

Original research paper

The influence of filamentous green algae on chlorococcal algae (*Desmodesmus* spp.)

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Abstract

The allelopathic influence of macroalgae has already been analysed both in marine and freshwater ecosystems. These organisms release allelopathically active compounds to prevent shading by epiphytic and planktonic primary producers. The aim of this study was to determine whether filamentous green algae represented by *Cladophora* sp. can exert an inhibiting effect on the growth of chlorococcal algae. Two coexisting culture systems differed in the species of *Desmodesmus* used, medium composition, light source and experiment duration. In both coexistence assays the abundance of microplankton in the controls was greater than in flasks with FGA, which seems to confirm an inhibiting effect by macroalgae on chlorococcal algae.

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INTRODUCTION

Allelopathy, as defined by Molisch (1937) as biochemical interactions among different primary producers is a well documented phenomenon in aquatic environments. Emerged and submerged macrophytes, algae and cyanobacteria may use active compounds to outcompete other species and gain dominance in their habitat. Allelochemical release has been confirmed for many common macrophytes e.g. *Ceratophyllum demersum*, *Elodea* spp., *Myriophyllum* spp., *Najas marina* and *Stratiotes aloides* (van Donk et al. 2002, Gross 2003, Mulderij et al. 2005). However, little is known about the impact of macroalgae on phytoplankton species. Nan et al. (2004) observed the allelopathic influence of *Ulva pertusa* on the growth of eight phytoplankton species, while Jin et al. (2005) reported the release of allelochemicals inhibiting *Prorocentrum micans* by *U. pertusa* and *U. linza*. As *Cladophora* sp. is a very common macroalgae in temperate freshwaters, there is a need to verify this type of ability for this species.

MATERIALS AND METHODS

Algal non-axenic assays were carried out using strains of two chlorococcal species, to analyze the effects of filamentous green algae on phytoplankton growth (culture system A and B). In coexistence culture system A – strain of *Desmodesmus quadricauda* was used, the pre-culture being incubated in a modified Chu-10 medium. Controls without FGA and FGA-*Desmodesmus* batches, all in triplicates, were prepared using the same growth medium. FGA's samples after washing in water were added as two different wet weights: 6.7 and 16.7 g l⁻¹. All of them were inoculated with algae at a cell density of 103 cells ml⁻¹. The experiment was conducted in natural light condition for 11 days. Microalgae growth was monitored by cell counts every two days.

In culture system B – the strain of *D. subspicatus* was cultured in a medium recommended by European standard ISO 8692: 2004. Test and control batches were inoculated with algae (about 75 × 10⁴ cells ml⁻¹) and maintained under a light intensity of 2000 lux for 5 days. The control and test flasks with FGA were also triplicated. Two different biomasses of *Cladophora* sp.: 5.0 and 16.6 g l⁻¹ were used. The cell density of microalgae in the samples was measured every day using an Olympus CX21 light microscope at a magnification of ×400. At the end of experiments the wet weight of FGA was also measured.

Fresh *Cladophora* sp. was collected from the shallow lowland Antoninek Reservoir (Poznań, Poland), in which it creates mats during the summer, while the abundance of phytoplankton remains low (Dondajewska et al. 2007). These

were used to analyse the possible allelopathic influence of FGA on chosen microalgae species.

RESULTS

Coexistence assay A showed that the abundance of *D. quadricauda* in the controls was significantly larger than in the flasks with FGA (Fig. 1). The number of cells decreased only at the beginning (on the third day), while on following days it increased to over 400×10^3 cells ml^{-1} on average by the end of experiment. The highest abundance of microalgae in the culture with FGA was 114×10^3 cells ml^{-1} for flasks with 6.7 g of FGA l^{-1} and 52×10^3 cells ml^{-1} for flasks with 16.7 g wet biomass of FGA l^{-1} . In all flasks with 6.7 g FGA l^{-1} and in two flasks with 16.7 g FGA l^{-1} a decrease was observed in the number of cells by the end of experiment. The wet weight of *Cladophora* sp. was on average slightly lower after the assay, especially in flasks with 6.7 g of FGA l^{-1} .

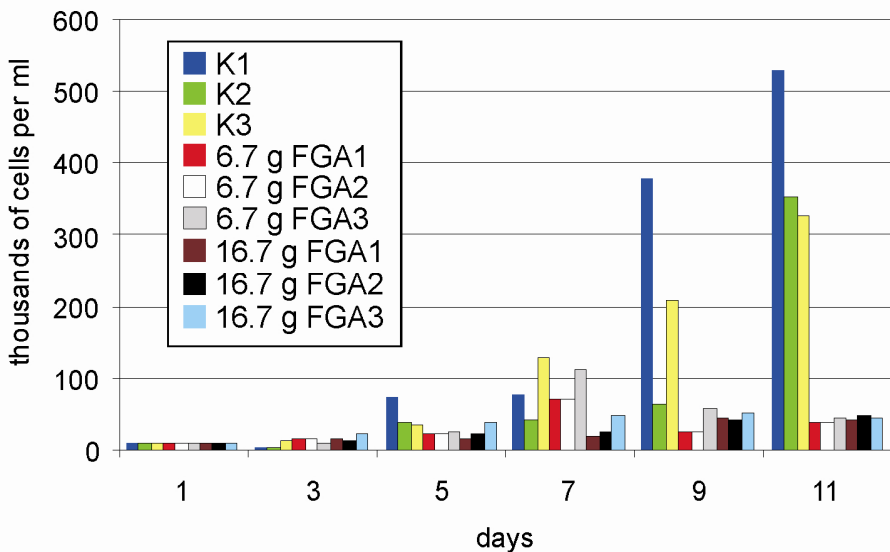


Fig. 1. The abundance of *Desmodesmus quadricauda* in flasks for coexistence assay A.

Coexistence assay B showed the abundance of *D. subspicatus* had decreased in all flasks by the second day of experiment, while from this time until the fifth day it increased progressively (Fig. 2). The number of microalgae cells in the controls was very much higher, reaching over 1000×10^3 cells ml^{-1} by the end of experiment, while in cultures with FGA the abundance was 4-5 times lower. The wet weight of *Cladophora* sp. decreased slightly (4.94 g l^{-1} on average) after the assay in flasks with 5.0 g l^{-1} of FGA and increased marginally (16.98 g l^{-1}) in flasks with 16.7 g l^{-1} .

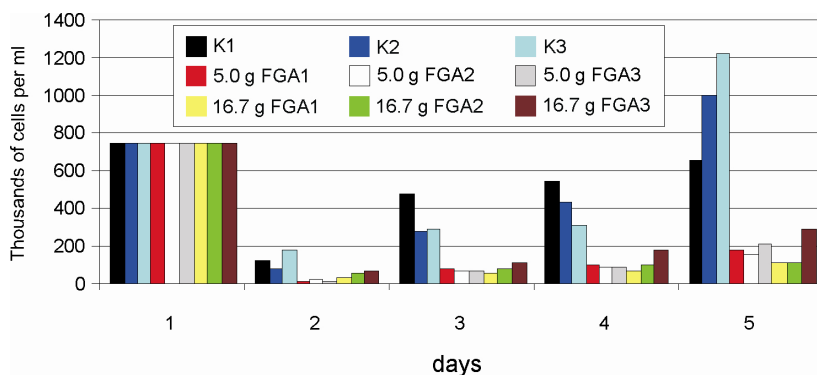


Fig. 2. The abundance of *Desmodesmus subspicatus* in flasks for coexistence assay B.

DISCUSSION

Both coexistence assay results showed that in the controls the abundance of microplankton was greater than in the flasks with FGA. Therefore, it seems to confirm a possible inhibiting effect of *Cladophora* sp. on chlorococcal algae *Desmodesmus* spp. Disturbing factors for the competition for light or space were minimized or excluded as far as possible by using flasks small enough to be equally permeated by light, but simultaneously sufficiently large in volume to create satisfactory conditions for both FGA and microalgae. Another important issue was potential nutrient competition, and nutrient concentrations were set at a non-limiting level by using a medium suitable for cultivating green algae species for the brief duration of the experiments. Nevertheless, nutrient competition can not be excluded completely from experimental conditions as both species actively absorb, metabolise and leak nutrients (Gross et al. 2007). Additionally, McGlathery et al. (1997) reported that filamentous macroalgae

can be a 'nitrogen sink' and inhibit the growth of phytoplankton by limiting the nitrogen concentration in water rather than allelopathically.

The wet biomass of FGA in cultures with FGA in both experiments was probably excessive and therefore caused slight differences between the abundance of *Desmodesmus* spp. in flasks with two different wet weight of FGA.

The coexistence experiments did not fulfil all of the Willis prerequisites to show the occurrence of allelopathy (Willis 1985) as none of available methods was able to achieve this (Gross et al. 2007). The assays proved that macroalgae can inhibit the growth of certain species of phytoplankton and that allelopathy is a co-factor in changes in algal abundance. Although the results are slightly imperfect due to competition for resources, this feature was minimized as far as possible for the conditions. Separation by chromatographic methods is necessary to identify the active compounds and their modes of action.

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