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Algae/bacteria consortium in high rate ponds: Influence of solar radiation on the phytoplankton community

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ABSTRACT

Using multivariate statistical tools, the composition of the phytoplankton community was related to the characteristics of the domestic sewage used as culture medium in three high rate ponds (HRPs) submitted to different solar radiation levels. A total of 32 genera of phytoplankton were identified in the ponds; the class *Chlorophyceae* was the most abundant during the entire sampling period, with a larger number of individuals of the genus *Desmodesmus* in the summer and fall, and of the genus *Chlorella* in the winter and spring. The lowest occurrence of phytoplankton was observed in the fall, with behavior similar to the evolution of solar radiation throughout the year. Blocking over 30% of the solar radiation allowed for less variability of the phytoplankton community and favored the growth of biomass with higher density of individuals, as well as higher concentrations of chlorophyll-*a* and dissolved oxygen. On the other hand, the pond with 80% of shading presented the lowest mean density of organisms; from the perspective of wastewater treatment, however, it can be considered the most efficient in terms of organic matter and nutrient removal. According to the regression analysis, the algal biomass in HRPs can be maximized mostly if we consider the positive effect of carbon and phosphorus and the limiting effect of nitrogen and non-biodegradable organic load. For the conditions evaluated in this study, the photoinhibition phenomenon was not observed. Other aspects such as competition with other microorganisms for space and nutrients, or predation by zooplankton, seemed to be more significant for the growth and development of algal biomass.

1. Introduction

Biological systems for wastewater treatment are usually controlled by operational variables, typically chemical or physical. Strategies to optimize such systems are usually regarded more as chemical than biological (Yuan and Blackall, 2002), compromising the development of the microorganisms responsible for degradation, and consequently affecting treatment efficiency.

High rate ponds (HRPs) are shallow, open ponds designed in a racetrack configuration, with continuous mixing provided by paddle-wheels (Oswald and Golueke, 1960). In open systems for microalgae cultivation, especially those associated with wastewater treatment, the combination between microalgae and bacteria has been widely studied. Algal photosynthesis produces the oxygen required for degradation of organic matter by heterotrophic bacteria. Nutrients and the CO_2 from oxidation are assimilated by the algae (Santiago et al., 2013). The production of microalgae in consortium with other microorganisms may represent an alternative for reducing the costs involved in the process of obtaining biofuels, since it presents competitive advantages such as less influence of environmental fluctuations and easier harvesting and processing of the biomass (Pires et al., 2013).

The characteristics of a HRP, such as the heavy load of mineral and organic pollution, combined with the shallow depth, intensify the strong daily and seasonal changes of the growth medium variables. The planktonic succession in HRPs can be considered similar to that in natural environments. The difference is that, depending on the characteristics of the effluent used as culture medium, the nutrients (nitrogen and phosphorus) are not limiting

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factors. The HRPs belong to the group of shallow hypereutrophic environments, and thus present some analogies with the functioning of a natural pond. Due to the permanent agitation and constant nutrient input, the HRP is very similar to a natural ecosystem, despite its reduced hydraulic retention time (HRT). The continuous agitation given by the paddlewheels submits the algae to fast cycles of vertical circulation, and consequently, luminosity: the cells frequently and rapidly pass through the clarity of the surface and the darkness of the bottom, which are translated into intense photosynthetic mechanisms and nutrient assimilation.

The most relevant variables to describe the dynamics of the phytoplankton are the ones directly related to the processes of the eutrophic or hypereutrophic medium. These variables are light intensity, temperature, nutrients and turbulence or mixture (Wium-Andersen et al., 2013). Directly involved in the photosynthetic mechanisms, the light intensity represents a primary factor for the development and multiplication of planktonic algae. As well as for vegetal organisms, there is a minimum light requirement for phytoplankton to perform photosynthesis. In a system fed with wastewater, the light frequently plays a limiting role, and the productivity is strongly related and dependent upon the amount of solar energy received (Cromar et al., 1996). It is well known that high light intensity for a considerable amount of time can be translated into a decrease in photosynthetic production (Chisti, 2007); however, on the other hand, it is believed that some algal species are highly capable of progressively adapting to the light intensity to which they are submitted, thus photoinhibition decreases or disappears in well adapted algae (Horne and Goldman, 1994). Moreover, for the specific case of the HRPs, since algae are temporarily submitted to strong light intensities due to turbulence, this inhibitory effect of solar radiation can be disregarded (Fernandes et al., 2014); however, the influence with respect to the composition of the community should not be neglected.

Despite its importance as a renewable energy source and the wide research on HRPs for wastewater treatment, the phytoplankton community in these systems is very little studied. There are few studies which exclusively address their interaction and behavior according to the characteristics of the culture medium and environmental variables (but see Barthel et al., 2008; Canovas et al., 1996; Park et al., 2011). Therefore, the objective of this study is to evaluate how solar radiation influences the phytoplankton community in HRPs. Using multivariate statistical tools, the composition of the phytoplankton community was associated with the characteristics of the domestic sewage used as culture medium in three HRPs submitted to different levels of solar radiation.

2. Material and methods

The experiment was carried out in Viçosa-MG, Brazil (20°45′14″S, 42°52′54″W). The climate in Viçosa is Cwa (humid subtropical climate) according to the Köppen classification, the annual average temperature is 19.4 °C, and the annual average relative humidity is 81%. The annual average precipitation is 1221.4 mm, concentrated in the period from November to March. The studied domestic sewage treatment system consisted of an upflow anaerobic sludge blanket (UASB) reactor followed by three HRPs.

In order to evaluate the influence of different solar radiation intensities in the biomass growth process, the HRPs were covered with screens that provided different shade levels. The first pond (HRP1) was the control unit without any cover, and the ponds 2 (HRP2) and 3 (HRP3) operated with screens that blocked 30% and 80% of the solar radiation, respectively. Fig. 1 illustrates the scheme of the experiment. The shading screens were made of high-density polyethylene, such as those used for agriculture crops, Sombrite® type 1003 and type 1008, for ponds 2 and 3, respectively.

2.1. Environmental and water quality variables

Combined samples for the analysis of physical and chemical variables were carried out every 2 h (from 8 am to 6 pm), on a weekly basis from January to November. Simple random samples for chlorophyll analyses were collected at 10:00 h. On sampling dates, the variables pH, dissolved oxygen (DO), and temperature (*T*) were measured at the site, every 2 h, using the Hach HQ40d portable meter (Luminescent Dissolved Oxygen–LDO–for DO). The photosynthetically active radiation (PAR) (400–700 nm) was also measured on the water surface using the LI-COR LI-193 Underwater Spherical Quantum Sensor.

The physical–chemical analyses of influent and effluent wastewater were carried out according to the Standard Methods for the examination of water and wastewater (APHA, 2005): total and soluble chemical oxygen demand (CODt and CODs, respectively) (5220D), total suspended solids (TSS) (2540D), volatile suspended solids (VSS) (2540 E), ammoniacal nitrogen (N-NH_4^+) (4500-NH3C), total kjeldahl nitrogen (TKN) (4500-NorgB), nitrate (N-NO_3^-) (4500-NO_3A), total phosphorus (TP) (4500P D), soluble phosphorus (SP) (4500 PC). The dissolved organic carbon (DOC) and the dissolved inorganic carbon (DIC) were determined using the Shimadzu TOC 5000 analyzer. The chlorophyll-*a* levels were determined by spectrophotometry according to APHA (2005), and the calculations were performed using equations described in the Nederlands Norm Nen 6520 (NEN, 1981)

2.2. Composition of the phytoplankton community

The samples for the characterization of the phytoplankton community were collected at a 2-week interval at each HRP from January to November. For qualitative analysis, the samples were collected using a 20-μm mesh-size plankton net and immediately fixed with formaldehyde 4% (1:1). The taxonomic identification was performed at the genera level using an inverted optical microscope (Olympus IX 70). The morphologic and morphometric characteristics of the vegetative and reproductive cycles of the relevant species (of significant taxonomic value) were analyzed according to the specialized literature (Bicudo and Menezes, 2006; Bourrelly, 1970; Parra et al., 1983).

For the quantitative analysis, 1-L effluent samples were collected in amber bottles and preserved in laboratory with Lugol's solution 5%. After sedimentation, the supernatant was discarded and the remaining concentrate of approximately 100 mL was homogenized and 1 mL was transferred using a pipette to the

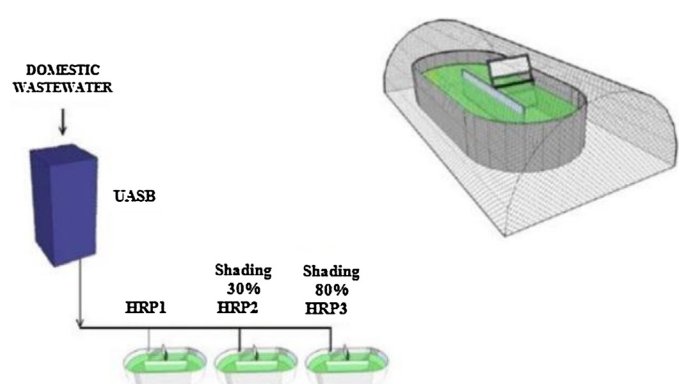

Fig. 1. Experiment scheme.