

*Досліджено вплив абіотичних факторів на ефективність видалення сполук нітрогену при очищенні циркуляційної води індустріальних рибницьких господарств у фітореакторі з рясковими. Здійснено аналіз доцільності використання люмінесцентних та світлодіодних ламп в фітореакторі. Визначено раціональні значення основних параметрів системи штучного освітлення для забезпечення видалення сполук нітрогену з циркуляційної води*

*Ключові слова: технологія біологічного очищення, фітореактор з рясковими, видалення сполук нітрогену*

*Исследовано влияние абиотических факторов на эффективность удаления соединений азота при очистке циркуляционной воды индустриальных рыбоводных хозяйств в фитореакторе с рясковыми. Проведен анализ целесообразности использования люминесцентных и светодиодных ламп в фитореакторе. Определены рациональные значения основных параметров системы искусственного освещения для обеспечения удаления соединений азота из циркуляционной воды*

*Ключевые слова: технология биологической очистки, фитореактор с рясковыми, удаление соединений азота*

# TREATMENT OF RECIRCULATING WATER OF INDUSTRIAL FISH FARMS IN PHYTOREACTOR WITH LEMNOIDEAE

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## 1. Introduction

In the strategies for the development of the fish farms in many developed countries, the transition to high-tech methods for the cultivation of fish and other hydrobionts in closed-circuit water systems (CWS) is considered one of the priority. Technological schemes for the cultivation of fish products with the repeated use of water provide minimal dependence on water supply sources, high controllability of the main water parameters and ecological purity. Implementation of such benefits is possible only if the circulating water contaminated in the pools is properly cleaned. Limiting pollution in CWS is the most toxic for fish dissolved metabolites: ammonium nitrogen and ammonia. Therefore, the main task of biological treatment facilities for circulating water is transformation and withdrawal of these compounds.

The efficiency of removing the main pollutants from the circulation water determines not only the coefficient of recycling of the fish farm, but also affects the cost of the grown produce. Reducing the dependence on the volume of water abstraction from a natural water source contributes to the growth of the geography of the CWS spread. Thanks to rational water use, the negative impact on the environment is minimized. Therefore, the development and implementation of reliable and environmentally friendly treatment technologies is an urgent task for the further development of aquaculture in systems with recirculation.

## 2. Literature review and formulation of the problem

Realization in the conditions of a closed CWS circuit of the traditional technology of nitrification is accompanied by a number of problematic aspects of biological and technical nature. The intensity of transformation of nitrogen compounds is limited by the relatively low rates of nitro bacteria metabolism, sensitivity to pH fluctuations, competitive relationships with heterotrophic biota of biofilters-nitrifiers. Also negative aspects of this technology are the need to use external carbon sources to ensure the denitrification process, the risks of poisoning fish with ethanol or methanol (reagents) residues.

Further sustainable development of world aquaculture is possible only if an inexpensive source of proteins is provided and the level of availability of nutrient components in feed is increased [1]. Ensuring the conversion of valuable components of fodder that are not used by fish, polluting circulating water, will significantly reduce the need for nutrients [2]. Therefore, the treatment technology of circulating water must correspond to the concept of integrated multi-trophic aquaculture systems (IMTA). The basic principles of this concept and the features of implementation within existing farms are considered in [1–3].

The most promising treatment agents capable of transforming ammonium nitrogen into fish-accessible protein mass are duckweeds – representatives of the family Araceae

Juss of the subfamily Lemnoideae. Lemnoideae is used as feed organisms for animals [4, 5], treatment agents for the treatment of domestic and industrial wastewater [6, 7], raw materials for biofuel production [8]. Interest in waste water as treatment agents of polluted water in fish farms is justified by the intensity of removal of biogenic elements and the possibility of efficient utilization of the growth of plant biomass as food for fish [9]. This biotechnology makes it possible to use the components of mixed fodders undeveloped by fishes more efficiently and to reduce the volumes of waste generated during the treatment process.

Estimation of the assimilation power of Lemnoideae by nitrogen can be carried out based on an analysis of plant growth rates and chemical composition. In the course of the research, the potential assimilation capacity of *Lemna minor* Linnaeus (1753) is determined by the main biogenic elements [7], which allowed the development of mathematical models for predicting the dynamics of plant biomass in the process of assimilation of dissolved contaminants by them.

With optimal parameters of the cultivation medium Lemnoideae is possible to extract up to 100 % nitrates and 82 % of ammonia for a short period of time [10]. Thus, even when treatment heavily contaminated wastewater in ponds with Lemnoideae processes of extraction of ammonium nitrogen and phosphates proceed several times faster than by nitrification followed by denitrification [5].

The most promising species for cultivating as treatment agents in ultrasound conditions is *Lemna minor* L., 1753 and *Wolffia arrhiza* (L.) Horkel ex Wimm, 1857, due to the fact that these species are better adapted to the contamination of the circulation water, have the highest growth rates and are well eaten by most species of fish that grow in CWS [11]. The positive effect of such vegetative feeding on the growth rate of fish mass has been confirmed in [9, 12].

To ensure the effective operation of the phytoreactor with Lemnoideae in CWS conditions it is necessary to arrange artificial lighting system. This is explained by the fact that even when the phytoreactor is located under a transparent roof, the expediency of artificial prolongation of the day's duration is justified by an increase in the extraction efficiency of the basic biogenic elements by plants. Since in modern conditions the cost of energy carriers can significantly affect the cost of production, to ensure the treatment of water in phytoreactors, rational organization of artificial lighting of the building is necessary.

Determination of rational parameters of artificial lighting of the phytoreactor will allow to provide the necessary effect of treatment by nitrogen with minimal energy costs. In [13] it is noted that the optimal boundaries of the light period for Lemnoideae are 14–16 hours. Therefore, the duration of artificial lighting in the presence of a transparent roof over the phytoreactor should be determined taking into account the duration of daylight hours. If there is no natural insolation in the room, the artificial lighting systems must independently provide the specified daily rate. In addition to the duration, for the Lemnoideae life, the intensity of the light flux is also important.

The accumulated experience of cultivation of Lemnoideae in natural and artificial conditions confirms the prospects of cultivation of these plants as purifying agents for the removal of ammonium nitrogen compounds from water in the CWS. Analysis of summary data on the chemical composition of Lemnoideae and dynamics of population growth in various nutrient media [9, 10, 13] allows for a preliminary

prediction of the elimination capacity by ammonium nitrogen in CWS conditions. Between the rates of plant growth and the assimilation capacity of the main biogenic elements, there is a direct relationship. Therefore, it is necessary to provide optimal lighting parameters for the phytoreactor. The indicated by the separate authors as the optimum limits of the intensity of lighting for the Lemnoideae differ substantially among themselves and do not take into account the physicochemical parameters of the circulating water in CWS. Accordingly, there is a need to determine rational limits of intensity and duration of phytoreactor lighting.

The growth rates of plants in the CWS circulating water obtained with natural insolation [14] can be considered as nominal for these water parameters and taken as a basis for determining the rational values of the parameters of the artificial lighting system. Accordingly, the research tasks include the determination of lighting parameters, which provide approximate the maximum growth rates of Lemnoideae.

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### 3. The aim and objectives of research

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The conducted researches aimed to define rational parameters of artificial lighting of the phytoreactor with Lemnoideae to ensure efficient removal of nitrogen compounds from the water.

To achieve this aim, it is necessary to solve the following tasks:

- to conduct an analytical review of technical characteristics and the possibility of using modern lighting devices in CWS water treatment systems;
- to determine the dependence of the biomass doubling time of plants on the duration of daylight in the conditions of the phytoreactor for treatment of the circulating water in CWS;
- to determine the purifying power of the phytoreactor with a Lemnoideae by nitrogen.

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### 4. Materials and methods for research of the removal of nitrogen and phosphorus compounds in phytoreactors with floating algae

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#### 4.1. The object of research, equipment and devices used for research

As an object of research, *Lemna minor* L. is chosen as one of the most promising biological agents for treatment of the circulating water of fish farms with closed water supply. The process of water treatment takes place in a bioreactor designed for the Lemnoideae cultivation (Fig. 1).

The dimensions of the phytoreactor in the plan are 1.3×0.75 m and a water depth of 0.35 m. To pump water into the phytoreactor, a centrifugal submerged pump Atman PF-1100 is used (pump head – 1.4 m, flow rate – 1.1 m<sup>3</sup>/h). Water flow regulation is carried out using a plastic ball valve for aquarium pumps installed on the supply line. The hydraulic regime is set in such way as to ensure the duration of contact of contaminated water with plants within the range of 60–90 min. Excess biomass of plants is removed manually and used as a fish feed.

The structure is supplied with the polluted water of the tilapia (*Oreochromis aureus*) growing farm after treatment on a flooded biofilter with expanded clay. Due to the ammonification processes that take place in the biofilter, the

concentration of ammonium nitrogen in water grows from 1.8–2.3 mg/dm<sup>3</sup> to 3.6–5 mg/dm<sup>3</sup>. The temperature regime is maintained at the level recommended for growing this fish species – from 25 to 27 °C, the water at the inlet to the phytoreactor has a temperature of 22–23 °C.

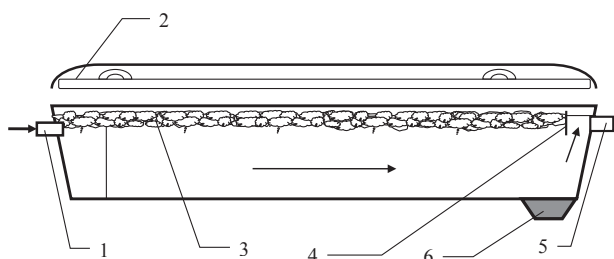


Fig. 1. Scheme of phytoreactor with floating algae: 1 – feeding pipeline with a system of uniform water distribution; 2 – lighting system; 3 – Lemnoideae layer; 4 – retaining partition; 5 – pipeline of tap cleared water removal; 6 – pit for collection and removal of sediment

The efficiency of water treatment is controlled by the main indices of CWS contamination – ammonium nitrogen and nitrates. The measurement is carried out using the ionometer I-160-MI with ion selective film electrodes ELIS-121 NH<sub>4</sub><sup>+</sup>, ELIS-121 NO<sub>3</sub><sup>-</sup>.

A characteristic feature of the phytoreactor with Lemnoideae is the need to illuminate only its surface. This makes it more efficient to use artificial light sources without loss of water reflection and dispersion in water. Selection of the necessary power and spectrum to ensure rational lighting by one or another type of lamps is carried out based on the technical characteristics of the products and the recommendations of the manufacturers. The characteristics of the lighting devices are chosen according to the passport data; the value of the specific power of the lighting system per unit area is taken in accordance with the nominal power of the studied lamps. To determine the lighting intensity of the water surface of the phytoreactor, a lux meter is used U-116.

The artificial lighting system of the phytoreactor with Lemnoideae can be constructed from fluorescent, metal halide or diode lamps. For the cultivation of macrophytes in artificial conditions, all the listed types are used. The main criterion for choosing the source of artificial lighting of the phytoreactor is the specific energy consumption per unit surface.

The most affordable fluorescent lamps are characterized by a relatively low cost, a wide range of spectra and the presence of typical cones in size. The linear length of the lamps, proportional to the power, makes it possible to effectively use to ensure uniform lighting of the water surface in the phytoreactors. To increase the lighting when using such lamps in ultrasound, it is advisable to use reflectors. Depending on the design and characteristics of the surface, the reflector can increase the illuminance of the phytoreactor surface by 20–60 %.

Metal halide lamps have is high brightness among the main advantages. This allows to penetrate a sufficiently deep layer of water (from 0.8 m to 1.5 m, depending on the lamp power and the water content of various impurities). The phytoreactor with floating algae requires lighting only of the surface, so in this case this advantage can't be realized in full. Lamps of this type are characterized by relatively high powers with relative compactness of the light source, which results in the effect of point lighting. Also, given the high heating temperature of lamps with metal halide lamps, it is

necessary to place them at a distance not less than 50 cm from the phytoreactor surface. Therefore, to provide uniform lighting of the phytoreactor by means of powerful lamps, if necessary, it is practically impossible to save energy at the same time. As a consequence, some areas of the construction site will be under intensive lighting, while others will be in shadow. An increase in the number of lamps per unit surface area in order to ensure uniform lighting entails a significant increase in current electricity consumption. Since the latter are characterized by low efficiency and limited choice on the spectrum, the expediency of using for plant lighting in phytoreactors is considered to be extremely doubtful.

LED lamps as the most promising sources of artificial lighting can be effectively used in the treatment system of circulating water. The main advantage is a high efficiency and a significant service life (up to 50 thousand hours). The quality of modern diode luminaires provides high brightness, besides, depending on the need, it is easy to increase or decrease. The only weighty factor, today significantly limits the widespread introduction of LED lamps, a relatively high cost.

#### 4. 2. Method for determining the reproductive potential and assimilation capacity of Lemnoideae by nitrogen

An evaluation of Lemnoideae reproductive potential in the conditions of a non-current aquarium is carried out according to the methodology [15]. The doubling time ( $t_d$ ) is calculated using the instantaneous population growth rate ( $r$ ).

The instantaneous population growth rate ( $r$ ) is calculated using formula (1):

$$r = \frac{\ln(N_t) - \ln(N_0)}{t}, \quad (1)$$

where  $N_0$  – the initial number of plates;  $N_t$  – the finite number of plates  $t$  – the exposure time (day).

The number of layers is determined by visual calculation on the entire surface of the aquarium. Further, the time is doubled (2):

$$t_d = \frac{\ln 2}{r} = \frac{0,6931}{r}. \quad (2)$$

The growth of Lemnoideae biomass in phytoreactors is determined by weighing the moist mass of plants at certain intervals. The obtained results allow to estimate the efficiency of the use of artificial lighting sources and to predict the intensity of removal of ammonium nitrogen by Lemnoideae.

To determine the assimilation capacity of the Lemnoideae by nitrogen, the dynamics of the growth of plant biomass during cultivation in circulating water is studied, and the chemical composition is analyzed (the content of nitrogen and phosphorus is determined). Determination of nitrogen content in the chemical composition of plants is carried out according to the procedure for determining the content of nitrogen, phosphorus and potassium from a single sample of plant material (wet ashing) in the certified laboratory of the Rivne State Agricultural Experimental Station UAAS (Ukraine).

### 5. Research results of the treatment process of circulating water in phytoreactors

Despite the fact that a wide range of abiotic factors influence the growth Lemnoideae dynamics, the most significant

are the water temperature, the intensity and duration of lighting, pH of water, the availability of macro- and micro-elements. Rational values of the artificial lighting parameters of the phytoreactor will make it possible to provide the necessary effect of treatment of circulating water in ammonium nitrogen with minimal energy expenditure. Lamps with low luminous flux and with an unbalanced spectral characteristic are not used for the studies, because the technical parameters do not allow to provide the proper level of lighting for the growth of plants in the phytoreactors. In studies of the influence of the spectrum of fluorescent lamps on the Lemnoideae growth rates (Table 1), it is found that the plants have approximately the same dynamics of biomass growth as using specialized lamps for greenhouses and aquariums (Sylvania Grolux, Osram Fluora) and lamps with improved luminous efficiency, intended for lighting of trading halls or use in premises (Sylvania Luxline Plus, Philips Master). The first type of lamps has a spectrum adapted to ensure a better vegetation period (maximums in the wavelength range of 440–450 nm and 650–670 nm). The second type of lamps is characterized by a balanced spectrum, close to natural light, visually perceived as a neutral or warm light.

Table 1

Dynamics of nitrogen compounds extraction by Lemna minor Linnaeus from sample of circulating water under various lighting conditions

Mark of lamps, marking	Technical specifications		Water treatment efficiency, %		$T_d$ , day
	power, W	luminous flux, lm	$NH_4^+$	$NO_3^-$	
Philips Master TLD 830	36	3350	97–95	92–84	2,9
Philips Master TLD 840	36	3350	97–95	94–80	2,8
Sylvania Luxline Plus Deluxe/865	36	3250	96–96	96–82	3
Sylvania Luxline Plus Deluxe/830	36	3350	95–93	90–83	2,8
Sylvania Grolux	36	2200	96–95	92–88	2,6
Osram Fluora 77	36	2250	97–95	96–82	2,8

The efficiency of removal of nitrogen compounds in the phytoreactors with Lemnoideae provides the possibility of reusing water for growing fish, since at the outlet of the structure the concentration of  $NH_4^+$  is 0.05–0.17 mg/dm<sup>3</sup> at an initial 1.8–2.4 mg/dm<sup>3</sup>, and the content of nitrates  $NO_3^-$  at the outlet of 0.8–7 mg/dm<sup>3</sup> at an initial 20–35 mg/dm<sup>3</sup>.

The luminous flux of the lamps of the Luxline Plus and Master series for types TLD 830–840 in terms of passport characteristics is 3350 lm, whereas the analogous characteristic for phytolamps varies between 2200–2250 lm. Since the growth rates of plants in both variants are approximately equal, the energy efficiency of such lamps under phytoreactor conditions can be considered the same. To provide the lower limit of the optimum range of lighting intensity in 4200 lux on each square meter of the phytoreactor surface it is necessary to place two phytolamps. The results of measuring the phytoreactor lighting by various types of lamps are given in Table 2. Therefore, to ensure rational lighting parameters of the phytoreactor, fluorescent lamps are advis-

able to have lines along the structure, the distance between the lines should be within 50 cm. For the purpose of uniform lighting of the entire surface, the fixtures must be placed at a height of 25 cm above the water. Thus, the installed power of the lighting system using fluorescent lamps will be about 60 W/m<sup>2</sup>, and when operating within 6 hours, the specific daily electricity consumption will be 0.36 kW/(m<sup>2</sup>·day). In the absence of natural insolation, the duration of the phytoreactor lighting should be maintained at a level of 12–14 hours, in which case the specific power wattage will grow to 0.72–0.84 kW/m<sup>2</sup> per day.

The lighting system using diode lamps also provide the proper intensity and meet the requirements of plants according to the spectrum of light. Using Epistar diodes rated at 10 W with a spectrum similar to TLD 830 and TLD 840 fluorescent lamps, 4–5 units should be set on each square meter of the phytoreactor area (a 10 W diode with a neutral spectrum is characterized by a light flux of up to 1000–1200 lm). Thus, the installed capacity of the diode luminaire will be 40–50 W/m<sup>2</sup>, and the daily electricity consumption, depending on the presence of natural insolation, can fluctuate within 0.24–0.6 kW/m<sup>2</sup>.

Table 2

The results of comparative studies of lamps for lighting of the phytoreactor with Lemnoideae

Lamp Type	Technical characteristics of the luminaire			light intensity, lux	Lighting time, hour/day	Biomass doubling time, day
	Number of lamps	Power, W	luminous flux, lm			
TLD	2	72	33502	5200–5450	12	2.6–3
Diode	5	50	51100	5130–5380	12	2.5–2.8

An investigation of the dependence of the increase in the Lemnoideae biomass on the duration of the lighting period (Fig. 2) is carried out in conditions of a non-current aquarium on samples of the circulating water of the farm for tilapia growing at a temperature regime of 25–26 °C. The intensity of artificial lighting is ensured on the indicated [17] level (4400 lux) with the help of fluorescent lamps with a spectrum specific for plants.

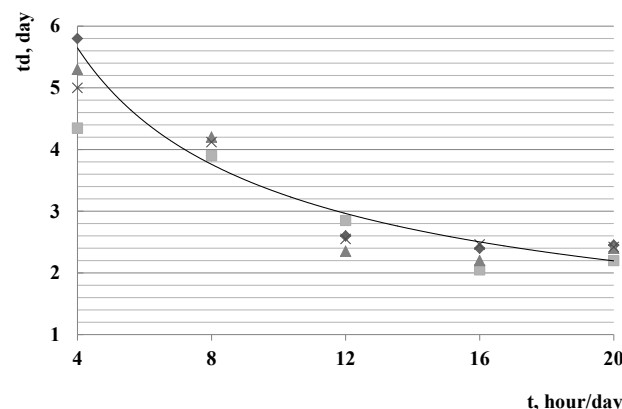


Fig. 2. Dependence of the Lemnoideae biomass doubling time ( $t_d$ ) on the duration of the light period ( $t$ )

Under ultrasonic conditions, the treatment power of the phytoreactor by nitrogen is determined, while the lighting intensity with fluorescent lamps is changed in the range 4200–6650 lux, and the lighting duration is from 6 to

16 hours per day. As confirmed by the results of the research, the treatment power of phytoreactor for nitrogen will be directly proportional to the rate of growth of plants and their specific mass per unit area. Slowing down the rate of plant growth as a result of limiting the duration of daylight hours or reducing the intensity of artificial (natural) lighting leads to a decrease in the intensity of assimilation of the basic nutrients, in particular, nitrogen.

Calculated data on the treatment power of the phytoreactor is obtained on the basis of an analysis of the chemical composition of cultivated plants and the increase in their biomass under various conditions of intensity and duration of lighting (Fig. 3). The specific Lemnoideae biomass is small in the phytoreactors, in which plants do not slow down the growth rate, can be up to 4–6 kg. Accordingly, if the Lemnoideae assimilating potential, reduced to 1 kg of wet weight, is about 2.4 g of nitrogen per day, the treatment power of the phytoreactor, depending on the amount of biomass on its surface, will be 9.6–14.4 gN/(m<sup>2</sup>·day). When recalculating to ammonium nitrogen, the maximum treatment capacity of the facility will be 12.3–18.5 g NH<sub>4</sub><sup>+</sup>/(m<sup>2</sup>·day).

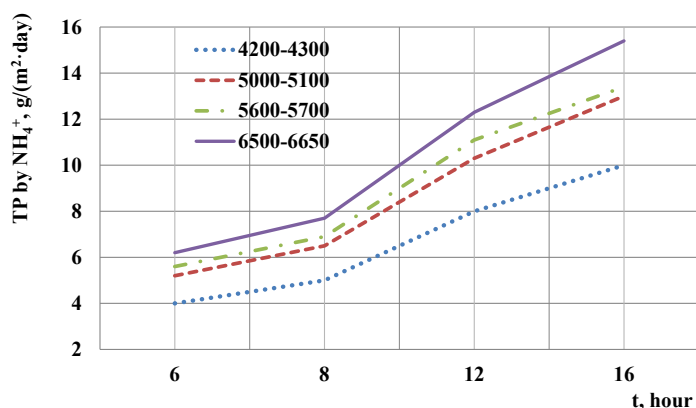


Fig. 3. Dependence of treatment power by ammonium nitrogen ( $TP$ ) on the light period duration ( $t$ ) at different light intensities, lux

Based on the obtained results, it is possible to establish such rational limits for the light period duration – 12–16 hours per day. The duration of the artificial lighting system in this case will depend on the period of natural insolation. Further artificial increase in the lighting duration is irrational, since the growth rates of plants practically do not change, while the cost of electricity will increase. The recommended range of light intensity at 6500–6700 lux when using fluorescent lamps is related to the technical characteristics (nominal luminous flux) and linear dimensions. With the use of diode lamps, the ability to effectively control the light flux makes it possible to change the lighting intensity depending on external factors (the level of natural insolation, the temperature of the circulating water, nitrogen loads on the phytoreactor).

## 6. Discussion of the research results of the treatment efficiency of circulating water depending on the lighting parameters

Determination of rational limits of lighting intensity of the phytoreactor by any source of artificial light, according to the duration of the light period, connected directly with

the optimization of the process of removal of nitrogen and phosphorus compounds in phytoreactors with Lemnoideae. Also, a controlled change in lighting intensity is an effective and rational tool for regulating the treatment power of the phytoreactor, since the technical characteristics of modern lighting systems make it possible to automate such process. A directional change in the lighting parameters of the phytoreactor can be considered as a mechanism for responding to certain changes in the ultrasound system. First, if possible to illuminate the phytoreactor due to natural insolation, the artificial lighting system allows compensating for the shortage of solar activity in winter, ensuring proper brightness on cloudy days. Secondly, the controlled change in the lighting parameters with a decrease in the nitrogen bond load on the phytoreactor can be used to maintain a stable mode of operation.

One of the main disadvantages of cultivation of Lemnoideae as treatment agents is limited capacity to increase specific biomass in phytobacteria. Maximum growth rates of Lemnoideae are observed with specific biomass in the range of 2–6 kg/m<sup>2</sup>, with an increase of this index to 8–10 kg/m<sup>2</sup>, the lower layers have a deficit of light, and the upper layers – a nutrient. Intensive mixing slows down the growth rate of Lemnoideae and leads to the death of a part of plants. Therefore, if necessary, intensify the work of the phytoreactor is possible only if grown in a polyculture with Lemnoideae of air-water plants or *Eichhornia crassipes* Solms, which will increase the biomass of plants to 10 kg of wet weight per square meter. The treatment power of the phytoreactor will increase in proportion to the specific mass of the plants per unit area and the rate of biomass growth. At the same time, this species requires more bright lighting and can't be used as a food for most fish that are cultured in CWS.

Since the removal of the bulk of the dissolved nitrogen compounds occurs when the plants perform metabolic processes, the calculation of the treatment power of the phytoreactor should be performed taking into account the specificities of the absorption of substances by macrophytes. The spectrum of compounds and elements that plants assimilate in the process of growth and reproduction is fairly wide, but the most important mineral elements in plant nutrition are elements such as carbon, oxygen, nitrogen, potassium, phosphorus, iron, magnesium and calcium. A positive feature of such properties of plant metabolism is the possibility of simultaneous removal together with ammonium nitrogen of other soluble impurities characteristic of fish farms – phosphates, potassium and sodium compounds, calcium and magnesium. At the same time, under the conditions of a phytoreactor with Lemnoideae, deficiency of any of the elements can potentially limit the removal rates of the most dangerous for fish ammonium nitrogen. Therefore, in the treatment of circulating water, it is desirable to analyze the dynamics of these elements.

## 7. Conclusions

1. The most promising in terms of energy efficiency are LED lamps, which, compared with luminescent, can reduce power consumption by 25–35%. However, fluorescent lamps will be advisable to use in the presence of natural insolation and if necessary short-term work (2–6 hours per day),

because such lamps are more affordable and characterized by low depreciation charges.

2. In the conditions of the phytoreactor for treatment of the circulating water in CWS, the Lemnoideae biomass doubling time is from 4.5 to 6 days with the lighting duration within 4 hours to 2–2.5 days with the lighting duration within 14–16 hours per day.

3. The treatment power of the phytoreactor with Lemnoideae by nitrogen when processing the circulating water in CWS is 9.6–14.4 gN/(m<sup>2</sup>·day), depending on the specific mass of plants per unit area. These indicators are provided with the organization of the duration of the light period in the range of 12–16 hours per day and with lighting intensity of 6500–6650 lux.

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