J. Egypt. Acad. Soc. Environ. Develop., 18 (1): 33-46 (2017) **(D-Environmental Studies)**
E.mail: easedjournal_egypt@yahoo.com **ISSN 1110-8770**
www.eased.byethost13.com **eISSN 2314-5471 (online)**

Bioremediation of some chemical pollutants from Fayoum industrial area, Egypt

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ABSTRACT

Bioremediation of industrial wastewater using algae and bacteria is the main goal of this study. Samples were collected from industrial drain water of chemical industrial area (Kom Oshem, Fayoum, Egypt). The main microorganisms tested in this study were *Chlorella vulgaris* and *Microccocus luteus*. The growth of algae and bacteria on wastewater was estimated either singly or dually regarding their efficiency in biodegradation of pollutants of wastewater.

The results revealed that *C. vulgaris* and *M. luteus* caused a removal of nitrogen, phosphorus, potassium and magnesium from wastewater either singly or dually. Dual bio treatment achieved the best removal of pollutants from waste water. It reduced phosphorus by a percentage of (78.71%), nitrate (65.46%), potassium (49.9%) and magnesium (78.8%) within incubation period of 16 days.

Key words: Bioremediation, industrial wastewater, *Chlorella vulgaris* and *Microccocus luteus.*

INTRODUCTION

One of Egypt's environmental dangers is increasing of soil salinity and acidity. The fertility of soil is threatened by untreated industrial wastewater from many factories that lacked pollution control. Untreated wastewater affects farm lands, agricultural productivity and public health of human and animals. Shi (2009) indicated that these problems are one of the serious concerns among different environmental issues in the society and environmental laws.

Treatment of wastewater is mandatory to safe human beings and to protect our environment. Around the world, most of the wastewater treatment use chemical precipitation methods to remove phosphorus from wastewater, but this is not efficient and has many disadvantages. Mehta and Gaur (2005) mentioned that these techniques may be ineffective when concentration of metals in wastewater is in range between 10–100 mg^{-1}.

Economic and energy efficient nitrogen and phosphorus removal technology is the right way to overcome these problems by introducing an alternative biological method called bioremediation. It is a pollution control technology that uses biological systems to catalyze the degradation or transformation of various toxic chemicals to less harmful forms. It is less expensive than other technologies that are used for cleanup of wastewater (Vidali, 2001). Microalgae have generally fast growth, low cultivation cost, capability to assimilate wastes, and efficient in converting solar energy into biomass. Prabha *et al*. (2016) reported that, algae are important bioremediation agents and are already being used in wastewater treatment. Kshirsagar (2013) reported that, *C. vulgaris* have high removal capacity for nitrate and COD. Sharma and Khan (2013) suggested that growing algae in nutrient-rich sewage wastewater offers a new option of applying algae to manage the nutrient load and after phycoremediation. According to Chalivendra (2014), *C. vulgaris* taken from Pleasant Hill Lake were used as candidate species for bioremediation of wastewater loaded with nitrogen (N) in the form of nitrates and phosphorous (P) in the form of phosphates. Kaoutar *et al*. (2014) mentioned that, algae have an important role in controlling and bio-monitoring of organic pollutants in aquatic ecosystems. El-Sheekh *et al*. (2016) revealed that, both *C. vulgaris* and *C. salina* were highly efficient and having a potential to reduce pH, total dissolved solids (TDS), biological oxygen demand (BOD), chemical oxygen demand (COD), nitrate, ammonia and phosphate.

According to Zhuang *et al*. (2010), halophilic microorganisms play an important role in the biological treatment of saline wastewater as decontamination pathways of organic contaminants, heavy metals and nutrients. Karigar *et al*. (2011) carried out an advanced bioprocess technology to reduce the toxicity of the pollutants and also to obtain novel useful substances by using enzymes from various microorganisms. Both algae and bacteria affect each other´s physiology and metabolism, although bacteria have often been considered as mere contamination of algae cultures. However, in the last few years, the scenario has changed. Nowadays, algae-bacteria interactions are being seen as promising in biotechnology, as some recent studies have shown a positive effect of algae-bacteria interaction on algal growth, which is the essential step in algal biotechnology (Fuentes *et al*. 2016). Safonova *et al*. (2004) showed significant decrease in the content of the pollutants by using algal-bacterial associations. According to Hernandez (2006), combination treatment of microalgae with bacteria was capable of removing up to 72% of phosphorus from the wastewater. De-Bashan and Bashan (2010) used immobilized eukaryotic microalgae and several prokaryotic photosynthetic cyanobacteria in removing nutrients with the support of plant growth-promoting bacteria. Olguín (2012) suggested dual purpose in algae-bacteria relationship, the first: microalgae-bacteria-based systems for treating wastewater and the second is production of biofuels and chemical products.

So, the main goal of this study is to bio-remediate industrial wastewater of the chemical area of Kom Oshem, Fayoum, Egypt, using algae and bacteria either singly or dually.

MATERIALS AND METHODS

1- Growth medium and culture conditions

1.1- Growth medium for algae

The stock algal cultures were received on agar slants obtained from the culture collection of algae in Botany Department, Faculty of Science, Cairo University. They were stored at room temperature (27 ^oC) and illuminated at (40-50 µE m^{-2} s^{-1}), then they were enriched in BG-11 medium (Allen and Stanier, 1968) and incubated for 8 days at 27 ^oC with illumination about (40-50 µE m^{-2} s^{-1}). BG-11constituents were: $NaNO_3$ (150 gL^{-1}), K_2HPO_4 (30 gL^{-1}), $MgSO_4.7H_2O$ (75gL^{-1}), $CaCl_2.2H_2O$ (36gL^{-1}), Citric Acid (6gL^{-1}), Ferric Ammonium Citrate (6 gL^{-1}), EDTA (1 gL^{-1}), Na_2CO_3 (20 gL^{-1}) and Trace Metal Solution. pH was approximately 7.5.

1.2- Culture conditions for algae

To obtain sufficient algal growth for use in wastewater treatment experiments, stock algal cultures were transferred and initially grown in 250 ml Erlenmeyer flasks containing 100 ml of BG-11 at 27 ± 2 °C with cool white fluorescent lamps giving a continuous irradiance of 40-50 µmol$m^{-2}$$sec^{-1}$.

1.3- Growth medium of bacteria

Bacteria were grown in Luria Bertani (LB) medium (Sambrook *et al*., 1989), which consists of tryptone (10.0 gL^{-1}) and yeast extract (5.0 gL^{-1}). The solution was bought to 1L by adding distilled water then autoclaved at 121 °C for 15 minutes. The composition of Luria agar (LA) medium (g L^{-1}) is the same LB media but the last one contains 20 g L^{-1} agar. The solution was supplemented with 1.5 g L^{-1} NaCl.The final pH was 7.5.

1.4- Culture conditions of bacteria

To obtain sufficient bacterial growth for use in wastewater treatment experiments, stock bacterial culture was incubated at 37 °C for 24 hours on an orbital shaker incubator operating at 120 rpm min^{-1}. A total of 10 mL of the pure culture was centrifuged to pellet out the cells, washed twice with sterile physiological saline solution and the suspension was adjusted to optical density of 0.1 at 600 nm which is equivalent to a cell population of about 10^6 cells mL^{-1} on the McFarland standard. Bacterial suspension was stored in test tubes in a refrigerator at 4 °C.

2.Experimental setup

2.1 Experimental Setup of bio treatment of wastewater by algae

Serial dilutions of wastewater of 0%, 20%, 40%, 60%, 80% and 100% were prepared in 250 mL Erlenmeyer flask containing the respective percentage of wastewater then completed to 100 mL by distilled water. The diluted wastewater autoclaved at 121°C for 20 minute, cooled then inoculated by equal volumes (5 mL) of each algal organism. All treatments were carried out in triplicates. Different treatments were subjected to four cool white fluorescent lamps (Philips F40T12/DX 40 Watts) giving a continuous irradiance of 40-50 $\mu molm^{-2}sec^{-1}$, placed horizontally and parallel to the front and back of Erlenmeyer flasks till ending the experiment. The temperature was about 27 ± 2 °C.

2.2 Experimental Setup of bio treatment of wastewater by bacteria

Serial dilutions of wastewater were prepared as described above. Each dilution was prepared in 100 mL Erlenmeyer flasks containing the respective percentage ofwastewater, diluted to 50 mL volume, then autoclaved as mentioned above and inoculated by equal volumes (1 mL) of each bacterial organism. All treatments were carried out in triplicates, and incubated at 37±2 °C till ending the experiment.

2.3 Experimental setup of dual bio treatment of wastewater by both of algae and bacteria

Serial dilutions of wastewater were prepared as described above. Each dilution was prepared in 250 mL Erlenmeyer flasks containing the respective percentage of waste water, then it was diluted to 100 mL volume, then autoclaved and inoculated by equal volumes (5 mL) of *C. vulgaris* and (1 mL) of *M. luteus.* All treatments were carried out in triplicates then subjected at room temperature 27 ± 2 °C with four cool white fluorescent lamps (Philips F40T12/DX 40 Watts) giving a continuous irradiance of (40-50 $\mu molm^{-2}sec^{-1}$) placed horizontally and parallel to the front and back of Erlenmeyer flasks till the experimental end.

3 Growth estimation

3.1.Extraction and determination of photosynthetic pigments of algae

The photosynthetic pigments chlorophyll-a were determined using the spectro-photometric method recommended by Metzner *et al*. (1965). A known volume of algal culture was homogenized in 85% aqueous acetone, then kept for 6 hours in a refrigerator. The homogenate was centrifuged and the supernatant was made up to a known volume with 85 % acetone, then measured against a blank of pure 85% acetone at three wave lengths: 452, 644 and

663 nm using Perkin Elmer UV spectrophotometer, taking into consideration the dilution made. It was possible to determine the concentration of pigment fractions as mg/mL using the following equations:

$C_a=10.3\ E_{663} - 0.918\ E_{644}$

$C_b=19.7\ E_{644} - 3.87\ E_{663}$

Carotenoid = $4.2\ E_{452} - (0.0264\ C_a + 0.426\ C_b)$

Where, C_a = Chlorophyll a, C_b = Chlorophyll b, C_{x+c} = Total carotene

3.2 Determination of algal cell counts

Algal cell counts were performed using Haemocytometer apparatus.

3.3 Optical density

Growth estimation of bacteria was recorded by determination of optical density using colorimeter (Bio system BTS 320) at 670 nm of liquid cultures.

3.4 Estimation of growth rate

Growth rate was estimated according to the equation of Wahidin *et al.* (2013):

$\mu = [\ln (N_2 - N_1)] / [t_2 - t_1]$

Where, N_1 and N_2 are the cell number concentration (cell mL^{-1}) at time t_1 and t_2

3.5 Estimation of division rate

The time required to duplicate the cell number: division rate (K) was estimated according to the equation of Wahidin *et al*. (2013):

$K = \mu \div \ln 2$

3.6 Removal efficiency of pollutants

The removal efficiency of pollutants was expressed as:

Percent removal W % = $100\% [(C_0 - C_i) / C_0]$

Where, C_0 and C_i are defined as the mean values of pollutants concentration at initial time t_0 and time t_i, respectively.

RESULTS AND DISCUSSION

1. Pretreatment and characteristics of wastewater

Wastewater was analyzed for a suite of chemical parameters commonly used to characterize chemical industrial water and are susceptible to affect algal growth (Table 1). The investigated samples of chemical industrial water contain high TDS (Over 1000 ppm) , acidic pH (5.9), It was obviously that, wastewater samples contain high amount of nitrate (27.65 ppm) and phosphorus (967.1 ppm). TDS, pH and phosphorus were detected at levels exceeding the permissible one for the Egyptian drinking water standards.

Table 1. Characteristics of wastewater sample.

No.	Parameter	Result	EPA drinking water standards	Egyptian drinking water standards
1	Color	Colorless	15 color units	-
2	pH	5.9	6.5-8.5	6-9.5
3	TDS	Over 1000 ppm	500 ppm	800 ppm
4	NO_3	27.65 ppm	10 ppm	-
5	P	967.1 ppm	-	25 ppm
6	K	350.0 ppm	-	-
7	Mg	50.0 ppm	0.05 ppm	-
8	Ca	70.9 ppm	-	-
9	Mn	2.76 ppm	0.05 ppm	-
10	Fe	0.71 ppm	0.3 ppm	-
11	Ni	150.0 ppb	1000 ppb	-
12	Zn	72.0 ppb	5000 ppb	-
13	Cr	11.6 ppb	100 ppb	-
14	Cd	5.5 ppb	5.0 ppb	-

Seven metals were detected at the highest level. These were iron, magnesium, manganese, potassium, cadmium, zinc and calcium which are represent the major metals detected in the industrial wastewater. These metals were detected at levels exceeding the primary US Environmental Protection Agency (US EPA, 2012) drinking water standards and/or health advisories, whereas levels of nickel and chromium were under the permissible levels of these standards.

2. Growth of microorganisms on serial dilutions of waste water

2.1 Bio treatment of wastewater using algae

C. vulgaris grew on all serial dilutions of waste water, after inoculating the algae on the wastewater, chlorophyll-a content and cell counts were measured at regular intervals. The initial measured chlorophyll-a content and cell counts which represented baseline was 0.05 ppm and 35×10^4 cell mL^{-1}, respectively. *C. vulgaris* showed the highest growth at 100% wastewater on 40^{th} day of inoculation, in which cell counts of 838×10^4 cell mL^{-1} (Fig. 1) and chlorophyll-a content of 3.69 ppm were detected (Fig. 2). One way analysis of variance indicated that the variation between sampling dates were highly significant (P = 1.3E-14), whereas the variation between different dilutions showed slightly significant difference (P = 0.054).

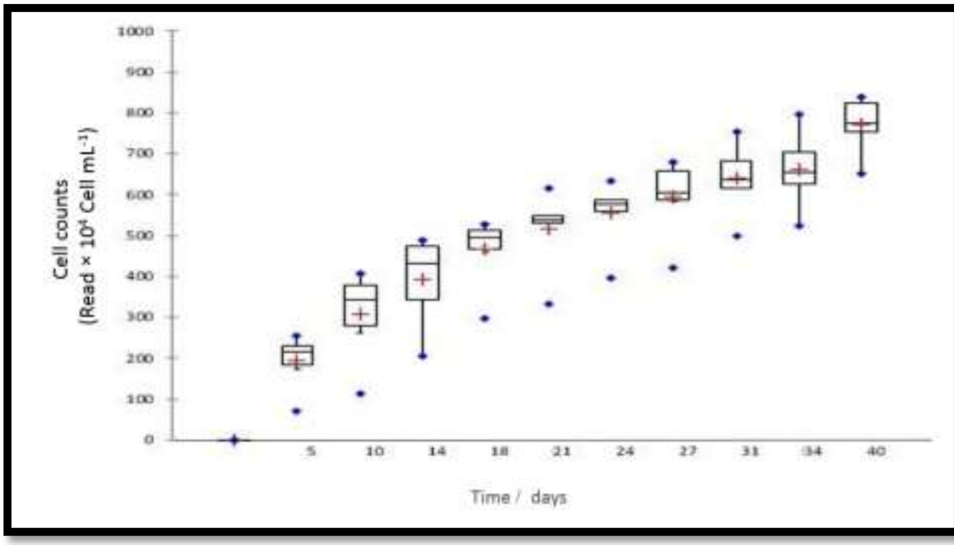

Fig. 1. Estimation of cell counts of *C. vulgaris*on 100% waste water.

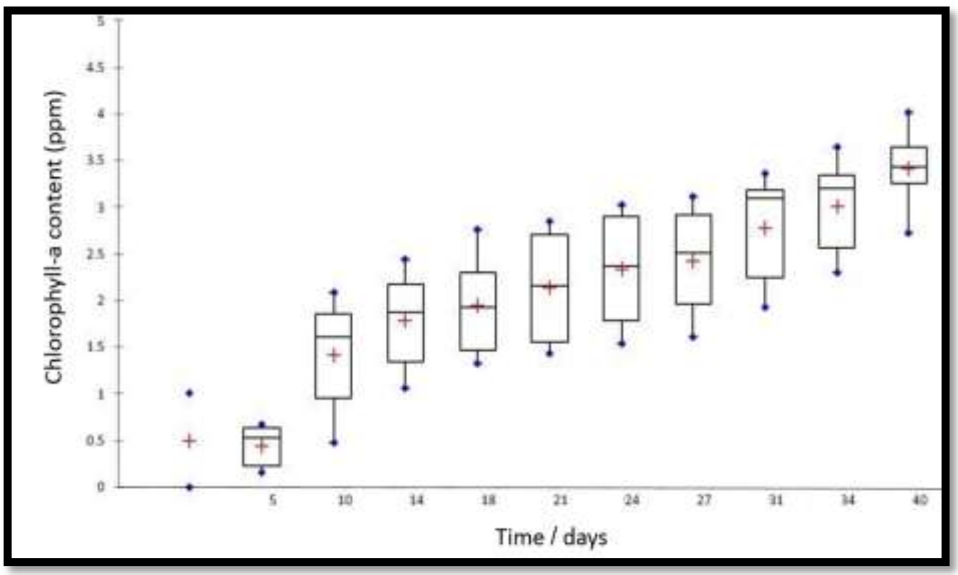

Fig. 2. Estimation of chlorophyll-a content of *C. vulgaris* on 100% waste water.

C. vulgaris showed both the highest and least growth rates of 1.51 and 0.63 at the control on days 27^{th} and 40^{th} of inoculation, respectively (Table 2). The highest average growth rate of 1.115 was measured at the control treatment whereas the least average of 0.993 was found at 60% dilution. One way analysis of variance indicated that the variation between sampling dates were highly significant (P = 6.12E-07), whereas the variation between different dilutions were non-significant (P = 0.84).

Table 2. Estimation of growth rate of *C. vulgaris* on serial dilutions of waste water.

Time / Dil. %	Growth rate (G.R.)										
	5	10	14	18	21	24	27	31	34	40	Average
Control	1.038	0.95	1.158	0.99	1.25	1.32	1.51	1.072	1.23	0.63	1.115
20 %	0.68	0.75	1.12	1.12	1.20	1.37	1.07	1.08	1.09	0.80	1.028
40 %	0.97	0.89	0.99	1.24	1.42	1.06	1.13	0.84	0.80	0.80	1.014
60 %	1.03	0.98	1.08	1.07	1.23	1.18	1.04	0.78	0.73	0.81	0.993
80 %	1.05	1.00	1.14	0.85	1.18	1.13	1.13	0.95	1.07	0.77	1.027
100 %	1.07	1.00	1.10	0.92	1.49	0.96	1.25	0.68	0.99	0.81	1.027

C. vulgaris showed both the highest and least division rate of 2.18 and 0.92 at the control on days 27th and 40th of inoculation, respectively (Table 2). The highest average division rate of 1.61 was measured at the control treatment whereas the least average of 1.439 was found at 60% dilution. One way analysis of variance indicated that the variation between sampling dates were highly significant (P = 7.57E-07), whereas the variation between different dilutions were non-significant (P = 0.84).

Table 3. Estimation of division rate of *C. vulgaris* on serial dilutions of waste water.

Time / Dil. %	Division rate (*k*)										
	5	10	14	18	21	24	27	31	34	40	Average
Control	1.49	1.37	1.67	1.43	1.80	1.91	**2.18**	1.55	1.79	**0.92**	1.611
20 %	0.98	1.09	1.63	1.63	1.74	1.99	1.55	1.57	1.59	1.16	1.493
40 %	1.42	1.29	1.43	1.80	2.05	1.53	1.64	1.22	1.15	1.16	1.469
60 %	1.48	1.42	1.56	1.56	1.78	1.71	1.51	1.13	1.06	1.18	1.439
80 %	1.52	1.45	1.65	1.23	1.71	1.64	1.64	1.37	1.55	1.12	1.488
100 %	1.56	1.45	1.60	1.34	2.16	1.39	1.81	0.98	1.45	1.17	1.491

At the 40th days of the experiment, *C. vulgaris* reduced nitrate concentration from 27.65 to 13.42 ppm by a percentage of 51.46%, and reduced phosphorus concentration from 967.1 to 280.6 ppm by a percentage of 70.98%. Potassium concentration was reduced from 350 to 196.1 ppm by a percentage of 43.97%, whereas magnesium concentration was reduced from 50 to 15.1 ppm by a percentage of 69.7%.

Phosphorus is very important for cell growth and reproduction. Also, photosynthesis requires large amounts of proteins whish are synthesized by phosphorus-rich ribosomes (Agren 2004). Algae use three different bio-processes to transform P into high energy organic compounds: phosphorylation at the substrate level, oxidative phosphorylation, and photophosphorylation (Sancho *et al*., 1997). According to Prescott (1968), green algae demand more nitrogen and phosphorous than do many other species, and they can take up generous nitrogen when the phosphorous content is relatively high.

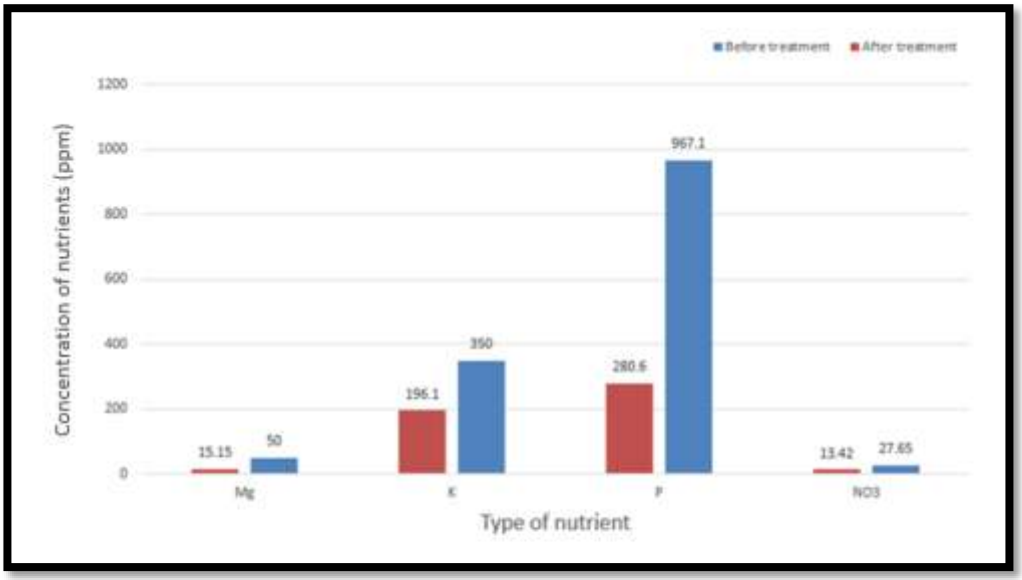

Fig..3. Removal efficiency of nutrients by *C. vulgaris* from 100% wastewater.

Nitrogen is an essential element of cells required for the biosynthesis of a large number of cell components including proteins, nucleic acids (RNA and DNA) and photosynthetic pigment. Chalivendra *et al.* (2013) mentioned that, the growth rate of algae increased with the increase in the nitrate concentration in the media.

The ability of algae to reduce other elements is that, potassium and magnesium play an important role in growth of *C. vulgaris*. Where, magnesium is important in photosynthesis as it is the central atom of chlorophyll molecules and it is also the cofactor of DNA polymerase which manages the cell division. Also, potassium is important for cell growth as it is a cofactor of several enzymes and plays important roles in protein synthesis and osmotic regulation. Abdel-Raouf *et al.* (2012) cited several studies using *C. vulgaris* which reported 50.2% - 86% nitrogen removal and 70% - 97.8% phosphorus removal. Woertz *et al*. (2009) were able to remove >98% of ammonium and >96% of phosphorus with microalgae. Valderrama *et al*. (2002) used *C. vulgaris* to treat recalcitrant wastewater by reduction of ammonium ion (71.6%) and phosphorus (28%).

C. vulgaris exhibited the fastest growth with the greatest biomass yield, this biomass contain protein content of 29.04 % and total phosphorus of 2.36%. Standard deviations and averages of nitrogen, protein and phosphorus contents of *C. vulgaris* on 100% wastewater treatment on 40th day were illustrated in Table (4). According to Richmond (2004), algae biomass typically contains 0.5% to 3.3% phosphorus content.

Table 4. *C. vulgaris* contents after 40th day of 100% wastewater treatment.

	Nitrogen	Protein	Total Phosphorus
Average	4.622 %±0.0024	29.04 % ±0.0017	2.36 % ±0.00027

2.2 Bio treatment of wastewater using *Microccocu luteus*

Microccocu luteus grew slowly on all serial dilutions of waste water. After inoculating *M. luteus*in the wastewater, initial measured optical density which represented as baseline was found to be around 0.01. *M. luteus* showed the highest growth on 80% wastewater on 12th day of inoculation, in which optical density was 0.329, but its optical density (0.207) was found on 100% wastewater.

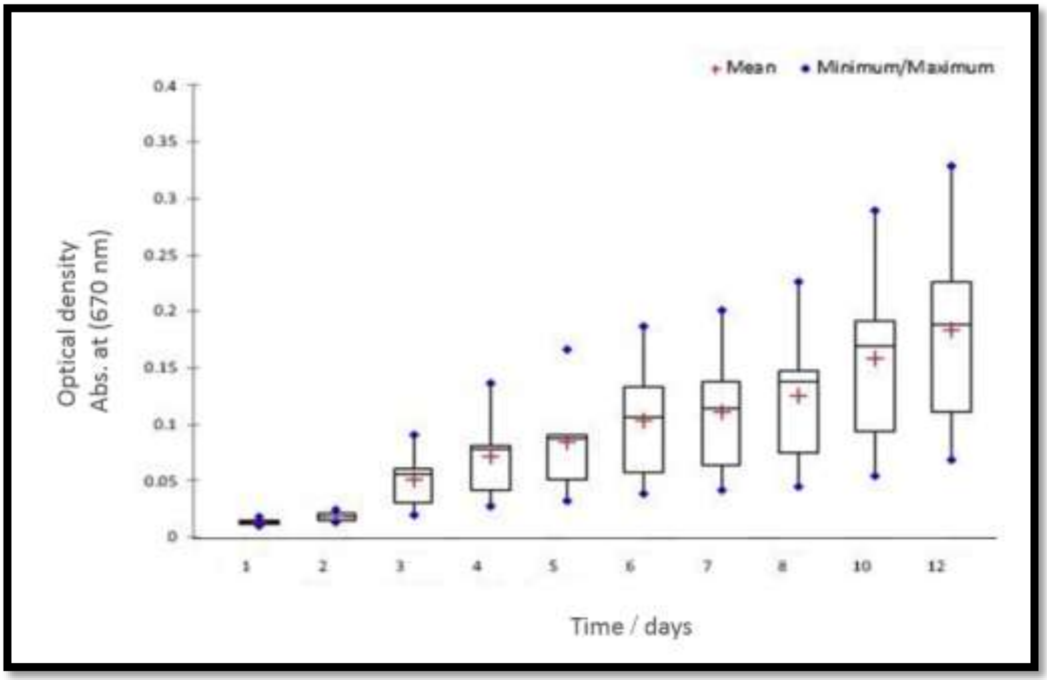

Fig.4. Estimation of optical densities of *M. luteus* on 100% waste water.

During 12th day, *M. luteus* reduced nitrate concentration of waste water from 27.65 to 16.75 ppm with a percentage of 39.4 %, and reduced phosphorus concentration from 967.1 to 675 ppm with a percentage of 30.2%. In the same time, potassium concentration was reduced from 350 to 244.2 ppm with a percentage of 30.2 %, magnesium concentration was reduced from 50 ppm to 35.5 ppm by a percentage of 29 %. Zhuang *et al*. (2010) confirmed that, halophilic microorganisms play an important role in the biological treatment of saline wastewater as decontamination pathways of organic contaminants, heavy metals and nutrients.

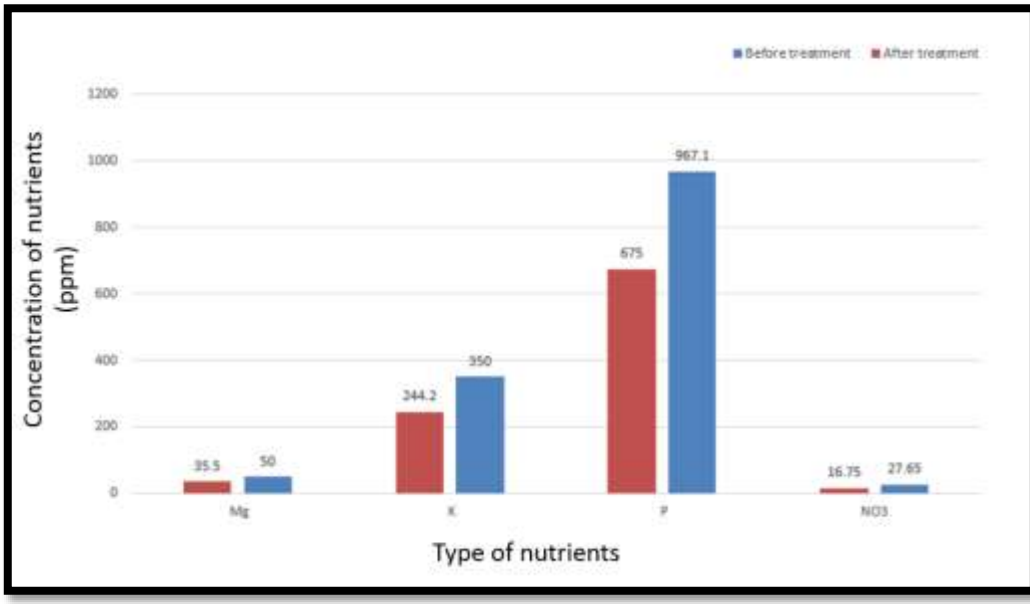

Fig. 5. Removal efficiency of nutrients by*M. luteus*from 80% wastewater.

2.3 Bio treatment of wastewater using both *C. vulgaris* and *M. luteus*

Regarding previous treatments, we designed this bio treatment by using both microorganisms that achieve high growth on wastewater (*C. vulgaris and M. luteus*) in ratio of 10 mL: 1 mL, to activate each other and achieve the best bioremediation. Also, both grew on all serial dilutions of wastewater. After inoculation, initial measured cell counts of *C. vulgaris* which represented as baseline was 25×10^4 cell mL^{-1} and the initial optical density measured of *M. luteus* which represented as baseline was 0.03 at 670 nm.

C. vulgaris showed the highest cell counts in 100% wastewater on 16^{th} day of inoculation, in which cell count was1450×10^4 cell mL^{-1}. It means that, growth of *C. vulgaris* was improved but *M. luteus* growth decreased into 0.081 if we compared it with optical density of single biotreatment of *M.luteus* in 100% wastewater. Some interpretations assumed that algal growth has been shown to be enhanced by growth promoting factors produced by bacteria in algal cultures (Fuentes *et al*., 2016).

On 16^{th} day, dual bio treatment reduced nitrate concentration from 27.65 to 9.55 ppm with a percentage of 65.5%, and reduced phosphorus concentration from 967.1 to 205.8 ppm with a percentage of 78.7%. Simultaneously, potassium concentration was reduced from 350 to 175.3 ppm with a percentage of 49.9% and magnesium concentration was reduced from 50 to 10.6 ppm with a percentage of 78.8%. Hernandez (2006) reported that, combined treatment of microalgae and bacteria was capable of removing up to 72% of phosphorus from the wastewater. De-Bashan and Bashan (2010) used immobilized eukaryotic microalgae and several prokaryotic photosynthetic cyanobacteria, with emphasis on removing nutrients with the support of microalgae growth-promoting bacteria.

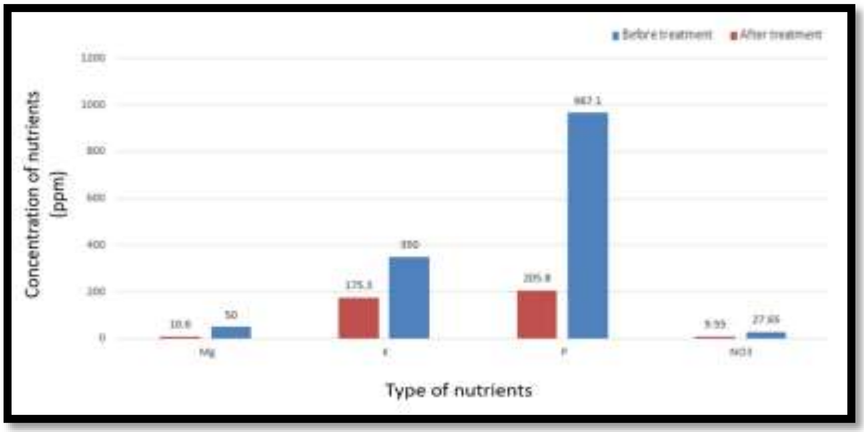

Fig. 6. Removal efficiency of nutrients from 100% wastewater usingdual bio treatment by*C. vulgaris* and *M. luteus*.

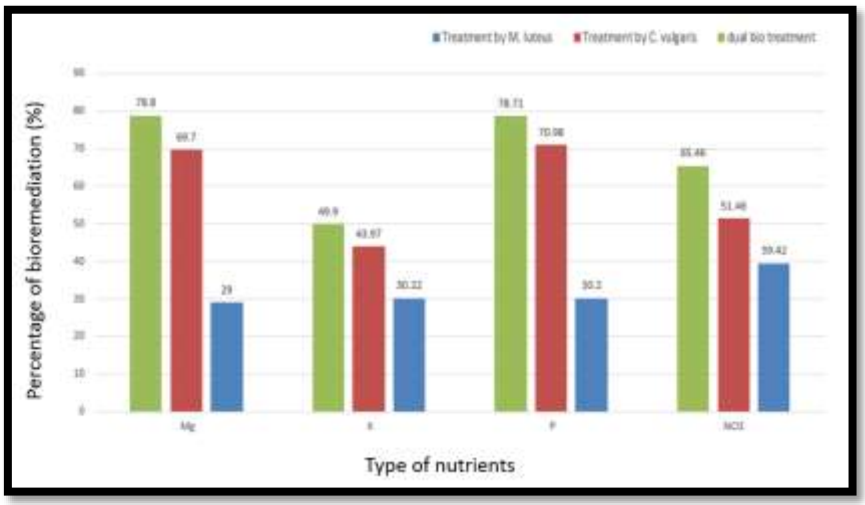

Fig. 7. Comparison between removal efficiency of nutrients in different bio treatment.

Table 5. Comparison of efficiency of bioremediation of P and NO_3using *C. vulgaris* among different researches.

NO_3	P	Reference
51.46%	70.98%	This study
77%		Chamberlin (2002)
	28%	Valderrama *et al*. (2002)
	>96%	Woertz *et al*. (2009)
	33.1–33.3%	Lim (2010)
50.2% - 86%	70% - 97.8%	Abdel-Raouf *et al*. (2012)
90%	62%	Chalivendra (2014)
96.4-99.4%		Halfhide (2014)

The phosphorus removal was much greater than those reported in many other studies using municipal wastewater (Valderrama *et al.*, 2002, Lim, 2010 and Chalivendra, 2014), suggesting that the algae species used in this study is high phosphorus concentration tolerance. The results of this study revealed that total phosphorus contents in *C. vulgaris* was 2.36% which is in consistency with that reported by Richmond (2004), who conducted that algae biomass typically contains 0.5% to 3.3% phosphorus. Thus it is reasonable to conclude that a considerable part of phosphorus was removed by sedimentation and did not assimilate to algal biomass. The removal capacity of NO_3-N is smaller than that reported by many other studies (Chamberlin, 2002,Abdel-Raouf *et al*., 2012,Chalivendra, 2014 and Halfhide, 2014) suggesting that NO_3-Nis not the only nitrogen form that can be assimilated by algae. Matusiak *et al.* (1976), Syrett, (1981), Barsanti and Gualtieri (2006) reported in their studies that, algae can assimilate NH4-N, nitrate, and simple organic nitrogen such as urea and amino acids in the wastewater, but the complicated organic nitrogen could not be directly used.

It was concluded from the present results that the best bio remediation of nitrate, phosphorus, potassium and magnesium from wastewater can be achieved by using dual bio treatment of *C. vulgaris* and *M. luteus*.

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المعالجة البيولوجية لبعض الملوثات الكيميائية من المنطقة الصناعية بالفيوم، مصر

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المستخلص

الهدف الرئيسى من تلك الدراسة هو عمل معالجة حيوية لمياه الصرف الصناعى بالمنطقة الصناعية بكوم أوشيم محافظة الفيوم-مصر بإستخدام الطحالب والبكتريا سواء كان كل على حده أو مجتمعين فى معالجة ثنائية ، و تم فى هذه الدراسة استخدام الطحلب *Chlorella vulgaris*، واستخدام البكتريا *Micrococcus luteus*.

أتضح من تحليل عينات مياه الصرف الصناعى أنها تحتوى على كميات عالية من الفوسفور والنترات والماغنسيوم والبوتاسيوم تجاوزت الحد المسموح به فى مياه الشرب المصرية . وبعد ستة عشر يوما من المعالجة الثنائية الحيوية المستخدم فيها طحلب *Chlorella vulgaris*وبكتريا *luteus Micrococcus* ، وجد أن تركيز النترات فى مياه الصرف الصناعي إنخفض بنسبة 65.5% ،و الفسفور انخفض بنسبة 78.7 % ، والبوتاسيوم انخفض بنسبة 49.9 % وكذلك الماغنسيوم انخفض بنسبة 78.8 % ، يتضح من خلال النتائج السابقة أن تلك المعالجة هى الأفضل لخفض نسبة الملوثات الموجودة بمياه الصرف الصناعي بالمنطقة الصناعية بكوم أوشيم – محافظة الفيوم .