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Interactive effects of salinity and light on growth of EPA and DHA rich-microalgae *Pavlova lutheri*

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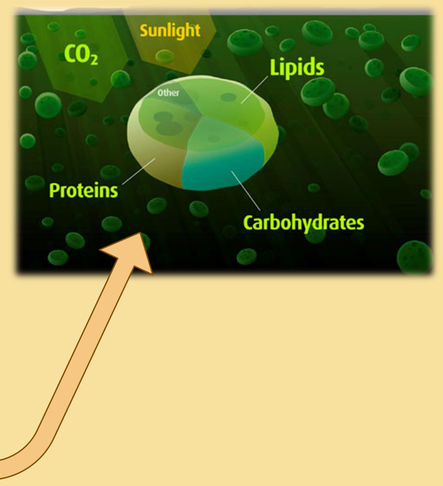
1 INTRODUCTION

Light intensity and salinity are some of the critical parameters for the regulation of autotrophic growth of microalgae and can bring significant changes in chemical composition of the biomass. Optimum light requirement and salinity level vary between different species and during cultivation needs to be adjusted for maximum biomass yields or accumulation of the desired compound (1). Effects of salinity on marine microalgae have been examined by several researchers (2, 3). However, limited reports are available examining interactive effects of salinity in combination with other regulatory parameters such as light intensity. *Pavlova lutheri* is a high lipid-producing marine microalgae, especially rich in EPA and DHA.

The aim of this study is to investigate growth trend of *P. lutheri* under different environmental conditions (salinity and light) in order to determine the most optimal combination for high growth rate.



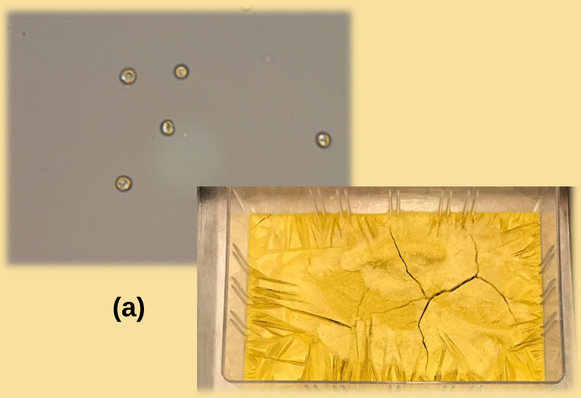
2 MATERIALS AND METHODS

Pavlova lutheri (SAG 926-1) cultivation experiments were carried out in F2P growth medium using 5 L GS Schott bottles (Table 1).

Table 1. Experimental set-up

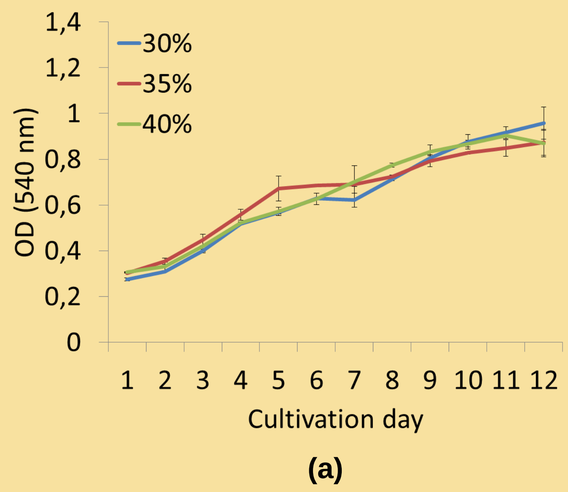
Light intensities (µmol photons $m^{-2}s^{-1}$)	Salinity (ppt)		
	40	**35**	**30**
75	X1	X4	X7
150	X2	X5	X8
300	X3	X6	X9

Cultures were aerated by mixture of sterile air and CO_2 (5%). Cultivation temperature range was 25 ± 1 °C. pH was kept at 8 by controlling CO_2 addition. Growth was monitored daily by taking 5 mL culture sample and measuring optical density at 540 nm. Biomass separation was performed by centrifugation at 6500 x g for 10 min followed by freeze-drying. In addition, lipid content and fatty acids profile were determined and compared during lag and stationary phase of cultivation.

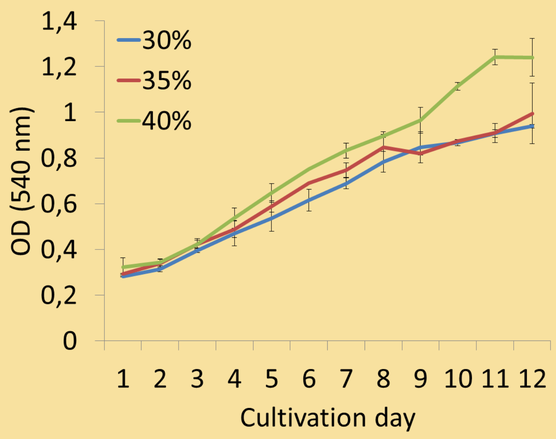


Picture 1. *Pavlova lutheri* **(a)** Optical micrographs (×100) **(b)** Freeze-dried biomass **(c)** Cultivation set-up

3 RESULTS



(a)



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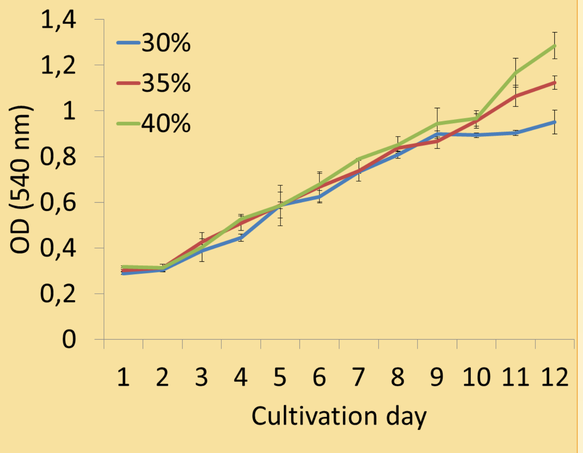

(c)

Figure 1. Effect of salt concentration on growth of *P. lutheri* during 12 days cultivation period under different light intesities: **(a)** 75 µmol photons $m^{-2}s^{-1}$ **(b)** 150 µmol photons $m^{-2}s^{-1}$ **(c)** 300 µmol photons $m^{-2}s^{-1}$. The results are presented as the means of n = 4 measurements from two biological replicates; error bars represent standard deviation .

Light intensity and salinity level showed to have interactive effect on the growth of *P. lutheri* (Fig 1). At low light intensities (Fig 1a) changes in the salinity showed no effect on the growth rate ($p>0.05$) while at higher light intensities (Fig 1ab) salt concentration affected the growth rate ($p<0.05$). The most optimal combination was salinity at 40 ppt with light intensity of 300 µmol photons $m^{-2}s^{-1}$ (**$\mu = 0.13$ day^{-1}**), comparing to the lowest specific growth rate (**$\mu = 0.09$ day^{-1}**) at salinity level 35 ppt and light intensity 75 µmol photons $m^{-2}s^{-1}$.

Total lipids and PUFAs (EPA and DHA) content increased significantly ($p<0.05$) with the cultivation time. EPA increased from **18.72±0.94%** to **24.79±0.27%** of total FA while DHA increased from **8.57±0.46%** to **11.95±0.66%** during 28 days of cultivation.

4 CONCLUSION

- Increase in light intensity in combination with high salinity level showed to have positive effect on the growth of *P. lutheri* resulting in higher growth rate.
- High content of EPA and DHA should be further tested in *P. lutheri* for their changes under different light and salinity levels in order to determine the most optimal condition for accumulation of these fatty acids at high growth rate.

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