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MERCURY TOXICITY ASSESSED USING ANABAENA FLOS-AQUAE, SCENEDESMUS QUADRICAUDA AND NILE WATER PHYTOPLANKTON

Sabah A. Badr and Azza M. Ashmawy
Water Pollution Control Dept.
NRC. Dokki, Cairo, Egypt.

ABSTRACT

Algal bioassays were performed to evaluate the toxic effect of Mercury on Anabaena flos-aquae, Scenedsmus quadricauda and Nile water algae. the results showed that the inhibitory effect of Mercury increased in the following order:

Scenedesmus > Anabaena > Nile water algae

Clear changes in domenancy and diversity of natural phytoplankton took place and Cyanobacteria were the most tolerant group. Statistical analysis showed a highly significant relationship between mercury concentration and algal growth. Mercury is highly accumulated in Nile water algae.

INTRODUCTION

Mercury is the most toxic metals in the non-essential group. Mercury compounds are used in electrical equipment, paints, thermometers, fungicides, as preservatives in pharmaceuticals and cosmetics (Berlin 1979). In, Egypt chlorolkali plants in particular are notorious sources of mercury pollution. One example is plant located in the surrounding complex at El-Max, west of Alexandria, Egypt (El-sokkary, 1989). In El max Bay, the average Hg concentrations were found to be 34±12 ng/L in dissolved from, 70±25 ng/L in particular form to be 20±12 μg/g in sediments. The objectives of this study are: (1) To assess the toxic effects of mercury on two isolated algal species, as well as natural phytoplankton assemblage (2) To asses the effect of mercury on the changes of diversity and dominance of Nile water algae and its accumulation in the produced biomass.

MATERIAL AND METHODS

A unicellular green algae *Scenedesmus quadricauda* and filamentous Cyanobacterium, *Anabaena flos-aquae* were chosen for the bioassays. The test organisms were isolated from Nile water and transferred into the standard culture media, namely, the Algal Assay, Procedure Bottle Test (US EPA, 1971) for *Scenedesmus* and the modified Watanaba medium (El-Nawawy1958) for *Anabaena.* When the organisms reached the logarithmic phase they were introduced into the pure cultures media. Natural assemblages of phytoplankton were

assayed to determine the effect of mercury on the dominance and diversity. Mercury was introduced to the culture as mercury chloride ($HgCl_2$) at different concentration namely. 0.02, 0.05, 0.1, 0.2, 0.5 and 1.0 mg/L.

Toxicity bioassays were performed under controlled conditions, using the change in chlorophyll" "a" concentration as an indicator of mercury toxicity. Bioassay flasks were incubated at 24±2°C for *Scenedesmus* and other freshwater algae, and 30±2°C for *Anabaena.* Continuous exposure of the system to fluorescent light (≈6000 Lux) was allowed. Flasks were shaken once per day to prevent clumping for the cells. From each flask a known quantity of test media was filtered through a 0.45 μm membrane filter to determine chlorophyll "a" in terms μg/L using methanol METHOD (Fitzgerald 1971) for test organisms and APHA (Standard Methods 1992) for natural phytoplankton.

Changes in the pH-values over the course of this study were recorded. Experiments were generally run for 10-days, this being a period designed to allow good growth but not long enough to cause nutrient shortages even in the control .

Atomic absorption cold vapour technique (EPA 1981) was used for mercury determination in the harvested algae (after7-day exposure time).

RESULTS AND DISCUSSION

Effect of Mercury on Scenedesmus Growth.

The results obtained (Figure 1) showed that slight variation in *Scenedesmus* growth at 0.02 and 0.5 mg/L mercury when compared with control. The maximum algal growth was 91.8% and 89.2 %of control (Figure2).

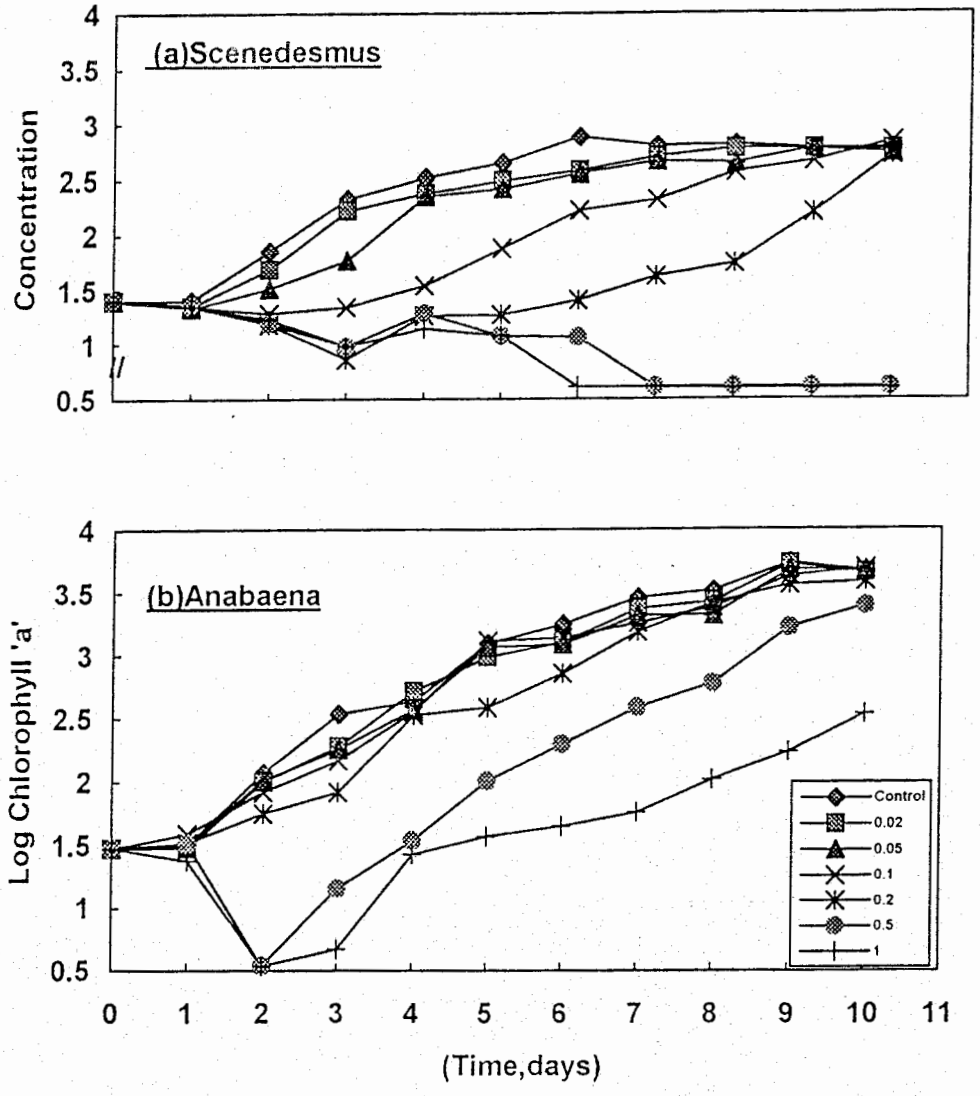
Inhibition levels increased with increasing mercury concentrations for 0.1 and 0.2 mg/L mercury especially during the first three days. then gradual increase in chlorophyll "a" content took place with maximum biomass 107% and 80% respectively. The inhibitory effect was clear at 0.5 and 1.0 mg/L mercury. Also, the specific growth rate of scenedesmus decreased by increasing mercury concentrations and reached zero at 0.5 and 1.0 mg/L mercury.

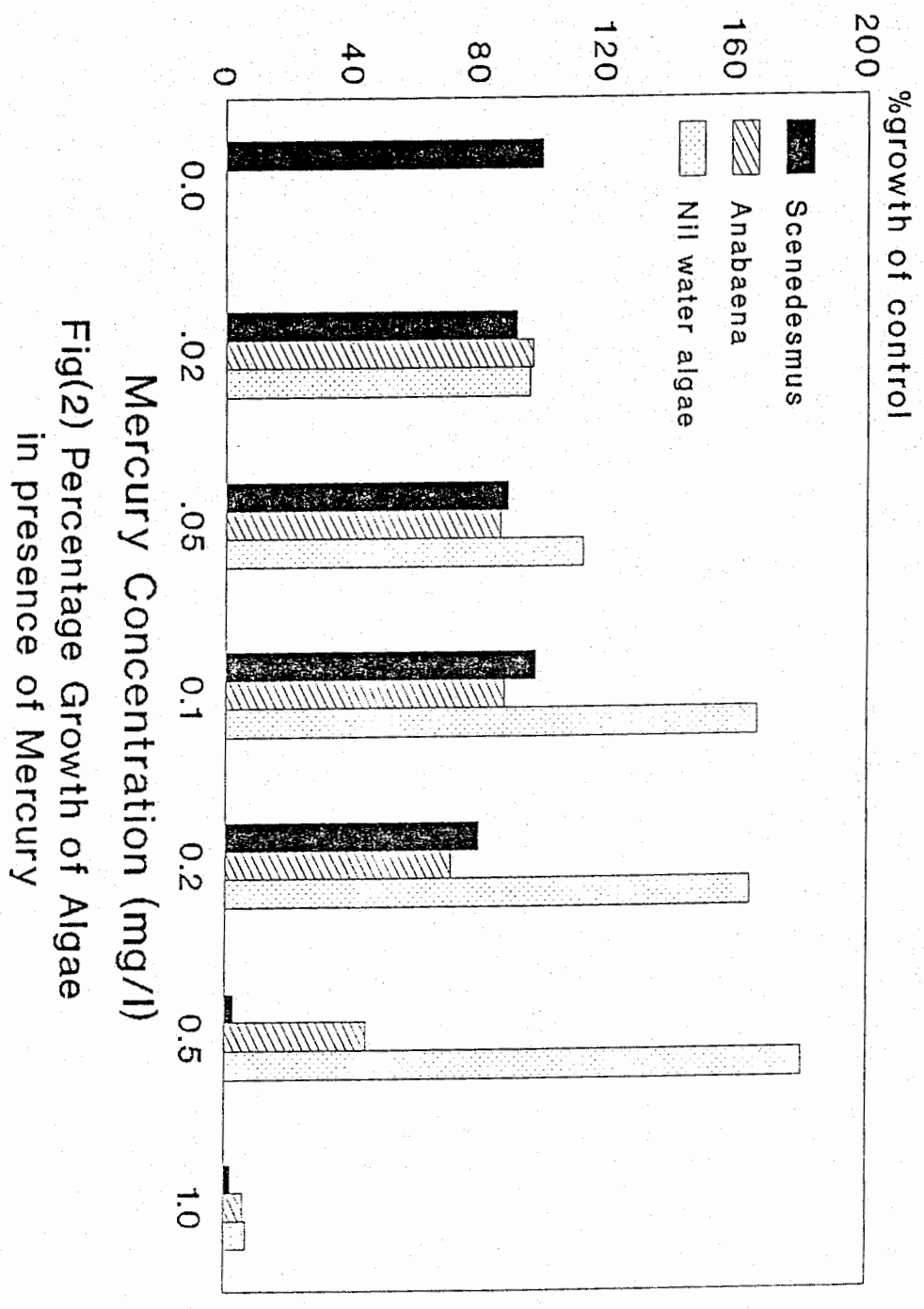
Rai. et al., (1981) showed that the specific growth rate of *Chorella vulgaris* decrease from 0.415 to 0.175 at 0.1 and 0.8 mg/L mercury. Trevors (1980) recorded that mercury at 1.0 mg/L inhibited the total growth of *Ankistrodesmus braunu*.

Effect of Mercury on Anabaena Growth.

The growth pattern of *Anabaena* in the presence of mercury is shown in (Figure 1). No clear variation in *Anabaena* growth at 0.02 mg/L mercury compared to that of control. The alga growth was slightly affected by 0.05, 0.1 and 0.2 mg/L mercury. When *Anabaena* subjected to 0.5 and 1.0 mg/L mercury sharp decline in alga growth up to the 2nd days took place, after which the alga recovered with maximum biomass 44,3% and 6.3% of control respectively (Figure 2).

Fig(1a,b) Effect of Mercury on the Algal growth





Fig(2) Percentage Growth of Algae in presence of Mercury

In addition the specific growth rate of *Anabaena* increased from 1.3 to 1.75 by increasing mercury concentrations (Figure 3). Strattan et al., 1979 found that mercury causes cell Lysis in the blue-green alga *Anabaena inaequalis* when used at concentration of 2 mg/L. Lower level still induce the visible leakage of cellular contents, leaching to the general inhibition of growth, photosynthesis and nitrogenase activity. Cell Lysis appear to be the primary toxic effect of mercury in this organism.

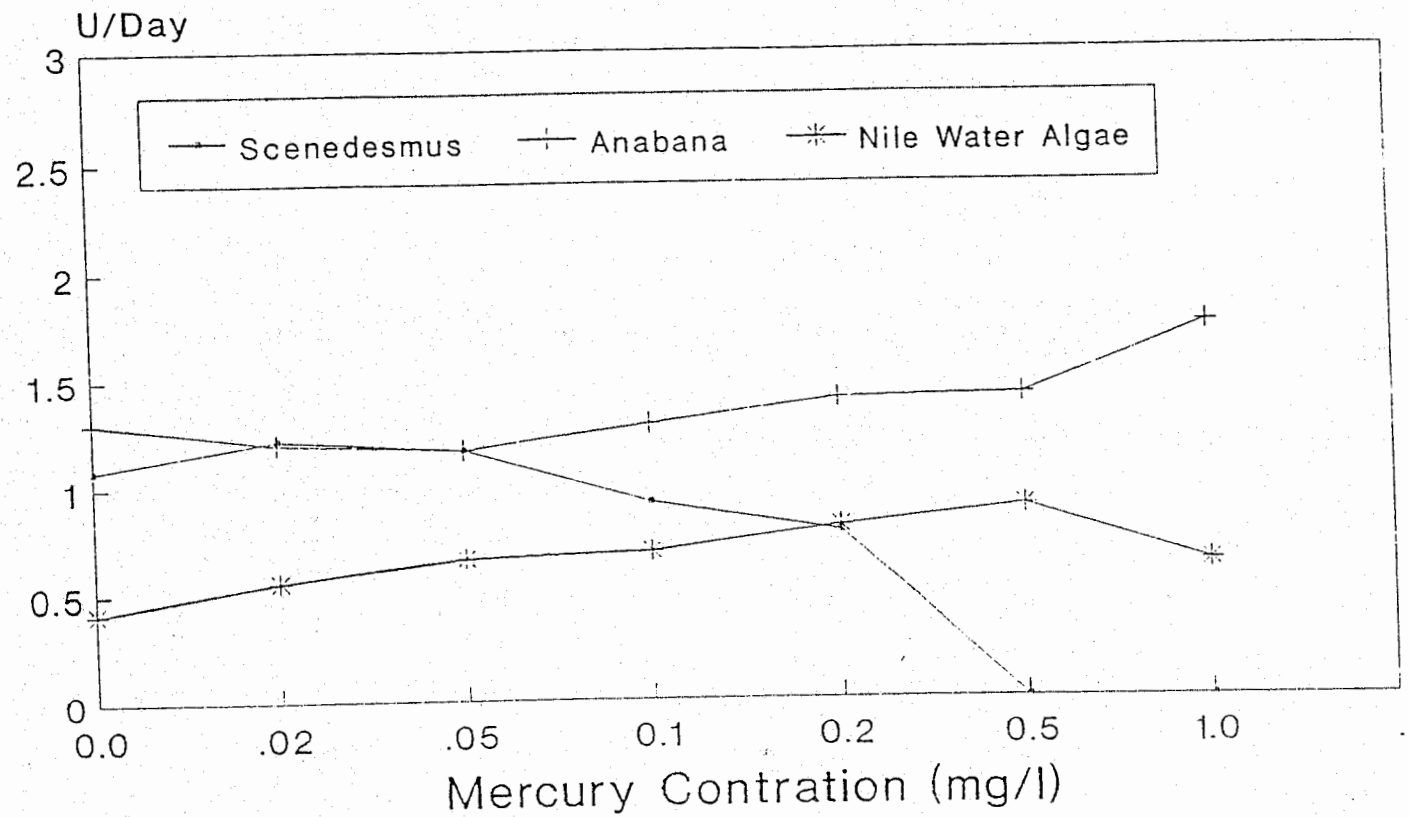
Effect of Mercury on Natural phytoplankton.

1-Algal growth.

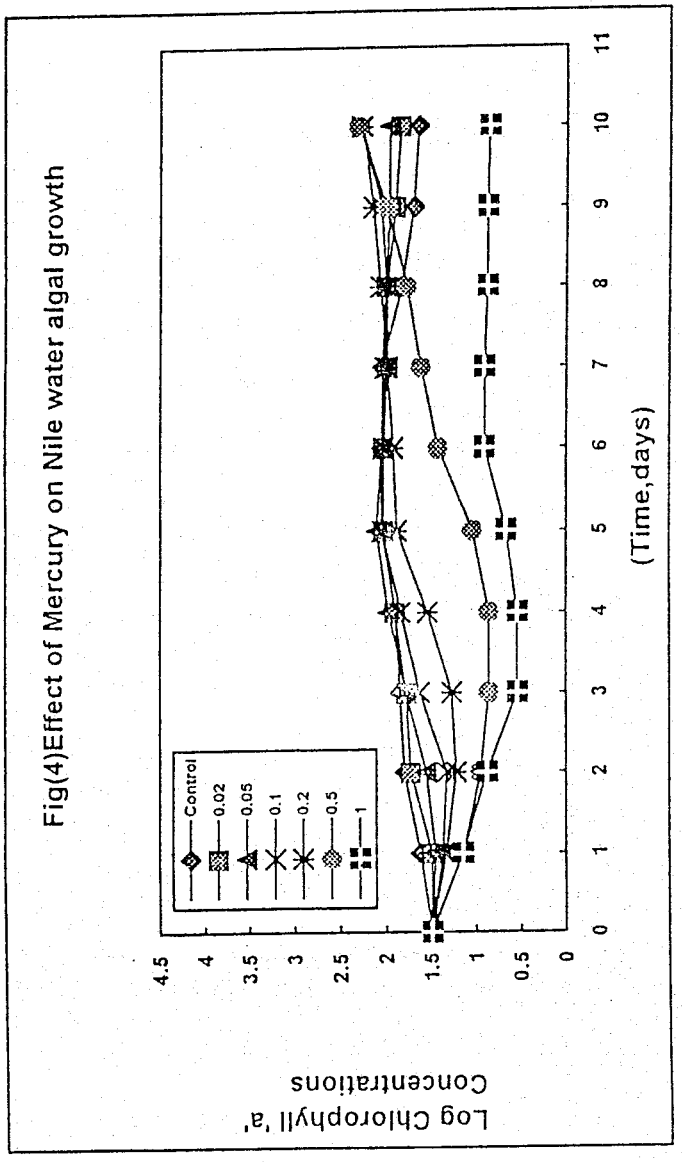
Natural phytoplankton were subjected to the same previous concentrations of mercury. From (Figure 4) it can seen that no clear variation in algal growth at 002 and 0.05 mg/L mercury. Inhibitory effect on algal growth up to the 2nd day was noticed at 0.1 and 0.2 mg/L meercury. However the tolerant algae species revealed high algal biomass in the successive days of exposure. Marked inhibitory effect was recorded up to the 4th day when mercury concentration increased to 0.5 mg/L mercury. After which gradual increase in algal growth took place and increased by 80% of control. Sever inhibitory effect was detected when natural algal flora exposed to 1.0 mg/L mercury.

Although, the maximum specific algal growth increase by increasing mercury concentration and reached its maximum at 0.5 mg/L mercury (Figure 3).

Kuiper et al., (1983) found that mercury significantly inhibited phytoplankton at 2µg/L mercury and Marshall et al., (1981) recorded that mercury at 4µg/L significantly inhibited the primary productivity of natural lake phytoplankton. In contrast Mora and Fabregas 1980 stated that mercury (0.15-20 mg/L) inhibited the growth of marine diatom, *Nitzschia acicularia*



Fig(3) Maximum Specific growth rat(U) of algae at different concentration of Mercury


Fig(4)Effect of Mercury on Nile water algal growth
Log Chlorophyll 'a' Concentrations
(Time,days)
Control
0.02
0.05
0.1
0.2
0.5
1
4.5
4
3.5
3
2.5
2
1.5
1
0.5
0
0
1
2
3
4
5
6
7
8
9
10
11

2- Changes in algal community structure

To correlate changes in algal community structure with mercury concentrations, regular microscopic examinations of algal populations were carried out. The results obtained showed that the initial flora were characterized by three major groups, namely *Chlorophyta, Cyanophyta* and *Bacillariophyta.* Diatoms represented the most dominant group, present in high numbers exceeding the other two groups. The most dominant genera were *Cyclotella* and *Melosira.* A good diversity of green algae were detected and *Eudorina* was found in high quantity. *Cyanophyta, Coelosphaerium* and *Cylindrospermum* were present in considerable amount (Tabe 1). No clear changes in the diversity of green algae was observed at the concentration of 0.02 and 0.05 mg/L mercury. When increasing mercury concentration from 0.1 to 1.0 mg/L mercury, the total number of species reduced from 31.2% to 68.7%. All investigated mercury concentrations reduce diatoms species by 50%. Blue-green alga *Oscillatoria* was the most tolerant species present in good quantity nearly at all mercury concentration.

Mercury accumulation

Mercury accumulation increased in the following order:

Nile water algae > *Anabaena* > *Scenedesmus*

and the average was 1.6, 1.1 and 0.3 μg/g mercury for NIle water algae, *Anabaena* and *Scenedesmus* respectively.

Statistical Analysis of the Results

Available data in terms of chlorophyll "a" were subjected to the regression formula (Ezakel and Fox 1959) to verify the relation between mercury concentrations and algal growth of *Scenedesmus*, *Anabaena* and natural flora. The regression lines recorded in Figure (5) revealed that a highly significant relation exists between mercury concentrations and chlorophyll "a" content.

CONCLUSIONS

The results indicated that Nile water algae has high ability to tolerate and accumulate mercury than the two isolated organisms. The inhibitory effect of mercury was in the following order:

Scenedesmus > *Anabaena* > Nile water algae.

Generally, the pure cultures and natural phytoplankton were almost equally sensitive to high mercury concentration.

Table (1) Changes in community structure at different Mercury concentrations.

Phytoplankton Taxa	Initial	Mercury concentrations mg/L						
	0.0	Control	0.02	0.05	0.1	0.2	0.5	1.0
Chlorophyta (Green algae)								
Actinastrum hantzshil	±	+	2+	+		+		
Ankistrodemus acicularis	±	+	2+	2+	2+			
Ankistrodemus falcatus.		±						
Botryococcus branii	±	2+	+	+	+	+		
Chodatella cillata.			±					
Coelasterum microporum.	±	2+	2+	+	+		+	
Dictyosphaeruim pulchellum.	±	+	+	+	+	+		
Eudorina elegans.	2+	±	+	+	+	+		
Gonium pectorale.		+	+	+	+	+		
Kirchneriella obesa.	+	+	+	+	+			
Microactinium pusillum.	+	2+	2+	2+	+	+	±	±
Oocystis parva.	+	+	+	+	+	+	+	+
Pediastrum duplex.			±					
Pediastrum simplex.	+	2+	2+	+	+		±	±
Pediastrum tetras.	±	±						
Scenedesmus obliquus.	±	±	±					
Scenedesmus quadricauda.	+	2+	2+	2+	2+	2+	±	±
Sphaerocystis schroeteri	±							
Spirogyra communis	+	+						
Staurastrum paradoxum	±	+						

Table (1) cont.

Phytoplankton Taxa	Initial	Mercury concentrations mg/L						
	0.0	Control	0.02	0.05	0.1	0.2	0.5	1.0
Tetraedron minimum	±							
Cyanophyta (Blue -green Algae)								
Anabaena spiroides .	±	+	±					
Chrococcus turgidus	±	+	±	±	+	+	+	+
Coelosphaerium kuetzingianum	+	2+	+	±	±	+	+	+
Cylindrospermum stagnale.	+	±						
Merismopedia glauca.	±	2+	2+	+	+	+	+	
Microcystis aeruginosa	±	+	±					
Oscillatoria mougeotii	±	4+	4+	3+	3+	2+	+	+
Bacillariophyta (Diatoms).								
Asterionella gracillima.	±	±						
Cocconies placentula	±	±	±					
Cyclotella Comta	4+	2+	2+	2+	2+	+	+	+
Cymbella prostrata	±	±						
Diatoma elongatum	2+	2+	2+	2+	2+	+	+	+
Melosira granulata	3+	2+	2+	2+	2+	+	+	+
Navicula cryptocephata	+	±	±					
Nitzschia linearis.	±	±	±	+	2+	2+	+	+
Peridinium cinctum	±	±						
Synedra ulna	2+	+	+	+	+	+	+	+

4+ Dominant 3+ Plenty 2+ Appreciable number ± Little

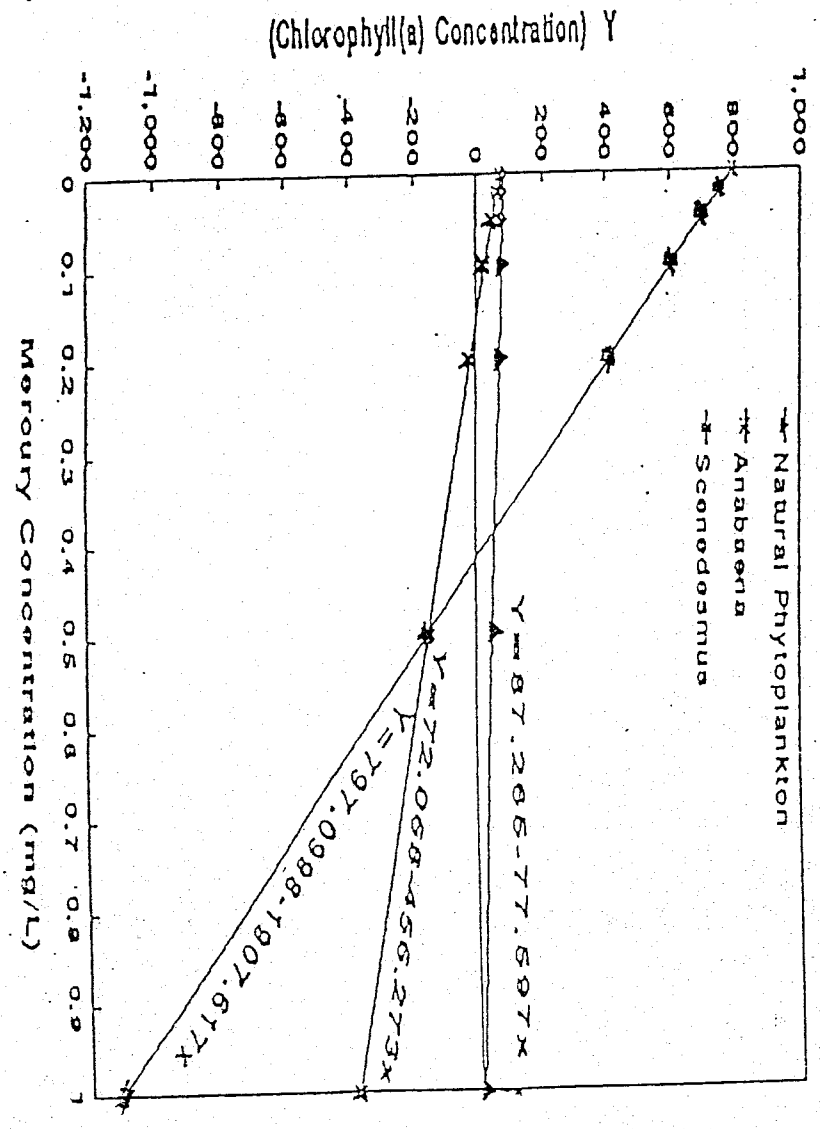


Fig. (5) Regression Between Chlorophyll(a) Content and Mercury Concentration

REFERENCES

Berlin, J. (1979): "Mercury in L. Friberg, G. F. Nordberg and V. B. Vouk Eds" Handbook on the Toxicology of Metals, Elsevier/North Holland biomedical Press, N. Y. P. 503.

El-Nawawy, A. S., Lotfi M. and Fahmy, M (1958): "Studies on the ability of some blue-green algae to fix atmospheric nitrogen and their effect on growth and yield of paddy, "Agric. Res. Rev. 36(27): 308-320.

Elsokkary, I. M. , (1989): " Mercury biogeochemistry in the aquatic systems of Alexandria District: an assessment of contamination and speciation. In :Proceedings, International Conference on Heavy Metals in the Environment, Geneva, Vol. 2, pp. 416-419.

Ezakel M and Fox, K. (1959): "Correlation and Regression Analysis", John Willy Sons Inc., New Yourk.

Fitzgerald G. P. (1971): "A manual on methods for measuring primary production in aquatic environment" Hand book No. 12, Blackwell Scientific publishers. Oxford and Edinburgh.

Kuiper J. Brokmann U. H., H. Van het Groenewond Hoornsman G. and Rocle P. (1983): "Effects of mercury on enclosed plankton communities in the Rosfjord during POSER "Mar. Ecol. Prog. Ser., 14, 93.

Marshall J . S. Parker J. I, Mellinger D. L. and lawerence S. G. (1981): "An in situ study of cadmium and mercury stress in the plankton community of lake 382, experimental lakes

area, northwesterm" Ontario, Can. J. Fish. Aquat, Sci. 38, 1209.

Mora B. and J. Fabregas (1980): "The effect of inorganic and organic mercury on growth kinetics of *Nitzschia acicularis* W. Sm. and *Tetraselmic Suecica* Batch Can. J. Microbiol. 26, 930(1980).

Standard Methods for the examination of water and wastewater, APHA, AWWA, WPCF, 18th ed, Washington (1992).

Stratton G. W., Huber A. L. and Corke, C. T. (1979): "Effect of mercuric ion on the growth, photosynthesis and nitrogenase activity of *Anabaena inaequalis"* Appl. Environ. Microbiol., 38, 537.

Trevors J. T. (1982): "A comparison of methods for assessing toxicant effects on algal growth, Biotechnol, Letters, 4, 243.

U. S. EPA (1971) "Algal Assay Procedure Bottle Test. Natural Eutrophication Research Program". Environmental Protection Agency.

U. S. EPA (1981), Interium Methods for sampling and analysis of priority pollutants in sediments and fish tissue" 600014/81/005.

التقييم السمى للزئبق على طحلب الأنابينا وطحلب السنيدسيميس وطحالب نهر النيل

صباح عبد العظيم بدر، عزة محمد عبد المنعم عشماوى،

قسم بحوث المياه – المركز القومى للبحوث

القاهرة – مصر

الهدف من هذا البحث هو تقييم مدى سمية التركيزات المختلفة للزئبق على طحلب الانابينا، طحلب السيندسيميس وطحالب نهر النيل وقد دلت النتائج على أن درجة سمية الزئبق تزيد على النحو التالى:

طحلب السيندسيميس < طحالب الانابينا < طحالب نهر النيل

كما اوضحت الدراسة أن هناك تغير واضح فى نوعية سيادة طحالب مياه النيل وكانت الطحالب الخضراء المزرقة ممثلة فى طحلب الأوسلاتوريا هى الأكثر تحملا وسيادة.

وقد اثبت التحليل الإحصائى الترابط الوثيق بين نمو الطحالب والتركيزات المختلفة للزئبق.

والخلاصة أن لطحالب مياه النيل القدرة على إستخلاص وتراكم عنصر الزئبق داخل خلاياه أكثر من السلالات المعزولة.