

Conventional process schemes of municipal sewage water treatment, advantages, and disadvantages of the methods applied when removing biogenic elements were considered. It was shown that the existing shortcomings cause additional explicit costs and difficulties when disposing of the resulting waste. Low efficiency of the removal processes themselves causing residual concentrations of biogenic elements in the treated sewage water was also shown. A process scheme for treating municipal sewage water was proposed. It includes the use of a photobioreactor of a proposed design for the removal of biogenic elements due to the metabolism of microalgae. It was experimentally shown that the use of *Euglena gracilis* strain for removal of phosphates in initial concentrations of 4, 7, and 14 mg/dm<sup>3</sup> from sewage water is the most efficient way. It makes it possible to reduce these concentrations to residual 0...0.55 mg/dm<sup>3</sup> in four days. A 3.75...5.58 times increment of microalgae biomass during this period was also shown. A mathematical model was constructed for calculating the time of staying the sewage water and microalgae mixture in a photobioreactor to achieve the required degree of removal of biogenic elements. Based on the proposed model and experimental studies, the required time of staying in the reactor working area was calculated. It was shown that with the use of *Euglena gracilis* strain but without removal of biogenic elements at previous purification stages (process schemes including only mechanical purification), the time of mixture staying in the working zone was 37.81 hrs. With partial removal of biogenic elements at the stage of biological treatment (0.55 mg/dm<sup>3</sup> total nitrogen, 0.91 mg/dm<sup>3</sup> ammonium nitrogen, 0.44 mg/dm<sup>3</sup> phosphates), this time was reduced to 26.66 hrs. It was found that the use of *Euglena gracilis* strain instead of *Chlorella vulgaris* (FC-16) in the removal of phosphates results in a 2-time increase in the process efficiency and a 50 % decrease in time of mixture staying in the working zone. Recommendations for calculating geometric parameters of photobioreactors of the proposed design were given for use in the process schemes

**Keywords:** biogenic elements, microalgae, nitrates, nitrites, process scheme, phosphates, photobioreactor

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# IMPROVING THE PHOTOBIOREACTOR OPERATION EFFICIENCY IN THE TECHNOLOGICAL SCHEME OF WASTEWATER TREATMENT

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## 1. Introduction

Eutrophication, the uncontrolled explosive development of microalgae, is often observed in surface water. One of these phenomena causes is the entry of biogenic elements into water

bodies. They enter with agricultural surface runoff, municipal sewage, sewage from industrial enterprises, etc.

According to Council Directive 91/271/EEC of 21 May 1991 named On Urban Waste Water Treatment, the following concentrations of biogenic elements must be ob-

served when discharging treated sewage water into water bodies: 2 mg/dm<sup>3</sup> for total phosphorus in settlements and their agglomerations with population up to 100,000 people and 1 mg/dm<sup>3</sup> in settlements with population more than 100,000 people; 15 mg/dm<sup>3</sup> for total nitrogen in settlements with population up to 100,000 people and 10 mg/dm<sup>3</sup> in settlements with population more than 100,000 people. At the same time, data for sewage subjected to just mechanical treatment often exceed 9 mg/dm<sup>3</sup> for total phosphorus and 55 mg/dm<sup>3</sup> for total nitrogen. Even after biological treatment, figures often exceed 6 mg/dm<sup>3</sup> for total phosphorus and 25 mg/dm<sup>3</sup> for total nitrogen. Methods of additional purification from biogenic elements involve their conversion to insoluble state with subsequent precipitation and disposal of sludge. This requires additional costs.

At the same time, the demand for microalgae biomass is constantly increasing. It is a raw material for manufacturing various marketable products. To obtain it, various cultivation methods are used. However, what they have in common is the use of biogenic elements.

There are different designs of photobioreactors for cultivation. Their vast majority can be used for sewage treatment by combining biomass cultivation with the removal of biogenic elements from the culture medium. This reduces the cost of both processes, unwanted amount of sludge, and risk of eutrophication.

Thus, we may assert that the use of microalgae in schemes of treating sewage of various origins is a promising line of improving water treatment processes and raising the safety of present-day sewerage systems.

There is an urgent problem of finding a microalgae culture for use in photobioreactors, improving the efficiency of their operation, and developing methods for calculating process variables and modes of photobioreactor operation.

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## 2. Literature review and problem statement

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One of the effective methods of removing biogenic elements from sewage water consists in the use of microalgae metabolism [1]. The metabolism results in the absorption of phosphorus and nitrogen compounds, carbon dioxide, and light energy [2]. The resulting biomass can be used for manufacturing various products [3].

A number of microalgae strains can be used to remove biogenic elements from sewage: *Chlorella* [4], *Botryococcus* [5], *Scenedesmus* [6], *Chlamydomonas* [7], *Spirulina* [8], *Haematococcus* [9], and others. Besides, these microorganisms can reduce the chemical and biochemical oxygen demand of treated sewage water [10]. Due to metabolism, it can also absorb significant amounts of radionuclides, phenols, and heavy metals [11]. An important positive consequence also consists in the absorption of carbon dioxide from the environment [12].

Mechanical, biological, and chemical treatment is mainly used for sewage treatment. Various schemes are applied. They often differ in the use of biological and chemical methods [13]. Their disadvantages include insufficient removal of biogenic elements and the formation of sediments that are difficult to dispose of. The schemes using sewage treatment with activated sludge and subsequent secondary settling for its removal can be considered basic ones for the facilities treating domestic sewage water [14]. Insufficient removal

of biogenic elements from sewage can also be considered a disadvantage of these schemes.

There are two main causes of using such schemes. In the first case, when treating with activated sludge to reduce biochemical oxygen demand, a nitrification process is used which makes it possible to convert ammonium nitrogen into nitrates. Denitrification is not envisaged in this case [15]. Thus, complete removal of nitrogen from sewage water cannot be provided. In the second case, partial denitrification is used. This can be considered a conventional scheme of removing biogenic elements from sewage. As a rule, it is used at sewage treatment plants of a rather small capacity. Anoxide reactors with sludge mixture recirculation for partial denitrification are provided in the head of aeration tanks [16]. Disadvantages include its ability to only partial removal of nitrogen while phosphorus compounds are not removed. To remove them before sending sewage to secondary settling tanks, chemical reagents (mainly iron or aluminum compounds) are added [17]. A disadvantage of this method consists also in that excess activated sludge becomes problematic to use as an organic fertilizer in agriculture. Besides, when using such a scheme, it becomes necessary to create and maintain a reagent farm which will significantly increase the cost of the cleaning processes.

Acidification methods are also used to remove phosphorus compounds. A small amount of easily oxidizable organic compounds entering the sewage water can be considered one of the main difficulties of biological methods of phosphorus removal. Acidification makes it possible to increase the concentration of such compounds in sewage water. Sewage water of direct settling is fed into an aeration tank or methanol or acetic acid compounds are fed into the anaerobic zone. During sludge acidification, it is possible to significantly increase the content of volatile fatty acids in sewage water entering for treatment. As a result, their concentration grows in the anaerobic zone of the aeration tank. To implement this method, changes must be introduced to the treatment scheme. A part of primary settling tanks should be left for sewage water treatment and a part should be used for compaction of raw sludge with acidification. The water drained from these settling tanks after compaction should be sent to aeration tanks [18]. A disadvantage of such a scheme consists in reduced productivity of the treatment plants or the necessity of building additional facilities which will increase capital costs.

Acidification in the first anaerobic zones of aeration tanks is also possible. If there is no mixing, activated sludge is compacted and refermentation of organic matter sorbed on the activated sludge takes place. Consumption of volatile fatty acids with a release of phosphates with the participation of phospho-accumulating bacteria continues in these zones. Increased concentration of these acids significantly activates the process. This scheme is more economical as it does not require additional tanks for settling and fermentation [19]. Its disadvantage consists in that its cost remains high.

To activate acidification, immobilization of activated sludge in the anaerobic zone using downloading is also applied. A biofilm is formed mainly of anaerobic heterotrophic bacteria which quickly adapt to the incoming organic matter. Such immobilization provides better resistance to changes in characteristics of incoming effluents and stabilizes and accelerates the process [20]. An increase in capital expenditures is another disadvantage.

The use of microalgae can be considered one of the most effective and environmentally friendly methods of removing biogenic elements from sewage water [4]. The advantage of such methods consists in that not only removal of biogenic elements but also accumulation of biomass rich in lipids, phosphates, and nitrates take place in the process of sewage water treatment. For a long time, microalgae were mainly considered as a raw material for lipid production as well as a means of reducing carbon dioxide emissions in industrial processes. Besides, the resulting excess biomass can be used in the manufacture of various marketable products [21]. The high biodiversity of energy-rich microalgae makes it possible to produce hydrogen by photolysis of water, lipids for the production of biodiesel or biokerosene, and sugar for fermentation of biomass (methane) or gas [22]. Potential production of biofertilizers [23] and, taking into account population growth, meeting food needs, and improving agriculture sustainability are of crucial importance among all agricultural issues. The use of such fertilizers increases agricultural production, increases yields, and crop quality while reducing carbon emissions into the environment.

Sewage water treatment plants are designed for various productivity levels (daily volume of treated sewage water), different concentrations of contaminants in sewage water, and different treatment efficiencies. The length of stay of treated sewage water in working zones is one of the main process variables for designing treatment plants. This variable makes it possible to determine the geometric dimensions of plants and their modes of operation. The problem of developing methods of their design is relevant for the introduction of photobioreactors in purification schemes. The development of a mathematical model of photobioreactor which will make it possible to determine its process variables in concrete conditions may be the first step.

Various types of photobioreactors can be used for sewage water treatment and simultaneous cultivation of microalgae. It can be affirmed that closed reactors are more promising because they have many advantages over open ones. There are many studies on the effectiveness of the use of closed reactors with various microalgae strains [24]. The use of freshwater as a culture medium is their disadvantage. Mathematical models of dynamics of biomass growth in photobioreactors of various designs are developed. The disadvantages of such models are as follows: the use of freshwater is assumed and only the increase in biomass is determined in [25]; the increase in biomass in a cylindrical reactor is determined in [26]; increase in biomass and CO<sub>2</sub> uptake taking into account characteristics of the airlift reactor is determined in [27]; the increase in biomass and CO<sub>2</sub> uptake taking into account changes in lighting is determined in [28]. All these models do not take into account the possibility of using sewage water as a culture medium and do not determine the dynamics of removal of biogenic elements.

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### 3. The aim and objectives of the study

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The study objective consists in checking the efficiency of phosphate uptake and biomass growth by *Euglena gracilis* microalgae (HPDP-114) strain. When using schemes with photobioreactors of ideal mixing at treatment facilities, the model will make it possible to calculate the required

duration of sewage water treatment with microalgae. The model should take into account the need to achieve the specified concentrations of biogenic elements at the outlet of treatment plants at different stages of their removal in the previous stages of treatment.

To achieve this objective, the following tasks were set:

- conduct experimental cultivation of *Euglena gracilis* (HPDP-114) microalgae strains in artificial sewage water at different concentrations of phosphates and determine the dynamics of their removal from sewage water as well as the dynamics of the microalgae biomass growth during cultivation;

- develop a mathematical model of the photobioreactor dynamics and calculate the duration of sewage water staying in the working zone of ideal mixing photobioreactor to achieve the required degree of phosphate release at their different initial concentrations;

- develop recommendations for designing ideal displacement photobioreactors for the use in schemes of sewage water treatment as well as for the use of microalgae strains to increase their efficiency.

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### 4. The study materials and methods

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Cultivation of *Euglena gracilis* (HPDP-114) microalgae strains in artificial sewage water with different concentrations of phosphates was performed in the study. The studied strain was obtained from the Bank of Microalgae Cultures of the Institute of Hydrobiology of the National Academy of Sciences of Ukraine. Artificial sewage water was prepared according to the recipe of biogenic culture medium containing 0.4 g/dm<sup>3</sup> magnesium sulfate solution (MgSO<sub>4</sub>·7H<sub>2</sub>O); 0.05 g/dm<sup>3</sup> calcium chloride solution (CaCl<sub>2</sub>·2H<sub>2</sub>O); 0.1 g/dm<sup>3</sup> sodium chloride (NaCl); 5 g/dm<sup>3</sup> glucose; 1 g/dm<sup>3</sup> α-glutamic acid; 1 g/dm<sup>3</sup> trace element solution; 1g/dm<sup>3</sup> ethylenediaminetetraacetic acid solution with iron ions (Fe-EDTA). To create a concentration of biogenic elements close to the concentration in real sewage water, potassium dihydro orthophosphate KH<sub>2</sub>PO<sub>4</sub>, and ammonium chloride NH<sub>4</sub>Cl were added to the culture medium in different amounts to achieve different phosphorus concentrations. Three experiments with different initial concentrations of phosphorus in synthetic sewage water were performed. In terms of orthophosphates, concentration was as follows: 4 mg/dm<sup>3</sup> in the first experiment; 7 mg/dm<sup>3</sup> in the second experiment; 14 mg/dm<sup>3</sup> in the third experiment.

Prior to the experiments (addition of microalgae to artificial sewage water), they were pre-adapted to the culture medium with the preparation of inoculum. The adaptation period was 7 days.

Inoculum of the adapted *Euglena gracilis* (HPDP-114) strain was mixed with three artificial sewage water samples with different concentrations of phosphorus in a ratio of 1:10. After mixing, the initial concentration of microalgae in each sewage water sample was 120 mg/dm<sup>3</sup>. The mixtures for cultivation were placed in 0.5 dm<sup>3</sup> clear glass flasks. Cultivation was performed by maintaining the temperature of 25...26.2 °C with artificial lighting at the intensity of 48–52 μmol-photon/m<sup>2</sup>·s and alternating light and darkness (16:8 hours).

Samples for measuring the concentration of phosphorus and microalgae cells were taken before the start of the experiment, after the 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup>, and 4<sup>th</sup> days of cultivation.

The concentration of reactive phosphorus (orthophosphates) in sewage water was measured by standard photometric method with amino acid [29]. A DR/3900VIS spectrophotometer manufactured by Hach Co. was used with the following characteristics: spectral wavelength range of 320–1100 nm; wavelength error of ±1.5 nm; a photometric range of 340–900 nm; wavelength resolution of 1 nm. The concentration of microalgae cells in artificial sewage water was determined by the weight method. The cell suspension was filtered through pre-dried and weighed 0.45 µm Sartorium membrane filters. Filters with precipitated algae were dried in a thermostat at 105 °C to a constant weight and weighed again. The weighing was performed using TBE-0.3-0.005-a laboratory electronic scales with a measurement resolution of 0.005 g. Concentration was determined by the formula  $C_p = \frac{M_{algae}}{V_{sew}}$ , where  $M_{alg}$  is the dry mass of microalgae retained by the filter,  $V_{sew}$  is the filtered volume of the selected sewage water sample.

All measurements were performed three times. The discrepancy between indicator values was estimated using the method of standard deviation and the Student's t-test. The result was considered trustworthy if  $p \leq 0.05$  condition was satisfied. The medium pH remained in the range of 7.5...8.0 in all experiments. The medium was monitored with Greisinger GMH 3351 pH meter.

**5. The results obtained in studying the efficiency of biogenic element removal in the photobioreactor**

**5.1. Dynamics of phosphate removal from sewage water and growth of microalgae biomass**

Concentrations of phosphorus and microalgae cells in artificial sewage water obtained in three variants of the experiment are given in Tables 1, 2. Results of phosphorus removal and increment of biomass of microalgae cells during each day are given in percent and absolute units in Tables 3, 4.

Table 1

Concentration of phosphorus in artificial sewage water samples in three experiments

Experiment No.	Phosphorus concentration, mg/dm <sup>3</sup>				
	Start	Day 1	Day 2	Day 3	Day 4
1	4.00	3.10	0.22	0.16	traces
2	7.00	5.53	1.66	0.22	traces
3	14.00	11.22	4.17	1.70	0.55

Table 2

Concentration of microalgae cells in artificial sewage water samples in three experiments

Experiment No.	Concentration of microalgae cells, mg/dm <sup>3</sup>				
	Start	Day 1	Day 2	Day 3	Day 4
1	120	160	240	380	570
2	120	165	280	450	670
3	120	170	310	505	790

Graphs in Fig. 1, a show the dynamics of decrease in the concentration of phosphorus in sewage water and Fig. 1, b shows the dynamics of growth of biomass of the cultured microalgae strain in four cultivation days.

Table 3

Removal of phosphorus from artificial sewage water each day

Experiment No.	Day 1	Day 2	Day 3	Day 4
Removal of phosphorus, %				
1	22.5	92.9	27.3	–
2	21.0	70.0	86.8	–
3	19.9	62.8	59.2	67.7
Removal of phosphorus, mg/dm <sup>3</sup>				
1	0.90	2.88	0.06	0.15
2	1.47	3.87	1.44	0.21
3	2.78	7.05	2.47	1.15

Table 4

Increment of microalgae cell biomass in artificial sewage water each day

Experiment No.	Day 1	Day 2	Day 3	Day 4
Biomass increment, %				
1	33.3	50.0	58.3	50.0
2	37.5	69.7	60.7	48.9
3	41.7	82.4	62.9	56.4
Biomass increment, mg/dm <sup>3</sup>				
1	40	80	140	190
2	45	115	170	220
3	50	140	195	285

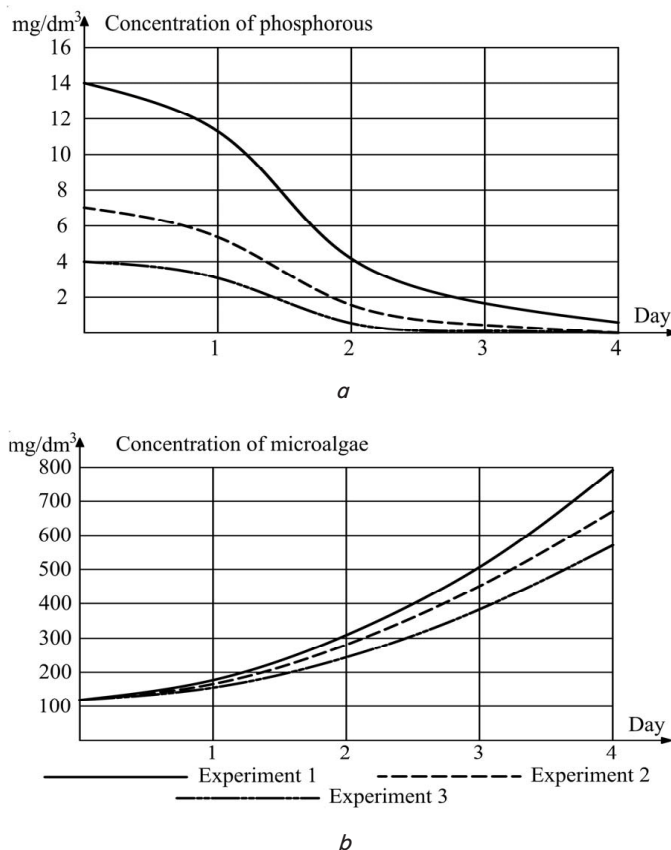


Fig. 1. Dynamics of concentration change: a – for phosphorus in sewage water; b – for microalgae biomass in sewage water

It is seen from Table 1 that phosphorus was almost completely removed from sewage water in four days. Only in the third variant of the experiment, its concentration of

0.55 mg/dm<sup>3</sup> was recorded at the end. It is seen from Table 2 that the concentration of microalgae cells has increased in all experiment variants: 3.75 times in variant 1; 4.58 times in variant 2 and 5.58 times in variant 3. Since initial cell concentration and culture condition were the same (120 mg/dm<sup>3</sup>), this suggests that there is a direct relationship between phosphorus concentration in the culture medium and the biomass increment. The biomass increment grows with concentration increment within 4...14 mg/dm<sup>3</sup>.

It is seen from Table 3 and Fig. 1 that the highest rate of phosphorus removal in absolute units (2.88...7.05 mg/dm<sup>3</sup>) and in percent (62.8...92.9 %) was achieved on the second day of cultivation in all experiment variants (except the second variant where the maximum percentage (86.8 %) was observed during the third day of cultivation). During the first day, this rate was much lower (0.9...2.78 mg/dm<sup>3</sup> and 19.9 (22.5 %). Phosphorus concentration continued to decrease effectively during the third and fourth days. Further cultivation (more than 4 days) to remove phosphorus was unnecessary as its concentration has become insignificant (traces in the first two experiment variants and 0.55 mg/dm<sup>3</sup> in the third variant).

Table 4 and Fig. 1, *b* show that the rate of biomass growth was higher in those experiment variants where the initial concentration of phosphorus was higher. A maximum percentage increase in biomass was observed: in the first variant of the experiment (53 %) during the third day. The second and third variants have demonstrated 69.7 % and 82.4 %, respectively, during the second day of cultivation. During the first day, the increment was much smaller (33.3...41.7 %) which coincided with less phosphorus uptake and may also indicate a possible adaptation of the culture to environmental conditions. During the fourth day, the concentration of phosphorus in sewage water was significantly reduced and the increment was also reduced but remained quite significant (48.9...56.4 %).

**5. 2. Mathematical model of photobioreactor dynamics and calculation of the duration of sewage water staying in the working zone of the photobioreactor**

Photobioreactor design offered by the author is presented in [30]. It is a batch photobioreactor that may be considered a perfect mixing reactor. Concentrations of microalgae and biogenic elements are the same in its entire volume at all times. It changes only over time. Sewage water with recycled microalgae biomass is periodically fed to the reactor working zone. After passing the stage of absorption of biogenic elements due to the photosynthesis processes, sewage water with microalgae is released from the working zone and subjected to the separation of the microalgae biomass. The separator was designed as a hydrocyclone.

To establish the photobioreactor dynamics, it is necessary to decide on the time of staying of the mixture of sewage water and microalgae in the working zone to achieve the required reduction of biogenic elements concentration in sewage water due to metabolism.

The following equation of material balance of biogenic elements in the working zone can be taken as a basic equation for calculation [31]:

$$\frac{\partial S_i}{\partial t} = \left( -v_{xi} \cdot \frac{\partial S_i}{\partial x} - v_{yi} \cdot \frac{\partial S_i}{\partial y} - v_{zi} \cdot \frac{\partial S_i}{\partial z} \right) + k_d \cdot \left( \frac{\partial^2 S_i}{\partial x^2} + \frac{\partial^2 S_i}{\partial y^2} + \frac{\partial^2 S_i}{\partial z^2} \right) + \mu_i, \tag{1}$$

where

- $S_i$  is the concentration of the  $i$ -th biogenic compound in sewage water;
- $t$  is time;
- $x, y, z$  are space coordinates;
- $v_x, v_y, v_z$  are components of sewage water velocity;
- $k_d$  is molecular and convective diffusion coefficient;
- $\mu_i$  is the rate of absorption of biogenic elements.

Since all parameters of the batch reactors are the same at any time and all points of the working zone space, derivatives of any order from biogenic element concentration in space will be zero. That is:

$$\left( -v_{xi} \cdot \frac{\partial S_i}{\partial x} - v_{yi} \cdot \frac{\partial S_i}{\partial y} - v_{zi} \cdot \frac{\partial S_i}{\partial z} \right) = 0, \tag{2}$$

$$k_d \cdot \left( \frac{\partial^2 S_i}{\partial x^2} + \frac{\partial^2 S_i}{\partial y^2} + \frac{\partial^2 S_i}{\partial z^2} \right) = 0. \tag{3}$$

Thus, (1) can be written as follows:

$$\frac{\partial S_i}{\partial t} = \mu_i. \tag{4}$$

If the volume of the mixture of sewage water with microalgae in the working zone is considered constant, the following can be written:

$$\frac{d[S_{i, start}(1 - K)]}{dt} = \mu_i, \tag{5}$$

where

- $S_{i, start}$  is the concentration of biogenic elements at the beginning of the process;
- $K_i$  is the degree of removal of the biogenic compound at the end of the process.

Expression (5) can also be written as follows:

$$dt = S_{i, start} \cdot \frac{dK_i}{-\mu}. \tag{6}$$

The rate of absorption of biogenic compounds by microalgae can be written as

$$-\mu_i = k_i \cdot S_i^n, \tag{7}$$

where  $k_i$  is the constant of the rate of absorption of the biogenic compound;  $n$  is the order of reaction (if absorption resulting from metabolism is considered as a chemical reaction).

Formula (6) can be written as follows taking into account (7):

$$dt = S_{i, start} \cdot \frac{dK_i}{k_i \cdot S_i^n}. \tag{8}$$

Taking into account (5), formula (8) can be represented as:

$$dt = S_{i, start} \cdot \frac{dK_i}{k_i \cdot S_{i, start}^n (1 - K_i)^n}. \tag{9}$$

Integration of (9) in the range from  $K_{i, start}$  (initial degree of removal of the biogenic compound at the beginning of the

process) to  $K_i$  (final degree of removal of the biogenic compound at the end of the process) gives

$$t = S_{i \text{ start}} \int_{K_{i \text{ start}}}^K \frac{dK_i}{k_i \cdot S_{i \text{ start}}^n (1 - K_i)^n}, \quad (10)$$

or

$$t = \frac{1}{k \cdot S_{i \text{ start}}^{n-1}} \int_{K_{i \text{ start}}}^K \frac{dK_i}{(1 - K_i)^n}. \quad (11)$$

The absorption of biogenic elements due to microalgae metabolism can be considered a first-order reaction [32]. Under such conditions, integration of formula (11) gives this expression:

$$t = \frac{1}{k} \ln \frac{1 - K_{i \text{ start}}}{1 - K_i}. \quad (12)$$

Thus, it can be affirmed that the time of staying of the sewage water in the photobioreactor working zone depends on the required degree of removal of biogenic elements before discharging the treated sewage water into reservoirs  $K_i$ . It also depends on the initial degree of removal of biogenic elements at previous stages of purification (biological purification) at the entrance to the photobioreactor  $K_{i \text{ start}}$ . If there is no removal of biogenic elements the previous stages of purification at all, the initial degree of extraction can be taken equal to zero ( $K_{i \text{ start}}=0$ ).

Using the results obtained in the experiments, the required time of sewage staying in the reactor working zone to achieve the required degree of removal of phosphorus compounds was determined. For this purpose, the equation of phosphorus absorption rate constant under specific conditions was used [31]:

$$k_i = \frac{\ln S_{i \text{ start}} - \ln S_i}{t}. \quad (13)$$

These calculations did not take into account the results of the first day of cultivation as it was believed that microalgae were undergoing a period of adaptation to the environment. The rate constant (0.052) was calculated in this way and averaged over three experiments.

Municipal sewage water may have different concentrations of phosphorus compounds depending on the effluent origin [33]. Average concentrations (of  $\text{PO}_4$ ) were taken for calculation: 7.2 mg/dm<sup>3</sup> for the sewage without biological treatment, 4.0 mg/dm<sup>3</sup> for the sewage subjected to biological treatment, and 1.0 mg/dm<sup>3</sup> for the sewage discharged into water bodies (regulatory requirements). The required degree of phosphate removal to achieve the regulatory requirements for concentration in sewage discharged into water bodies (1.0 mg/dm<sup>3</sup>) at an initial concentration of 7.2 mg/dm<sup>3</sup> will be  $K_{\text{PO}_4}=0.86$ .

In absence of prior removal of biogenic elements (non-pre-treated sewage), the initial degree of phosphate removal will be  $K_{\text{PO}_4, \text{no}, 0}=0$ . In this case, the time of staying of sewage in the reactor working zone calculated from (12) will be

$$t_{\text{PO}_4}^0 = \frac{1}{0.052} \ln \frac{1-0}{1-0.86} = 37.81 \text{ hrs.}$$

For effluents after biological treatment (phosphate concentration of 4.0 mg/dm<sup>3</sup>), the initial degree of phosphate

removal will be  $K_{\text{PO}_4, \text{start}}=0.44$ . Under such conditions, the time of staying of sewage in the working zone will be

$$t_{\text{PO}_4} = \frac{1}{0.052} \ln \frac{1-0.44}{1-0.86} = 26.66 \text{ hrs.}$$

### 5.3. Recommendations on the design of photobioreactors for sewage water treatment

The proposed mathematical model (12), (13) makes it possible to determine the efficiency of removal of biogenic elements from sewage water in the photobioreactor and take into account different efficiency of plants for sewage pre-treatment before its delivery to the photobioreactor.

When preparing the process design of ideal mixing photobioreactors for their use in municipal sewage treatment, (12) and (13) can also be used. The time of sewage staying in the photobioreactor calculated according to these formulas will make it possible to calculate the overall dimensions of the main reactor elements. It will also enable the calculation of operating modes at different concentrations of biogenic elements at the entrance to the treatment plant and various concentrations required at the outlet of purification facilities.

To compare the results obtained with *Euglena gracilis* strain, results of experiments with biomass of the *Chlorella Vulgaris* (FC-16) strain given in [34] were used. Phosphorus and nitrogen compounds were removed from actual municipal sewage water with concentrations of 3.09...5.15 mg/dm<sup>3</sup> for phosphates ( $\text{PO}_4$ ), 37.53...63.24 mg/dm<sup>3</sup> for total nitrogen (N), and 8.80...13.45 mg/dm<sup>3</sup> for ammonium nitrogen ( $\text{NH}_4$ ). Microalgae concentrations were introduced: 1 mg/dm<sup>3</sup>, 2 mg/dm<sup>3</sup>, 4 mg/dm<sup>3</sup>, 6 mg/dm<sup>3</sup> and 10 mg/dm<sup>3</sup>. The process was carried out at 25±1 °C, pH=7.5 and light intensity of 100 mol×s<sup>-1</sup>×m<sup>-2</sup>. The results show that final concentrations of 2.17 mg/dm<sup>3</sup> for phosphates, 8.84 mg/dm<sup>3</sup> for total nitrogen, and 1.00 mg/dm<sup>3</sup> for ammonium nitrogen were obtained at microalgae concentration of 2 mg/dm<sup>3</sup> and initial concentrations of 4.04 mg/dm<sup>3</sup> for phosphates, 53.33 mg/dm<sup>3</sup> for total nitrogen, 11.30 mg/dm<sup>3</sup> for ammonium nitrogen and a 1 day (24 hrs.) stay of microalgae in sewage water.

Besides phosphorus compounds, municipal sewage may contain different concentrations of nitrogen compounds [35, 36]. Their average concentrations are given in Table 1.

Table 1

Concentration of biogenic elements in sewage water at different treatment stages

Sewage treatment stage	Concentration of biogenic elements, mg/dm <sup>3</sup>		
	Total nitrogen (N)	Ammonia nitrogen (NH <sub>4</sub> )	Phosphates (PO <sub>4</sub> )
Sewage without biological treatment	55.0	20.0	7.2
Sewage after biological treatment	25.0	1.8	4.0
Sewage discharged to water bodies (normative requirements)	15.0	0.5	1.0

The degree of removal of biogenic elements will be different after different stages of conventional treatment. Their values are given in Table 2.

**Table 2**  
Degrees of removal of biogenic elements after different stages of treatment

Sewage treatment stage	Degree of removal of biogenic elements		
	Total nitrogen (N)	Ammonia nitrogen (NH <sub>4</sub> )	Phosphates (PO <sub>4</sub> )
After biological treatment	0.55	0.91	0.44
After additional treatment in a photobioreactor	0.73	0.98	0.86

The time of staying of non-pretreated sewage in the working zone of the reactor when using *Chlorella Vulgaris* (FC-16) strain and in the need of achieving regulatory concentrations allowing sewage discharge to water bodies was calculated using (12):  $t_{PO_4}^0=75,62$  hrs;  $t_{NH_4}^0=38,73$  hrs;  $t_N^0=17,46$  hrs.

For sewage subjected to biological treatment, the time spent in the working zone will be  $t_{PO_4}^0=53,32$  hrs;  $t_{NH_4}^0=14,89$  hrs;  $t_N^0=6,80$  hrs.

As the calculations performed for the *Chlorella Vulgaris* strain show, phosphorus is the dictating element among those considered above. Therefore, the duration of sewage staying in the photobioreactor is 75.62 hrs. if the sewage is not pretreated and 53.32 hours in the case of biological pretreatment.

The efficiency of the treatment process is defined as a value inverse to the time of sewage staying in the working zone of the photobioreactor to achieve the required degree of biogenic element removal. The study results show that the efficiency of phosphate removal when using *Euglena gracilis* (HPDP-114) strain is 2.0 times higher compared to the use of the *Chlorella Vulgaris* (FC-16) strain.

The authors have proposed the design of a photobioreactor of ideal mixing [30]. Its working zone volume is proportional to the time of sewage staying in it. Therefore, it is advisable to use the *Euglena gracilis* strain to remove phosphates from sewage water. In this case, the estimated time of sewage staying in the working zone of the proposed photobioreactor and hence its volume is reduced by 50 %.

**6. Discussion of the results obtained in the study of the efficiency of removal of biogenic elements and the recommendation on photobioreactor design to be used in the process schemes of sewage treatment**

It can be seen from the obtained experimental results that the use of *Euglena gracilis* strain makes it possible to effectively remove phosphorus from sewage water and get a significant increment of biomass. At the same time, the higher the phosphorus concentration the higher biomass increment. This can be explained by the fact that phosphorus is a limiting factor determining the rate of metabolism under these cultivation conditions. The dependence of the increment rate on concentration is nonlinear and the rate obviously has a limiting value.

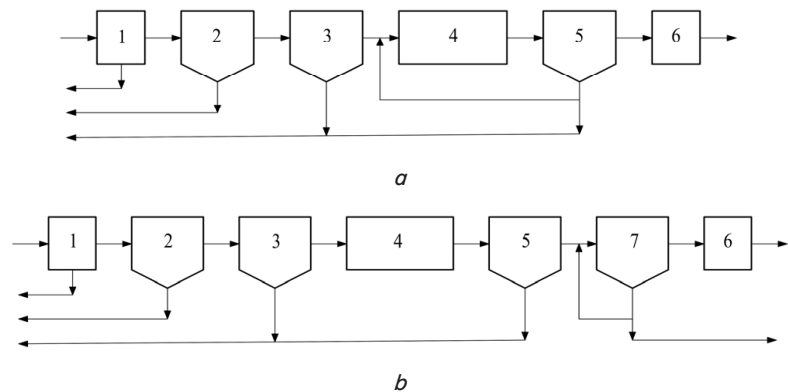
The change in phosphorus removal rate observed in different days of cultivation in each of the experiments was

also nonlinear. The rate during the first day was lower than the average rate. This can be explained by the fact that cells adapt to environmental conditions on the first day (cells are moved from inoculum to sewage water). A further increase in rate may indicate that adaptation is completed and the metabolic rate reaches its highest values.

During the fourth day, along with the reduction of phosphorus concentration to very low values, the biomass growth rate decreases but its value remains significant. This may indicate that phosphorus accumulated in the biomass during the first three days helps maintain metabolism and the lack of phosphorus in sewage water starts suppressing it.

Thus, the determination of the dependence of the biomass growth rate on phosphorus concentration in sewage requires further experimental studies to better understand the process dynamics.

The closed-type photobioreactor proposed by the authors in [30] may be used as a structural element of the conventional process of domestic and similar sewage treatment. The conventional process combining mechanical and biological treatment may include grates (cutting screens), sand traps, primary settling tanks, aeration tanks (biological filters), secondary settling tanks and units for disinfection of sewage water (Fig. 2, a). Besides the above items, a diagram including a photobioreactor installed downstream of the secondary settler [37] can be recommended (Fig. 2, b). The location of the photobioreactor downstream of the secondary settling tank is explained by the need for light access to the microalgae biomass in the entire space of the working zone. Prior to the secondary tank, sewage has a significant concentration of activated sludge, hence it is turbid.



**Fig. 2.** Process schemes of sewage treatment (including removal of biogenic elements): a – without a photobioreactor; b – with a photobioreactor; 1 – gratings; 2 – sand traps; 3 – primary settling tanks; 4 – aeration tanks (biological filters); 5 – secondary settling tanks; 6 – sewage disinfection units; 7 – photobioreactors

The photobioreactor in this scheme operates as follows. Biologically treated and clarified sewage water is mixed with *Euglena gracilis* (HPDP-114) microalgae strain to a concentration of 120 mg/dm<sup>3</sup> and fed to the reactor working zone. The sewage water and microalgae mixture is kept in the working zone of the reactor for at least 26.66 hrs. The working zone temperature of 25...26.2 °C and pH of 7.5...8.0 are maintained. After the process completion, the mixture is drained from the working zone and the algae biomass is removed by means of a hydrocyclone. As the biomass increment takes place during treatment, it is divided after its removal into a “recirculation” biomass which is taken for

mixing with clarified sewage water and then fed to the photobioreactor working zone for the next treatment cycle, and a “surplus” biomass which is taken away as a raw material for making commodity products.

In comparison with existing schemes, the one proposed in this study features the use of a photobioreactor that not only enables the removal of nitrogen and phosphorus compounds from sewage water but also does not form hard to dispose of sediments. To compare, if the sewage water contains 25 mg/dm<sup>3</sup> total nitrogen (N), 1.8 mg/dm<sup>3</sup> ammonium nitrogen (NH<sub>4</sub>), 4.0 mg/dm<sup>3</sup> phosphates (PO<sub>4</sub>) after the stage of biological treatment, these values reduce to 15 mg/dm<sup>3</sup>, 0.5 mg/dm<sup>3</sup>, 1.0 mg/dm<sup>3</sup>, respectively, after treatment in a photobioreactor. At the same time, instead of sediments, microalgae biomass is obtained which can be used in the manufacture of marketable products.

This study limitation consists in that it was based on the use of only one microalgae strain: *Euglena gracilis* (HPDP-114). However, the proposed mathematical model makes any strain applicable. This requires trial cultivation studies to experimentally determine constants of the rate of absorption of total nitrogen, ammonium nitrogen, and phosphates. This can be done either with model solutions or using actual sewage water. A compulsory condition consists in ensuring environmental parameters as close as possible to real ones.

The difficulty of finding a site for the reactor may be the disadvantage of using photobioreactors. Existing treatment plants are often arranged in such a way that sewage flow between successively located treatment plants relies on self-motion due to gravitational forces. The reactor location in the purification scheme cannot be arbitrary. It must be located downstream of the biological treatment plants. Therefore, its recommended location should be as close to them as possible. Finding and preparation of a large enough site with local conditions satisfying it can entail significant capital costs. If there is such a problem, it can be solved by placing the photobioreactor at a distance from the main buildings. In this case, sewage will be supplied to it by pumping via pressure pipelines. Such a decision will require additional operating costs for electricity. The final decision on the design should be made based on technical and economic calculations.

The proposed mathematical model takes into account the dynamics of absorption of biogenic elements with microalgae. However, it does not take into account the dynamics

of the growth of algae biomass itself. It is advisable to use a domestic model with such growth in further studies. Since biomass can be considered a commodity product, the dynamics of its growth will significantly affect the technical and economic performance of the proposed process scheme.

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## 7. Conclusions

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1. *Euglena gracilis* (HPDP-114) microalgae strain was experimentally cultivated in artificial sewage water with different concentrations of phosphates: 4, 7, and 14 mg/dm<sup>3</sup>. It was shown that phosphates were removed from sewage water (residual concentration: 0...0.55 mg/dm<sup>3</sup>) during the cultivation period (four days). A 3.75...5.58 times increment of microalgae biomass was also shown.

2. A mathematical calculation model was constructed and the necessary duration of sewage stay in the photobioreactor working zone was also calculated. It was shown that time of the sewage water stay in the photobioreactor to achieve normative phosphate concentration (1.0 mg/dm<sup>3</sup>) allowable for discharge to water bodies when using *Euglena gracilis* (HPDP-114) strain at average starting phosphate concentration (7.2 mg/dm<sup>3</sup>) in sewage water is 37.81 hrs. without prior biological treatment and 26.66 hrs. with biological pretreatment.

3. Recommendations were elaborated for designing ideal mixing photobioreactors to be used in sewage treatment process schemes. It was shown that the concentration of phosphorus compounds in sewage water is a dictating indicator in the calculation of geometric parameters and modes of photobioreactor operation. To improve the efficiency of phosphate removal from sewage water, the use of *Euglena gracilis* (HPDP-114) strain has been proposed to reduce the photobioreactor working zone size by 50 % compared to the *Chlorella vulgaris* (FC-16) strain.

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