

# SEGMENTATION OF MULTISPECTRAL IMAGES USING MATHEMATICAL MORPHOLOGY AND AUTOMATIC CLASSIFICATION

## *Application to Microscopic Medical Images*

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**Abstract:** In this paper, a new color segmentation scheme of microscopic color images is proposed. The approach combines a region growing method and a clustering method. Each channel plane of the color images is represented by a set of regions using a watershed algorithm. Those regions are represented and modeled by a Region Adjacency Graph (RAG). A novel method is introduced to simplify the RAG by merging candidate regions until the violation of a stopping aggregation criterion determined using a statistical method which combines the generalized likelihood ratio (GLR) and the Bayesian information criterion (BIC). From the resulting segmented and simplified images, the RGB image is computed. Structural features as cells area, shape indicator and cells color are extracted using the simplified graph and then stored in a database in order to elaborate meaningful queries. A regularization step based on the use of an automatic classification will take place. Results show that our method that does not involve any a priori knowledge is suitable for several types of cytology images.

## 1 INTRODUCTION

The image segmentation is an essential step of low-level processing of imagery. It aims to split the image into disjoint regions that are generally homogenous in terms of color and texture. Various algorithms and segmentation methods can be found in the literature and can be divided into several categories: clustering methods, edge-based techniques, region growing process and mathematical morphology. Please refer to (Lucchese and Mitra, 2001) for more details about those state-of-the-art techniques. Approaches combining some of these methods were also proposed in (Lucchese and Mitra, 2001) and (Lezoray and Lecluse, 2007).

In the context of automated analysis of medical microscopic images, we are interested on studying the color image segmentation in order to decompose those images into meaningful entities.

Many of the existing medical images segmentation methods involve *a priori* knowledge as the desired number of classes and use many parameters which are generally difficult to tune

(Lezoray, 2003). In this work, a novel unsupervised method of microscopic color images segmentation is presented in order to cure those weaknesses by reducing the number of parameters. We propose to combine different approaches of segmentation towards this goal.

In this paper, we focused on studying the region growing process based on morphological operations applied to Region Adjacency Graph (RAG). The aim of the region growing process is to simplify the over-segmented images by merging the candidate adjacent regions using morphological operations until the violation of a stopping aggregation criterion. This criterion is determined using the combination of the Generalized Likelihood Ratio (GLR) and the Bayesian Information Criterion (BIC) used in the segmentation method proposed in (El-Khoury et al., 2007).

This paper is organized as follows: in section 2, we describe our morphological segmentation process. In section 3 an original clustering method is proposed. In section 4 we present the hypercube classification. Then, results on cytological images

are shown in section 5. Conclusions and future works are described in section 6.

## 2 MORPHOLOGICAL SEGMENTATION

As the segmentation of color images may be time-consuming and due to the numerous gray-scale methods developed in the literature, we choose to segment independently each RGB channel. First, the images are represented and modeled by a set of regions using a watershed algorithm (Lucchese and Mitra, 2001). An important step in morphological segmentation is to detect the edges of the objects in the image to be segmented. Thus, we proposed to use the color gradient image as input image for watershed algorithm in order to provide the first set of homogenous regions. Instead of using grayscale gradient techniques to individual channels which seems to be inadequate, we decide to apply a RGB color gradient which provides more accurate description of the image.

### 2.1 Color Gradient Watershed

Watershed algorithm constitutes an image segmentation tool based on the mathematical morphology (Lucchese and Mitra, 2001). This process considers the image as a topographic surface on which a flooding action starting from its minima is applied. The basic idea of the watershed construction is to create an influence zone for each regional minima of the image. Generally, the watershed transformation is applied on the gradient of the image representing pixels altitudes.

Different gradients of color images were defined in the literature (Hirata et al., 2000) to detect edges. In our work, we used the supremum of the morphological gradient computed on the red, green and blue images. It is defined as follow:

$$\nabla_B^{\text{sup}} = \vee [\nabla_B^R, \nabla_B^G, \nabla_B^B] \quad (1)$$

where  $\nabla_B$  is the classical Beucher gradient and B the structuring element (Beucher and Lantuéjoul, 1979) applied on every spectral channel and defined as the arithmetic difference between dilation and erosion:

$$\nabla_B = \delta_B - \varepsilon_B \quad (2)$$

This operator yields a grayscale image where each point is the difference between the maximum and the minimum gray levels of the image inside the structuring element.

The resulting gradient image  $\nabla_B^{\text{sup}}$  is used as an input image for the watershed algorithm. The application of watershed algorithm provides an over-segmented image represented by a set of disjoint homogeneous regions  $R = \{R_1, R_2, \dots, R_n\}$  of any sizes and shapes.

The proposed process will constitute a good starting point to carry out the morphological process on RAG.

### 2.2 Region Adjacency Graph

The Region Adjacency Graph (RAG) is an efficient way to manipulate image information because it provides a spatial adjacency view of the regions. One way to represent a RAG consists of associating a node  $P_i$  to each region  $R_i$  and an edge  $A_{i,j}$  to each pair of adjacent regions  $(R_i, R_j)$ . Two regions are defined to be adjacent if they share the same boundary. For more details about the RAG construction, please refer to (Mestar et al., 2007).

To each node  $P_i$  are associated the relevant attributes of the region it represents such as area, perimeter, the mean gray level values of the region, the length of the boundaries shared by adjacent regions and a compactness factor of the regions.

#### 2.2.1 Morphological Region Growing Process on the RAG

In order to simplify the over-segmented regions and obtain only the meaningful ones, we develop an algorithm based on region growing process applied on the RAG. In our case, the region growing process starts with a region  $R_i$  already provided by the watershed algorithm and then iteratively adds to  $R_i$  neighboring regions  $V_A(R_i)$  which share some spectral and spatial properties.

$V_A(R_i)$  is defined as follow:

$$V_A(R_i) = \{R_j \in R, (R_i, R_j) \in A\} \quad (3)$$

where A is the set of edges separating pairs of adjacent regions.

This region growing process on RAG is based on applying morphological operations such as opening and closing operations ((Mestar et al., 2007),(Pesaresi and Benediktsson, 2001)) defined respectively as follow:

$$\gamma(\text{RAG})(R_i) = \text{Min}(\text{RAG}(R_i), \text{Max}(\text{RAG}(R_j))) \quad (4)$$

$$\phi(\text{RAG})(R_i) = \text{Max}(\text{RAG}(R_i), \text{Min}(\text{RAG}(R_j))) \quad (5)$$

where

$$R_j = \{V_A(R_j)\} - R_i \quad (6)$$

The geometrical action of the opening and closing operations  $\gamma(\text{RAG})(R_i)$  and  $\phi(\text{RAG})(R_i)$  respectively, consists of merging two candidate adjacent regions  $R_i$  and  $R_j$  on the RAG in only one area  $(R_i \cup R_j)$  if the aggregation criterion is verified. This criterion is determined using a clustering method.

### 3 CLUSTERING

To determine the aggregation criterion, we propose to perform a thresholding using a statistical method proposed in (El-Khoury et al., 2007). This approach splits the histogram of the grayscale images into several sections by computing automatically the thresholds that separate the different representative classes of pixels in the image without introducing any *a priori* knowledge.

This method supposes that the probability density function of each cluster is Gaussian and then finds the most probable spectral points of change that separated two consecutive clusters by using the generalized likelihood ratio (GLR) and the Bayesian information criterion (BIC). In our case, the hypothesis test is defined as:

$H_0$  :  $R_i$  and  $R_j$ , two adjacent regions belong to the same cluster.

$H_1$ :  $R_i$  and  $R_j$ , two adjacent regions belong to different clusters separated by a point of change  $C$ .

The GLR is computed as followed:

$$\text{GLR} = \frac{P(H_1)}{P(H_0)} \quad (7)$$

Once the points of change are detected a re-adjustment step takes place in which GLR is applied several times until stabilization. Finally the definitive change detection step is processed using BIC.

The clustering result is used to control the morphological operations on the RAG. Therefore two adjacent regions  $R_i$  and  $R_j$  that belong to the same cluster will be merged on the RAG using the opening and closing operations as described above.

The three segmented maps are fused together giving the final segmented RGB image. A final stage yields a segmentation refinement using the hypercube classification.

## 4 HYPERCUBE CLASSIFICATION

This method suppresses the isolated pixels and filters the classes that contain less than 3 pixels that are incorporated into the larger adjacent class with which the minimum difference in color is verified. It consists in detecting the valley on the gradient of the histogram 1D of R, G and B on which an interval  $[S_\lambda; S_\lambda + \Delta S]$  is defined for each of the three components. The intersection of all intervals defines classes. This process is shown in Figure 1.

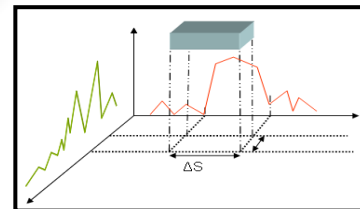


Figure 1: Hypercube classification.

The final segmented RGB image is shown in Figure 2.

## 5 EXPERIMENTAL RESULTS

In this section we present some results obtained by applying our segmentation scheme on microscopic medical test images. Here we present the process of the iterative RAG processing on a cytological image that decreases the number of regions by 80% without introducing clustering errors. Segmentation results are presented in Figure 3 by showing the



region borders showing the ability of the proposed segmentation scheme to simplify the original image. The use of morphological process as opening and closing operations applied on the RAG yields an interesting feature extraction as the photometric value, the area, the perimeter, the compactness factor, the number of neighbors of a region and their relationship for each region. This description represents the simplified information and contains potentially an elaborate knowledge. After the interpretation of the image, a list of retained objects and their associated features are stored in an XML (eXtensible Markup Language) file and ready to be integrated into a medical information system.

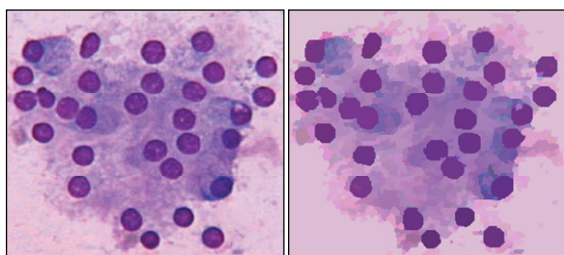


Figure 2: The original RGB image and its corresponding segmented image.

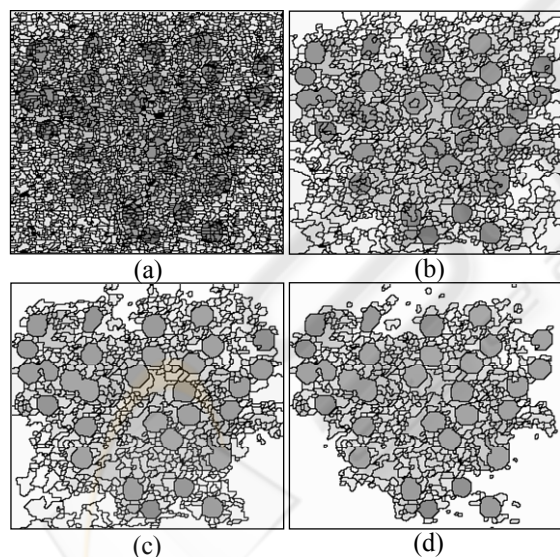


Figure 3: Hierarchical RAG levels shown the step of region merging.

## 6 CONCLUSIONS

We have proposed a new method of microscopic medical images segmentation using mathematical morphology applied on RAG and an automatic clustering method followed by a regularization step

using an automatic hypercube classification. Due to the unsupervised nature of the procedure especially the use of automatic thresholds detection, it can be reliable to the huge variability of intensities and shapes of the image regions and will be tested as a part of future work in other color space without introducing *a priori* knowledge and pre-processing stages.

Results show the effectiveness of our method for medical image applications as cytology images and the impact that it introduces on the semantic high level search for any disease or abnormal cells.

In this paper, the morphological operations consider only the extrema of region neighborhood. For future works, we will pursue the aggregation operations beyond the limits presented by the morphological processing avoiding the refinement segmentation step that uses the hypercube classification.

## REFERENCES

- Lucchese, L., Mitra, S.K., 2001. Color image segmentation: A state-of-the art survey. In *Image Process. Vis. Pattern Recog. Proc. Indian Nat. Sci. Acad. (INSA-A)*, Vol.67 i2, pp. 207-221.
- Lezoray, O., Lecluse, M., November 2007. Automatic segmentation and classification of cells from broncho alveolar lavage. In *Image Analysis and Stereology*, Vol.26, pp. 111-119.
- Lezoray, O., Juin 2003. Supervised automatic histogram clustering and watershed segmentation. Application to microscopic medical images. In *Image Analysis and Stereology*, Vol. 22, Numéro 2, pp.113-120.
- El-Khoury, E., Senac, C., André-Obrecht, R., 2007. Speaker Diarization: Towards a more Robust and Portable System. *Int. Conf. On Acoustics, Speech, and Signal Processing (ICASSP'2007)*, Honolulu, Hawaii, USA.
- Hirata Jr., R., César Flores, F., Barrera, J., Lotufo, R., Meyer, F., 2000. Color Image Gradients for Morphological Segmentation. In *XIII Brazilian Symposium on Computer Graphics and Image Processing (SIBGRAPI'00)*, pp.316-326.
- Beucher, S., Lantuéjoul, C., Sept. 1979. Use of watersheds in contour detection. In *Int.workshop on image processing, real-time edge and motion detection*.
- Mestar, A., Vannoorenberghe, P., Flouzat, G., April 2007. Mathematical Morphology applied to Very High Resolution Spatial images interpretation. In *Urban Remote Sensing Joint Event*, pp. 1-7.
- Pesaresi, M., Benediktsson, J.A., Feb. 2001. A new approach for the morphological segmentation of high-resolution satellite imagery. In *Geoscience and Remote Sensing, IEEE Transactions*, Vol 39, Issue 2.