Preliminary observations on the effect of increased concentrations of total dissolved salts on growth and photosynthetic rates in different algal species

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Abstract

The effect of increased Vaal River total dissolved salts (TDS) concentrations was investigated on algal growth and photosynthetic rates of 3 different algal species i.e. *Cyclotella meneghiniana* Kütz., *Monoraphidium circinale* (Nyg.) Nyg. and *Microcystis aeruginosa* Kütz. The algal species were introduced to different Vaal River salt concentrations in a GBG-11 growth medium for a period of 14 d. The salt concentrations varied between 100 mg·t^{-1} and 2 000 mg·t^{-1}. Turbidity was used as a measure of growth. On day 8 of the experiment, water samples were taken and chlorophyll-*a* was measured as well as primary productivity by the ^{14}C-uptake method (acid-bubbling). With increased TDS concentrations, the growth of *C. meneghiniana* and *M. aeruginosa* was inhibited, but the growth of *M. circinale* was unaffected. With increasing salinity concentrations the chlorophyll-*a* concentration in *C. meneghiniana*, *M. aeruginosa* and *M. circinale* increased up to salinities of 250 mg·t^{-1} and then decreased at salinities of between 500 mg·t^{-1} and 2 000 mg·t^{-1}. The carbon assimilation rate of *C. meneghiniana* and *M. aeruginosa* increased with an increase in TDS concentration, and that of *M. circinale* was very low at salinities of 250 mg·t^{-1} and above. These results show that if the salt concentration of the river increases with time, algal species that can adapt to a much wider range of salinities may become dominant, e.g. *M. circinale*. *C. meneghiniana* and *M. aeruginosa*, on the other hand, can be expected to be eliminated from the water under conditions of increased salinity.

Introduction

Salinity is a growing problem in freshwater ecosystems in many parts of the world. On a world-wide basis, an area of about 950m. ha is affected by salt. This is apparently due to dryland salinity as well as salinity in irrigation regions (Hart et al., 1990). According to Hart and co-workers, the main concern is the increase in salinity caused by human disturbances such as agricultural practices, pollution and other human activities. This kind of salinisation is often referred to as secondary salinisation.

Rivers or lotic systems differ from lakes and wetlands in that movement of the water has an effect on plant and animal life. The increase in turbulence increases the input and the breakdown of organic matter which also influences life in the water. In addition, a river is an open system which receives allogenic substances through precipitation and rainfall as well as from streams in its catchment area (Wetzel, 1983). Parts of the Vaal River, which drain densely populated areas, i.e. the Witwatersrand and West Rand mining regions, are also affected by human intervention which contributes to an increase in allogenic substances (Stander et al., 1962).

According to Brock (1985) high salinity conditions could affect autotrophic macrophytic communities in freshwater environments in the following ways. As salinity levels increase, the diversity of macrophyte species declines. At salinities of approximately 4 000 mg·t^{-1} TDS most freshwater macrophytes will be replaced by halophytic macrophytes which can persist over a much wider range of salinities (Brock, 1981). A similar pattern of change can be expected to occur in algal communities as a result of increased salinity, an aspect that should, at this stage, only be regarded as speculative because there is apparently no information available in the literature on the response of algal communities to increased salinities in running waters. In this preliminary study the aim was to investigate the effect of an increase in total dissolved salts on freshwater algal species in order to provide specific information in this regard.

The Vaal River is one of the most important rivers in South Africa because of economic activities and the concentration of the human population in its catchment area (Oliveira, 1986). In addition to being eutrophied and polluted, the Vaal River is also salinised and mineralised because of the extensive utilisation of the water through household, mining and industrial activities as well as agricultural practices (Triebel, 1986).

Because of the increased TDS load, the water in the Vaal River will probably become clearer (Grobler *et al.*, 1986; 1987). Clearer water due to salinisation and the high nutrient supply (eutrophication), will probably result in more intensive algal blooms. In some cases the blooms may be by algal species that have not, until now, caused problems (Pieterse, 1986). One possibility is the development of dinoflagellate blooms which could cause phenomena similar to red-tides in the ocean and should be considered as potentially toxic (Pieterse, 1986). However, recently a strategy was developed by the South African Department of Water Affairs and Forestry to control the dissolved salts concentration in the middle reaches of the river by releasing water from the Vaal Dam into the Vaal River Barrage (Van Vliet, 1993).

Temporal changes in phytoplankton composition occur in the Vaal River (Pieterse, 1986). It has been found that a *Sphaerodinium* species (a dinoflagellate) becomes dominant in the Vaal River when TDS concentrations increase to levels above approximately 1 000 mg·t^{-1} (Pieterse, unpublished information).

Material and methods

Uni-algal cultures from the algal culture collection of the Department of Botany and Genetics, University of the Orange Free State, were used as experimental material, namely *C. meneghiniana*

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Kütz. (diatom), *M. circinale* (Nyg.) Nyg. (green alga) and *M. aeruginosa* Kütz. (blue-green alga). *Cyclotella meneghiniana* was isolated from the Vaal River (by MC Steynberg, Rand Water, Vereeniging), *M. aeruginosa* from the Hartbeespoortdam (by W Scott, now at the Dept. of Environment Affairs, Pretoria) and *M. circinale* from Lake Kinneret (by B Kaplan, Kinneret Limnological Laboratory, Israel). All 3 of these species occur in the Vaal River (Pieterse, unpublished information).

Uni-algal stock cultures were made of these 3 algal species and were kept in a light chamber prior to each experiment. The cultures were grown in 100 m*l* GBG-11 growth medium (Krüger, 1978) in 250 m*l* Ehrlenmeyer flasks equipped with a side-arm and stoppered with a cellulose stopper.

An inoculum of 1 m*l* was transferred, under sterile conditions, from each stock culture to the experimental and control flasks with side-arms at the beginning of each experiment.

The effect of TDS concentration on growth and photosynthetic rates was investigated by adding different concentrations of a natural salts mix prepared from Vaal River water. The natural salts mix was prepared in the following way: Water from the Vaal River was evaporated at 80°C for 24 h. The remaining salts were used as follows: Distilled water was used to dilute 4 g of the salts to 1 *l* to give a concentration of 4 000 mg·t^{-1} mixed salts. The different experimental concentrations (100 to 2 000 mg·t^{-1}; see Table 1) were then prepared from the salts mix solution in combination with the nutrient medium. The different volumes of nutrient medium used are given in Table 1.

TABLE 1
THE PREPARATION OF DIFFERENT EXPERIMENTAL TDS CONCENTRATIONS

Treatment (mg/*l*)	Salts mix (m*l*)	GBG-11 (m*l*)
0 (control)	-	100
100	2.5	97.5
250	6.25	93.75
500	12.5	87.5
1 000	25	75
1 500	37.5	62.5
2 000	50	50

After an inoculum of 1 m*l* of each species had been transferred to experimental flasks with side-arms, the cultures were placed in a light chamber at a temperature of 23°C and at a light intensity of 60 µE·m^{-1} ·s^{-1} for a period of 14 d. Turbidity (in Klett units) was used as a measure of biomass and growth. Side-arms of the Ehrlenmeyer flasks are suitable for fitting into a Klett-Summerson photoelectric colorimeter equipped with a 540 nm green filter for the determination of turbidity.

On day 8 of the experiment subsamples were taken from the flasks and chlorophyll-*a* was measured by using a pigment extraction method described by Sartory (1982). Carbon assimilation rates were also determined on day 8 by using the acid-bubbling method as described by Schindler *et al.* (1972). A 4 m*l* suspension of each experimental flask was placed in 20 m*l* glass scintillation vials. Acid-bubbling was done on each vial according to the method of Schindler and co-workers, and thereafter 0.5 m*l* Carbosorb and 10 m*l* scintilla ion liquid were added. The samples were then counted in a LKB 1217 liquid scintillation counter and counts were automatically corrected for quenching. Carbon assimilation for each sample was calculated using the following formula:

$$P = \frac{A \times C \times D \times E}{B \times T}$$ (Vollenweider, 1969)

where:

P = carbon assimilated
A = ^{14}C assimilated (DPM - DPM background ~ 100)
B = ^{14}C available (DPM/m*l* x sample volume filtered)
C = ^{12}C available (mg C/*l*)
D = isotope factor of 1.06
E = 1 000
T = incubation time (h).

Results and discussion

Figure 1 shows the effect of increased TDS concentrations on the growth (in Klett units) of *M. aeruginosa*. At concentrations of 0 to 250 mg·t^{-1} the growth of *M. aeruginosa* was stimulated (higher Klett units were recorded at these concentrations than at the control) and at concentrations above 250 mg·t^{-1} the growth of *M. aeruginosa* was lower than in the control. These observations are in support of information given by Wetzel (1983) that most freshwater bacteria and blue-green algae (blue-green bacteria) are relatively homoiosmotic, tolerating a narrow range of salinity. Wetzel (1983) also indicated that bacteria and blue-green algae could adapt to increasing salinity by means of genetic change.

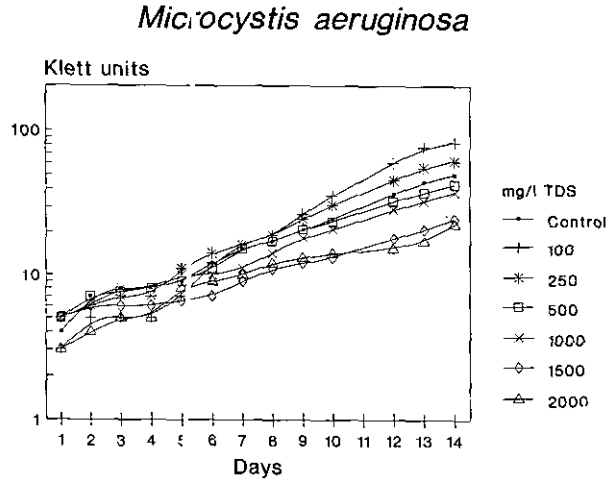

Figure 1
The effect of increased TDS concentration on the growth of Microcystis aeruginosa *over a 14 d period*

It is also possible that at concentrations of 100 to 250 mg·t^{-1} salts added, the growth of *M. aeruginosa* was stimulated by the nutrients added. At concentrations above 250 mg·t^{-1} salts, high salt concentration most probably inhibited growth despite the fact that additional nutrients were added.

Figure 2 shows the effect of increased TDS concentration on the growth of *M. circinale*. With an increase in TDS concen-

tration, the growth of this species was neither stimulated nor inhibited. The apparent lack of effect of dissolved salts within the range investigated could be attributed to the fact that *M. circinale* was isolated from Lake Kinneret in Israel. The TDS concentration in Lake Kinneret is approximately 1 300 meq·t^{-1} (Serruya, 1978) while the Vaal River has an average TDS concentration of approximately 16 meq·t^{-1} (Roos, 1991). According to Wetzel (1983) most algal species in aquatic systems in semi-arid and arid regions, like Lake Kinneret, are adapted genetically to persist over a wider range of salinities.

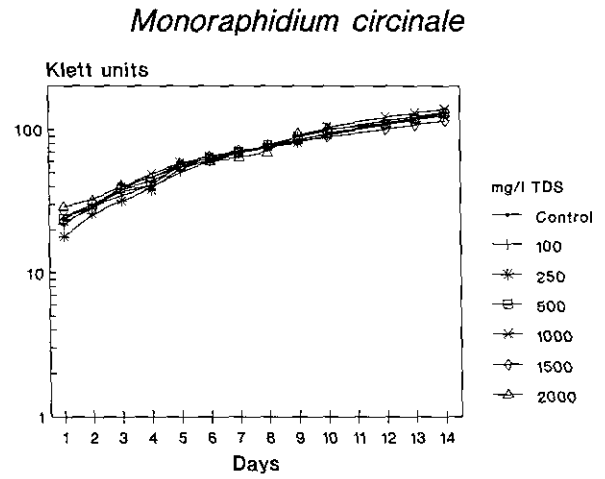

Figure 2
The effect of increased TDS concentration on the growth of Monoraphidium circinale *over a 14 d period*

The growth of *C. meneghiniana* (Fig. 3) was apparently stimulated after 8 d at a concentration of 100 mg·t^{-1}. At salinity concentrations of between 250 mg·t^{-1} and 2 000 mg·t^{-1}, no growth occurred after 2 to 5 d indicating that *C. meneghiniana* is sensitive to dissolved salts concentration. The observed sensitivity corresponds to Kolbe's (1932) classification of *C. meneghiniana* as an oligohalobien species with optimum growth under conditions of low salt concentration. The low growth rate shown for *C. meneghiniana* could also be attributed to the temperature (23°C) at which the cells were incubated. According to Patrick (1971) the development of large populations of a given diatom species is clearly correlated with temperature. Although the effect of temperature on the growth of *C. meneghiniana* from the Vaal River has to be determined, it is possible that *C. meneghiniana* is adapted to conditions cooler than 23°C for optimal growth.

Figure 4 shows the chlorophyll-*a* concentrations in *C. meneghiniana, M. aeruginosa* and *M. circinale* cultures after 8 d of growth. With an increase in salinity the chlorophyll-*a* concentrations in *C. meneghiniana, M. aeruginosa* and *M. circinale* cultures increased at TDS concentrations up to 250 mg·t^{-1} and then decreased at the higher concentrations. *Monoraphidium circinale* had the highest chlorophyll-*a* concentrations of the 3 species investigated.

Figure 5 shows that the carbon assimilation rate of *C. meneghiniana* decreased with an increase in TDS concentration in accordance with the growth results illustrated in Fig. 1. The carbon assimilation rate of *M. circinale* was very low at TDS concentrations of 250 mg·t^{-1} and above, while TDS showed no effect on the growth of this species (Fig. 3). The carbon assimila-

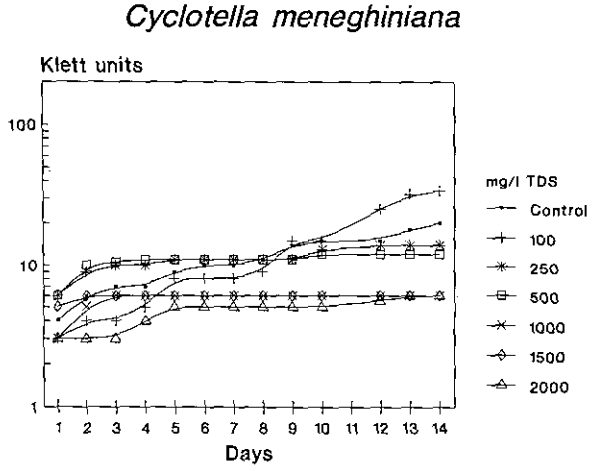

Figure 3
The effect of increased TDS concentration on the growth of Cyclotella meneghiniana *over a 14 d period*

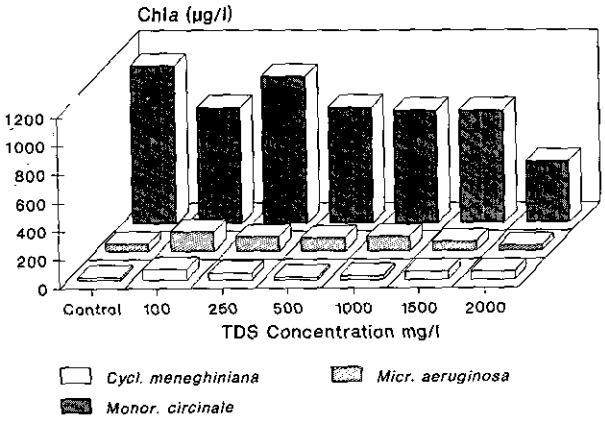

Figure 4
Chlorophyll-a concentrations in Cyclotella meneghiniana, Microcystis aeruginosa *and* Monoraphidium circinale *cultures in µg·t^{-1} as determined after 8 d*

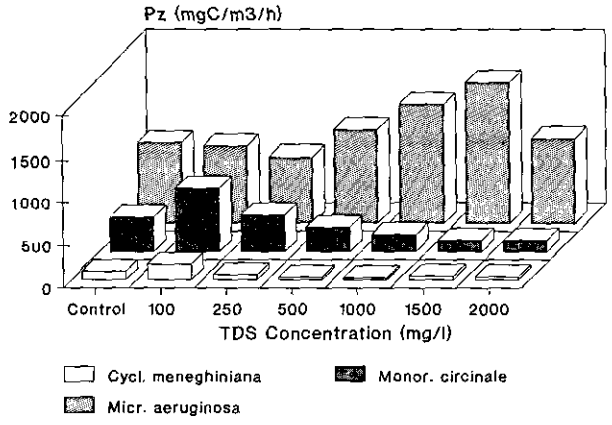

Figure 5
Carbon assimilation by Cyclotella meneghiniana, Microcystis aeruginosa *and* Monoraphidium circinale *in mg C·mg Chl* $a^{-1}·h^{-1}$.

tion of *M. aeruginosa* generally increased with an increase in the concentration of TDS (Fig. 5) in contrast to the effect that TDS had on the growth of this species (Fig. 2). The maximum photosynthetic rate measured in *C. meneghiniana* was at a concentration of a 100 $mg \cdot l^{-1}$ TDS (182 mg $C \cdot m^{-3} \cdot h^{-1}$); it then decreases with increasing TDS concentration. *Monoraphidium circinale* also showed a decrease in photosynthetic rate with an increase in salinity. In *M. circinale* a maximum photosynthetic rate of 742 mg $C \cdot m^{-3} \cdot h^{-1}$ was measured at a concentration of a 100 $mg \cdot l^{-1}$. The maximum rate of carbon-assimilation in *M. aeruginosa* was at 1 500 $mg \cdot l^{-1}$ (1 629 mg $C \cdot m^{-3} \cdot h^{-1}$).

According to Roos and Pieterse (1992) the maximum photosynthetic rate in the Vaal River, measured by the ^{14}C method after a 4 h incubation period, was 285 mg $C \cdot m^{-3} \cdot h^{-1}$. In both *M. aeruginosa* and *M. circinale* the average rate of carbon assimilation in culture was much higher than in the river.

Conclusions

The different algae showed different sensitivities to TDS. Of the 3 algae investigated, *C. meneghiniana* was the most sensitive and *M. circinale* the least sensitive to increased dissolved salts.

If the salt concentration of Vaal River water increases with time, algal species that can persist over a much wider range of salinities could become dominant. Of the species investigated, *M. circinale* can be expected to become dominant under conditions of increased salinity. *Cyclotella meneghiniana* and *M. aeruginosa* on the other hand, can be expected to be excluded from the water under conditions of increased salinity above 250 $mg \cdot l^{-1}$.

These preliminary experiments must be regarded as a basic guideline for more intensive investigations to clarify aspects of the effect of salinity on the growth and photosynthetic rates of algal species in rivers and especially the Vaal River.

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References

BROCK, MA (1981) The ecology of halophytes in the south-east of South Australia. *Hydrobiol.* **81** 23-32.

BROCK, MA (1985) Are Australian salt lake ecosystems different? Evidence from the submerged aquatic plant communities. *Proc. Ecol. Soc. Aust.* **14** 43-50.

GROBLER, DC, TOERIEN, D F and ROSSOUW, JW (1986) Effects of sediment on water quality in the Vaal River System. In: *Proc. Joint Symp. Vaal River Ecosystem: Status and Problems.* Convened by the FRD and the VRCA and held at the CSIR, Pretoria, March 20 1986. Occasional report Nr.5, FRD, CSIR, Pretoria: 115-148.

GROBLER, DC, TOERIEN, DF and ROSSOUW, JW (1987) A review of sediment water quality interaction with particular reference to the Vaal River system. *Water SA* **13** (1) 15-22.

HART, BT, BAILY P, EDWARDS, R, HORTLE, K, JAMES, K, Mc MAHON, A, MEREDITH, C and SWADLING, K (1991) Effects of salinity on river, stream and wetland ecosystems in Victoria, Australia. *Water Res.* **24** 1103-1117.

KOLBE, RW (1932) Grundlinien einer allgemeinen Ökologie der Diatomeen. *Ergebn. Biol. Berlin.* **8** 221-348.

KRÜGER, GHJ (1978) The Effect of Physico-chemical Factors on Growth Relevant to the Mass Culture of *Microcystis* under Sterile Conditions. Unpublished Ph.D. thesis, UOFS, Bloemfontein.

OLIVEIRA, MP (1986) Sources of pollution in the Vaal River.In: *Proc. Joint Symp. Vaal River Ecosystem: Status and Problems.* Convened by the FRD and the VRCA and held at the CSIR, Pretoria. March 20,1986. Occasional report nr. 5, FRD, CSIR, Pretoria:32-45.

PATRICK, R (1971) The effect of increasing light and temperature on the structure of diatom communities. *Limnol. Oceanogr.* **16** 405-421.

PIETERSE, AJH (1986) Aspects of the ecology and the significance of algal populations of the Vaal River. In: *Proc. Joint Symp. Vaal River Ecosystem: Status and Problems.* Convened by the FRD and VRCA and held at the CSIR, Pretoria. March 20, 1986. Occasional Report nr. 5, FRD, CSIR, Pretoria.

ROOS, JC (1991) Primary Productivity of the Vaal River Phytoplankton. Unpublished Ph.D. thesis, UOFS, Bloemfontein.

ROOS, JC AND PIETERSE, AJH (1992) Diurnal variations in the Vaal, a turbid South African river: primary productivity and community metabolism. *Arch. Hydrobiol.* **124** 459-473.

SARTORY, DP (1982 Spectrophotometric Analysis of Chlorophyll *a* in Freshwater Phytoplankton. Technical report TR 115, Dept. of Environm. Affairs, HRI, Pretoria.

SCINDLER, DW, SCHMIDT, RV and REID, RA (1972) Acidification and bubbling as an alternative to filtration in determining phytoplankton production by the ^{14}C method. *J. Fish. Res.Bd. Canada* **29** 1627-1631.

STANDER, GJ, MALAN, WC and HENZEN, MR (1962) The Chemical Quality of the Water of the Lower Vaal River between the Barrage and Kimberley. CSIR, Special Report **No. 14** 1-14.

SERRUYA, C (1978) *Lake Kinneret* (Lake of Tiberias, Sea of Galilee). Dr W Junk Publishers. The Hague, Boston, London.

TRIEBEL, C (1986) Sources of pollution in the Vaal River. In: *Proc. Joint Symp. Vaal River Ecosystem: Status and Problems.* Convened by the FRD and the VRCA and held at the CSIR, Pretoria. March 20,1986. Occasional report Nr. 5, FRD, CSIR, Pretoria.

VAN VLIET, H (1993) Personal communication. Director, HRI, Pretoria.

VOLLENWEIDER, RA (1969) Primary Production in Aquatic Environments. IBP 12, Blackwell Scientific Publications, Oxford, 41-127.

WETZEL, RG (1983 *Limnology* (2nd edn.) Saunders College Publishing, Philadelphia.