

RESPONSE OF NILE PHYTOPLANKTON TO ALGICIDE ( $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ )  
IN MULTISPECIES CHEMOSTAT EXPERIMENTS

BY

A. A. ISSA\* and ZEINAB A. AHMED\*\*

\* Department of Botany, Faculty of Science, Assiut University

\*\* Department of Botany, Faculty of Science (Sohag), Assiut University

إستجابة هائمات النيل النباتية لمبيد الطحالب (كبريتات النحاس)  
في تجارب المزارع المستمرة (الكيموستات)

أحمد عبد السلام عيسى و زينب عبد الفتاح

- لقد استهدفت هذه الدراسة تأثير كبريتات النحاس (مبيد للطحالب) على مزارع من هائمات النيل النباتية والنامية تحت ظروف ثابتة باستخدام الكيموستات ووجد أن إضافة هذا المبيد تسبب عموماً في حدوث تغيرات ملحوظة في تكوين الأنواع والعدد الكلي للطحالب وسمية هذا المبيد على الأنواع المختلفة .

وقد إنخفض العدد الكلي للطحالب وكذلك عدد الأنواع بصورة ملحوظة بزيادة تركيز المبيد في أوعية المزارع مهما كان معدل التخفيف المستخدم .

وكانت الأجناس فراجيلاريا ، أنابينا ، ميكروسيستس وسيلناسترم الأكثر حساسية لكبريتات النحاس ( مبيد الطحالب ) بينما كانت الكلوريللا والبيديا سترم والسينيدي سماس الأكثر مقاومة لهذا المبيد .

*Key Words:* Algicide,  $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ , Nile phytoplankton.

ABSTRACT

The addition of  $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$  (algicide) to cultures of phytoplankton populations from the river Nile, grown under steady-state conditions using chemostat, resulted in remarkable alterations in species composition, total algal counts and toxicity of algicide to various species. The total algal population and the number of species were considerably reduced by increasing the concentration of the algicide in culture vessels, irrespective of the dilution rates used. *Fragilaria*, *Anabaena*, *Microcystis*, and *Selenastrum* were considered to be very susceptible to copper sulfate, while *Chlorella*, *Pediastrum* and *Scenedesmus* were resistant.

INTRODUCTION

The ecological and environmental importance of most algae are clouded somewhat by problems caused by a relatively, associated with their massive rapid growth under favourable conditions. Eutrophication of aquatic environments, accelerated in recent years by abnormal nutrient loading from domestic and industrial wastes and drainage from agricultural land, is one cause of the increasing number of a algal fouling problems especially those associated with blue-green algae [1-4]. Chemicals can be used to prevent development of algal growth (algistatic) or to kill existing populations (algicides). Copper sulfate pentahydrate ( $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ ), first advocated as an algicide in 1904, is widely used for controlling planktonic algae [3, 5 & 6]. The lowest concentration of copper sulfate which is toxic for a particular alga also varies according to the abundance of the alga, temperature and alkalinity of the water, the amount of organic

material in the water, and other factors. Thus, the listing of a specific concentration of al algicide as the minimum effective dosage is not reliable unless these other factors have been taken into consideration.

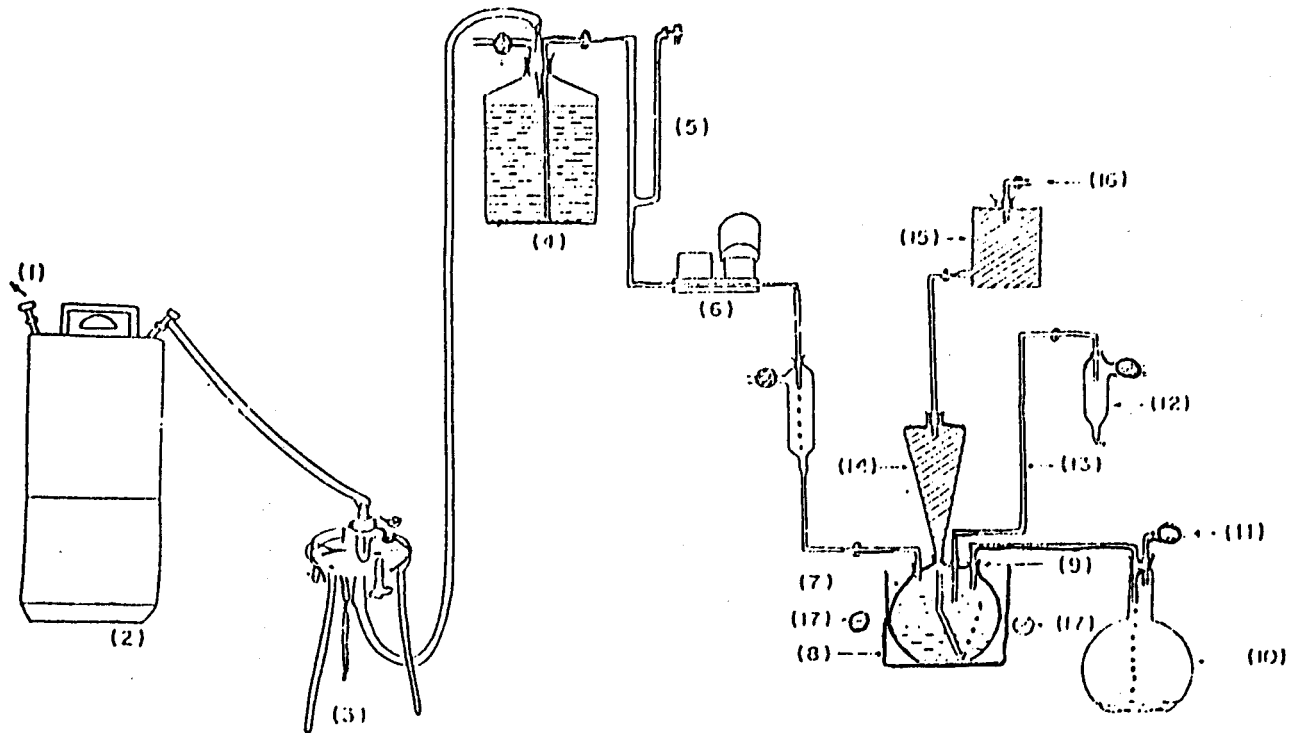
This work was planned to study the effect of copper sulfate on the density and diversity of Nile phytoplankton in multispecies chemostat experiments under steady-state conditions.

MATERIALS AND METHODS

The inoculation consisted of 10L of natural phytoplankton suspension taken from the light saturated layer of river Nile (Assiut, Egypt.) at the beginning of each experiment. Before being filled into the culture vessel, the plankton suspension was bubbled for 1 hour by nitrogen in order to kill zooplankton by oxygen depletion. The dilution rates ranged

from  $0.0077 \text{ h}^{-1}$  to  $0.0417 \text{ h}^{-1}$  by micro-pump (Fig. 1); temperature was adjusted to  $25 \pm 1^\circ\text{C}$  and surface light intensity of  $1.6 \times 10^{16} \text{ quanta Cm}^{-1} \text{ S}^{-1}$ . pH was regulated to 7.0 by addition of  $\text{CO}_2$  in order to prevent carbon limitation. The nutrient solution was CHU 12 as modified by Müller [7], with all nutrients in excess augmented with an algicide  $\text{CuSO}_4$ . Three concentrations, 5, 10 and 50 ppm, were used.

Absorbance at 750nm was measured to monitor changes more than 5% over 3 days, the culture was considered to be in a steady-state than 5% over 3 days, the culture was considered to be in a steady-state (growth rate  $\mu$  equals the dilution rate  $D$ ). At that time, the entire culture volumes were sampled for identification and counts.



1- Air pump. 2- Pressure vessel. 3- Filter holder. 4- Medium reservoir. 5- Measuring tube. 6-Micro pump. 7-Chemostat culture vessel. 8-Waterbath 9-Overflow tube. 10-Overflow vessel. 11-Air outlet. 12-Sampling device. 13-Withdrawal tube. 14-Sterile glass wool filter. 15-Prefilter. 16-Air inlet 17-Fluorescent lamps.

Fig. (1): Diagrammatic representation for the chemostat culture of Nile phytoplankton.

## RESULTS

Continuous culture technique enables to use very low nutrient concentrations, and to obtain a dynamic equilibrium between the nutrient input and algal growth. A desired cell density can be maintained either by controlling the levels of the limiting nutrients in the reservoir or by controlling the rate of their inflow. In this well defined chemical environment, the desired growth can be easily selected and maintained for a long period at any rate between zero and maximum. The total algal counts were considerably reduced by raising the concentration of  $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$  in culture vessel. Similarly, the number of species were lowered with the rise of algicide concentration (Table 1).

The effect of various dilution rates ( $0.0077$ ;  $0.0165$ ;  $0.0215$ ;  $0.0260$ ;  $0.0344$  &  $0.0417 \text{ h}^{-1}$ ) on algal counts and algal diversity at various algicide concentration were studied. These parameters increased with the increased dilution rate up to  $0.0260 \text{ h}^{-1}$ , while at relatively higher dilution rates, a continuous decrease was observed.

The various genera and species of algae respond differently in their reaction to copper sulfate, and this factor has frequently been neglected in determining the concentration of the algicide to be applied (Table 2).

## DISCUSSION

The grouping of algae is by very general ranges in the dosage required for treatment. The total algal counts and number of species decreased with the increase in algicide concentration, irrespective of dilution rate. This is in accordance with the results obtained by some other authors using herbicides [8-14]. Bryfogle and McDiffett [13] who summarized the effect of herbicides on algal communities, observed that at higher levels, the structure of the communities, observed that at higher levels, the structure of the community is altered with loss of diversity and changed in species dominance.

At relatively higher dilution rates ( $< 0.0260 \text{ h}^{-1}$ ), algal counts and diversity were continuously reduced. These results are in harmony with those obtained earlier [15, 16].

**Table 1**  
Effect of algicide (copper sulfate) on species composition and total algal counts of Nile phytoplankton

Algal group & Species	Dilution rates D (1/h)																															
	0.0077				0.0119				0.0165				0.0215				0.0260				0.0344				0.0417							
	con	5	10	50	con	5	10	50	con	5	10	50	con	5	10	50	con	5	10	50	con	5	10	50	con	5	10	50				
Chlorophyta:-																																
<i>Actinastrum cracillium</i> Smith	3	3	1	-	4	4	2	-	4	4	3	-	6	6	-	-	8	5	4	-	6	5	-	-	5	4	-	-				
<i>Ankistrodesmus jalcatus</i> Ralis	5	5	-	-	5	4	-	-	6	4	-	-	8	5	-	-	8	5	-	-	7	3	-	-	6	2	-	-				
<i>A. convolutus</i>	2	1	-	-	3	2	-	-	6	4	-	-	9	6	-	-	11	8	-	-	8	4	-	-	5	1	-	-				
<i>Chodatella ciliata</i> Lemm.	1	-	-	-	2	-	-	-	4	-	-	-	4	-	-	-	5	-	-	-	4	-	-	-	3	-	-	-				
<i>Chlorella</i> sp.	3	3	3	3	5	4	3	3	8	5	4	2	11	8	6	3	13	7	6	5	11	10	6	3	9	2	1	1				
<i>Dictyosphaerium pulchellu</i> Wood	5	2	-	-	5	2	-	-	5	1	-	-	6	3	-	-	6	2	-	-	4	1	-	-	2	1	-	-				
<i>Eudorina elegans</i> Ehrbg	1	1	-	-	1	-	-	-	2	-	-	-	2	-	-	-	2	-	-	-	1	-	-	-	1	-	-	-				
<i>Gonium pectorale</i> Mull	1	-	-	-	1	-	-	-	3	-	-	-	4	-	-	-	6	-	-	-	3	-	-	-	2	-	-	-				
<i>Microactinium pusillum</i> Fresenius	3	-	-	-	5	-	-	-	7	-	-	-	7	-	-	-	9	-	-	-	5	-	-	-	4	-	-	-				
<i>Microspora</i> sp.	1	1	-	-	1	1	-	-	1	1	-	-	1	1	-	1	1	-	-	-	-	-	-	-	-	-	-	-				
<i>Oocystis borgeri</i> Snow	3	2	1	-	3	1	1	-	5	2	1	-	6	3	2	-	8	4	3	-	4	2	1	-	2	1	-	-				
<i>O. parva</i> West	1	-	-	-	2	-	-	-	2	-	-	-	4	-	-	-	5	-	-	-	1	-	-	-	-	-	-	-				
<i>Pandorina morum</i> Mull	1	1	1	1	2	1	1	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-				
<i>Pediastrum duplex</i> Meyen	4	2	1	1	6	3	2	2	6	2	1	1	7	4	3	1	8	5	3	2	5	1	1	1	3	1	1	1				
<i>P. integrum</i> Meyen	3	1	-	-	5	1	-	-	5	2	-	-	5	3	-	-	4	1	-	-	3	2	-	-	1	1	-	-				
<i>P. simplex</i> Meyen	1	-	-	-	2	-	-	-	3	-	-	-	3	-	-	-	1	-	-	-	-	-	-	-	-	-	-	-				
<i>Selenastrum gracile</i> Reinsch	2	-	-	-	3	-	-	-	3	-	-	-	5	-	-	-	7	-	-	-	6	-	-	-	3	-	-	-				
<i>Scenedesmus arcuatus</i> Lemm.	6	3	3	3	8	3	2	3	11	8	5	5	15	10	7	8	21	8	3	2	18	7	4	4	14	5	3	3				
<i>S. quadricauda</i> Breb	8	4	3	3	10	8	7	3	14	10	8	5	16	10	8	8	23	11	10	10	13	11	10	7	11	8	5	3				
<i>Staurastrum uniseriatum</i> Nyg.	6	3	2	-	8	3	3	-	8	3	3	-	9	3	2	-	13	6	3	-	10	8	2	-	8	5	1	-				
<i>Spirogyra</i> sp.	2	1	-	-	3	1	-	-	3	1	-	-	3	1	-	-	8	1	-	-	4	1	-	-	3	1	1	-				
<i>Tetraspora</i> sp.	2	-	-	-	4	-	-	-	5	-	-	-	6	-	-	-	7	-	-	-	4	-	-	-	1	-	-	-				
<i>Tetraedron minimum</i> Hansg.	3	-	-	-	3	-	-	-	4	-	-	-	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-				
<b>Total count.</b>	<b>66</b>	<b>32</b>	<b>13</b>	<b>11</b>	<b>101</b>	<b>38</b>	<b>21</b>	<b>12</b>	<b>116</b>	<b>47</b>	<b>25</b>	<b>13</b>	<b>138</b>	<b>43</b>	<b>28</b>	<b>20</b>	<b>174</b>	<b>65</b>	<b>32</b>	<b>19</b>	<b>117</b>	<b>55</b>	<b>24</b>	<b>15</b>	<b>83</b>	<b>29</b>	<b>11</b>	<b>8</b>				

counts : organism / ml

Table 1 Contd.

Algal group & Species	Dilution rates (1/h)																											
	0.0077				0.0119				0.0165				0.0215				0.0260				0.0344				0.0417			
	con	5	10	50	con	5	10	50	con	5	10	50	con	5	10	50	con	5	10	50	con	5	10	50	con	5	10	50
Bacillariophyta:-																												
<i>Amphora ovalis</i> Kutz	5	1	1	-	5	2	3	-	6	4	1	1	8	5	3	1	8	4	3	-	6	3	2	-	4	1	1	-
<i>Bacillaria paradoxa</i> Gmelin	4	-	-	-	5	-	-	-	7	-	-	-	9	-	-	-	13	-	-	-	10	-	-	-	8	-	-	-
<i>Cyclotella stelligrea</i> Cleve	5	2	2	-	7	3	3	-	7	3	2	-	11	7	5	-	6	4	3	-	4	1	1	-	4	1	1	-
<i>Cymbella tumida</i> Breb	6	3	3	-	5	3	1	-	6	4	3	-	9	5	3	-	10	8	7	-	7	6	3	-	3	1	2	-
<i>Diatoma vulgare</i>	3	-	-	-	4	-	-	-	4	-	-	-	4	-	-	-	5	-	-	-	3	-	-	-	1	-	-	-
<i>Fraguikarua oubbatam</i> Ehrbg	6	-	-	-	5	-	-	-	5	-	-	-	5	-	-	-	8	-	-	-	3	-	-	-	-	-	-	-
<i>Melosira granulata</i> Ehrbg	8	-	-	-	10	-	-	-	10	-	-	-	13	-	-	-	15	-	-	-	11	-	-	-	8	-	-	-
<i>M. islandica</i>	2	-	-	-	2	-	-	-	2	-	-	-	2	-	-	-	3	-	-	-	1	-	-	-	1	-	-	-
<i>M. italica</i> Ehrbg	1	-	-	-	1	-	-	-	1	-	-	-	1	-	-	-	1	-	-	-	-	-	-	-	-	-	-	-
<i>Navicula pupula</i> Kutz	4	2	1	-	6	3	2	-	7	4	4	-	7	4	3	-	9	3	2	-	3	2	1	-	3	1	1	-
<i>N. radiosa</i> Kutz	3	1	1	-	3	2	1	-	4	3	2	-	3	2	2	-	8	5	3	-	1	1	1	-	2	1	1	-
<i>Nitzschia calida</i> Grun	6	5	2	-	9	6	4	-	11	8	7	-	15	10	7	-	22	10	5	-	19	8	7	-	10	3	1	-
<i>N. palea</i> Kutz	8	5	3	-	8	5	3	-	8	5	4	-	13	8	8	-	16	9	7	-	11	5	3	-	10	2	1	-
<i>Pinnularia</i> sp.	1	-	-	-	1	-	-	-	1	-	-	-	1	-	-	-	1	-	-	-	1	-	-	-	1	-	-	-
<i>Synedra acus</i> Kutz	7	-	-	-	9	-	-	-	13	-	-	-	17	-	-	-	22	-	-	-	15	-	-	-	13	-	-	-
<i>S. ulna</i> Ehrbg	3	-	-	-	2	-	-	-	5	-	-	-	7	-	-	-	10	-	-	-	8	-	-	-	3	-	-	-
<i>Surireila ovata</i> Kutz	2	-	-	-	2	-	-	-	1	-	-	-	1	-	-	-	1	-	-	-	1	-	-	-	1	-	-	-
<i>S. robusta</i> Ehrbg	2	-	-	-	2	-	-	-	1	-	-	-	1	-	-	-	2	-	-	-	-	-	-	-	-	-	-	-
Total count	76	19	13	-	86	24	17	-	99	31	23	-	123	37	28	-	165	43	32	-	106	29	20	-	72	10	8	-
Cyanophyta:-																												
<i>Anabaena</i> sp.	2	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
<i>Chroococcus</i> sp.	2	-	-	-	5	-	-	-	7	-	-	-	13	-	-	-	16	-	-	-	16	-	-	-	11	-	-	-
<i>Lyngbya</i> sp.	1	1	1	1	1	1	1	1	2	1	1	1	2	1	1	1	2	1	1	1	2	1	1	1	1	1	1	1
<i>Merismopedia elegans</i> A. Braun	1	-	-	-	1	-	-	-	1	-	-	-	2	-	-	-	2	-	-	-	-	-	-	-	-	-	-	-

Table 1 Contd.

Algal group & Species	Dilution rates (1/h)																											
	0.0077				0.0119				0.0165				0.0215				0.0260				0.0344				0.0417			
	con	5	10	50	con	5	10	50	con	5	10	50	con	5	10	50	con	5	10	50	con	5	10	50	con	5	10	50
Cyanophyta:-																												
<i>Microcystis incerta</i> Lemm.	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
<i>Oscillatoria agardhii</i>	3	2	2	1	4	2	3	1	6	4	3	1	6	5	5	4	8	6	3	4	6	5	3	1	5	3	1	1
<i>O. formosa</i> Bory	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
<i>O. limosa</i> Lemm.	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
<i>O. limnetica</i> Lemm.	2	2	1	1	3	2	1	1	3	2	1	1	2	1	1	3	2	1	1	-	-	-	-	-	-	-	-	-
<i>Phormidium molle</i> (Kutz) Gomont	4	3	2	2	5	4	3	3	7	5	3	3	9	7	2	2	9	6	3	3	3	2	1	1	2	1	1	1
<i>Spirulina laxa</i>	1		1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
Total count	19	11	9	8	17	12	11	9	29	15	11	9	37	17	12	10	42	17	10	11	28	10	7	5	19	7	3	3
Euglenophyta:-																												
<i>Euglena</i> sp.	2	1	1	-	3	1	1	-	2	1	-	-	1	-	-	-	1	-	-	-	-	-	-	-	-	-	-	-
<i>Phacus</i> sp.	3	1	1	-	2	1	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Total count	5	2	2	-	5	2	2	-	2	1	-	-	1	-	-	-	1	-	-	-	-	-	-	-	-	-	-	-
Phyrophyta:-																												
<i>Chroomonas acuta</i> Utermohl	1	1	-	-	1	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
<i>Ceratium</i> sp.	1	1	-	-	1	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Total count	2	2	-	-	2	2	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
No of species	56	32	24	12	54	32	24	12	50	28	21	11	50	27	20	10	47	25	20	10	41	24	16	9	39	20	15	7
Total Counts	168	66	37	19	211	78	61	21	246	94	59	22	299	97	68	30	385	125	74	30	251	94	51	29	213	46	22	11

**Table 2**  
Relative toxicity of copper sulfate to Nile phytoplankton grown in chemostat culture

Algal genera	very susceptible (-)	susceptible (-)	Resistant (+)	Very resistant (++)
Chlorophyta:-				
<i>Chodatella</i>	--			
<i>Chlorella</i>				++
<i>Dictyosphaerium</i>		-		
<i>Micractinium</i>	--			
<i>Eudrina</i>			+	
<i>Gonium</i>			+	
<i>Selenastum</i>	--			
<i>Scenedesmus</i>				++
Bacillariophyta:-				
<i>Bacillaria</i>	--			
<i>Cyclotella</i>		-		
<i>Melosira</i>	--			
<i>Synedra</i>		-		
Cyanophyta:-				
<i>Anabaena</i>	--			
<i>Lyngbya</i>			+	
<i>Microcystis</i>	--			
<i>Oscillatoria</i>			+	++
<i>Phormidium</i>				++
<i>Spirulina</i>				++

The diatoms as a group are relatively susceptible, but they have often developed in large numbers following the destruction of other algae through treatment with copper sulfate. *Fragilaria*, *Anabaena*, *Microcystis* and *Selenastrum* were considered to be very susceptible to copper sulfate (present only in control). However, a number of the very minute planktonic green algae were very resistant to the toxic effects of copper sulfate such as *Chlorella*, *Pediastrum* and *Scenedesmus*. The green flagellates and some of the filamentous blue-green algae were considered to be resistant.

It can be generally concluded that the results obtained during this investigation applying chemostat culture are in conformity with those obtained from the field studies by other authors.

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#### REFERENCES

- [1] Fitzgerald, G.P., G.C. Gerloff and F. Skoog, 1952. Studies on chemicals with selective toxicity to blue-green algae. *Sewage Ind. Wastes* 24: 888-896.
- [2] Echlin, P., 1966. The blue-green algae. *Sci.*, 214: 74-83.
- [3] Hawkins, A.F., 1972. Control of algae. *Outlook on Agriculture* 7: 21-26.
- [4] Starmach, K., 1974. Biological characteristics of blue-green algae and conditions of their development. A review. *Pol Arch. Hydrobiol.* 21: 29-39.
- [5] Grance, J.H., 1963. The effect of copper sulphate on *Microcystis* and zooplankton in ponds. *Provege Fish Cult.* 25: 198-202.
- [6] Fogg, G.E., W.D. P. Stewart, P. Fay and A. E. Walsby, 1973. The blue-green algae. Academic Press London.
- [7] Müller, H., 1972. Wachstum und phosphatbedarf von *Nitzschia actinostroids* (LEMM) von Good in statischer und homokontinuierlicher Kultur unter phosphatlimitierung *Arch. Fur Hydrobiol. Suppl.* 38: 399-484.
- [8] Westerfield, W.W., 1965. Biological response curves. *Science*, 123: 1017-1019.
- [9] Cooke, G.D., 1967. The pattern of autotrophic succession in laboratory microcosmos *Bioscience*, 17: 717-722.
- [10] Wurster, C.F., 1968. DDT reduces photosynthesis by marine phytoplankton. *Science*, 159: 1474-1475.
- [11] Kormondy, E.J., 1969. Comparative ecology of sandspit ponds. *Am. Midl. Nat.* 28: 28-61.
- [12] Menzel, D. W., J. Anderson and A. Randtke, 1970. Marine phytoplankton vary in their response to chlorinated hydrocarbons. *Science*, 167: 1724-1726.
- [13] Bryfogle, B.M. and W. F. McDiffett, 1979. Algal succession in laboratory microcosmos as affected by an herbicide stress. *Am. Midi Nat.*, 101: 344-354.
- [14] Bogdanovic, M., P. Brikic, M. Tamirdizija and M. Plesnicar, 1980. The effect of herbicide San. 9789 on chlorophyll synthesis and photochemical activities in Black pine cotyledon. Fifth International Congress on photosynthesis. Greece, 71.
- [15] Mohammed, A.A., 1978. Freilandmessungen und Experimente zur Frage der Nährstofflimitierung von planktonalgenen Bodense. *Diss. Fak. Fur Biologie, Albert-Ludwigs-Leniversitat, Freiburg.*
- [16] Mohammed, A. A., A. M. Ahmed and H. M. Shafik, 1991. Effect of nitrogen limitation on growth of *Ankistrodesmus falcatus* and *Scenedesmus obliquus*. *Arch. Pratischenkd.* 139: 261-273.