

## *Microcystis aeruginosa* as an organohalogen precursor

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### Abstract

*Microcystis aeruginosa*, the most common algal bloom former in South Africa was found to be a prolific organohalogen precursor agent. Precursors (extracellular products) are produced by the alga during its growth cycle and upon the death and lysis of the algal cells. In the absence of phosphate, a laboratory culture of *M. aeruginosa* ceased to grow and died after eight days. Relative to algal cultures containing 60 and 600  $\mu\text{g}/\ell$  phosphate, the organohalogen concentration in the zero phosphate culture increased a hundredfold after eight days.

### Introduction

The development of large amounts of algae, especially cyanobacteria (or blue-green algae), in lakes or impoundments as a result of excessive phosphorus loading or eutrophication is well known (e.g. Toerien, 1977; OECD, 1982). *Microcystis aeruginosa* is the most common algal bloom former in South Africa and is of considerable importance in local eutrophication problems. The Hartbeespoort Dam situated 60 km west of Pretoria is a typical example. The dam is hypertrophic and phytoplankton populations are dominated by *M. aeruginosa* (Scott *et al.*, 1980; Robarts, 1984; Zohary, 1985). Precursor values ranging between 670 and 2 300  $\mu\text{g}/\ell$  have been reported for this dam (Tenth Annual Health Report, 1985). Treated water from this dam is used as a drinking-water supply by a number of municipalities. A considerable amount of literature on water chlorination and its effect on the formation of organohalogen compounds has been published (Jolley, 1978; Jolley *et al.*, 1978; 1980; 1983; Van Steenderen *et al.*, 1983; 1984; 1987). However, comparatively little has been published concerning the role of algae or algal extracellular products (Fogg, 1966; 1971) as organohalogen precursors (Hoehn *et al.*, 1978; 1980; Morris and Baum, 1978; Briley *et al.*, 1980; Oliver and Shindler, 1980; Bernhardt, 1982; Wachter and Andelman, 1984).

Two morphological forms of *M. aeruginosa* have been distinguished in field material from the Transvaal (Scott, 1974). Irregularly shaped colonies with a net-like appearance containing individual cells with a diameter of 4 to 6  $\mu\text{m}$  are known as *M. aeruginosa* forma *aeruginosa*, while compact spherical or lens-shaped colonies containing individual cells with a diameter of 2,5 to 5,5  $\mu\text{m}$  are known as *M. aeruginosa* forma *flos-aquae* (Komárek, 1958).

In relation to South African legislation that by 1985 effluent discharge to rivers in specified sensitive areas should comply with a 1  $\text{mg } \ell^{-1}$  phosphate (as P) standard, the role that *M. aeruginosa* could play as a source of organohalogen precursors, when using phosphate as a limiting nutrient, was investigated.

### Experimental procedures

A uni-algal NIWR laboratory culture of *Microcystis aeruginosa*, strain WR133 (Scott, 1986), originating from the Hartbeespoort Dam, was used for this study. The culture morphologically resembled *M. aeruginosa* forma *flos-aquae*. Cultures were grown at 30°C in five litres modified Volk and Phinney's culture medium (Scott *et al.*, 1981) under a light : dark regime of 16:8 at three levels of added phosphate viz. 600  $\mu\text{g } \text{P}/\ell$ , 60  $\mu\text{g } \text{P}/\ell$  and no added P. Growth of these cultures was monitored as chlorophyll *a*.

At selected intervals over a period of 14 d, as sample was collected from each algal culture, filtered, and then chlorinated with enough NaOCl to contain a residual free chlorine concentration of 10  $\text{mg}/\ell$  after 24 h. The medium was maintained at pH 9. Chlorophyll *a*, dissolved organic carbon (DOC) and total organohalogen (TOH) concentrations were measured according to methods described by Zohary (1985), Van Steenderen and Lin (1981) and Van Steenderen (1980) respectively. Background concentrations for these three determinands in the algal-free cultures were monitored and incorporated in the final calculations.

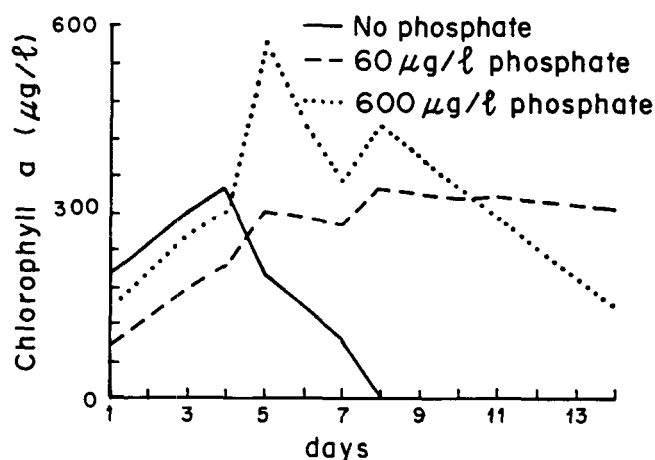


Figure 1  
Changes in chlorophyll *a* values in three cultures of *M. aeruginosa* containing phosphate concentrations of 0, 60 and 600  $\mu\text{g}/\ell$  respectively.

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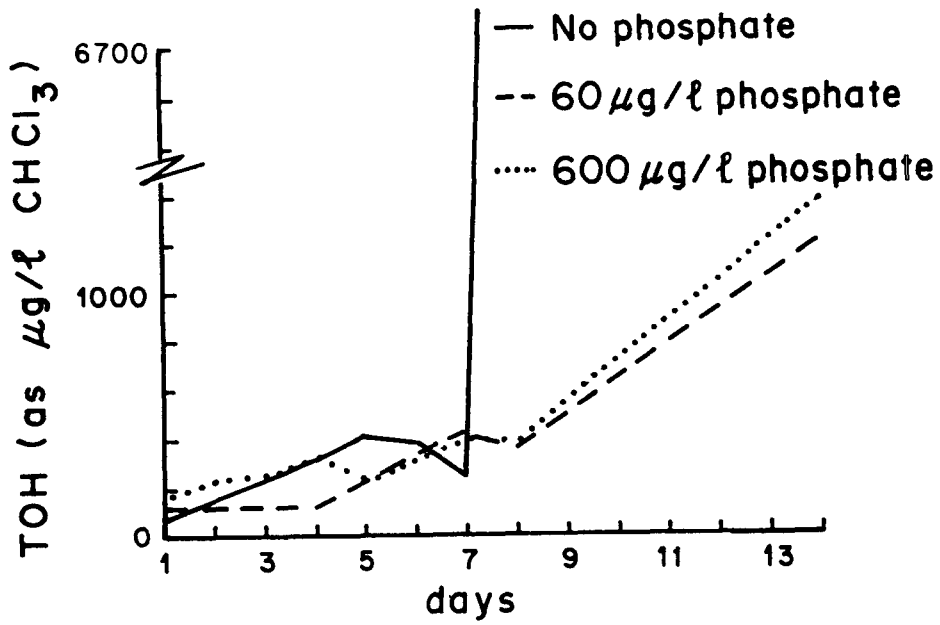


Figure 2  
Changes in organohalogen precursor values in three cultures of *M. aeruginosa* containing phosphate concentrations of 0, 60 and 600 µg/l respectively.

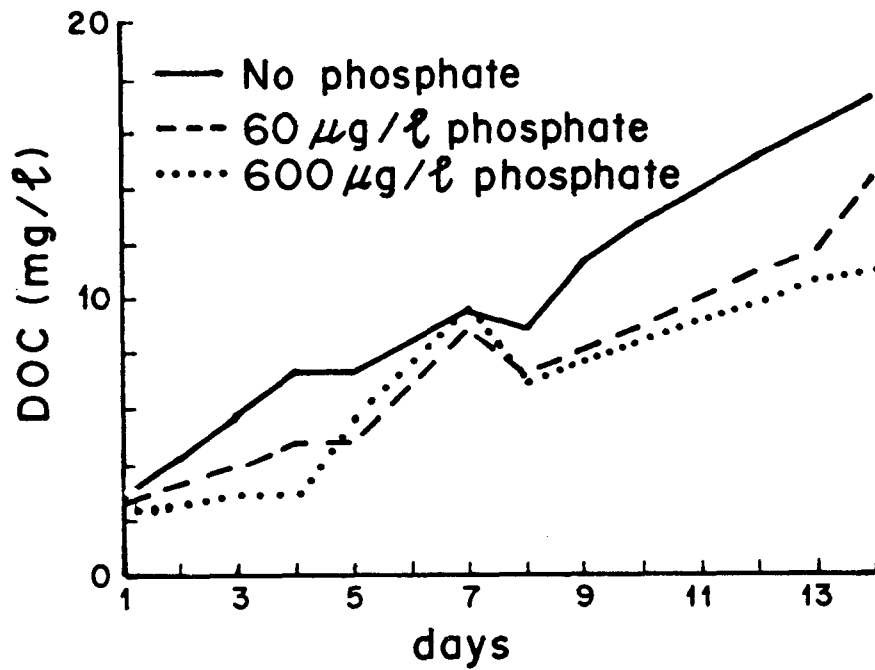


Figure 3  
Changes in dissolved organic carbon values in three cultures of *M. aeruginosa* containing phosphate concentrations of 0, 60 and 600 µg/l respectively.

## Results

Maximum growth occurred in the 600 µg/l phosphate culture where the chlorophyll *a* concentration reached a value of 600 µg/l. (Fig. 1) After the eighth day both phosphate-containing cultures showed negative growth (Fig. 1). The cultures were not bacteria-free and were regularly examined for contamination under phase contrast microscopy. According to visual estimates the bacterial biomass was never more than 1% of the algal biomass in the growing cultures. It was noticed that on the seventh day the 600 µg/l culture had a larger infection of bacteria (bacterial biomass approximately 5% of the algal biomass). Additional contamination may have been introduced during subsampling. The other two cultures did not appear to suffer heavily from bacterial infection. In the culture containing no phosphate, the growth reached a maximum after four days after which negative growth occurred until the algal culture died on the eighth day. (Fig. 1).

The death of the algae in this culture was accompanied by a simultaneous release of organohalogen precursors (Fig. 2). From the eighth day the precursor concentrations in both phosphate-containing cultures increased steadily and at approximately the same rate (Fig. 2). The dissolved organic carbon (DOC) concentration in all three cultures maintained a steady increase throughout the experiment although the production of DOC in the culture containing no phosphate was higher at every stage of the experiment relative to the phosphate-containing cultures. Relative to the first eight days the rate of DOC release in this culture increased over the last six days (Fig. 3).

## Discussion

Organohalogen compounds are formed when chlorine reacts with organic precursor molecules which in most instances are the aquatic humic substances derived from vegetal decay (Stevens *et al.*, 1976). According to Black and Christman (1963) approximately eighty five per cent of dissolved organics in a natural environment consists of aquatic humic substances. It has, however, recently been shown that algae and their extracellular products (ECP), upon chlorination, produce trihalomethane (THM) levels comparable to those obtained from the chlorination of humic material (Briley *et al.*, 1980). Oliver and Shindler (1980) actually fractionated algal components and found that the component which was the most active THM precursor was chlorophyll *a*. Hoehn *et al.* (1980) furthermore concluded that, in general, algal ECP's yielded even greater quantities of trihalomethanes upon chlorination than did the algal biomass.

The prolific algal growth in the Hartbeespoort Dam and many other water impoundments in South Africa is a continuous source of problems to water authorities. These problems are often 'seemingly' solved by prechlorination practice or by the inclusion of a more recently reinstated process, namely that of dissolved air flotation (DAF) whereby algae are removed – prior to conventional treatment.

Research has established that the physiological state of algae is determined by the interaction of environmental factors such as available free carbon dioxide, light intensity, temperature, nutrients, pH and dissolved organic carbon (see for example Platt, 1981). It can therefore be accepted that as these conditions and the physiological state of the algae vary, the TOH precursor value of the algal extracellular products (ECP) will differ. In this experiment, limiting the phosphate concentration was shown to

be an additional factor giving rise to the production of TOH precursors.

## Conclusions

*Microcystis aeruginosa*, the most common algal bloom former in South Africa was found to be a prolific organohalogen precursor agent. Upon the death and lysis of these algae even greater amounts of organohalogen precursors were released into the water. Physical removal of *M. aeruginosa* prior to water purification is essential especially if prechlorination is practiced and organohalogen precursor formation is to be limited. The term limited is used here since it has been shown that in addition to the algae, dissolved algal extracellular products can yield even greater quantities of organo-chloro compounds upon chlorination than the algal biomass itself.

## Acknowledgement

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