**Effect of a double stress Light-Salinity, on the polysaccharides compartmentalization of *Porphyridium cruentum***

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**Highlights**

* Identification of the critical light stress for polysaccharide accumulation
* Identification of the optimal osmotic/light stress combination for floridoside accumulation
* Optimization of the added value of the biomass of *Porphyridium cruentum*

**1. Introduction**

*Porphyridium cruentum* is a photosynthetic micro-organism which could produce many valuable metabolites depending on the growing conditions. In the present work, the effect of the salinity and light on the carbohydrates biosynthesis has been particularly investigated. Floridoside is the major glycoside produced by *Porphyridium* (S. Y. Li *et al.*, 2001). This osmoregulator has been identified as a potential precursor for exopolysaccharide (S. Li & Arad, 2002; S. Y. Li *et al.,* 2001) and it is also an interesting molecule for pharmaceutics and cosmetics application like Antiviral/Antitumoral (Bessie & Magne, 2007), anticomplementary agent focus in therapeutic complement depletion (Courtois, Simon-colin, Boisset, & Berthou, 2008), bone formation promoter (Gyu, 2015) or for other application like biofouling (Taylor et al., 2010). In a previous study, salinity has been identified has the major parameter which interfers in floridoside accumulation and light stress has been identified has the most effective parameter to induce polysaccharide production (bound polysaccharide and released polysaccharides). The aim of this study is to define a treatment post harvesting for polysaccharide and floridoside accumulation.

**2. Methods**

For that purpose an original approach was developed, combining small scale screening photobioreactors for the testing of the various environmental conditions (EOSS 2), to an original rapid quantification method of the main carbohydrate fractions impacted by the tested cultivation parameters (the starch, the intracellular low weight carbohydrates - LWC, the EPS, BPS, etc…). In this presentation, salt and light stress have been combined, the objective was to accumulate floridoside after an osmotic stress then apply a light stress to produce valuable polysaccharides. Furthermore another protocole has been tried, light stress before the osmotic stress, the objective was to accumulate both molecule in order to add value to the final biomass. For the first protocol, salinities have been studied from 22, 50 and 72 g NaCl.L-1 for two MRPA (Mean Rate of Photon Absorption) respectively 0 and 5,3 µmoles photon.g-1.s-1 light stress was applied after 6 hours of osmotic stress for 12 hours. For the second protocol, different MRPA have been applied (from 2 to 25 µmoles photon.g-1.s-1) and after 24 hours salinity has been increase to 50 g NaCl.L-1 in order to identify the effect of osmotic stress on a starch-rich biomass (Floridoside and starch has a common precursor).

**3. Results and discussion**

Experimental design identified a relation between light and salinity for floridoside accumulation. Indeed, during the osmotic stress, *Porphyridium* accumulate floridoside for MRPA=0 (from 1,60 mg Floridoside.10-6 cells to 2,60 mg Floridoside.10-6 cells, so +39% in 3 hours) and more for MRPA= 5,3 (from 1,60 mg Floridoside.10-6 cells to 3,41 mg Floridoside.10-6 cells so +53% in 3 hours). BPS (bound polysaccharides) accumulation is highly affected by the osmotic stress. Indeed, osmotic pressure block the carbohydrate metabolism. After the light stress, only lower salt concentration cultures have produced a large amount of BPS (+59 % polysaccharide after 14h of light stress). A significant relation appears between salinity concentration and BPS accumulation rate, indeed, BPS accumulation rate is inversely proportional to the salt concentration.

**4. Conclusions**

These experimentations leads to the determination of the critical light stress (measured by evaluation of the mean rate of photon absorption in µmole.g-1.s-1) and the critical salt concentration for polysaccharides and floridoside accumulation by *Porphyridium cruentum*. *Porphyridium cruentum* has a particular behavior in chemostat and could not produce a large amount of floridoside and bps. This treatment could be applied after harvesting in order to produce floridoside and BPS in the same time which will add value to the final biomass.

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