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SEWAGE TREATMENT WITH MICROALGAE

BY

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ABSTRACT

Microalgal bioassay methods were performed to test their efficiency in removing residual nutrients responsible for the eutrophication problems in municipal wastewaters. Tertiary treatment proved to be effective in removing most pollutants. Percentage removal ranged from 70.1% to 100%; 41.7% to 100% and 33.3 to 96.1% for ammonia, nitrate and phosphate, respectively. Maximum reduction in COD and BOD recorded 90.6% and 84.1%. Moreover, the algal biomass produced may be used as a source of energy, food and fertilizers (2.7 gL^{-1}; 2.2 $gL.^{-1}$ 2.4 gL^{-1} and 3.1 gL^{-1} for Chlorella, Scenedesmus, Spirulina and algal population, respectively.

INTRODUCTION

Aquatic ecosystems are hyper eutrophic because of massive nutrient introduction by wastewater influents. Wastewaters resulting from human activities include wastes of municipal, agricultural and industrial origin which contain high concentrations of residual phosphorus, nitrogen and carbon. Tertiary treatment of wastewater with planktonic algae was extensively used to purify municipal sewage (Shelef et al., 1980; Van-Cillie et al. 1990; Ganter et al. 1991; Tripathi and Shukla, 1991 and Travieso et al. 1992; Oswald 1992). Oxygen production by algae for waste oxidation by bacteria in ponds, is now generally recognized, but algal contributions in enhancing sedimentation and disinfection and in nutrient, heavy metal and toxic organic removal are not so well known (Oswald, 1970). In addition algae represent an efficient and inexpensivewastewater treatment system, yet the harvested tissues are a cheap source of protein, energy, fertilizers and fodder.

The present study deals with th performance of different microalgae on domestic sewage as a tertiary treatment to improve its water quality through utilization of the dissolved nutrients and reduction of biological oxygen demand. Biomass production at different waste dilutions was also followed.

MATERIALS AND METHODS

The domestic wastewater used in this study was collected from Zenein Wastewater Treatment Plant Giza, at 11 am in big plastic jars. Primary treatment of sewage was carried out by sedimentation and flotation of suspended matter. This was accomplished when the effluent was left for 24 h (Al Salem, 1988). The wastes are converted to their inorganic forms by biological oxidation with activated sludge (secondary treatment). Bacteria will decompose the organic matter into mineral components (Stover and kincannon, 1975). The microalgal species selected for this study (*Chlorella pyrenoidosa, Scenedesmus obliquus,* and *spirulina platensis*) are usuially employed in algal wastewater treatment systems.

Culture media consists of a mixture of tap water free of chlorine (left 48 h in jars before use) and secondary treated sewage effluent to obtain concentrations of 10%, 20%, 40%, 80% and 100% respectively. In case of *Spirulina* 1.6% of NaHCO3 were added to all concentrations. Initial chlorophyll a and dry weight contents of algal inoculum were determined. Triblicate flasks from all treatments each containing 50 ml to which algal inoculum was added. For comparison in calculations, control cultures with the same effluent dilutions but with no algal inoculum were prepared and incubated under the same conditions parallel to the experimental cultures under continuous illumination (58.5 uEm-2 S^{-1}) at 28°C ± 1.

The experiments were extended until the inorganic N and P were depleted or algal tissue begin to disintegrate (7 days). In another experiment the same inoculum for the three organisms together were added to 150 ml of each waste dilution to study the effeciency of algal population on wastewater purification. The yield of the microalgae was harvested at the end of the incubation period by centrifugation and drying till constant weight. Chlorophyll a content was estimated in 100% hot methanol extract according to sartory and Grobbelaar (1984). Total suspended solids (T.S.S.) were determined gravimetrically after filtration and drying to constant weight at 105 C°. Sub-samples were taken from the culture media for water analysis at the begining and at the end of each experiment as follows. pH; dissolved ammonia; reactive phosphates, nitrates and chemical oxygen demand (COD) were estimated according to standard methods for the examination of water and wastewater (1985). Biochemical oxygen demand (BOD) was determined according to U.S. Environmental Protection Agency Office of Research and Development (1978).

RESULTS AND DISCUSSION

The results presented in Fig. 1 show that biomass yield increased to reach 2.7 gL^{-1}; 2.2 gL^{-1} and 2.4 gL^{-1} for *Chlorella, Scenedesmus* and *Spirulina* at 100% waste concentration after 7 days incubation. Such values are nearer to those obtained with the standard media. This may be attributed to the fact that municipal wastes contain all or most macro and micronutrients required to support optimum algal growth. Dilution of the effluent to 80% concentration resulted in subsequent decrease in biomass yield in both *Chlorella* and *Spirulina* but increase *in Scenedesmus* harvest of 2.5 gL^{-1}. Further dilution of the original effluent led to remarkable drop in biomass yield which may be

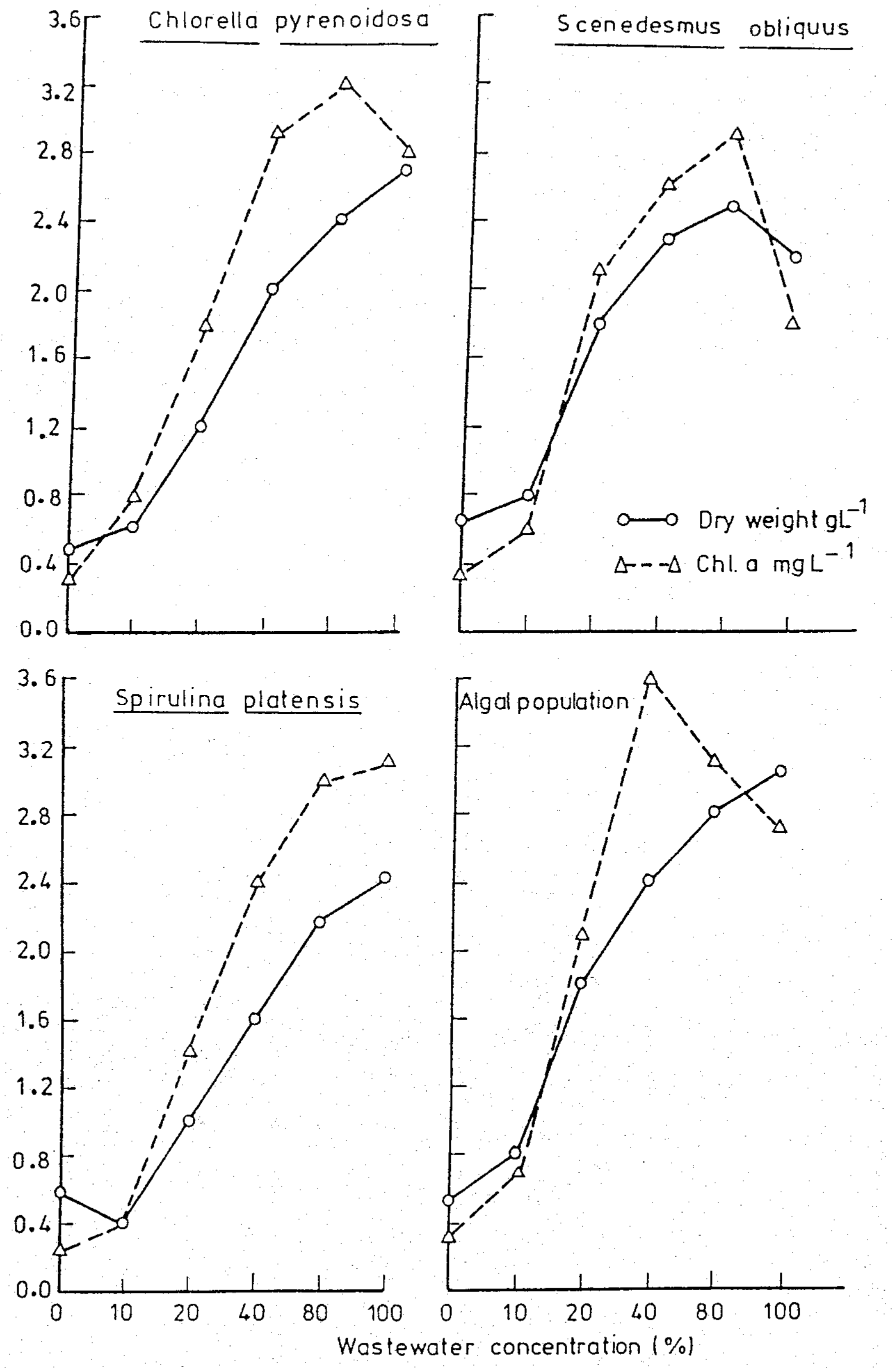

Fig.1. Effect of various concentrations of secondary treated sewage effluent on the biomass yield and chlorophyll a content of some microalgae after 7 days incubation.

due to insufficient nutrients in the diluted wastewater. Microalgal population growth showed the same trend recorded for unialgal cultures with increase in biomass yield to reach 3.1 gL^{-1} at 100% waste concentration. These results are in harmony with those obtained by Lahaniatis et al., (1991) who reported a continuous increase in the wet and dry biomass in the batch experiment for sewage treatment by employing algae and plants. However, the high yield of *Chlorella* recorded in most sewage concentration levels by the end of the experiment indicates high tolerance of such alga to extreme conditions (Rzeczycka et al. 1987). In contrary to this Dunstan and Menzel (1971) mentioned that the lower growth rates of algae at high levels of sewage is attributed to the toxicity of one or more of the major nutrients in addition to the high organic load and increased turbidity of water, all of which create highly stressed conditions. On the other hand, Saini et al. (1992) found that the highest average algal production (20 gL^{-1}) was obtained from dose 33% concentration of urban sewage. The data in Fig . 1, also indicate that chlorophyll a content as a parameter for growth went parallel to biomass yield with some minor fluctuations. Maximum values recorded at 80% waste effluent in *Chlorella, Scenedesmus* and *Spirulina* cultures were 3.2 mgL^{-1}, 2.9 mgL^{-1}; 2.9 mgL^{-1}, respectively. In microalgal population cultures maximum chlorophyll a (3.6 mgL^{-1}) was recorded at 40% waste concentration. Generally, higher levels of chlorophyll a are accompanied in most treatments with increase in dry weight gain of the microalgae under investigation.

The microalgal efficiency for removing the major nutrients are presented in Table 1. *Chlorella Pyrenoidosae* efficiency for removing ammonia ranged between 75.8 and 100%. At 20% waste concentration the initial concentration (7.2 mgL^{-1}) was consumed totally at the end of incubation period. Nitrate removal by *Chlorella* ranged from 43.5% at 100% waste and 100% at 10% waste concentration. Maximum inorganic phosphate removal by *Chlarella* (96.1%) was attained at 100% waste concentration. *Scenedesmus Obliquus* efficiency for removing the major pollutants ranged between 68.4% and 93.1 of initial for ammonia; 41.7% and 87% of initial for nitrate and 38.5% to 80.4% of initial for reactive phosphate (Table 1) . At 10% and 20% effluent concentrations nitrates were not utilized by *Scenedesmus*, removal (negative or zero). Ammonia was depleted totally by *Spirulina* at 10% 20% and 40% effluent dilutions, yet it recorded 84.1% and 70.1% of initial at 80% and 100% concentrations. Nitrates and phosphats were utilized completely by *spirulina* at 10% concentration. Phosphate removal ranged from 62.5% to 84.6% of initial (Table 1). The efficiency of the three microalgae together in a population in removing the major pollutants amounted to 78.1% to 94.5% of initial for ammonia, 50% to 85% of initial for nitrate and 33.3% to 80.4% of initial for reactive phosphates. Maximum percentage removal of ammonia by microalgal population was attained at 40% while maximum nitrate and phosphate removal were recorded at 80% sewage concentration. It is of particular interest to notice that *Chlorella* and *Spirulina* showed higher efficiency in nutrient removal than *Scenedesmus* and microalgal population particularly at 10%, 20% and 40% waste dilutions.

Table (1): Initial and final concentrations of nutrients, their amounts consumed after 7 days incubation in culture media of some microalgae with various dilutions of sewage effluent.

Concentration %	Organism	NH_4 mgL^{-1}			NO_3 mgL^{-1}			PO_4 mgL^{-1}		
		Initial	Final	% R.	Initial	Final	% R	Initial	Final	% R
10	Chlorella	3.8	0.2	94.7	0.3	0.0	100	0.6	0.3	50.0
	Scenedesmus		1.2	68.4		0.3	---		0.2	66.7
	Spirulina		0.0	100.0		0.0	100		0.0	100.0
	Population		0.5	86.8		0.15	50.0		0.4	33.3
20	Chlorella	7.2	0.0	100.0	0.7	0.3	57.2	1.3	0.6	53.9
	Scenedesmus		1.1	84.2		0.8	---		0.8	38.5
	Spirulina		0.0	100.0		0.4	42.9		0.2	84.6
	Population		0.7	90.3		0.2	71.4		0.7	46.2
40	Chlorella	14.5	2.0	86.2	1.2	0.3	75.0	2.4	0.7	70.8
	Scenedesmus		1.0	93.1		0.7	41.7		1.2	50.0
	Spirulina		0.0	100.0		0.5	58.3		0.9	62.5
	Population		0.8	94.5		0.3	75.0		0.6	75.0
80	Chlorella	28.3	5.1	81.9	2.0	0.9	55.0	4.5	1.0	77.8
	Scenedesmus		2.5	91.2		0.6	70.0		1.1	75.6
	Spirulina		4.5	84.1		0.8	60.0		1.2	73.3
	Population		6.2	78.1		0.2	85.0		0.9	80.0
100	Chlorella	38.8	9.4	75.8	2.3	1.3	43.5	5.1	0.2	96.1
	Scenedesmus		7.6	80.4		0.3	87.0		1.0	80.4
	Spirulina		11.6	70.1		0.8	65.2		1.6	68.6
	Population		6.5	83.3		0.4	82.6		1.1	78.4

% R = Percentage removal.

Furthermore, it is to be noted that the removal of ammonia was more effective than that of nitrates and phosphates. This is in consistent with the findings of Crumpton and Isenhart (1987) who reported that ammonia assimilation by algae reduces the impact of ammonia loads from secondary treated wastewater since they preferentially assimilate it rather than nitrate or nitrite. In this connection, Li et al. (1991) mentioned that the removal of N was more effective than that of P in the algal ponds with efficiencies of up to 99.3% for N and 48.1% for total P. Also, a relatively high efficiency in ammonia- N removal was reported in high rate algal ponds (Picot et al., 1991 and Colak and Kaya, 1988). On the other hand, Hensman (1986) found that the amount of nitrogen and phosphorus which may be removed from the effluent by algal assimilation mechanism is dependent upon the intracellular content of algal biomass and the amount of biomass which may be abstracted daily as productivity or yield. Moreover, Li et al. (1991) mentioned that nitrogen and phosphorus are necessary for the algae to grow and reproduce, and the increase in algal chl a in the colony was markedly related to the removal of N and P. The higher the content of chl a in the water, the more effective the removal of N and P from the water in high rate algal ponds. El-Halouani et al. (1993) concluded that nitrogen and phosphorus removal by high rate algal ponds is correlated mainly with phytoplanktonic activity which controls biological nutrients assimilation and pH levels.

The results in Table 2 show that the secondary treatment of sewage with activated sludge resulted in reduction in total suspended solids, BOD, COD nitrate and phosphate. Percentage removal of BOD and COD was 67.9% and 74.4% initial respectively. In this stage ammonia content increased from 31.7 mgL^{-1} to 38.8 mgL^{-1}, with no removal since bacteria and protozoa can grow normally without light. pH value ranged from 6.5 to 8.9 due to algal photosynthesis. When the wastewater samples were filtered before use in tests, the effect of both suspended matter and the toxicants adsorbed on its surface were eliminated. The tertiary treatment of sewage with different microalgae singly and in population resulted in reduction of both COD and BOD values. Percentage removal ranged from 71.3% to 84.1% of initial for BOD and 61.8 to 90.6% of initial for COD (Table2). Algal population represented the maximum removal percentage of BOD in this study while *Chlorella* showed the maximum reduction in COD. Decreasing trend in water quality parameters were reported by (Colak and Kaya, 1988; Tripathi and Shukla, 1991; Aziz and NG 1992). Percentage reduction in BOD and COD from 75 to 95 and 72 to 91, respectively were reported by Govindan (1984).

According to the classificatio of the National Water the water quality of finally treated effluents after cultivation of *Chlorella* in media enriched with 10%, 20% and 40% sewage effluents lie within the category of good quality class in relation to ammonia and nitrate levels while it lies within the category of moderate quality class in relation to phosphate and BOD. At 80% and 100% sewage dilutions ammonia level in *Chlorella* treated cultures reported poor or bad quality (ammonia < 3.11 mgL^{-1} council unit) (Table 1). The water quality of *Scenedesmus*

Table (2): Efficiency of tertiary treatment with different microalgae on 100 % sewage after 7 days incubation.

Parameter (mgL^{-1})	Raw sewage	2ry treatment		3ry treatment							
				Chlorella		Scenedesmus		Spirulina		Population	
		Value	% R	Value	% R	Value	% R	Value	% R	Value	% R
pH	6.5	7.2		7.5		7.4		8.9		7.7	
T.S.S.	82.0	12.0	85.4	3.0	75.0	3.0	75.0	5.0	8.3	3.0	75.0
BOD	97.7	31.4	67.9	6.0	80.9	9.0	71.3	8.0	74.5	5.0	84.1
COD	122.5	31.4	74.4	7.0	90.6	12.0	61.8	8.0	74.5	9.0	71.3
Ammonia	31.7	38.8	--	9.4	75.8	7.6	80.4	11.6	70.1	2.5	93.6
Nitrate	5.7	2.3	59.7	1.3	43.5	0.3	87.0	0.8	65.2	0.4	82.6
Phosphate	7.2	5.1	29.2	0.5	90.2	0.7	86.3	1.6	86.6	0.3	94.1

% R = Percentage removal.

cultures treated with 1010%, 20% and 40% sewage dilutions fall within the category of good quality when dealing with nitrates while effluents derived from media with 80% and 100% sewage are classified as poor quality dealing with ammonia and BOD. The water quality of *Spirulina* treated cultures treated with 10%, 20% and 40% waste dilutions falls within the category of good quality when dealing with ammonia and nitrate and moderate quality when dealing with phosphates. Complete consumption of nutrients was noticed at 10% sewage *Spirulina* treated cultures. The water quality of algal population treated cultures represents the highest amount of BOD removal in this study (84.1).

In conclusion, application of tertiary treatment by microalgae provides high quality effluent that can be reused in many agricultural and industrial uses. In addition, the produced algal biomass is important economically because of their high nutritive value.

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معالجة المخلفات السائلة بإستخدام الطحالب

عفت فهمى شبانة
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فى هذه الدراسة تم استخدام المزارع الطحلبية المنفردة والمجتمعة لاختيار مدى كفاءتها فى عملية معالجة مياه الصرف الصحى. اثبتت المعالجة بواسطة الطحالب كفاءة عالية فى إزالة أغلب الملوثات حيث تدرجت نسب التخلص من العناصر كالآتى: ٧٠,١- ١٠٠٪ للأمونيا، ٤١,٧-١٠٠٪ للنترات وكذلك ٣٣,٣٪ الى ٩٦,١٪ للفوسفات.

كما تسببت المعالجة بالطحالب فى انخفاض تدريجى فى محتوى الأكسجين الحيوى الممتص ٨٤,١٪ والاكسجين الكيماوى المستهلك الى ٩٠,٦٪، كما سجلت الطحالب المستخدمة معدلات نمو وأصباغ كلوروفيل أ عالية فى التركيزات ١٠، ٢٠ وكذلك ٤٠٪ من المخلف حيث سجلت ٢,٧جم/ لتر لطحلب Chlorella ٢,٢جم/ لتر طحلب Scenedesmus ، ٢,٤جم /لتر لطحلب Spirulina وكذلك سجلت ٣,١جم/ لتر من العشائر الطحلبية.

كان معدل المعالجة اكثر كفاءة فى الامونيا عنها فى النترات والفوسفور وكذلك أثبتت Chlorella كفاءة اعلى للمعالجة من باقى الكائنات.