Bioresource Technology 203 (2016) 357–363

Contents lists available at ScienceDirect

Bioresource Technology

journal homepage: www.elsevier.com/locate/biortech

Review

Flashing light in microalgae biotechnology

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HIGHLIGHTS

- Flashing light effect on microalgae is comprehensively reviewed.
- Recent studies in flashing light use in microalgae biotechnology are discussed.
- Novel bioreactors that create flashing light effect are introduced.
- Flashing light is still not applied on large scale in microalgae biotechnology.
- Future aspects and suggestions related to flashing light are outlined.

ARTICLE INFO

Article history:
Received 15 October 2015
Received in revised form 17 December 2015
Accepted 19 December 2015
Available online 23 December 2015

Keywords:
Flashing light
Microalgae biotechnology
Energy efficiency
Biomass
PUFA

ABSTRACT

Flashing light can enhance photosynthesis and improve the quality and quantity of microalgal biomass, as it can increase the products of interest by magnitudes. Therefore, the integration of flashing light effect into microalgal cultivation systems should be considered. However, microalgae require a balanced mix of the light/dark cycle for higher growth rates, and respond to light intensity differently according to the pigments acquired or lost during the growth. This review highlights recently published results on flashing light effect on microalgae and its applications in biotechnology, as well as the recently developed bioreactors designed to fulfill this effect. It also discusses how this knowledge can be applied in selecting the optimal light frequencies and intensities with specific technical properties for increasing biomass production and/or the yield of the chemicals of interest by microalgae belonging to different genera.

1. Introduction

Microalgae have been used as food, feed supplements, high-value chemicals, and in cosmetics, and are considered to be a promising feedstock for biofuel production. However, the success of any agricultural or algae-based industrial product is dependent on two key factors; high biomass productivity and quality (Georgianna and Mayfield, 2012) with a low production cost. Photoautotrophic and mixotrophic microalgae primarily require CO_2 and a light source to carry out photosynthesis (Chen et al., 2011). Sunlight is the most cost-effective energy source for microalgal production but the exploitation of sunlight as a light source has drawbacks, including changes in weather, day and night cycles, and seasonal changes, which affect light intensity and its spectrum. In case of the closed photobioreactors the high costs and complexities of the cooling systems increase energy inputs during the cultivation process. Moreover, the light intensity at which the culture growth becomes dense upon time is an important factor in determining light utilization efficiency. Therefore, optimization of the light supply remains a critical issue in microalgae biotechnology due to low photosynthetic quantum efficiency (Raven, 2011).

Alternatively, artificial illumination can be produced from renewable energy sources and is economically feasible (wind, running water, and the excess energy from power plants, but sunlight should be the preferred energy source as it costs least). For biomass used for fine products such as nutraceuticals, carotenoids, and polyunsaturated fatty acids (PUFA), artificial illumination provides better regulation of the photosynthetic photon flux density (PPFD), photoperiod, and light spectra. These illumination conditions can result in enhanced photosynthesis and thus higher biomasses and valuable-content productivities (Abu-Ghosh et al., 2015a; Heining and Buchholz, 2015; Schulze et al., 2014; Solovchenko and Chekanov, 2014). Artificial illumination, thus, can make the bioreactor design simpler.

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http://dx.doi.org/10.1016/j.biortech.2015.12.057
0960-8524/

Productive algal cultures are characterized by tremendous light attenuation along the light path (Kim et al., 2006). In other words, light impinging on the surface of algal cultures is almost completely absorbed along the light path, resulting in a light gradient and, possibly, a dark zone that limits photosynthesis. The length of the dark zone depends on many factors: algal concentration, algal pigmentation, light path, and light intensity at the surface. Light energy should be delivered evenly over the algal culture and with an adequate amount of PPFD to enable the photons to reach the cells. Excessive intensity might lead to photooxidation and photoinhibition (Raven, 2011) (see Glossary for terminology), while low light levels are growth-limiting (Loera-Quezada et al., 2011). As microalgal cells travel – by the active mixing of the culture – between the saturating light at the surface of the pond or the transparent wall of the reactor, and the complete-dark depth of the culture, they experience a flashing-light regime. Fig. 1 illustrates the movement of algal cells between the saturating light zone at the surface to almost a complete dark in depth, resembling the flashing light effect, which leads to an optimal integrated light dose for photosynthesis and growth.

Flashing light has been experimentally proved to be one of the most promising light regimes in microalgal cultivation, however; its use in microalgae biotechnology to obtain high-value biochemical traits has been poorly reviewed. This review is the first report in the literature that provides an overview of the application of various flashing lights to microalgal production, giving a theoretical background of the flashing light effects, and discusses possibilities to improve algal-facility productivity by its applications.

2. Flashing light effect

It was first reported that when algal cells were illuminated by a succession of very short flashes, the maximum carbon dioxide uptake and oxygen production rates under these light conditions could be the same as those under continuous light (Emerson and Arnold, 1932). This means energy saving, and even enhanced algal productivity under optimal flashing light parameters. Moreover, the use of flashing light in microalgae biotechnology can have other advantage over continuous light, e.g. according to the existence of the light-off cycle, the cooling period is shorter and that will, in turn, reduce the electrical energy consumption, eventually reducing costs significantly and making the process easier.

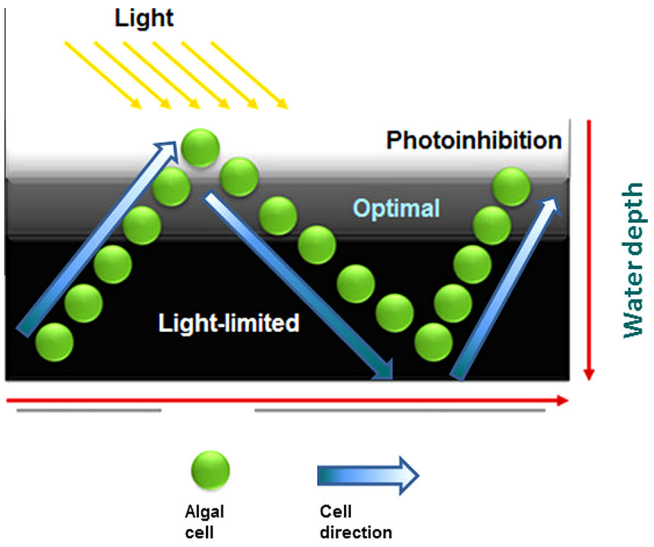

Fig. 1. Schematic diagram of the flashing light effects on an algal cell as a result of mixing in the algal cultivation systems. By this, algal cells receive an integrated light intensity equals to the optimal one.

The properties of the flashing light determined mainly on the light source and its intensity, spectral distribution and beam geometry of the light rays (light propagation in terms of rays), culture density, reactor/pond architecture and depth, and the hydrodynamics of mixing (Baroukh et al., 2015; Iluz et al., 2012). The three main parameters characterizing the flashing cycles are light intensity, light frequency, and duty cycles or light/dark (L/D) cycles that is the fraction of time in which the algae spend in the light (Fig. 2). These parameters define the range of light intensities to which the cells are exposed, as well as their frequencies. Therefore, the kinetics of mixing cycles varies greatly and changes between a millisecond time-scale in algal reactors to longer times by several orders (Iluz et al., 2012). It has been suggested that the alternation of L/D periods is beneficial to photosynthetic efficiency (Grobbelaar, 2010; Vejrazka et al., 2013).

During the early growth stages in the outdoor photobioreactor, shades and/or cooling systems are needed to protect the microalgal cultures from the high PPFD of the sun in order to prevent photoinhibition due to the continuous emission of photons over time. In contrast, under flashing light of the same high photon dose, algae can utilize light energy more efficiently without undergoing moments of photoinhibition in which the chlorophyll is both excited and relaxed in the L/D cycle (Abu-Ghosh et al., 2015b; Béchet et al., 2013), i.e. better matching photon input-rate to the limiting steps of photosynthesis.

The enhancement of photosynthesis under flashing light could be as a result of a relatively enhanced dark reactions' rate, i.e. efficient transfer electron rate (ETR) between photosystem I and Photosystem II, than under continuous illumination. This hypothesis is based on the fact that under full sunlight the limiting factor of the oxygenic photosynthesis is ETR which is about ten times slower than the rate of light-capture by chlorophyll (Kok, 1973).

Falkowski and Raven (1997) suggested two possible physiological mechanisms for the flashing light effect in photosynthetic organisms: (1) enhanced dark respiration rates following a period of photosynthesis in light, a mechanism know as enhanced post-illumination respiration (EPIR). This suggestion based on that respiration increases with increasing light intensity, or with the length of the illumination period. Under flashing light, especially with high frequencies, the respiration rate is hypothesized to be much lower for the light signals, thus, less energy consumption that leads to an enhanced photosynthesis. (2) The disequilibrium occurs between the photosynthetic electron transport and the Calvin cycle. This suggestion follows the logic that the Calvin cycle is the bottle neck of the photosynthesis final product under continuous light. Therefore, under high flashing light-frequencies, the amount of ATP generated in the electron transfer chain is sufficient to support the, comparably, slow Calvin cycle under these conditions throughout the dark phase and allows for its continuous operation.

Iluz et al. (2012) reported that the enhancement of photosynthesis under flashing light could be due to less photodynamic damage to the 32 kD protein of photosystem II (PSII) than under continuous light. According to them, under appropriate frequencies photoinhibition is reduced since light exposure is too short to cause damage or that the periodic dark intervals facilitate the *de novo* repair of the damage.

Recently, Abu-Ghosh et al. (2015b) showed that microalgal cells acclimated to flashing light pronounce less activity of the photoprotective mechanism, the xanthophyll cycle, when exposed to high light, and thus less energy loss by thermal dissipation than continuous-light acclimated cells, since the photoprotective and photoinhibition mechanisms work on similar time scales. However, the study revealed that there were no significant differences in chlorophyll content. In other words, the cells successively photoacclimate to the average light intensity of the flashing light.