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The study of blood smear as the analysis of images of various objects

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Abstract
Processing of microscope images in medicine is one of the priority research areas. Among the many medical imaging follows allocate the image of blood preparations. This is due to the fact that study of the image of blood preparations allows to conduct a comprehensive diagnosis of human health state. However, the specific complexity of visualization process of blood preparations and their subsequent processing with the use of automated processing determine the necessity to study different possibilities to use any approaches for image processing. We consider the image of blood preparations is a complex image. For image analysis of blood preparations' structure, we use the method of color segmentation. For image analysis, we also use the methodology of the human-machine. This allows you to clarify the structure of the image of blood preparations. We give some of examples that show the effectiveness of the proposed image processing of blood preparations.

Keywords: Erythrocytes, segmentation, blood plasma, blood cell, white blood cells, microscope image analysis

Introduction

Blood is a body fluid in human. Blood delivers nutrients and oxygen to the cells and transports metabolic waste products away from those cells. Blood consists of a liquid portion (blood plasma) and cells – the formed elements (erythrocytes, leukocytes and thrombocytes) [1, 2]. Blood is a complex structure.

Blood test is important for the diagnosis of human diseases. The calculation and classification of blood cells allow to evaluate and diagnose a vast number of diseases. But, nowadays the morphological analysis of blood cells is performed manually by skilled operators. This involves numerous drawbacks, such as slowness of the analysis and a nonstandard accuracy, depending on the operator skills.

It is necessary to automate the calculation of blood cells. One such directions of the analysis is the use of a common ideology of image processing. This is due to the fact that such studies allow conducting the comprehensive diagnosis of human health state, identifying and preventing the development of diseases in the early stages, providing an additional research in non-standard symptomatic forms of rare diseases.

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However, the specific complexity of visualization process of blood preparations and their subsequent processing with the use of automated processing determine the necessity to study new possibilities to use new approaches for image processing. This fact was the basis for considering the possibility to use different methods for image processing of blood preparations.

Materials and Methods

Methodology of image processing in medical image study

Image processing is one of the areas of data mining and method for extracting additional information about processes under experimental study. Image processing methods allow us to determine the information that is difficult to see. At the same time such information is an objective as obtained by using interactive analysis.

The standards of perception of medical information can be formed in different systems such as roentgenograms, tomography pictures and other pictures received with different special devices in optical or not in optical range. These circumstances impose certain features and restrictions, both on the nature of considered standards of perception, and on possibilities of their analysis, additional data accessing about outward things. We can also talk about different methods that form the basis of functioning of different computer vision system. Nevertheless, despite the ability of using the various methods of processing and analysis of received visual image in different computer vision systems, it should be considered the specifics of

how these images are displayed, as well as key tasks these computer vision systems fulfill.

In this study we consider medical images, which are made under the microscope. This defines the processing methods for images of blood preparations.

Color segmentation as a tool for image analysis of blood preparations

The main objective of the use of image methodology in the study of blood cells should be called clustering of objects in the image. It allows realizing the various procedures of image segmentation for their subsequent analysis and resulting of certain conceptual positions for acceptance of corresponding solutions on the basis of the analysis of incoming images. Then, based on the ratio of different clusters we can talk about the degree of manifestation of human diseases.

The image of blood preparations can be represented by the following formula:

$$D = P \cup \bigcup K_1 \cup \bigcup K_2 ... \bigcup K_n , \quad (1)$$

where D – general field the image of blood preparations,

P – blood plasma,

$\bigcup K_1$ – aggregate of blood cells type 1,

$\bigcup K_2$ – aggregate of blood cells type 2,

$\bigcup K_n$ – aggregate of blood cells type n.

Each set of blood cell has a different color. It makes possible to apply the segmentation by color. It can also calculate the portion of each cell group in the general image of blood preparations ($\bigcup K_n$). But we should bear in mind that the staining of blood preparations can affect the quality of segmentation. Therefore we say about the need to apply the methodology of the human-machine in image processing of blood preparations.

This problem can be solved on the basis of:

the choice of markers for individual objects (performed by a person);

the detection of objects from an image (performed by a computer);

the calculating the proportion of an object in the image (performed by a computer).

The detection of objects from an image is based on identification of the object by color. This is a standard procedure in the image processing, which allows you to select an area of a predetermined set of colors [3].

The following formula is used for calculating the proportion of an object in the image:

$$V_i = \frac{s_i}{S} 100\% , \quad (2)$$

where V_i – proportion of i subjects in the image ($i = \{P, \bigcup K_n\}$);

s_i – area of the i subjects in the image ($i = \{P, \bigcup K_n\}$);

S – image area D.

Then,

$$S(D) = s_P + \sum_{j=1}^{n} s_{\bigcup K_j} , \quad (3)$$

or

$$V(D) = V_P + \sum_{j=1}^{n} V_{\bigcup K_j} = 100\% . \quad (4)$$

We can also write:

$$S(D) = S(D_1) + S(D_2) + ... + S(D_m) , \quad (5)$$

where $S(D_m)$ – area of individual m parts of the image D ($D = D_1 \cup D_2 \cup ... \cup D_m$). It makes possible to analyze the parts of the image of blood preparations.

This approach allows to study the image of blood cells for different magnification of the microscope.

Data analysis

For the analysis, we use different images of blood preparations (these images are in public internet access):

Fig. 1: The cells of human blood (using the 10X objective lens, image size 300x254 pixels).

Fig. 2: The cells of human blood (using the 60X objective lens, Image size 872x699 pixels).

For Fig. 1 and Fig. 2 are used two different types of staining of blood preparations.

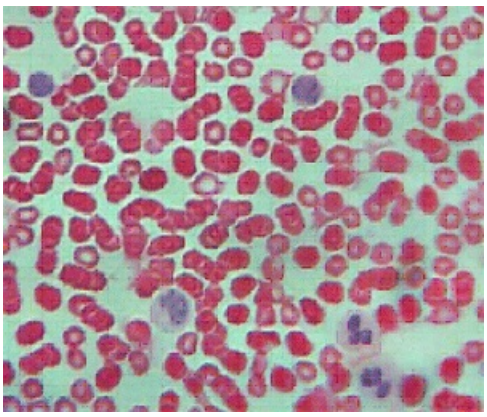

Figure 1. Cells of human blood (using the 10X objective lens)

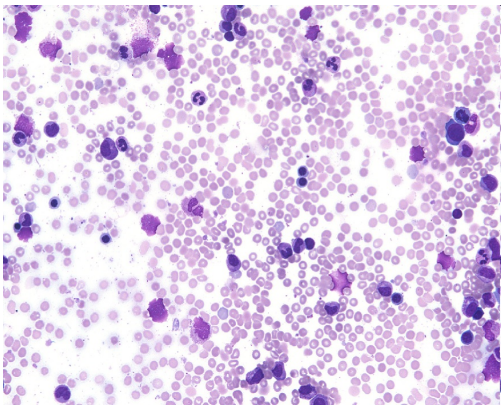

Figure 2. Cells of human blood (using the 60X objective lens)

On Fig. 1 and Fig. 2 we can see different blood cells. On Fig. 1 we can see erythrocytes (red color and pink color) and white blood cells (cells with dark blue nuclei). On Fig. 2 we can see erythrocytes (violet color) and white blood cells (cells with dark blue nuclei).

Results

For image analysis of blood preparation, shown in Fig. 1, we define the markers of individual image objects. Fig. 3 shows markers of individual objects (for Fig. 1).

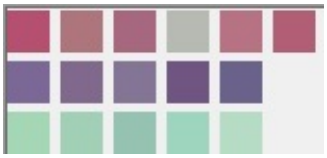

Figure 3. Markers of individual objects for Fig. 1.

The first line of Fig. 3 shows the markers for the identification of red blood cells. The second line of Fig. 3 shows the markers for the identification of white blood cells. The third line of Fig. 3 shows the markers for the identification of blood plasma. The result of detection of objects from the image (for Fig. 1) is shown in Fig. 4.

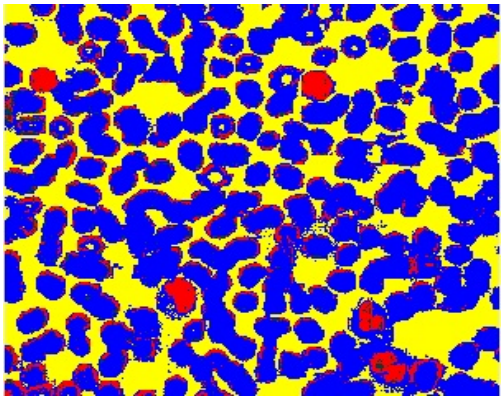

Figure 4. Result of detection of objects for the image for Fig. 1.

Fig.4 shows: erythrocytes (blue color), blood plasma (yellow color), white blood cells (red). Analysis of Fig. 4 shows that in the Fig. 1 the erythrocytes make 54.4% of the image of blood preparation, blood plasma – 35.7% and white blood cells – 8.9%.

By comparison Fig. 1 and Fig. 4 some errors in identification of objects on the analyzed image of blood preparations can be seen. In order to solve this problem we divide the image of Fig. 1 into several images. At the same time we will choose those parts of the image that are questionable. Such transformations are possible in accordance with formulas 3-5. After clarification of the identification of objects, we received the following result: erythrocytes make 54.1% of the image of blood preparation, blood plasma – 36.2% and white blood cells – 9.7%.

For image analysis of blood preparation, shown in Fig. 2, we also define the markers of individual image objects. Fig. 5 shows markers of individual objects (for Fig. 2).

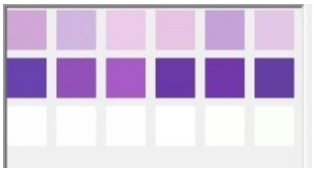

Figure 5. Markers of individual objects for Fig. 2.

The first line of Fig. 5 shows the markers for the identification of red blood cells. The second line of Fig. 5 shows the markers for the identification of white blood cells. The third line of Fig. 5 shows the markers for the identification of blood plasma. The result of detection of objects from the image (for Fig. 2) is shown in Fig. 6.

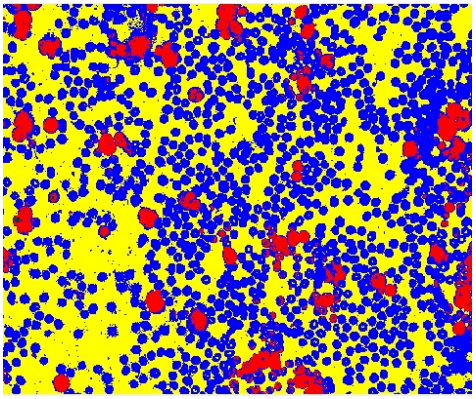

Figure 6. Result of detection of objects for an image for Fig. 2.

Fig. 6 shows: erythrocytes (blue color), blood plasma (yellow color) and white blood cells (red). Analysis of Fig. 6 shows that in the Fig. 2 the erythrocytes make 45.6% of the image of blood preparation, blood plasma – 45.9% and white blood cells – 8.5%. After clarification of the identification of objects, we received the following result: erythrocytes make 45.4% of the image of blood preparation, blood plasma – 46.4% and white blood cells – 8.2%.

The results of identification are better for image on Fig. 2 than for image on Fig. 1. This is due to the use of different staining techniques.

We asked the experts to determine the structure of blood preparations. We compared our results and the results of the experts (Table 1).

Table 1. The results of the comparison in the study of the structure of blood preparations. Shows that the results are comparable.

Blood elements	Our result	Expert 1	Expert 2	Expert3	Expert 4	Expert 5
for the image on Fig. 1						
Erythrocytes, %	54,1	53,8	54,0	54,1	53,4	53,0
Blood plasma, %	36,2	36,6	36,0	35,7	36,8	37,5
White blood cells, %	9,7	9,6	10,0	10,2	9,8	9,5
for the image on Fig. 2						
Erythrocytes, %	45,4	45,1	45,2	45,0	45,1	45,3
Blood plasma, %	46,4	46,5	46,6	46,9	46,8	46,4
White blood cells, %	8,2	8,4	8,2	8,1	8,1	8,3

Discussion

As an example of separate works that use the ideology of image processing for studying of blood preparations, the different research work can be provided:

L. Putzu and C. Di Roberto offer to make calculation of blood cells based on their morphological characteristics [4]. For this purpose each blood cell is described and investigated. At the same time G. Apostolopoulos, S. V. Sinopoulos, E. Dermatas study the shape of blood cells [5]. Identification of the shape of blood cells includes image normalization, features extraction using both histogram of oriented gradients and region covariance features, dimensionality reduction using the independent. But it increases the time of the analysis of blood preparations.

L. K. Jha, B. K. Das, H. S. Dutta offer to study blood preparations on the basis of classical pattern recognition methods [6]. For this purpose the blood cells database was used. But the images of blood cells can be modified. Thus, the result shows a small precision for the identification and enumeration of various types of blood cells.

V. von Hagen, P. Saint-Jean, A. van Driel-Kulker, R. Le Go and J. C. Bisconte consider texture model analysis for the classification of the image of blood cells [7]. Also the work shows that the biologic significance of the texture of blood preparations can not be assessed by mathematical methods alone. V. von Hagen, P. Saint-Jean, A. van Driel-Kulker, R. Le Go and J. C. Bisconte indicate that these methods should be tested with appropriate biologic models. In this case, the analysis of the blood preparations should consider the model of the human-machine. It underscores the importance of using interactive analysis that is used in our approach.

S. Wienert et al. allocate contour of blood cells for their analysis [8]. Thus it is necessary to notice that the classical approaches use a priori information on cell shape features to obtain the contour. Therefore there are questions on determining the accuracy contour of a cell.

J. M. Sharif et al. [9], Z. Liu et al. [10], S. H. Shirazi, A. I. Umar, S. Naz and M. I. Razzak [11] study the segmentation algorithms to highlight the blood cells, and then to determine their quantity. For this reason transform techniques for image enhancement and noise elimination are also used. However, such techniques can introduce distortion in the original image. Therefore, the methods for image enhancement and noise elimination should be used carefully.

Staining of blood smear is also important to study the blood cells. Staining of blood smears allows a better look at the blood cells. But it also creates new difficulties in blood cells analysis. It is important to maintain image contrast [12-15]. At the same time we also need to say that the blood smear is a complex image that consists of a plurality of individual objects. Therefore, we emphasize the importance of using the model of the human-machine for the blood cells analysis.

Disadvantages of the methodology that was used for the study of blood preparations: false point in the calculation of the area affected by the disease may occur. This is due to the automatic allocation of the selected color and also poor quality of the sample of blood preparation.

Advantages of the methodology that was used for the study of blood preparations: can change the size of the marker to select the colors in the image segmentation and to clarify the marker group. This will avoid the occurrence of false points.

The method we used can be applied to study the structure of blood preparations. This is confirmed by the results of research (Table 1).

Conclusion

This work showed the method for the study of structure of blood preparations in order to provide an automated procedure as support for medical activity. In this case we consider the image of blood preparations as a complex image with many different objects. We use color

segmentation method to separate blood components. We also use colored markers. The choice of color markers allows to adjust the results of image processing of blood preparations. We compared our results and the results of experts for the analysis of structure of blood preparations. The received results show that the proposed method is able to identify the various elements of blood preparations.

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