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CHROME BIOSORPTION AND CHEMICAL OXYGEN DEMAND REDUCTION FROM TANNERY WASTEWATER BY AEROBIC ACTIVATED SLUDGE

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ABSTRACT

The leather industry is considered a polluting industry as it generates effluents which are characterized by high amounts of organic and inorganic load. One of the most important elements in the inorganic fraction is chrome. In this investigation, the capability of aerobic activated sludge to biosorp chrome was evaluated in a synthetic chrome solution and in tannery effluent. The wastewater contained a high levels of chrome (3014 mg/l) and COD (10306 mg/l). The estimated COD:N:P ratio was as high as 2015:30:1. In the synthetic chrome experiment, three concentrations of chrome were studied *i.e.* 100, 500, 1000 mg chrome/l, where and chrome removal percentages reached 10.6 , 9.3 and 11.9 , respectively. While in the tannery effluent , the removal percentage was very high; the chrome removal reached 99.98% after 6 days in undiluted and water diluted (1:2) effluent. The chemical oxygen demand (COD) was monitored during the tannery effluent treatment , where a 72.5% reduction at the sixth day was achieved.

***Key words**: activated sludge, chrome biosorption, COD reduction, tannery effluent.*

1. INTRODUCTION

Chromium is found extensively in tanning industry effluent. When wastewater effluent, containing chromium, is discharged into the environment, it poses a serious problem to the quality of such an environment, therefore, removal of chromium from waste water is obligatory in order to avoid water pollution through removal of chromium before wastewater discharge. Legislation by different governments, demands that the concentration of Cr in discharges should be less than 0.5 mg/l (Water quality standards hand book 2nd edition, 1993).

The current methods being employed, such as chemical precipitation, are not able to reduce the chrome concentration to the desired levels. Using various microorganisms as biosorbents for chromium removal, offers a potential alternative to existing methods for its recovery from industrial wastewater (Onyancha *et al.*, 2008).

The discharge of effluents containing Cr (VI) and Cr (III) contains concentrations ranging from tenths to hundreds of mg/l. Hexavalent chromium is considered the most toxic form of chromium, whereas trivalent chromium is much less toxic. Because of these differences in its toxicity, the discharge of Cr (VI) into surface water is regulated below 0.05 mg/l by the U.S. EPA, while total Cr including Cr (III) and its other forms is regulated below 2 mg/l (Park *et al.*, 2004).

The tannery, commonly use basic Cr (III) sulphate [Cr $(H_2O)_5(OH)SO_4$] in the tanning process (Sharma and Adholeya, 2011).

The first report on using bacteria in chrome reduction came out in the late of 1970's. Number of bacterial species have been isolated and shown to be capable of reducing Cr (VI). These species belong to *Pseudomonas, Escherichia, Bacillus, Enterobacter* and Sulphate-reducing bacteria (SRB) including *Desulfovibrio, Desulfomicrobium* and *Desulphotomaculum*. (Park *et al.*, 2004).

Also, the activated sludge can be used as a detoxification agent to reduce Cr (VI) to Cr (III) and to adsorb Cr (III) on the suspended solids (Stasinakis *et al.*, 2004).

The treatment and safe disposal of hazardous organic waste material in an environmentally acceptable manner and at a reasonable cost is a topic of great universal importance. Usually treatment of organic pollutants in wastewater is performed by applying two steps, firstly a chemical treatment, which targets the most non-biodegradable fractions of wastewater; followed by a biological treatment (Mantzavinos and Psillakis, 2004).

Biomass synthesis, oxidation, denitrification

and accumulation are elementary processes that take place in activated sludge during organic compounds COD removal (Dobrzynska *et al.*, 2004).

The aim of this study was to monitor the chrome biosorption and chemical oxygen demand reduction from tannery wastewater using aerobic activated sludge.

2. MATERIALS AND METHODS

2.1. Materials

2.1.1. Activated sludge

Aerobic activated sludge was obtained from Zenien station for sewage water treatment, Giza, Egypt. It was aerated at 200 rpm for 24 h after freshly obtained teo oxidize the organic matter that might be present.

2.1.2. Heavy metal

In a synthetic chrome batch system, $K_2Cr_2O_7$ was used in three Cr^+ concentrations *i.e.*, 100, 500 and 1000 mg/l. Three replicates for each concentration were set up for 500 ml Erlenmeyer flasks.

2.1.3. Instruments

An atomic absorption spectrophotometer (Thermo Electron Corporation) was used to measure heavy metal concentration, a cooling centrifuge (3K30) was used at 4800 rpm for 20 min to separate the cells, a water bath adjusted at 75°C/18 h for digesting cells, a rotary shaker (SI-100) 100 rpm, thermo reactor (CR 3200) for the COD reaction (150°C for 2 h) and a visible spectrophotometer (JEN WAY 6300) for COD determination at 600 nm.

2.1.4. Wastewater samples

Tannery effluent wastewater was obtained from the tannery house in the region of Ein El-Sera, Cairo, Egypt. At the chrome tanning stage of the leather tanning process, chemicals such as chrome sulfate were added to the pretreated leather.

All glassware used for the experimental purposes were washed with 0.1 M HCl before and after each experiment and subsequently rinsed with distilled water (Hussien *et al.*, 2004).

2.2. Methods

2.2.1. Biosorption of chrome from synthetic chromium solution by aerobic activated sludge

The added activated sludge wt/v was 4.93 g/l. Three concentrations (100, 500 and 1000 mg/l) with three replicates of each concentration was placed in 500 ml of distilled water plus $K_2Cr_2O_7$ with activated sludge cells in Erlenmeyer flasks of 1000 ml and shaked on a rotatory shaker at 150 rpm for up to 61 minutes. Periodical samples (20 ml) were taken every 15 min. and centrifuged at 4800 rpm for 20 min. The pellet was digested in 3 ml nitric acid (60%) in a water bath at 75° C for 18 h. The supernatant was acidified by adding one milliliter of nitric acid to ensure availability of the chromium ions for detection by the atomic absorption spectrophotometer. The chromium concentration in both pellet and supernatant was measured using the atomic absorption spectrophotometer. If the sample was diluted the corresponding multiplication was made.

2.2.2. Tannery effluent treatment

Experiments were performed using 500 ml shaking flasks containing 250 ml of the tannery effluent and shaked on a rotary shaker for up to 16 days. Two hundred and fifty of the tannery effluent was placed in 500 ml flasks for each of the following treatments: undiluted control (pH adjusted to 6.0 by NaOH 0.1N), water diluted control (1:2) (without pH modification) and water diluted control (1:2) (pH adjusted to 6 by NaOH 0.1 N). 10% inoculum of activated sludge was added to undiluted tannery effluent (pH adjusted to 7 bt 0.1 NaOH 0.1 N) and to diluted tannery effluent (1:2) (pH adjusted to 6 by NaOH 0.1 N). Periodical samples (20 ml) were withdrawn and centrifuged at 4800 rpm for 20 minutes. The supernatant was acidified by adding one milliliter of nitric acid to ensure availability of chrome for detection by atomic spectrophotometer; chrome was analyzed using the atomic adsorption spectrophotometer after dilution to be in the range of the standard curve.

2.2.3. COD determination

The COD was performed according to APHA (1992). Two and half of the sample to be examined was added into the COD glass tube followed by 1.5 ml of digestion solution and 3.5 ml of sulfuric reagent. Tubes were then covered with screw caps, mixed well and put in the COD reactor at 150 °C for 2 h. Blank and standard solutions were treated with the same manner as well. After digestion, tubes (samples, blank and standard) were allowed to cool and inverted several times and the solids allowed to settle before measuring absorbance. Measurements of the color intensity were carried out by spectrophotometer at 600 nm.

2.2.4. Statistical analysis

Data obtained was statistically analyzed by least significant difference according to Gomez and Gomez (1984), and L.S.D. values at the 0.01 level of significance were used for comparison between means.

3. RESULTS AND DISCUSSION

The tannery effluent was characterized by a high level of COD reached to 10360 mg/l. Considering the N and P contents and the COD:N:P ratio was estimated to 2015:30:1. While the chrome content exceeded 3000 mg/l. but the number of microorganisms did not exceed 3.5cfu/ml.

3.1. Batch biosorption by activated sludge

Fig.(1). illustrates the 100 ppm chrome batch using the activated sludge. Experiment conditions were as follows, temperature 28°C, shaking speed 150 rpm, pH 6 and cell weight 4.93g/L. The supernatant behavior of the 100 ppm chrome batch was totally random and the net reduction at the best points was in the range of 10%. While, the pellet behavior was unlike what was shown in the supernatant in that there was an increase in chrome level at 16 minutes which was maintained till the end of the batch.

Fig.(2). illustrates the 500ppm chrome batch using the activated sludge. Experiment conditions were as follows, temperature 28°C, shaking speed 150 rpm, pH 5.3 and cell weight 4.93g/L. Concerning supernatant of the 500 ppm chrome batch, there was a decrease in chrome from the first minute and this decline was maintained till the end of the batch. This was going in parallel with the increase of the chrome content in the pellet at the same time (first minute) and the continuity of this increment till nearly the end of the batch.

Fig.(3). illustrates the 1000 ppm chrome batch for the activated sludge. Experiment conditions were as follows, temperature 28°C, shaking speed 150 rpm, pH 5.3 and cell weight 4.93g/l. Concerning the supernatant of the 1000 ppm chrome batch, there is a slight decrease in the overall chrome content of the batch, while in the pellet the increase in chrome accumulated started from the first minute and maintained till the end of the batch.

The L.S.D. 0.01 values were 3.52 and 0.003 for the supernatant and pellet, respectively for the three batches illustrated in Figures1, 2, and 3.

The reason for inefficient chrome removal in the three batches might be due to two factors. The first one is that, the chrome used was Cr(VI) which is according to (Ksheminska *et al.* 2005) considered to be highly toxic as it can be easily transported into microbial cells by the sulphate transport system and cause immediate reduction reactions leading to formation of various intermediates which are harmful to cell organelles, proteins and nucleic acids. The second factor is the pH. According to Lin and Yu (2012), the optimum pH for maximum chromium removal is pH 5, where as the mean of pH replicates in the three batches recorded during this study were 6, 5.3 and 5.3 for the 100, 500 and 1000 ppm chrome batches respectively. Those pH values are considered higher than the optimum, and therefore, the chromium removal efficiency decreased due to osmotic and hydrolyzing effect (Sepehr *et al.*, 2005).

Unexpectedly, only a marginal decrease in chromium concentration from solution was observed over the incubation period. Obviously, the biosorption capacity of the microbial cells was low, since the adsorbed chromium on the cells was very low. This could be explained by the activity of the functional groups and metal chemistry in solution (Yao *et al.*, 2009). They also found that, the biosorption capacity began to decrease when pH increased (>5.0).

3.2. Tannery effluent treatment

In the undiluted effluent, the chromium decreased in the solution significantly below 1 mg/l after 6 days of incubation either without or after addition of activated sludge (Table, 1). The observed growth of natural microflora in the bottles without sludge reaching >300 cfu/ml might be responsible for chromium biosorption.

In diluted effluents, however, another trend was observed. There was no growth of the natural microflora until day six; consequently very low chromium reduction (2.6%) was recovered. After addition of activated sludge, the chrome could be reduced significantly to a concentration below 0.1mg/l representing a removal efficiency of more than 99%.

The results indicated that this high removal percentage did not occur in the batch experiments. This may be due to interactions between activated sludge and tannery wastewater components. Sepehr *et al.* (2005) reported that tannery water contains carbohydrates and organic acids (formic acid) that act as a carbon source of bacteria It also contains enzymes such as (tripsin and protease) as well as proteins of hide that act as sources of nitrogen. A tannery wastewater contains phosphorous so it has the most important elements which are essential for the microbial cells activity.

The overgrowth that was observed through all experiments duration confirms the activity of activated sludge. Nies, (1999) stated that, in order to grow in a chromate containing media, a biological system would need to develop a detoxifying tool by either extracellular or intracellular accumulation.

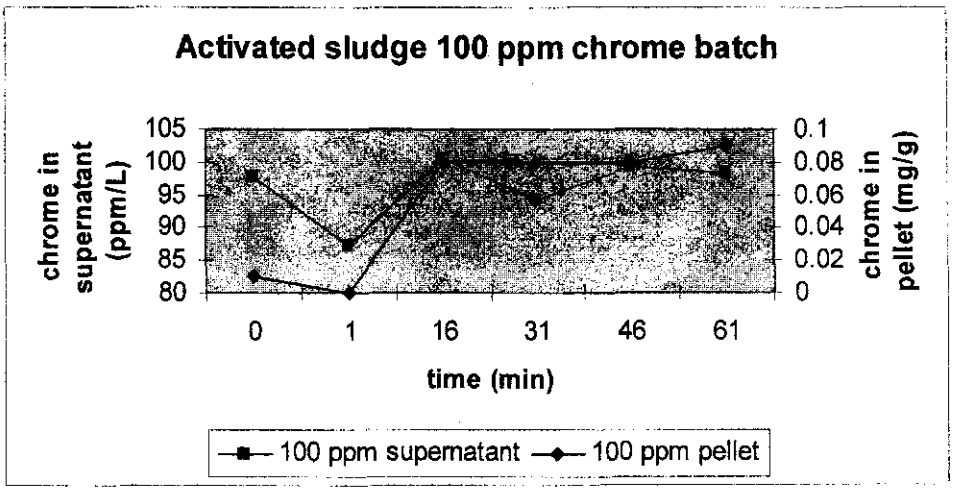

Fig. (1) : 100 ppm chrome batch of the activated sludge.

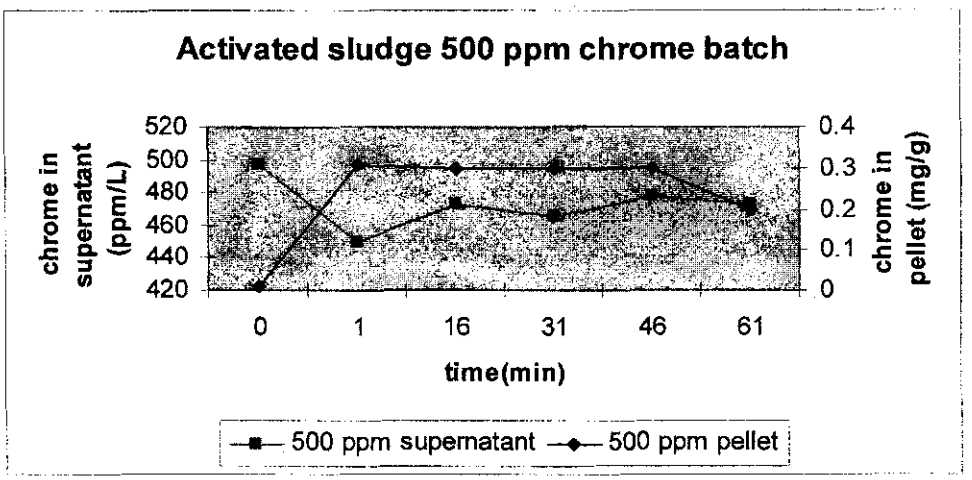

Fig. (2) : 500 ppm chrome batch of the activated sludge.

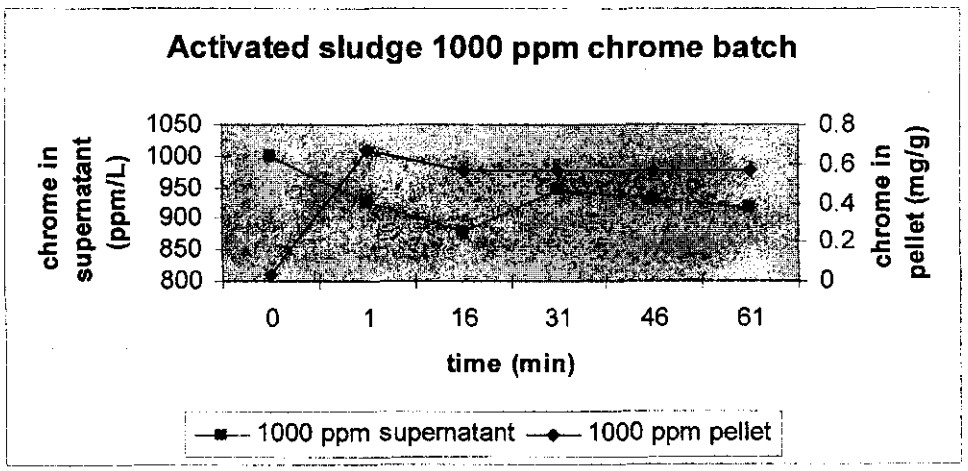

Fig. (3): 1000 ppm chrome batch of the activated sludge

Table (1): Effect of control and activated sludge treatments of tannery effluent experiment.

Treatment	Parameters	Time (days)						
		0 day	2 days	4 days	6 days	10 days	13 day	16 day
Control tannery effluent (undiluted) pH=6	COD(ppm)	10360	9060	9760	6860	6810	10610	8960
	pH	6.08	5.9	6.1	6.7	5.5	6.5	7.4
	T.C.(cfu/ml)	3.5	5	100	Over growth	Over growth	Over growth	Over growth
	Cr(ppm)	3104	---	---	0.440	---	0.195	---
Control Tannery effluent (diluted1:2) No pH modification	COD(ppm)	6010	5810	4060	3410	3960	8010	5710
	pH	2.4	2.9	6	5.7	5.7	5.3	3
	TC(cfu/ml)	No growth	No growth	No growth	No growth	1	4	20
	Cr(ppm)	1035	---	---	1007.90	---	1149.70	---
Tannery effluent treated with activated sludge (undiluted) pH modified to 6	COD(ppm)	29513	9360	8710	4860	8910	8210	8260
	pH	6.3	5.5	6.9	5.4	4.5	5.4	8
	TC(cfu/ml)	Over growth	Over growth	Over growth	Over growth	Over growth	Over growth	Over growth
	Cr(ppm)	3104	---	---	0.310	---	2.535	---
Wastewater (1:2) treated with activated sludge (diluted) pH not modified	COD(ppm)	9210	5360	5310	930	3210	3060	3310
	pH	3.4	3	3.6	4.4	3.9	3.9	4
	TC(cfu/ml)	Over growth	Over growth	Over growth	Over growth	Over growth	Over growth	Over growth
	Cr(ppm)	1035	---	---	0.060	---	0.815	---
L.S.D. 0.01	**L.S.D. for the COD values for the interaction between time and wastewater treatments =8.896** **L.S.D. for the chrome values for the interaction between time and concentration=5.250**							

----= not determined

3.3. Chemical oxygen demand (COD) reduction

In undiluted effluent, results indicated that the COD significantly decreased in both treatments either with or without activated sludge (Table.1). However, the highest COD removal efficiency of 48.9% (representing 775.5 mg COD removed $l^{-1}d^{-1}$) was achieved in the presence of activated sludge after 6 days compared to only 33.8% (representing 583.3 mg COD removed $L^{-1}d^{-1}$) without sludge after the same time. Further incubation up to 16 day did not give any obvious decrease in COD concentration. The observed decrease in COD concentration in the absence of activated sludge might be due to the increase in natural microflora counts, which was doubled 30 fold by the 6^{th} day of incubation.

While in a diluted effluent, a similar trend of COD decrease was observed in both treatments, before and after addition of activated sludge. Although the initial pH value was within the acidic range the removal percentages of COD in both treatments were relatively as high as (43.3 and 64.1, respectively) than those observed in undiluted effluent (Table 1).

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الإدمصاص الحيوى للكروم وإختزال المتطلب الكيماوى للأكسجين من مخلفات مياه المدابغ بواسطه الحمأة الهوائيه النشطة

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ملخص

تعتبر صناعه الجلود من الصناعات الملوثه للبيئه حيث ينتج عنها مخلفات سائله تتميز بمحتواها المرتفع من المكونات العضوية وغير العضوية. ويعتبر الكروم من أهم العناصر التى تتواجد ضمن المكونات غير العضوية. يتضمن هذا البحث اختبار قدرة الحمأه الهوائية النشطة على إدمصاص الكروم على مستوى مياه الكروم المجهزة معمليا وعلى مستوى مياه مخلفات صناعة الدباغة. اشارت النتائج المتحصل عليها من تحليل مياه مخلفات المدابغ انها تحتوى على محتوى عالى من الكروم (٣٠١٤ ميلليجرام /لتر) وكذلك كانت نسبه المحتوى العضوى : النيتروجين : الفسفور ١:٣٠:٢٠١٥

بالنسبه لمياه الكروم المجهزة معمليا تم عمل ثلاث تركيزات من الكروم (١٠٠، ٥٠٠ و ١٠٠٠ ميلليجرام/ لتر) حيث قدرت نسبه الازاله المئويه للكروم ب ١٠,٦ ، ٩,٣ و ١١,٩ بعد ١ ، ١٠ و ١٦ دقيقة على التوالى.

أما بالنسبه لمعدل أزاله الكروم فى مياه مخلفات المدابغ فقد كانت مرتفعه جدا حيث و صلت نسبه الكروم المزال الى ٩٩,٩٨% بعد ٦ ايام فى كل من مياه المدابغ المخففه و غير المخففه . كذلك تم تقدير قيم المتطلب الكيماوى للاكسجين خلال عمليه المعالجة الحيويه لمياه مخلفات المدابغ حيث بلغت نسبه التناقص ٧٢,٥% فى اليوم السادس من المعالجة .