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Effects of a surfactant (FFD-6) on *Scenedesmus* morphology and growth under different nutrient conditions

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Abstract

Surfactants are man-made compounds that are meanwhile omnipresent in the environment, but environmental concentrations of surfactants are such that they are thought to have little risk for aquatic systems. The major anionic surfactants currently on the global market are linear alkylbenzene sulfonates (LAS), a class where the commercially available FFD-6 belongs to. The hypothesis was tested that sublethal effects of FFD-6, i.e. the morphological effect of colony formation in the common test alga *Scenedesmus obliquus*, occurs at a concentration lower than the no-observed-effect concentrations for endpoints commonly used in regulatory toxicity testing with algae. The surfactant FFD-6 induced colonies in *Scenedesmus* at concentrations a few orders of magnitude lower (i.e. between 0.001 and 0.01 $g\,l^{-1}$) than at which growth inhibition was observed (i.e. between 1 and 10 $g\,l^{-1}$). Growth rates were lowest for *Scenedesmus* grown in P-limited medium, intermediate for algae reared in N-limited medium and highest for algae cultured in non-limited standard medium. Growth inhibition due to FFD-6 was similar for non-limited and nutrient-limited *Scenedesmus*, but colony formation was stronger in non-limited *Scenedesmus* than in nutrient limited cultures. The colony inducing effect of the surfactant FFD-6 on *Scenedesmus* occurs at much lower concentrations than growth inhibition and might affect species interactions, the survival of species and the energy flow along the food chain.

Keywords: Algal toxicity testing; Colony formation; Detergents; Morphogenetic chemicals; Phenotypic plasticity

1. Introduction

One of the major global changes is the discharge of numerous anthropogenic compounds of which many end-up in aquatic ecosystems. Direct discharge to surface waters or via effluents of wastewater treatment plants has resulted in an omnipresence of surfactants in the aquatic environment. Surfactants are man-made compounds that arise mainly from the worldwide large volume consumption of commercial detergents and cleaning agents (Lewis, 1992). The widespread discharge has led to environmental concentrations of surfactants varying between 0.001 and 10 $mg\,l^{-1}$, but generally those are below 0.5 $mg\,l^{-1}$ (Kimerle, 1989; Rapaport and Eckhoff, 1990; Lewis, 1991; McAvoy et al., 1993; Temara et al., 2001). Although being worldwide present in aquatic environments, the environmental concentrations of surfactants are such that they are thought to

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have little risk for aquatic ecosystems (Fairchild et al., 1993; van den Plassche et al., 1997, 1999; Vandepitte and Feijtel, 2000).

Aquatic ecosystems can only exist when there is a continuous flow of energy along the food chains. In typical aquatic food chains, algae comprise an important component as these organisms convert solar energy into biomass. This algal material forms the potential energy that can be transferred to higher trophic levels. One of the most common, well-known and widespread green algae is *Scenedesmus obliquus*; being true cosmopolites they can be found in freshwater bodies all around the world (Trainor, 1998). *Scenedesmus* species are polymorphic, which means that depending on environmental conditions they can be unicellular or colonial (Trainor, 1998). Unicellular *Scenedesmus* are easily harvested by grazing zooplankton, but typical eight-celled colonies are considered grazing resistant (Hessen and Van Donk, 1993; Lürling and Van Donk, 1996).

Recently it was discovered that the commercially available surfactant FFD-6 (Skalar chemical) could transform unicellular *Scenedesmus* into colonial ones (Lürling and Beekman, 2002). The study of Lürling and Beekman (2002) focussed on membrane filter extractable compounds, but the morphological effect of the surfactant observed in that study suggest the surfactant could affect the energy flow from algae to higher trophic levels at concentrations previously thought to be safe. Hence, in the current study, the effect of the surfactant FFD-6 on the green alga *Scenedesmus* was examined further. The hypothesis was tested that the morphological effect of formation of colonies occurs at a concentration lower than the no-observed-effect concentrations for endpoints commonly used in regulatory toxicity testing with algae. This was achieved by determining and comparing EC_{50}- and NOEC-values of FFD-6 for algal biovolume-based growth, chlorophyll-based growth, photosystem II efficiency and colony formation. In addition, because in nature algae may be limited by either nitrogen (N) or phosphorus (P), and sensitivity to compounds of algae under nutrient-limitation may be greater than under nutrient-sufficient conditions (e.g. Hall et al., 1989; Lin et al., 1996), I included series with N- and P-limited *Scenedesmus*.

2. Materials and methods

2.1. Chemicals

The surfactant FFD-6 was obtained from Skalar BV (Breda, The Netherlands). The product (Skalar 00429040590) was delivered as a FFD-6 surfactant solution that consists of 55% water and 45% mono and didodecyl disulphanated diphenyloxide, sodium salt (Fig. 1). The compound was tested in the following concentrations of 0, 0.001, 0.01, 0.1, 1.0 and 10 g l^{-1}, without the use of additional solvents.

2.2. Algal culture

The green alga used in this study, *Scenedesmus* (Turpin) Kützing SAG 276/3a, originated from the culture collection of the University of Göttingen (Germany). This alga has been maintained for a few years in our laboratory in a 1.0 l chemostat system on a modified WC (Woods Hole modified CHU-10) medium (Lürling and Beekman, 1999). The chemostat was continuously illuminated at an irradiance of 100 μmol quanta m^{-2} s^{-1} provided by circular fluorescent tubes (Philips TLEM 40 W/33RS) in a temperature controlled chamber at 20 °C and at a dilution rate of 1.1 day^{-1}. Inocula were taken from the chemostat and subjected to the surfactant in batch systems.

2.3. Experimental protocol

An experiment was conducted to examine the effect of the surfactant FFD-6 on *Scenedesmus* growth and morphology under non-limiting and either N- or

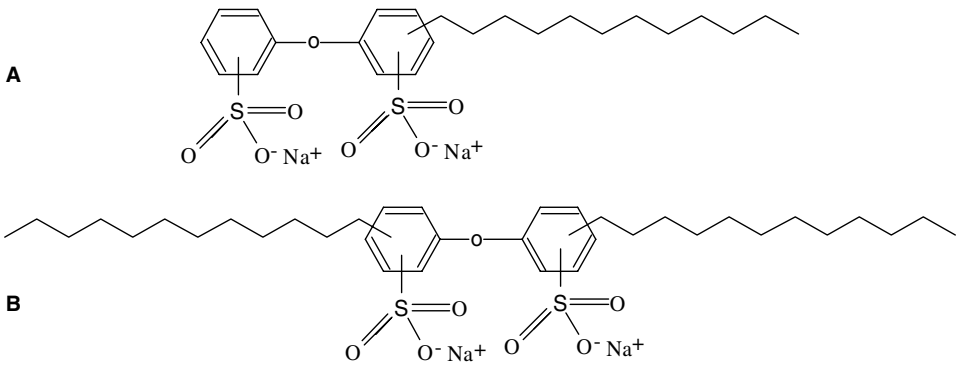

Fig. 1. Structure of anionic surfactant FFD-6, (A) monododecyl disulfonate diphenyloxide, (B) didodecyl disulfonate diphenyloxide.