



## ARAŞTIRMA/RESEARCH

# The methodology of wavelet analysis as a tool for cytology preparations image processing

Sitoloji preparatlarının görüntü işlenmesi için bir araç olarak dalgacık analizi metodolojisi

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### Abstract

**Purpose:** The aim of this study was to determine the possibility of using wavelet analysis for processing images of cytology preparations.

**Material and Methods:** A set of different images of cytology preparations were analyzed through changes in their contrast and application of the methodology of the wavelet analysis.

**Results:** Developed procedure of processing of cytology preparations images. Procedure of processing of cytology preparations images allows to qualitatively (in terms of their visualization) allocating: cells' edges, cell nuclei, revealing in more detail textural features of cells' images, which allows analyzing cell structure.

**Conclusion:** Consider the possibility and feasibility issues of applying wavelet analysis for processing cytology preparations images. This improves the quality of the analysis of cytology preparations images. This allows the to properly diagnose.

**Key words:** Wavelet analysis, image, contrast enhancement, cell, medicine, cytology preparation

### Öz

**Amaç:** Bu çalışmanın amacı sitoloji preparatlarının görüntülerinin işlenmesinde dalgacık analizlerinin kullanılma olasılığını belirlemektir.

**Gereç ve Yöntem:** Sitoloji preparatlarının farklı bir görüntü seti, kontrastlarındaki değişimler ve dalgacık analizlerinin metodolojik uygulamalarıyla analiz edildi.

**Bulgular:** Sitoloji preparat görüntülerinin işlenmesi prosedürü geliştirildi. Sitolojik preparat görüntü işlenmesi prosedürü kalitatif olarak (görüntüleme açısından) birçok yapının (hücre yapısının analizi, hücre görüntülerinin yapısal özellikleri, hücre sınırları, ve hücre nükleusları) tanımlanmasına izin verir

**Sonuç:** Sitolojik preparatların görüntüleme işlenmesi için dalgacık analizlerinin uygulanabilirliği ve yapılabiliğinin düşünülmesi. Bu işlem sitolojik preparatların görüntüleme analizi kalitesini iyileştirir dolayısıyla bu da daha uygun şekilde tanıya olanak tanıyacaktır.

**Anahtar kelimeler:** Dalgacık analizi, görüntü, kontrast geliştirme, hücre, tıp, sitoloji preparasyonu

## INTRODUCTION

Processing real objects' images, processes and phenomena is one of the ways of perception of the world around us. This processing allows studying not only the immediate changes that occur in the real world, but also discovering and studying possible pattern of such changes. At the same time, image processing allows studying the processes that cannot be seen or analyzed by means of human

vision. This allows talking about various directions in the use of image processing techniques and methods. One such directions of application of a common ideology of image processing is medicine. In this case we are talking about the possibility to apply common ideology of image processing when studying human health state, diagnosing and detecting symptoms of various diseases. In this case, images of real objects are those of different organs, tissues, parts of human skeleton, obtained with the

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help of special methods of their visualization<sup>1-5</sup>. Thus, medical image is a complex structural and functional image of different human organs, intended for the study of functional characteristics of human body and diagnostics of possible illnesses.

Among the many real objects that allow studying human body, one can underline the cytology preparations images. It is connected with the following facts: On the one hand, cytology preparations are objects of microcosm, which allow for a more in-depth studies of the human body, to study the dynamics of its operation and to diagnose possible diseases in the early stages of their development. On the other hand, these are special images that differ in their visualization of microcosm objects, which necessitates the use of a variety of image processing techniques to obtain information about objects, processes, and phenomenon under study. Primary, which is directly formed when rendering images of the real world, and that reflects the qualitative characteristics of objects represented in images: the ability to analyze the obtained images, the ability to identify individual objects in the image, etc. Additional information which is based on additional calculations of the original image: the number of objects represented in the image, the dimensions of the objects represented, the structural characteristics of the presented objects, etc.

Thus there are still quite a lot of different tasks for processing images and information represented on them/ In particular, such set of tasks may include the initial presentation of cytology preparations images, the selection of methods for post-processing of cytology preparations images. In the end, the necessity to solve such complex of tasks in their combination has determined the selection of research fields addressed in this article.

## MATERIALS AND METHODS

### The general ideology of post-processing of cytology preparations images

As a background for the selection of separate research tasks discussed in this article, the common ideology of processing of cytology preparations images is elaborated. This can be done by studying the works of various authors that deal with the solution of similar tasks. As an example of separate works that use the ideology of imaging processing

for studying of cytology preparations the following research work can be provided: Saha et. al, which deals with the cytological image segmentation to isolate the cell nucleus<sup>6</sup>.

The following image processing methods have been chosen as the main ones for this study: preliminary processing, which aims at improving the perceptual quality and subsequent processing of the original image; threshold segmentation, which allows isolating the cell nucleus; Mahendran et al. discussing the issues of segmentation and classification of cells cytology preparations images<sup>7</sup>. Particular attention is given to the pre-processing of source images in order to obtain more reliable results; Singh and Gupta, who examine the possibility of applying the texture analysis methods for cytology preparations<sup>8</sup>. At the same time, the authors point out that the isolation and analysis of the texture of the original image involves implementation of images pre-processing, where filtration and change of contrast can be allotted. Ensink et. al, who study the issues of the selection of threshold for image segmentation of cytology preparations<sup>9</sup>. However, as the authors point out, the selection of this threshold depends to a large extent on the baseline characteristics of the original images. Therefore, the authors talk about the necessity of pre-processing of the original image as of some tuning procedure for selecting the optimal threshold for further segmentation. At the same time, the authors offer their approach to the selection of such a threshold, while rejecting traditional methods of calculating the threshold for image segmentation - fixed threshold and Otsu's method<sup>9</sup>.

George et al. offered to conduct automated segmentation of cells in the images of the cytology preparation under study<sup>10</sup>. In addition, for the implementation of such process of segmentation authors talk about the necessity to change the histogram of the input image in order to enhance its contrast. Malviya et al. dealt with nucleus localization in the cytology preparations images under study<sup>11</sup>. To implement this image processing procedure, a special technique is used with staining of clinical specimens. This allows using a simple threshold processing of the input image without its pre-processing. Nevertheless, the authors point out that there may be some ambiguity while localizing nucleus. The reason for such ambiguity is the emerging difference in the relative staining intensity

of the clinical samples examined. Possible errors in segmentation of cells on cytology preparations images as a result of the arising differences in relative intensity of their staining is also studied by E. M. van Ingen, L. Leyte-Veldstra, I. Al, G. Wielenga and I. S. Ploem<sup>12</sup>. At the same time N. Dey, A. S. Ashour, A. S. Ashour and A. Singh talk not only about the possible influence of the relative staining intensity of the preparations under study on the quality of their image processing<sup>13</sup>. N. Dey, A. S. Ashour, A. S. Ashour and A. Singh determine the whole range of problems connected with the processing of microscopic images in medicine, where the primary goal is to obtain high quality image for its further thematic processing<sup>13</sup>.

Thus, the overall ideology of cytology preparations image processing pursues its goal as the selection of certain parts of the image (cells, nucleus) for further study of their changes (changes in cell shape, the change in the area of a cell) or for the calculation of certain quantitative characteristics (number of cells, the number of nuclei, cells' area). At the same time, particular attention is paid to the methods of cytology preparations source images (filtering, change of contrast, histogram equalization) in order to enhance the information they contain. However, it should be noted that by simply changing the brightness, contrast or by filtering it is impossible to solve arising issues with proper quality while processing cytology preparation images. Based on noted above, the objectives of this study are to explain the method of cytology preparation images processing, to review the ideology of preprocessing of cytology preparation images for their processing method under discussion and to conduct experiments based on the suggested method of cytology preparation images processing.

### Wavelet analysis as a tool for cytology preparations image processing

In order to solve the set of issues connected with cytology preparations image processing the methodology of wavelet analysis will be considered. The selection of wavelet analysis method for further cytology preparations images processing is based on the fact that wavelet processing allows taking into account the particular characteristics of the images under study by decomposing source data into a plurality of approximate and detail coefficients, in particular by image edge detection<sup>14</sup>. Moreover, wavelet analysis has become an integral part of

processing of complex stochastic processes of different nature, which include the visualization of cytology preparations objects, in terms of complexity of the representation of wild life clinical objects under study. In addition, image processing results obtained with the help of wavelet analysis, are often more informative<sup>15,16</sup>.

Wavelet analysis is based on wavelet transform. The wavelet transform is a signal decomposition (e.g. of some image) by the system of wavelets. Wavelets are obtained by shifting and scaling a single function – parent wavelet<sup>17</sup>.

Wavelet in this case is a function, rapidly decreasing at infinity, the average value of which equals to zero. Unlike Fourier analysis, each scale value of wavelet analysis corresponds with the infinite number of shifted in relation to each other spatially localized functions. If the signal is discontinuous, only those wavelets will have high amplitudes, which maxima will appear near the discontinuity point. This allows detecting image edge on the image under study. At the same time, discontinuity point is a sharp intermittent transition during some process. Quantitatively, it can be estimated by the value of the first derivative of such process. In the places of intermittent transitions the first derivative is very high. If the transition is in the form of discontinuity point, then the first derivative tends to infinity. However, the real processes, measured by real devices, cannot have perfect discontinuity points. In fact, the measured fractal transitions are characterized by the finite value of the derivative. The sharper the transition, the higher the derivative value is. Smooth transitions will have small derivative values. This allows you to determine the presence of special characteristics of the image analyzed, as well as the point where these characteristics may arise.

Behind the formalization of the continuous wavelet transform (CWT) there's the use of two continuous and integrable along the whole axis functions<sup>17,18</sup>.

- wavelet – function  $\phi(t)$  with zero integral value

$$\int_{-\infty}^{\infty} \phi(t) dt = 0, \quad (1)$$

determining the details of the signal and generating extended fractions;

- scaling function  $\varphi(t)$  with a unit value of integral

$$\int_{-\infty}^{\infty} \varphi(t) dt = 1, \tag{2}$$

determining a rough approximation of signal and generating approximation coefficients.

However, CWT function can be applied only for one-dimensional signals, and image is a two-dimensional signal. Therefore, in order to be able to apply CWT to detect image edges it is suggested to consider the following analysis and edge detection procedure: 14

- let's perform calculation for horizontal discontinuities of the original image  $F$ , represented by matrix defined by its readings  $f_{ij} \in \{0, 1, \dots, P\}$ ,  $i = 1, 2, \dots, N$ ,  $j = 1, 2, \dots, M$  on a square lattice  $N \times M$ . To do this, we use the following formula to get the so-called matrix of wavelet spectrogram  $W$  (based on the sequential processing of each line of the original image  $F$ ):

$$W[f_{ij}] = \frac{1}{\sqrt{a}} \int_{-\infty}^{+\infty} f_{ij} \phi\left(\frac{t-b}{a}\right) dt, \tag{3}$$

where  $\phi\left(\frac{t-b}{a}\right)$  is a mother wavelet that meets the condition (1),

$a, b$  – scale and center of temporary localization which determine the scale and bias function  $\phi(t)$  in accordance with the terms of scaling (2);

$[f_{ij}]$  indicates the number of the processed string of the original image  $F$  to get a plurality of values of its wavelet spectrogram.

Parameters  $a, b$  are chosen so that the corresponding linear dimensions of the matrix of wavelet spectrogram  $W$  correlate with linear dimensions of the original image  $F$ , and at the same time possible parameter of wavelet transform are taken into account. Then, based on the analysis of the obtained spectrogram ( $W$  for each row of the original image  $F$ ) we select its certain line  $NN$  based on the condition:

$$NN = \max_m \left( \frac{1}{M} \sum_{d=1}^M w_{md} \right) \tag{4}$$

where  $w_{md}$  is the element of wavelet spectrogram of the analyzed row (line) of the original image  $F$  ( $m = \overline{1, a}, d = \overline{1, b}, b = M$ ).

This selection is determined by the fact that we select that part of spectrum of the original image row (line), which corresponds to the largest discontinuity area of the original signal between its readings (see comments above).

The selected in such a way line (row), will correspond to the line (row) in matrix  $F_g$  which characterized the matrix of horizontal discontinuities of the original image  $F$ .

Processing of all lines of the original image  $F$  allows obtaining the matrix of horizontal discontinuities  $F_g$  through the following sequence of transformations:

$$\xrightarrow{\text{F CWT lines}} \xrightarrow{\text{W selection}} \text{line } F_g$$

- in a similar way we calculate the vertical discontinuities of the original image  $F$  for each column. For this purpose, use formula (3) and the formula similar to formula (4) to select certain line from the obtained wavelet spectrograms of each column of the original image  $F$ :

$$MM = \max_m \left( \frac{1}{N} \sum_{d=1}^N w_{md} \right), \tag{5}$$

where  $w_{md}$  is the element of wavelet spectrogram of the analyzed column of the original image  $F$  ( $m = \overline{1, a}, d = \overline{1, b}, b = N$ ).

Processing of all columns of the original image  $F$  allows as a result obtaining the matrix of vertical discontinuities  $F_v$ , due to the following sequence of transformations:

$$\xrightarrow{\text{F CWT column}} \xrightarrow{\text{W selection}} \text{column } F_v$$

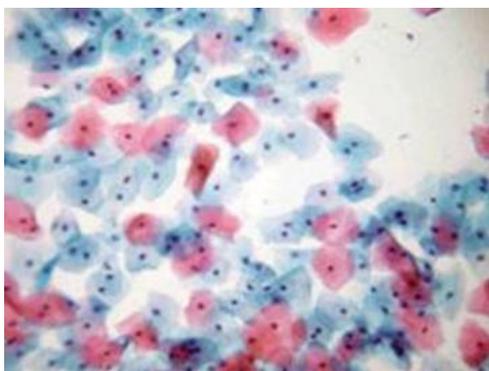
- add matrixes of vertical and horizontal discontinuities into one matrix that displays the edge of the original image based on CWT methods. For visual clarity, matrixes are horizontal, vertical discontinuities, as well as generalized matrix showing the edge of the original image can be inverted.

It should be noted that the construction wavelet spectral pattern  $W$  is largely determines by the size of the original image and the used scale parameter  $a$  when conducting wavelet transform of the image under study. In this work, to consider the possibility of using wavelet analysis as a tool for processing cytology preparations images, parameter  $a=20$ , and parameter  $b$  correlates with the linear dimensions of the original image in accordance with the procedure of constructing the matrix of wavelet spectral pattern for rows and columns of the image respectively.

However, before we proceed to the consideration of the obtained results that relate to conducting wavelet transform for the cytology preparations images under study, let's focus on the preliminary processing of the original images as on one of the main elements of processing of microscopic images in medicine.

### Data for analysis

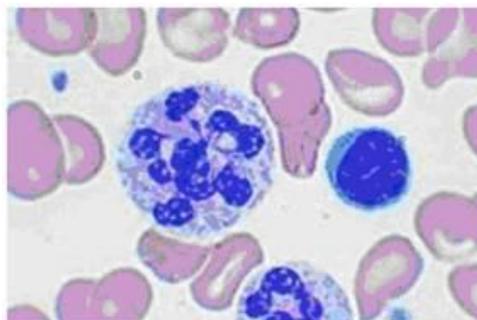
In order to identify the possibility of using wavelet analysis as a processing tool for cytology preparations images, some images have been selected. Test images - a image of different cellular structures have been presented in Figure 1 to Figure 4 and numbered accordingly



**Figure 1. Cellular composition of cervical smear**

The presented images of cytology preparations are different in their structure and complexity of perception, which allows evaluating the possibility of using wavelet analysis methodology as a tool for their processing. Moreover, all images are presented in color. However, the implementation of certain functions of the general methodology of wavelet analysis involves the work with gray-level images. Therefore, the color does not affect the selection contour of objects with the help of wavelet analysis.

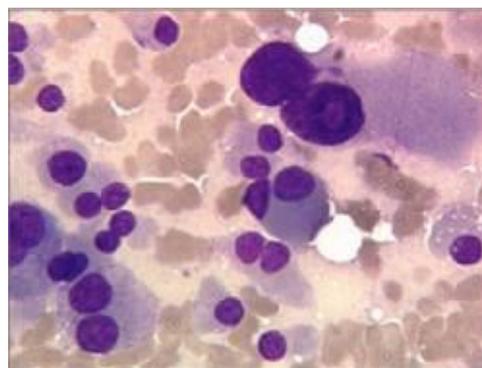
We are working with gray-level images. This feature the use of wavelet analysis for cytology preparations image processing.



**Figure 2. Image cells in pernicious anemia**



**Figure 3. Transparent structure of cells**



**Figure 4. Medullary (C-cell) cancer**

## RESULTS

### Pre-processing of images

Therefore, all the original images are preliminary presented in the form of corresponding gray-level images, which can be considered as the first stage of the original images pre-processing (Figure 5, 6, 7, 8).

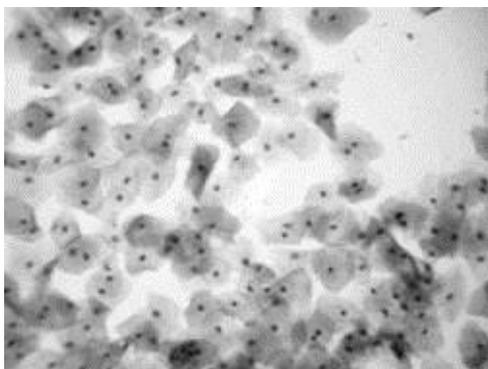


Figure 5. Gray-level (halftone) image No.1

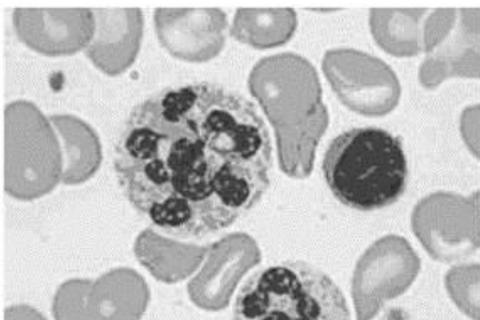


Figure 6. Gray-level (halftone) image No.2

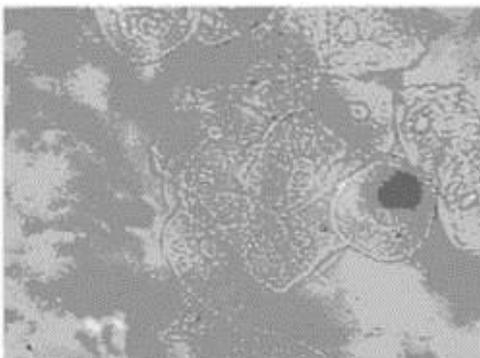


Figure 7. Gray-level (halftone) image No.3

A visual comparison of the original color images and their representations in the form of gray-level images shows that the initial information about cytology preparations is not lost. However, as noted above, one of the necessary stages of preprocessing of microscopic images in medicine is their contrasting. Contrast is one of the main characteristics of the image; it is directly related to the brightness of pixels that are the sources of information about the objects in the image. Therefore, changing the contrast of the image allows

improving both image perception accuracy, as well as the accuracy (efficiency) of its further processing. By increasing the contrast of the image (pixels - individual image points) highlights become lighter and dark image regions become darker.

As a result, there occurs a redistribution of pixels as a result of the average gray-level range. When reducing image contrast, on the contrary, there is an expansion of the average gray-level range. Dark pixels become lighter, and light pixels become darker and partially transform into the midtones. Thus, changing the contrast of the image, and above all increasing the contrast allows making some image details more distinct. It is very important for microscopic images in medicine, an example of which are images of cytology preparations. Therefore, to further analyze the halftone images, they all were contrasted.

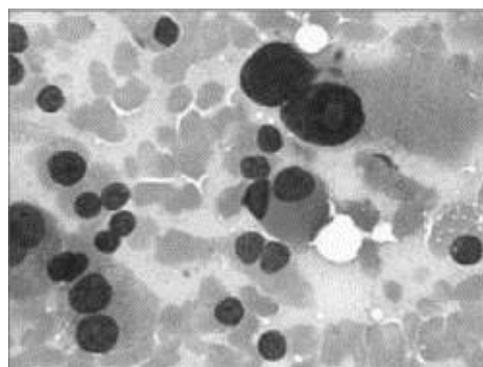


Figure 8. Gray-level (halftone) image No.4

The selection of different levels of contrast enhancement for the images under study is first of all determined by the necessity to test the possibility of using wavelet analysis for cytology preparation image processing.

### Results of wavelet transform of cytology preparations images

Thus, wavelet transform of cytology preparations images will be held on halftone images, one of which is the source (primary) image obtained from the corresponding color image, and the second one is a contrasted image of the original grayscale (halftone) image. As a wavelet transform of cytology preparations images the method of selecting special features of the images was used, described in the part "Wavelet analysis as a tool for processing of cytology preparations images".

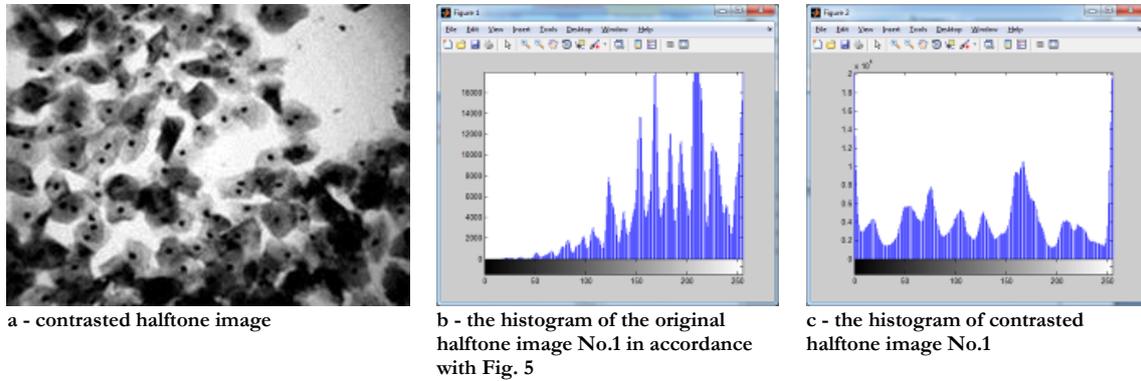


Figure 9. Result of contrasting of half-tone image No.1 by 35%

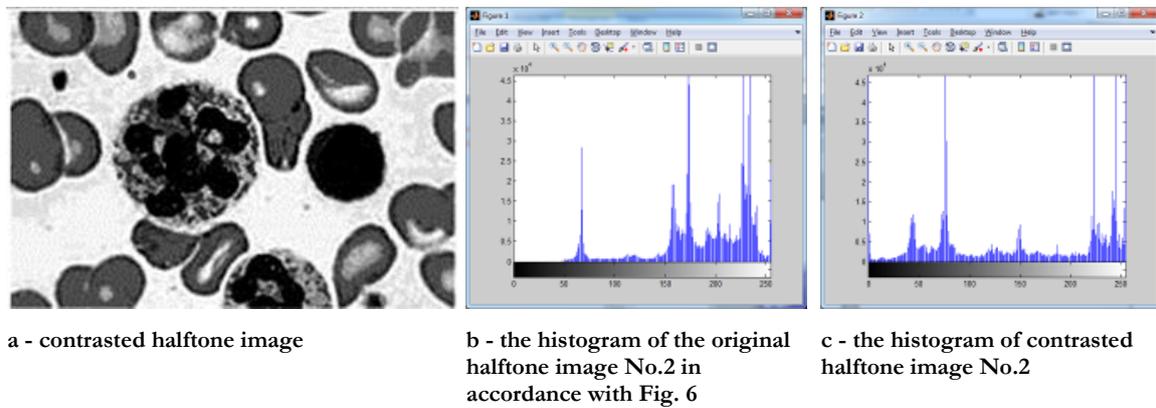


Figure 10. Result of contrasting of half-tone image No. 2 by 40%

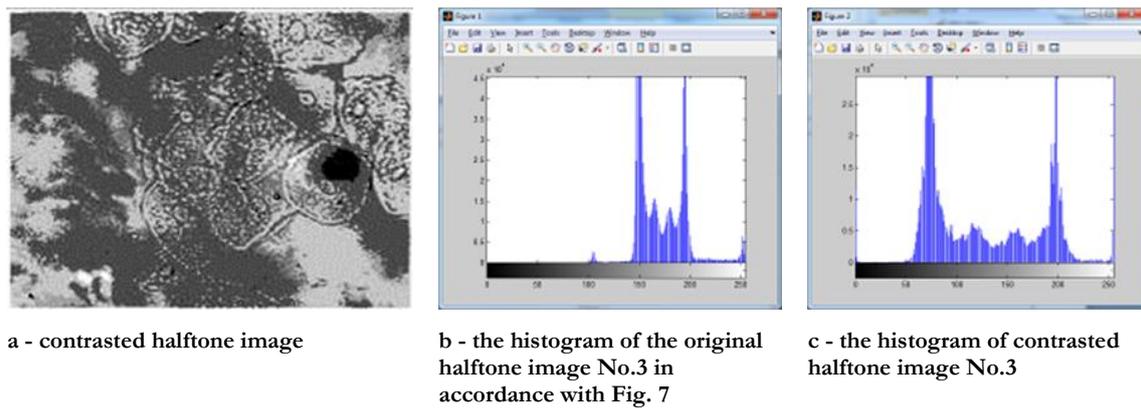
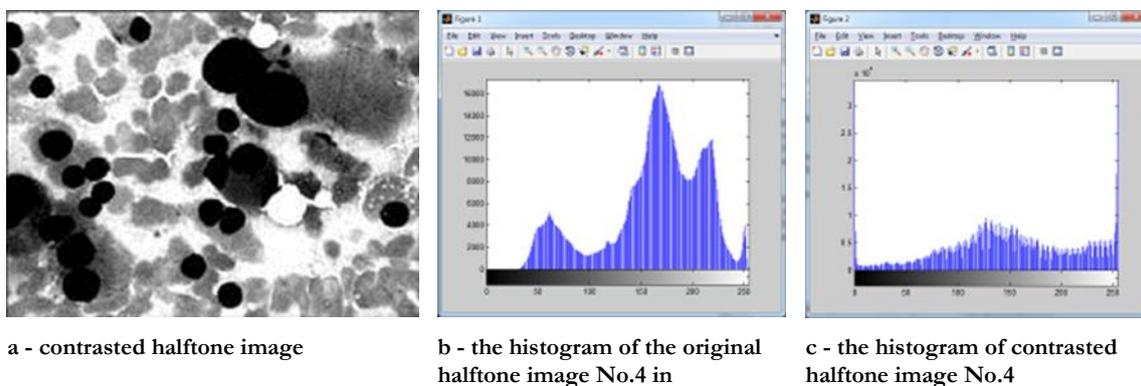


Figure 11. Result of contrasting of half-tone image No. 3 by 40%



a - contrasted halftone image

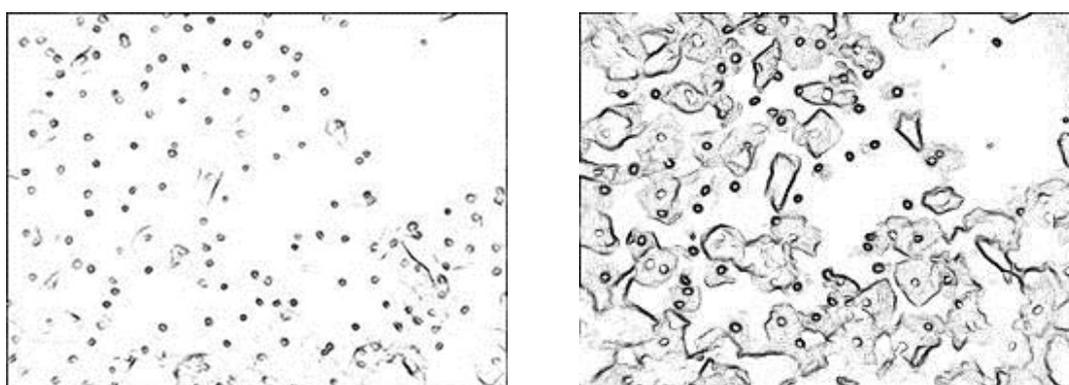
b - the histogram of the original halftone image No.4 in accordance with Fig. 8

c - the histogram of contrasted halftone image No.4

Figure 12. Result of contrasting of halftone image No. 4 by 35%

As it can be seen from data on Figure13 to Figure 16 the described method of image wavelet transform allows detecting first of all edges of separate objects represented on the corresponding images. The used wavelet transform also allows highlighting the specific features of cytology preparations of separate objects (cells) in the images. At the same time, on basis of shown in Figure 13 to Figure 16, it can be stated that the use of the studied wavelet transform provides more information for images that have been contrasted. In the case where wavelet processing was applied to a more contrasted image, the result is not only more accurate cell edge detection, but the allocation of the internal structure of these cells (Figure 13b, Figure 14 b, Figure 15b). This allows for a more detailed qualitative and quantitative analysis of the internal structure of the

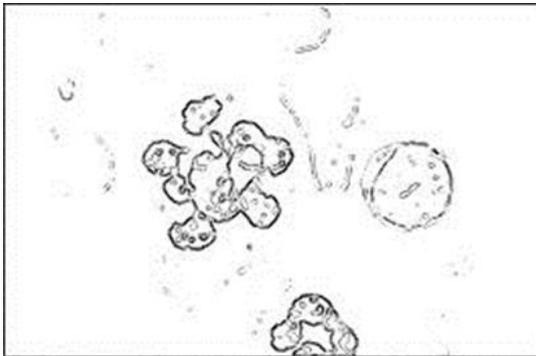
cells represented in the images of cytology preparations. In particular, it is possible to analyze the textural changes that occur within the cell, to analyze in more details the individual elements of cells' structure, to calculate the dynamics of change in the cell nucleus, the nucleolus, intracellular filaments, etc. (see Figure 14b). Nevertheless, it is possible to combine the results of wavelet processing of images with different contrast. This will help solving different problems: from localizing only cell nuclei to the study of the internal structure of cells. In any case, the discussed above one the procedures of wavelet analysis shows that it is possible and feasible to use wavelet analysis as a tool for processing cytology preparations images in order to obtain additional information to conduct diagnostics and assess the state of human health.



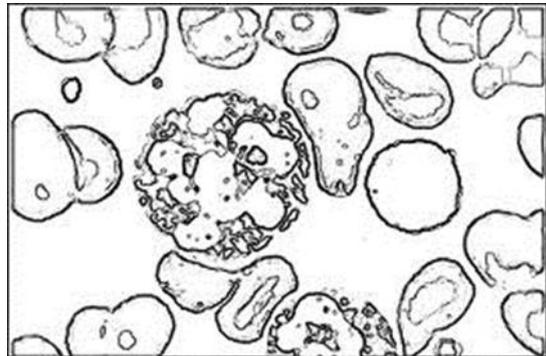
a. processing of the original halftone image

b. processing of contrasted halftone image

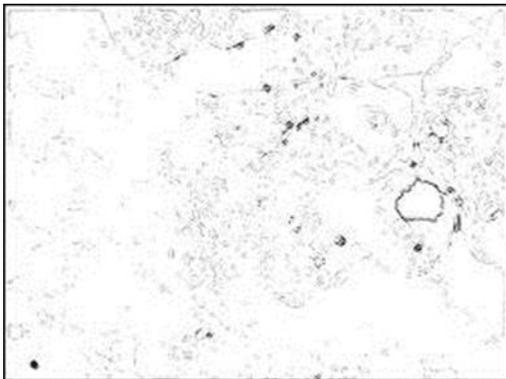
Figure 13. Results of wavelet transform for image No.1



a. processing of the original halftone image  
Figure 14. Results of wavelet transform for image No.2



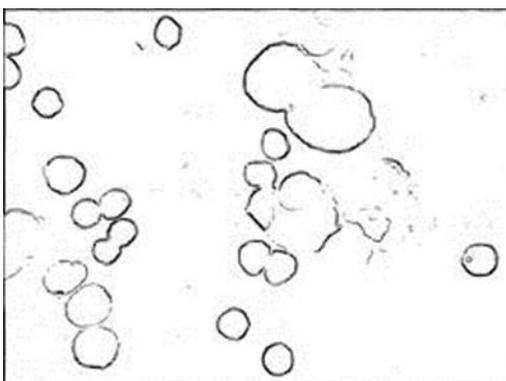
b. processing of contrasted halftone image



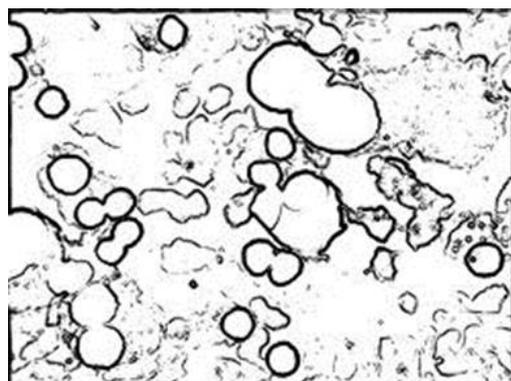
a. processing of the original halftone image  
Figure 15. Results of wavelet transform for image No.3



b. processing of contrasted halftone image



a. processing of the original halftone image  
Figure 16. Results of wavelet transform for image No.4



b. processing of contrasted halftone image

## DISCUSSION

Edge detection is a critical step, which allows the analysis of images. But edge detection of objects on the image of cytology preparations is a challenge. For edge detection can use different algorithms. It is important have a quality (accuracy) for edge detection here. To select a contour, usually used classical approaches which include image edge detection using the Roberts, Sobel, Prewitt, and Canny operators. These methods are well described and reviewed in the book "Histochemical and Cytochemical Methods of Visualization"<sup>19</sup>.

But as pointed out by Dey et al., among disadvantages of edge detection techniques listed above are the following: dependence of edge detection quality on the changes of image brightness value and on differences between the potential zones of edge detection, which, in general, is associated with the lack of the ability to automatically select a threshold for conducting the appropriate convolutions<sup>13</sup>. CWT technique does not have this drawback. We are changing the contrast of the image. This improves the accuracy of edge detection.

Classical image edge detection techniques provide computational simplicity in their implementation. The key point in the time for their implementation is the number of possible enumerations for calculating convolutions between the masks of specific edge detection operators and various image areas. CWT technique for edge detection doesn't require a large number of enumerations in order to generate image contour, as it allows working as a whole, both with columns and with rows of the original image matrix. However, the real computational complexity of the CWT technique that influences the time required for image edge detection is the wavelet transform technique itself – for each column and row of the original image.

At the same time, it should be noted that as compared to the classical methods, CWT technique has the following advantages. First the processing is conducted not on the image display area, but consequentially by columns (rows) of the image matrix, which allows to accurately identify gradients on each column (row), and therefore, increase the quality of edge detection. Second for processing columns (rows) of the image matrix to identify boundaries of object on the image high-sensitivity wavelet spectral analysis has been used. This can be

considered as the main advantage of CWT technique, since only wavelets can allocate even small discontinuities on one-dimensional signals in the most accurate way, which are interpreted as object boundaries in this case.

There is an interesting study of Pise et al.<sup>20</sup>. The study covers the different pre-segmentation processes, like circular Hough transform. But this method allows to select you only the cell nuclei. CWT technique allows to select: the circuit cells and emphasize the internal structure of cells.

Many methods for edge detection object in the image cytology preparations based on the choice of color image points. This idea was considered by Al-Kofahi et al.<sup>21-23</sup> This method improves the accuracy of edge detection. But to implement such a method it is necessary to set point, which can be segmented. This limits the possibility of automatic selection contour to know threshold segmentation. This is difficult to do in an online of image processing. As the limit is necessary, one should specify the application of wavelet analysis methodology that would be used for halftone images cytology preparations. The format should be .BMP or .JPG at 100 dpi or greater resolution.

Analyzing images cytology preparations allow for a more in-depth studies of the human body, to study the dynamics of its operation and to diagnose possible diseases in the early stages of their development. Complexity of visualization process of cytology preparations and their subsequent processing with the use of automated processing determines the necessity to study new possibilities to use new approaches to image processing.

In summary, the paper deals with the possibility and feasibility issues of applying wavelet analysis for processing cytology preparations images. As a separate wavelet analysis procedure, which is proposed to be applied to processing of cytology preparations images, the procedure of allocating specific features on the presented images is discussed. Then the general ideology of the procedure for the use of wavelet analysis as a tool for cytology preparations images processing can be presented as follows. Firstly the necessity to transform the original image is determined. Then color image is converted to halftone (gray-level) image. The necessity to change contrast of the original halftone image is determined. Later wavelet transform of the original halftone image and of

contrasted halftone image is conducted. Conclusions are made on basis of wavelet transform results (additional processing procedures are applied to the obtained images in this case: calculating cell nuclei, cells, cells' area, etc.). The proposed procedure of processing of cytology preparations images allows to qualitatively (in terms of their visualization) allocating cells' edges, cell nuclei, revealing in more detail textural features of cells' images, which allows analyzing cell structure.

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