The influence of diverse optical wavelengths on growth rate of Chlorella Vulgaris microalgae

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Abstract. The effect of different optical wavelengths on growth and development of microalgae had been examined repeatedly. Many articles had been published over the subject, although the results of these investigations were frequently controversial. Depending on the number of days spent on cultivation process, yield gain could be both significant and barely noticeable. Also, irradiation intensity plays vital part in microalgae functioning.

1. Introduction
Microalgae grow and develop in the process of photosynthesis, like other plants. In other words, they assimilate inorganic carbon to convert it into organic constituents. Stimulation of this reaction occurs with the help of light, as a source of energy. Based on this information, the spectral composition and energy component of light should be taken into account when cultivating microalgae. It was revealed that microalgae consume optical radiation in the range from 400 to 700 nm for the photosynthesis process. The wavelengths of light absorbed by microalgae cells differ in groups. Green microalgae, for example, is fed with light energy for photosynthesis, using chlorophylls as the main pigments that absorb light energy in the ranges 450-475 nm and 630-675 nm, and carotenoids as secondary ones - absorption occurs only in the wavelength range 400-550 nm [1]. Various research mentions the yield gain of microalgae under different wavelengths conditions. Red (600-700 nm) and blue (400-500 nm) light stimulate microalgae development, when the rate of growth and lipid content vary due to diverse light intensity [1-3].

2. Materials and methods
In their work Blair M. F. et al had proven that adequately selected optical wavelengths affect remarkably yield gain of microalgae. Red and green lights did not cause any positive effect on growth rate in comparison to wavelengths from white and blue bands (figure 1). Compact fluorescent lamps with 60 W power level (276 µmol/m²s) were applied for suspension irradiating. To avoid thermal effect, light sources were located 20 cm apart from the bioreactors.

Figure 1 b illustrates the highest growth rate under white, blue and red lights on the 3rd day of cultivation: 0.369 d⁻¹, 0.235 d⁻¹ and 0.140 d⁻¹ respectively. Green lighting has the highest growth rate on
the 2nd day: 0.137 d\(^{-1}\). Green-band photons have 20% more energy than red photons (680 nm) and 15.5% less than blue ones (470 nm). Therefore, green light could have decelerating effect, caused by power of provided energy. However, it is obvious, that blue photon has more energy than green. It is depicted in the figure 1 c that the highest biomass productivity was 0.038 g/L\(-\)d, 0.0199 g/L\(-\)d and 0.0096 g/L\(-\)d for white, blue and red lights on the 3rd day respectively. For green-light exposure this criterion was far less – 0.0088 g/L\(-\)d on the 2nd day.

It appeared that 7-day results shouldn’t be represented as complete growth cycle of microalgae, apart from red and green lights, which sequentially demonstrate lower growth rates and mismatch practical appliance as provided by the results of the experiment. Another set of experiments was conducted with the use of blue and white lights for 12 days. Figure 1 d depicts that growth rate of microalgae under blue light conditions surpassed growth rate with white lighting (after 10 days). This confirms the intensification of microalgae Chlorella growth rate under blue light after the adaptation period [1].

In that investigation red light did not demonstrate high growth rate. This tendency can be explained with the help of previous research group that claimed that red light could actually cause damage in cells [3, 4]. Moreover, it is noteworthy, that specific ability of optical wavelengths absorption depends on the microalgae species. During the experiment, Yan et al had detected that white and red lights performed with far higher yield gain of microalgae, although the study was conducted between 6th and 10th days and the cultivation medium was synthetic high-tension wastewater [2]. In addition, growth properties were affected by light intensity [5].

Figure 1 d shows the approximate linearity of growth rate under blue-light irradiation (\(R^2 = 0.916\)), in contrast to white-light irradiation (\(R^2 = 0.8764\)), or highlights the trend towards exponential phase reaching. Cultivation under blue-light irradiation demanded serious time input for exponential phase achieving. In this case, slow growth rate under blue light does not satisfy the requirements for practical
use, because the more rapid is the exponential phase reaching, the more efficient would be biomass production and harvesting time would be less [1].

In another research by Barghbani R. et al more yielding microalgae cultivation was obtained under blue and yellow lights. The results from the study above match with the conclusions of other scientists. Throughout the actual testing the irradiance was continuous, the light sources were luminescent lamps of different colors and power level was 23 W (figure 2).

Four diverse irradiation colors were used in this examination (white, yellow, blue and red) (figure 2). Microalgae demonstrated peak growth rate under yellow-light impact (low energy). Other colors (blue, white and red) led to slightly less biomass productivity [6].

In the following research by Kim D. G. et al, continuous irradiation was introduced at the light intensity level of 100 µmol/m²s under monochromatic blue (λ_max = 430-465 nm) and red (λ_max = 630-665 nm) lights. It was illustrated that particular wavelength could dramatically affect the growth rate of microalgae (figure 3).

Cells number depended remarkably on wavelength of light due to the fact that Chlorella microalgae cumulated the highest and the lowest cells density under red and blue light relatively. Cells density of Chlorella grown under red light was 1.5 times higher than density of cells, irradiated by blue light during
post-inoculation time period (figure 3). These results correlate with the conclusion of scientists, who conducted similar experiments with other microalgae species. Red-band wavelengths appeared the most optimal for effective Spirulina platensis growth as well as for Chlorella pyrenoidosa [8]. Pigmentation enhancement of chlorophyll was initiated by red light, what could be referred to the positive effect of this exact irradiation [9]. Yet white-light irradiation of suspension led to average cells number increase, while blue light gradually was dwindling cells gain. One of the possible assumptions is that an excess of light wavelengths from the blue spectrum can inhibit the division of microalgae cells, which leads to a decrease in the growth of microalgae biomass.

3. Results
It is indicative that cytomorphology of sizes was significantly modified as a function of light wavelength. Red light multiplied the number of cell divisions, causing small-sized cells appearance, and at the same time blue light provided size development of cells. On that basis, it could be assumed that microalgae cells generation with larger dimensions at the initiation of cultivation could encourage further cell fission at advanced development stage, and biomass productiveness raise consequently [7].

During microalgae stock culture transfer into photobioreactor adaptation to ambient conditions occurs at first, and cells growth prediction becomes insufficient. Also, to obtain valid results, microalgae should be cultivated before cells growth rate reaches stationary phase. Under different wavelengths and irradiation level conditions stationary response time varies for all samples.

4. Conclusion
Nowadays, microalgae are widely applied in diverse branches of human activity such as pharmaceutics, wastewater treatment, agriculture, biodiesel fuel production et al. To achieve accelerated growth of microalgae, vast number of parameters should be taken into consideration, particularly, optimum spectral range selection. Artificial lighting sources have advantage over natural – the possibility to control spectral structure which enables choosing the most efficient light conditions while cultivating. In this case, there is a possibility to obtain high rates of valuable substances, which are included into Chlorella, biosynthesis, and enhance reproduction and yield gain of the culture. Cultivation time of microalgae could be noticeably reduced, if chosen spectral structure of irradiation sources would correspond to absorption spectrum of Chlorella.

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