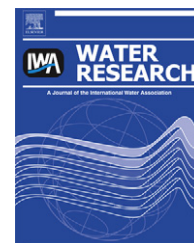


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Review

Heterotrophic cultures of microalgae: Metabolism and potential products

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ABSTRACT

This review analyzes the current state of a specific niche of microalgae cultivation; heterotrophic growth in the dark supported by a carbon source replacing the traditional support of light energy. This unique ability of essentially photosynthetic microorganisms is shared by several species of microalgae. Where possible, heterotrophic growth overcomes major limitations of producing useful products from microalgae: dependency on light which significantly complicates the process, increase costs, and reduced production of potentially useful products. As a general rule, and in most cases, heterotrophic cultivation is far cheaper, simpler to construct facilities, and easier than autotrophic cultivation to maintain on a large scale. This capacity allows expansion of useful applications from diverse species that is now very limited as a result of elevated costs of autotrophy; consequently, exploitation of microalgae is restricted to small volume of high-value products. Heterotrophic cultivation may allow large volume applications such as wastewater treatment combined, or separated, with production of biofuels. In this review, we present a general perspective of the field, describing the specific cellular metabolisms involved and the best-known examples from the literature and analyze the prospect of potential products from heterotrophic cultures.

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1. Introduction

Large-scale microalgal production has been studied for decades (Becker, 1994; Lee, 2001), given the wide variety of practical and potential metabolic products, such as food supplements, lipids, enzymes, biomass, polymers, toxins, pigments, tertiary wastewater treatment, and “green energy” products that can be obtained. These products were achieved by cultivating the microalgae on diverse mineral media, organic substrates, and synthetic or real wastewaters (Pulz, 2001; de-Bashan et al., 2002, 2004; Pulz and Gross, 2004; Lebeau and Robert, 2006; Harun et al., 2010).

Today, the most common procedure for cultivation of microalgae is autotrophic growth. Because all microalgae are photosynthetic, and many microalgae are especially efficient solar energy converters, microalgae are cultivated in illuminated environments naturally or artificially. Under autotrophic cultivation, the cells harvest light energy and use CO₂ as a carbon source. The introduction of sufficient natural or artificial light to allow massive growth and dense populations is the main objective and a limiting factor of the cultivation: the more light, up to a limit for the species, the better (Mandalam and Palsson, 1998; Yang et al., 2000; Suh and Lee, 2003). Therefore, as practiced with other microbial communities producing economic products, open ponds that mimic natural environments of microalgae are the most common option for mass cultivation (Oswald, 1992; Tredici, 2004).

Large open outdoor pond cultivation for mass algal production of single-cell protein, health food, and β-carotene (Borowitzka and Borowitzka, 1989; Wen and Chen, 2003; Carvalho et al., 2006; Chisti, 2007) is one of the oldest industrial systems since the 1950s (Oswald, 1992). Large open ponds can be built of glass, plastic, concrete, bricks, or compacted earth in a variety of shapes and sizes. The most common is the “raceway pond”, an oval form resembling a car-racing circuit (Lee, 2001; Pulz, 2001; Chisti, 2007). These cultivation systems present relatively low construction and operating costs and the large ones can be constructed on degraded and nonagricultural lands that avoid use of high-value lands and crop producing areas (Chen, 1996; Tredici, 2004).

All these benefits notwithstanding, open ponds have several inherent disadvantages: (1) Poor light diffusion inside

the pond, decreasing with depth. It is aggravated when cultivation is intensive and causes self-shading. Consequently, shallow depth is required for ponds and they have a low volume to area ratio; (2) Mono-cultivation of the desired microalgae is difficult to maintain for most microalgae species because of constant airborne contamination, except for extremophile species; (3) Environmental growth parameters of cultivation rely primarily on local weather conditions, which may not be controlled and make production seasonal; (4) Harvesting is laborious, costly, and sometimes limited by low cell densities; (5) Continuous and clean water is needed; and (6) Production of pharmaceutical or food ingredients is not feasible or is very limited (Chen, 1996; Tredici, 1999; Molina Grima et al., 1999, 2003; Lee, 2001; Pulz, 2001; Wen and Chen, 2003; Sansawa and Endo, 2004; Carvalho et al., 2006; Chen and Chen, 2006; Chisti, 2007; Patil et al., 2008).

To overcome inherent disadvantages of using open, less controlled environments, numerous closed photo-bioreactors (PBR) of various volumes and shapes have been designed (Molina Grima et al., 1999; Tredici, 1999, 2004; Tsygankov, 2000; Zhang et al., 2001; Barbosa, 2003; Suh and Lee, 2003; Zijffers et al., 2008). The principle final goal of any PBR is reduction in biomass production costs. This has been done by improving catalysts, shaping of the PBR, controlling environmental parameters during cultivation, aseptic designs, and operational approaches to overcome rate-limiting of growth, such as pH, temperature, and gas diffusion. Overcoming these limitations make monocultures and production of pharmaceutical and food goods possible (Cooney, 1983; Chen, 1996; Apt and Behrens, 1999; Pulz, 2001; Wen and Chen, 2003; Lebeau and Robert, 2006).

Similar to the open-pond concept, large-scale PBRs have three major disadvantages that make them uneconomical for low-cost end-products: At operational volumes of 50–100 L or higher, it is no longer possible to disperse light efficiently and evenly inside the PBR (Chen, 1996; Pulz, 2001); development of algal biofilm fouls PBR surfaces and thereby limiting light penetration into the culture. A high initial investment in infrastructure and continuous maintenance is required (Carvalho et al., 2006). Nonetheless, numerous applications of PBR for microalgae were proposed and were reviewed (Apt and Behrens, 1999; Lebeau and Robert, 2006; Muñoz and Guieysse,

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