DIGITAL MICROSCOPE

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APRIL - 2018

Digital Microscope

Idea LabProject

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Declaration

We do hereby declare that the technical project report submitted is original, and is the outcome of the independent investigations/research carried out by us and contains no plagiarism. Their search is leading to the discovery of new facts/ techniques/ correlation of scientific facts already known. This work has not been submitted to or supported by any other University or funding agency.

We do hereby further declare that the text, diagrams or any other material taken from other sources (including but not limited to books, journals, and web) have been acknowledging, referred and cited to the best of our knowledge and understanding.

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IDEALAB

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Annual/Final Report of the work done on the Idea Lab Project.

(Reporttobesubmittedwithin3weeksaftercompletionoftheproject)

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5. Name of Student 2: Vinayak Sharma (15BEC125)
6. Name of Student 3: Kshitij Saxena (15BME062)
7. Name of Mentor: Prof. Tanish H. Zaveri
8. Total Amount Approved: Rs. 31,000/-

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1. <u>Introduction</u>

A Microscope is an instrument used to see objects that are too small to be sensed/ viewed by naked eyes. The word Microscope comes from Greek Word “Micro” meaning “small” and “scope” meaning “to look or to see”. Microscopy is the science of dealing with small objects and structures for the purpose of investigation and discovery. There are a number of microscopes available in the market today in terms of design, features, and techniques used to detect objects under the microscope.

Some of the available microscopes are:

1. Optical Microscope, which uses light to pass through the sample and produce an image onto the lens in the forward direction only.

Figure 1: Normal optical Microscope

2. Fluorescence Microscope uses reflection property of light to detect the objects under the microscope, where the slide is coated with a fluorescent material, which is then bombarded with light rays from a source near eyepiece the reflected light rays are the cause of formation of an image in this technique.

Figure 2: Fluorescence Microscope

3. Electron Microscope uses a beam of accelerated electrons as a source of illumination. Electrons have approx. 100,000 times shorter wavelength than light thus providing higher resolving power and better revelation of structures of smaller objects.

Figure 3: Electron Microscope

4. Digital Microscope is a variation of traditional microscopes using a camera to output images created onto a screen and provide complete details onto hard copy.

Figure 4: Digital Microscope

1.1 Uses of Microscope

Microscopes are used in various scientific fields and for many different purposes. Some of the places where microscopes are used are:

- **Tissue Analysis**

  Histologists use microscopes to study cells and tissues & determine some useful information regarding the generation and degeneration of cells in the body.

- **Examining Forensic Evidence**

  Evidence collected at a crime scene may contain some information that is not directly visible to naked eyes, those are covered under microscopes such as bullet examination and all.

- **Determining the health of an Ecosystem**

  Field biologists used to monitor the health of the particular ecosystem in terms of identification of a number of micro-organisms and their diversity.

- **Studying atomic structures**

  Very powerful microscopes such as atomic force microscopes have allowed scientists to go in depth of atomic studies and study surfaces of atoms.

1.2 Objective

The aim of this project is to develop a Low-cost digital Microscope for the purpose of reducing the time lag between the process of sample collection and report generation. Further due to its low cost and mobility feature it can also be used for surveying and testing at remote areas where Medical facilities in terms of pathological labs and other testing areas are not viable and available.

1.3 Scope of Work

In the past decade or so, there have been many advances in the field of Microscopy but the problem still persists in the remote areas and the rural areas of the country. People suffer due to long wait time for report generation procedures. Digital Microscopes were introduced to reduce eye strain of the Lab technicians, but we changed the definition of the digital microscope by using them as a way to automate the process of microscopy and report generation. This would be of great help to the people who can't afford or are not been able to avail the services of pathologists.

2. Literature Survey

In today's world, almost all the pathological tests are done under a microscope by a Lab technician or doctor itself in some cases. In big labs where the work-load is around 1000-1500 samples per day require LTs to work for several hours continuously on the microscope which may result in a backache, eye strain, and other mental and physical problems. Further, these facilities are only available in the urban areas and not in rural areas due to lack of qualified personnel. As a result, people from rural areas have to come a long way down to some lab in an urban area to get the test done.

In the medical analysis, blood cell count plays a major role in depicting the condition of a patient suffering from a disease. It has been found out that a variation in blood cell count is the result of many diseases in human body. The complete blood count (CBC) is extensively used for detecting infections, allergies, anemia, leukemia etc. in the human body. In order to get CBC test done, a blood sample is taken and blood film is stained which is then imaged using microscopes by LTs. Currently, this process is done manually in order to determine/identify disorders in the sample collected, under a microscope. As we all know the factor of fatigue plays a major role in the accuracy and perfection of report generation process, in every scenario and here also it plays a vital role. The process here is time-consuming and may lead to undesirable human errors. The following are the cellular elements of Blood:

- White Blood Cells or leukocytes:

  WBCs are an important part of human body's immune system, which helps us to fight against certain bacteria, viruses and other infectious diseases. The normal range of leukocytes in blood is around 5000-7000/mm^3. The low count indicates risk of infection in the body while High count indicates existing infection, leukemia, or tissue damage.

- Red Blood Cells:

  RBCs are the most important and numerous blood cells in the body which gain color red from a protein called as hemoglobin, which also carries out the main function of RBCs i.e. carrying oxygen from lungs to each and every part of the body. Low RBC count can be due to severe diseases.

- Platelets:

  Platelets are the cell fragment that circulates in the blood and helps in clotting blood from wounds. A low count can cause a person to bleed out and a high count of platelets increases the risk of blood clots in blood vessels known as thrombosis.

This project currently deals with one of the basic microscopy application i.e. blood count and blood analysis. For this, we have referred several books used by professional LTs such as “Medical Laboratory Technology by Ramnik Sood” & “Notes on Clinical Lab Techniques by K.M. Samuel”.

2.1 Traditional Method

The slide is prepared by Lab Technician using grease-free slides. The blood drop is spread with the help of a second slide. As soon as blood has spread entirely across the end of the spreader slide, with a quick movement spreader is pushed towards another end of the under the slide. Blood film should not be too thin or too thick. Blood slide is stained using Leishman’s method, or Giemsa’s stain method, or Jaswanth Singh & Bhattacharji stain method, or Peroxide Reaction (Sato and Seiya's Method).

Each one of the methods is specialized for one or the other tests and they give some or the other way to analyze one of the specific components in the blood slide such as Giemsa’s stain method is used as best stain for malarial and other blood parasites as well as is also a satisfactory staining method for routine blood stain.

After staining a general examination of the film is done to see if RBCs, WBCs, and Platelets are present in the normal proportion or not and apart from that, noticing that if any abnormal or immature cells are there among RBCs, and WBCs and shape and size of red cells for the inclusion or parasites.

If everything goes correctly then, the slides go for Differential Leucocytes count (DLC). Leucocytes are WBCs in common terms. About 200-500 cells are counted and the average count is taken per cent. According to experts, the best place to get the DLC is an ideal thickness which is the third part of blood film from the head of the smear.

The conventional device used by pathologists for CBC test is known as Haemocytometer. It has a thick glass microscope slide with rectangular indentation creating chambers of certain dimensions, which are etched with a grid of perpendicular lines. To count blood cells, LT/physician must view hemocytometer through a microscope and manually count the blood cells using hand tally counter, or some other conventional method.

Drawbacks of the manual method:

- Time-consuming and laborious causes fatigue.
- Counting overlapped cells are a major problem.
- Consistent results are not guaranteed by visual inspection.

To overcome all these drawbacks of manual counting and report generation "Automated methods" come into the market. Here the CBC is done using automated analyzer, blood is well mixed and not shaken & then placed on a rack in the analyzer. The instrument has a number of distinct chambers to analyze different elements in blood. The results are then either printed out directly of sent to a computer for review.

Drawbacks of automated methods:

- High cost
- Irregularities and variations in shape and size cannot be detected.

Using this technique traditionally blood count and analysis is done. But as one can see all this require a whole lot of human effort and intelligence, and without trained personnel, one cannot get good results for the tests and this is the reason which is holding back both Government and private organizations to provide a good and easy access to pathological facilities.

2.2 Proposed Method using Digital Microscopes

The slide preparation part is completely same as the traditional method, but the analysis part is different in terms of approach and speed. The slide is prepared and put under a digital microscope which has a smartphone attached to its eyepiece. The smartphone camera captures the image generated by eyepiece and produces a good high-resolution image that can be used for the purpose for image processing and producing results of the tests using preliminary image processing algorithms and basic transformations.

The image captured is uploaded to a server or high-performance machine where the image is used to find out the number of RBCs and WBCs in our project using the best of the 3 component images of an image viz. R, G & B component images. These images are further passed through different thresholding methods to correctly identify shapes such as circles and ellipses using Hough circle transformation and finally counting a number of circles detected in the range defined for the radius which determines whether detected circles denote RBCs or WBCs.

The method is explained using a flow chart as follows:

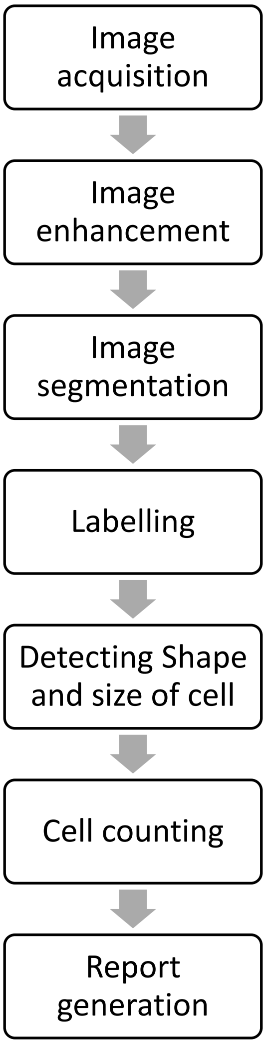

Figure 5: Process Flow of the report generation procedure

The process of CBC using image processing is done using a specific method or we can say by following a specific flow of methods. The process starts with image acquisition from smart-phone camera to a computer system or onto a centralized server. For better segmentation and extracting most of the information, image enhancement is done and this is done by first separating the R G & B components of the image and selecting the Green plane, which according to several experimentations contains most of the information as compared to Red and Blue plane. Then to enhance the image, histogram equalization is used to adjust its contrast and get a more suitable image to work on.

Next step is to detect all the cells in the image; this process requires a basic image-processing algorithm also known as image segmentation. As the shape we need to detect is almost a circle, we use circular Hough transform, which reduces the work of programmer and draws circles around the detected cells. Here even the overlapped cells are also detected.

Finally, the program counts the number of cells in the image.

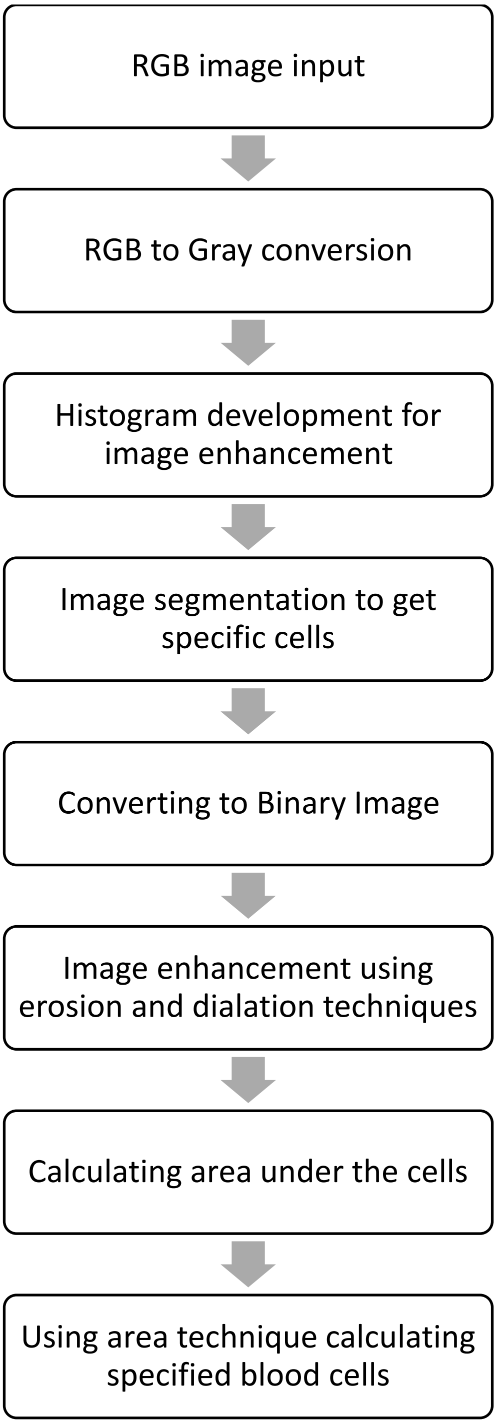

Figure 6: Algorithm to detect cells in given image

2.3 Test Results on the proposed algorithm

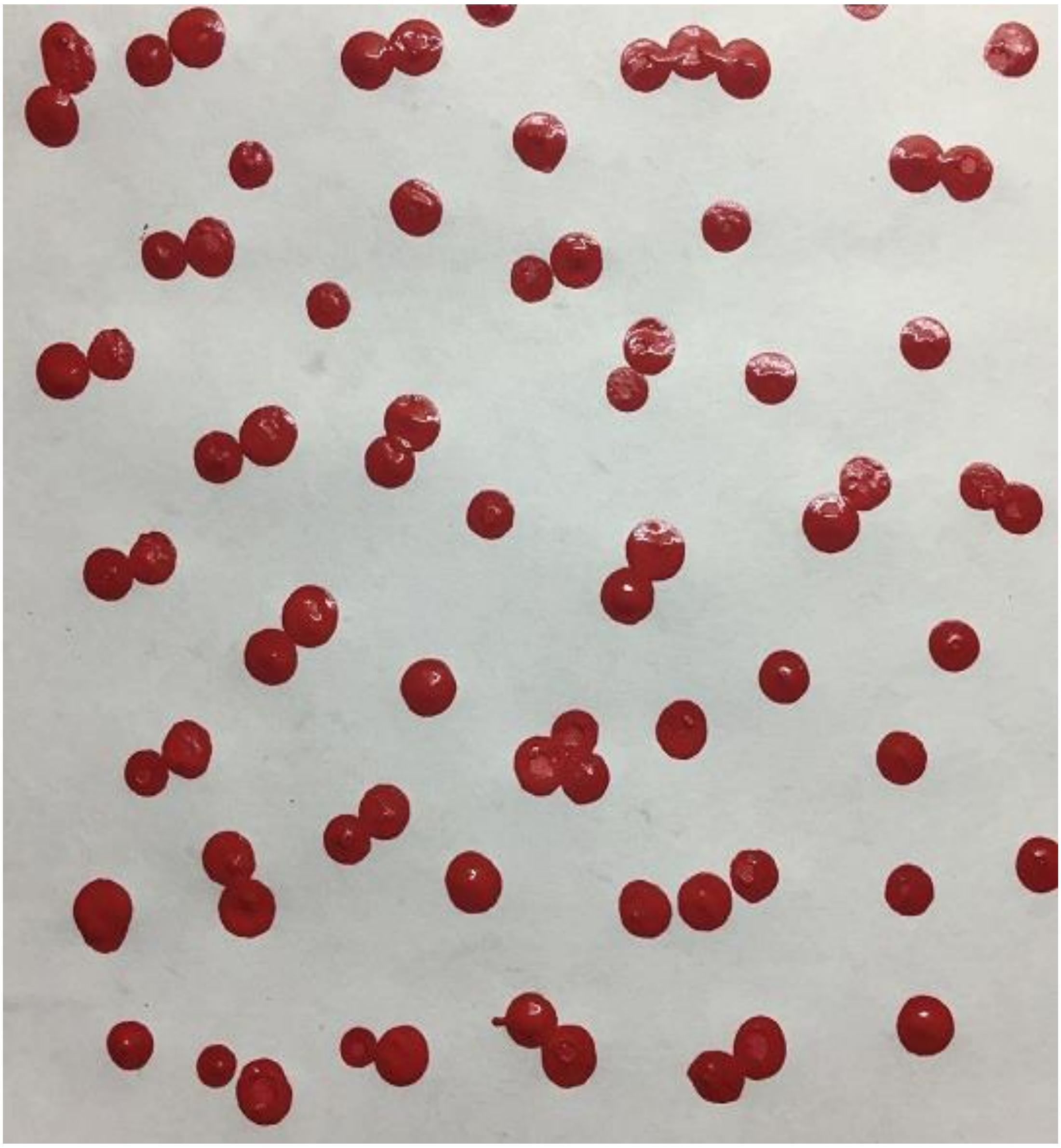

Figure 7: Input image to the system with different orientations of cells

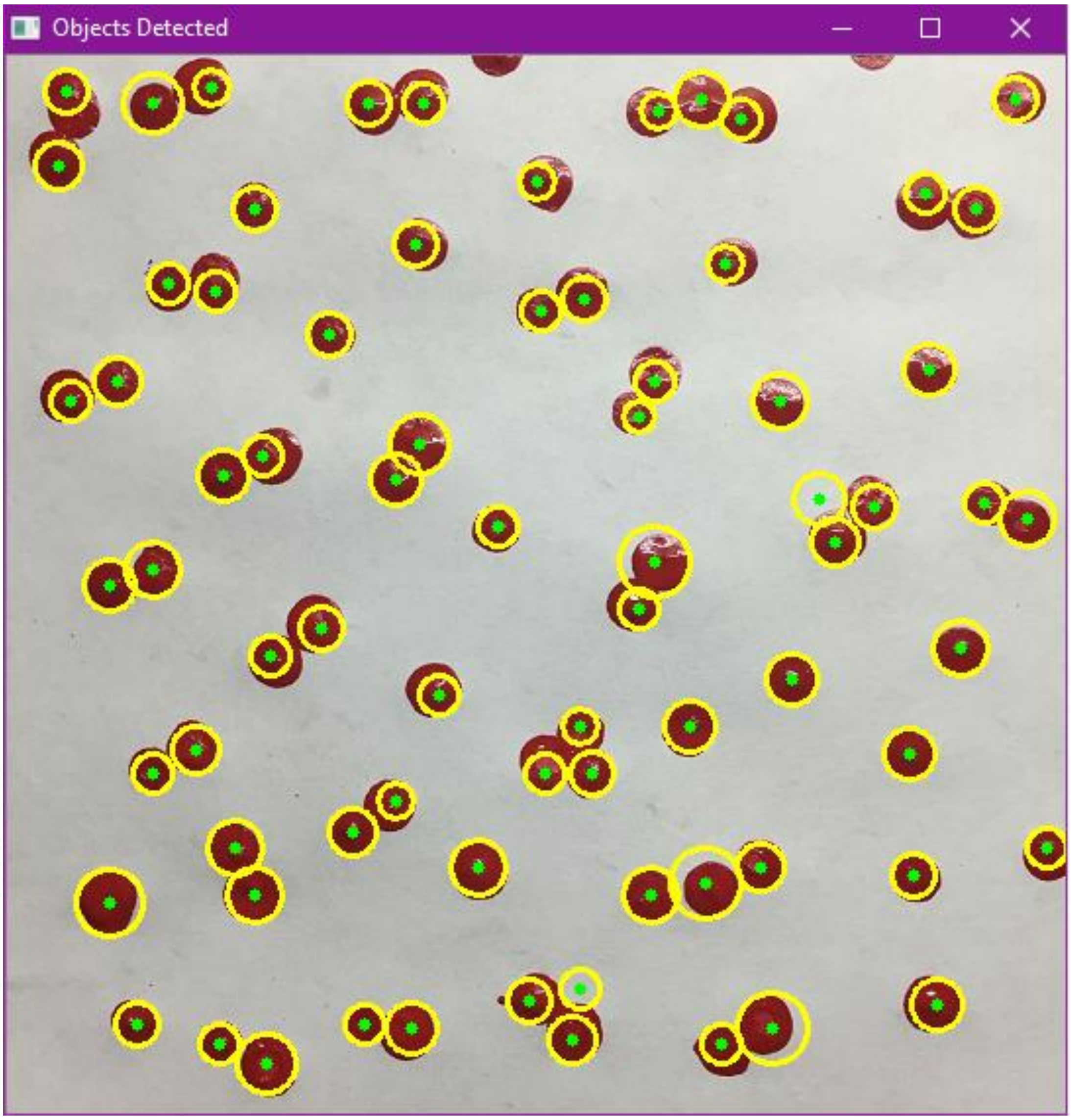

Figure 8: Detected cells in output image

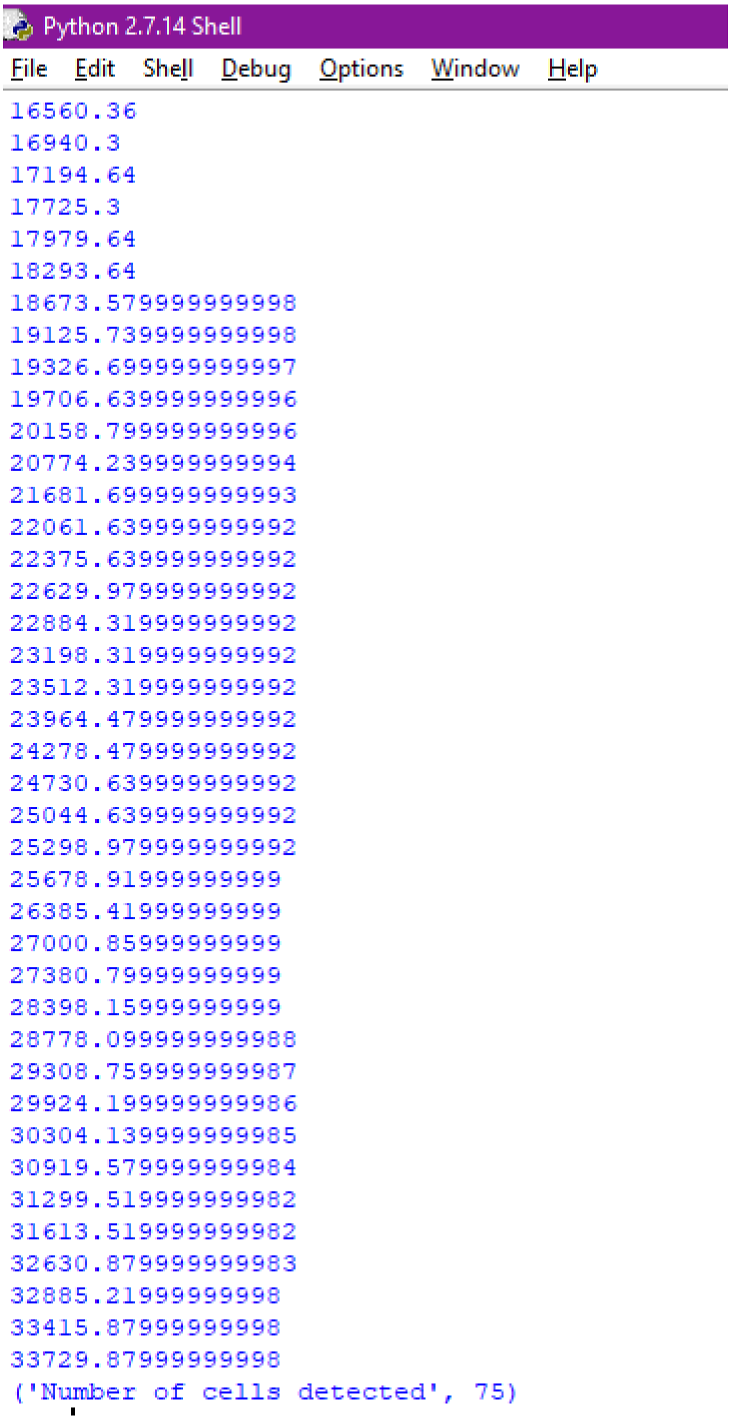

Figure 9: Detected cell number and total area covered by the cells

As can be seen from the results of one of the test data, that the current algorithm is working fine on detecting circles and counting the number of cells present in the image. The same algorithm and code can be modified a bit to detect other cells in blood smear and with shape recognition, we can even go on to next stage of detecting different microorganisms in the smear to detect other diseases as well.

3. Hardware Design

The microscope is designed such that it ensures small size factor, ability to work with off the shelf optics and the ability to accommodate any smartphone available in the market. The design is based on the commercially available microscope to ensure proper integration with the lenses that are available in the market.

Design Highlights

- The design uses prisms for bending the optical path of the light to ensure small form factor of the setup.
- The tube length is based on the commercially available microscopes in order to use the commonly available lens and other optical components.
- The design uses a 3 degree of freedom mobile phone holder to ensure that any smartphone available in the market can be used with the setup.
- All the parts are designed to be easily dismantledand assembled to provide mobility and ease in transportation.
- The slide and lock design ensure ease of maintenance and any part can be easily changed in case of its failure.
- The setup has its own power source and high-efficiency LEDs for the light source and can be used in remote locations.

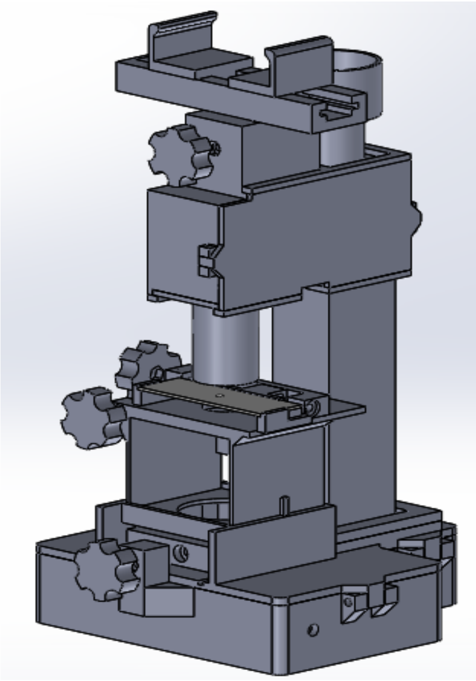

Figure 10: SolidWorks design part view

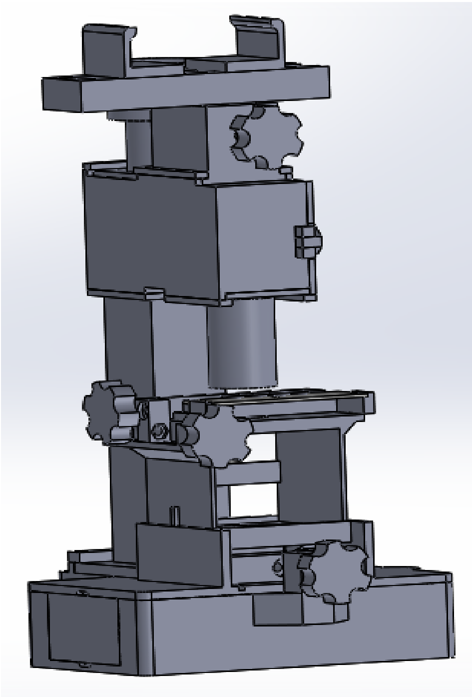

Figure 11: Solid Works part view

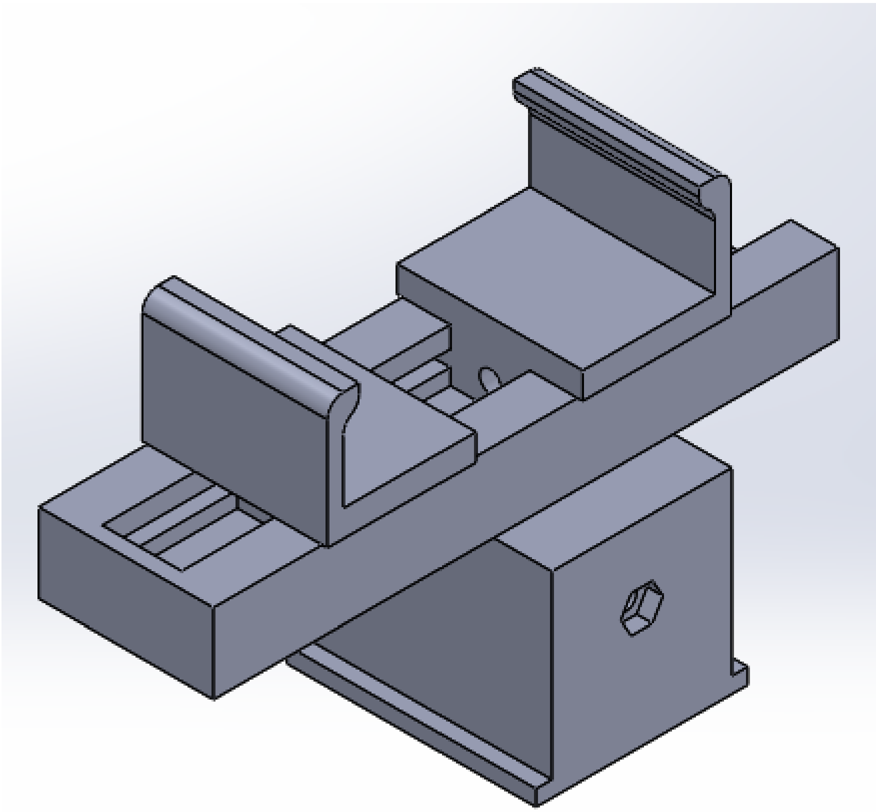

Figure 12: Phone mounting part design view

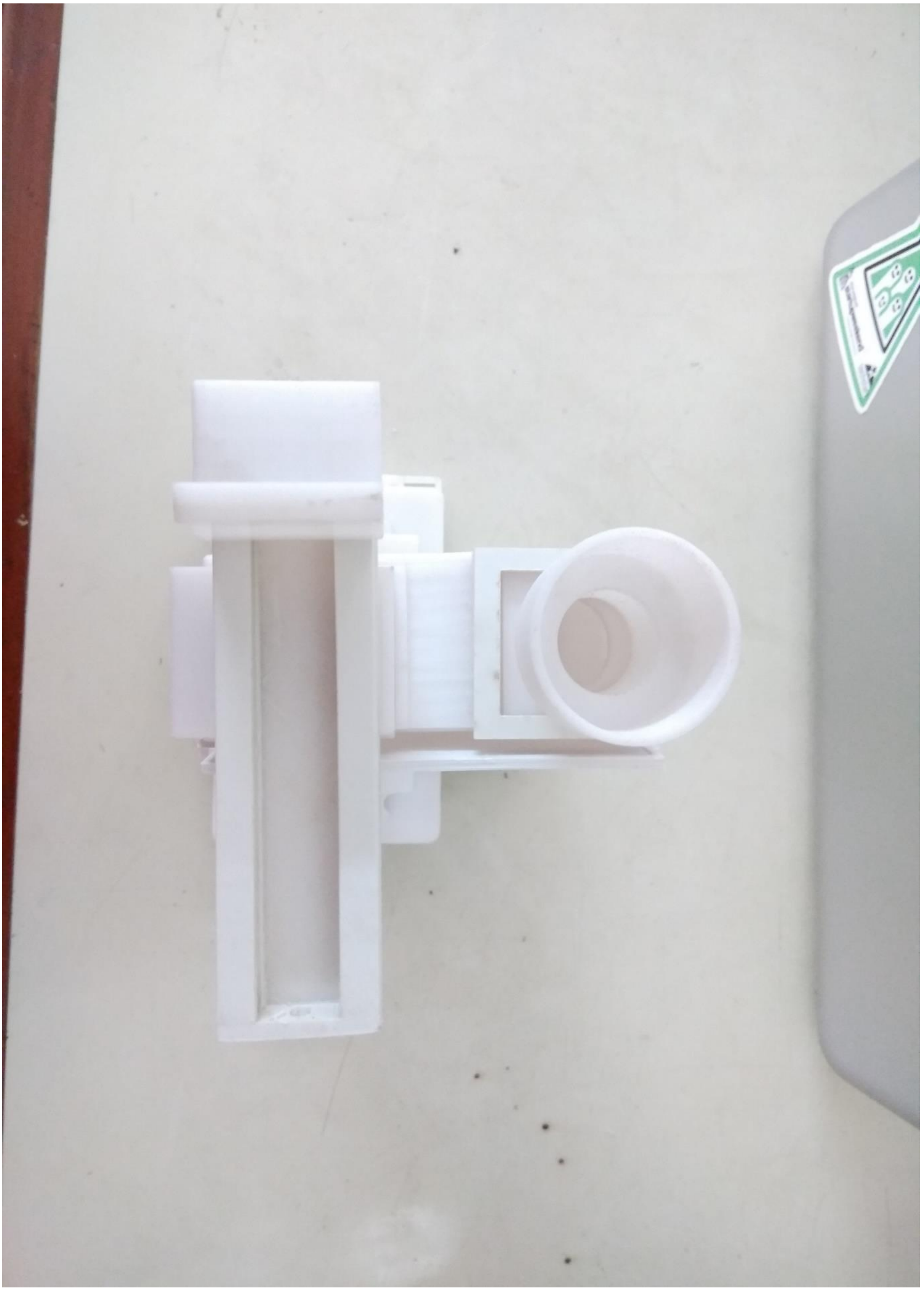

Figure 13: Top view of 3-D printed Model of Microscope

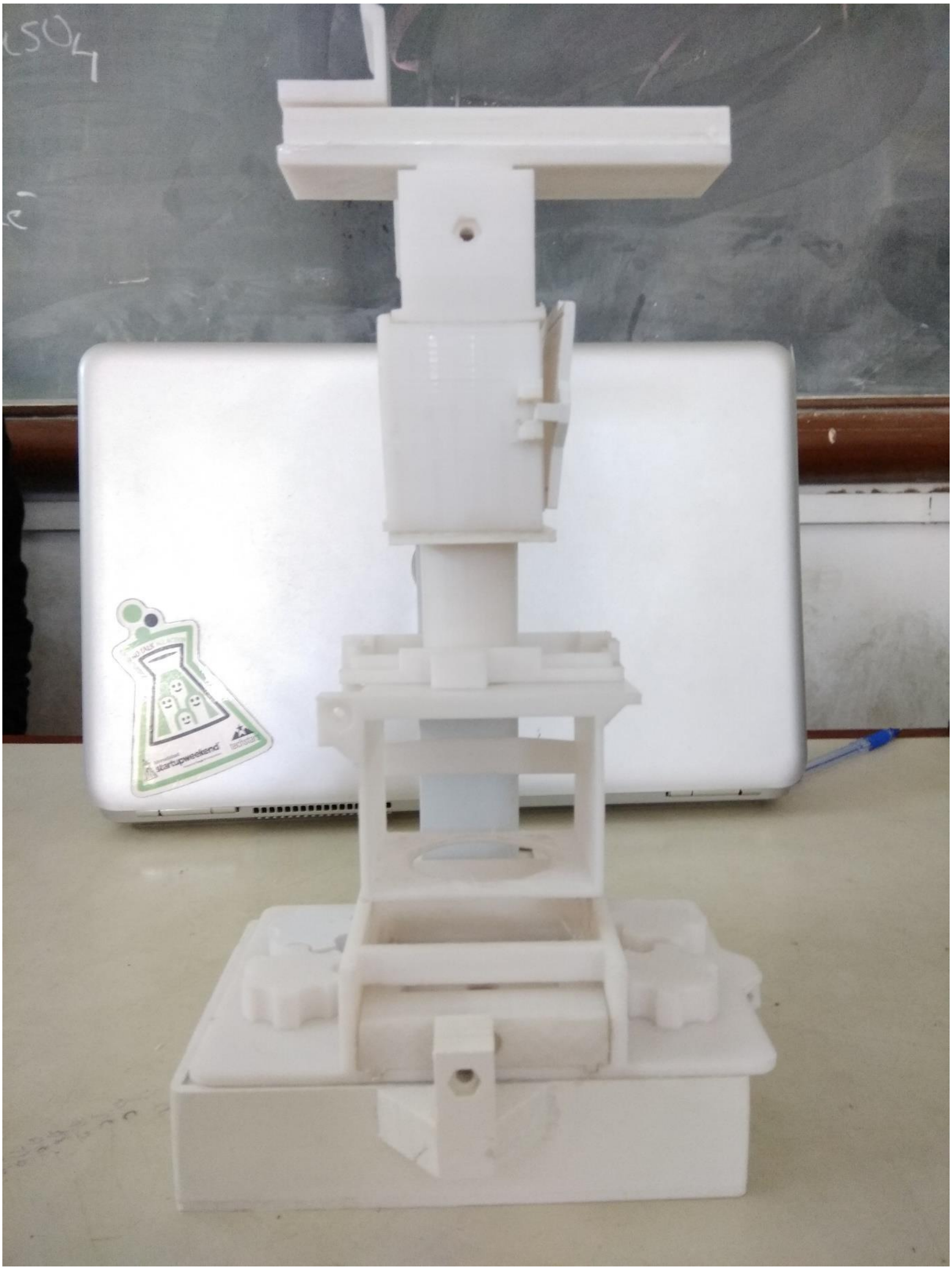

Figure 14: Front view of 3-D printed Microscope

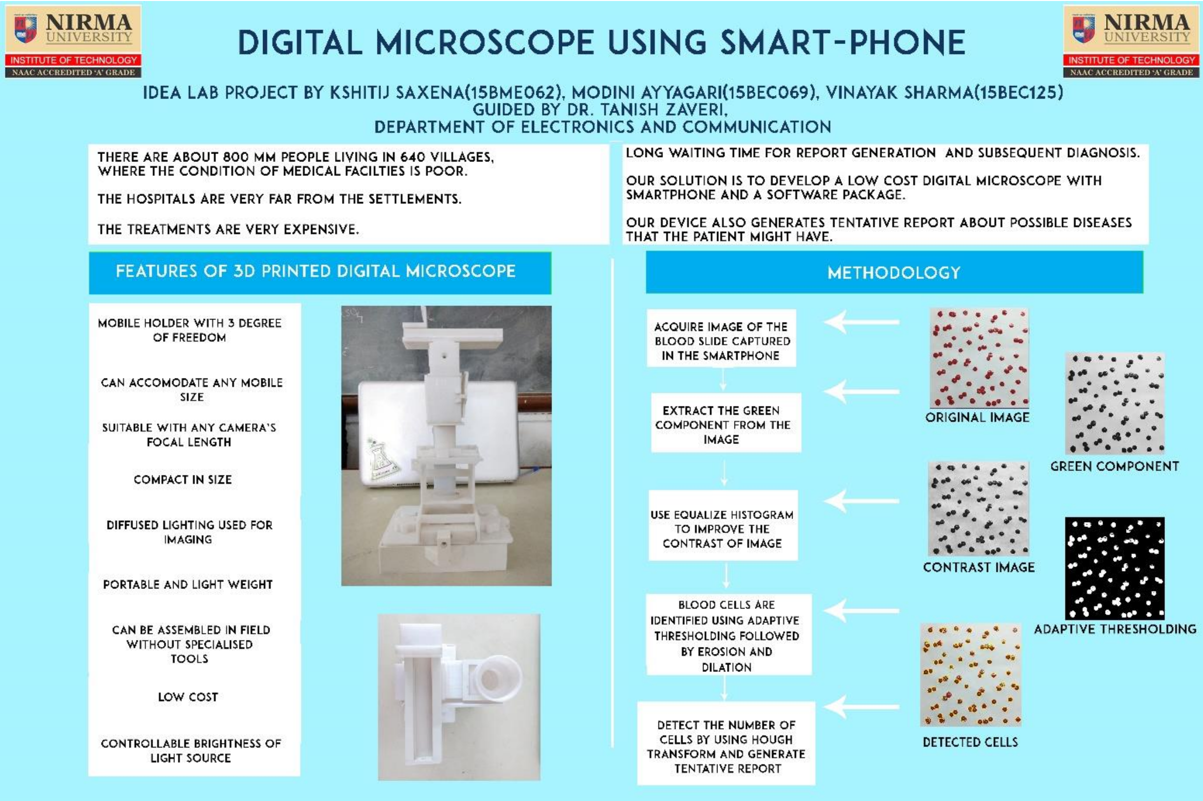

Figure 15: Final Poster showing gist of the project Idea & process flow

4. Conclusion

The demands of pathological tests are increasing day by day and the time to get the reports increases with complexity of test. Thus the requirement of digital microscopes that can reduce work of human and increase the efficiency of the procedure are required. Not only that, using digital microscopes we can get results in very less time as compared to conventional methods. The use of image processing in such cases provided with a way to use the computation power of processors and GPUs to be used for the survival of mankind and produce something big rather than to be used only for gaming purposes.

5. References

[1]. Ramnik Sood "Medical laboratory Tevhnology Methods and Interpretations" by JAYPEE BROTHERS

[2]. K.M. Samuel "Notes on Clinical Lab Techniques "

[3]. https://en.wikipedia.org/wiki/Digital_microscope

[4]. "Microscopic images dataset for automation of RBCs counting" research paper by Sherif Abbas.

[5]. Practical OpenCV book by Samarth Brahmbhatt.