

Blood Microscopic Image Segmentation & Acute Leukemia Detection

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Abstract—

In this paper the visual inspection of microscopic images of blood is done for the identification of blood disorder. The results obtained from this identification can be used for classification of disease related to blood. Pathologists mostly use the leukocytes to identify the various diseases related to blood. The traditional methods like fluorescent in situ hybridization (FISH), immunophenotyping, cytogenetic analysis, and cytochemistry are time consuming. So the proposed method is used for leukemia detection which is very fast and used for analysis of normal and malignant cells. In acute lymphoblastic leukemia the infected lymphocytes are afterward converted as lymphoblast. The disease grows very fast in short duration so that it may destroy life. Since the early detection is important for doctors to treat the patient. In this paper segmentation, shape features with contour signature and texture features are calculated and the results are explained. The MATLAB software is used for this technique.

Keywords— Acute Lymphoblastic Leukemia, White Blood Cells, Median filtering, Automatic thresholding, segmentation.

I. INTRODUCTION

The term disease means the absence of ease within the body. Because of that the normal functioning of the body and any other part of the body becomes impaired. When diseases take place the medical treatment is necessary. In general, disease can be classified on the basis of their cause and cell origin i. e. infectious, immunological, endocrine, genetic, neoplastic and traumatic etc. Physicians are interested in understanding the biology of the diseases and how it can be prevented or treated. All cancers are characterized by the uncontrolled growth of abnormal cells, invade surrounding tissues, metastasize, and eventually killing the host where it originates [1]. Cancer can develop in any race, gender, age, socioeconomic status, or culture and can involve any type of cells, tissues or organs of the human body. Cancer is the second leading growth of death, after cardiovascular diseases and 12.7 million people are diagnosed with cancer out of which 7.6 million deaths occurred in the year 2008 [2]. Haematological malignancies such as leukemia, lymphoma and myeloma are the types of blood cancer that can affect blood, bone marrow, lymphatic system and the major contributors for the cancer death [3].

Leukemia is a disease which affects blood, bone marrow and lymph nodes. The diagnosis of leukemia is based on the blood test and also on the bone marrow test. An immature White Blood Cells (WBC) can also occur in gonads, meninges, thymus, liver, spleen and lymph nodes. Leukemia diagnosis is based on WBC count if white cell count is increased and platelets and neutrophils are decreased then the leukemia is present. Pathologically there are two main types of leukemia that are acute and chronic. In acute lymphoblastic leukemia there is rapid growth of blast cells and in chronic lymphoblastic leukemia there is slow growth of blast cells. It can be also classified as,

1. Lymphoblastic leukemia
2. Myelogenous leukemia

Above two subtypes can also be classified into several subcategories. In the given paper only the Acute Lymphoblastic Leukemia (ALL) is considered. Acute Lymphoblastic Leukemia (ALL) generally presents in 0-10 years old children. So the above methods are useful for analysis of lymphocyte as a normal cell or a lymphoblast cell. Instead of using traditional methods like immunophenotyping, flow cytometer, molecular probing etc., and the microscopic examination of blood slides still remains standard leukemia diagnosis technique. The symptoms of leukemia patient are fever, weakness, pallor, infection bleeding and enlarged lymph nodes and spleen. So the cost effective and automated system is used for the improvement purpose of leukemia detection.

II. LITERATURE REVIEW

Over years several works has been introduced for the detection of leukemia which is based on segmentation method of blood smear images. Generally most of the methods are based on the local image information. In this method automated white blood cell segmentation is used. A two-step segmentation process using HSV colour model is used in [4]. Cell segmentation using active contour model is used in [5]. The use of shape analysis into WBC segmentation is used in [6]. The colour segmentation procedure which is applied to leukocyte images using mean-shift is described in [7]. Extensive research has been carried out to implement quantitative microscopy on histopathological images, studies on the evaluation of haematological images for disease recognition and classification is limited.

There are many researches present on blood cell segmentation and detection in the literature. An automated segmentation is more useful and also it is proper method. In the present paper we propose an automated segmentation method which can help the physician for better diagnosis and treatment. In an automated segmentation the leukocytes are separated from the blood cells and then lymphocytes are extracted from the subclass. These lymphocytes are used for feature extraction. Further classification is done by using Support Vector Machine (SVM) classifier. Support Vector Machine (SVM) classifier used to separate healthy and leukemic cells.

III. METHODOLOGY

The following steps used for leukocyte classification procedure that are: - 1) Pre-processing, 2) Segmentation, 3) Feature extraction, 4) Classification. The blood smear image consists of Red Blood Cells (RBC) along with White Blood Cells (WBC) and platelets. Our main aim is to separate the lymphocytes from all these blood components for further work. Our main goal is to separate white blood cells and finally to separate nucleus and cytoplasm. For Acute Lymphoblastic Leukemia (ALL) we have to consider only nucleus as the region of interest and from that the essential features are extracted. The whole process is shown in figure 1,

A. Image Grabbing

The image is grabbed by using digital microscope under 100X oil immersed setting and with an effective magnification of 1000.

B. Pre-processing

The noise which accumulated during image acquisition and due to excessive staining is removed by using pre-processing technique. In the present paper the pre-processing is done by selective median filtering [8] followed by unsharp masking [9]. These are more reliable and more efficient methods for pre-processing. Median filtering is used to remove the noise present in an image and unsharp masking is used for better enhancement of an image. So the segmentation procedure is easier.

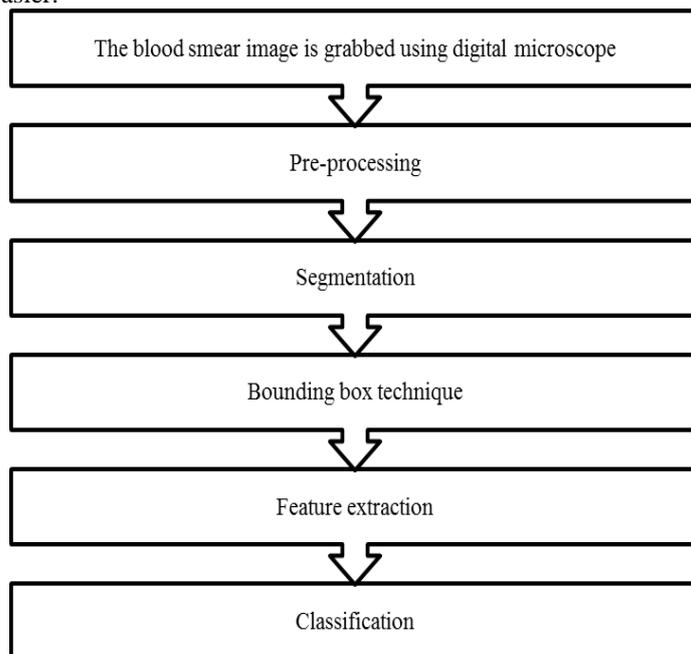


Figure 1:- Typical system overview

1. Median filtering:- The steps which are used for median filtering are as follows:
 - 1) Assume 3 x 3 empty mask.
 - 2) Place the empty mask at the hand corner.
 - 3) Arrange the 9 pixels in ascending or descending order.
 - 4) The median is chosen from 9 values.
 - 5) Finally place the median at the centre.
2. Unsharp masking:- It is a technique used for the sharpening an image. Digital unsharp masking is powerful and flexible method to increase sharpness.

C. Colour conversion

By using digital microscope the RGB images are generated. RGB images are difficult for segmentation. The RGB colour space is converted into the HSV colour space to make segmentation easy. HSV stands for Hue, Saturation and Value. For better separation of cells, we have to select saturation plane because saturation plane shows better contrast. Histogram equalization and contrast stretching is used for better segmentation.

Blood smear microscopic images are acquired in RGB colour space. Colorimetric transformation of initial colour coordinate system i. e. RGB is essential to obtain colour space in which the representation of data is the best to optimally perform the segmentation process.

D. Segmentation

Image segmentation is one of the early computer vision problems. Segmentation has wide range of application. It is used to divide the image into a set of homogeneous and meaningful regions, such that the pixels of each divided region has identical set of properties and attributes. The result of segmentation is nothing but the number of homogeneous regions, each having a unique label.

We have to perform first the thresholding in segmentation procedure which is main step. The result of thresholding is nothing but the RGB image is converted into black and white image. In automated image segmentation the thresholding is done by Otsu's method because in Otsu's method the threshold value is automatically selected. After thresholding it is necessary to remove the background pixels and artefacts from the image. To clear the artefacts the area opening operation is used. Area opening operation allows deleting the unwanted cells and objects which are smaller in size than structuring element. It is easy for the segment further for overlapping cells when the image with only leukocytes is obtained. The components present in binary image are labelled by various numbers. The labelled numbers are nothing but the nuclei of WBC's which is used to find out the features and hence used for leukemia detection.

E. Sub imaging

The disaster of blue nuclei present in peripheral blood smear image. For accurate detection of leukemia it is important to extract individual nucleus for classifying it as blast cell. Therefore sub image which consist single nucleus per image is necessary. For sub imaging purpose the bounding box technique is proposed.

F. Feature extraction

The common properties present within an image are isolated by using image feature. An image feature is distinguishing attribute of an image. The transformation of input data into the set of features is nothing but the feature extraction [10]. By using the segmentation for further processing the shape features are extracted.

1. Shape features:- The shape of the nucleus is an important feature for describing the cell as blast or normal according to doctors. The boundary based features are evaluated here. The evaluation of each cell is independently done using these features.

- 1) Area: It is nothing but total number of nonzero pixels in an image.
- 2) Perimeter: Total number of pixels in the boundary of the shape of an object.
- 3) Eccentricity: This parameter is used to measure the deviation of an object being circular. This is important feature because lymphocytes are more circular than blast cells. To measure this the formula is in (1),

$$\text{Eccentricity} = \frac{a^2 - b^2}{a} \quad (1)$$

4) Solidity: It is the ratio of actual area and convex hull area. It is essential for blast cell classification. To measure this the formula is in (2),

$$\text{Solidity} = \frac{\text{Actual Area}}{\text{Convex hull Area}} \quad (2)$$

5) Form factor: It changes the surface irregularities and it is dimensionless factor. To measure this the formula is shown in (3),

$$\text{Form Factor} = \frac{4 \times \pi \times \text{Area}}{\text{perimeter} \times \text{perimeter}} \quad (3)$$

2. Texture features:- This features includes,

- 1) Energy: Uniformity is measured using energy.
- 2) Correlation: Correlation between pixel values and neighbourhood is represented.

G. Classification

The Support Vector Machine (SVM) classifier is used for classification because SVM is powerful tool for data classification [11]. It is achieved by separating linear or non- linear surface in input space of data set.

IV. RESULTS

The segmentation technique is applied to various images of blood smear. The input image is segmented by using Otsu's method. From the input image only the clusters of blue nucleus are considered for evaluation. The obtained results are shown in figure below,

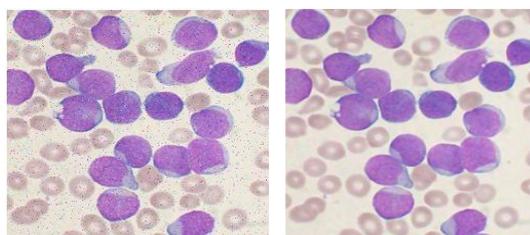


Figure 2. Noisy image and Filtered image

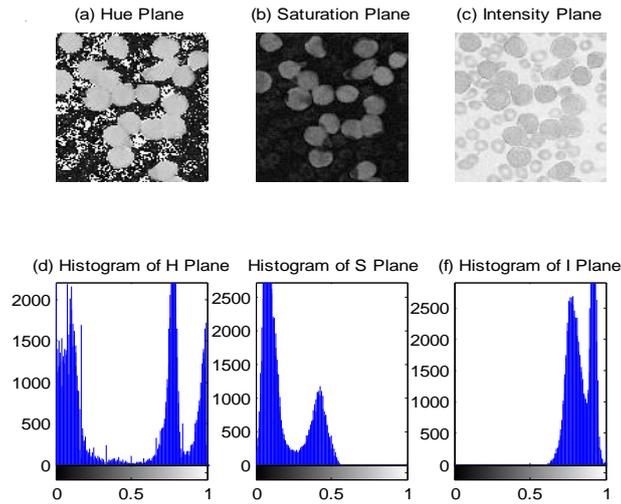


Figure 3. RGB to HSV colour conversion



Figure 4. Segmentation output

Then the lymphocytes from the segmented output images are separated using bounding box. As shown in fig (5)

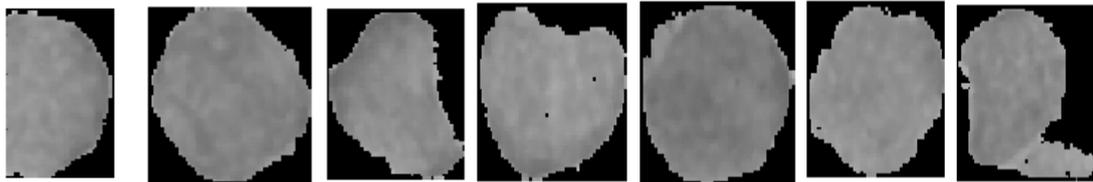


Figure 5. Separated nucleus sub images using bounding box technique

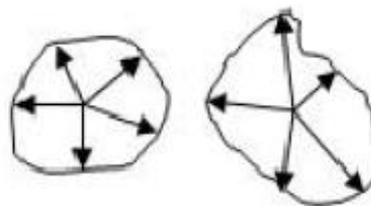


Figure 6. Contour Signature

In above results input noisy image and filtered image is shown in figure 2. The image and histogram of 'h', 's', and 'v' component shown in figure 3. The final segmented output is shown in figure 4 and result of sub imaging is shown in figure 5. The result of counter signature is shown in figure 6. In leukemia detection segmentation is the main part. The segmented output is used for the further processing that is for the feature extraction and classification. The shape feature and counter signature are calculated and result shown in table below.

Table I. Result of various shape measurements

Measure	N1	N2	N3	N4	N5	N6	N7
Area	1811	1964	1807	2000	1880	2086	2118
Perimeter	171.9828	168.8528	186.4092	182.7523	172.0244	183.6812	233.8234
Eccentricity	0.773	0.4226	0.7252	0.5960	0.3957	0.5044	0.6344
Solidity	0.9789	0.9713	0.9099	0.9324	0.9701	0.9600	0.8448
Form Factor	0.7694	0.8656	0.6535	0.7525	0.7983	0.7770	0.4868

Table II. Result of counter signature

ROI	N1	N2	N3	N4	N5	N6	N7
Standard Deviation	0.1403	0.0487	0.1497	0.1120	0.0386	0.0599	0.1200

Table III. Result of Texture feature measurement

Measure	N1	N2	N3	N4	N5	N6	N7
GLCM	192.4468	184.5712	201.5650	238.1986	170.5425	258.3318	266.8758
Energy	0.4737	0.3092	0.3459	0.4716	0.3097	0.5296	0.4001
Correlation	0.9033	0.8864	0.8971	0.8953	0.8446	0.8934	0.9052

V. CONCLUSION

The basic goal of the proposed method is to detect the leukemia which is childhood cancer. Acute Lymphoblastic Leukemia (ALL) growth is faster so for the treatment of patient we need early detection. The traditional methods like Fluorescent in Situ Hybridization (FISH), Immunophenotyping, Cytogenetic analysis and Cytochemistry are time consuming. So for the fast detection the proposed method is useful because the segmentation used in this method is an automated segmentation which is based on the Otsu's method. Thus the automated segmentation performs the important role for the detection of leukemia. The result of shape features also important for leukemia detection. The result of contour signature is used for feature extraction. Results obtained encourage the future work to develop automated segmentation system which is independent of stains used in blood smear image and which is helpful for segmenting overlapping cells.

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