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To cite this article: V G Nikitaev *et al* 2019 *J. Phys.: Conf. Ser.* **1205** 012041

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Model of image sensor signal correction in the computer microscopy system

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Abstract. The article is devoted to the analysis of the possibilities of improving the accuracy of the analysis of low-contrast images in the computer microscopy systems. The image distortion factors are considered in the system of computer microscopy with a camera based on a light-sensitive electronic matrix. The level of distortion of the image sensor signal in the computer microscopy system was estimated on the results of the experiment. The model of correction of distortions of the image sensor signal is offered. The application of the proposed model will provide an increase in the accuracy of automated analysis of microscopic images of low-contrast objects in the computer microscopy systems

1. Introduction

Light microscopy is widely used in various fields of science, industry and medicine [1, 2]. The basis of the analysis of microscopic images is the identification of the structural elements of the objects of study on the basis of their color or brightness, contrasting with the surrounding background. In a number of tasks this contrast is visually unobtrusive [3, 4, 5]. Electronic means of image recording based on optical radiation sensors can be used to increase the contrast of objects in the image and their subsequent analysis in computer microscopy systems. The formation of a digital image allows to automate image analysis, highlight objects of interest and measure their characteristics. Automatic detection of low-contrast objects is limited by the capabilities of image sensors.

2. Materials and methods

The formation of a digital image in the system of computer microscopy is as follows (in the example, the mode of transmitted light is considered): the light flux from the light source is directed by the microscope condenser to the sample under study, part of the light is absorbed and scattered by the structural elements of the sample, the transmitted light is projected onto the photosensitive matrix of the electronic camera using a microscope lens. The light energy absorbed in the cell of the photosensitive matrix is converted into an electric charge, which is converted into voltage at the input of the analog-to-digital converter, the output of which is formed by a digital code recorded in the computer memory.

The formation of the image in the computer microscopy system is influenced by a number of factors along with the light flux on the light-sensitive matrix. A set of factors determining the distortion of the signal generated by the optical radiation sensor for each cell can be represented by the model $\langle Er, Kr, T, Tr, P, Pr, Ur \rangle$.



Here E_r is a random component of the light energy captured in the cell (due to the fluctuation of the light flux density at a constant brightness of the light source). K_r is a characteristic of the deviation of the quantum yield of one light-sensitive cell from the average value of the quantum yield for all cells of the matrix (different cells have different quantum yield). T , T_r - constant and random components of the dark current (dark current fluctuation) due to the generation of a parasitic charge in each cell, even in the absence of lighting. P , P_r - constant and random components of other factors contributing to the formation of the charge package, regardless of the light flux. For example, in the case of a camera in which the image sensor is a CCD, such a factor is a constant and random component of the charge transfer efficiency in the CCD (part of the charge packet is lost during the transfer of charge packets on the CCD). U_r - noise caused by the process of converting the charge package into voltage and its amplification to the level required for the analog-to-digital converter, which provides the formation of a digital signal.

If the useful recorded signal is separated from the side factors, it is possible to present the image signal $I(x,y,t)$ by the following generalized model:

$$I(x, y, t) = K * G(x,y) + S(x,y,t) \quad (1)$$

Here $G(x, y)$ is a function that determines the distribution of the light flux passing through the sample through the site, the projection of which on the photosensitive matrix corresponds to the area of the photosensitive cell of the image sensor with coordinates (x, y) ; (x,y) – integer spatial coordinates of the photosensitive cell of the matrix reduced to the plane of the sample (the numbers x and y correspond to the row and column of the cell of the photosensitive matrix); K - light-signal conversion factor (in the ideal system K does not depend on the coordinates (x,y) , but may depend on the value of the light flux), t – the time at which the image was recorded. $S(x,y,t)$ is a function of distortions associated with the imperfection of the image recording system. Under constant lighting conditions and a fixed sample, the function $G(x,y)$ is time-independent. The dependence of the function $S(x,y,t)$ on time reflects the random nature of the distortions on time.

Deterministic and random components can be identified in these side factors affecting the formation of the image. Generalizing influence of the specified factors, it is possible to present function of distortions in the form of:

$$S(x,y,t) = C(x,y, G(x,y)) + V(x,y, t, G(x,y)) \quad (2)$$

The first term in formula (2) defines a time-independent constant distortion component for each cell with coordinates (x,y) . The second term corresponds to the time-random changes in the signal under constant lighting conditions of a stationary sample.

Since the mathematical expectation of the second term in formula (2) is equal to zero, it is possible to estimate the first summand in the formula (2) with the required accuracy by experiment with statistical averaging of measurement results. The resulting estimate can be used to reduce the first term in formula (2) by subtracting the resulting estimate from the image signal. The influence of the second term on the image analysis results of low-contrast objects can be reduced by forming a calculated image, which is obtained as a result of statistical averaging of a series of images, shooting of which is performed under constant conditions.

An experiment was conducted to evaluate the components of the proposed model in a computer microscopy system using the Imperx IPX-4M15T-GCFB digital camera. The camera mode was used with a signal encoding of 24bit per pixel (8 bits per each RGB component) in the non-cooling mode. The frame size of the image is 2048x2048 pixels. The image was recorded in a file in the BMP24 format. A series of shots was taken under constant conditions to estimate the second term in formula (2). A uniformly illuminated background at different lighting levels was chosen as the subject to estimate the first term in formula (2).

3. Discussion

The results of the experiment (for example, G component with 256 brightness gradations) showed that the standard deviation of the values of the second term in the formula (2) is approximately in the range from 1.5 to 3 brightness gradations, while the values of the first term reach 5 brightness gradations. Thus, it is enough to shoot a series of 81 frames and compute the component average in each pixel for a series of frames to reduce the influence of the second term in the formula (3) to the level of less than one gradation (taking into account the spread of random values in the interval $\pm 3\sigma$). The random component of the distortion of the image sensor signal in the averaged image will not exceed two-thirds of the brightness gradation if you shoot an image in a series of 183 frames, followed by averaging the frames. The image signal can be corrected by subtracting the values of the obtained estimates from the image sensor signal based on the experimentally obtained estimates of the first term in the formula (2). The accuracy of the correction is determined by the number of frames in a series of images taken to estimate the systematic component of the signal distortion. So the error of correction of a systematic signal distortions of the image sensor did not exceed one third of component brightness gradation in shooting of images by a series from 729 frames (for a confidence interval of an assessment in ± 3 standard deviation). This operation is performed once during system calibration. Thus, it is possible to reduce the influence of signal distortion of the image sensor in the computer microscopy system, using a camera Imperx IPX-4M15T-GCFB, to one brightness gradation if having the correction of the systematic signal distortion of the image sensor as it is showed above(it reduces the influence of the systematic signal distortion to a third of a brightness gradation), and then averaging the images in a series of 183 frames(it reduces the influence of the random signal distortion to two thirds of brightness gradation).

4. Conclusion

A model of image signal distortion in a computer microscopy system is proposed for subsequent experimental evaluation of systematic and random components of signal distortion. Experimental evaluation of the components of the image signal distortion is performed. It is revealed that the values of the random and systematic component of the signal distortion in the computer microscopy system using the Imperx IPX-4M15T-GCFB digital camera are several units of the brightness gradations in 256 gradations signal range (the random component is up to three gradations, the systematic component is up to five gradations). The distortion correction model was proposed, it ensures the correction accuracy to one brightness gradation. The proposed method of distortions correction in microscopic images automated analysis systems will improve the accuracy of measurements of the characteristics of low-contrast objects.

Acknowledgement

The reported study was funded by RFBR according to the research project № 18-07-01456

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