An assessment of optimal growth conditions for microalgal strains using crossed gradients of light and temperature – a prerequisite for toxicity tests

Martina Pichrtová, Kateřina Černá, Magda Škaloudová, Jana Veselá, Yvonne Němcová, Jiří Neustupa & Pavel Škaloud

Charles University in Prague, Faculty of Science, Department of Botany, Benátská 2, CZ-12801, Praha 2, Czech Republic

e-mail: pichrtov@natur.cuni.cz

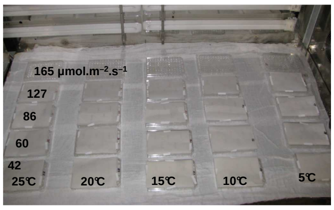
Introduction

Algal biofilms growing attached to submerged surfaces (so-called fouling) represent a serious problem for watercraft transport. Fouling negatively affects the sailing speed and therefore increases the fuel consumption and total costs of transport. Therefore, special antifouling coating solutions have been applied that should inhibit the algal growth. However, the development of new coatings and subsequent testing of their efficiency and suitability usually takes about 5 years and is also quite expensive.

The aim of the IATS project (7th Framework Programme) is to design and develop a fully innovative, completely automated antifouling test system for professional examinations of marine coatings. Such system shall be able to analyze antifouling potential of tested coating solutions in laboratory conditions within only few weeks and completely automatically. Consequently, tests of newly developed coatings will be performed consederably faster, at lower costs and in a more effective way.

The role of the Charles University in Prague (Phycology research group and the Culture collection CAUP) within this project is to select a set of suitable microalgal strains that will be used for the antifouling coatings tests. However, before performing any toxicity tests, the optimal growth conditions of test species should be investigated, which was the objective of the presented work.

An example of biofouling of algae and invertebrtes on a vessel (www.daff.gov.au)



The crossed gradient unit of light and temperature

Methods

The microalgal strains used in the experiments were obtained directly from biofilms growing on ships and isolated into clonal cultures using glass micropipettes. The algae were cultivated on two main types of culture medium – WC was selected for diatoms and BBM for the rest of the strains.

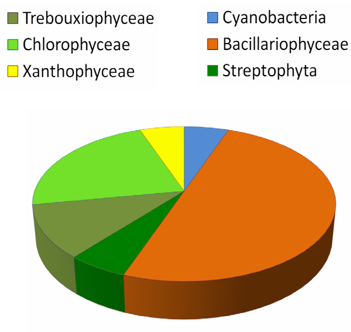
Optimal conditions for growth were assessed using the crossed gradient unit of light and temperature which represents an effective way how to test various combinations of cultivation conditions at once (Kvíderová & Lukavský 2001). Diluted, exponentially growing cultures were pipetted into 96 or 9-well culture plates and kept under continuous illumination for 9-15 days. The algae were cultivated in 25 combinations of temperature (5°C, 10°C, 15°C, 20°C, 25°C) and irradiance (42, 60, 86, 127, 165 μmol m-2 s-1) with three replicates in each combination of factors.

Biomass was estimated as percentage cover or by using image analysis software NAJA (Hauer & Jirka 2007)

Experimental strains

Out of all isolated strains 18 were selected for further investigations. The main criterion for the strain selection was satisfactory growth in laboraory conditions which is neccessarry for the long-time maintenance of the cultures.

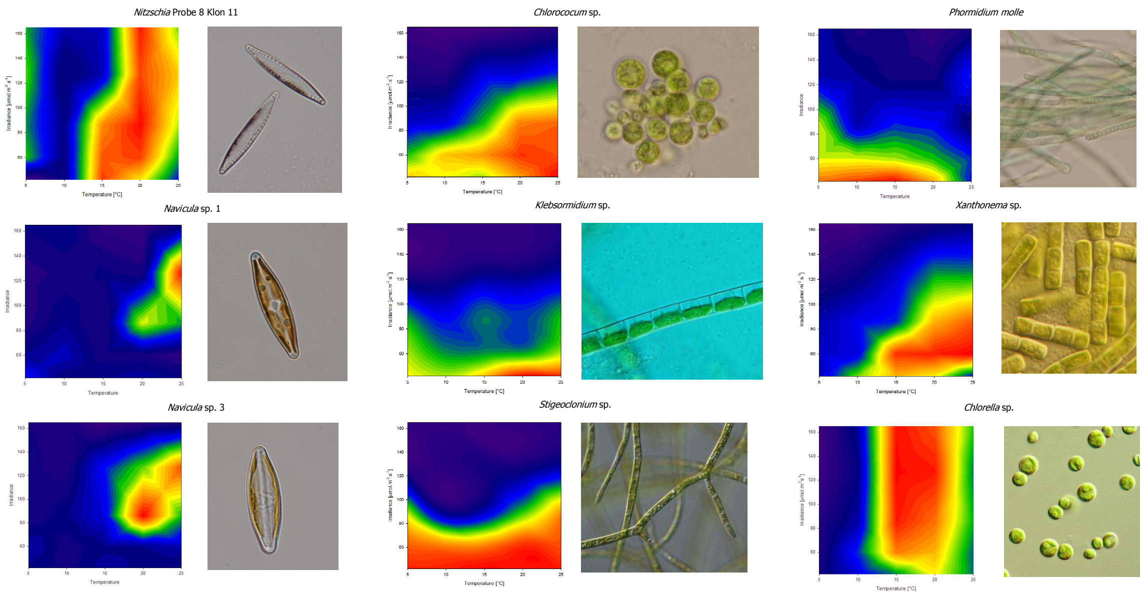
The selected strains belonged to different taxonomic groups, the proportion of which can be seen in the following chart:



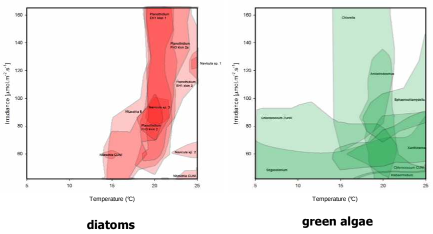
Results

Assessment of optimal growing conditions of selected species. The higest biomass production is indicated in red, no growth in dark blue. Optimal conditions of all tested strains are presented in a table:

Strain	Optimal growth	
	temp. °C	irradiance $\mu mol.m^{-2}.s^{-1}$
Cyanobacteria		
Phormidium molle SAG 26.99	15	42
Diatoms		
Achnanthidium sp.EH1 Klon 1	20	42-60
Navicula sp.1 Lovosice	25	127
Navicula sp.2 Lovosice	25	60
Navicula sp.3 Lovosice	20	86
Gomphocymbella sp. Lovosice	20-25	42-60
Nitzschia sp. Lovosice	25	42
Achnanthidium sp. EH1 Klon 3	20	140-160
Achnanthidium sp. FH3 Kl. 2A	15	60
Nitzschia sp. Probe 8 Klon 11	20	86-127
Xanthophyceae		
Tribonema sp. Lovosice	20-25	60
Trebouxiophyceae		
Chlorella sp. Lovosice	15-20	86-127
Sphaerochlamydella sp. Lovos.	20	86
Chlorophyceae		
Stigeoclonium sp. Lovosice	15-25	42-60
Chlorococcum sp. Zurek 04	5-15	42-60
Chlorococcum sp. Lovosice	25	42
Ankistrodesmus sp. V.H.S 5	20	86
Streptophyta		
Klebsormidium sp.Lovosice	20-25	42



The summary of the optimal growth conditions in diatoms and green algae:



Summary

Temperature and illumination are considered leading abiotic factors determining successful cultivation of microalgae. Therefore, we cultivated selected microalgal strains using crossed gradients of temperature and light to assess their optimal growth conditions.

The results showed that the investigated strains differed both in their optima and the range of conditions for growth. Nevertheless, some generalizations can be concluded. Most diatoms grew optimally at 20 °C and 86 μmol m-2 s-1. Several strains were able to sustain a wide range of light intensity, but none of the diatom strains was able to grow well in a wide range of temperature. On the other hand, green algae (including a Xanthophyte alga *Xanthonema*) provided more diverse response, and wide ranges of optimal temperature and light intensity were observed. However, most of the green algal strains grew optimally at 20 °C and 60 μmol m-2 s-1. The only blue-green alga in our experiments, *Phormidium molle*, grew in the whole temperature gradient, but had narow irradiance optimum of 42 μmol m-2 s-1.

Knowledge of optimum conditions will be used in subsequent laboratory anti-fouling tests, where marine coating samples will be submeged and exposed to the selected species. Then, strains with the best response will be further selected for the development of the IATS automatic test system.

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References: Hauer, T. & Jirka, L. (2007): Image analysis – a simple method of algal ulture growth assessment. – J. Appl. Phycol. 19: 599-601. Kvíderová, J. & Lukavský, J. (2001): A new unit for crossed gradients of temperature and light. – Nova Hedwigia Beiheft 123: 541-550.