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## **Experimental studies of phytoplankton concentrations in water bodies by using of multispectral images**

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### ***Abstract***

The aim is to improve performance and reliability of monitoring of nutrient pollution from bioindication by phytoplankton and use of multispectral images. Actuality of the theme caused by the necessity development of new methods of monitoring the state of water bodies based on bioindication by phytoplankton. We improved method of environmental monitoring groundwater multispectral methods and conducted experimental studies of phytoplankton concentrations in water bodies. The paper analyzed the evaluation methods of ecological state of water bodies based on bioindication by phytoplankton and features experimental techniques. Comparison bioassay results wastewater using phytoplankton culture *Scenedesmus subspicatus* and experimental studies of pollutants specialized laboratory environmental inspection. For identification phytoplankton particles carried compare arrays of multispectral images obtained at characteristic wavelengths of phytoplankton pigments exemplary multispectral imaging using Bayesian classifier on discriminant function based on Mahalanobis distance. Through this increased accuracy of identifying particles of phytoplankton in comparison with classical algologically methods, based on visual comparison of images of phytoplankton particles obtained by the microscope, with exemplary images taken from inventories and determinants, and performance monitoring of the ecological state of water bodies increases by 10 ..20 times.

### ***Keywords***

multispectral image, environmental monitoring, phytoplankton, water

## **1 Introduction**

Water resources are a national wealth of each state. Currently, there is a tendency to water pollution by industrial effluents. This disturbs the equilibrium of ecological systems and leads to the loss of their ability to recover. One of the most significant factors affecting the quality of surface waters is their anthropogenic eutrophication. It is the result of human activity and is fast deteriorating water quality due to the receipt of these nutrients and organic substances in quantities far exceeding the usual natural level. At present solve water management problems can not be performed without ensuring the stable functioning of ecosystems of water bodies, without preserving their integrity and stability (*Romanenko et al., 1998; Wright et al., 1993*).

The study aims to improve performance and reliability of control nutrient pollution from bioindication by phytoplankton. Actuality of the theme caused by the necessity development of new methods and means of control of water facilities based on biological indication of phytoplankton, as typical for traditional low value performance and reliability of monitoring.

## **2 Multispectral monitoring of water pollution based biological indication by phytoplankton**

Monitoring of natural aquatic environments may be based by bioindication indexes of phytoplankton. The functional role in ecosystems of phytoplankton - the primary element converting solar energy stream, producer of autochthonous organic matter, an important self-cleaning agent and photosynthetic aeration of water. Phytoplankton is one of the elements of biological classification of ecological status of water bodies according the Water Framework Directive EU 2000 (*2000/60/EC*). The object of control in this study are plankton with particle size to 50 microns. Phytoplankton algae are mostly unicellular, although there are many colonial and filamentous forms, especially in freshwater. In each field reservoir volume concentration of phytoplankton determined by physical, chemical and biological-coenotic conditions (*Dell'Uomo, 1997*). Information on the phytoplankton concentration in water bodies with his great spatial and temporal variation. Volume concentration and ratio of different kinds of phytoplankton depends on the time scale of annual seasonal changes and the depth of the reservoir, its cross section and along the length of the water body.

For identification phytoplankton particles carried compare arrays of multispectral images obtained at characteristic wavelengths of phytoplankton pigments exemplary multispectral imaging using Bayesian classifier on discriminant function based on Mahalanobis distance. Through this increased

accuracy of identifying particles of phytoplankton in comparison with classical algologically methods, based on visual comparison of images of phytoplankton particles obtained by the microscope, with exemplary images taken from inventories and determinants, and performance monitoring of the ecological state of water bodies increases by 10..20 times. The main pigment particles present in phytoplankton is chlorophyll a (characteristic wavelengths of 430 nm and 663 nm). Green algae contain chlorophyll b (435 nm, 645 nm). Diatoms and dynofitovyh algae contain chlorophyll c (440 nm, 583 nm, 634 nm). In red algae contain chlorophyll d. In addition to chlorophyll in chloroplasts always available carotenoids content equivalent to estimated beta-carotene (480 nm). Blue-green and red algae contain two types phycobilins (fikotsianyn and fikoeretryn) in different ratios. The proposed method is technically more complex than existing indirect methods of integrated assessment of phytoplankton pigment characteristics for the groups, as can more accurately determine the relationship between certain truck phytoplankton (Petruk *et al.*, 2012; Petruk *et al.*, 2015a,b).

The authors developed a structural diagram of the device measuring multispectral television monitoring of the ecological state of water bodies in the parameters of phytoplankton (Fig.1). The device contains a water sample with particles of phytoplankton 1, pump 2, CCD-camera 3, microscope 4, measuring the flow cell 5, capacity 6, database particles of phytoplankton 7, specialized processor 8, clarifier 9, index calculation unit 10, indicator 11.

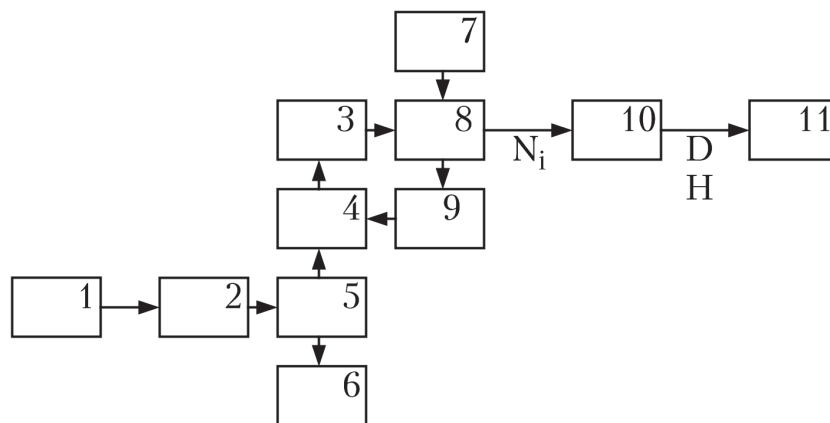


Fig. 1. Block diagram of the device measuring multispectral television monitoring of the ecological state of water bodies in the parameters of phytoplankton

When changing environmental ecosystem water bodies in eutrophication begins with the rapid growth of the number of certain species of phytoplankton, these species begin to dominate in the ecosystem gradually replacing other species of ecosystems. Therefore the relative number of dominant species  $p_i$  will grow,

which will increase the index of Simpson. In contrast to this in the ecosystem of water body that has good ecological status any species of phytoplankton are not dominant ecosystem is balanced and the relative numbers of different species are small, which reduces the Simpson index. With the deterioration of the ecological state of water body ecosystems, such as anthropogenic pollution because of its most sensitive species of phytoplankton reduce their number and subsequently disappear and are replaced by more resistant to contamination species of phytoplankton, which reduces the index Shannon.

### 3 *Image processing and identification of phytoplankton particles*

Is possible identify a certain type of phytoplankton particles based on statistical data on average particle size  $\langle \rho \rangle$  or their refractive index  $\langle m \rangle$ . However, a significant drawback of this approach is the necessity of construction calibration curve to calculate the empirical coefficients also does not take into account the structure and shape of particles because the particles to form complex structures and can not pick up a suspension of artificially created particles whose parameters are known. It is therefore advisable to carry out a direct comparison of multispectral images with images of particles to particles known type, structure, shape and size determined by means of exemplary means of measurements, for example, using an electron microscope. The simplest method of comparison is to compute the difference image ( $\Delta N_{ij}$ ) between elements measured ( $N_{ij}$ ) and model ( $N_{et.ij}$ ) images

$$\Delta N_{ij} = N_{ij} - N_{et.ij} \quad (1)$$

In terms of the theory of decision a comparison of two dimensional vectors  $x$  and  $y$  comes to finding the Euclidean distance between them (*Gonzalez et al., 2009*)

$$d(x, y) = \|x - y\| = \|y - x\| = \left[ (x_1 - y_1)^2 + \dots + (x_n - y_n)^2 \right]^{1/2} \quad (2)$$

If the Euclidean distance does not exceed the allowable threshold, the particle corresponds to the same type as the reference image. It should be noted that the size and shape of the particles of a certain type are random variables distributed by the law close to Gaussian, and therefore the threshold should be chosen so as to cover 99.9% of the particles of a certain type, as well as to secure the separation of particles of different types.

The choice of the shortest Euclidean distance is equivalent to calculation of the function

$$d_j(x) = x^T m_j - \frac{1}{2} m_j^T m_j \quad (3)$$

where  $m_j$  – the average vectors for  $j$  class.

Identification of particles to a certain type carried out at the highest value  $d_j(x)$ . Hypersurface distinguishing between classes  $\omega_i$  and  $\omega_j$  in the case of the classifier on a minimum Euclidean distance is determined by the equation (Gonzalez et al., 2009)

$$d_{ij}(x) = d_i(x) - d_j(x) = x^T (m_i - m_j) - \frac{1}{2} (m_i - m_j)^T (m_i + m_j) = 0 \quad (4)$$

It is advisable to use this method of detection of particles, allowing the particles to compare images of different sizes and provide the least value recognition errors.

One such approach is to search for spatial correlation images. In the analysis of the original image  $f_1(x, y)$  correlation task is to find the position of the image that best match the reference image  $f_2(x, y)$ . Because the spatial correlation of images, according to the theorem of correlation reduces to convolution in the frequency domain and search spatial correlation is reduced to multiplication of the transformed images

$$f_1(x, y) \circ f_2(x, y) \Leftrightarrow F_1(u, v) \cdot F_2^*(u, v) \quad (5)$$

where  $f_1(x, y) \circ f_2(x, y)$  – spatial correlation,  $F_1(u, v) \cdot F_2^*(u, v)$  – a product of transformations images.

To compare images of different dimensions using their processing in the frequency domain using the discrete Fourier transform (DFT), direct DFT  $F(u, v) = \text{fft2}(f(x, y))$  is as follows

$$F(u, v) = \sum_{x=0}^{M-1} \sum_{y=0}^{N-1} f(x, y) e^{-j2\pi(ux/M+vy/N)} \quad (6)$$

where  $f(x, y)$  – investigated image  $M \times N$  ;

$x = 0, 1, 2, \dots, M - 1$  ;  $y = 0, 1, 2, \dots, N - 1$  – spatial coordinates;

$u = 0, 1, 2, \dots, M - 1$  ;  $v = 0, 1, 2, \dots, N - 1$  – frequency coordinates.

That is the dimension of matrix is stored in the frequency domain, but its elements are complex numbers. Inverse DFT  $f(x, y) = \text{ifft2}(F(u, v))$  is as follows

$$f(x, y) = \frac{1}{MN} \sum_{u=0}^{M-1} \sum_{v=0}^{N-1} F(u, v) e^{-j2\pi(ux/M+vy/N)} \quad (7)$$

Comparing the two images  $f_1(x, y)$  and  $f_2(x, y)$  using DFT carried out by convolution in the frequency domain:

$$F_1(u, v) = \text{fft2}(f_1(x, y)) \quad (8)$$

$$F_2(u, v) = \text{fft2}(f_2(x, y)) \quad (9)$$

$$F_{con}(u, v) = F_1(u, v) \cdot F_2^*(u, v) \quad (10)$$

$$f_{con}(x, y) = \text{Re}(\text{ifft2}(F_{con}(u, v))) \quad (11)$$

where  $F_2^*(u, v)$  – DFT matrix of complex conjugated elements.

When comparing of investigated  $f_i(x, y)$  and model  $f_s(x, y)$  images define the correlation coefficient between them.

$$k_{corr} = \frac{\frac{1}{M_i N_i} \sum_{x=0}^{M_i-1} \sum_{y=0}^{N_i-1} \left[ \text{Re}(\text{ifft2}(\text{fft2}(f_i(x, y)) \cdot \text{fft2}^*(f_s(x, y)))) \right]}{\frac{1}{M_s N_s} \sum_{x=0}^{M_s-1} \sum_{y=0}^{N_s-1} \left[ \text{Re}(\text{ifft2}(\text{fft2}(f_s(x, y)) \cdot \text{fft2}^*(f_s(x, y)))) \right]} \quad (12)$$

Then conducted comparison image investigated particles  $x_i$  with a set of sample images  $m_j$  of  $k$  types (groups) of particles. The decision to belong to a group of particles performed on the maximum level of the coefficient of correlation. If the resulting value of the maximum correlation coefficient does not exceed a specified threshold value, the particle does not match any of the available types.

On the basis of recognition particle using convolution in the frequency domain using DFT calculate the ratio between the particles of certain groups in total number.

Using spatial correlation images with convolution in the frequency domain with application DFT allows for the detection of particles series of multispectral images and get the ratio of particles of different groups. However, error recognition at the same time is commensurate with the results of visual microscopy.

To increase the probability of control of the concentration of phytoplankton particles should be reduced recognition error of type particles. For this we use the method of detection based on a combination of one particle images obtained at different wavelengths, and comparing them with exemplary multispectral images (Fig. 2).

It should find a set of Euclidean distances between each of the matrices of the family, which will require considerable time for calculation. Another method is to calculate a weighted average of the distance between vectors and families, and the distance is determined by the weight of the matrix, the inverse covariance matrix  $C_Y$  model image. This metric is defined Mahalanobis distance (Gonzalez *et al.*, 2009)

$$d(m_Y, m_X) = (m_Y - m_X)^T C_X^{-1} (m_Y - m_X) \quad (13)$$

where average families vectors defined as follows:

$$m_X = \frac{1}{L} \sum_{l=1}^L x_l \quad (14)$$

$$m_Y = \frac{1}{L} \sum_{l=1}^L y_l \tag{15}$$

and the covariance matrix of the family model images obtained so

$$C_Y = \frac{1}{L-1} \sum_{l=1}^L (y_l - m_Y)(y_l - m_Y)^T \tag{16}$$

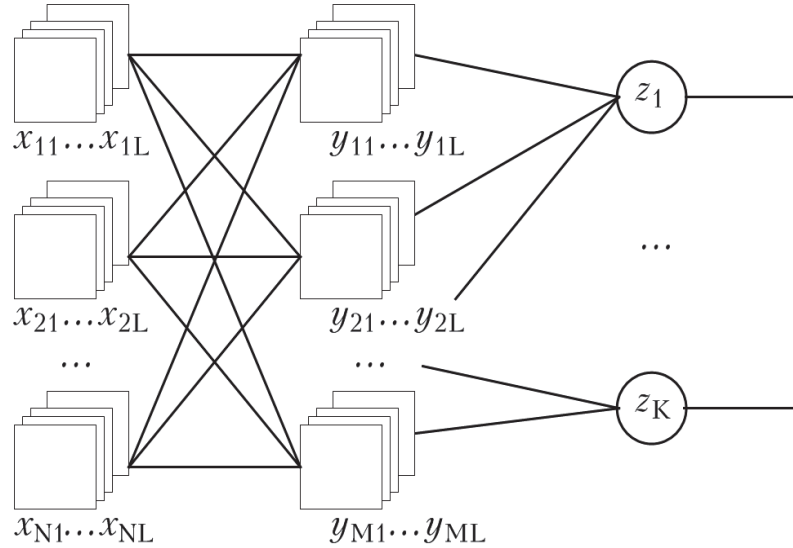


Fig. 2. The scheme of recognition particle based on comparison with exemplary multispectral imaging using Bayesian classifier

We carry out multispectral detection of particles using a statistically optimal classifier Bayesian that has the following crucial function for zero-unit loss function

$$d_j(x|\omega_j) = p(x|\omega_j)P(\omega_j) \tag{17}$$

where  $j = 1, 2 \dots K$ ,  $p(\mathbf{x}|\omega_j)$  – density function probability distribution for the feature vector class  $\omega_j$ ,  $P(\omega_j)$  – class probability of detection  $\omega_j$ . At a certain feature vector classification process is to calculate all the key rules and to appoint the image class, the decisive rule which gives the number increases.

The probability density function for the feature vector n-dimensional Gaussian random variable have the form:

$$p(m_x|\omega_j) = \frac{1}{(2\pi)^{n/2} |C_j|^{1/2}} e^{-\frac{1}{2}[(m_x - m_j)^T C_j^{-1} (m_x - m_j)]} \tag{18}$$



where  $C_j$  and  $m_j$  – covariance matrix and middle-class vector of family of exemplary pictures;  $m_x$  – middle-class vector of family of investigated images;  $|C_j|$  – determinant of matrix  $C_j$ .

Since finding of maximum function  $d_j(m_x)$  is equivalent to finding the maximum  $\ln(d_j(m_x))$ , we can write a function in such a way

$$d'_j(m_x) = \ln(p(m_x|\omega_j)P(\omega_j)) = \ln(p(m_x|\omega_j)) + \ln(P(\omega_j)) \quad (19)$$

Substituting in (19) a particular function for multivariate Gaussian values, we obtain

$$d'_j(m_x) = \ln(P(\omega_j)) - \frac{n}{2} \ln 2\pi - \frac{1}{2} \ln |C_j| - \frac{1}{2} \left[ (m_x - m_j)^T C_j^{-1} (m_x - m_j) \right] \quad (20)$$

Discarding is the same for all classes of constant value  $\frac{n}{2} \ln 2\pi$ , we obtain a decisive function with the Mahalanobis distance in square brackets

$$d''_j(m_x) = \ln(P(\omega_j)) - \frac{1}{2} \ln |C_j| - \frac{1}{2} \left[ (m_x - m_j)^T C_j^{-1} (m_x - m_j) \right] \quad (21)$$

By increasing the probability of a correct recognition of the particles when using multispectral recognition using the classifier Bayes with decision functions based on Mahalanobis distance in feature space has improved accuracy in determining the ratio of particles phytoplankton groups compared to their recognition on the basis of a correlation processing using a convolution in the frequency domain using DFT.

#### **4 Experimental analysis of the results of monitoring of phytoplankton**

We carry out an assessment of ecological status of water bodies on the basis of indicators of bioindication by phytoplankton. Selection of phytoplankton samples was carried out in water bodies Vinnitsa using bathometer, as well as with the use of filtration and sedimentation method using a membrane filter. Samples are taken at various locations of the water body at different depths using a bathometer for field study of its hydro-biological parameters such as of phytoplankton concentrations of various species. The study of phytoplankton samples was carried out in vitro in a fixed state (16% formalin solution (2 ml / 200 ml)). The samples are protected from direct sunlight and stored at a constant temperature.

The differences between the spectral absorption characteristics of different groups of algae pigments and different character of the effect of temperature on the relative speed of propagation of phytoplankton leads to a seasonally adjusted number of different groups of algae (Fig. 3).

On the basis of the results of research found seasonal variation changes the relationship between different groups of phytoplankton ponds, caused by natural factors - changes in temperature, solar irradiance, concentration and chemical composition of substances entering the waters with sewage.

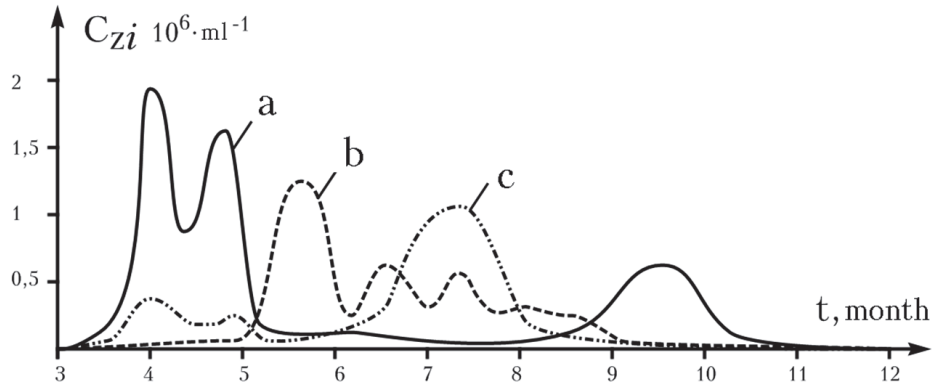


Fig. 3. Seasonal changes in the relations between different groups of phytoplankton:

a – diatoms, b – green, c – blue-green algae

Therefore, it is appropriate to use bioindication indexes to assess the human impact. The degree of indicator species is established using pivot tables and atlases saprobity organisms and monographic treatments of a particular group aquatic organisms (*Barinova et al., 2004, 2005, 2006; Fiorani et al., 2008; Dubelaar et al., 2000; Kopilov et al., 2002; Giakos; 2006; Malkiel et al., 1999*).

The evaluation of water quality based on the results of bioindication by of phytoplankton carry out such a method. Pollution index is developed based on the method Zelinka-Marwan implemented so (*Dell’Uomo, 1997; Barinova et al., 2004*)

$$S_{EPI} = \frac{\sum_{i=1}^N s_i C_{zi} J_i}{\sum_{i=1}^N C_{zi} J_i} \tag{22}$$

where  $N$  – number of phytoplankton species that are bioindicators;  $C_{zi}$  – the concentration of particles of phytoplankton  $i$ -species;  $s_i, J_i$  – saprobity a valence and indicator weight species are taken from the reference tables for the species bioindicators.

## 5 Conclusions

The development of processing industry resulting increase in man-made impact on aquatic ecosystems primarily by increasing the discharge of pollutants from wastewater into local water bodies. Even if new modern technological equipment a number of chemical substances used in manufacturing can get into waste water, such as violations of technological processes or failure of treatment equipment. Therefore the theme of operational control of wastewater toxicity is particularly relevant for environmental inspections.

The paper analyzed the evaluation methods of ecological state of water bodies based on bioindication by phytoplankton and features experimental techniques.

Comparison bioassay results wastewater using phytoplankton culture *Scenedesmus subspicatus* and experimental studies of pollutants specialized laboratory environmental inspection. We conducted a correlation analysis to identify relationships between concentrations of pollutants and the ratio of the concentrations of phytoplankton in the investigated and the control sample. We found correlations between some parameters of pollutants (concentration of sulfate, ammonium ions, nitrite ions, chloride ions, biochemical oxygen demand) and concentration of phytoplankton, which confirms the possibility of using *Scenedesmus subspicatus* cultures phytoplankton as bioindicator contamination of waste water. According to the methodology of environmental assessment of surface water quality categories for the relevant water samples are classified as moderately polluted. The results of this study can be used in specialized laboratories for environmental inspections rapid control parameters wastewater.

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