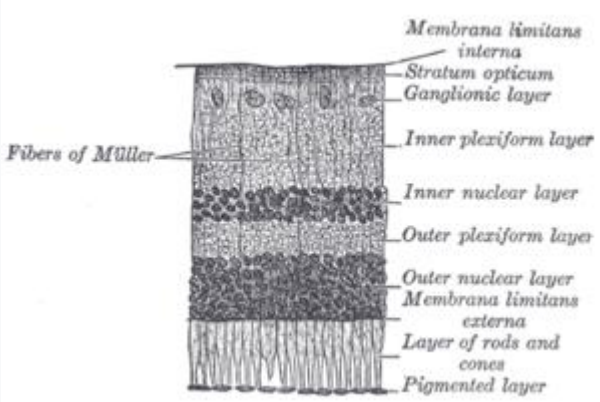

Figure 3:Section of retina

Rods, cones and nerve layers in the retina. The front (anterior) of the eye is on the left. Light (from the left) passes through several transparent nerve layers to reach the rods and cones (far right). A chemical change in the rods and cones send a signal back to the nerves. The signal goes first to the bipolar and horizontal cells (yellow layer), then to the amacrine cells and ganglion cells (purple layer), then to the optic nerve fibres. The signals are processed in these layers. First, the signals start as raw outputs of points in the rod and cone cells. Then the

nerve layers identify simple shapes, such as bright points surrounded by dark points, edges, and movement. (Based on a drawing by Ramón y Cajal, 1911.)

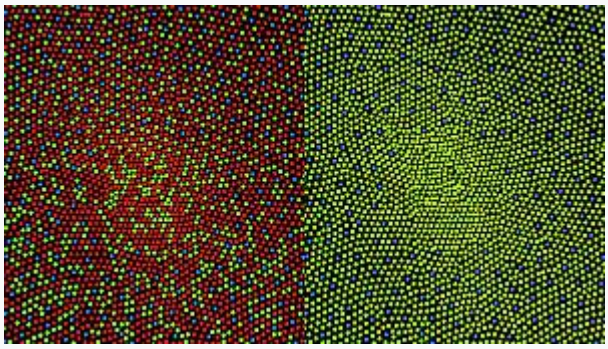

Figure 4:Distribution of rods and cones

The optic nerve is a central tract of many axons of ganglion cells connecting primarily to the lateral geniculate body, a visual relay station in the diencephalon (the rear of the forebrain). It also projects to the superior colliculus, the suprachiasmatic nucleus, and the nucleus of the optic tract. It passes through the other layers, creating the optic disc in primates.

Additional structures, not directly associated with vision, are found as outgrowths of the retina in some vertebrate groups. In birds, the pecten is a vascular structure of complex shape that projects from the retina into the vitreous humour; it supplies oxygen and nutrients to the eye, and may also aid in vision. Reptiles have a similar, but much simpler, structure.

In adult humans, the entire retina is approximately 72% of a sphere about 22 mm in diameter. The entire retina contains about 7 million cones and 75 to 150 million rods. The optic disc, a part of the retina sometimes called "the blind spot" because it lacks photoreceptors, is located at the optic papilla, where the optic-nerve fibres leave the eye. It appears as an oval white area of 3 mm^2.

Temporal (in the direction of the temples) to this disc is the macula, at whose centre is the fovea, a pit that is responsible for our sharp central vision but is actually less sensitive to light because of its lack of rods. Human and non-human primatespossess one fovea, as opposed to certain bird species, such as hawks, who are bifoviate, and dogs and cats, who possess no fovea but a central band known as the visual streak. Around the fovea extends the central retina for about 6 mm and then the peripheral retina. The farthest edge of the retina is defined by the ora

serrata. The distance from one ora to the other (or macula), the most sensitive area along the horizontal meridian is about 32 mm.

Functions of Retina:

The retina translates an optical image into neural impulses by the patterned excitation of the colour-sensitive pigments of its rods and cones, the retina's photoreceptor cells. The excitation is processed by the neural system and various parts of the brain working in parallel to form a representation of the external environment in the brain.

The cones respond to bright light and mediate high-resolution colour vision during daylight illumination (also called photopic vision). The rods are saturated at daylight levels and don't contribute to pattern vision. However, rods do respond to dim light and mediate lower-resolution, monochromatic vision under very low levels of illumination (called scotopic vision). The illumination in most office settings falls between these two levels and is called mesopic vision. At mesopic light levels, both the rods and cones are actively contributing pattern information. What contribution the rod information makes to pattern vision under these circumstances is unclear.

The response of cones to various wavelengths of light is called their spectral sensitivity. In normal human vision, the spectral sensitivity of a cone falls into one of three subtypes, often called blue, green, or red but more accurately known as short, medium, or long wavelength-sensitive cone subtypes. It is a lack of one or more of the cone subtypes that causes individuals to have deficiencies in colour vision or various kinds of colour blindness. These individuals are not blind to objects of a particular colour but are unable to distinguish between colours that can be distinguished by people with normal vision.

Humans have this trichromatic vision, while most other mammals lack cones with red sensitive pigment and therefore have poorer dichromatic colour vision. However, some animals have four spectral subtypes, e.g. the trout adds an ultraviolet subgroup to short, medium, and long subtypes that are similar to humans. Some fish are sensitive to the polarization of light as well.

In the photoreceptors, exposure to light hyperpolarizes the membrane in a series of graded shifts. The outer cell segment contains a photopigment. Inside the cell the normal levels

of cyclic guanosine monophosphate (cGMP) keep the Na+ channel open, and thus in the resting state the cell is depolarised. The photon causes the retinal bound to the receptor protein to isomerise to trans-retinal. This causes the receptor to activate multiple G-proteins. This in turn causes the Ga-subunit of the protein to activate a phosphodiesterase (PDE6), which degrades cGMP, resulting in the closing of Na+ cyclic nucleotide-gated ion channels (CNGs). Thus the cell is hyperpolarised. The amount of neurotransmitter released is reduced in bright light and increases as light levels fall. The actual photopigment is bleached away in bright light and only replaced as a chemical process, so in a transition from bright light to darkness the eye can take up to thirty minutes to reach full sensitivity.

When thus excited by light, the photoceptor sends a proportional response synaptically to bipolar cells which in turn signal the retinal ganglion cells. The photoreceptors are also cross-linked by horizontal cells and the neural signals being intermixed and combined. Of the retina's nerve cells, only the retinal ganglion cells and few amacrine cells create action potentials.

In the retinal ganglion cells there are two types of response, depending on the receptive field of the cell. The receptive fields of retinal ganglion cells comprise a central, approximately circular area, where light has one effect on the firing of the cell, and an annular surround. In ON cells, an increment in light intensity in the centre of the receptive field causes the firing rate to increase. In OFF cells, it makes it decrease. In a linear model, this response profile is well described by a difference of Gaussians and is the basis for edge detection algorithms.

Retinal vasculature structure implicates important information helps the ophthalmologist in detecting and diagnosing a variety of retinal pathology such as Retinopathy of Prematurity (RoP),diabetic retinopathy, glaucoma, hypertension, and age-related macular degeneration or in diagnosis of diseases related to brain and heart stocks, which are associated with the abnormal variations in retinal vascular structure. Therefore, changes in retina’s arterioles and venules morphology have a principal diagnostic value.

In general, vessels segmentation occupy a remarkable place in medical image segmentation field,retinal vessels segmentation belongs to this category where a broad variety of algorithms and methodologies have been developed and implemented for the sake of automatic identification, localization and extraction of retinal vasculature structures .

Fundus(EYE):

The **fundus** of the eye is the interior surface of the eye opposite the lens and includes the retina, optic disc, macula, fovea, and posterior pole. The fundus can be examined by ophthalmoscopy and/or fundus photography.

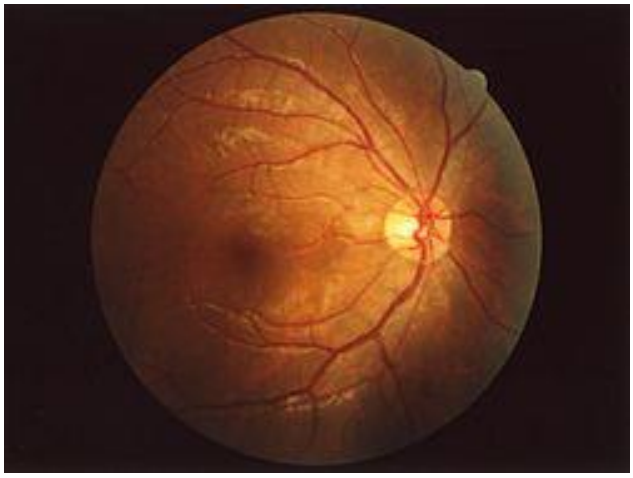

Figure 5:Fundus

Retinal Fundus Imaging:

Retina photography is typically conducted via an optical apparatus called fundus camera, fundus camera can be viewed as a low power microscope that specializes in retina fundus imaging, where the retina is illuminated and imaged via the attached camera. In particular, fundus camera is designed to capture an image for the interior surface of human eye, which is composed of major parts, including macula, optic disk, retina and posterior pole.

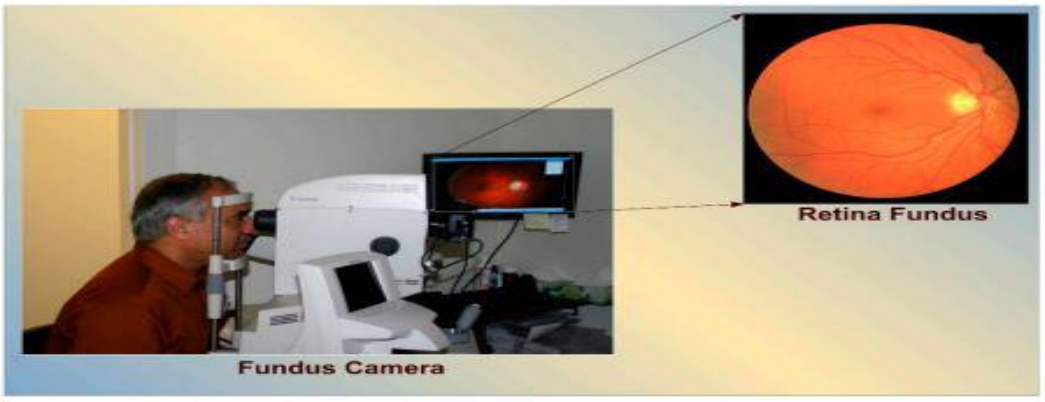

Figure 6:Fundus Camera

Fundus photography can be viewed as a sort of documentation process for the retinal interior structure and retinal neurosensory tissues. The retinal neurosensory tissues convert the optical images reflection, that we see, into electrical signals in the form of pulses sent to our brain where it decoded and understood. Retina photography can be conducted based on the idea that the eye pupil is utilized as both an entrance and exit for the illuminating and imaging light rays that are used by the fundus camera. During the fundus photography, patients' foreheads are placed against the bar and their chins placed in the chin rest.After the oculist aligns the fundus camera, the camera shutter is released so a flash light is fired and a two-dimensional picture for retina fundus has been taken. the fundus of the human eye is the back portion of the interior of the eye ball. The optic nerve resides at the center of retina, which can be seen as a white area of circular to oval shape and measuring about 3 × 3 mm across diameter. The major blood vessels of the retina radiate from the center of the area of optic nerve and then radiate and branched to fill the entire area excepting the fovea zone, which is a blood-vessel free reddish spot with an oval-shaped that lies directly to the left of the optic disc and resides in the center of an area that is known by ophthalmologists as the "macula" region.

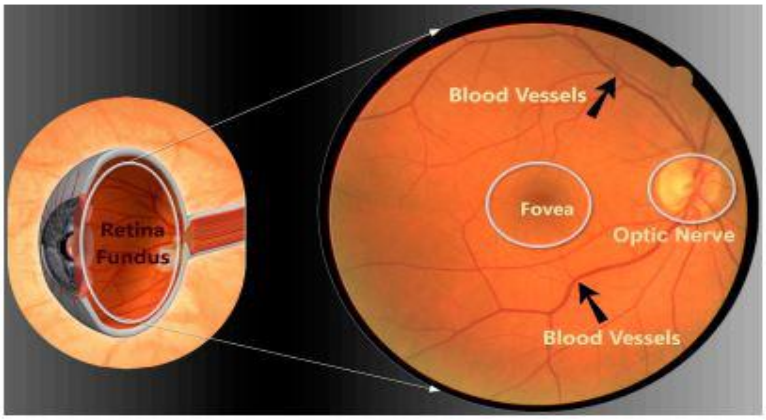

Figure 7:Fundus Image

In General, the photographic process involves grasping the light that reflected off the subject under consideration. In our case, the subject is the fundus of retina. Since the internal room of the eye has no light source of its own, in the retina photography, we need to flash or shine a light into eye room to capture a good photograph. The ocular fundus imaging has three major photography modes:

1. Full-color Imaging
2. Monochromatic (Filtered) Imaging
3. Fluorescence Angiogram

In the case of full-color photography mode, no light-filtration is used and it is totally non-invasive contrary to other modes of fundus imaging. The resultant retina fundus image is a two-dimensional full color image.

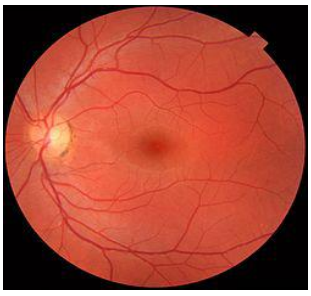

Figure 8:Full-color Imaging

On the other hand, if the fundus is imaged via a monochromatic filter or via particular colored illumination, then the fundus photography is called “monochromatic”, This type of fundus photography is built based on the idea that the visibility of different structures in a retinal image is enhanced if the spectral range of illumination is changed correspondingly. In other words, instead of using white light of a broad scale of wavelengths, we use a light of a specified wavelength that corresponds to a specific color, for example, a red object in an image would appear lighter if the image is taken through a red filter and it would appear darker if it is taken through green filter. As the white light can be divided into red, green and blue lights, the ocular fundus can be photographed via one of these gradient lights where each light has the capability to enhance the visibility of specific retinal anatomical structures based on their colors. For example, blue filter (filter with blue light) enhances the visibility of the interior of retina, which in full-color photo (taken by white light) appears almost transparent. On the other hand, we can get the best overall view of retina fundus and the most enhanced contrast if we use the green filter. Moreover, green filters have the capability to enhance the visibility of common lesions such as exudates, hemorrhage and drusen.

Another alternative to monochromatic filters is to split the full-color fundus image into its basic components, namely, red, green and blue. It operates similar to colored filters, except we lose the resolution, and is adopted in a variety of retinal vessel identification approaches in the stage of image preprocessing framework.

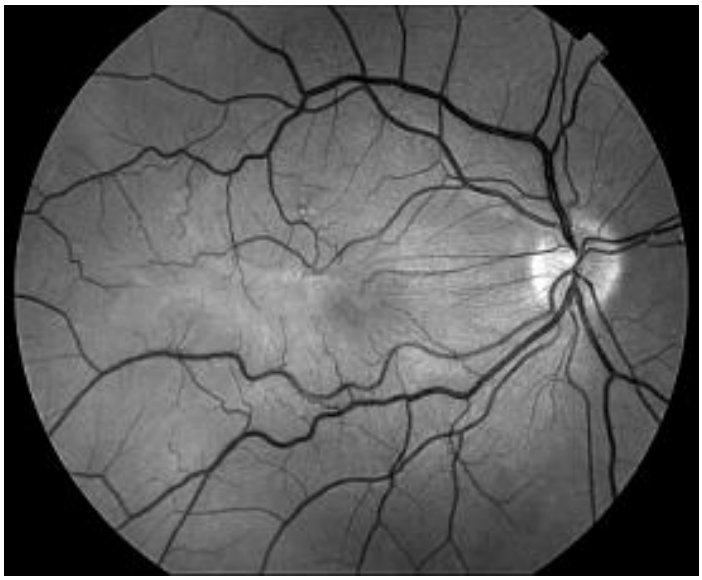

Figure 9:Monochromatic Imaging

Fundus angiography is the most invasive fundus imaging, and involves injecting a tiny amount of fluorescein dye into a vein of patient's arm; the dye makes its way to the main blood stream leading to retina vessels, and then the retina fundus is photographed. Originally, the word "angiography" is derived from the Greek words Angeion, which means "vessels", and "graphien", which means to record or to write. Once the sodium-fluorescein has been injected, and reaches retina, the retina fundus is illuminated with a blue light, and then is flashed in a yellow-green color. Later, specialized filters in the fundus camera allow the fluorescent light to be imaged, leading to high contrast (grey-scaled) retinal vascular structure images,Retina angiography is considered the photography mode that highly revolutionized the ophthalmologists' ability to understand both retina physiology and pathology. Moreover, it is used in the process of diagnosing and treating choroidal diseases.However, this mode is considered the most invasive one, due to injecting dyes in the human veins directly. Thus, as recently reported, it is important to consider the potential risk associated with using such mode of retina fundus photography, especially for neonatal people.

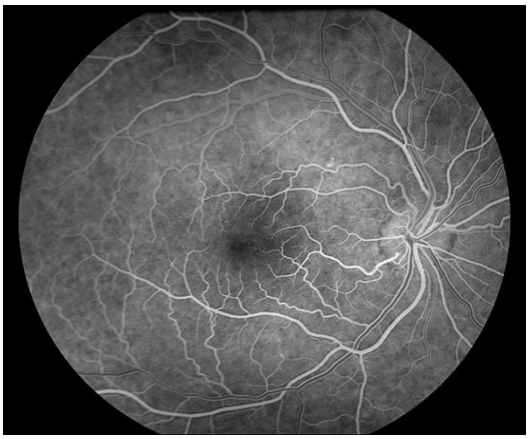

Figure 10:Fluorescence Angiogram

Image Processing:

Image processing is a method to convert an image into digital form and perform some operations on it, in order to get an enhanced image or to extract some useful information from it.

It is a type of signal dispensation in which input is image, like video frame or photograph and output may be image or characteristics associated with that image. Usually **Image Processing** system includes treating images as two dimensional signals while applying already set signal processing methods to them.

Image processing basically includes the following three steps.

1.Importing the image with optical scanner or by digital photography.

2. Analyzing and manipulating the image which includes data compression and image enhancement and spotting patterns that are not to human eyes like satellite photographs.

3.Output is the last stage in which result can be altered image or report that is based on image analysis.

Image

An image is defined as a two-dimensional function,**F(x,y)**, where x and y are spatial coordinates, and the amplitude of **F** at any pair of coordinates (x,y) is called the **intensity** of that image at that point. When x,y, and amplitude values of **F** are finite, we call it a **digital image**.

In other words, an image can be defined by a two-dimensional array specifically arranged in rows and columns.Digital Image is composed of a finite number of elements, each of which elements have a particular value at a particular location.These elements are referred to as picture elements,image elements,and pixels.A *Pixel* is most widely used to denote the elements of a Digital Image.

Types

The two types of **methods used for Image Processing** are **Analog and Digital** Image Processing. Analog or visual techniques of image processing can be used for the hard copies like printouts and photographs. Image analysts use various fundamentals of interpretation while using these visual techniques. The image processing is not just confined to area that has to be studied but on knowledge of analyst. Association is another important tool in image processing through visual techniques. So analysts apply a combination of personal knowledge and collateral data to image processing.

Digital Processing techniques help in manipulation of the digital images by using computers. As raw data from imaging sensors from satellite platform contains deficiencies. To get over such flaws and to get originality of information, it has to undergo various phases of processing. The three general phases that all types of data have to undergo while using digital technique are Pre- processing, enhancement and display, information extraction.

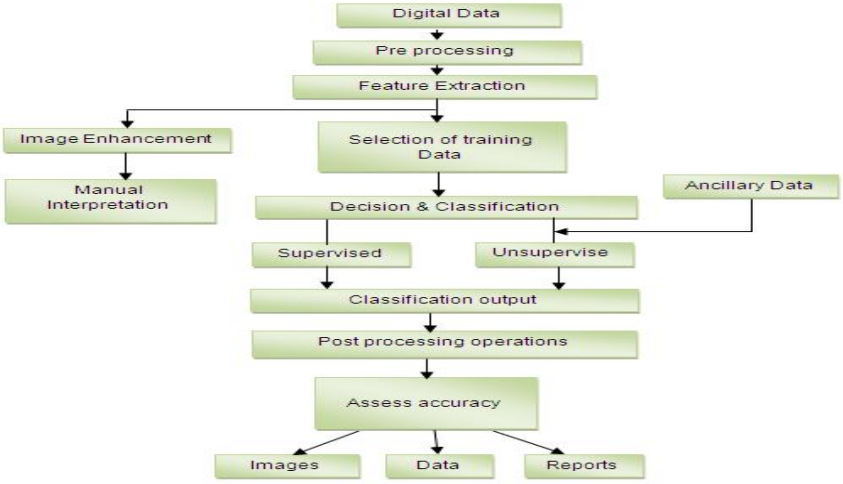

Figure 11:Flowchart of process in image processing

PHASES OF IMAGE PROCESSING:

1.**ACQUISITION**– It could be as simple as being given an image which is in digital form. The main work involves:

a) Scaling

b) Color conversion(RGB to Gray or vice-versa)

2.**IMAGE ENHANCEMENT**– It is amongst the simplest and most appealing in areas of Image Processing it is also used to extract some hidden details from an image and is subjective.

3.**IMAGE RESTORATION**– It also deals with appealing of an image but it is objective(Restoration is based on mathematical or probabilistic model or image degradation).

4.**COLOR IMAGE PROCESSING**– It deals with pseudocolor and full color image processing color models are applicable to digital image processing.

5.**WAVELETS AND MULTI-RESOLUTION PROCESSING**– It is foundation of representing images in various degrees.

6.**IMAGE COMPRESSION**-It involves in developing some functions to perform this operation. It mainly deals with image size or resolution.

7.**MORPHOLOGICAL PROCESSING**-It deals with tools for extracting image components that are useful in the representation & description of shape.

8.**SEGMENTATION PROCEDURE**-It includes partitioning an image into its constituent parts or objects. Autonomous segmentation is the most difficult task in Image Processing.

9.**REPRESENTATION & DESCRIPTION**-It follows output of segmentation stage, choosing a representation is only the part of solution for transforming raw data into processed data.

10.**OBJECT DETECTION AND RECOGNITION**-It is a process that assigns a label to an object based on its descriptor.

Retinal Image Processing:

The oculists scan the retina of patients using fundus camera with high resolution. Accordingly,the situation of retina blood vessels is probed to diagnose retinal diseases. In many cases, it is found that the retinal vascular structure has low contrast with regard to their background. Thus, the diagnosis of retinal diseases becomes a hard task, and applying a suitable image segmentation technique becomes a must for highly accurate retinal vascular structure detection, since it leads to accurate diagnosis.Retina vessel identification and extraction faces many challenges that may be outlined as follows.

Firstly, the retinal vessels' widths take a wide range of color intensity range from less than one pixel up to more than five pixels in the retinal image,which requires an identification technique with high flexibility.

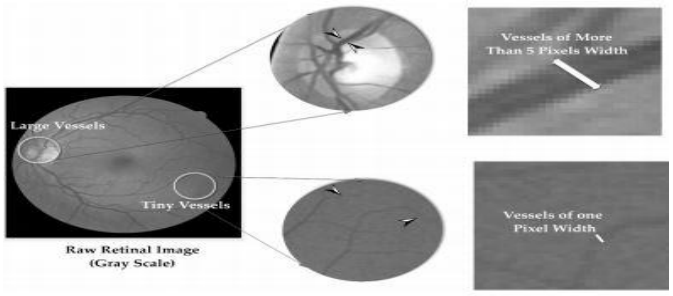

Figure 12:Retinal Image Processing

To elaborate this challenge, a snippet of MATLAB code has been developed for sake of grey levels substitution in retinal image; the different grey levels of a raw retinal image have been replaced by color ones.It can be noted that many retinal vessels, either large or tiny ones, take the same background color intensities. This reveals the broad range of colors that may be taken by the retinal vasculature structure, making the identification process more complicated rather than that found in other identification problems.

First challenge of retinal image segmentation: (a) Vessels types distribution on retina surfaces; and (b) different sizes of retinal vessels. This challenge opens the room for a field of research specialized in detecting and segmenting thin retinal vascular structures, as in . Secondly, Vessels identification in pathological retinal images faces a tension between accurate vascular structure extraction and false responses near pathologies and other nonvascular structures (such as optic disc and fovea region). The retinal blood vasculature is a tree-like structure that disperses across the fundus image surface including pathologies. Thin and filamentary retinal vessels melt in the retinal abnormal regions burden the task of accurate vessel segmentation

In summary, retinal vascular structure, inside either normal or abnormal retina images, has low contrast with respect to the retinal background. Conversely, other retinal anatomical structures have high contrast to other background tissues but with indistinct features in comparison with abnormal structures; optic disc and exudates lesions represent typical examples.

All these challenges, in terms of medical image processing, make the classical segmentation techniques such as Sobel operators,Prewitt operators,gradient operators and Robert and Krish differential operations inefficient and inaccurate. Consequently, various algorithms and methodologies have been developed and implemented for sake of automatic identification, localization and extraction of retinal anatomical structures and can be broadly divided into rule-based and machine learning techniques.

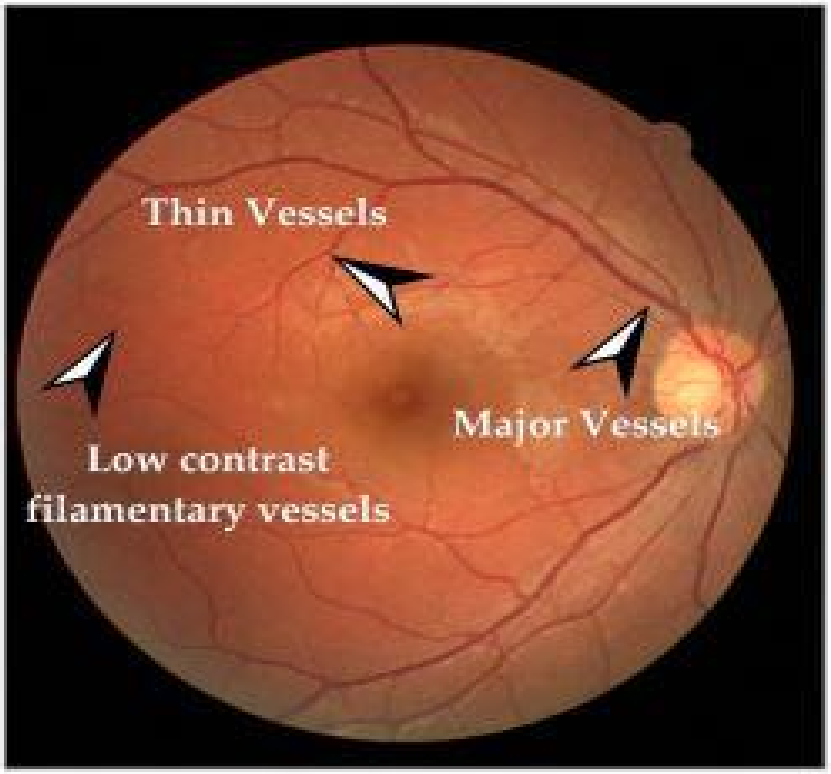

Figure 13:Filamentary Vessels

1.1 LITERATURE SURVEY

This chapter deals with different research works that were carried out in past and contributed to this field of Retinal Blood Vessel Segmentation. So many approaches are developed for the segmentation of blood vessels which played important role to carry out this work. Some of the related research work is described below:

Pawel Liskowski, Krzysztof Krawiec, Member, Citation information: DOI 10.1109/TMI.2016.2546227, IEEE Transactions on Medical Imaging "Segmenting Retinal Blood Vessels with Deep Neural Networks".

The condition of the vascular network of human eye is an important diagnostic factor in ophthalmology. Its segmentation in fundus imaging is a nontrivial task due to variable size of vessels, relatively low contrast, and potential presence of pathologies like microaneurysms and hemorrhages.ised and supervised, have been proposed for this purpose in the past[1].He proposed a supervised segmentation technique that uses a deep neural network trained on a large sample of examples pre processed with global contrast normalization, zero-phase whitening, and augmented using geometric transformations and gamma corrections. When applied to standard benchmarks of fundus imaging, the DRIVE, STARE, and CHASE databases, the networks significantly outperform the previous algorithms on the area under ROC curve measure (up to > 0.99) and accuracy of classification (up to > 0.97). The method is also resistant to the phenomenon of central vessel reflex, sensitive in detection of fine vessels (sensitivity > 0.87), and fares well on pathological cases.

Automatic segmentation of blood vessels from retinal fundus images through image processing and data mining techniques ,R GEETHARAMANI and LAKSHMI BALASUBRAMANIAN

Machine Learning techniques have been useful in almost every field of concern. Data Mining, a branch of Machine Learning is one of the most extensively used techniques. The ever-

increasing demands in the field of medicine are being addressed by computational approaches in which Big Data analysis, image processing and data mining are on top priority. [12] Vessel segmentation can also be a pre-processing step for segmentation of other retinal structures like optic disc, fovea, microneurysms, etc. In this paper, blood vessel segmentation is attempted through image processing and data mining techniques. The retinal blood vessels were segmented through color space conversion and color channel extraction, image pre-processing, Gabor filtering, image postprocessing, feature construction through application of principal component analysis, k-means clustering and first level classification using Naïve–Bayes classification algorithm . A comparison of these results with the existing methodologies is also reported. This methodology can help ophthalmologists in better and faster analysis and hence early treatment to the patients.

Retinal Blood Vessel Segmentation Based on Multi-Scale Deep Learning Ming Li ; Qingbo Yin ; Mingyu Lu

Fundus images are one of the main methods for diagnosing eye diseases in modern medicine. The vascular segmentation of fundus images is an essential step in quantitative disease analysis. Based on the previous studies, we found that the category imbalance is one of the main reasons that restrict the improvement of segmentation accuracy[17]. This paper presents a new method for supervised retinal vessel segmentation that can effectively solve the above problems.

In recent years, it is a popular method that using deep learning to solve retinal vessel segmentation. We have improved the loss function for deep learning in order to better handle category imbalances. By using a multi-scale convolutional neural network structure and label processing approach, our results have reached the most advanced level. Our approach is a meaningful attempt to improve blood vessel segmentation and further improve the diagnostic level of eye diseases.

IMAGE RESTORATION BASED ON MORPHOLOGICAL OPERATIONS- A.M.Raid, W.M.Khedr, M.A.El-dosuky and Mona Aoud

Image processing including noise suppression, feature extraction, edge detection, image s egmentation,shape recognition, texture analysis, image restoration and reconstruction, image compression etc uses mathematical morphology which is a method of nonlinear filters. It is modulated from traditional morphology to order morphology, soft mathematical morphology and fuzzy soft mathematical morphology.[18]This paper is covers 6 morphological operations which are implemented in the matlab program, including erosion, dilation, opening, closing,boundary extraction and region filling.

Detecting Morphological Filtering of Binary Images -Francesco G. B. De Natale Giulia Boato

Morphological operators are widely used in binary image processing for several purposes, such as removing noise, detecting contours or particular structures, and regularizing shapes. In particular, morphological filters are largely adopted in scanned documents to correct the artifacts caused by acquisition and binarization, as well as other processing[20]. In this paper, we propose a novel approach for forensics detection of morphological filtering on binary images. The proposed technique exploits some mathematical properties of the two basic morphologic operators, erosion and dilation, to define an algorithm able not only to detect the application of the filter, but also to estimate the shape of the relevant structuring element. Experimental tests demonstrate that the technique is effective and robust to the most common operations performed on binary image documents.

2.SOFTWARE REQUIREMENTS SPECIFICATIONS

- **Functional Requirements:**
    1. **Pre-processing(MODULE 1):** In the pre-processing stage, we convert RGB2Green channel and then it is converted to grey image.We use median filtering and canny edge detectors to enhance the image.
    2. **Segmentation(MODULE 2):** We use Line Tracking algorithm to segment the vessels from pre-processed image.
    3. **Post segmentation(MODULE 3):**Morphological operators such as erosion and dilation are used to remove noise in the output image and output displays blood vessels in a much clearer manner.
- **Non-Functional Requirements:**
    1. **Availability:** The Software for the detection of blood vessels in the eye is available on systems where MATLAB is installed.
    2. **Reliability**: This system attempts to ensure appropriate content but assume no responsibility for external manipulations.
    3. **Response Time:** The System shall give response in 5 seconds after loading the MR image.
    4. **Capacity:** The System will support processing of one image at a time.
    5. **User-interface:** The System shall respond within 3-4 seconds.

- **Software Requirements:**
    - MATLAB R2013a
    - Operating System : Windows

- **Hardware Requirements:**
    - Hard disk: 256GB
    - RAM: 1GB
    - Processor: Pentium(R)Dual-core CPU

3.DESIGN

The database used in the work is obtained from the different hospitals and the DRIVE & STARE data base in the internet. In this work we took different fundus images of retinas .

METHODOLOGY& FLOW CHART OF PROPOSED METHOD

The proposed algorithm aims to work upon the fundus images obtained from DRIVE and STARE database.The algorithm consists of several stages in which the fundus image is read which is taken from fundus camera.The fundus image may be either of .tif,.jpg,.jpeg or .png format.The fundus image is converted into a green channel image,which is in turn converted into gray scale image.The next step is to check if there is any noise present in the gray scale image.Canny edge detector is implemented for enhancing edges and corners.If there is noise present,the median filter is applied to the gray scale image.After enhancing the image the next step is to convert the gray scale image in to binary image by a process called thresholding which comes under segmentation.The threshold value is used in a technique called Line tracking algorithm which is used to segment the blood vessels.Any unwanted pixel information is removed using the morphological operations.

The detailed process is shown in flow chart and explained in the following sections.

Flowchart: The Flow chart for the proposed approach

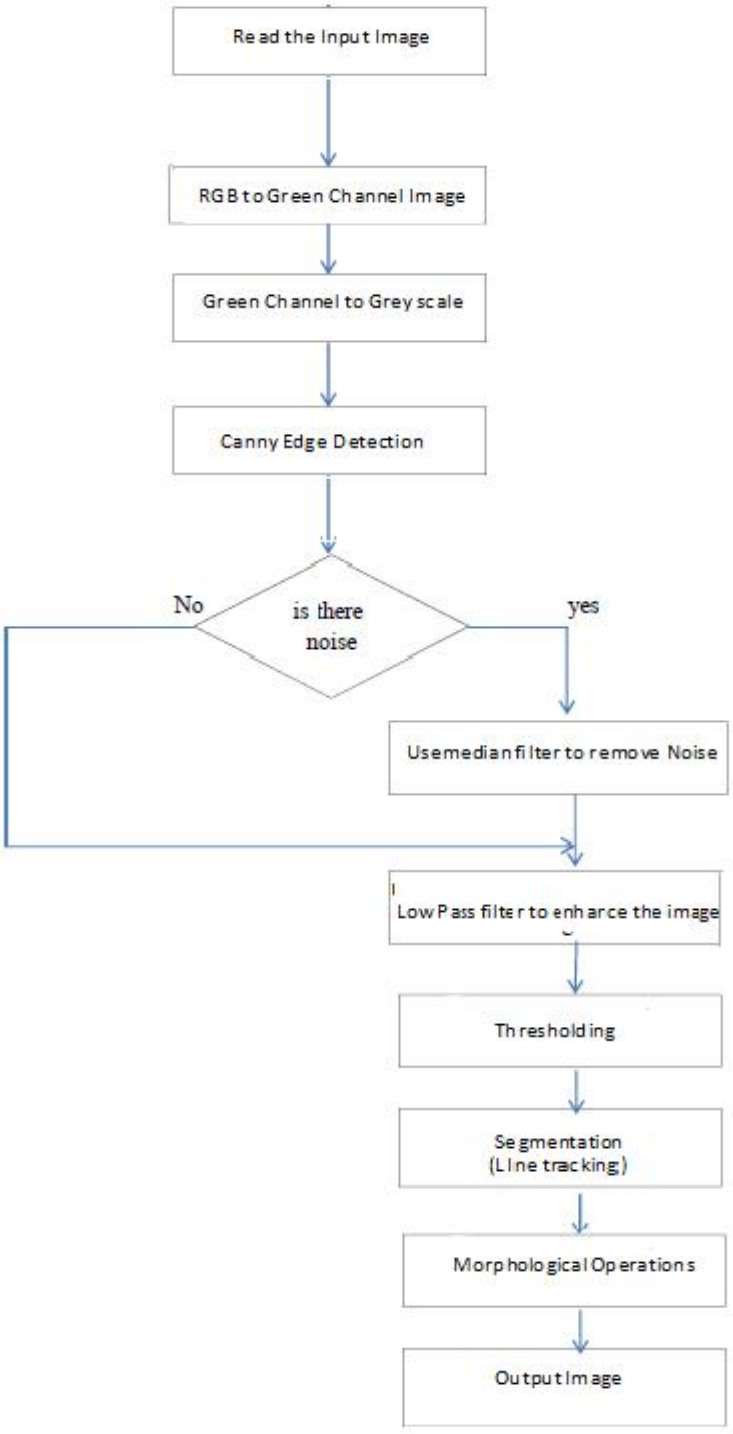

Figure 14: Flowchart for showing the methodology for the proposed approach

4. IMPLEMENTATION

IMAGE SEGMENTATION

In computer vision, **image segmentation** is the process of partitioning a digital image into multiple segments. The goal of segmentation is to simplify and/or change the representation of an image into something that is more meaningful and easier to analyze. Image segmentation is typically used to locate objects and boundaries (lines, curves, etc.) in images. More precisely, image segmentation is the process of assigning a label to every pixel in an image such that pixels with the same label share certain characteristics.

The result of image segmentation is a set of segments that collectively cover the entire image, or a set of contours extracted from the image (edge detection). Each of the pixels in a region are similar with respect to some characteristic or computed property, such as color, intensity, or texture. Adjacent regions are significantly different with respect to the same characteristic(s).

MODULE 1
GRAPHICAL USER INTERFACE

GUI is designed such that usage of every function of the application is easy to use and the picture is given in the further sections.

RGB TO GREEN CHANNEL CONVERSION

The **RGB color model** is an additive color model in which red, green and blue light are added together in various ways to reproduce a broad array of colors. The name of the model comes from the initials of the three additive primary colors, red, green, and blue.

The main purpose of the RGB color model is for the sensing, representation and display of images in electronic systems, such as televisions and computers, though it has also been used in conventional photography. Before the electronic age, the RGB color model already had a solid theory behind it, based in human perception of colors.

RGB is a *device-dependent* color model: different devices detect or reproduce a given RGB value differently, since the color elements (such as phosphors or dyes) and their response to

the individual R, G, and B levels vary from manufacturer to manufacturer, or even in the same device over time. Thus an RGB value does not define the same *color* across devices without some kind of color management.

In general the medical images are all gray scale images in some cases if the image is present in available in the RGB pattern we have to convert these RGB image to the gray scale image. Here as in the chapter 4 we discussed about the different types of images RGB images have different colors for every pixels here in this conversion it converts every pixels in to certain range 0(black)-255(white) which uses the 8 bits to storage this image. The question is why we are converting the RGB image to the gray scale image is for storage and to do process easily.

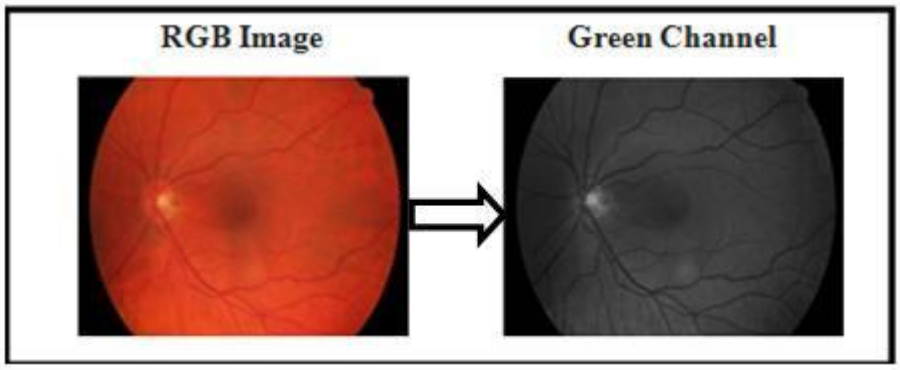

Figure 15 - RGB Image & Green channel Image

GREEN CHANNEL TO GRAYSCALE CONVERSION: Green channel image is converted into Grayscale on which further operations are done using the following snippet:

```
for i =1:h
    for j=1:w
gray(i,j) =0.29* rgb(:,:,1)+0.59* rgb(:,:,2) +0.11* rgb(:,:,3);
end
end
```

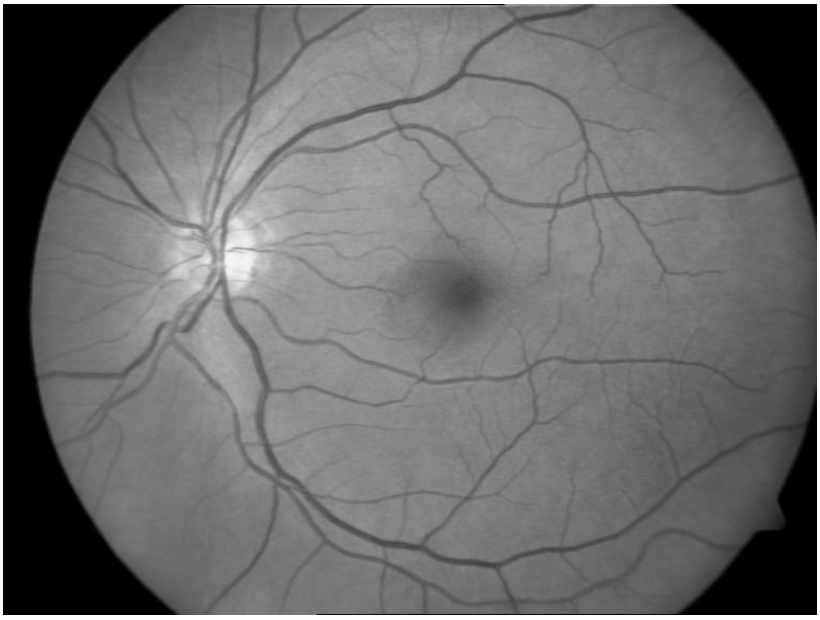

Figure 16 :Grey-Scale Image

MODULE 2

PRE-PROCESSING STAGE

In this stage image is enhanced in the way that finer details are improved and noise is removed from the image. Most commonly used enhancement and noise reduction techniques are implemented that can give best possible results. Enhancement will result in more prominent edges and a sharpened image is obtained, noise will be reduced thus reducing the blurring effect from the image. Filtering is done to remove noise from the medical images because medical images are somewhat noisy. In proposed work we will use filter to smoothing and removing noise from the image. In addition to enhancement, image segmentation will also be applied. This improved and enhanced image will help in detecting edges and improving the quality of the overall image.

ENHANCING THE IMAGE

For the better visualization of the image we have to enhance the image , it means using certain algorithms we have to enhance the clarity of the images.

CANNY EDGE DETECTORS

The Process of Canny edge detection algorithm can be broken down to 5 different steps:

1. Apply Gaussian filter to smooth the image in order to remove the noise
2. Find the intensity gradients of the image
3. Apply non-maximum suppression to get rid of spurious response to edge detection
4. Apply double threshold to determine potential edges.
5. Track edge by hysteresis: Finalize the detection of edges by suppressing all the other edges that are weak and not connected to strong edges.

Gaussian Filter

Since all edge detection results are easily affected by image noise, it is essential to filter out the noise to prevent false detection caused by noise. To smooth the image, a Gaussian filter is applied to convolve with the image. This step will slightly smooth the image to reduce the effects of obvious noise on the edge detector. The equation for a Gaussian filter kernel of size $(2k+1)\times(2k+1)$ is given by:

$$H_{ij} = \frac{1}{2\pi\sigma^2} \exp\left(-\frac{(i-(k+1))^2 + (j-(k+1))^2}{2\sigma^2}\right); 1 \leq i, j \leq (2k+1)$$

Canny edge detection is a multi-step algorithm that can detect edges with noise supressed at the same time.

MEDIAN FILTERING

We have to increase the intensity of the every pixel of the image for this in this proposed algorithm we are using the median filter to enhance the image . The main idea of the median filter is to run to each and every pixel of the image and replaces the each pixel by the median of

all neighboring pixels, so that every pixel will be enhanced by some value and hence the intensity distribution is done to each and every pixel. The main purpose of the median filter is to enhance the image.

The **Median Filter** is a non-linear digital filtering technique, often used to remove noise from an image or signal. Such noise reduction is a typical pre-processing step to improve the results of later processing (for example, edge detection on an image). Median filtering is very widely used in digital image processing because, under certain conditions, it preserves edges while removing noise , also having applications in signal processing.

It is widely used as it is very effective at removing noise while preserving edges. It is particularly effective at removing ‘salt and pepper’ type noise. The median filter works by moving through the image pixel by pixel, replacing each value with the median value of neighboring pixels. The pattern of neighbors is called the "window", which slides, pixel by pixel over the entire image 2 pixels, over the entire image.

LOW PASS FILTERING(BLURRING)

The most basic of filtering operations is called "low-pass". A low-pass filter, also called a "blurring" or "smoothing" filter, averages out rapid changes in intensity. The simplest low-pass filter just calculates the average of a pixel and all of its eight immediate neighbors. The result replaces the original value of the pixel. The process is repeated for every pixel in the image.

Figure 17:Before and After Low-Pass Filter

This low-pass filtered image looks a lot blurrier. But why would you want a blurrier image? Often images can be noisy – no matter how good the camera is, it always adds an amount of ”snow” into the image. The statistical nature of light itself also contributes noise into the image.

Noise always changes rapidly from pixel to pixel because each pixel generates its own independent noise. The image from the telescope isn't "uncorrelated" in this fashion because real images are spread over many pixels. So the low-pass filter affects the noise more than it does the image. By suppressing the noise, gradual changes can be seen that were invisible before. Therefore a low-pass filter can sometimes be used to bring out faint details that were smothered by noise.

MaxIm DL allows you to selectively apply a low-pass filter to a certain brightness range in the image. This allows you to selectively smooth the image background, while leaving the bright areas untouched. This is an excellent compromise because the fainter objects in the background are the noisiest, and it does not degrade the sharpness of bright foreground objects.

HISTOGRAM and THRESHOLD

Intensity Histogram

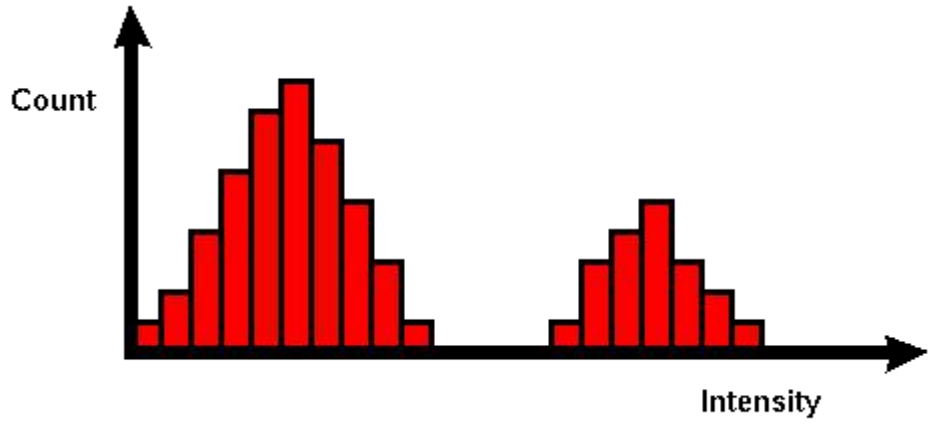

Figure 18:Intensity Histogram

In an image processing context, the histogram of an image normally refers to a histogram of the pixel intensity values. This histogram is a graph showing the number of pixels in an image at each different intensity value found in that image. For an 8-bit grayscale image there are 256 different possible intensities, and so the histogram will graphically display 256 numbers showing the distribution of pixels amongst those grayscale values. Histograms can also be taken of color images - either individual histograms of red, green and blue channels can be taken, or a 3-D histogram can be produced, with the three axes representing the red, blue and green channels, and brightness at each point representing the pixel count. The exact output from the operation depends upon the implementation --- it may simply be a picture of the required histogram in a suitable image format, or it may be a data file of some sort representing the histogram statistics.

How It Works

The operation is very simple. The image is scanned in a single pass and a running count of the number of pixels found at each intensity value is kept. This is then used to construct a suitable histogram.

Guidelines for Use

Histograms have many uses. One of the more common is to decide what value of threshold to use when converting a grayscale image to a binary one by thresholding. If the image is suitable for thresholding then the histogram will be bi-modal --- i.e. the pixel intensities will be clustered around two well-separated values. A suitable threshold for separating these two groups will be found somewhere in between the two peaks in the histogram. If the distribution is not like this then it is unlikely that a good segmentation can be produced by thresholding.

USING THRESHOLD TO SEGMENT

Sometimes in image processing, we need to separate an image into some regions (or their contours), thresholding which is nothing but separation of dark and light regions. Let me explain a bit more simpler. If you have a gray scale image(whose intensity varies from 0 to 255) then it can be converted into binary image using thresholding by setting some threshold value.

For example-

Set threshold =60(on a scale from 0 to 255).Now if intensity of a particular pixel exceeds the threshold value it symbolizes light region or binary 1 else it represents dark region or binary 0.

You can also implement it yourself on MATLAB. Here is the program-

```
A=imread('einstein.bmp'); % Input image

B=im2bw(A,0.50);

imshowpair(A,B,'montage');
```

Sample output is shown here:

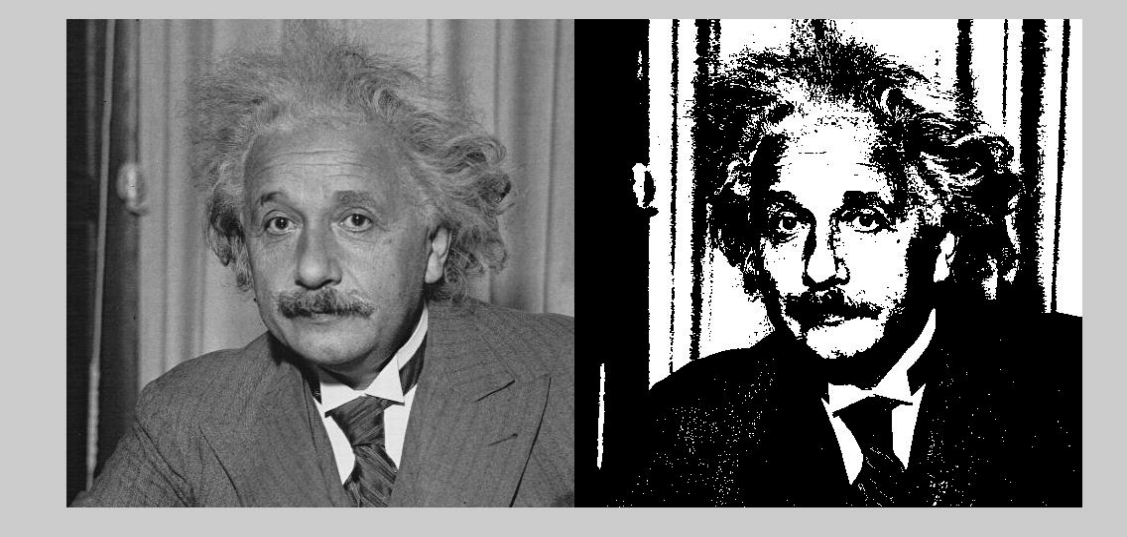

Figure 19 : Threshold to segment

MODULE 3

SEGMENTATION:

Segmentation is an essential process to extract information from complex medical images. The main objective of segmentation is to simplify or change the representation of an image into

something that is more meaningful and easier to analyze and the pixels within the region are homogeneous with respect to a predefined criterion.

LINE TRACKING ALGORITHM:

Line Tracking Method used to trace a line on the image with a certain angular orientation and diameter. By utilizing the image histogram, the pixel area boundaries will be determined to be tracked by the threshold value corresponding to the frequency of the intensity image. After getting the tracking area, it will be done early in the initialization process for tracking pixel pixel neighbors with direction and a predetermined diameter. By calculating the value of the weight of each pixel neighbors, it will be selected the pixels that have the greatest weight and the value exceeds a predetermined threshold weight. If it is not eligible, it will be re-initialization process early pixels. If there is one that meets the pixel, the pixel is marked as a line pixel by providing trust value of "1", while the other pixels set to "0". Furthermore, this process is repeated until all of the pixel area is completed tracking. This is our interface/ visualization of program part 2 :

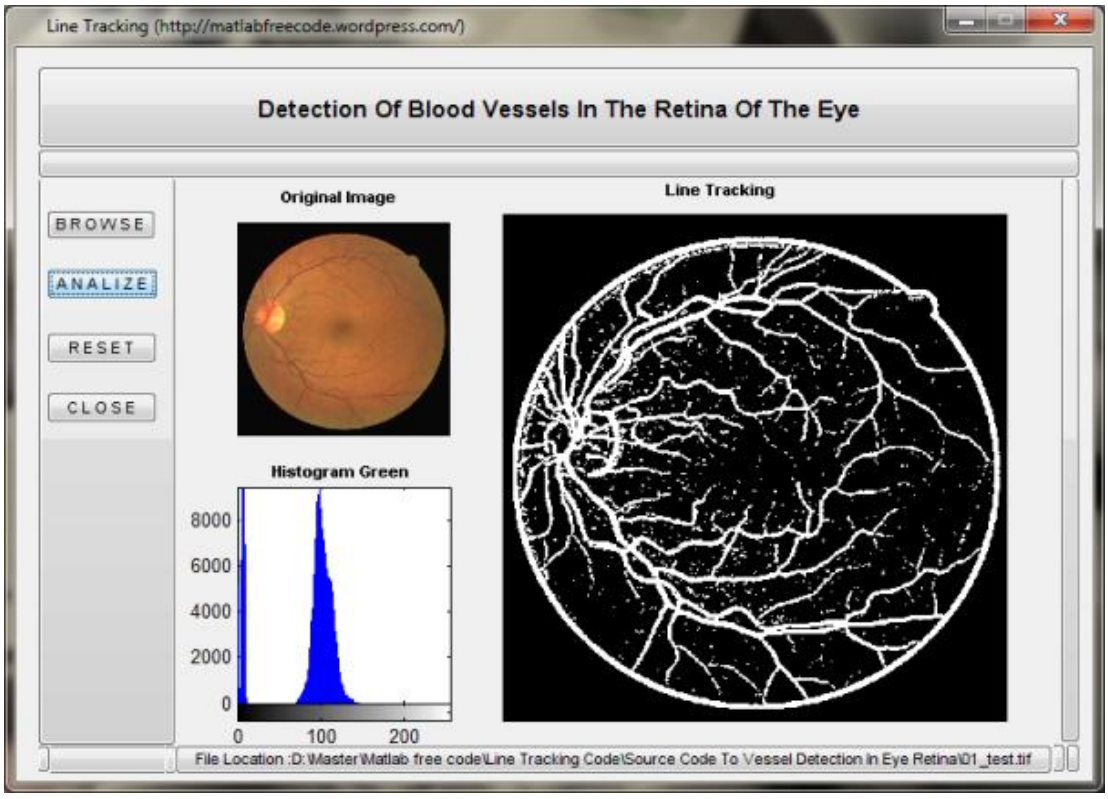

Figure 20 : Calculation of Histogram and Detection of blood vessels

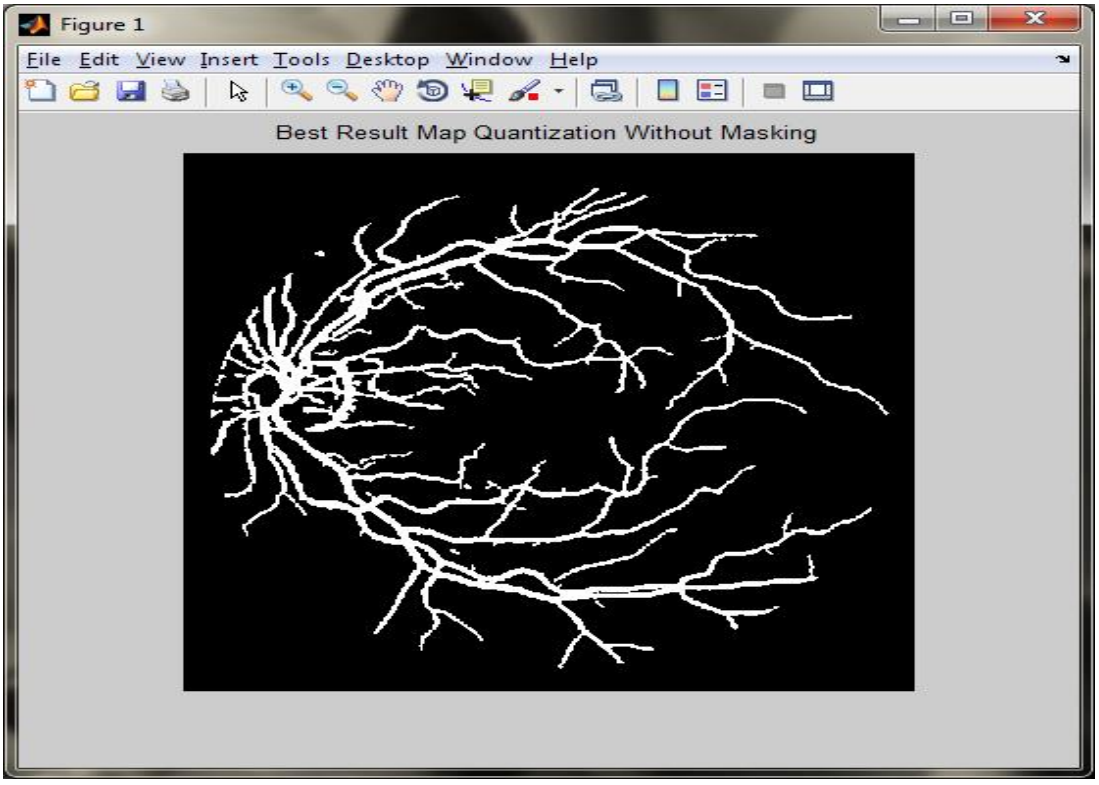

Figure 21 :Detection Of Vessels In Eye Retina Using Line Tracking Algorithm

MORPHOLOGICAL OPERATIONS

The term Morphology denotes a branch of biology that deals with the form and structure of animals and plants. To detect and extract boundaries, skeletons, shapes the morphological operations plays an important role. Here in this morphological image processing the image is traversed with a block called as structural element. When this structural element traversed over the image and is compared with the neighborhood pixels of the image .We can do the morphological operations on the binary images and it results the binary images.

Eroding and Dilating Image Objects

The basic morphological operations, erosion and dilation, produce contrasting results when applied to either grayscale or binary images. Erosion shrinks image objects while dilation expands them.

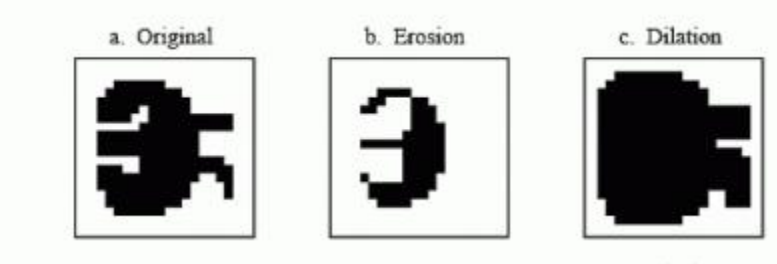

Figure 22 :a)Original b)Erosion c)Dilation

The specific actions of each operation are covered in the following sections.

Characteristics of Erosion

- Erosion generally decreases the sizes of objects and removes small anomalies by subtracting objects with a radius smaller than the structuring element.
- With grayscale images, erosion reduces the brightness (and therefore the size) of bright objects on a dark background by taking the neighborhood minimum when passing the structuring element over the image.
- With binary images, erosion completely removes objects smaller than the structuring element and removes perimeter pixels from larger image objects.
- It shrinks or thins objects in a binary image. The manner and extent of shrinking is controlled by a structuring element.
- The erosion of A by B is the set of all points x such that B, translated by x, is contained in A.

$$A \ominus B = \{z \in E | B_z \subseteq A\}$$

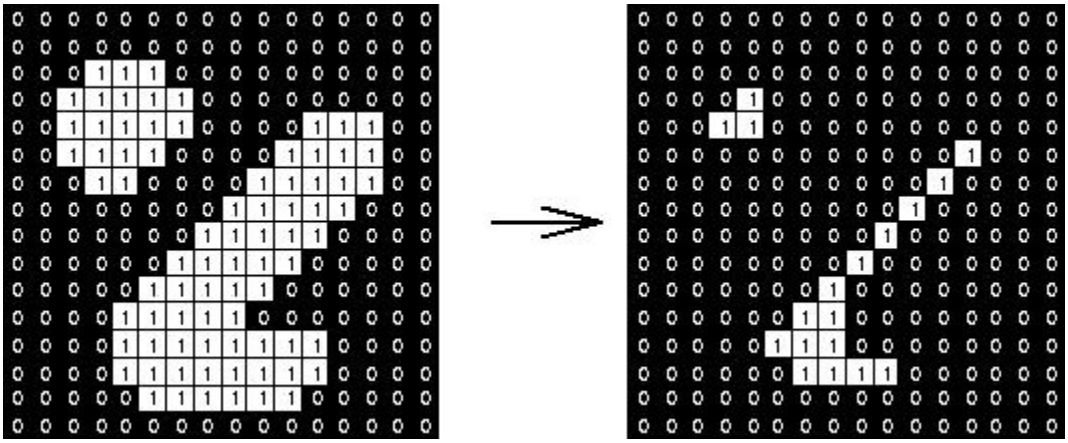

Figure 23 :a)Before erosion b)After erosion

Characteristics of Dilation

- Dilation generally increases the sizes of objects, filling in holes and broken areas, and connecting areas that are separated by spaces smaller than the size of the structuring element.
- With grayscale images, dilation increases the brightness of objects by taking the neighborhood maximum when passing the structuring element over the image.
- With binary images, dilation connects areas that are separated by spaces smaller than the structuring element and adds pixels to the perimeter of each image object.It is an operation that grows or thickens objects in an image.
- The dilation of A by B is defined as the set operation

$$A \oplus B = \{z \in E \mid (B^s)_z \cap A \neq \varnothing\}$$

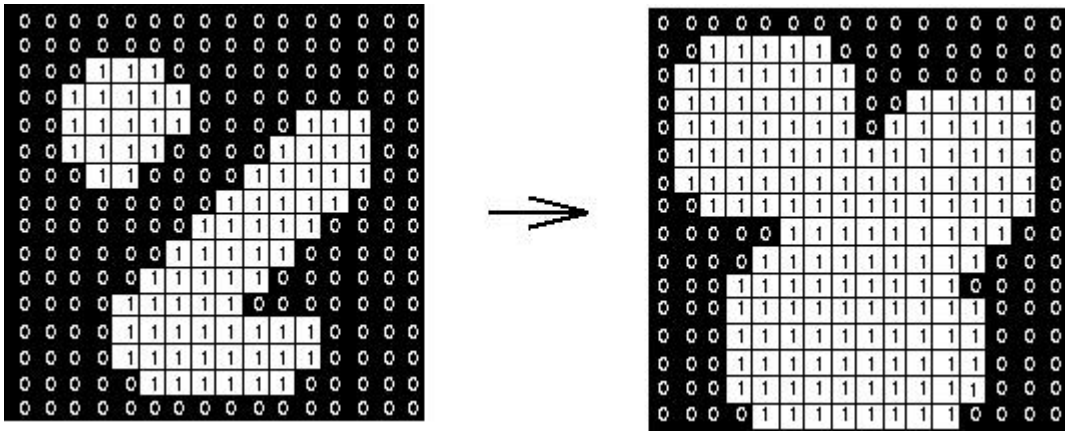

Figure 24 :a)Before dilation b)After dilation

After detecting the blood vessels present in the image by performing the morphological operations ,erosion followed by dilation with the structural element as the disk with radius 5.This post segmentation operations helps in much more clear and accurate output.

5.TESTING

TestCase 1

The application takes input fundus image and then analyses it to retrieve histograms and threshold values.Using that,we get the segmented output.

Step1: The GUI (Graphical User Interface) of the application is shown in figure below

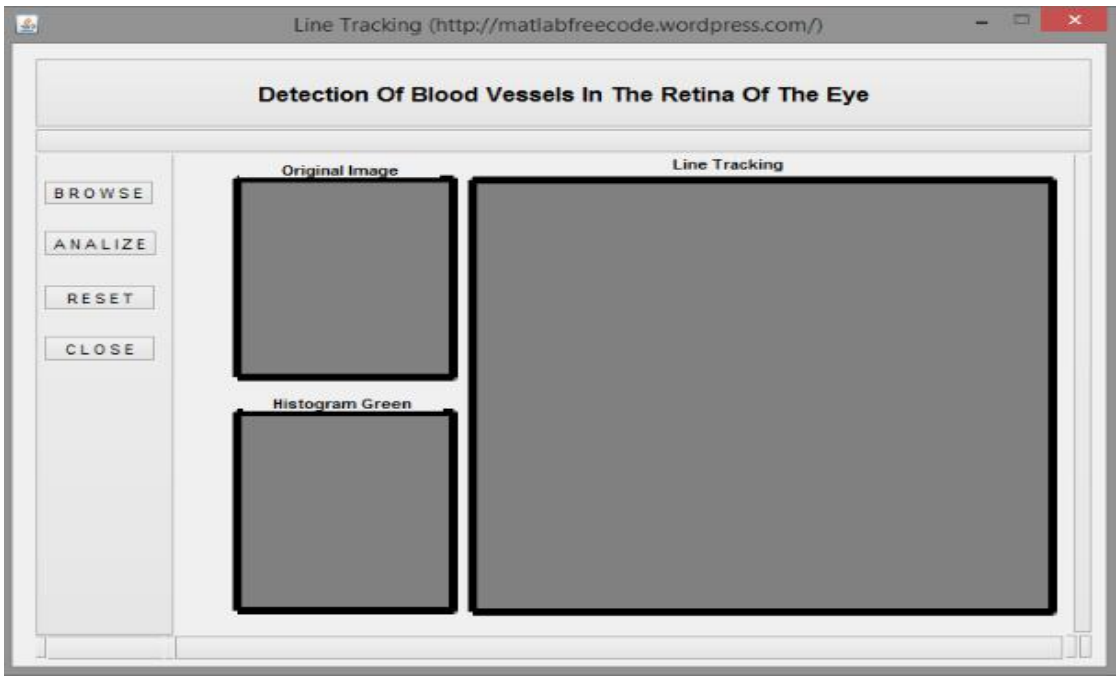

Figure 25:GUI of application

Step2: In figure the image is loaded and analysis is done with the help of a Histogram.

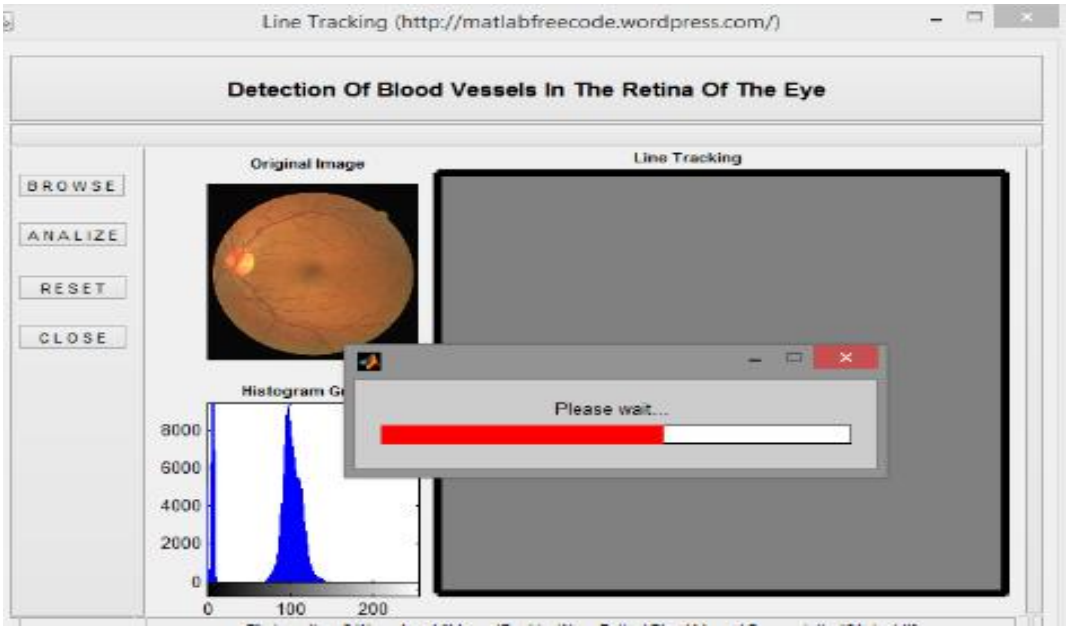

Figure 26:Loading and analysis of Fundus image

Step 3: In figure conversion of gray scale image to binary image with threshold value,which is derived from the values of Histogram.

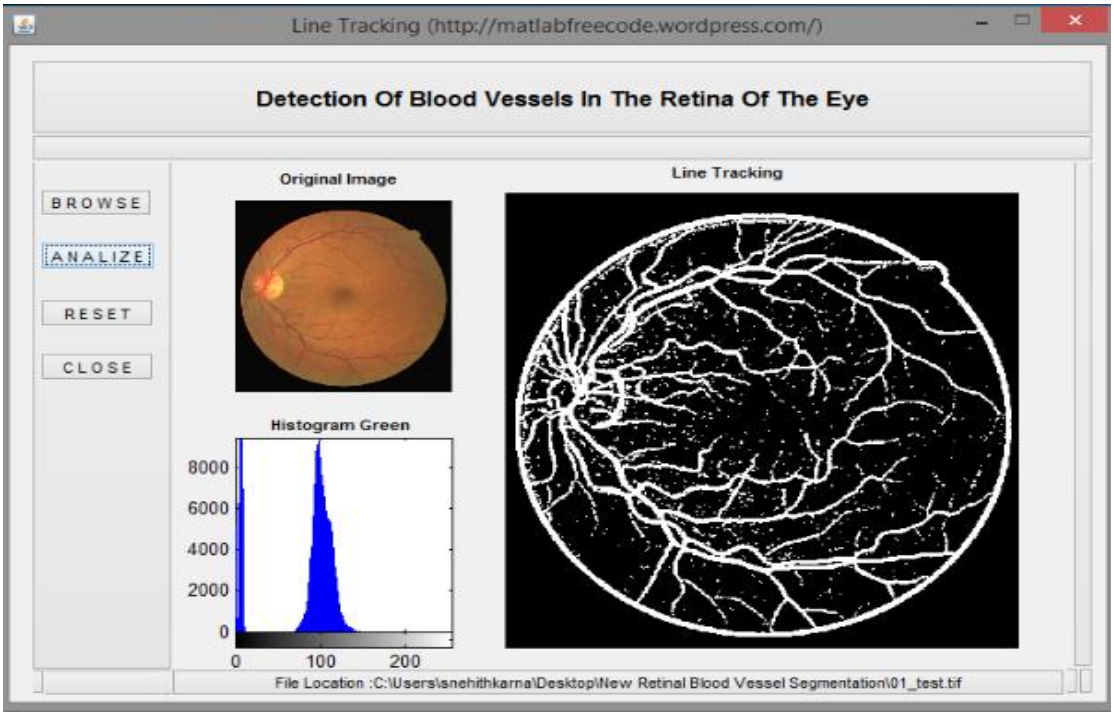

Figure 27:Derivation of Histogram

Step 4: In figure the noise is cleared and output is displayed separately.

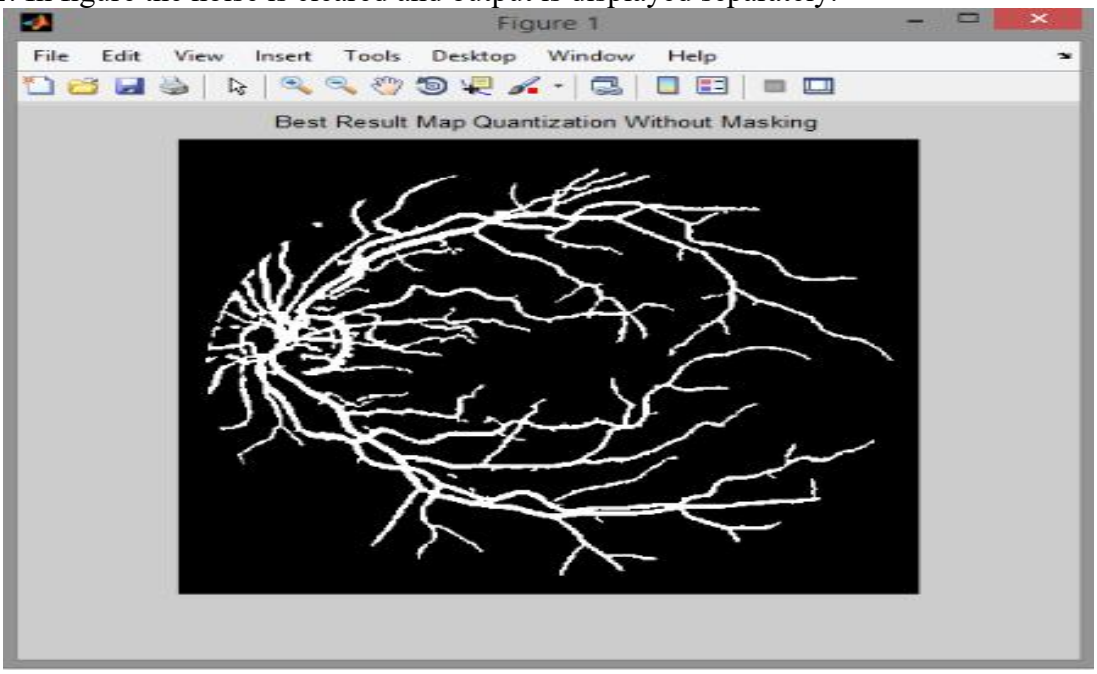

Figure 28: Segmented blood vessels

Test Case 2(Successful): In figure a new fundus image is taken for analysis again.

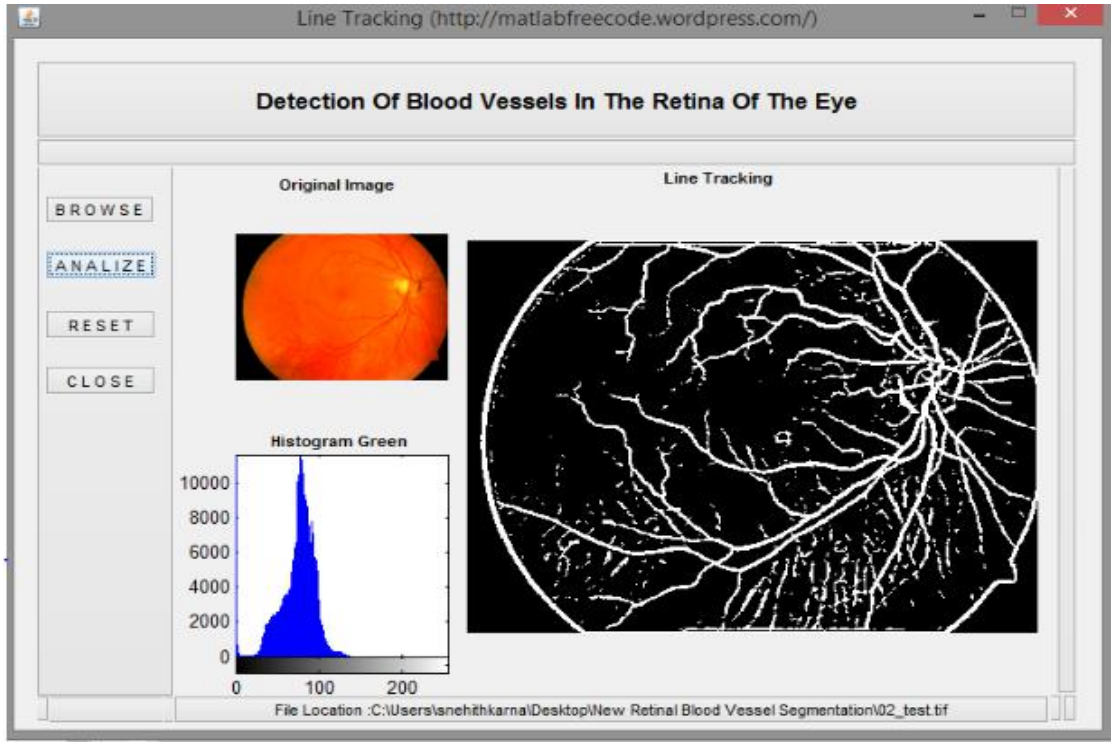

Figure 29:Calculation of Histogram and Detection of blood vessels.

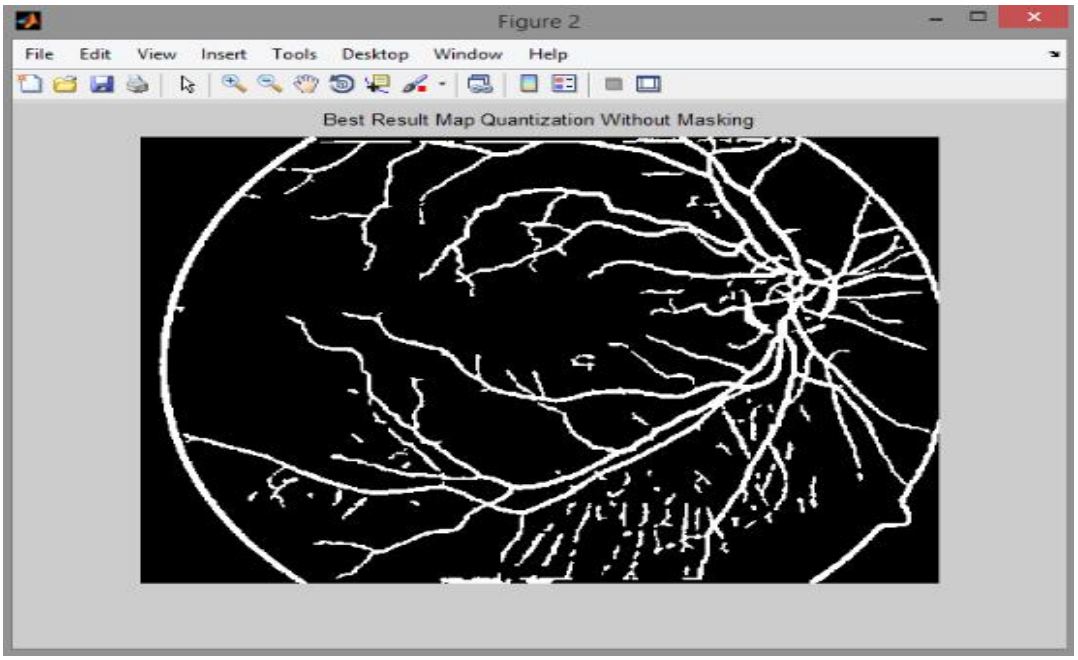

Figure 30:Segmented blood vessels for test case 2

Test Case 3(Unsuccessful):

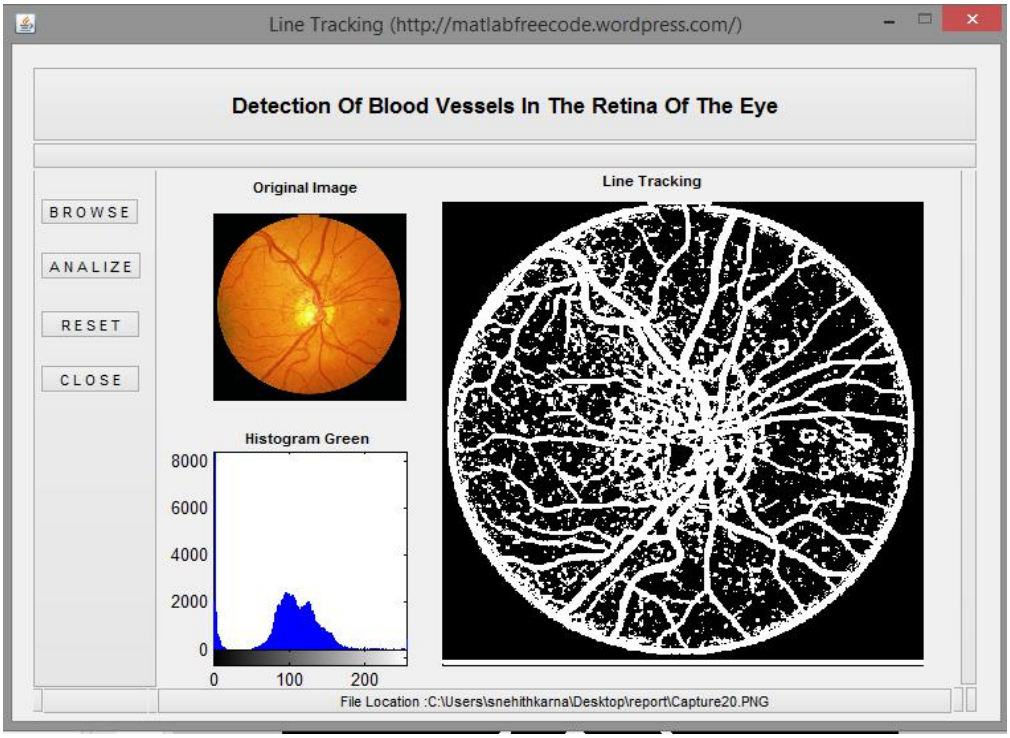

Figure 31:Output for Test Case 3

Test case 4(Successful):

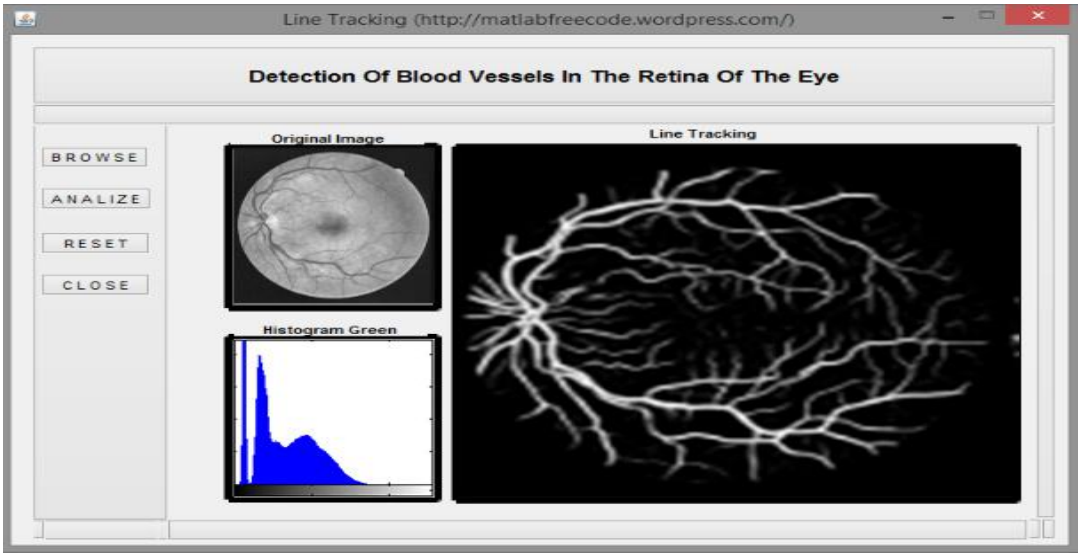

Figure 32:Output for Test case 4

Test case 5(Successful):

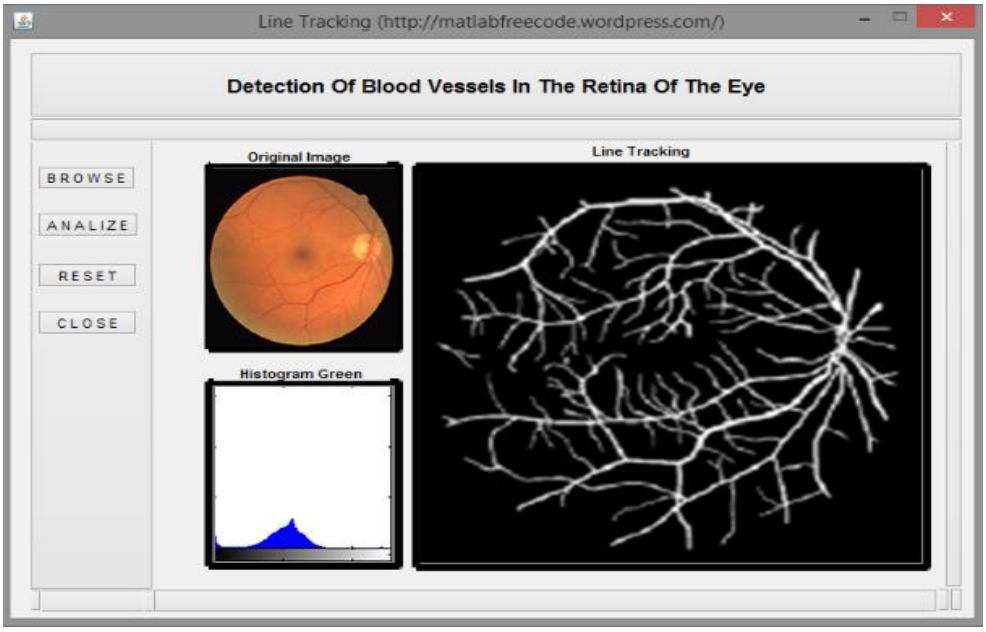

Figure 33:Output for test case 5

Testcase 6(Successful):

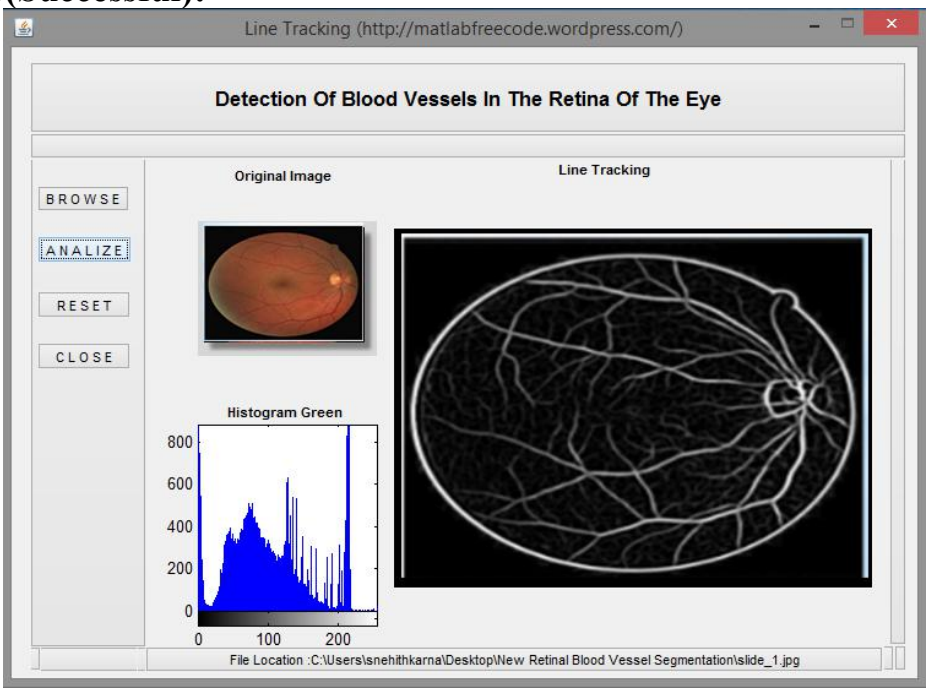

Figure 34:Output for test case 5

CONCLUSION

In this work, imaging is one of the important and essential aspects in the medical field. To analyze and diagnose a disease related to eye,blood vessel segmentation is necessary.

The Retinal image analysis through efficient detection of vessels and exudates for retinal vasculature disorder analysis. In this work, we proposed an efficient and simple algorithm for the detection of blood vessels. It plays important roles in detection of some diseases in early stages, such as diabetes, which can be performed by comparison of the states of retinal blood vessels. . Here, we proposed a new algorithm to detect the retinal blood vessels effectively. Experimental result proves that the blood vessels and exudates can be effectively detected by applying this method on the retinal image.

In this proposed research work, we have explored and developed novel computerised techniques to segment the retinal blood vessels and detect the presence of exudates in retinal images automatically, which serve as a clinical aid for the diagnosis of DR in mass screening programs. This method has proved itself to be efficient for image segmentation and classification by identifying the blood vessels and detecting the exudates in retinal images. The precise extraction of the retinal vascular structure forms the backbone of numerous automated computer aided systems for screening and diagnosis of cardiovascular and ophthalmologic diseases. One of the practical applications of automatic blood vessel segmentation in health care includes screening of DR.

FUTURE ENHANCEMENTS

The future scope is the development of research software applications, which amalgamate various computer assisted retinal image analysis algorithms like vessel segmentation, image registration, detection of pathologies, vessel calibre measurement and crossover detection. The development of integrating all these individual tasks is the new research direction. These kinds of applications can facilitate the progress of studying the correlations of ocular fundus anatomy with retinal diseases. Furthermore, there is a requirement of resolution for screening programs where the evaluation of large image datasets and the association between the retinal experts and healthcare units is essential.

Presently, a set of individual techniques has been proposed which evaluates and computes the retinal vessel morphology. All these procedures are based on the vessel segmentation and form a common scheme that directs to a set of quantitative indices depicting the retinal vessel morphology. These include vessel tortuosity, branching angle, branching coefficient, vessel width, and fractal dimensions. In the future work, all these techniques can be combined together, to develop a software package, that accepts retinal colour fundus image as input and provide a set of numerical indices as output that describes the current status of the vessel morphology.

Even though varied progress could be performed in future, the overall research demonstrated in this thesis reveal a significant progress towards the development of an automated system, which complements the diagnosis of DR that assists the ophthalmologists in their screening programs.

REFERENCES

[1]Pawel Liskowski, Krzysztof Krawiec, Member, Citation information: DOI 10.1109/TMI.2016.2546227, IEEE Transactions on Medical Imaging “Segmenting Retinal Blood Vessels with Deep Neural Networks”.

[2] R. Nekovei and Y. Sun, “Back-propagation network and its configurationfor blood vessel detection in angiograms.” IEEE Transactions on Neural Networks, vol. 6, no. 1, pp. 64–72, 1995. [Online]. Available: http://dblp.uni trier.de/db/journals/ tnn/tnn6.html #NekoveiS95

[3] G. E. Hinton, N. Srivastava, A. Krizhevsky, I. Sutskever, and R. R. Salakhutdinov, “Improving neural networks by preventing co-adaptation of feature detectors,” arXiv preprint arXiv:1207.0580, 2012.

[4] Y. Bengio, “Learning deep architectures for ai,” Foundations and trendsR in Machine Learning, vol. 2, no. 1, pp. 1–127, 2009.

[5] J. Schmidhuber, “Deep learning in neural networks: An overview,”

Neural Networks, vol. 61, pp. 85–117, 2015.

[6] A. Krizhevsky and G. Hinton, “Learning multiple layers of features from tiny images,” Computer Science Department, University of Toronto, Tech. Rep, vol. 1, no. 4, p. 7, 2009.

[7] A. Hyvärinen and E. Oja, “Independent component analysis: algorithms and applications,” Neural networks, vol. 13, no. 4, pp. 411–430, 2000.

[8] A. J. Bell and T. J. Sejnowski, “Edges are the" independent components" of natural scenes,” in NIPS, 1996, pp. 831–837.

[9] A. Dosovitskiy, J. T. Springenberg, M. Riedmiller, and T. Brox, “Discriminative unsupervised feature learning with convolutional neural networks,” in Advances in Neural Information Processing Systems, 2014, pp. 766–774.

[10] X. Jiang and D. Mojon, “Adaptive local thresholding by verificationbased multithreshold probing with application to vessel detection in retinal images,” Pattern Analysis and Machine Intelligence, IEEE Transactions on, vol. 25, no. 1, pp. 131–137, 2003.

[11] A. Osareh and B. Shadgar, “Automatic blood vessel segmentation incolor images of retina,” Iran. J. Sci. Technol. Trans. B: Engineering, vol. 33, no. B2, pp. 191–206, 2009.

[12] “Automatic segmentation of blood vessels from retinal fundus images through image processing and data mining techniques” ,R GEETHARAMANI and LAKSHMI BALASUBRAMANIAN,IEEE Transactions on, vol. 14, no. 1, pp. 101–143, 2007.

[13] A. Hoover, V. Kouznetsova, M. Goldbaum, "Locating blood vessels in retinal images by piece-wise threshold probing of a matched filter response", *IEEE Trans. Med. Imaging*, vol. 19, no. 3, pp. 203-210, 2000.

[14] A.L. Pal, "Extraction of retinal blood vessels from retinal fundus image for computer aided diagnosis", *Proc. of National Joint Conf. on Innovations in Engineering & Technology*, 2015.

[15] M. Niemeijer, J.J. Staal, B. van Ginneken et al., "Comparative study of retinal vessel segmentation methods on a new publicly available database", SPIE Medical Imaging, pp. 648-656, 2004.

[16] Soares J.V., Leandro J.J., Cesar R.M., Jelinek H.F., Cree M.J., (2006): Retinal

vessel segmentation using the 2-D Gabor wavelet and supervised classification. IEEE Transaction of Medical Imaging, vol.25, pp.1214–1222.

[17] Retinal Blood Vessel Segmentation Based on Multi-Scale Deep Learning Ming Li ; Qingbo Yin ; Mingyu Lu-JSPE Medical Imaging,vol56, pp. 778-786, 2004.

[18] IMAGE RESTORATION BASED ON MORPHOLOGICAL OPERATIONS-A.M.Raid, W.M.Khedr,M.A.El-dosuky and Mona Aoud,in NIPS, 1999, pp. 279-299.

[19] Blood Vessel Segmentation and Centerline Tracking Using Local Structure Analysis , Rahul Prasanna Kumar, Fritz Albregtsen , Martin Reimers , Bjørn Edwin ,Thomas Langø and Ole Jakob Elle

[20]Detecting Morphological Filtering of Binary Images -Francesco G. B. De Natale Giulia Boato, vol. 4, no. 1, pp. 12–27, 2010.

APPENDIX A-Abbreviations

RGB - Red Green Blue

MATLAB - MATrix LABoratory

CCD - Central Cloudy Dystrophy

IRMA - IntraRetinal Microvascular Abnormalities

NVE - NeoVascularization Elsewhere

NVD - NeoVascularization of the Disc

DR-Diabetic Retinipathy

APPENDIX-B SOFTWARE INSTALLATION PROCESS

Step 1: Preparation. ...

Step 2: Start the Installer.

Step 3: **Install** With a File **Installation** Key. ...

Step 4: Review the License Agreement. ...

Step 5: Specify the File **Installation** Key. ...

Step 6: Specify the **Installation** Folder. ...

Step 7: Specify Products

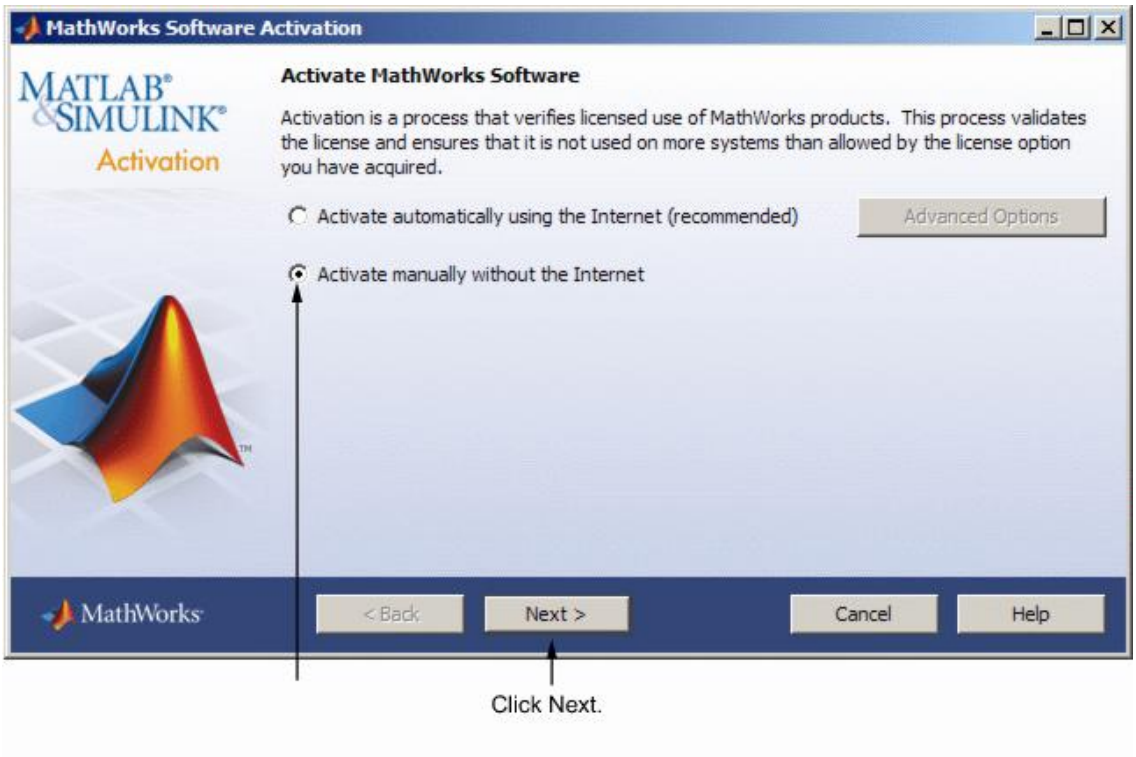

Figure 35:Activation of MATLAB

Figure 36:Accepting the licence agreement

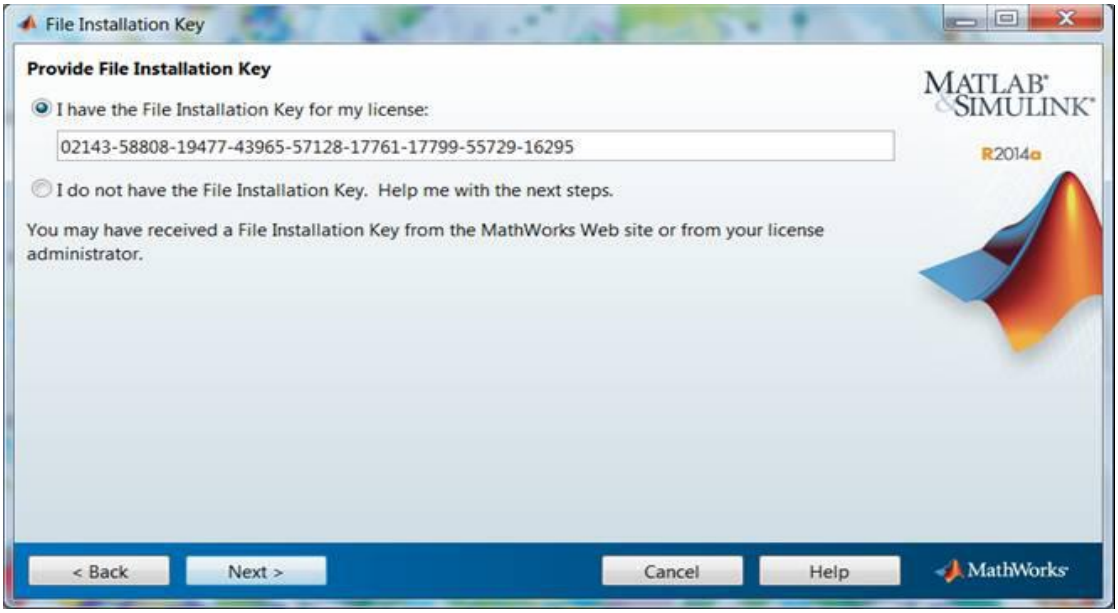

Figure 37:File Installation Key

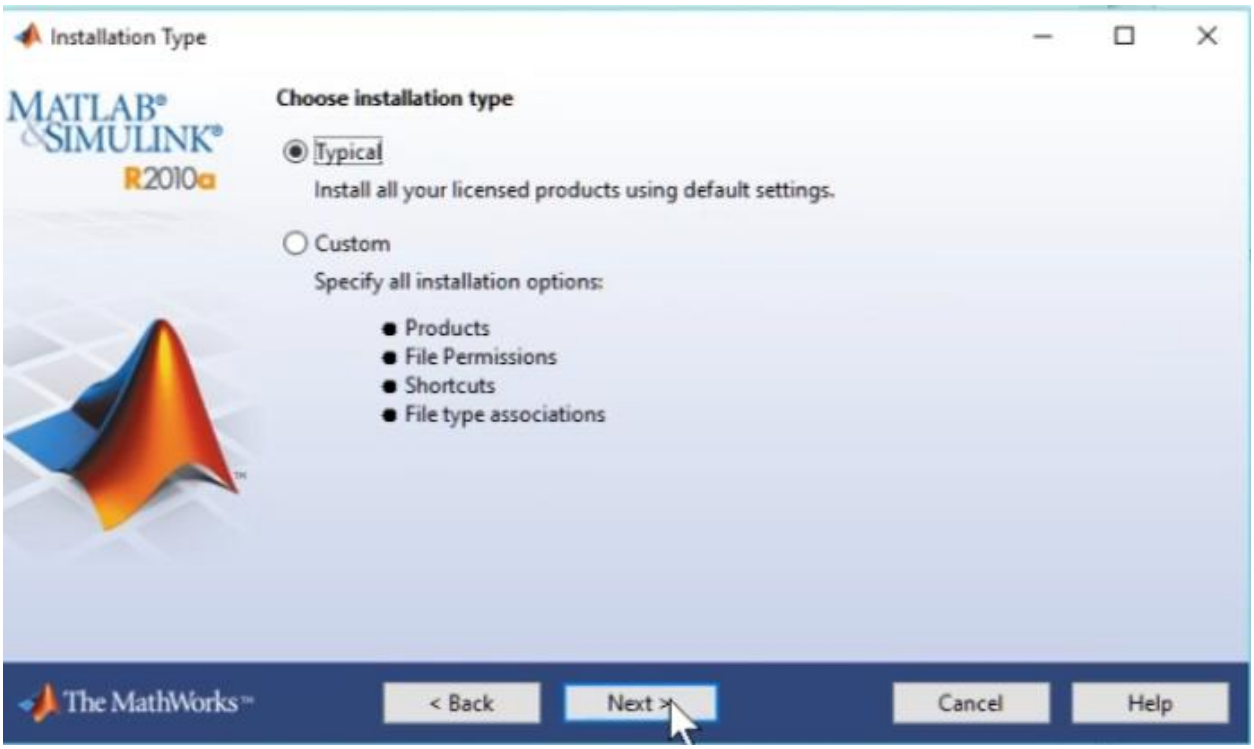

Figure 38:Choosing of installation type

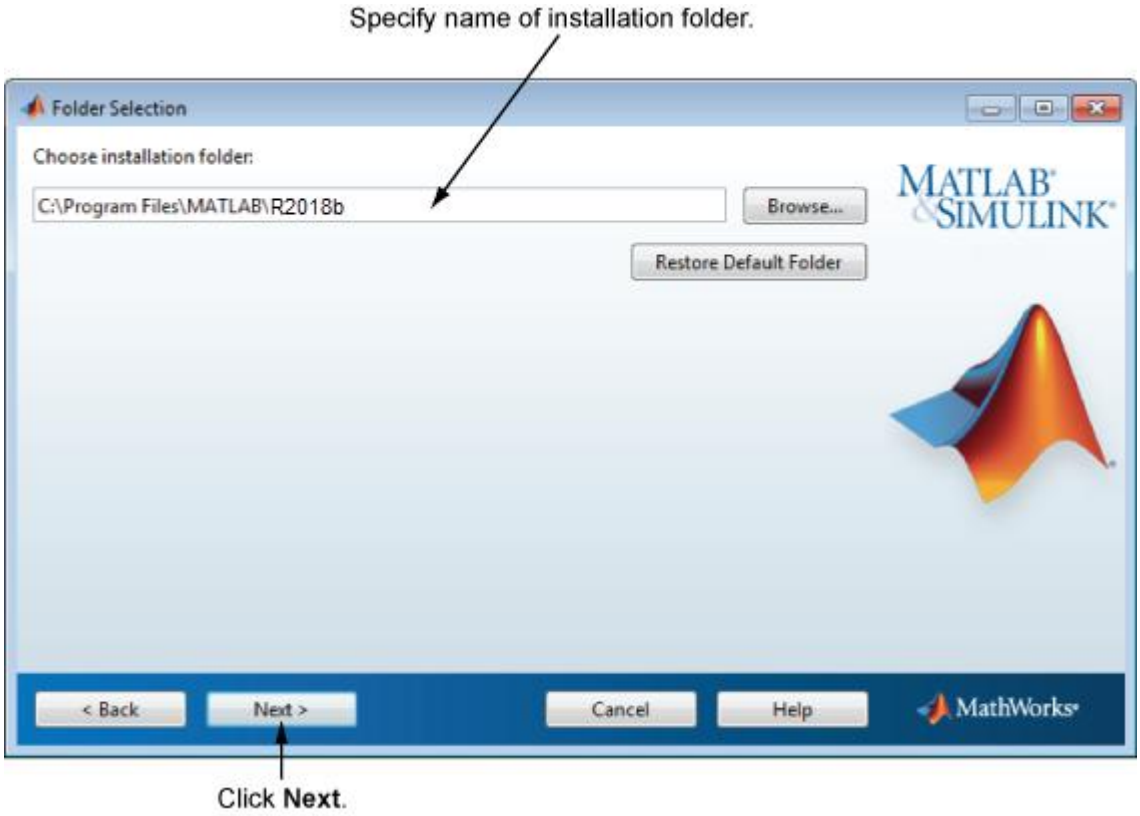

Figure 39:Choosing Installation folder

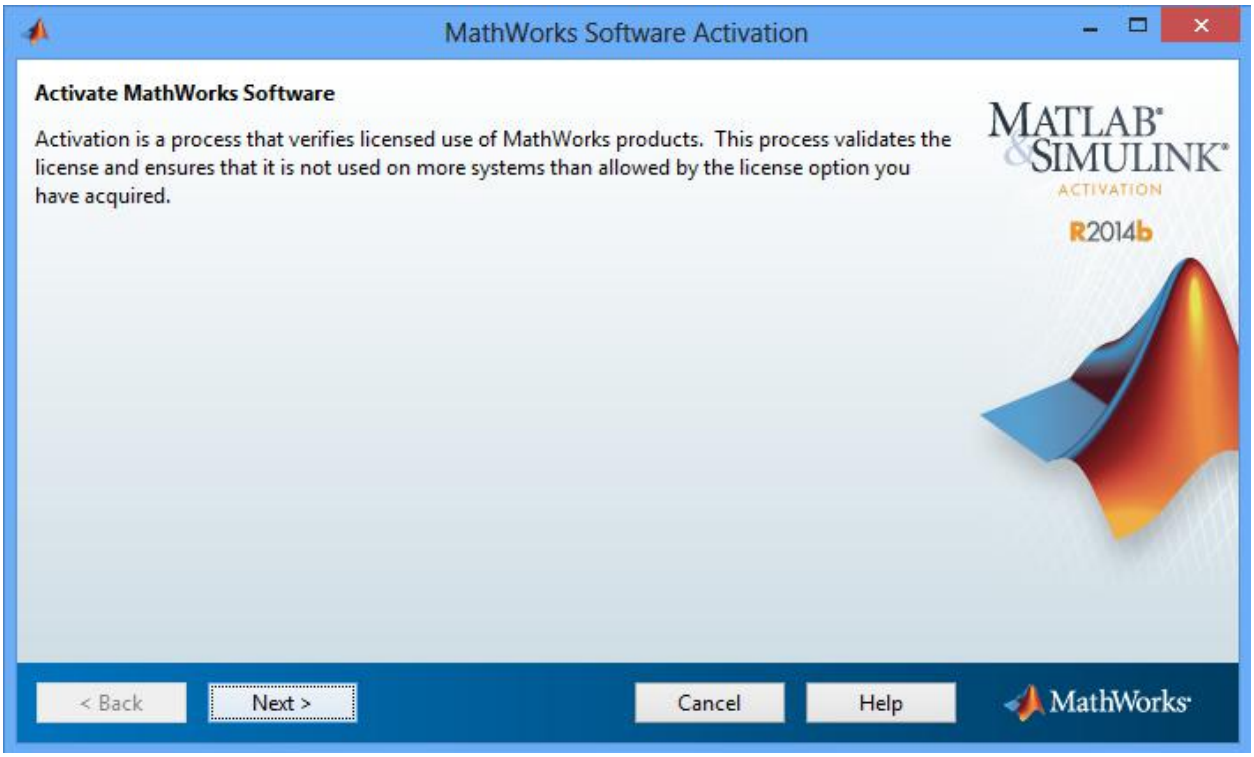

Figure 40:Verification of license

Figure 41:Closing the setup wizard after Installation

APPENDIX C Software Usage Process

We built this Software(Retinal Blood Vessel Segmentation) for Segmentation of retina by using a fundus camera.The following are the steps to understand the usage of the software.

1. Open MATLAB software.
2. To open the user interface(UI), first select the folder which contains MATLAB files and run the "line.m" file.
3. After opening the user interface(UI), press the button named "BROWSE" and then a window is opened which consists of Fundus Images of EYE where you can choose an image(format of an image can be '.jpeg', '.bmp', '.png','.tif').
4. After an image is loaded, then press the button named "ANALYZE" to segment the Fundus image.
5.Then the image is segmented and also a Histogram is also displayed in the bottom left corner.
6.Finally, the segmented image is displayed in a display dialog box.