Detection and classification of abnormalities in erythrocytes by techniques of image analysis and pattern recognition

María Elena Cruz Meza, M. en C[1], *Graciela Vázquez Álvarez, Dra.* [2], and *José Cruz Martínez Perales, M. en C* [1]
[1]First and Third Author's *Instituto Politécnico Nacional, Escuela Superior de Cómputo, Departamento en Sistemas Computacionales, Academia de Ingeniería de Software & Academia de Proyectos Estratégicos y Toma de Decisiones, Ciudad de México, mcruzm@ipn.mx, jmartinezp@ipn.mx*
[2]Second Author's *Instituto Politécnico Nacional, Escuela Superior de Ingeniería Mecánica y Eléctrica, Sección de Estudios de Posgrado e Investigación, Departamento de Sistemas, Ciudad de México, gravalvarez15@gmail.com*

Abstract

This document shows the detection of anomalies known as acanthocytes, equinocytes and spherocytes present in blood cells called erythrocytes or red blood cells, through the analysis and processing of images and pattern recognition techniques, in order to assist the specialist doctor in hematology During the medical diagnosis process. The particular interest of this work is that these blood tests are performed with specialized devices and the use of these machines to analyze a blood draw is not very common and time consuming, in addition to the acquisition of these machines requires a high cost .

Keywords. Patterns. Recognition, Image Analysis, Cytology, Hematology, Acanthocytes, Equinocytes

Resumen

Este documento muestra la detección de anomalías conocidas como acantocitos, equinocitos y esferocitos presentes en células sanguíneas llamadas eritrocitos o glóbulos rojos, a través de técnicas de análisis de imágenes y reconocimiento de patrones, con el fin de auxiliar al médico especialista en hematología durante el proceso de diagnóstico médico. El interés particular de este trabajo se debe a que los análisis de sangre se realizan con dispositivos especializados y el uso de estas máquinas para analizar una extracción de sangre no es muy común y lleva mucho tiempo, además de que la adquisición de estas máquinas requiere un alto costo.

Palabras Clave. Reconocimiento de Patrones, Análisis de Imágenes, Citología, Hematología, Acantocitos, Equinocitos

I. Introduction

Doctors rely on the practice of blood analysis because with this it is possible to investigate almost all diseases, from an infectious or parasitic disease, to cancer in the blood. However, the current procedures to diagnose some of these types of diseases through blood analysis take an exhaustive process with a high cost, both technical and economic. This and the fact that it is important for patients and physicians to have the medical diagnosis quickly and effectively induced to study the problem and support the medical area in the detection of these possible diseases in less time and less cost.

II. process

The values of a complete blood count are usually presented as a function of the number of cells in a specific volume of blood. Normal values may vary slightly depending on the reference range and the machine used in the laboratory and, therefore, the results may be slightly different from one laboratory to another. The normal reference range is typically provided and printed with the results of complete blood count for accurate interpretation. In addition to these results, it is convenient to analyze the samples under a microscope to detect abnormalities in the cells that provide more information for diagnosis. With this work we try to support the specialist analyzing only the morphology of blood cells to automatically detect by means of image analysis and pattern recognition any possible anomaly in blood cells, specifically the possible malformations that may occur in erythrocytes or commonly known as red blood cells.

Hematology is the part of medicine that studies the functioning of the cells that circulate in the blood, the organs that produce them, the diseases of the blood and aspects related to transfusion medicine. The blood cells (Figure 1) are formed in the bone marrow and only when they mature leave this compartment and circulate through the blood, which in turn is a fluid formed by a liquid part (the plasma) and a solid part, constituted by blood cells: erythrocytes (red blood cells), leukocytes (white blood cells) and platelets [1].

The above can be observed graphically in the following image 1.

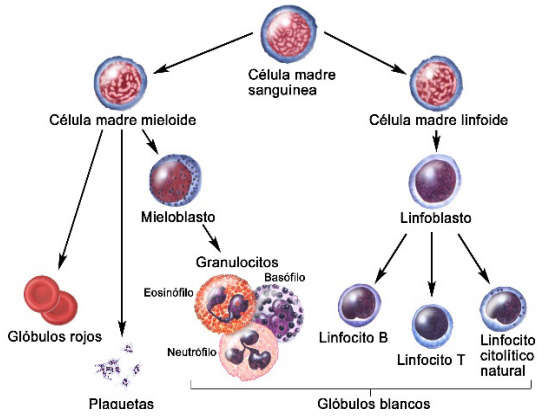

Image 1. Structure of blood cells Source [5].

Abnormalities in erythrocytes Of the previously described elements, our objects of interest will be the red blood cells that present some abnormality, specifically the anomalies of acanthocytes, echinocytes and spherocytes, as illustrated in Image 2.

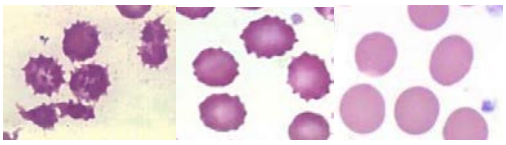

Image 2 Image of erythrocytes with normal forms type a) acanthus, b) echinocytes, c) spheroid

The anomalies suffered by erythrocytes can be classified into three main types that are:

a Shape: erythrocytes that undergo a deformation in their morphology ceasing to be biconcave discs and taking other forms such as half moon, ovals, pencil shape, teardrop shape, among others.

b Size: the erythrocytes can retain their shape and only change in size of the cell: the increase of this is known as macrocytosis and the decrease as a microcyte.

c Color: they retain the shape of a biconcave disc, but their top view shows change and can form figures inside the cell as a form of bullseye target, fish mouth shape, among others. Basically we would be talking about the distribution of hemoglobin.

The anomalies considered in this study have the following characteristics:

Acanthocyte: Erythrocyte with a dented and prickly profile with spicules (3 to 12) of different lengths [1]. Associated diseases are: alcoholic cirrhosis, neonatal hepatitis, poor absorption states, after splenectomy [2].

Equinocyte: This erythrocyte shows short spicules with blunt ends, usually more than 30, distributed regularly throughout the surface of the cell that resembles a sea urchin, from which it derives its name [1]. The associated diseases are: kidney failure, extensive burns, severe dehydration, in the erythrocytes of stored blood, in liver diseases [2]

Spherocyte: Spherical erythrocytes whose diameter is smaller than normal and which appear hyperchromic due to their shape. Characteristically they are found in hereditary spherocytosis but can be seen in newborns with hemolytic disease due to ABO incompatibility and in adults with autoimmune hemolytic anemias and isoimmune [1]. The diseases in which it occurs are: Hereditary spherocytosis (Minkowsky-Chau_ard disease), immunohemolytic anemias, hypersplenism, severe burns, hypophosphatemia [2]

III. Proposed Solution

It is important to note that the anomalies acanthocytes and echinocytes are very similar, so the complexity of their correct detection is high. As for the spherocyte abnormality, it is clearly distinguishable from the previous ones. So to solve the detection of these possible anomalies, the following process is performed:

1 Input: normalization of the images that are to be pre-processed.
2 Pre-processing of the image: various filters are applied to adapt the image for a correct analysis.

The objective of the pre-processing of the image consists of eliminating objects that are not of interest for the analysis of the anomalies, such as platelets, leukocytes or objects caused by the staining of the smear. For this, the images are subjected to a medium filter using a 9x9 dimension kernel, a larger kernel generates loss of information because it deforms the original shape of the cells and a smaller kernel leaves a lot of noise inside the image . To separate the objects from the background the binarization of otsu is applied, since it adequately separates the objects of interest from the background. To eliminate the noise presented in the images we use a morphological operation called opening [6], which will disappear all small objects according to the dimension of the structure element. After the tests it was determined that the dimension of the structure element should be 9x9 and elliptical in shape, since this size and shape disappears most of the noise without having alterations in the objects of interest. In image3 we can see that most of the noise has disappeared, in a) all the small objects that left the smear of the smear disappeared, in b) we can see

how the platelets present after Otsu binarization have been eliminated [9

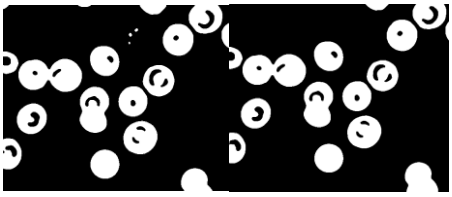

Image 3. Binarized image, on the left Platelets present (noise), right platelet removal

At the moment of eliminating the noise present in the image, the problem of holes or holes in the erythrocytes is generated, which is why we proceed to eliminate the pepper-like noise. In this way all that object is filled with an area greater than 2092 pixels. This process, in turn, sometimes leaves a salt-like noise present, so it is eliminated by applying the medium filter.

3. Segmentation of the objects present in the image to generate the descriptors (selection and extraction of the characteristics).

Once the objects of all the sample images were segmented, they were divided manually into the different classes according to the experience to identify them. This is for the purpose of generating the training files necessary for the classification.

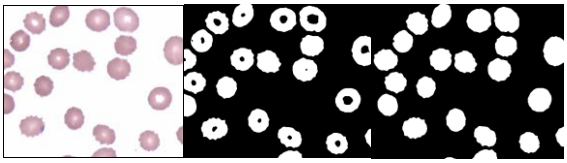

Image 4. a) Original image b) Presence of holes after Otsu, c) Elimination of holes

The process of extracting traits is one of the most important stages in the image classification process, since the effectiveness of the designed classifier depends on the correct selection. Because in the abnormal erythrocytes of the types (classes) acanthocytes, echinocytes and spherocytes produce changes in the shape of the cell, with respect to healthy erythrocytes, the traits that are selected for the classification of the cells and with them form the descriptors of The classes to be discriminated against are [4]:

• Chain code. (Image5)
• Firms. (Image 6)
• Euler's number.
• Circularity and circularity form factor.
• Skeleton. (Image 7)

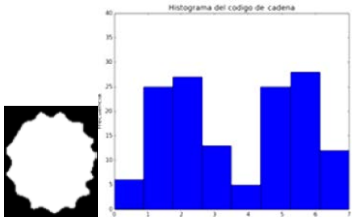

Image 5. On the left an equinocyte and on the right the histogram of its generated chain code.

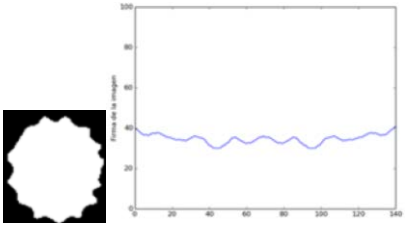

Image 6. On the left an equinocyte and on the right the histogram of his signature

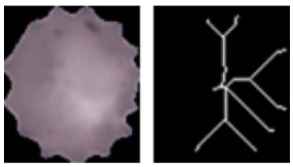

Image 7. On the left an equinocyte and on the right its morphological skeleton

4. Classification and detection of anomalies present in the image (acanthocytes, echinocytes or spherocytes) through pattern recognition techniques.

The k-NN method is a very simple technique [8] and for the case of the chosen descriptors, this method is sufficient to carry out the discrimination of the three abnormal forms of erythrocytes.

IV. RESULTS AND CONCLUSIONS

Blood extension was carried out, that is to say, the maintenance of the samples, since the digital images of the anomalies that are intended to be studied were not available, thus the necessary smears with such anomalies could be obtained. To carry out this process, the collaboration and authorization of the General Hospital "Dr. Eduardo Liceaga ", under the support and supervision of the Pharmaceutical Chemistry Emma R. Mendoza, in charge of the Hematology laboratory To obtain the digital images of the erythrocyte cells, a specialized team was used and facilitated by the National School of Biological Sciences, with the collaboration of Dr. Elba Reyes Maldonado and Master Erika Rosales, thanks to the vast experience of Mendoza Chemistry and the researchers, Ms. Rosales and Dr. Reyes, the preparation of blood samples and the digitalization of In this way, the bank of images with a ".TIFF" format of 2560x1920 pixels corresponding to 47 samples (15 samples of acatocytes, 15 samples of echinocytes and 17 samples of spherocytes) was successfully carried out.

Detection of erythrocytes with abnormalities.

The graphs below illustrate the results obtained in the classification tests, with the re-substitution method for a k-NN that considers three neighbors and a Euclidean metric.

Table 1. Matrix of confusion in anomaly of cell shape

Anomalía	Acantocitos	Equinocitos	Esferocitos	Total	% clasificación
Acantocitos	254	65	6	325	78.15%
Equinocitos	0	91	6	99	93.81%
Esferocitos	0	0	99	99	100.00%

As shown in the table above, the effectiveness of the detection of the abnormal cells corresponding to the erythrocytes, the type of the echinocytes yielded 93.81% and the spherocytes 100%. This was guaranteed because the characteristics selected to perform the classification of the echinocytes and spherocytes were appropriate, so we believe that the percentage of correct classifications for the shape of the cell is high.

The lowest percentage was given in the recognition of acanthocyte type abnormality with 78.5%. The 21.5% that were not classified correctly, we believe is mainly due to the fact that the number of samples of this normal form was small and for the training of the system, these were obtained by modifying the cells by a chemical procedure, the effect is known as a cell cre- ation, which means that they acquire a serrated appearance by decreasing their volume [16]. Although the results are very encouraging to those expected, it would be convenient to perform laboratory tests with images completely from diseased cells and a larger volume of these.

REFERENCES

[1] Campuzano Maya G. (2007), Del hemograma manual al hemograma de cuarta generación, Editora Médica Colombiana S.A

[2] Jaime Pérez, J. C.; Gómez Almaguer D., (2015). Hematología La sangre y sus enfermedades. Mc Graw Hill,4a. Ed.

[3] López Farre, C. A.; Mayaca Miguel (2009). Libro de la salud cardiovascular del Hospital Clínico San Carlos y de la Fundación BBVA. Fundación BBVA, primera edición ed.

[4] Gonzalez R, Wood R., (2008). Digital Image Processing. Prentice Hall. 3a. Ed.

[5] Pajares Gonzalez J. M., Martinsanz; Cruz García, (2008). Visión por computador, imágenes digitales y aplicaciones. Ed. RA-MA.

[6] Shih F. Y., (2017), Image Processing and Mathematical Morphology: Fundamentals and Applications, pp. 344-371, CRC Press. Copyright.

[7] A. Bradski, Gary; Kaehler, (2008). Learning OpenCV. O'Reilly Media, Inc.

[8] Vitria J., Sanches J. M., Hernández M. (2011). Pattern Recognition and Image Analysis: 5th Iberian Conference, IbPRIA.

[9] Beucher S., (2010). Image segmentation and mathematical morphology. http://cmm.ensmp.fr/

[10] Mejía M, Alzate M, Rodríguez J. (2016). Clasificación automática de glóbulos rojos en Frotis de sangre periférica. Rev Univ Ind Santander Salud. http://dx.doi.org/10.18273/revsal.v48n3-2016005

[11] Cerecedo Díaz B. S. R. (2013). Fisiopatología y métodos de diagnóstico de las anemias hemolíticas por alteración de la membrana eritrocitaria, Master's thesis, Escuela Nacional de Ciencias Biológicas.

[12] Asunción Batugo G. K., (2012), Tésis de Maestria, Reconocimiento automático de melanomas mediante técnicas de visión por ordenador y reconocimiento de patrones. http://e-archivo.uc3m.es/bitstream/handle/10016/17028/

[13] Ramírez-Manzanares, (2017). Reporte de búsqueda, detección y conteo de objetos, Centro de Investigación en Matemáticas. http://www.cimat.mx/~alram/VC/ramirez_segmObjDetecc.pdf

[14] Sysmex, (Mayo 2017) Ematology Sistemization, http://www.sysmex-ap.com/products/diagnostics/hematology/

[15] Grau J. F. P., (2003). Técnicas de análisis de imágenes, Aplicaciones en Biología. PUV, segunda edición ed. Universitat de Valencia, Servei de Publicaciones

[16] Bottani E. J., Odetti H. S, (2006). Química general. Ed. Universidad Nacional de Litoral, segunda edición.

[17] Olachea Cota R., (2010). Tesis de Maestría, Reconocimiento y cuantificación de células de peces en imágenes de cortes histológicos, Instituto Tecnológico de Puebla.