



Characterization of planktonic and biofilm cells from two filamentous cyanobacteria using a shotgun proteomic approach

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ABSTRACT

Cyanobacteria promote marine biofouling with significant impacts. A qualitative proteomic analysis, by LC-MS/MS, of planktonic and biofilm cells from two cyanobacteria was performed. Biofilms were formed on glass and perspex at two relevant hydrodynamic conditions for marine environments (average shear rates of 4 s^{-1} and 40 s^{-1}). For both strains and surfaces, biofilm development was higher at 4 s^{-1} . Biofilm development of *Nodosilinea* sp. LEGE 06145 was substantially higher than *Nodosilinea* sp. LEGE 06119, but no significant differences were found between surfaces. Overall, 377 and 301 different proteins were identified for *Nodosilinea* sp. LEGE 06145 and *Nodosilinea* sp. LEGE 06119. Differences in protein composition were more noticeable in biofilms formed under different hydrodynamic conditions than in those formed on different surfaces. Ribosomal and photosynthetic proteins were identified in most conditions. The characterization performed gives new insights into how shear rate and surface affect the planktonic to biofilm transition, from a structural and proteomics perspective.

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Introduction


Marine biofouling causes several ecological problems and severe economic impacts on aquatic environments and marine industries (Carvalho 2018). Biofilm formation on submerged equipment containing optical and electrochemical sensors (which are used for on-site monitoring with continuous measuring) can affect readings and lead to incorrect measurements (Delauney et al. 2010). Cyanobacteria and diatoms are responsible for the initiation of biofilm formation by the production of large amounts of extracellular polymeric substances (EPS), and they also represent the major components of marine biofilms (Bharti et al. 2017). Additionally, these microfooler organisms can promote the adhesion of macrofouler organisms such as invertebrate larvae, mussels, seaweeds and barnacles (Mieszkin et al. 2013).

Physico-chemical factors related to the surface being colonized (Crawford et al. 2012) and flow velocity and shear rate (Allen et al. 2018) are some of

the most critical parameters that affect biofilm composition, structure and development. Moreover, these factors can also impact EPS production, energy metabolism and induce molecular changes in biofilms (Moreira et al. 2015). The development of novel anti-fouling strategies for marine applications is of paramount importance (Telegdi et al. 2016). Therefore, molecular studies capable of identifying the agents and pathways affecting the settlement of biofouling organisms are necessary to obtain new targets and develop more effective biofilm mitigation strategies.

Omics approaches such as genomics, transcriptomics, proteomics and metabolomics have been widely used to characterize bacterial virulence factors (Alexova et al. 2011; Babele et al. 2019) and provide new information about biofilm development and regulation (Leary et al. 2014; Parnasa et al. 2016). Specifically, proteomic studies have also been used to understand the molecular mechanisms involved in important cellular processes such as cell division, metabolism, transport, stress response and motility in these microbial communities (Parnasa et al. 2016;

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