

## Differential Response of Green Algal Species to Solvents

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Unicellular algae in aquatic ecosystems are subjected to a variety of pollutants from sources such as runoff from agricultural lands and industrial outfalls. Organic solvants are natural components of oil deposits and commonly find their way into surface waters as a result discharges from refineries, waste oil, disposal, and accidental spills. Organic solvents can make their way into the environment as industrial wastes. Because of their carcinogenic potential (Ward et al. 1986), contamination of soil and water by solvents is cause for serious concern. Relatively few reports have been published on the comparative toxicity of solvents toward test organisms, and these dealt primarily with fish and aquatic invertebrates (Bringmann and Kuhn 1959; Bringmann and Kuhn 1980; Bouman et al. 1981; LeBlanc and Suprenant 1983). However, limited data of toxicity effects of solvents on algae have been published (Pearson and McConnell 1975; Lay et al. 1984; Ward et al. 1986; Stratton 1987).

Algae have been considered to be good indicators of bioactivity of industrial wastes (Walsh et al. 1984) Unicellular algae vary in their response to a variety of toxicants (Hollister and Walsh 1973; Tadros et al. 1980). Little is known, however, about toxicity of solvents to freshwater unicelluar green algae.

The work reported here was done to examine the effect of selected solvents on unicelluar green algae species to determine whether they differed in their responses to these chemicals.

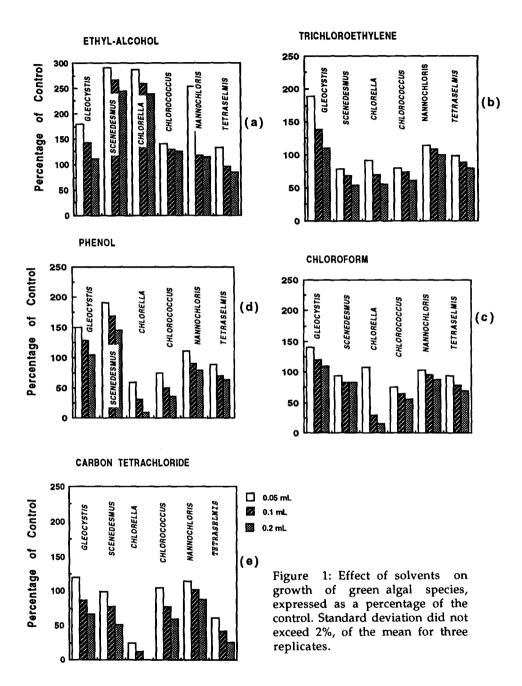
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## MATERIALS AND METHODS

The unicelluar green algal genera: Gleocystis ampla, Scenedesmus obliquus, Nannochloris sp., Tetraselmis sp., Chlorella ellipsoidea., and Chlorococcum sp., were obtained from the University of Texas Algal Collection (UTEX). The growth medium for the algal species was prepared according to Bold (1949). The pH of the growth medium was adjusted to 8.0 with dilute HCl or NaOH. The following chemicals were tested: ethanol, chloroform, carbon tetrachloride, trichloroethyl -ene and phenol. The chemicals were obtained from I.T.Baker Chemicals Co (Phillipsburg, New Jersey, USA). All test organisms were assayed in water-soluble fraction concentrations of 0.05, 0.1, 0.2 ml added to 100mL medium. Several higher concentrations were tested, but they caused bleaching of all tested organisms and therefore they were not considered in the assay. The 100 % solution was prepared by adding 1 ml of chemical to 100 mL water (volume to volume) and stirring in covered glass bottles with Teflon-coating-lined screw caps for 2 hr. After allowing the solution to settle for 1 hr the water-soluble fraction was siphoned into another container for distribution to the test containers. The assay was carried out in tubes containing 25 mL medium. All assays were conducted in triplicate test tubes. Each test tube was inoculated from exponential growing cells at an initial density of approximately 4 x 10<sup>3</sup> cells/mL. All cultures were incubated on a shaker for 96 hr at a temperature of 30 °C under cool-white light producing 100 uEm-2 s1 irradiation in continuous cycle. The growth was measured after 96 hr spectrophotometrically at 525 nm on a Fisher electrophotometer (Pittsburgh, Pennsylvania USA). All experi-ments were performed in a environmental chamber in the laboratory. Data in figures are the means and standard deviations from three independent experiments with triplicate cultures and triplicate samples within each individual experiment. The statistical significance of the data was estimated by means of a student's t test for p<0.05.

## RESULTS AND DISCUSSION

Results of the assays are presented in Figure 1. All data are expressed as percentage of the control levels. Algae treated with ethanol (Figure 1a) at three concentrations, the data show that all species were stimulated. Chlorella and Scenedesmus almost doubled in their growth with all alcohol concentrations. Nannochloris was stimulated at lowest concentration (0.05 %). Gleocystis, Chlorococcum and Tetraselmis were stimulated. In general, all species were not inhibited in media treated with ethyl alcohol.



Trichloroethylene stimulated Gleocyctis to almost double the control in a concentration of 0.05% (Figure 1b). Increasing the concentration of trichloroethylene, lowered the response, but still it was positive. Nannochloris and Tetraselmis tolerated the chemical in all concentrations while Scenedesmus Chlorella and Chlorococcum were inhibited, with increasing trichloroethylene concentrations. Chloroform in media stimulated Gleocystis at all concentrations (Figure 1c). Scenedesmus, Chlorococcum, Nannochloris and Tetraselmis tolerated the chemical. Chlorella, on the other hand, tolerated the chemical in lowest concentration of 0.05 but was inhibited in higher concentrations (0.1, 0.2 %). Gleocystis and Scenedesmus were stimulated with phenol in media treated with all concentrations (Figure 1d). Nannochloris and Tetraselmis tolerated the chemical at lowest concentration (0.05%). However, Chlorella and Chlorococcus were inhibited by phenol at all concentrations. Nannochloris tolerated carbon tetrachloride at all concentrations (Figure 1e). However, Gleocystis, Scenedesmus and Chlorococcus tolerated the chemical in concentration of 0.05 and started to be inhibited in higher concentrations (0.1 %, 0.2 %). The results were statistically different from the control. Chlorella was very sensitive to carbon tetrachloride at all concentrations.

In this work, wide variations occurred in response to the chemicals among individual species of the green algae. Gleocystis, Scenedesmus, Nannochloris and Tetraselmis proved to be the most tolerant genera while, Chlorococcus and Chlorella were the most sensitive to chemicals. In terms of solvents, carbon tetrachloride and phenol were more inhibitory than the other compounds. Investigations using different algal species as test organisms have shown that algae vary greatly in their response to chemicals (Walsh and Merrill 1984; Tadros et al. 1990). Differential sensitivity of the green species to the compounds could induce species shifts within communities (Brigmann and Kuhn 1980; Brand et al. 1986; Genter et al. 1987).

The data presented suggest that when bioassays are conducted to determine the effect of solvents on algal species, one should consider several species to obtain realistic data (Holister and Walsh 1973). Applying different organisms would provide a broader basis for assessing the damaging action of water pollutants. Ecotoxicological testing of potential water pollutants to evaluate their toxicity involving only one organism would give an incomplete and biased picture of the effects of pollutants.

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