Detection Of White Blood Cells Nuclei Using Automatic Segmentation

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Abstract - In clinical and laboratory tests white blood cell recognition and also classification of leukocytes into various distinct subtypes is very important and useful. In most of the cases the nucleus features are more adequate to identify the type of the cell, the traditional morphology test to look at the cell under the microscope done by a hematology expert is a time consuming and tedious job, beside this to do the test the medical instrument which is used are very expensive and costly and this may not be exist in all the hospitals and clinics. The main aim of this research is to use image processing techniques in order to automate the process of detection of white blood cells and classification of leukocytes. The inspection procedure of blood smear get much easier, simpler and faster and also the amount of data can be analyzed by using an automatic image segmentation system by such a clinician that they handled more than they normally handled. In white blood cell segmentation only the very most crucial step of such systems is present. In this research we focused mainly on white blood cell nucleus segmentation that they are used to separate the nucleus from the whole cell body by using a combination of automatic contrast stretching followed by image arithmetic operation, global threshold and minimum filter.

Keywords: - Blood cell, Leucocyte, Dataset, Segmentation, MATLAB Code, WBC,RBC.

1. INTRODUCTION

Blood disorder is the one that consider as the most dangerous of all the diseases which can lead to death. Many of this type of blood diseases they are related to white blood cells they are called as Leukemia. Under the microscope we observe different types of blood smear that gives invaluable qualitative and quantitative information, this helps to diagnose a variety of diseases such as Leukemia. Blood tests are very important to know the occurrence of diseases and also can investigate many diseases such as cancer, HIV/AIDS, diabetes, anemia, and coronary heart disease and so on [1]. Therefore the blood tests are given high importance for diagnosis of many and more diseases and also investigates functions of body organs like kidney, liver, thyroid, and heart. Manual microscopic examination is must and should when there is a suspicion of abnormality in the blood samples but it is tedious job, time consuming, and subjective too. If the visual sample inspection is automated then to increase productivity and to reduce costs it will help the pathologists.

The automation process is done for white blood cells that includes various techniques such as image acquisition, image processing, segmentation, feature extraction, and classification. In this paper Segmentation is considered as the most important technique and critical step in the automation process as that it affects the rest of all the following steps . We proposed an efficient technique for white Blood cells (WBC) nuclei automatic segmentation. The algorithm proposed by Madhloom is modified to account for more and more general situations.The proposed modifies of madhloom that is to reduce dependence on the image initial contrast. This contrast dependence on images may leads to the capturing of all the objects which have the same gray-level as well as the WBCs. To overcome this type of disadvantage we proposed to use some type of constraints to eliminate the false and negative objects.

Blood is a red color liquid (that is vital of life) which is composed of an isotonic fluid (plasma) in which various cells (hemocytes) are suspended. Here there are three major groups of these types of cells, all that types of blood cells are manufactured in the bone marrow, which are growing from a cell which is known to be hematopoietic stem cell .

The red blood cells (also called as Erythrocytes) which contain hemoglobin that carry the oxygen to the tissues, platelets (also called as thymocytes) which is responsible for the clotting of blood, The Leukocytes (also known as white blood cells), which are the cells of the immune system defending the body against and over both infectious diseases and foreign materials.

Blood cells are five kinds of white blood cells that form part of the **immune response**; each with different functions, namely, neutrophils, lymphocytes, monocytes, eosinophils and basophils and these are the white blood cells belong to two groups granulocytes or agranulocytes. Blood smear are routinely and commonly used in the clinical laboratories to give indications about the various and different types of diseases and also help to diagnose, monitor and evaluate the patient case immediately.

Whole blood count i.e. total number of RBC, WBC and platelets in given blood sample is the first and most important requirement for the diagnosis of any disease. And if there are excess of any of these types or any of these is few in number then it assures the doctor that the person is not healthy for sure. Manual counting of them is very tedious task. First of all we will differentiate RBC and WBC. Blood consists of mainly red blood cells (RBC), white blood cells (WBC), and platelets. Each has its own function in our body and posses equal importance. Blood contain three types of cells and cell fragments which are floating in liquid called plasma. These elements are given as follows:

- Red Blood Cells ("erythrocytes," "RBCs") - oxygencarrying cells
- White Blood Cells ("leukocytes," "WBCs") - cells that help to protect our body against diseases and prepare the body's immune system.
- Platelets ("thrombocytes") - fragments of cells that perform an important role in formation of blood clots.

II. WHITE BLOOD CELLS

WBC and RBC have fixed count in our body. If their count is less than the ideal Both count then it gives signal that our body is not healthy. Therefore blood count assists in detecting many diseases in their initial stage. White Blood Cells shield the body from disease. They are many less in number than red platelets, representing around 1 percent of your blood.

The most widely recognized kind of White Blood Cells is the neutrophil, which is the "quick reaction" cell and records for 55 to 70 percent of the aggregate White Blood Cells check. Every neutrophil lives not as much as a day, so your bone marrow should always make new neutrophils to keep up insurance against contamination. Transfusion of neutrophils is for the most part not successful since they don't stay in the body for long.

The other real kind of White Blood Cells is a lymphocyte. There are two principle populaces of these cells. T lymphocytes help manage the capacity of other resistant cells and straightforwardly assault different tainted cells and tumors. B lymphocytes make antibodies, which are proteins that particularly target microbes, infections, and other remote materials.

White Blood Cells assume a critical part in your insusceptible framework. They are likewise called as leukocytes or once in a while WBCs by specialists. White Blood Cells stream in your circulation system, assaulting attacking microscopic organisms, parasites and some other cells and items that are not intended to drift around inside your blood. White Blood Cells are not all equivalent. Entirely are six unique sorts of White Blood Cells, every one of which has a to some degree diverse part to play in your safe resistance. The six sorts of WBCs are part in to two noteworthy parts, granulocytes and granulocytes.

2.1 Neutrophils

Neutrophils are the most ordinarily happening kind of White Blood Cells. They represent around 65% of all White Blood Cells, and are otherwise called polymorphonuclear leukocytes or a PMN for short. They are the essential protectors when bacterial and parasitic contamination happened. Neutrophils can be thought of as the "specialists on call" in an attack of outside microscopic organisms or growths.

2.2 Basophils

Basophils are the minimum basic sort of White Blood Cells. Just 1% of your White Blood Cells comprise of basophils. The essential capacity of a basophil is to discharge a substance known as histamine accordingly a contamination. Histamine is a compound that has numerous capacities, yet it is basically in charge of starting an incendiary response.

2.3 Eosinophils

Their offer is around 4% of aggregate leukocytes. These White Blood Cells perform two noteworthy capacities as: Primary protectors against parasitic contaminations and lifts in instances of unfavorably susceptible responses, for example, hives, or even asthma identified with sensitivities.

2.4 Lymphocytes

The first of the agranulocytes are lymphocytes. Like all agranulocytes they do not have the film bound granules found in the other classification of White Blood Cells. Lyphocytes represent 25% of White Blood Cells. There are really three unique sorts of lymphocytes; B cells, T cells, and Natural Killer Cells. Every one of the three kinds have minutely fluctuating capacities.

2.5 Monocytes

Monocytes share around 6% of White Blood Cells and have a to some degree one of a kind and fascinating part to play in your invulnerable framework. Monocytes are fairly extensive contrasted with other White Blood Cells. They go around in your blood, searching for microscopic organisms, infections and other "waste" that necessities expulsion. When they discover something that requirements tidying up, they swallow the culpable molecule in a procedure known as "phagocytosis". Subsequent to gulping these bits, the monocyte will soften the trespasser up tolittler pieces and present them on its cell surface with the goal that passing T cells can "learn" more about the substance make-up of the intruder and make it simpler to execute a greater amount of them. Here more concern is tied in with grouping the WBC pictures in light of the fact that every cell compose has distinctive shape and shading as appeared in Figure and which influences the characterization exactness.

III. LITERATURE REVIEW

Platelet division basically removes the phones from bewildered establishment and parts every phone into morphological sections, for instance, center, cytoplasm, and some others. The count proposed by Ongun[5] et al. pieces the WBCs using dynamic shape models (snakes and inflatables). These shape based and surface based features are utilized for the gathering errand. Close around twelve classes of WBCs are considered for this computation.

Adollah[6] et al. present an expansive report about division procedures. The principal focus of his examination is to develop a robotized structure on platelet course of action. His work shortens the most surely understood and recognized methodology important for the evaluation of picture examination prominently in the division system.

Theerapattanakul[7] et al. use division by using dynamic structures. He uses twofold thresholding and a short time later by looking at the matched picture to find WBCs whose energy of center outperforms thresholding regard. At first round shape (wind) is put on the center zones discovered finally, dynamic frame demonstrate is used with slant stream vector constrain as an energy to drive the snake shape fitting the WBC to be evacuated.

F. Sadeghian[8] et al. show a structure to portion WBC in two sections as: Nucleus division Based on morphological examination and gives 92% precision Cytoplasm division Based on pixel-force thresholding giving 78% exactness if there should be an occurrence of previously mentioned system the significant impediment is that for simple usage, system has done on sub-pictures.

Madhloom[4] et al. recommends a calculation to mechanize the procedure of discovery and order of leukocytes into different particular subtypes. Here for the most part white platelet acknowledgment and grouping into different particular subtypes is focussed. Here essentially center is given around white platelet core division which separate core from the entire cell body by utilizing a blend of programmed balance extending with the guide of picture number juggling activity, least channel and worldwide edge methods. This proposed technique gives exactness between 85-98%.

Markiewicz[9] et al proposed a programmed framework for platelet acknowledgment. The acknowledgment is for the most part completed based on the bone marrow pictures. He apply the morphological pre-handling of the picture for singular cell extraction, age and determination of the indicative highlights and the acknowledgment framework utilizing Gaussian piece Support Vector Machine (SVM) naturally and yields 87% exactness.

In a programmed division method created by Theera-Umpon [10], he utilizes the fluffy C-implies (FCM) calculation to excessively fragment every cell picture to frame patches and numerical morphology and core smoothing and little fixes evacuation. Angulo and Flandrin research a system to consequently identify the working territory of fringe blood smears recolored with May-Grünwuald Giemsa.

The ideal region is characterized by the well spread piece of the spread. In this calculation two phases are available. In first stage, a picture investigation is completed utilizing numerical morphology to separate the erythrocytes. Also, if there should arise an occurrence of second, the quantity of associated segments from the three sorts of particles is checked and the coefficient of spreading and the coefficient of covering are figured.

Cao[12] et al. show a calculation that spotlights on the location of red platelets in pee picture. For recognition of red platelets, at begin he pre-handled pee picture by an enhanced Sobel administrator and restricted RBCs utilizing Hough Transform. Extraction and determination of highlights is completed with the assistance of Principal Component Analysis (PCA) and after that arrangement is finished with LDA (Linear Discriminant Analysis).

Ramoser[13] et al. display a completely computerized framework for leukocyte division which is hearty as for cell appearance and picture quality. Here arrangement of highlights portrays cytoplasm and core properties and pairwise SVM order separates between various cell writes. Assessment on an arrangement of 1166 pictures (13 classes) brought about 95% right divisions and 75% to 99% right characterization.

Vromen and McCane depict a model based form following way to deal with handle with the issue of consequently sectioning a Scanning Electron Microscope picture of red platelets. They utilize a moment arrange polynomial model and a basic Bayesian way to deal with guarantee smooth limits.

Poomcokrak and Neatpisarnvanit [15] point by point a strategy which is utilized to recognize ordinary red platelets (RBCs).Here neural system is utilized for arrangement. This investigation found that the proposed strategy gives affectability 0.86, specificity 0.76 and exactness 0.74. N. Sharma and L.M.Aggarwal proposed a paper which subtle elements procedures for mechanized therapeutic picture division. These strategies are particularly talked about with regards to CT and MR pictures. The methodologies clarified in this audit can be requested by materialness, reasonableness, execution, and computational cost.

Execution of division strategies as thresholding, and area based systems can be enhananced by coordinating them with counterfeit consciousness methods. Systems in light of textural highlights using chart book or look-into table have phenomenal outcomes on therapeutic picture division. In any case it is hard to effectively choose and name information, portion complex structure with variable shape, size, and properties with chart book based system. For this situation we can utilize unsupervised strategies, for example, fluffy means calculation. Numerous neural system based calculations are additionally accessible for surface based division and order having great precision. Be that as it may, they require broad supervision and preparing.

G. Lebrun, C. Charrier [21]proposed a quick and effective division conspire for cell minute picture. This plan for the most part focuses on the best way to diminish the intricacy of choice capacities delivered by help vector machines (SVM) while protecting acknowledgment rate. Vector quantization is utilized to decrease the inalienable excess present in tremendous pixel databases. Half and half shading space configuration is additionally utilized as a part of request to enhance informational collection measure decrease rate and acknowledgment rate.

Another choice capacity quality basis is displayed to choose great tradeoff between acknowledgment rate and handling time of pixel choice capacity. At that point another division conspire utilizing probabilistic pixel grouping with a few free parameters and a programmed choice is created. Another imperative commitment here is the meaning of another quality paradigm for assessment of cell division. The outcomes infer that the determination of free parameters of the division plot by streamlining of the new quality cell division model produces effective cell division.

S. Chinwaraphat1, A. Sanpanich [22]itemized a plan of altered fuzzy clustering for white platelet division. In this investigation most importantly the division is done by utilizing a standard FCM bunching strategy to characterize the picture of blood test slide into 4 essential gatherings as white platelet core, white platelet cytoplasm, plasma and red platelet. At that point FCM is altered to take out a dispersing or false bunching which was available due to an indistinct or shading pixel comparability amongst cytoplasm and plasma foundation was actualized again proficiently until the point that those blunders were limited. The minimization in every cycle circle was done by utilizing a neighboring shading pixel of its dissipating as a kind of perspective. At last the yield demonstrates that the altered technique can extricate core and cytoplasm district more productive than typical FCM.

Zhaozheng Yin, Ryoma Bise, Mei Chen and Takeo Kanade[12] proposed a strategy for cell division in microscopy symbolism utilizing a sack of nearby Bayesian classifiers. In microscopy symbolism cell division is essential for some bioimage applications, for example, cell following. For division of cells from the foundation precisely, a pixel order approach which is free of cell write or imaging methodology is displayed. This technique prepared an arrangement of Bayesian classifiers from bunched neighborhood preparing picture patches. Each Bayesian classifier is a specialist to settle on choice in its specific area. The choice from the blend of specialists decides how likely another pixel is a cell pixel.

The proposed technique points of interest the adequacy of this approach on four cell writes with various morphologies under various microscopy imaging modalities.A procedure was introduced by Tiago William Pinto,Marco Antonio Garcia de Carvalho, Roberto Marcondes C'esar J'unior to perform Image Segmentation Using Watershed and Normalized Cut. This system proposes a picture division methodology which utilizes two approaches to change over pictures into diagrams: Pixel proclivity and watershed change. Both ways give us result as a likeness network that is utilized to figure the phantom chart properties (eigenvalues and eigenvectors).

Proposed calculation decreases clamor and upgrades exactness contrasted with one proposed by Madhloom et al. By looking at Madhloom et al. with proposed calculation by visual review utilizing a test picture we can without much of a stretch foresee that the relative size test is critical to dispose of all the non-core objects which at last suggests predominance of proposed calculation.

IV. PROPOSED ALGORITHM

Only the nucleus part of the cell will appear as the darkest part of the image because in the beginning all the images are converted into grayscale images so that the nucleus part of the cell will appear as the darkest part of the image. The white blood cells nucleus localization is obtained due to automatic contrast stretching, histogram equalization and image arithmetic.

STEP 1: Take a WBC microscopic image say

STEP 2: Convert it to grayscale image

STEP 3: First one copy of the image will be enhanced with a linear contrast stretching (in this research this is referred to as L) .

STEP 4: another copy will be enhanced with histogram equalization (in this research this is referred to as H)

STEP 5: Obtain the image R1=L+H

STEP 6: Obtain the image R2=L-H.

STEP7: Obtain the image R3=R1+R2.

STEP 8: Implement, three times, 3-by-3 minimum filter on the image R3.

STEP9: Convert R3 to binary image using the threshold from step 8.

STEP10: Use morphological opening to remove small pixel groups. Use a disk structuring element with a radius of 9 pixels.

STEP 11: Connect the neighboring pixels to form objects.

STEP 12: Apply the size test to remove all objects that are less than 50% of average RBC area.

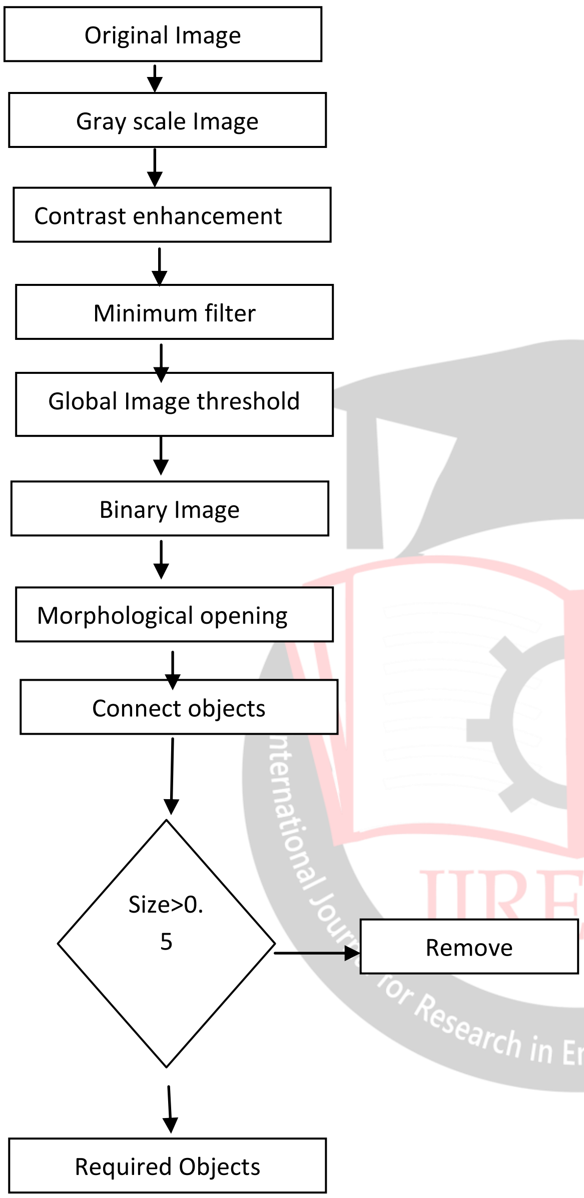

Fig1: Design Flow of Automatic Segmentation

V. CONCLUSION

The proposed method manages to show results and obtain accuracy results between 85-98%. The results showed that the proposed method is promising and better when compared to the result from the expert. This study introduces a method for white blood cell nucleus localization and segmentation as a first step towards a fully automatic system for leukemia diagnosis and classification using peripheral blood microscope image. White blood cell segmentation is the key procedure in the automatic leukemia diagnosis system. The variation in the accuracy rate between the proposed result and the manual segmentation is due to the type of the cell and also the method that is used to prepare the slides beside that the manual segmentation is highly depend on the hematologist capabilites. The best segmentation results is manifest from the Monocyte and Neutrophill image where the boundaries of the nucleus and cytoplasm are very distinct.

VI. RESULTS

Fig 2 : Input image

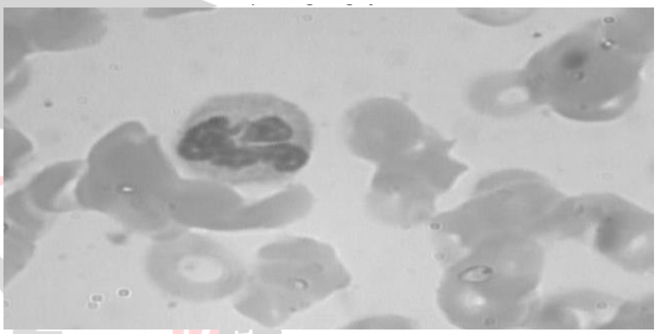

Fig 3:Input image in gray scale

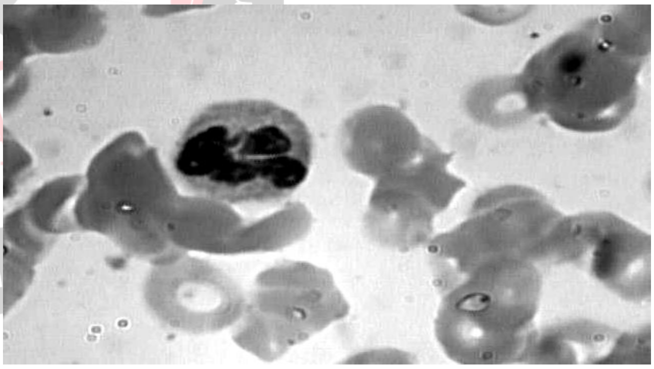

Fig 4:Input image with intensity values adjusted by a linear contrast stretching

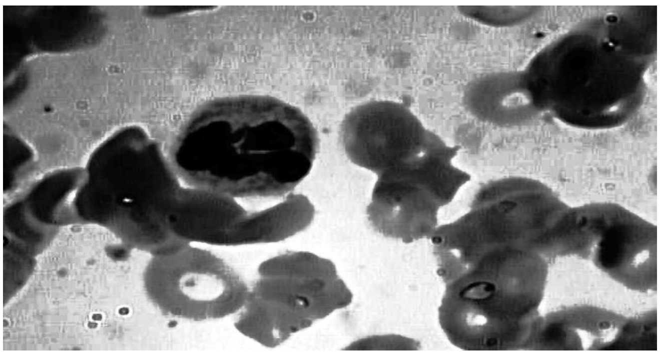

Fig 5: Enhance contrast using histogram equalization

ISSN : 2454-9150

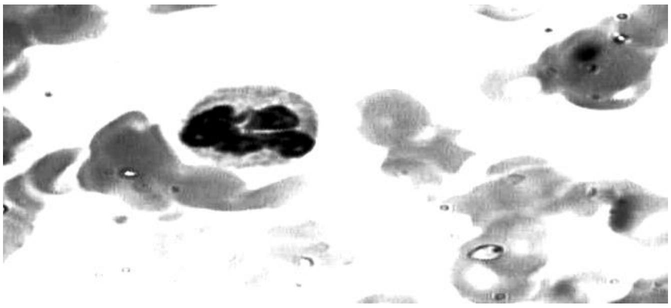

Fig 6: Brighten most of the details in the image except the nucleus.

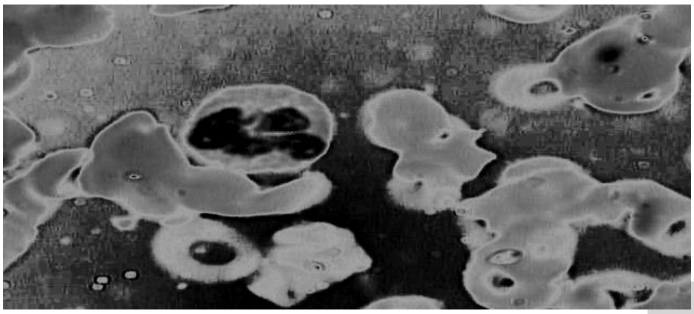

Fig 7: Highlight all the objects and its borders in the image including the cell nucleues

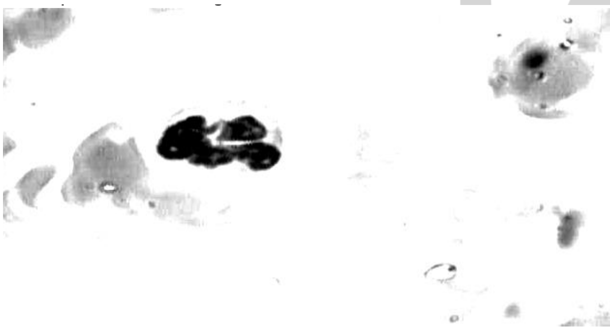

Fig 8: Remove almost all the other blood components while retaining the nucleus with minimum effect of distortion on the nucleus part of WBC.

Fig 9: Global threshold using Otsu's method

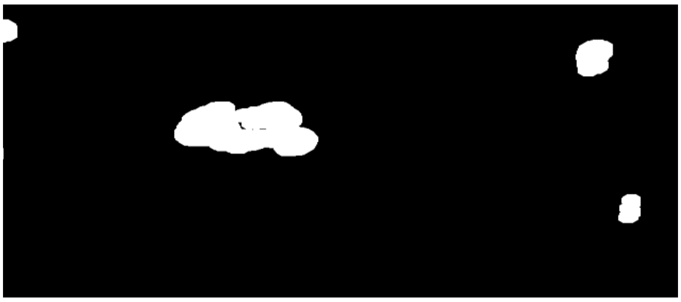

Fig 10: After morphological opening

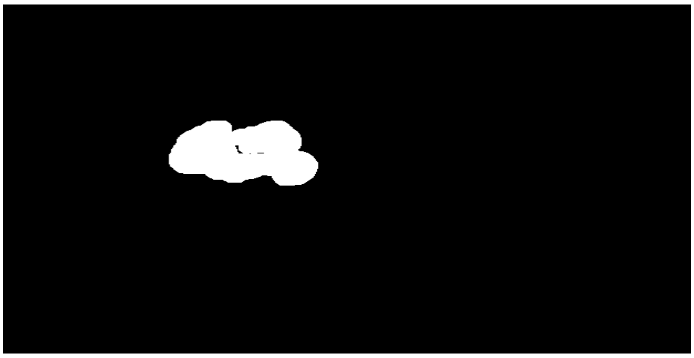

Fig 11: After minimum area check and remove false objects.

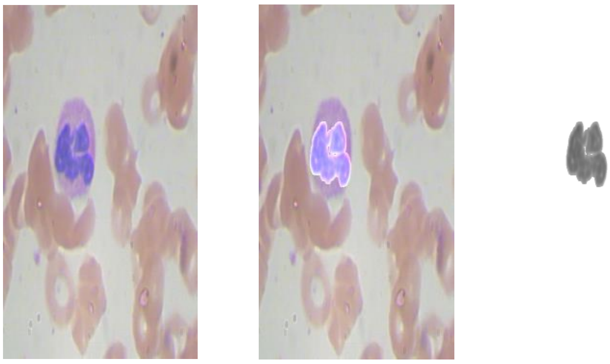

Fig 12: Final segmented results

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