

# A Review paper on White Blood Cells Segmentation

Priyanka Premnath<sup>1</sup> Srilatha.L.Rao<sup>2</sup> Anand Jatti<sup>3</sup>

<sup>1,2,3</sup> Department of Biomedical Signal processing & Instrumentation  
<sup>1,2,3</sup> RVCE, Bengaluru, India

**Abstract**— Diagnosis of various blood related diseases adopts differential WBC count which plays an important and vital role. Counting the cells manually is a tedious task and, time consuming. Hence many automatic techniques have been developed which gives a faster and more accurate ways to count the blood cells. This paper describes the various techniques for WBC segmentation the basic steps involved are segmentation, feature extraction and classification. Few techniques discussed are Fuzzy C, Snake contours, NDA, Segmentation Based on Gram-Schmidt Orthogonalization, White Blood Cell Segmentation in Microscopic Blood Images Using Digital Image Processing, Modified Fuzzy Clustering, and Magnetophoretic separation of blood cells at the microscale.

**Key words:** Feature extraction, NDA, Magnetophoretic, Thresholding, Fuzzy cellular neural networks (FCNN), image segmentation, mathematical morphology, and white blood cell detection. Snake contour.

## I. INTRODUCTION

Disorders results in variations in the number of white blood cells. Hemanalysis provide an analysis of the quantitative information, life study and the diagnosis of the various diseases. Complete Blood count (CBC) and the Differential Blood count(DBC) are the two types of blood count required for the diagnosis .CBC are usually performed automatically by cytometers and DBC is a manual procedure done using a microscope. DBC includes all of the cell types within a blood sample which are five different white blood cell types, red blood cells and platelets. The five different types of white blood cells and their relative percentages in blood are neutrophils: 40-70%, eosinophil's: 5%, basophils: 0.5%, lymphocytes: 20-50% and monocytes: 1-5%. WBC composition reveals important information about the patients. However, DBC is highly tedious and complex. Advancements in the field of computers and the Image processing have led to the automatic detection of the WBC which has reduced the analysis time and improved the efficiency.

Automatic Detection /Recognitions of the WBC have accelerated the diagnosis of various diseases. It usually consists of four major steps, including: preprocessing, image segmentation, feature extraction and classification as shown in Fig 1.

The segmentation of WBC is the most critical step as the accuracy of the feature extraction and classification depends on it. Automatic white blood cell detection is very complex. Besides white blood cells, there exist red blood cells, platelets, and other objects mixing in the microscopic blood images. To distinguish one element from others becomes very difficult. Thus various enhancements have

been done to the traditional segmentation techniques which are discussed here.

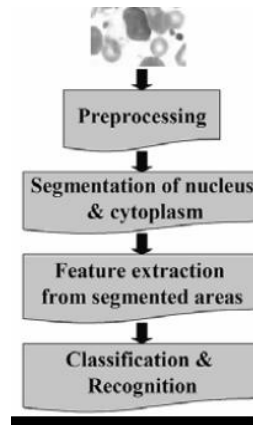


Fig 1: Common Block Diagram for WBC detection

## II. VARIOUS SEGMENTATION TECHNIQUES USED

### A. WHITE BLOOD CELL NUCLEUS SEGMENTATION BASED ON GRAM-SCHMIDT ORTHOGONALIZATION

The nucleus of the WBC gives the information on the type of WBC. In this automatic process, the segmentation of the WBC is an important stage which is based on orthogonality theory and Gram Schmidt process. The color of the nuclei is often violet with different intensities and saturation. Here, a 3-D feature vector is defined for each pixel using the RGB components of the images, to apply Gram-Schmidt orthogonalization for the segmentation of nucleus of white blood cells. A weighting vector  $w$  is calculated for amplifying the desired color vectors and weakening the undesired color vectors, according to the Gram-Schmidt method, As shown in Fig. 2, the pixel vectors and the inner product of the weighting vector defined from the original image produce a composite image which has maximum intensity in regions with violet color and minimum intensity in other regions. [1]

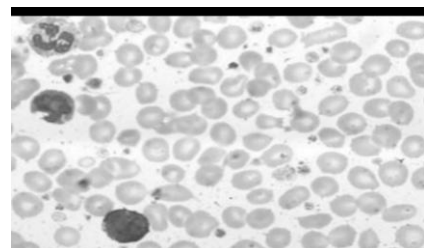


Fig. 2: Original Image

Due to this large variation of the changing intensities and saturation of the blood samples, three different weighting vectors are calculated for the Gram-Schmidt orthogonalization. For each vector, an image which is similar to Fig. 3 is obtained and an appropriate threshold is

calculated for each image, which is computed based on weighted mean and maximum of their histograms. Finally, after thresholding and removing small (fine) components, logical “AND” operation to the results. Fig. 4 shows a flow chart for our proposed scheme.

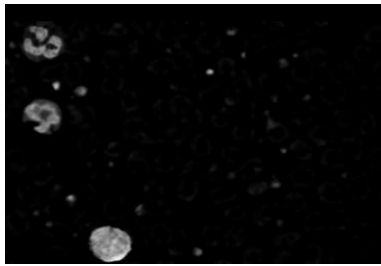


Fig. 3: Resulting Image after multiplying pixels of original image by vector calculated by Gram-Schmidt method [1]

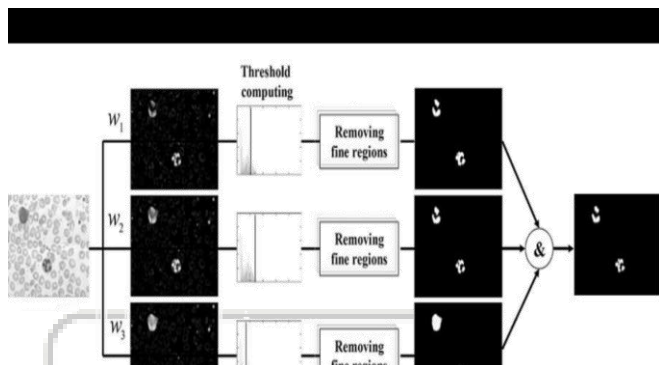


Fig. 4: Block diagram for proposed scheme [1]

This method is simple to implement, quick and efficient.

#### B. A Framework for White Blood Cell Segmentation in Microscopic Blood Images Using Digital Image Processing

In this method the main goal is to separate the leucocytes from different components in blood image. A Typical blood smear consists of four components which are red cells (un-nucleated cells), white cell's nucleus, background, and cytoplasm. WBC appears rather darker than the background, and red cells appear in an intermediate intensity level. Also, there is shape variation in cells and their nucleus. In the process of nucleus segmentation, snake algorithm is used that are not related to size and color of nucleoli because there are various shapes of nucleus in a different kind of white blood cells. It gives high accuracy result in segmenting nucleus in any type of WBCs and in any capture individual WBC is preferred to get a better result. This method is very simple with trustable accuracy and high speed illumination that cause different color space in images. And, in a cytoplasm segmentation method, the thresholding technique is used where background is completely segmented from other components. Thus, based on the difference between RBC color in blood image and cytoplasm area, thresholding value is easily segmented in the cytoplasm part. But, sub-image that contains.

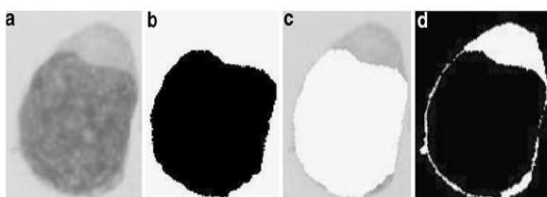


Fig. 5: an Original image, b segmented nucleus, c image resulting from Subtracting a with b, d segmented cytoplasm. [2]

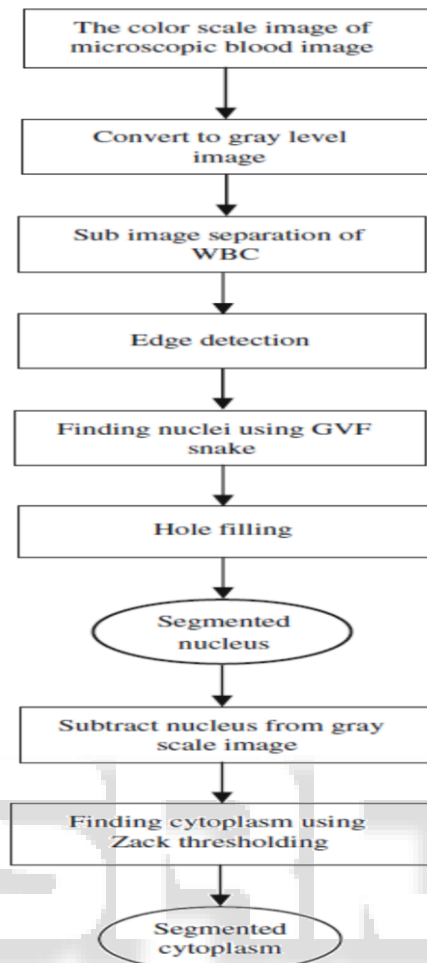


Fig. 6: The proposed framework of the WBC segmentation scheme. [2]

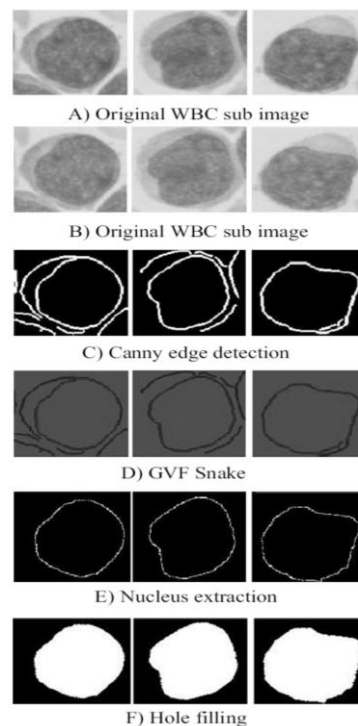


Fig. 7: Nucleus segmentation procedure. A Original WBC sub image. B Original WBC sub image. C Canny edge detection. D GVF snake. E Nucleus extraction. F Hole filling. [2]

Thus, this method able to yield 92% accuracy for nucleus segmentation and 78% for cytoplasm segmentation. [2]

### C. A MODIFIED FUZZY CLUSTERING FOR WHITE BLOOD CELL SEGMENTATION

Firstly, the segmentation is performed using a standard FCM clustering technique to classify the image of blood sample slide into 4 groups such as white blood cell nucleus, white blood cell cytoplasm, plasma and red blood cell.

Thus, the segmentation algorithm starts from a color transformation of WBC image slide into HIS space due to its color tone dominance. Color data is then clustered by a standard FCM to obtain 4 groups of primary clustered information. Since there is an unclearness or color pixel similarity between cytoplasm area and plasma background area, some scattering or misgrouping of color pixel always occurred inherently. Clustering with a modification was performed iterately in order to minimize these errors. Using a neighbouring color pixel of its scattering as a reference, the minimization in each iteration loop was accomplished.

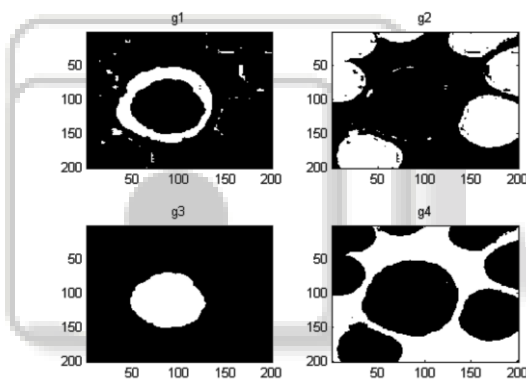


Fig. 8: (g1) WBC cytoplasm group, (g2) RBC

Group, (g3) WBC nucleus group, (g4) plasma group

The segmentation due to modified FCM shows that this method is able to enhance a clustering promisingly.

The nucleus portion and cytoplasm portion that are obtained can be used to identify a WBC type in the next classification step of automatic WBC counting process efficiently. However, there may be some overlaps between cytoplasm and plasma portion. These errors can be overcome in the slide preparing process.

The final output shows that our modified method is able to extract nucleus and cytoplasm region more efficient than normal FCM. In this method we are able to segment white blood cell promisingly. [3]

### D. PATCH-BASED WHITE BLOOD CELL NUCLEUS SEGMENTATION USING FUZZY CLUSTERING

This technique is based on fuzzy c means clustering and mathematical morphological where a single cell WBC in bone marrow is segmented into two regions, nucleus and non-nucleus and this segmented count is compared with manually segmented images. This uses Bayes classifiers style for classification of patches. Patches are basically

group of connected pixels. This count is used as a basis for analysis of many diseases such a blood cancer, AIDS etc. The count of these WBC in bone marrow is done according to their maturation levels. FCM aims to classify all the similar cells into a single category and dissimilar cells into different category or groups. Here various types of morphological processing techniques and is applied on the image such as enhancements, edgedetection, segmentation etc are applied. Along with this many morphological operations like erosion, dilatation are performed on the image. Next Bayes theorem is used for analyses of the function obtained. The cells are then classified based on the classes obtained. Usually there are 6 classes that are got which usually classified as myeloblast, promyelocyte, myelocyte, metamyelocyte, band, and PMN. Every class of the above WBC cells has 3 distinct regions nucleus, cytoplasm, and background with certain distinguishable gray level values. Certain evaluation measures are calculated to get the percentage of differently labeled pixels in the image. Here a median filter is used to overcome the difficulty caused by the varying intensity in each cell regions. After segmentation is completed the image now consists of 2 regions nucleus and non-nucleus which are obtained by patch combining techniques using FCM centers. These patches are classified as nucleus or non-nucleus depending on the mean of all centers value. Closings of the structural elements are used to remove Smooth patches and smoothen edges. This technique is much simpler than pixel based segmentation. [4]

### E. WHITE BLOOD CELL SEGMENTATION AND CLASSIFICATION IN MICROSCOPIC BONE MARROW IMAGES

Microscopic WBC are automatically segmented into its 3 basic regions namely nucleus, cytoplasm, and background. The probability of error generated when the resultant segments is compared with manually segmented images by an expert is then used for evaluation. Manual procedures are usually tedious and time consuming as it involves steps like locating cells, feature extraction, classification etc. Here differential counting is difficult due to high density of cells. Immature WBC also appears in bone marrow which again complicates the analysis. [6]. Here WBC differential counting in bone marrow is obtained by mixing of mathematical morphologies. Also to get the count of differential cell classes a new training algorithm for neural networks were used. [7][8]. Here bone marrow cells WBC are segmented as nucleus and cytoplasm and FCM is applied on it to get cell patches in images. These patches are then combined to form nucleus, cytoplasm, and background. The segmentation errors are evaluated by comparing the automatic segmented images to the corresponding images segmented by an expert using the probability of error in image segmentation. Features such as its shape, size etc are also extracted using neural techniques.

The nucleus boundary of a mature cell is more clearly distinguishable than a young cell. Thus PE in nucleus segmentation is smaller for mature cells. Here automatic classification is achieved by using neural networks.

Here fivefold validation is performed. For every segmented image the nucleus spectrum is calculated. Four

features, i.e., cell area, pattern spectrum's peak location, first and second granulometric moments, as described in section 4.2, are extracted. The classification rates achieved by using the automatic segmented images are close to that achieved by using the images segmented manually by the expert. [5]

#### F. A New Detection Algorithm (NDA) Based on Fuzzy Cellular Neural Networks for White Blood Cell Detection

For diagnosis of any blood related disease, analysis of White blood Cell segmentation using automatic microscopic images is one of most important and preferred method. Hence the accuracy and speed of operation should be very high. Two basic methods which are used are threshold segmentation followed by mathematical morphology (TSMM), and the fuzzy logic method—a new detection algorithm (NDA) based on fuzzy cellular neural networks. Though the individual methods pose certain drawbacks, these are overcome using the combination of two methods. Using the NDA almost all the WBC and their contour are detected.

In Gray value of nuclei is the smallest in the image hence in TSSM, binary threshold segmentation is the first step performed. Then the WBC is detected individually according to their shape.

The following procedure is used 1) the image compressed using pyramidal method 2) binary segmentation is done after taking the average of cytoplasm as threshold 3) next perform dilation, erosion on the image 4) segment the nuclei with the shape features such as area and round degree; 5) locate each nucleus, set the proper size of window, recover the original image inside the window. NDA technique is an adoption of FCNN. FCNN is applicable to color images too. Hence when NDA is used along with FCNN we are able to extract information about all the WBC as well as its structural information during the same processing. Also, blood cell detected is nearly complete as a result of the advanced morphological gray reconstruction. Due to proper color transformation NDA technique has strong adaptability to staining and illumination. [9]

#### G. SEGMENTATION THE WHITE BLOOD CELL BY UTILIZING ACTIVE CONTOUR

In this technique, first the image is converted into binary using double thresholding. Then the image is scanned to find the nucleus of each WBC cells whose intensity is exceeding the threshold value. Next circular shape (snake) is placed on inside of each WBC cells detected and active contour model using the gradient flow vector force is applied as driving force. Extracted contour is then used to separate these WBC. This type of segmentation technique works well even in the case of touching neighbouring cells. The main draw back with this technique is the initial position and initial size of the snake contour. If the initial contour of circle has very small radius then there is small gradient vector force and as a result the snake contour does not change with iteration. If the initial contour has very big radius then the snake contour grows towards the gradient vector which belongs to RBC. [10]

#### H. MAGNETOPHORETIC SEPARATION OF BLOOD CELLS AT THE MICROSCALE

This is a direct and continuous method to separate RBC and WBC present in plasma. In this method a microfluidic system that consists of an array of integrated soft-magnetic elements embedded beneath a microfluidic channel is used. This microsystem is activated by applying a bias field that magnetizes the elements. Once the elements are magnetized it develops weak magnetic forces which help its motion in the microsystem. White blood cells behave as diamagnetic micro particles while red blood cells behave diamagnetic or paramagnetic depending on the oxygenation of their hemoglobin. A mathematical model is developed to predict motion of white blood cells and deoxygenated red blood cells microsystem. And the magnetic force developed is sufficient to separate the two types of cells as they flow through the micro channel. Thus the cells are separated. It has advantages over competing techniques such as centrifuging, or magnetophoresis that involves magnetically labeled materials. [11]

### III. CONCLUSION

The various methods of white blood cell detection in the automatic recognition systems of microscopic blood images have been discussed in this paper. Different techniques used in WBC segmentation have been analyzed along with their principle of operation and the algorithm used advantages, disadvantages and enhancements made in the field relating to it. Also the various techniques involved in separating the WBC and RBC are discussed.

### REFERENCES

- [1] S. H. Rezatofghi H. Soltanian-Zadeh R. Sharifian R.A. Zoroofi, a New Approach to White Blood Cell Nucleus Segmentation Based on Gram-Schmidt Orthogonalization.
- [2] FarnooshSadeghian, ZaininaSeman, Abdul Rahman Ramli, BadrulHisham Abdul Kahar, and M-Iqbal Saripan. A Framework for White Blood Cell Segmentation in Microscopic Blood Images Using Digital Image Processing.
- [3] S. Chinwaraphat<sup>1</sup>, A.Sanpanich, C. Pintavirooj, M. Sangworasil<sup>1</sup> and P. Tosranon a modified fuzzy clustering for white blood cell segmentation.
- [4] Nipon Theera-Umpon: Patch-Based White Blood Cell Nucleus Segmentation Using Fuzzy Clustering.
- [5] Nipon Theera-Umpon: White Blood Cell Segmentation and Classification in Microscopic Bone Marrow Images.
- [6] Minnich V.: Immature Cells in the Granulocytic, Monocytic, and Lymphocytic Series, American Society of Clinical Pathologists Press, Chicago (1982).
- [7] Theera-Umpon N., Gader P.D.: Training Neural Networks to Count White Blood Cells via a Minimum Counting Error Objective Function. In: Proc 15th Intl Conf on Pattern Recog, (2000) 299–302.
- [8] Theera-Umpon N., Gader P.D.: System Level Training of Neural Networks for Counting White Blood Cells. In: IEEE Trans Systems, Man, and Cybern Part C: App and Reviews. Vol. 32. No. 1. (2002) 48–53

- [9] Wang Shitong and Wang Min: A New Detection Algorithm (NDA) Based on Fuzzy Cellular Neural Networks for White Blood Cell Detection.
- [10] J. Theerapattanakul, J. Plodpai and C. Pintavirooj: Segmentation the White Blood Cell by Utilizing Active Contour.
- [11] E. P. Furlani: Magnetophoretic separation of blood cells at the microscale.

