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Influence of Walls in a Container on the Growth of the *Chlorella Vulgaris Algae*

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ABSTRACT

Most of the algae are eukaryotic organisms commonly found in the aquatic environment. They are characterized by a great variety of species and the possibility of growing under various conditions. They photosynthesize, mainly needing light, water and carbon dioxide to grow. Algae can be used in various branches of the economy for the production of food, animal feed, bio-fertilizers, pigments, they can be used for sewage treatment or carbon dioxide sequestration. The aim of the work was to investigate the effect of the material from which the walls of containers are made on the bioreactors for algae cultivation. Two wall materials were used in the research: shiny aluminium foil and matte black light-absorbing paper. The content of photosynthetic pigments in algae cells, optical density, temperature and pH were examined. The tests were performed in triplicate and the standard error was calculated with the 95% confidence interval. It was observed that the glossy aluminium foil wall significantly improved the growth of the *Chlorella vulgaris* algae at the lowest light intensities by more than 4 times chlorophyll *a* compared to the sample placed in a container with walls of matte black paper. This means that the use of walls in shiny aluminium foil containers can reduce the lighting costs and contribute to an increase in the produced biomass.

Keywords: illumination, algae, *Chlorella vulgaris*, impact, aluminium foil, reflection, black paper

INTRODUCTION

Algae are eukaryotic or prokaryotic organisms. The commonly occur in the aquatic environment (salt or fresh waters) and may also be found on land (on tree bark or on soil) [Czerwik-Marcinkowska, 2019]. It is estimated that the number of algae is around 800 000 species [Suganya et al., 2016]. According to AlgaeBase, about 52 491 species of algae are already known [Guiry and Guiry, 2021]. The large number of species, and their diversity growth opportunities in various climatic zones have contributed to the colonization of ecosystems all over the globe by algae. In nature, these organisms play an important role in the circulation of elements [Czerwik-Marcinkowska, 2019]. Most of them can photosynthesize (e.g. they are autotrophs), but there are also heterotrophs, saprophages and parasites. They take various forms of cell structure and organization [Morales-Sánchez et al., 2017; Kondzior and Butarewicz, 2018; Czerwik-Marcinkowska, 2019].

Algae are more and more important to humans. They are used in food products of humans and animals, in the processes of carbon dioxide sequestration, wastewater treatment, production of bio-fertilizers, synthesis of pigments and stable biochemical isotopes. They can also become an alternative third-generation fuel source [Dogaris et al., 2015; Suganya et al., 2016]. The algae are cultivated in open or closed systems. Open systems, which are cheaper to maintain, but less controlled, are most often used for energy purposes. Closed algae growing systems are more expensive

to build and maintain [Singh and Sharma, 2012; Kutsay et al., 2020].

In order to use the potential of algae, it is necessary to create optimal conditions for their growth [Blair et al., 2014]. The growth of algae will increase the biomass that can be used in various industries. For the production of algae biomass, it is necessary to provide, among others: nitrogen and phosphorus compounds, carbon dioxide, water and light. As algae thrive, they need organic compounds which are produced through photosynthesis. For photosynthesis to proceed, the light of the correct wavelength is required. One of the light sources may be a light-emitting diode (LED). Their spectral emission range can be adjusted to the absorption range of photosynthetic pigments so that the photosynthesis process is optimal, which can lead to an increase in algae growth by adjusting the emission spectrum range to the specific needs of individual algae species [Wahidin et al., 2013; Ramanna et al., 2017]. Most species of algae increase their photosynthetic activity in the range of light exposure of 200–400 μmol E/m^{2}s. For the *Chlorella vulgaris* algae, the growing intensity range is 232 to 465 μmol E/m^{2}s in the PAR range [Dębowski et al., 2020]. The another challenge in the cultivation of autotrophic algae is to reduce the impact of the attenuation of light which passes through the dense culture of algae in the bioreactor [Salmean et al., 2019].

The use of LED lighting as a light source can contribute to reducing the energy requirement in the algae cultivation process. Reflective walls can be employed in order to use lighting and the energy emitted by lighting more efficiently. Owing to them, the light, although diffused and of lower intensity, will be delivered to the bioreactor in which the algae are grown. The shorter the distance the light travels, the more energy it will transfer to the photosynthetic system, which will be used by the algae. When illuminating the installation on one side, the use of a cultivation container with light-reflecting walls increases the area of the bioreactor that is illuminated.

The aim of the work was to determine the influence of the wall used in the construction of the container on the growth of the *Chlorella vulgaris* algae. A wall made of shiny aluminium foil and another made of matte black paper were used in the research. Bioreactors were exposed to a light of three intensities.

MATERIAL AND METHODS

Biological materials

The *Chlorella vulgaris* BA02 algae strain from the Culture Collection of Baltic Algae (CCBA) of the Institute of Oceanography, Faculty of Oceanography and Geography of the University of Gdańsk in Gdynia, al. Marszałka Piłsudskiego 46, were used in the study. The photo of *Chlorella vulgaris* BA02 taken using an optical microscope is presented in Figure 1.

Research stand and the culture medium

The research was carried out in the bioreactors made of Dreschla washers. The BG-11 Medium for Blue Green Algae (ATCC Medium 616) was used to grow the algae. The composition of the BG 11 medium is shown in table 1. The volume of culture medium was 400 ml. The algae culture was incubated at room temperature (22 ± 2°C). The culture was illuminated for 8 hours, then there was a period of 10 hours without lighting (8h light/10h dark). LED (light-emitting diode) growth panels based on 84 diodes (red to blue in a ratio of 1:3) SMD 5630 (surface-mount devices) were used for lighting. The influence of each type walls was tested at three intensity levels: 500, 250 and 50 μmol/m^{2}s. The walls of the container were made of shiny aluminium foil in order to reflect light or matte black paper to absorb light. The photos of the research setup are presented in Figure 2.

MEASUREMENT METHODS

The intensity of illumination was measured with the Delta OHM HD 2102.1 photoradiometer with the LP 471 PAR sensor. Temperature and pH were measured with a Mettler Toledo pH-meter FiveGo F2. Optical density was measured at a wavelength of 686 nm [Dziosa and Makowska, 2016]. The pigment content was measured spectrophotometrically with a Lambda Bio+ Perkin Elmeron at 0, 4 and 8 days of exposure. The tests were repeated three times and the standard error was calculated with the 95% confidence interval of the result. The test samples were taken before the lighting was turned on. In order to measure the content of photosynthetic pigments a 2 cm^3

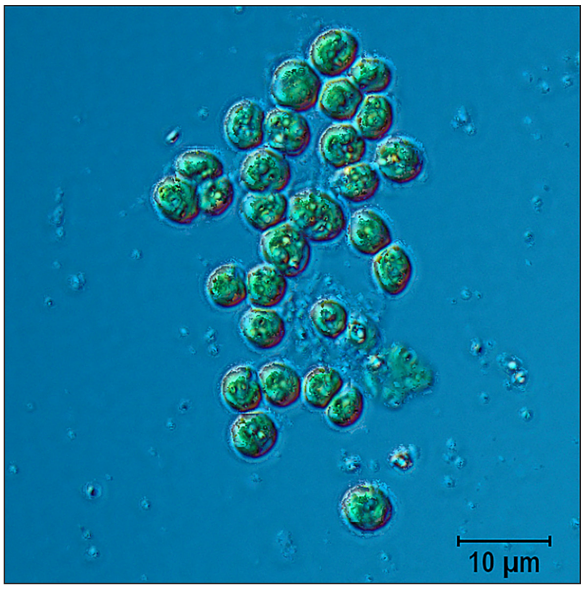

Figure 1. *Chlorella vulgaris* BA02 algae. The samples were examined under an Olympus BX61 microscope with objective UPLFLN60XOI, Differential Interference Contrast (DIC) technique was used with the function of deepened depth of field – Extended Focal Image (EFI)

culture was taken as a test sample. The test sample centrifuged at 4500 RPM for 10 minutes using the Sigma 3–16KL centrifuge. The supernatant was decanted and 2 cm^3 of 90% methanol was added to the centrifuged mass of the algae. Then, the test-tubes were closed and then mixed and placed in a thermoblock Biometra TB2 set at 60°C for 10 minutes. After this time, the sample was centrifuged again for 10 minutes at 4500 RPM and the resulting supernatant was subjected to a spectrophotometric analysis at wavelengths 470; 652 and 665 nm. The pigments content was calculated from the formulas published in Xiong et al. [2016]:

$$Chlorophyll\ a\ =\ 16.82A_{665} - 9.28A_{652}\ [\frac{mg}{\mathrm{L}}] \quad (1)$$

$$Chlorophyll\ b\ =\ 36.92A_{652} - 16.54A_{665}\ [\frac{mg}{\mathrm{L}}] \quad (2)$$

$$C_{carotenoid}\ = \frac{(1000A_{470} - 1.91C_a - 95.15C_b)}{225}\ [\frac{mg}{\mathrm{L}}] \quad (3)$$

Electricity consumption was measured on the basis of readings from the Whiteenergy H&O Electricity Consumption Meter PN: 05993 device.

Table 1. Composition of BG 11 medium used for the cultivation of *Chlorella vulgaris* BA02

Component	Value [mg·L^{-1}]
$NaNO_3$	1500
K_2HPO_4	40
$MgSO_4 \cdot 7H_2O$	75
$CaCl_2 \cdot 2H_2O$	36
$C_6H_8O_7$	6
$(NH_4)_5[Fe(C_6H_4O_7)_2]$	6
$C_{10}H_{16}N_2O_8$	1
Na_2CO_3	20
Microelements	
H_3BO_3	0.00286
$MnCl_2 \cdot 4H_2O$	0.00181
$ZnSO_4 \cdot 7H_2O$	0.000222
$NaMoO_4 \cdot 2H_2O$	0.00039
$CuSO_4 \cdot 5H_2O$	0.000079
$Co(NO_3)_2 \cdot 6H_2O$	0.0000494

RESULTS AND DISCUSSION

Characteristics of LED panels

The emitted spectrum of LED growth panels was measured. Figure 3 shows the results of the spectrum emitted by the panels, compared to the spectrum emitted by the Sun at specific wavelengths of 350 to 800 nm, previously published in Kondzior et al. [2019]. The panels emit a spectrum in the range of about 410 nm to 525 nm with a peak at 456 nm and emission from about 570 nm to 675 nm with a peak at 636 nm. The spectrum emitted by the Sun covers almost the entire studied area from 350 nm to 800 nm with very high intensity. Lysenko et al. [2021] investigated the effect of red and blue light at different intensities of 40, 130, 350 µmol/m^2s on oxygen production and the kinetics of chlorophyll fluorescence in the *Chlorella vulgaris* algae cells. Researchers found that there were large differences in the behavior of the photosynthetic system in terms of color and light intensity within one species of algae. This means that the color and intensity of lighting should be more carefully considered during research [Lysenko et al., 2021].

The content of pigments

The content of photosynthetic pigment chlorophyll *a* was measured three times, while the

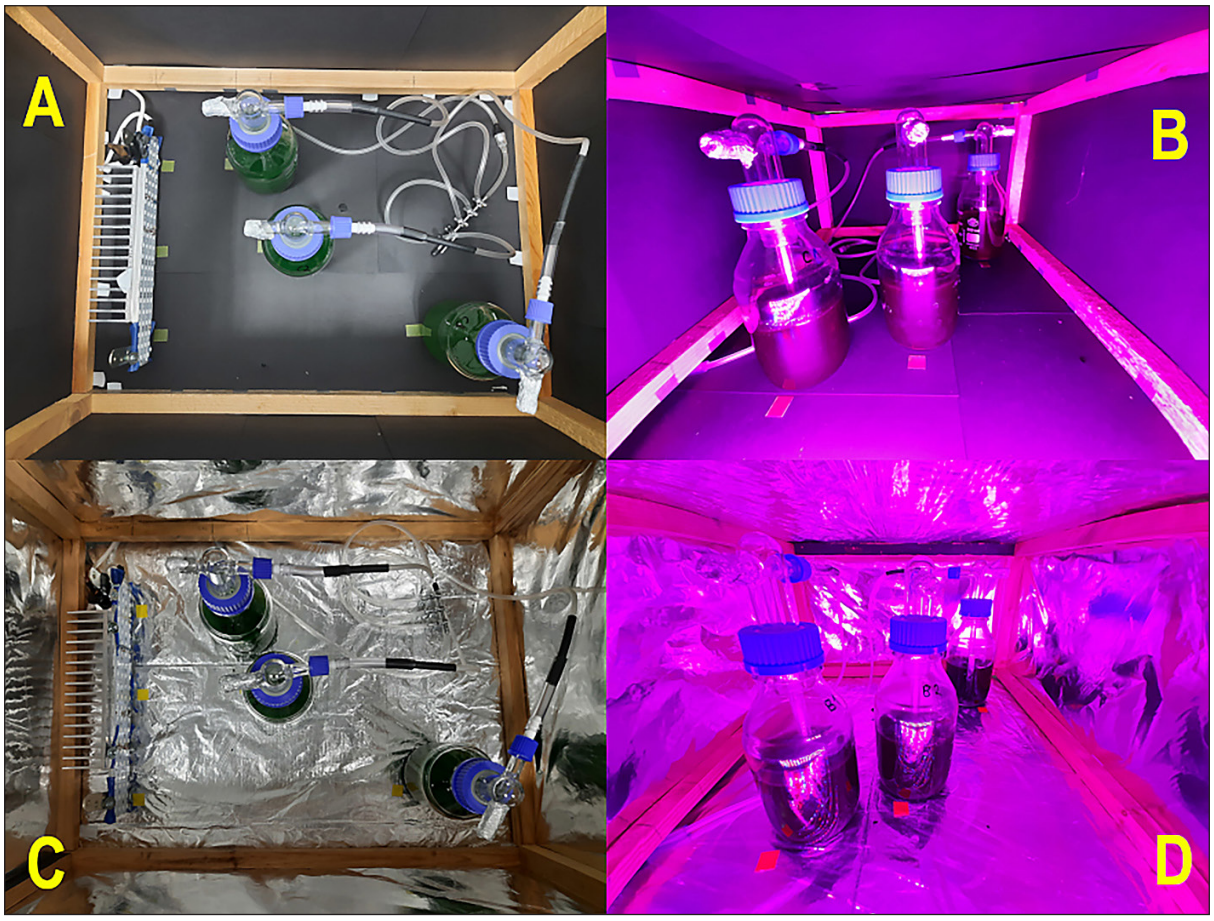

Figure 2. Research setup with containers on bioreactors for algae cultivation. Photo A and B – walls made of matte black light-absorbing paper. Photo C and D – walls made of shiny aluminium foil. Photo B and D with LED growth lighting on

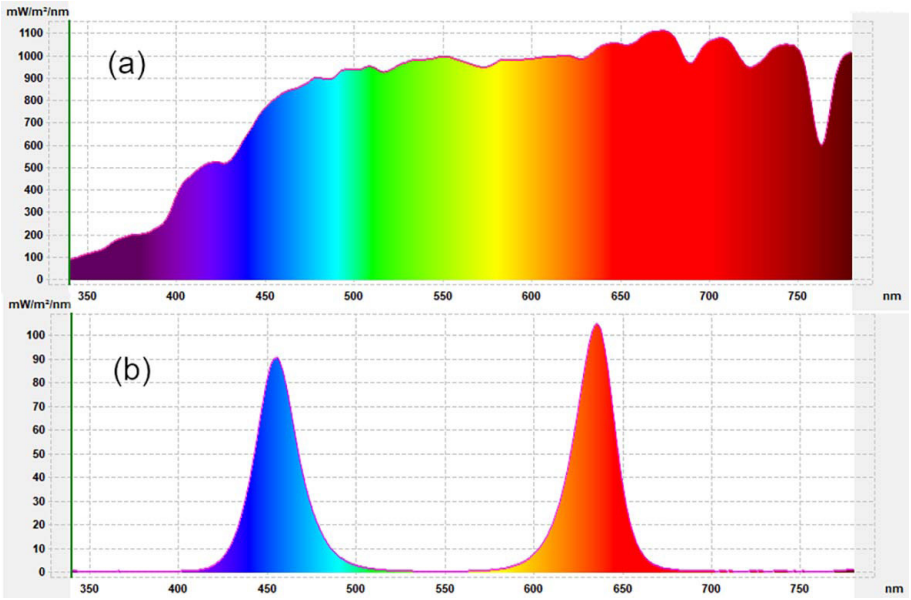

Figure 3. Spectrum emitted by the: (a) Sun (in the range from 350 to 800 nm) published in Kondzior et al. [2019], (b) LED growth [Kondzior and Butarewicz, 2021]

arithmetic mean and standard error with 95% confidence interval were calculated for individual samples. The results are presented in Figure 4. More chlorophyll *a* was determined in the culture container made of shiny aluminium foil walls than of matte black paper. The highest amount of chlorophyll a was tested in sample 2S on the 8th day of exposure to light and amounted to 29.9 mg/L. This is 51% higher pigment concentration than in the 2M parallel sample in the second container made of black paper. The smallest amount of pigment on individual days was determined in the 3M bioreactor illuminated with the intensity of 50 µmol/m^{2}s. It may be related to the insufficient amount of light supplied. The content of chlorophyll *a* in the sample illuminated with the same intensity of 50 µmol/m^{2}s, but placed in a container with walls made of shiny aluminium foil is 4 times greater on the 8th day of exposure. The introduction of a small amount of additional lighting caused by light reflection from the aluminium foil allowed for a much more efficient development of the *Chlorella vulgaris* algae. According to the Lambert-Beer law (photometric distance law), the illumination intensity decreases with the square of the distance. Thus, the light beam reflected from the aluminium foil has a longer path to the bioreactor, but it can reflect many times and the light, although with a weak intensity, can reach around the bioreactor, so that the sum of the energy of the incident light is greater than when using the walls made of light-absorbing material. The research showed that there cannot be too much light, because in the bioreactor illuminated with the intensity of 500 µmol/m^{2}s in the container with walls made of aluminium foil, the pigment content is determined by 10.2% lower than the content determined in the bioreactor illuminated with 250 µmol/m^{2}s in the same container.

Siedlewicz et al. [2020] examining the effect of oxytetracyline on algae, including *Chlorella vulgaris*, determined the content of chlorophyll *a,* in the control sample after 11 days of incubation it was 3.74 mg/L. The researchers used a low illumination intensity of 80 µmol/m^{2}s. In own research, the result was 5.8 mg/L determined in the 3M test with an intensity of 50 µmol/m^{2}s in a container with walls made of matte black paper.

Ajayan el al. [2019] conducted a similar research using the LED bio-box. It is a system consisting of LED lighting in the investigated variants of light: white, red, green and blue, and a cover into which the bioreactor is inserted. The cover or the bag is covered on the inside with a material reflecting the rays of light. The short distance from the cover to the bioreactor minimizes the loss of light energy and reduces the effect of algae obscuring during cultivation (self-shading mechanism). Ajayan el al. [2019] found that the highest biomass was obtained under red lighting, and under blue lighting, the highest lipid productivity, the content of fatty acids and total chlorophyll on the 10th day of incubation was 30.4 ± 2.2 mg/L, consisting of chlorophyll *a*, chlorophyll *b* and carotenoids. They also found that the use of a algal culture box with light reflecting walls

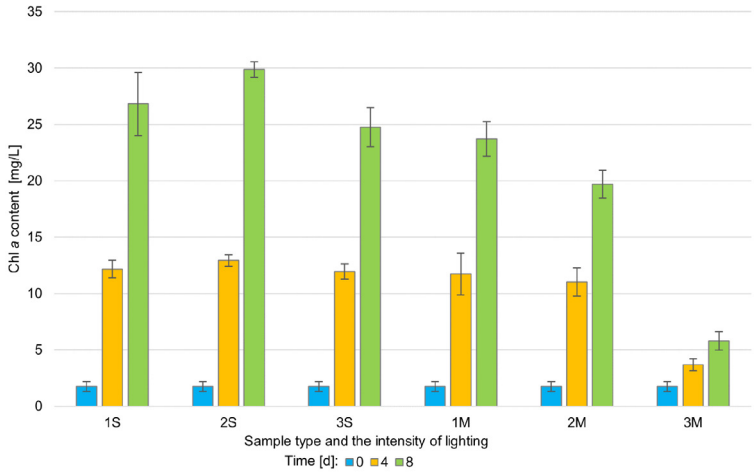

Figure 4. Content of photosynthetic pigment chlorophyll *a* in cells of the *Chlorella vulgaris* BA02 algae in the following days of exposure to light

Legend: walls made of shiny aluminium foil: 1S – 500 µmol/m^{2}s, 2S – 250 µmol/m^{2}s, 3S – 50 µmol/m^{2}s; walls made of matte black paper: 1M – 500 µmol/m^{2}s, 2M – 250 µmol/m^{2}s, 3M – 50 µmol/m^{2}s. Whiskers – standard error with 95% confidence interval [Kondzior and Butarewicz, 2021]

ensured high efficiency, even light penetration which improved the cultivation time and growth rate compared to normal cultivation conditions [Ajayan et al., 2019].

The measurement results were collected and presented in the form of a bar chart in Figure 5 which shows the content of chlorophyll *b* pigment in various tested configurations on individual days. The content of chlorophyll *b* pigment in individual samples 1S, 2S, 3S, 1M, 2M, 3M was: 13.3; 14.8; 12.0; 12.0; 9.5; 2.45 mg/L, respectively. The greatest amount of pigment was determined in the 2S sample, and the lowest in the 3M sample. The determined concentration of chlorophyll *b* was lower than the 2S sample for: 1S – by 10.1%; 3S – 19.1%; 1M – 19.1%; 2M – 35.9%; 3M – 83.4%. Yu et al. [2017] extracted chlorophyll from two algae *Chlorella sp.* and *Nannochloropsis sp.* They were grown at a temperature of 25°C in the light: dark photoperiod of 18:6h. After 14 days of incubation, they obtained the concentration of pigment for *Chlorella sp.:* chlorophyll *a* 8.45 g/L, chlorophyll *b* 4.33 g/L and for *Nannochloropsis sp*.: chlorophyll *a* 21.2 g/L, chlorophyll *b* 9.66 g/L.

Benavente-Valdés et al. [2017] conducted research on the *Chlorella vulgaris* algae in two bioreactors of own design, which were illuminated with the intensity of 100 µmol/m^{2}s for 12 hours and 12 hours without light. Researchers tested the effects of various algae breeding and nutrition conditions (autotrophic or heterotrophic way). After 8 days, incubation in the autotrophic system in the bioreactor the a flat panel airlift (FPA) reached the concentration of total chlorophyll (a+b) at the level of 17.38 mg/L, the concentration of carotenoids at the level of 1.55 mg/L and a biomass concentration of 0.69 g/L. When the nutrition conditions were heterotrophic *Chlorella vulgaris* alga in the same medium with the addition of glucose (2 g/L), much higher concentrations were achieved, amounting to total chlorophyll 43.34 mg/L, carotenoids 4.66 mg/L and biomass 1.43 g/L for the same FPA bioreactor. In own research, the cultivation was carried out in the autotrophic system. The lighting intensities used for the culture were different from those in the studies by Benavente-Valdés et al. [2017]. The intensities of 50 and 250 µmol/m^{2}s were the closest. The researchers did not mention whether or not the materials reflected lighting. However, in the sample 2S (illuminated with 250 µmol/m^{2}s in a container with walls made of shiny aluminum foil), achieved the highest content of total chlorophyll (a+b) 44.64 mg/L and carotenoids 6.3 mg/L, which was similar to that achieved in the studies of Benavente-Valdés et al. [2017] in a heterotrophic system.

In order to improve light transmission through algae cultures (self-shading mechanism limitations), Huang el al. [2016] proposed collection of preliminary harvesting partial microalgae *Chlorella vulgaris* cells. They tested light intensities of 100, 160 and 220 µmol/m^{2}s. They found that they achieved a 46% increase in biomass production with a 30% preliminary harvest daily of algae cells at illumination of 160 µmol/m^{2}s. Researchers estimated that the average light intensity in the

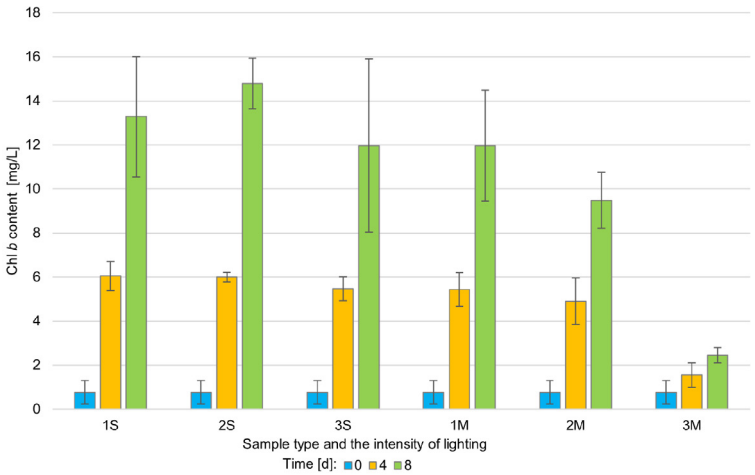

Figure 5. Content of photosynthetic pigment chlorophyll *b* in cells of the *Chlorella vulgaris* BA02 algae in the following days of exposure to light. Legend: walls made of shiny aluminium foil: 1S – 500 µmol/m^{2}s, 2S – 250 µmol/m^{2}s, 3S – 50 µmol/m^{2}s; walls made of matte black paper: 1M – 500 µmol/m^{2}s, 2M – 250 µmol/m^{2}s, 3M – 50 µmol/m^{2}s. Whiskers – standard error with 95% confidence interval

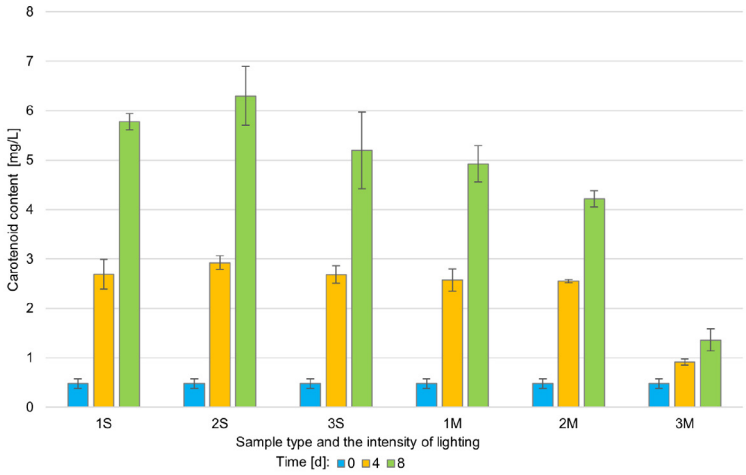

Figure 6. Content of photosynthetic pigment carotenoid in cells of the *Chlorella vulgaris* BA02 algae in the following days of exposure to light

Legend: walls made of shiny aluminium foil: 1S – 500 µmol/m²s, 2S – 250 µmol/m²s, 3S – 50 µmol/m²s; walls made of matte black paper: 1M – 500 µmol/m²s, 2M – 250 µmol/m²s, 3M – 50 µmol/m²s. Whiskers – standard error with 95% confidence interval

bioreactor increased by 27–122%. Huang el al. [2016] determined the total chlorophyll content in the described configuration to be about 58 mg/L already on the 3rd day of incubation. The chlorophyll content was maintained at a similar level for the following days until the end of the experiment on day 7.

The content of the identified concentrations of carotenoids in the samples is shown in Figure 6. The highest content of carotenoids was determined in sample 2S, and the lowest in sample 3M. In the 1M bioreactor illuminated with the highest tested intensity of 500 µmol/m²s in a container with walls made of black paper, a slightly lower content of carotenoids was determined amounting to 4.93 mg/L than in the 3S sample, amounting to 5.2 mg/L illuminated with the lowest tested intensity of 50 µmol/m²s in a container with walls made of aluminium foil.

The ratio of the content of chlorophyll *a* to *b* is shown in Figure 7. The highest ratio of chlorophyll *a* to *b* was observed in the 3M sample and amounted to 2.37, the smallest ratio was recorded in the 1M sample and amounted to 1.99. The differences between the individual results are not large. However, it can be concluded that the higher the light intensity, the lower the ratio of chlorophyll *a* to *b* in the individual containers. This can

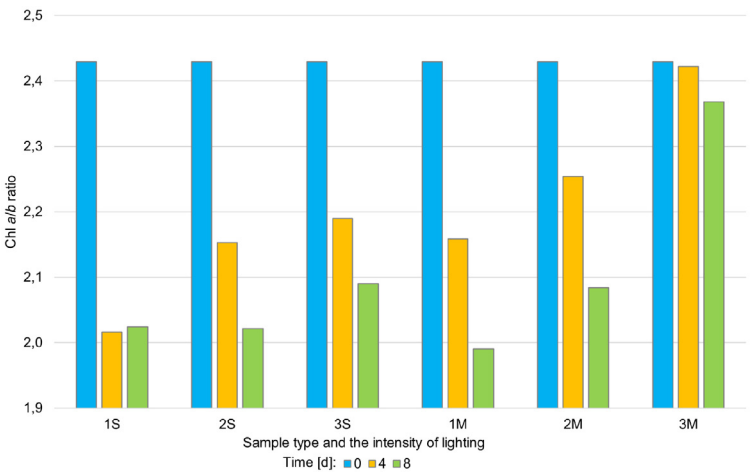

Figure 7. Chlorophyll pigment ratio *a*/*b* in cells of the *Chlorella vulgaris* BA02 algae in the following days of exposure to light. Legend: walls made of shiny aluminium foil: 1S – 500 µmol/m²s, 2S – 250 µmol/m²s, 3S – 50 µmol/m²s; walls made of matte black paper: 1M – 500 µmol/m²s, 2M – 250 µmol/m²s, 3M – 50 µmol/m²s.

be especially seen on day 4 of light exposure, on day 8, sample 1S has a similar ratio of 2.024 to sample 2S of 2.021. It was also noticed, in addition to the 1S sample, the chlorophyll *a* to *b* ratio decreased in the following days of exposure.

Optical density

The optical density was measured on days 0, 4 and 8. The highest absorbance of 3.48 was measured for the sample placed in a container with walls made of shiny aluminium foil with the code 2S after 8 days of incubation at an intensity of 250 µmol/m^{2}s. This result is 8% higher than the sample illuminated with the intensity at 500 µmol/m^{2}s and 27.2% higher than the sample illuminated at 50 µmol/m^{2}s in the same container. In the second container with walls made of matte black paper, the samples were smaller than the samples of the same intensity in the container with walls made of shiny aluminum foil by 14.2% for 1M; 33.2% for 2M and 74.6% for 3M. The optical density is shown in Figure 8.

Galler et al. [2018] tested four species of algae in three intensities of 50, 100, 300 µmol/m^{2}s and at four temperatures of 5, 10, 15, 20°C . The researchers determined the optical density and then converted it into the growth rate expressed in mg/L/day. During the incubation of the algae at a temperature of 20°C, the determined growth rate of the culture did not increase linearly with the increase of the intensity of lighting. For *Scenedesmus bijuga* and *Chlorella sorokiniana* species, the highest growth rate was achieved for the samples illuminated with 300 µmol/m^{2}s, then for 50 µmol/m^{2}s, and the lowest growth rate was recorded for 100 µmol/m^{2}s. The next two species, *Chlamydomonas yellowstonensis* and *Chlamydomonas augustae*, at a temperature of 20°C, the highest determined growth rate of algae was recorded for the intensity of 50 µmol/m^{2}s.

The pH of the culture medium

The measured pH values for the culture medium is shown in Figure 9 and was between 7.85 and 8.88. It was observed that, apart from the sample 1S, the pH acted similarly in the first cultivation period and on day 4 of incubation it decreased and then increased on 8 day.

Energy consumption

Electricity consumption of the installation, which includes individual elements: control device, aerator, and lighting. The consumption results are presented in Table 2. The energy consumption of the aerator and the control device are not shown in the tables due to the low energy consumption. Table 3 shows the costs related to electricity consumption. The calculations assume the average electricity price announced by the Energy Regulatory Office in information No. 25/2021 in Q1 2021, the average electricity price was 237.27 PLN/MWh [Energy Regulatory Office, 2021]. The formulas were used for the calculations:

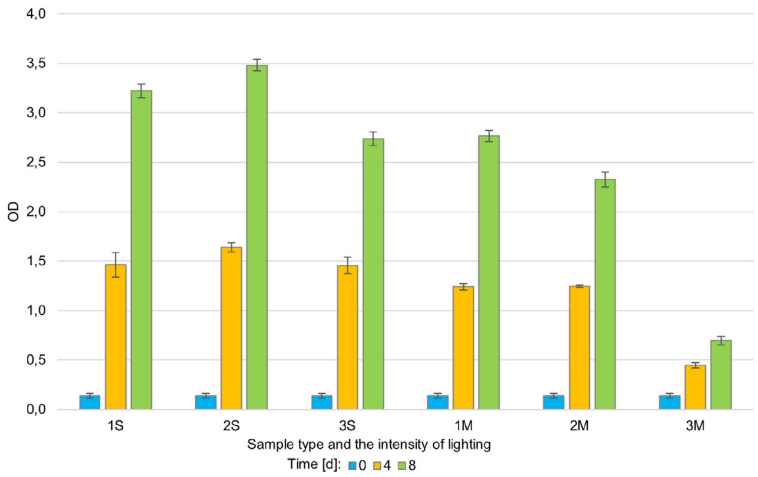

Figure 8. Optical density of the *Chlorella vulgaris* BA02 algae in the following days of exposure to light. Legend: walls made of shiny aluminium foil: 1S – 500 µmol/m^{2}s, 2S – 250 µmol/m^{2}s, 3S – 50 µmol/m^{2}s; walls made of matte black paper: 1M – 500 µmol/m^{2}s, 2M – 250 µmol/m^{2}s, 3M – 50 µmol/m^{2}s. Whiskers – standard error with 95% confidence interval [Kondzior and Butarewicz, 2021]

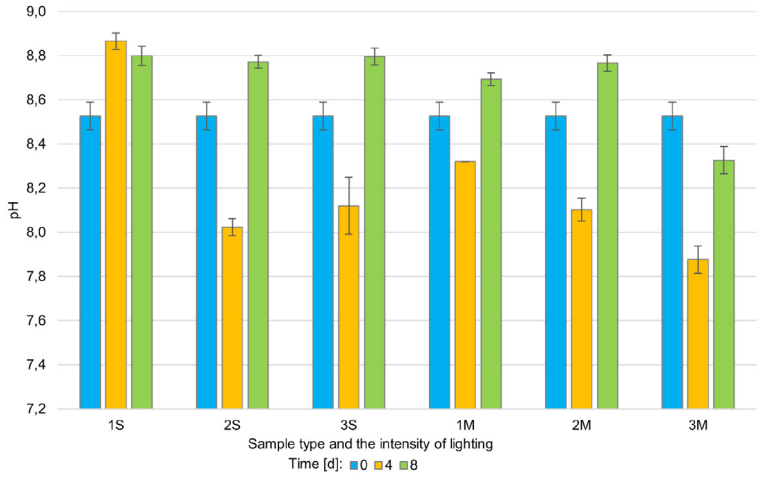

Figure 9. The pH of the BG 11 medium for the cultivation of the *Chlorella vulgaris* BA02 algae in the following days of exposure to light

Legend: walls made of shiny aluminium foil: 1S – 500 µmol/m²s, 2S – 250 µmol/m²s, 3S – 50 µmol/m²s; walls made of matte black paper: 1M – 500 µmol/m²s, 2M – 250 µmol/m²s, 3M – 50 µmol/m²s. Whiskers – standard error with 95% confidence interval

$$Cost_{per\ container}\ [\mathrm{PLN}] = \\ = \frac{Power\ [W]}{1000} \times time[d] \times 24 \times \\ \times \frac{Average\ energy\ cost\ [\frac{\mathrm{PLN}}{\mathrm{MWh}}]}{1000} \quad (4)$$

$$Cost_{per\ sample}\ [\frac{PLN}{g/L}] = \\ = \frac{Cost_{per\ container}}{s} \times \\ \times \frac{1000}{(Chl\ a\ content_{\,n}[\frac{mg}{L}] - Chl\ a\ content_i[\frac{mg}{L}])} \quad (5)$$

where: s – the number of samples in the container,
$Chl\ a\ content_{\,n}$ – chlorophyll *a* content in the next day of exposure to light,
$Chl\ a\ content_{\,i}$ – chlorophyll *a* content in the starting inoculum

In order to evaluate the cost savings related to electricity consumption, the cost of energy consumption for the production of 1 g of chlorophyll *a* was calculated for individual samples. The results of the calculations are presented in Table 4. The obtained estimates showed that the most economical solution is to use the walls made of shiny aluminum foil in the container. At an intensity of 50 µmol/m²s, an almost 6 times more economical solution was obtained. With the use of lighting 250 µmol/m²s the estimated saving was 39%, with 500 µmol/m²s 16%.

CONCLUSIONS

It was found that the use of an algae culture container with walls made of shiny aluminum foil resulted in an increase in the pigments and the optical density of the algae culture. The chlorophyll *a* in sample 3S, compared to 3M illuminated with the intensity of 50 µmol/m²s, increased by about 4 times. It has been estimated that for the production of 1 gram of chlorophyll *a* for the sample illuminated with the intensity of 50 µmol/m²s, it is possible to achieve almost six times lower cost of energy consumption for lighting and aerating the culture using a container with walls made of shiny aluminum foil.

Table 2. Energy consumption by installation and light

Specification	Shiny aluminium foil		Matte black paper	
	Power [W]	Ampere [A]	Power [W]	Ampere [A]
Entire installation	40.8	0.302	42.6	0.331
Only light	38.4	0.295	41.5	0.331

Table 3. Cost related to the cultivation process in individual algae containers for eight days of incubation [in PLN]

Type of walls in the container	Cost
Unit	[PLN]
Shiny aluminium foil	1.86
Matte black paper	1.94

Table 4. Costs related to the supply of energy for lighting and aeration of the installation producing 1 g Chl *a* for individual configurations for eight days of incubation [in PLN/g/L] (the initial value of inoculum of algae was subtracted)

Samples	Costs
Unit	[PLN/g/L]
1S	24.73
2S	22.06
3S	26.94
1M	29.46
2M	36.05
3M	160.60

Note: walls made of shiny aluminium foil: 1S – 500 µmol/m^{2}s, 2S – 250 µmol/m^{2}s, 3S – 50 µmol/m^{2}s; walls made of matte black paper: 1M – 500 µmol/m^{2}s, 2M – 250 µmol/m^{2}s, 3M – 50 µmol/m^{2}s.

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