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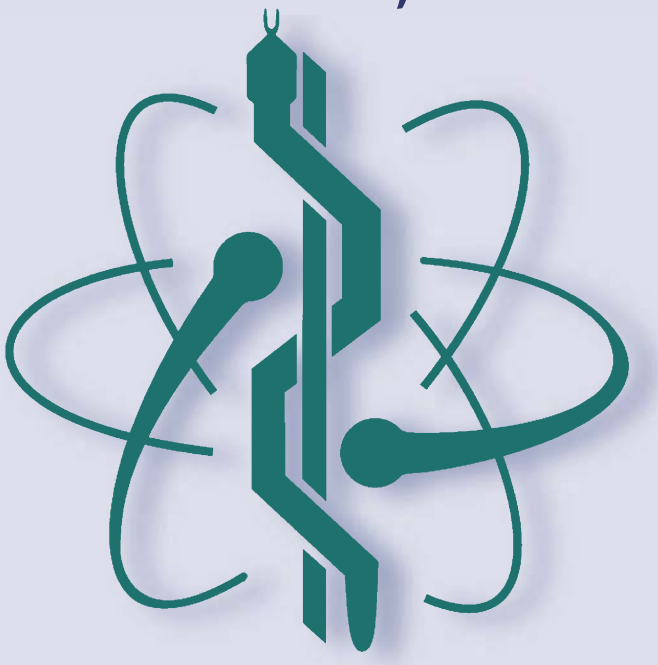

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Editors
Isnardo Torres
Presidente
Asociación Colombiana de Bioingeniería
y Electrónica Médica - ABIOIN
Bucaramanga
Colombia

John Bustamante
Centro de Bioingeniería
Universidad Pontificia Bolivariana - UPB
Medellín
Colombia

Daniel A. Sierra
Escuela de Ing. Eléctrica, Electrónica
Universidad Industrial de Santander
Bucaramanga
Colombia

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Contents

Color morphological reconstruction as a segmentation tool for microscope cell images

J.I. Pastore[12], M. Brun[12], A. Bouchet[12] and V.L. Ballarin[23]

[1] ICYTE, CONICET - UNMDP, Universidad Nacional de Mar del Plata, Argentina.
[2] Department of Mathematics, School of Engineering, UNMDP, Argentina
[3] Department of Electronics, School of Engineering, UNMDP, Argentina

Abstract— **There are numerous segmentation methods for gray level microscope cell images, however in some situations the texture features and roughness are not as relevant as the color for the segmentation task. For example, for detection of histological analysis, the relative sizes of nucleus and cytoplasm, as well as their shapes, are the relevant features, while other characteristics such as texture and roughness have no value in the diagnosis.**

In this context, geodesic reconstruction is one of the image operators, of mathematical morphology, that facilitates image segmentation of individual objects. This operator uses markers to highlight, in the image, objects of interest, in order to separate them from the rest of the scene.

This paper presents a new segmentation method, for microscope cell images, based on mathematical morphology color reconstruction, where the markers can be obtained automatically or semi-automatically. Automatically, when you want to detect those cells that were not removed at the generation of the markers. Semiautomatically when the expert manually selects one pixel for each cell of interest.

Keywords—**Mathematical morphology, color, segmentation, reconstructions, cells.**

I. Introduction

Modern technological development has enabled a huge advance in the histological knowledge. Currently, the use of segmentation algorithms aids the detection and treatment of different lesions. Thus, lots of researchers devote their efforts to develop more powerful algorithms that support diagnostics issued by medical specialists [1]. However, they are still insufficient achievements in this regard. This paper proposes a new segmentation method, for microscope cell images, based on mathematical morphology color reconstruction, where the markers can be obtained automatically or semi-automatically.

The automatic segmentation of cells microscopic images can be considered to be one of the major hurdles for a robust analysis. Automatic or semiautomatic segmentation techniques attempt to improve sensitivity and specificity by detecting, segmenting and classifying the cells and later obtain the cytoplasm or nuclei shape. To solve this problem, in this work, color mathematical morphology reconstruction technique was developed based on a new local defined ordering [2]. The marker image can be obtained automatically or semi-automatically. Automatically, when you want to detect those cells that were not removed at the generation of the markers. Semiautomatically when the expert manually selects one pixel for each cell of interest. In the first case, the marker image is obtained performing successive dilations with a structuring element which size is related to the size of the nucleus of the cell that wants to be removed. In that way we obtained the image mask automatically. Also the expert can also select by clicking a particular cell to segment it.

Experimental results show that the proposed color morphological reconstruction can be efficiently used in the segmentation of cells.

II. Mathematical morphology in color space

Mathematical Morphology (MM) has a strong theoretical basis supported on concepts of geometry, algebra, topology and set theory. The principal idea of this theory is to compare the objects of interest with a set of predefined and known geometry, called "structuring element". The use of different shapes and sizes for the structuring element (SE) allows testing and quantifying how the structuring element "is, or is not contained" in the image [3-4].

The MM has been studied and successfully applied in biomedical image segmentation. The process of segmenting an image consists in generating a partitioning of the image set in groups of pixels. Its objective is to simplify or change the representation of an image in a more meaningful one to facilitate the analysis. The segmentation is used either to locate objects of interest or to determine the boundaries within an image [5].

The MM had been introduced as a processing technique for binary images, which were regarded as sets; therefore, its elementary operations are based on the set theory [3-4]. However, the extension to sets of grayscale images, using the umbra concept [3], introduced a generalization of the basic morphological operations. The grayscale morphology is based on the lattice theory, which implies a partial ordering of the data within the grayscale images. Therefore

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erosion and dilation, the fundamental operations of MM, are defined within a complete lattice as the operations which distribute over the infimum and the supremum [6].

The erosion and the dilation of an image f, using a flat SE g, are defined as follows [6]:

$$\varepsilon_B^{\leq_\Im}(f) = \inf_{s\in B}^{\leq_\Im}\{f \circ \tau_s\} \quad (1)$$

$$\delta_B^{\leq_\Im}(f) = \sup_{s\in B}^{\leq_\Im}\{f \circ \tau_{-s}\} \quad (2)$$

It can be noticed that the basic morphological operations involve finding an infimum and a supremum for the points within a local region, given by the SE positioning. For a general account on mathematical morphology the interested reader should refer to the two pioneer books by Serra [3-4]. Fundamental references to works which have studied the theory of vector morphology theory are [7-11].

While the extension from binary to grayscale images is a natural one, the extension to color or multivariate images is not straightforward, because of the vectorial nature of the data and the difficulty in finding a suitable ordering for it.

A color can be represented by different algebraic structures. The color spaces provide a way to specify order and manipulate colors. These representations correspond to a subset of a three-dimensional coordinate system in which each color is represented by a point univocally. A color image can be modeled as a function $f:\Omega\subset\mathbb{R}^2\to\Im\subset\mathbb{R}^3$ where $\Im$ represents a color space. As mentioned above, the definition of morphological operators needs a complete lattice structure to spatial structures [6], i.e., the possibility of defining an ordering relationship among the points to be processed.

Being $\Im\subset\mathbb{R}^3$ a color space and $\leq_\Im$ a proposed order that gives to $\Im$ a structure of complete lattice. The space of functions $f:\Omega\subset\mathbb{R}^2\to\Im\subset\mathbb{R}^3$ with the order $\leq$ defined as: $f,g:\Omega\subset\mathbb{R}^2\to\Im\subset\mathbb{R}^3$, $f\leq g$ if and only if $f(x)\leq_\Im g(x)\ \forall x\in D$, has a lattice structure (Serra, 1982; Talbot et al., 1998). Indeed, $\forall f,g:\Omega\subset\mathbb{R}^2\to\Im\subset\mathbb{R}^3$, $\exists\inf(f,g),\sup(f,g):\Omega\subset\mathbb{R}^2\to\Im\subset\mathbb{R}^3$ defined by:

$$\left.\begin{aligned}\inf(f,g)(x) &= \inf_{(\Im,\leq_\Im)}\left(f(x),g(x)\right)\\ \sup(f,g)(x) &= \sup_{(\Im,\leq_\Im)}\left(f(x),g(x)\right)\end{aligned}\right\}\forall x\in\Omega$$

This allows defining the basic operations of erosion and dilation for a color image $f:D_f\subset\mathbb{R}^2\to\Im\subset\mathbb{R}^3$ by a SE B following Eq.1 and Eq.2.

A central aspect of the MM is the construction of any operator as a combination of basic operators: erosion, dilation and the operations supreme and infimum. The formal definition of this decomposition is given by a formal language called Morphological Language [7-8]. Particularly, the reconstruction operator, which is a very useful tool provided by mathematical morphology, is employed not only in binary images but also in grey levels. Besides, it can be applied to different stages of image processing such as filtering, segmentation or features extraction. In general, it is introduced as part of a set of operators called geodesic operators [12-13]; and it basically consists in extracting the connected components of an image from a marker image.

A. Geodesic morphological operators

The geodesic reconstruction is one of the operators of mathematical morphology that facilitates image segmentation. This operator allows, by using markers, highlighting an image objects of interest, in order to remove them from the rest of the scene. That means geodesic operators are useful when you want to process a subset of the space analyzed. The geodesic reconstruction operator employs successive dilations (erosions) to a marker image whose result is delimited by a mask image [12-13].

The definitions of geodesic erosion and dilation are closely related to the geodesic distance [14].

The *geodesic erosion* of an image f (called mark), by an structuring element *b*, conditioned to g (called mask), $g\leq f$, is defined as:

$$\varepsilon_{b,g}^{(1)}(f) = \varepsilon_b(f)\vee g \quad (3)$$

being $\vee$ the supremum.

First, the mark image f is eroded. Then the supremum between the eroded image and the mask is calculated. The visual effect of this type of erosion is that the mask retains the marker so that it does not disappear (the contraction of the marker is limited). In this case the geodesic erosion is greater than or equal to the mask, it is also an increasing and anti-extensive operation.

The geodesic erosion of size n, with $n\geq 1$, of an image f, by an structuring element *b*, conditioned to g, is defined as the iteration of geodesic erosions of increasing size, i.e.:

$$\varepsilon_{b,g}^{(n)}(f) = \underbrace{\varepsilon_{b,g}^{(1)}(\varepsilon_{b,g}^{(1)}(.....\varepsilon_{b,g}^{(1)}(f)))}_{n-times} \quad (4)$$

The *geodesic dilation* of an image f (mark), by a structuring element *b*, conditioned to g (mask), $f\leq g$, is defined as:

$$\delta_{b,g}^{(1)}(f) = \delta_b(f)\wedge g \quad (5)$$

being $\wedge$ the infimum.

The mask acts as the limit of the marker image dilated therefore $\delta_{b,g}^{(1)}(f) \leq f$. The geodesic dilation, like classical dilating, is a growing and extensive operator.

The *geodesic dilation* of size n, $n \geq 1$, of an image f, by an structuring element b, conditioned to g, like the geodesic erosion, is defined as the iteration of geodesic dilations of increasing size, i.e.:

$$\delta_{b,g}^{(n)} = \underbrace{\delta_{b,g}^{(1)}(\delta_{b,g}^{(1)}(.....\delta_{b,g}^{(1)}(f)))}_{n-times} \quad (6)$$

The geodesic erosion and dilation have the particularity that they allow, when iterated until stability, the definition of powerful algorithms of morphological reconstruction. Both the geodesic erosion and dilation converge in a finite number of iterations [15].

Reconstruction by dilation of a mark image f from a mask image g, both with the same domain $f \leq g$, is define as the geodesic dilation of f conditioned to g until stability and it denotes $\rho_g(f)$.

The *gray level reconstruction* $\rho_g(f)$ of an image g with respect a marker image f, $f < g$, is obtained iterating successively geodesic dilations in gray levels, by a structuring element b, until new changes do not occur. That means:

$$\rho_g(f) = \bigvee_{n \geq 1} \delta_{b,g}^{(n)}(f) \quad (7)$$

As g acts like constraint, $\rho_g(f) \leq g$, so at the end of the geodesic dilation produces no change in the image. Reconstruction by dilation is an anti-extensive operation.

Each geodesic dilation of the reconstruction is performed from the geodesic dilation of the previous iteration. Thus the marker can progressively reducing its intensity in the spread under the mask

Similarly the dual reconstruction $\rho_g^*(f)$ with respect to an image g is defined $g \leq f$, as:

$$\rho_{b,g}^*(f) = \bigwedge_{n \geq 1} \varepsilon_{b,g}^{(n)}(f) \quad (8)$$

The idea is to iterate several times to spread the minimum (maximum) value of each component till the entire component is homogenized. That minimum (maximum) represents the gray level of the reconstruction of the object, so that the reconstruction of an image in gray levels will not be perfectly reconstructed, as it happens in the binary case.

B. Proposed Method

The proposed method to segment the cells can be summarized in the following four steps:

Step 1: First we define a total order in the RGB color space. In order to define the morphological operators involved in the segmentation, we use the previously defined order in [16]. This order have three stages: The order between two colors is defined first observing the average intensities, if it matches we take the distance to the color of the central element of the structural element and finally if there is a new match we used the order predefined in the decision window.

Step 2: we define the geodetic operators in the complete lattice generated by the former order.

Step 3: the marker image is determined.

Step 4: the dual reconstruction is applied using the marker image of the previous step.

Figure 1 show the proposed color morphological reconstruction applied to a synthetic image. The marker image was obtained performing a dilation with a structuring element. The size of the structuring element is related to the size of the object that wants to be removed by reconstruction. In that way we obtained the marker image automatically.

III. RESULTS

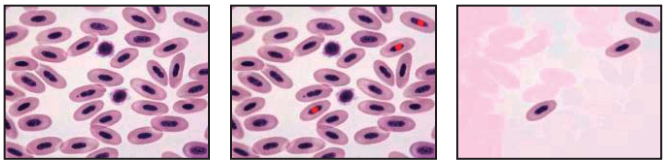
As an example of application, we applied the proposed color segmentation method to cell images from different sources.

Figure 2 shows an example of an image of avian erythrocyte [17], or red blood cells, which also show their nuclei.

We also tried our segmentation approach on these images, first with manual selection of cells, and then with automatic marker detection, with an structuring element of size 11. Figures 2 and 3 show the respective results.

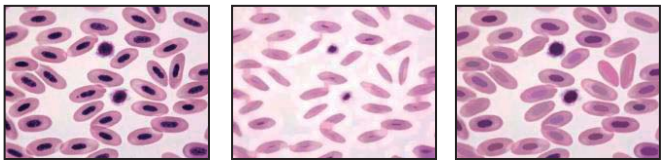
(a) (b) (c)

Figure. 1: Result of the propose method. (a) Original image. (b) Marker image. (c) Result of the dual reconstruction.



(a) (b) (c)

Figure 2. Avian erythrocyte sample. b) Manually selected markers in red. c) Segmented image, where we can see the three selected cells.



(a) (b) (c)

Figure 3. Avian erythrocyte sample b) Automatic selection of markers. c) Reconstructed image.

We can see that in all cases we obtain a good recovery of the marked cells. In the last case, because of the overlap between cells, the reconstructed image contains additional cell regions. This issue is not present on the previous examples, where the cells were shaped in a more consistent shape.

We also applied the proposed segmentation method to frog red blood cells, called also erythrocytes and also to nasal mucosa in allergic rhinitis obtained from a nassal cytology. Both cases were successfully segmented.

IV. CONCLUSIONS

We proposed here a new method for color image segmentation based on color reconstruction, for application on segmentation of cells images. The algorithm works directly on color images, avoiding the need to convert them to gray scale images, with the loss of information that it produces, and the final results are the full selected cells, reconstructed from original markers. Another advantage of this approach is that, because of the use of the new color image operators, no false colors are created in the process, and the resulting image contains exactly the same color present in the original image. As we can see in the result, the algorithm can perform properly in different situations, where cell nucleous are well defined, with option to select the cells manually, or using an automatic marker algorithm.

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