The exopolysaccharide production by *Cyanothece* sp. PCC 7822 through an adapted metabolism

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**Introduction**

The Algotech project was established to progress on the field of high-added value compounds production by Microalgae/Cyanobacteria. In this way, we investigate the exopolysaccharide (EPS) production of *Cyanothece* sp. PCC 7822 well-known for its diazotrophic metabolism in light/dark cycle condition. The influence of several parameters on EPS production and composition will be examined throughout this study.

**Impact of nitrogen sources**

Nitrate, ammonium and N₂ conditions are characterised by comparable lag and exponential phases. The main difference consists in the highest reached OD₂₅₀ mm. In opposition, the strain can’t grow in presence of 8.5 mM urea

At 300h of culture in N₂ condition, a second exponential rise in the OD follows the stationary phase. It could be related to a complete use of NO₃ by the bacterium and a switch to a diazotrophic metabolism (Fig. 1). However, NO₃ quantification exhibits 8 mM of NO₃ when the OD increases for the second time (results not shown).

![Fig 1 - Growth curve of *Cyanothece* sp. PCC 7822 in BG 11 medium supplemented with different nitrogen sources: NO₃ 17mM, NH₄Cl 17 mM, urea 8.5 mM and N free. The experiment was realised in 4 replicates.](image)

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**Fig. 2 - Observation of *Cyanothece* sp. PCC 7822 exopolysaccharides by using a positive alcian blue staining at pH 2.5 in presence of 4 different nitrogen sources (17 mM NO₃, 17 mM NH₄Cl and 8.5 mM urea). Exopolysaccharides are observed at different moments of the growth curve and experiment are performed in 4 replicates.**

Fig. 2: Observation of *Cyanothece* sp. PCC 7822 exopolysaccharides by using a positive alcian blue staining at pH 2.5 in presence of 4 different nitrogen sources (17 mM NO₃, 17 mM NH₄Cl and 8.5 mM urea). Exopolysaccharides are observed at different moments of the growth curve and experiment are performed in 4 replicates.

The presence of nitrate enhances exopolysaccharide production in the form of RPS (Released PolySaccharide) (Fig. 3). Usually, the presence of combined nitrogen sources increases EPS formation because of the lower energy required for their assimilation unlike N₂ fixation. In addition, a quantification of EPS (Capsular PolySaccharide) is necessary to obtain a global view of EPS produced by the strain. The rise of RPS measured at the end of the curve in N₂ condition could be related to bacteria degradation and intracellular polysaccharide release.

![Fig 3 - Total carbohydrate quantification of released polysaccharides by the phenol/sulfuric acid method when *Cyanothece* sp. PCC 7822 is cultivated in BG 11 supplemented by 3 different nitrogen sources: NO₃ 17mM, NH₄Cl 17 mM and N free.](image)

**Conclusion**

We chose to modify the N source because of its influence on C:N ratio and so on carbon metabolism. Interestingly, modification of this parameter has an impact on *Cyanothece* sp. PCC7822 growth but also on EPS production and appearance. Future experiments will go further into the effect on EPS composition and structure but also on EPS biosynthesis pathways. The N source is not the only parameter regulating EPS production. Effectively carbon source, light intensity or salt concentration could also affect EPS. Therefore, some of these will be attractive to analyse in addition to nitrogen supply variation.

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