

## A Novel Color Image Segmentation Method and Its Application to White Blood Cell Image Analysis

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### Abstract

*According to the fact that the H component in HSI color space contains most of the white blood cell information, and the S component contains the structure information of the white blood cell nucleus, we develop an iterative Otsu's approach based on circular histogram for the leukocyte segmentation by taking full advantage of this knowledge. Experimental results show that this method works successfully in the segmentation of color smear microscopic images.*

### 1. Introduction

White blood cell composition reveals important diagnostic information about the patients. The counting result gives important information about patient's health status and plays an important role in diagnosis. So an accurate and automatic differential white blood cell counter is crucial in the counting process. The process usually contains three steps: image segmentation, feature extraction and classification.

The paper is mainly about the first step of automatic analysis: the segmentation step. This step is very crucial because the accuracy of the subsequent feature extraction and classification depends on the correct segmentation of white blood cells. It is also a difficult and challenging problem due to the complex nature of the cells and uncertainty in the microscopic images.

Many image segmentation methods are based on threshold selection, edge detection or region growing. Conventional thresholding techniques can not always produce meaningful results since not all information is made use of during the process of the segmentation,

although they are usually combined with mathematical morphology operations to improve the accuracy of segmentation [1,2] in the case of cell segmentation: edge detection performs poorly on cell images because not all cell boundaries are sharp and hence it is difficult to get all the edge information and locate the cells accurately [3]. In this paper, we process the smear images in the Hue-Saturation-Intensity (HSI) space. Leukocytes are detected successfully in hue component, while nuclei are found out in the saturation component subsequently. Both procedures are based on the Otsu's segmentation approach [4]. An important improvement to Otsu's approach is that the segmentation in hue component images is based on circular histograms instead of linear programs as in the case of gray level images. This takes full advantage of the fact that as the angle of hue increases clockwise from 0° to 360°, the color it represents changes gradually from red, to yellow, green, cyan, blue, magenta, ..., and back to red. The color function behaves a periodical or circular one.

This paper is organized as follows. A brief overview of the mathematical foundation of our method is given in Section 2. The segmentation procedures are thoroughly discussed in Section 3. Finally, discussions, conclusions and perspectives are presented in Section 4.

### 2. Iterative Otsu's approach based on circular histogram

#### 2.1 Otsu's approach

Otsu's approach is generalized on the base of least square method [4]. Its basic principle is to get the variance of the two parts of an image that is segmented by a threshold. We can use the threshold to segment the

image when the inter-variance of the two parts gets the maximum value.

Let a histogram  $h(x)$ ,  $x = 0, 1, \dots, L-1$ , where  $L$  is the number of gray levels in the case of gray level images. Suppose  $u_i, i = 1, 2$  is the mean gray value of the two parts separated by a threshold  $k$ , while  $w_i, i = 1, 2$  is respectively the occurrence probability of  $u_i, i = 1, 2$ . So the mean gray value is  $u = w_1u_1 + w_2u_2$ , the inter-variance between two parts is as follow:

$$d(k) = w_1(u_1 - u)^2 + w_2(u - u_2)^2 \quad (1)$$

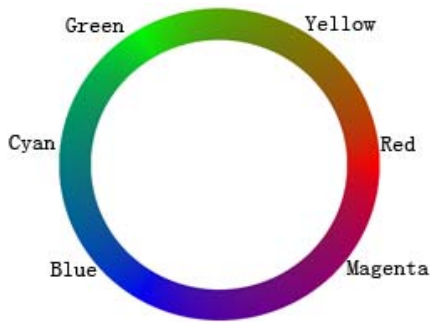
Here  $k$  is the Otsu threshold and  $u_i$  are functions of  $k$ . The mean gray value of two parts is used to the formula of (1), we can get results as follow:

$$d(k) = w_0w_1(u_1 - u_2)^2 \quad (2)$$

Shift  $k$  from 0 to  $L-1$  in order to find out a  $k^*$  satisfying  $d(k^*) = \max\{d(k)\}$ . Afterwards, the best segmentation can be realized by the threshold  $k^*$ , the larger the variance, the clearer the difference between the two parts: background and object. Otsu's approach is a steady one in image segmentation though it is not the best method in some cases.

## 2.2 Iterative Otsu's approach based on Circular Histogram

A perceptual HSI color space is a 3-D cylindrical coordinate system  $(r, \theta, z)$ , the attribute  $H$  being



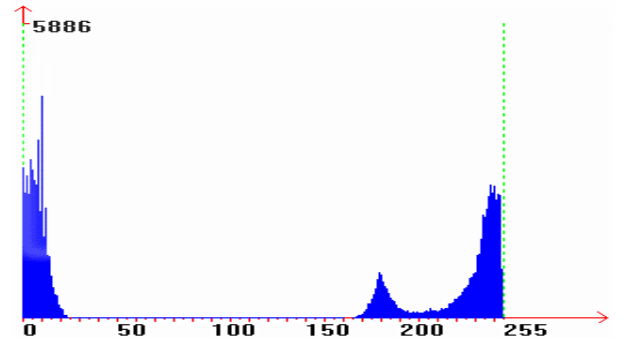
**Figure 1.** Hue changes periodically as the angle increases

represented by the coordinate  $\theta$  in the range of  $[0^\circ, 360^\circ)$ .

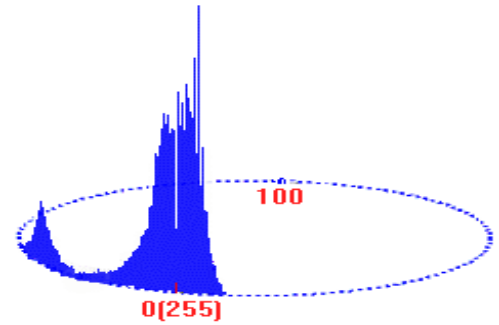
As  $\theta$  increases counterclockwise from  $0^\circ$  to  $360^\circ$ , the human visual perception of hue changes gradually from red, to yellow, green, blue, magenta, ....., and back

to red. The color function behaves a periodical or circular one, as shown in Fig. 1.

However, if we directly represent a hue histogram as a traditional linear histogram (Fig. 2 (a)), we will lose the periodicity of the hue attribute. A single threshold will divide all colors into two meaningless clusters. Thus we adopt the circular histogram shown in Fig. 2 (b) [5]. And we propose a modified Otsu's algorithm, an iterated Otsu's algorithm, based on circular histogram. In the algorithm, we try to find two thresholds  $k_1^*$  and  $k_2^*$  to separate the loop in Fig. 1 into two parts, as in Fig. 2 (b), in place of one threshold  $k^*$ , which is selected to separate the linear histogram into two parts, as in Fig. 2 (a). Note that in Fig. 2, the hue has been quantized to 256 levels.



(a) Linear histogram



(b) Circular histogram

**Figure 2.** Histogram of hue image

The iterated Otsu's algorithm is described as follows:

**Step 1** Initialize the iteration number:  $i = 0$ ;

**Step 2** Get the first two hue values  $p_1$  and  $p_2$  which are corresponding to two peaks of the circular histogram. This can be done by smoothing the histogram with a Gaussian filter of a changing variance  $\sigma^2$ . As  $\sigma^2$  decreases from an initial value, the histogram becomes smoother

and smoother. The process stops until exact 2 peaks are remained. The filtering can be easily realized by FFT, because the circular convolution can be realized by multiplication in frequency domain, provided the histogram signal to be filtered is periodical. Fortunately, this is true in the case of circular histogram.

**Step 3** Get the initial threshold for  $k_1$  :

$$k_1^i = (p_1 + p_2) / 2.$$

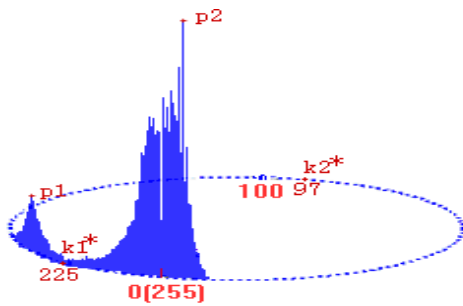
**Step 4** Divide the circular histogram into a linear histogram by  $k_1^i$ .

**Step 5** Calculate a new threshold  $k_2^i$  with Otsu's approach.

**Step 6** Divide the circular histogram again into a linear histogram again by  $k_2^i$ .

**Step 7**  $i = i + 1$ . Calculate the new threshold  $k_1^i$ . If  $i = 1$  or 2, go to step 4. Otherwise, if the current thresholds  $(k_1^i, k_2^{i-1})$  are equal to the previous thresholds  $(k_1^{i-1}, k_2^{i-2})$ , go to step 8, otherwise go to step 4.

**Step 8** The iteration stops,  $k_1^i, k_2^{i-1}$  being set respectively to the two optimal thresholds  $k_1^*, k_2^*$ .

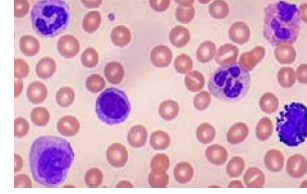


**Figure 3.** Results of our iterated approach

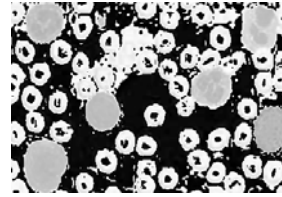
### 3. Segmentation Procedures

The microscopic blood images are shown in Fig. 4. Among them, (a) is the original color image, (b) is the H component of image (a), and (c) is the S component.

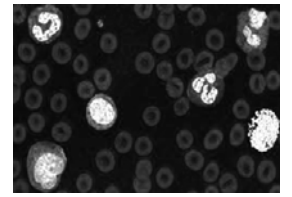
Observations and experiments have shown that the H component can be used to locate the cells, while the S component can be used for nuclei detecting. The I component is not used because its values are sensitive to uneven illuminations.



(a)



(b)



(c)

**Figure 4.** Original Blood image

Upon the above discussions, three steps are taken here to implement the segmentation procedures:

(i) Locating cells. The aim of this step is to locate all the cells within the H component images, but not to get the final segmentation results. The proposed iterative Otsu's approach based on circular histogram is applied here in order to threshold this image. Then, mathematical morphology operations are applied to fill the possible holes and gaps, eliminating the fragments at the same time.

(ii) Detecting cells. The cells can be detected out with the help of the rectangle of each cell gotten from the above step. This detecting processing is done inside the rectangles, like in step (i), the iterative Otsu approach based on circular histogram, mathematical morphology operations are applied. The detected cells image is shown in Fig. 5. The closed-boundary of each cell is defined here for next step.

(iii) Detecting nuclei. The last task is to find out the nucleus (or nuclei) for each cell. Traditional Otsu's approach is employed in the S component image to threshold each single cell image and mathematical morphological operations are employed. The result is shown in Fig. 6.



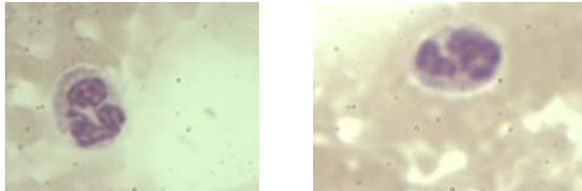
Figure 5. Detected cells image area



Figure 6. Detected nucleus image areas

#### 4. Discussions, Conclusions and Perspectives

The above described method is applied to some other blood microscopic images. The results are shown in Fig. 7 on two typical images, where (a) are the original color images, the segmented cell image areas are shown in (b) and the segmented nucleus image areas are shown in (c). Experimental results show that our method works successfully in the segmentation of white blood cell images: the cells and their nuclei are detected out effectively.



(a) Original cell images



(b) Detected cell image areas



(c) Detected cell nuclei areas

Figure 7. More segmentation results

Still, there are more works to be done in order to perfect our approach, the current segmentation results rely too much on the staining methods effects, and the shapes of the nuclei are not so good-looking.

#### Acknowledgements

This paper is partly supported by Provincial Science Foundation of Jiangxi (No 0511076)

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