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Wastewater treatment and biomass generation by Nordic microalgae

Growth in subarctic climate and microbial interactions

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Akademisk avhandling

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Avhandlingen kommer att försvaras på engelska.

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Title
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Abstract

Nordic native microalgal strains were isolated, genetically classified and tested for their ability to grow in municipal wastewater. Eight of the isolated strains could efficiently remove nitrogen and phosphate in less than two weeks. Two of these strains, *Coelastrella* sp. and *Chlorella vulgaris*, were found to have high biomass concentration and total lipid content; also two *Desmodesmus* sp. strains showed desirable traits for biofuel-feedstock, due to their fast growth rates and high oil content. The adaptation to subarctic climate was comparatively evaluated in three Nordic strains (*C. vulgaris*, *Scenedesmus* sp. and *Desmodesmus* sp.) and a collection strain (*S. obliquus*). Their growth performance, biomass composition and nutrients removal was investigated at standard (25°C) or low temperature (5°C), under continuous light at short photoperiod (3 h light, 25°C) or moderate winter conditions (6 h light, 15°C). Only the Nordic strains could grow and produce biomass at low temperature, and efficiently removed nitrogen and phosphate during both cold- and dark-stress. Phenotypic plasticity was observed in *Scenedesmus* and *Desmodesmus* under different growth conditions, adaptation to low temperature increased their carbohydrate content. Short photoperiod strongly reduced growth rates, biomass and storage compounds in all strains and induced flocculation in *C. vulgaris*, which, however, performed best under moderate winter conditions.

The symbiotic relationships between the Nordic microalga *C. vulgaris* and the naturally co-occurring bacterium *Rhizobium* sp. were investigated batchwise under photoautotrophic, heterotrophic and mixotrophic conditions, comparing the co-culture to the axenic cultures. The photoautotrophic algal growth in BG11 medium mainly supported *Rhizobium* activity in the co-culture, with no significant effects on *C. vulgaris*. In synthetic wastewater, a synergistic interaction only occurred under mixotrophic conditions, supported by CO_2/O_2 exchange and a lower pH in the culture, resulting in higher biomass and fatty acids content and more efficient wastewater treatment in the co-culture. Under heterotrophic conditions, the lower biomass production in the co-culture suggested a competition for nutrients, although nutrients removal remained efficient.

A pilot-scale high rate algal pond (HRAP) located in Northern Sweden was inoculated with the collection strain *Scenedesmus dimorphus* UTEX 417 and operated from spring to autumn. Using metabarcoding of 18S and 16S rRNA genes, the microbial diversity of eukaryotic and prokaryotic communities was revealed. *S. dimorphus* was initially stable in the culture, but other microalgal species later colonized the system, mainly due to parasitic infections and predation by zooplankton in summer. The main competitor algal species were *Desmodesmus*, *Pseudocharaciopsis*, *Chlorella*, *Characium* and *Oocystis*. *Proteobacteria*, *Firmicutes*, *Bacteroidetes* and *Actinobacteria* were the most abundant bacterial phyla in the HRAP. The structure of the microbial communities followed a seasonal variation and partially correlated to environmental factors such as light, temperature and nutrients concentrations.

Overall, these results contribute with new knowledge on the establishment and optimization of microalgal-based wastewater treatment systems coupled with biomass generation in Nordic areas. The use of native microalgal species is proposed as a potential strategy to overcome the limitations posed to algal cultivation in subarctic regions.

Keywords
Microalgae, Wastewater, Nitrogen, Phosphate, Biomass, Lipids, Subarctic Climate, Light, Temperature, Bacteria, Photoautotrophy, Heterotrophy, Mixotrophy, HRAP, Metabarcoding, Microbial Communities, Alpha-diversity, Environmental Factors.

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