

A Study on The Effect Of Temperature on The Treatment of Industrial Wastewater Using *Chlorella Vulgaris* Alga

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Abstract

Laboratory experiments were performed to study nutrient uptake by the unicellular green microalgae (*Chlorella vulgaris*) grown in batch system uses Al-Asady Factory for Seeds and Animal Feed (Mahmodia/Iraq) industrial wastewater as culture media. The initial concentration of *C.vulgaris* was 1×10^6 cells/ml with 48 hrs of incubation in wastewater at different temperatures ranges (20-35) $^{\circ}$ C where changes in COD, BOD, total nitrogen and total phosphorus concentration in the effluent were calculated.

The results show that the removal efficiencies of COD, BOD, nitrogen and phosphorus are 88%, 89%, 92% and 89% respectively. The optimum temperature at which highest removal efficiencies were obtained was 30 $^{\circ}$ C.

Keywords : wastewater treatment, nutrient removal, microalgae, *Chlorella vulgaris*

دراسة تأثير درجة الحرارة على معالجة المياه الصناعية بأستعمال طحلب (*Chlorella vulgaris*)

الخلاصة

في مزرعة اعتمد فيها نظام الوجبات بأستعمال المياه الصناعية لمعمل الأسدي لإنتاج البذور والعلف الحيواني (المحمودية/عراق) كمزرعة لتنمية هذا الطحلب. كان التركيز الابتدائي للطحلب (cell) انجزت دراسة مختبرية لمعالجة المياه الصناعية بأستعمال طحلب من نوع *Chlorella vulgaris* 1×10^6 لفترة حضن 48 ساعة تحت درجات حرارة تتراوح ما بين (20-35) $^{\circ}$ م. اوضحت النتائج ان أعلى كفاءات الفصل في كلامن للـ COD 88% , BOD 89% , للنتروجين 92% و للفسفور 89% عند درجة حرارة 30 مئوية .

Introduction

Biological treatment processes accomplish oxidation of organic materials in wastewater by microbial activity such as activated sludge, lagoons or anaerobic processes and photosynthesis of micro algae which are being used to reduce some physiochemical factors such as pH, biochemical oxygen demand (BOD) and chemical oxygen demand (COD) [1,2].

Microalgae are microscopic photosynthetic organisms which are found in both marine and fresh water environments [3]. They have received more attention in recent years. They have several uses, they can serve as water bioremediation agents, as feed for aquaculture ,sometimes are used as feed for human in Chinese vegetable known as (*fat chay*) and animals feed, and in pigment production [4] . Microalgae also have ability to absorb heavy metals [5]. It can produce biogas, hydrogen, methane and biodiesel for use as fuel. Using micro algae to treat waste water saves energy and reduces greenhouse emissions [6]. They can utilize various organic compounds especially eutrophic compounds containing phosphorus and nitrogen. Phosphorus is often the limiting nutrient for algal blooms or algal growth in seas and lakes. Both nitrogen and phosphorus are major sources of eutrophication and high concentration of nitrogen or phosphorus can cause algal blooms and other hazardous environmental problems [7].

Algae are used for wastewater treatment because of their low cost of the operation: on aeration equipment is used oxygen requirement are provided by natural surface aeration and photosynthesis by micro algae. Bacteria can consume the oxygen released by the micro algae to decompose the organic matter producing carbon dioxide, ammonia and phosphates, which are assimilated by the micro algae [8,9].

Successful treatment of wastewater, with microalgae requires good growth, therefore understanding of the factors that affect growth is essential. The growth rate of algae and cyanobacteria is influenced by physical, chemical and biological factors Table (1) [3, 10].

Many authors studied the effect of algae on the treatment of wastewater. Tarlan *et.al* [2] studied the ability of *Chlorella* to treat a wood-based pulp industry wastewater. They found that 58% of COD, 84% of color and 80% of AOX (Absorbable Organic Xenobiotics) have been removed at 20°C. Travieso *et.al* [8] tested the effect of initial concentration of settled piggery wastewater in the range (250-1100)mg/L on mixed culture of *Chlorella vulgaris* and bacteria, they have found 88% removal efficiencies of COD from initial concentration (250mg/l) for 190 hrs at 20°C. Valderramna *et.al* [10] obtained 61%, 76.6% and 28% removal efficiencies for COD, nitrogen and phosphorus, respectively. by *Chlorella vulgaris* in

the treatment of diluted ethanol and citric acid production industry wastewater. There are no previous studies that deal with the temperature effect on treatment of wastewater. Therefore the aim of this study is to investigate the substrate removal by *Chlorella vulgaris* as well as determine the biokinetic coefficients fluorescent light mounted about 30 cm above the jar in a shaking incubator with glass cover at 200rpm in batch experiments, represented by specific growth rates (μ) and ultimate oxygen demand (L_0).

2- Materials and Methods

The algae (*Chlorella vulgaris*) was taken from laboratories of Biochemical Technology Division/ University of Technology –Baghdad

The composition of inorganic nutrient medium used in algae growth tests was as follows: 0.6 M NaCl, 6.0mM,EDAT (Ethylenediamine tetra-acetic acid), 1.5 mM FeCl₃, 0.4 mM KH₂PO₄, 5.0 mM KNO₃, 20.0 mM NaHCO₃, 5.0 mM MgSO₄.

2-1 Experimental Work

The sets of experiments were carried out at temperature range from (20-35) °C and exposed to natural and. Lighting was provided for 12 hrs with 18W fluorescent lamp to simulate natural field condition [2,7]. pH ranged from (6.8-7), All the tests were run using glass jars of (1L) volume as batch reactors within the experimental period of 2 days (removal efficiency reached its maximum in 2 days) [2]. The microalgae inoculation volume (suspended in microalgae growth media) was placed in each glass jars with an initial concentration of *C. vulgaris* (1×10^6) cell per ml, the seed and animal feed production

industry waste water used in batch experiments as substrate. The characteristics of the raw wastewater are summarized in Table (2) (were tested according to [12]).

2-2 Analytical Techniques

The analyses carried out in the experiments were chemical oxygen demand (COD), biochemical oxygen demand (BOD), total nitrogen, and total phosphorus. After the removal of algal cells by centrifugation at 300rpm, analysis was performed according to standard methods for the examination of water [12]. COD was measured by excess potassium dichromate and organic compounds were oxidized by using chromic and sulfuric acid at 150°C for 2 hrs. After oxidative digestion, the remaining potassium dichromate was titrated with ferrous ammonium sulfate, then the oxygen equivalent was calculated. BOD was measured as oxygen difference between the initial sample and after incubation for 5 days at 20°C. Total nitrogen and phosphorus tests were carried out in the Ministry of Environment laboratories. Specific growth rate (μ) was measured by taking the samples after 48 hr and counting the number of cells by light microscopy using Neubauer Hematocytometer: Specific growth rate (μ) was determined by the formula

$$\mu = \frac{\ln N_{t_1} - \ln N_{t_0}}{t_1 - t_0}$$

where N_{t_0} = indicates the number of the cells at the beginning and N_{t_1} =indicates, the duration of incubation [10]. and (L_0) ultimate oxygen demand = $BOD_5/0.68$ [1]

3-Results and Discussion

Figures (1) and (2) show the influence of temperature on COD and BOD values. It was found that at temperature 30°C there is a decrease in COD from 560mg/l to 66mg/l and BOD decrease from to 135mg/l to 14mg/l. It could be observed that at temperature higher than 30°C the removal efficiency of BOD and COD decreases because the activity of microorganism becomes lower after this temperature which means ability to remove pollutants decreases. The removal efficiencies were 88%, 89% for COD and BOD respectively as shown in Table (3). This result is in a good agreement with the conditions detected by environmental protection agency (EPA) [1]. Previous studies in Europe and U.S.A have shown that the nutrient removal efficiency by ponds is higher in summer than in winter [13]. Figure(3) shows that the highest removal of nitrogen and phosphorus is achieved at temperature 30°C and they were 92%, 89% respectively.

Microalgae can utilize nitrogen for their growth and phosphorus is a micro- nutrient essential for growth, which is taken up by algae as inorganic orthophosphate (PO_4^{-3}) [7,8]. The nitrogen and phosphorus removal efficiencies vary depending on the media composition and environmental conditions such as algae species, the light/dark cycle, the initial nutrient concentration, aeration, retention time and temperature [13].

In addition to COD and BOD removal, biokinetic constants (μ and L_0) were also calculated to help in understanding the behavior of *C.vulgaris* in the treatment of

wastewater at different temperatures, as shown in Table (3). Figure (4) shows the relationship between temperature and growth rate constant (μ), where growth rate constant increases with increasing temperature of growth until reaches maximum value of (1.5 day^{-1}) at 30°C, then it begins to decrease which means that beyond 30°C, the activity of microorganism (algae) begins to decrease and this agrees with the theory of growth phases for batch culture (lag- growth- death phases) as described in detail by Fogler [14].

4- Conclusions

The experimental study of the treatment of Al.Asady Factory for Seeds and Animal Feed industrial wastewater by using microalgae (*Chlorella Vulgris*). The following conclusions ar drawn :

- 1- Temperature has been considered as the most important physical factor influencing the efficiency of nutrient removal because it directly affects the metabolic rate of micro-organisms , algae.
- 2- Chemical oxygen demand (COD) and biochemical oxygen demand (BOD) removal efficiency were (88%) and (89.6%) respectively, at 30°C during 48 hrs.
- 3- Culture of algae provides efficient nitrogen and phosphours removal in the treatment of the industrial waste water.
- 4- Growth rate constant (μ) is affected by temperature of growth where maximam growth was (1.5 day^{-1}) at 30°C.

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Table [1] Factors that influence algae growth in a high rate alga pond [4, 11]

Physical and Chemical	Light(quality, quantity) Temperature Nutrient concentration O ₂ and CO ₂ pH Salinity Toxic chemical
Operational Factors	Mixing Pollution rate Depth Addition of bicarbonate Harvesting frequencz

Table[2] Characteristics of Al-Asady Plant for Seed and Animal Feed Industrial Wastewater

Property	Unit	Value
Temperature	°C	25
pH		7.2
Total dissolved solid (TDS)	mg/L	960
Conductivity	m.s/cm	1840
Total suspended solid (TSS)	mg/L	612
Total phosphorus	mg/L	10.5
Total nitrogen	mg/L	13.0
Biochemical Oxygen Demond (BOD)	mg/L	135
Chemical Oxygen Demond (BOD)	mg/L	560
Turbidity	NTU	22
Chloride	mg/L	138

Table[3] Removal efficiencies and biokinetic constant

Different Temperature	COD% removal	BOD% removal	$\mu_{1/d}$	L ^o
20	73.2	68.88	0.8	61.76
25	80.3	76.3	1.015	47.05
30	88	89.6	1.5	20.58
35	83.9	80.	1.2	39.7

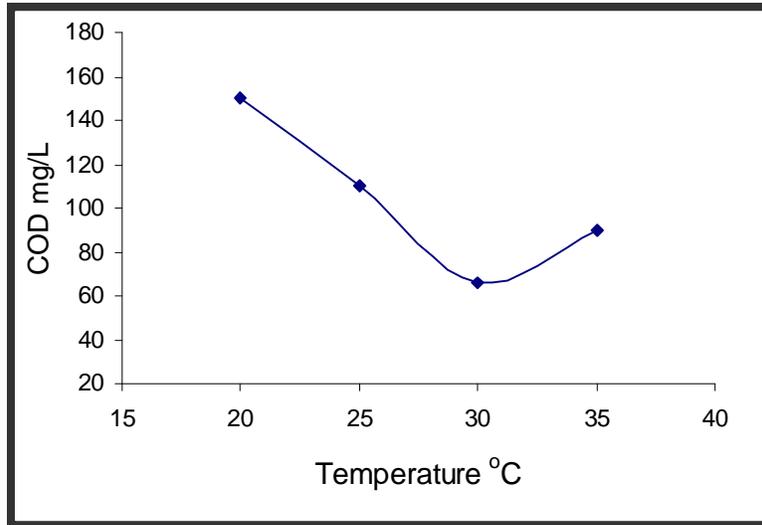


Figure (1) Effect of different temperatures on COD(original concentration was 560 mg/l as COD)

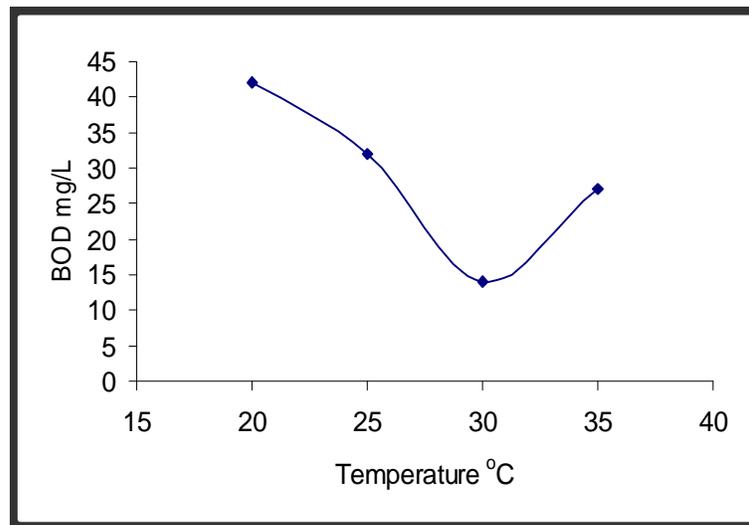


Figure (2) Effect of different temperatures on BOD (original concentration was 135 mg/l as BOD)

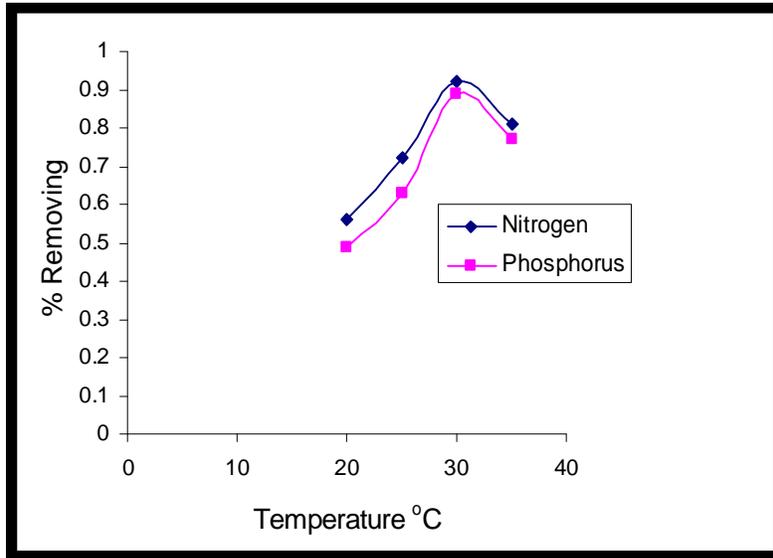


Figure (3) Removing percentage of total nitrogen and total phosphorus from wastewater by microalgae at different temperatures for 2 days

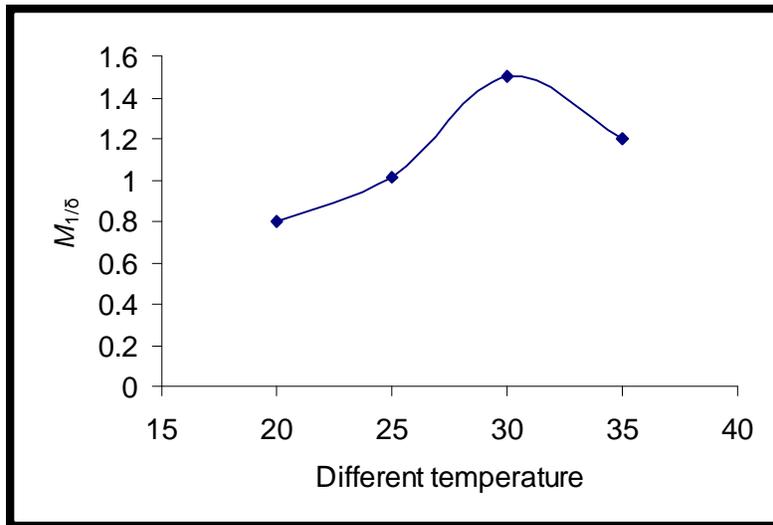


Figure (4) Relationship between growth rate (μ^{-1}) and heat for 2 days