

Conditions for cyanobacteria biomass development and selection for further processing

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Abstract – the purpose of research is to define place of cyanobacteria in the system of micro and macroalgae, discover peculiarities for conditions of their development and biomass selection for further processing.

Key words – cyanobacteria, microalgae, alternative sources, biomass development, nutrients

I. Introduction

Cyanobacteria appear to be one of the oldest, biggest and most important bacteria on earth, since they launched process of oxygen production. Because they are photosynthetic, they are often called `blue-green algae`. Cyanobacteria can modify in special cell types that are called heterocysts and fix nitrogen, tolerate high levels of temperature which proves in its blooming in hot springs and ice lakes of Antarctica. Cyanobacteria can produce wide range of biologically active compounds, with some of which have antiviral, anticancer and UV resistant qualities. As nutrient source for biomass development, it is important today to use CO₂ aiming minimization of its negative impact on environment, and to use degistate in biogas production.

II. Conditions for cyanobacteria development

Cyanobacteria don't need vitamins for existence. They can use nitrates or ammonia as a source of nitrogen and they also need phosphorus and such microelements as iron. Most of cyanobacteria types are phototrophs, but some filamentous types can bloom in darkness, using sugars, glucose or sucrose as carbohydrates and source of energy.

Conditions for cyanobacteria development depend on many factors and are interconnected.

Problem of impact factors on cyanobacteria development based on example of *Anabaena*, *Aphanizomenon*, *Cylindrospermopsis*, *Nodularia*, *Lyngbya*, *Oscillatoria*, *Microcystis*, *Planktothrix* was a research issue for large amount of scientists (Chorus and Bartram 1999; Carmichael 2008; Paerl and Huisman 2008; Hudnell 2008, 2010; O'Neill et al. 2012; Paerl and Paul 2012). Among factors that affect cyanobacteria development conditions, these scientists were focusing on: salinity, enough nutrient supply. The following factors were also taken into account: calm water, stratified conditions, a lot of irradiance, high levels of water temperature.

Salinity

Marine cyanobacteria such as *Prochlorococcus*, *Synechococcus sp.* and *Trichodesmium sp.* showed presence of high level of salinity as a result of the research done in the laboratory. They also demonstrate wide salinity tolerance ranges. For instance, the least salinity tolerant *Cylindrospermopsis* thrives under level of salinity of 2,5 ppt. At the same time, the most salinity tolerant are *Anabaenopsis* and *Nodularia* that thrive under 5-20 ppt levels of salinity (Moisander et al. 2002). As an example, *Microcystis aeruginosa* is tolerant up to level of 10 ppt without changes in its growth, compared to those, that are in freshwater (Tonk et al. 2007). Based on such results one may conclude, that under optimal growth conditions, these species might potentially thrive in regions, where water is darker. Over the last decades one may observe a spread in the geographical extent such species into the mesohaline (5-15 ppt) (Paerl and Paul 2012). As example, blooming of *Microcystis aeruginosa* can be met in Baltic Sea (Maestrini et al. 1999) and at the San Francisco Estuary (Lehman et al. 2013). These species as a result of research showed that other factors among salinity influence geographical distribution.

Level of nutrient concentration

As in other photosynthetic phytoplankton, under optimal temperature level and irradiance, cyanobacteria biomass accumulation is directly proportional to the amount of nutrients, available in water column.

Concentrations of nitrogen and phosphorus

Researches of great number of scientists have shown that cyanobacteria growth in freshwater systems (rivers and lakes) is frequently linked with excessive phosphorus loading (Likens 1972, Schindler 1977, Edmondson and Lehman 1981, Elmgren and Larsson 2001, Paerl 2008, Schindler et al. 2008). Supply of nutrients in stationary and non-stationary sources in agriculture or wastewater effluents leads to simultaneous increase of phosphorus and nitrogen concentrations (Paerl and Paul 2012, Paerl et al. 2014b). During summer researches the results showed that when cyanobacteria biomass is at its highest peak and given minimal nutrient concentrations, nitrogen and phosphorus exert equal control over biomass accumulation in this system (Paerl et al. 2014a). In general, dominance of both N₂-fixing and non-N₂ fixing cyanobacteria such as *Aphanizomenon flos aquae*, *Nodularia spumigena*, *Microcystis aeruginosa* and *Cylindrospermopsis raciborskii*, have increased worldwide in concert with increased loads of both nitrogen and phosphorus (Chapman and Schelske 1997, Jacoby et al. 2000, Gobler et al. 2007, Burford et al. 2006, Burford and O'Donahue 2006, Hong et al. 2006, Suikkanen et al. 2007, O'Neill et al. 2012). In order to change nutrient ratio aiming to affect phytoplankton growth, concentrations of nutrients should be so low (phytoplankton biomass relatively) that in the end either phosphorus or nitrogen will reduce their growth.

Phosphorus and nitrogen supplement of cyanobacteria is greater than of other eukaryotic group due to the large protein demand of the peripheral light harvesting antennae.

Irradiance and Water Clarity

Carotenoid pigments that are concentrated in cyanobacteria serve a photoprotective function by dissipating excess light energy when required allowing cyanobacteria to be exposed to high irradiances without experiencing photoinhibition (Paerl *et al.* 1983, 1985). Aided by their positive buoyancy, cyanobacteria such as *Microcystis*, can grow very close to the surface by tolerating irradiance levels that are inhibitory to other members of the phytoplankton community. As a result, these cyanobacteria can increase their cell densities past the point where they would ordinarily become light-limited by self-shading. Growing close to the surface can also help cyanobacteria avoid light limitation if there is a high concentration of suspended sediment matter in the water.

Temperature

Perhaps one of the most important factors in controlling the growth rate of cyanobacteria is temperature (Robarts and Zohary 1987, Butterwick *et al.* 2005, Reynolds 2006, Paerl and Huisman 2008). Cyanobacteria isolated from temperate latitudes (i.e. excluding polar regions) typically have temperature growth optima between 25 and 35°C (Reynolds 2006, Lurling *et al.* 2013). For example, in a survey of eight cyanobacteria the growth optima of two *Microcystis aeruginosa* strains were 30-32.5°C and that of *Aphanizomenon gracile* was 32.5°C. Lower growth temperature optima were observed in *Cylindrospermopsis raciborskii* and *Planktothrix agardhii*, both at 27.5°C while *Anabaena sp* had an optimum of 25°C (Lurling *et al.* 2013).

Cyanobacteria growth is reported from cryophilic (+4 °C) up to thermophilic conditions (e.g. *Synechococcus lividus*, 75 °C). Photosynthetic activity, without observable growth, was reported by De Vera even at -30 °C. Miyake and Nishioka reported PHA production with *Synechococcus* MA19 at 50 °C, while almost all the other authors had done their cultivation experiments in the range between 20 and 30 °C. Thermophilic conditions are beneficial because of increased metabolic turnover and because of a significantly reduced contamination risk. However, thermophilic cyanobacteria able to produce PHA are rare, and thermophilic production in a largescale photobioreactor will cause very high effort for thermal insulation.

Level of pH

According to Brock, cyanobacteria generally seem to be unable to grow at a pH lower than 4 to 5. In fact, most are alkalophiles having their growth optima between pH 7.5 to 10.

Although pH next to alkalinity and temperature influences the interspeciation of dissolved inorganic carbon, it has an effect on growth independently.

Overall, the optimal pH for maximal growth rate cannot be generalized as it varies from strain to strain and depends on their natural environment.

III. Selection of biomass for further processing

As a result of cyanobacteria biomass development, water and nutrients after harvesting cyanobacterial biomass and product extraction can be directly recycled. Biomass can also be anaerobically digested or hydrothermally liquefied via HTL (mineralization of organic nutrients) and then recycled. Recycling process water directly can increase the concentration of inhibitory substances and dissolved organic matter from the previous batch produced by cyanobacteria, which decrease the productivity of cyanobacteria. Furthermore, nutrient competition may arise by enhanced bacterial growth.

Auto-selectivity, a combination of cultivation conditions favourable for the intended strain and unfavourable for all potential contaminants, is a serious goal for all biotechnological processes. For cyanobacteria, this selectivity can be achieved by setting several parameters simultaneously: the lack of dissolved organic carbon, limiting concentrations of nitrogen and phosphorous, and a pH-value at or above 8.5. One can also observe repeatedly some growth of green algae (*Chlorellasp.*) in the relevant research. As the culture reaches its stationary phase, cells will die and release their content. This may be a carbon and energy source for heterotrophic contaminants, making long time running batch processes critical.

In cyanobacteria cultivation it is important to mind, that under unsanitary conditions the contamination is inevitable. Thus, purification will be one of integral elements of cyanobacteria selection for further processing.

Conclusion

Cyanobacteria growth conditions arrive from several factors that have different effect for every species. Cyanobacteria are important in the system of balanced environmental management, since their nutrients can be wastewater and industrial-agricultural effluents.

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