

# Biological treatment of saline wastewater in a rotating biodisc contactor by using halophilic organisms

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**Abstract** Biological treatment of saline wastewater presents unique difficulties as a result of plasmolysis of microorganisms in the presence of salt. Removal of salt from wastewater before biological treatment by reverse osmosis or ion exchange operations are rather expensive. Inclusion of halophilic organisms in activated sludge culture seems to be a more practical approach in biological treatment of saline wastewater.

A synthetic wastewater composed of diluted molasses, urea,  $\text{KH}_2\text{PO}_4$ ,  $\text{MgSO}_4$  and various concentrations of salt (0–5% NaCl) was treated in a rotating biodisc contactor (RBC). A salt tolerant organism *Halobacter halobium* was added onto activated sludge culture (50%) and used as inoculum. Effects of important process variables such as A/Q ratio, COD loading rate, feed COD concentration, salt concentration and liquid phase aeration on system performance were investigated. An empirical mathematical model describing the system's performance as a function of important process variables was developed and constants were determined by using the experimental data.

## 1

### Introduction

Biological treatment of saline wastewater usually results in low BOD removal performance because of adverse effects of salt on microbial flora. High salt concentrations (>1% salt) cause plasmolysis and/or loss of activity of cells. There are limited number of studies on biological treatment of saline wastewater. Early studies with *Bacillus cereus* indicated that endogenous respiration rate of the culture increased with NaCl concentrations up to 1%, but decreased above this concentration (1). Adaptation of *E. coli* to NaCl was studied by Doudoroff (2). It was found that the cells exhibited the greatest degree of adaptability in the early stationary growth phase as compared to other growth phases (2).

Kincannon and Gaudy found that reductions in salt concentration caused more severe effects on microorgan-

isms than increases in salt concentration (3, 4). Approximately 30% decrease in BOD removal efficiency was observed when fresh-water sludge was dosed with 30 g/l NaCl solution. However, when an activated sludge acclimated to 30 g/l NaCl was dosed with fresh medium, BOD removal efficiency decreased approximately 75%. Rapid changes in salt concentration caused immediate release of cellular constituents resulting in an increase in soluble COD(4). Sludges grown in high salt concentrations exhibited low carbohydrate and protein but high lipid and RNA content (3, 4). Rapid shifts in salt concentration was reported to have more adverse effects than gradual shifts (3, 4, 5, 6).

Lawton and Eggert have studied the effects of salt on trickling filter slimes. Lower degree of reduction in BOD was observed when salt concentration exceeded 20 g/l (7).

Effects of salt on extended aeration process was investigated by Stewart et al. Temporary reductions in BOD removal efficiency were observed when severe changes in salinity were combined with heavy organic and hydraulic loadings (8).

Ludzack and Noran investigated the effects of salt concentrations up to 20 g/l on the activated sludge process (9). High salt concentrations resulted in low BOD removal and flocculation efficiencies. Nitrification was also adversely affected by salt concentration (9).

Effects of salt on performance of a RBC unit was investigated by Kinner et al. in treatment of a domestic wastewater containing seawater salinity (10). It was shown that salinity levels used did not significantly effect the performance of RBC unit and up to 64% COD removals were reported at a hydraulic loading rate of  $0.08 \text{ m}^3/\text{m}^2 \cdot \text{d}$  (10).

Wastewaters rich in halogenated organics were treated at different salt concentrations and BOD removal performance was found to decrease with increasing salt concentration (11).

Woolard and Irvine studied treatment of hypersaline wastewater by a moderate halophile in a sequencing batch reactor (12) and also in a biofilm reactor (13). Up to 99% phenol removal was obtained from a 15% saline wastewater.

Kargi and Uygur tested various types of microbial culture for treatment of saline wastewater in an aerated percolator unit (14). *Halobacter* added activated sludge culture was found to be the best among other cultures tested. Effects of salt concentration on the system's performance were also investigated in this study.

Kargi and Dinçer studied biological treatment of synthetic saline wastewater in an aerated tank operating in

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fed-batch mode (15, 16). Adverse effects of high salt concentrations were significantly alleviated by using salt tolerant organisms (*Halobacter*) along with activated sludge culture. Nearly 85% COD removal efficiency was obtained with 5% salt concentration in the presence of *halobacter* within 9 hours of operation time.

There are numerous studies in literature on biological treatment of domestic and industrial wastewaters by RBC systems (17–21). However, biological treatment of saline wastewater in an RBC system was not studied extensively. Domestic wastewaters with seawater salinity were treated in an RBC unit (10). However, low COD removal efficiencies (nearly 60%) were obtained because of salt content of wastewater.

Major problems encountered in biological treatment of saline wastewaters can be summarized as follows:

1. Limited extent of adaptation: Conventional cultures cannot be effectively used to treat saline wastewaters of larger than 3–5% salt. Salt adaptations of cultures are easily lost when subjected to salt free medium.
2. Sensitivity to changes in ionic strength: Shifts in salt concentration from 0.5 to 2% usually cause significant disruptions in system performance. Even with acclimated cultures adequate performance requires constant ionic composition. Rapid changes in salt concentrations cause more adverse effects than gradual changes. Equalization to constant salt concentration is essential before treatment of saline wastewaters.
3. Reduced degradation kinetics: Biological degradation rates of organic compounds decrease with increasing salt concentration. Therefore, saline wastewaters should be treated at lower F/M ratios.
4. High effluent suspended solids concentration: Salt content in wastewater reduces the populations of protozoa and filamentous organisms resulting in low sedimentation efficiencies.

In the lights of aforementioned studies, this study was designed to investigate biological treatment of saline wastewater in an RBC unit by using halophilic organisms along with activated sludge culture. A continuously operating rotating biodisc contactor was used since it is commonly encountered in treatment of domestic wastewaters for small communities. *Halobacter* was added to activated sludge inoculum culture in order to overcome low treatment efficiencies reported with saline wastewaters. Effects of important process variables (A/Q ratio, COD loading rate, salt and feed COD concentrations) on system performance were investigated. An empirical mathematical model was developed describing the effects of important process variables on system's performance.

## 2 Materials and methods

### 2.1 Experimental set-up

A schematic of the biodisc system used in this study is shown in Fig. 1. The system consists of the following units: wastewater aeration tank containing biodiscs, driving motor, sedimentation tank, recycle pump and pipes, air

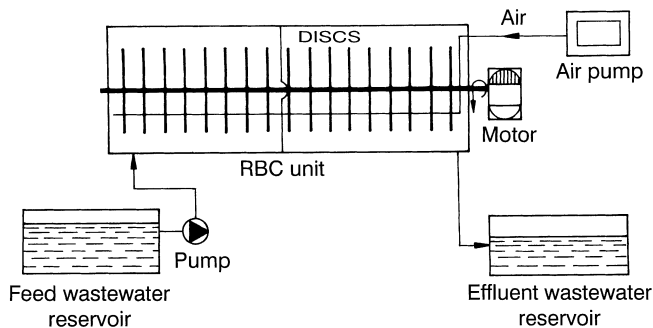


Fig. 1. A schematic diagram of the Rotating Bio-Contactor (RBC)

pump and perforated tubes immersed in liquid phase, feed wastewater pump, feed waste water reservoir. Wastewater tank had two sections containing 20 discs in each amounting a total of 40 discs. Discs were made of polypropylene with diameters of 20 cm. Total disc surface area was  $A_T = 2.512 \text{ m}^2$ . Discs were 40% immersed in water resulting in wet surface area of  $A_w = 1.5 \text{ m}^2$ . Total liquid volume in aeration tank was  $V_L = 9.5 \text{ l}$ . Disc surface area per unit liquid volume was  $a = 264 \text{ m}^2/\text{m}^3$ . Discs were mounted on a shaft and were rotated with a constant speed of  $n = 5 \text{ rpm}$  with a driving motor. Liquid phase aeration was provided by an air pump and perforated tubes placed in wastewater tank underneath the discs.

### 2.2 Wastewater composition

Synthetic wastewater used throughout the studies was composed of diluted molasses, urea,  $\text{KH}_2\text{PO}_4$ ,  $\text{MgSO}_4$  and various concentrations of salt (0–5% NaCl) resulting in COD/N/P = 100/10/1 in the feed. COD, total nitrogen and phosphorous concentrations in the feed wastewater were  $\text{COD}_o = 5000 \pm 200 \text{ mg/l}$ ,  $N_T = 500 \pm 20 \text{ mg/l}$  and  $P = 50 \pm 2 \text{ mg/l}$ , respectively.  $\text{MgSO}_4$  concentration in the feed was 0.05 g/l throughout the studies. Salt concentrations were adjusted by adding required amounts of NaCl into the feed wastewater.

### 2.3 Organisms

Activated sludge culture was obtained from the wastewater treatment plant of PAK-MAYA Bakers yeast company in Izmir, Turkey. *Halobacter halobium* was obtained from the ATCC, USA and cultivated in our laboratory. Activated sludge and *Halobacter* cultures were mixed in 1/1 ratio (i.e., 50%–50%) and used as inoculum throughout the studies.

### 2.4 Experimental procedure

Experiments were started batch wise. About 9 l of synthetic wastewater was placed in the wastewater tank containing biodiscs, inoculated with activated sludge + *Halobacter* culture (50%/50%) and was operated batchwise for about ten days. Biofilm thickness on the discs reached about 1.5–2 mm at the end of batch operation. During continuous operation, feed wastewater placed in a deep refrigerator ( $T < 4 \text{ }^\circ\text{C}$ ) was fed to the biodisc unit with a constant flow rate and the effluent was discharged to the sewerage. Temperature, pH and dissolved oxygen of the medium

during operation were  $T = 27 \pm 1 \text{ }^\circ\text{C}$ ,  $\text{pH} = 8 \pm 0.2$  and  $\text{DO} = 3 \pm 0.5 \text{ mg/l}$ , respectively. Temperature was controlled with the aid of a sensor, heating element and an automatic controller. pH was controlled manually with the addition of dilute sulfuric acid. Aeration rate was adjusted to result in  $\text{DO} > 3 \text{ mg/l}$  in wastewater.

**2.5 Analytical methods**

Samples were removed from the feed reservoir and effluent twice a day and were centrifuged at 6000 rpm for 1/2 hour. COD analysis were carried out on clear supernatant according to Standard Methods (22).

Biomass concentrations in liquid phase were determined by filtering 10 ml liquid samples from a milipore filter (0.45  $\mu$ ) and drying the filter paper in an oven at 103  $^\circ\text{C}$  until constant weight. Total weight of biomass on the biodiscs were determined by washing several (2 or 3) biodiscs and determining the biomass concentrations by filtration and drying.

Dissolved oxygen (DO) measurements were made by using a Solomat 520 C, DO Analyzer and a DO probe.

**3 Results and discussion**

Experiments were designed to investigate the effects of major process variables on COD removal efficiency and rate. Major variables considered were A/Q ratio, COD loading rate, salt concentration and feed COD concentration.

**3.1 Effect of A/Q ratio**

Experiments were performed with a constant disc surface area (total of 40 discs with a surface area of  $A_T = 2.512 \text{ m}^2$ ) at different wastewater flow rates for 1% salt concentration. Each experiment was conducted for about six days at steady-state until three consecutive COD measurements of the effluent were the same. Variation of effluent COD concentrations and COD removal efficiencies with A/Q ratio are depicted in Fig. 2. Effluent COD concentration decreased from a value of  $S_e = 1700 \text{ mg/l}$  at  $A/Q =$

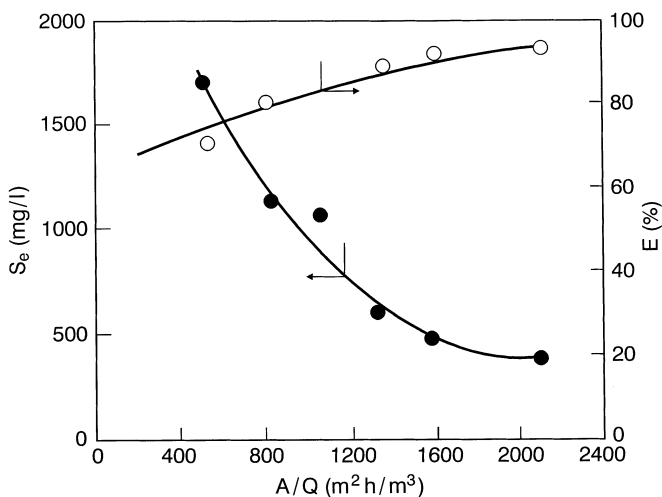


Fig. 2. Variation of effluent COD concentration ( $S_e$ ) and COD removal efficiency (E) with A/Q ratio. (1% salt)

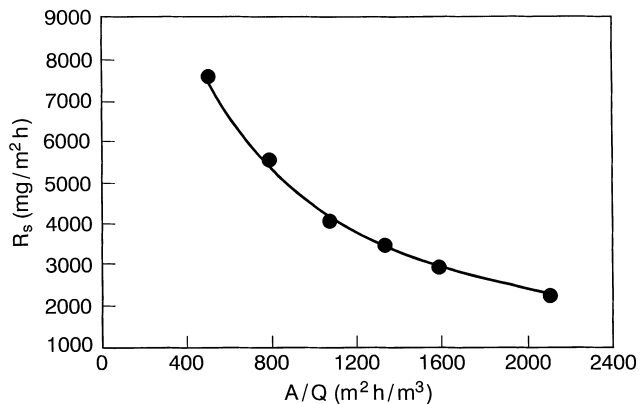


Fig. 3. Variation of COD removal rate ( $R_s$ ) with A/Q ratio (1% salt)

$530 \text{ m}^2 \cdot \text{h/m}^3$  ( $E = \%71$ ) to  $S_e = 400 \text{ mg/l}$  at  $A/Q = 2110 \text{ m}^2 \cdot \text{h/m}^3$  ( $E = 92\%$ ). This sharp decrease in effluent COD concentration with increasing A/Q ratio is a result of high biomass intensity on disc surfaces at high A/Q values. COD removal efficiency did not change significantly for  $A/Q > 1600 \text{ m}^2 \cdot \text{h/m}^3$ .

Variation of COD removal rate ( $R_s$ ,  $\text{mgCOD/m}^2 \cdot \text{h}$ ) with A/Q ratio is depicted in Fig. 3. COD removal rate based on disc surface area decreased with increasing A/Q ratio as a result of decreasing effluent COD concentration (i.e, the rate is a function of effluent COD concentration based on Monod equation). Maximum COD removal rate was  $R_s = 7700 \text{ mg/m}^2 \cdot \text{h}$  at  $A/Q = 530 \text{ m}^2 \cdot \text{h/m}^3$  and the minimum was  $R_s = 2300 \text{ mg/m}^2 \cdot \text{h}$  at  $A/Q = 2110 \text{ m}^2 \cdot \text{h/m}^3$ .

**3.2 Effect of COD loading rate**

COD loading rate ( $L_s = QS_o/A$ ,  $\text{mgCOD/m}^2 \cdot \text{h}$ ) was varied by varying feed wastewater flow rate at constant  $S_o = 5000 \pm 200 \text{ mg/l}$  and  $A = 2.512 \text{ m}^2$ . Variation of effluent COD concentration and COD removal efficiency as a function of COD loading rate is depicted in Fig. 4. As COD loading rate increased from  $L_s = 2500 \text{ mg/m}^2 \cdot \text{h}$  to  $L_s = 11000 \text{ mg/m}^2 \cdot \text{h}$ , the effluent COD concentration

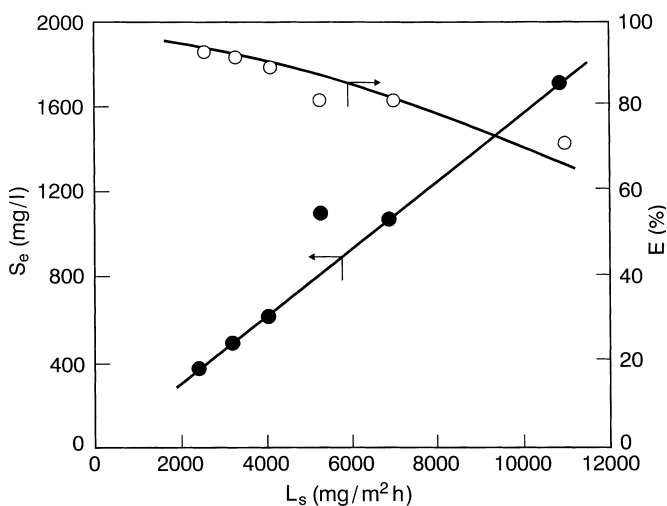


Fig. 4. Variation of effluent COD concentration ( $S_e$ ) and COD removal efficiency (E) with COD loading rate ( $L_s$ ). (1% salt)

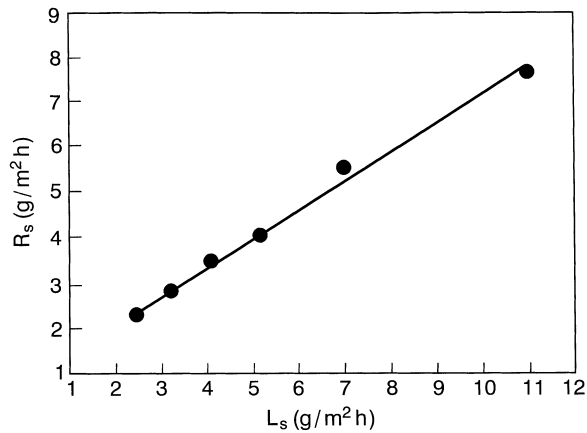


Fig. 5. Variation of COD removal rate ( $R_s$ ) with COD loading rate ( $L_s$ ). (1% salt)

increased almost linearly from  $S_e = 400$  mg/l ( $E = \%92$ ) to  $S_e = 1700$  mg/l ( $E = \%71$ ). For COD removal efficiency of larger than  $E > 90\%$  the system should be operated at  $L_s = 2000\text{--}3000$  mgCOD/ $m^2 \cdot h$ .

Figure 5 presents variation of COD removal rate ( $R_s = Q(S_o - S)/A$ , mgCOD/ $m^2 \cdot h$ ) with COD loading rate ( $L_s$ ). COD removal rate based on disc surface area increases almost linearly with COD loading rate as a result of increasing effluent COD concentration (i.e., the rate is a function of the effluent COD concentration based on the Monod equation).

### 3.3 Effect of salt concentration

Feed wastewater salt (NaCl) concentration was varied between 0% and 5% at  $A/Q = 1060$   $m^2 \cdot h/m^3$  ( $O_H = 4$  h) and  $S_o = 5000 + 200$  mg/l constant values. Fig. 6 depicts variation of effluent COD concentration and COD removal efficiency with salt concentration. COD removal efficiency with salt free wastewater was  $E = 95\%$  ( $S_e = 320$  mg/l). As

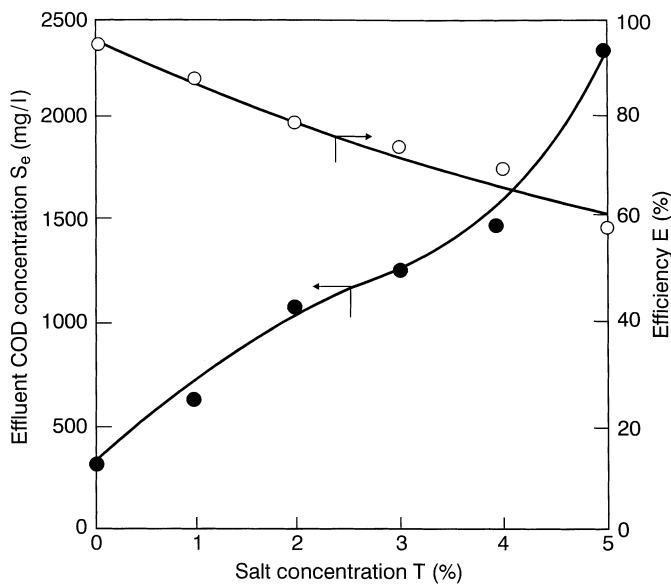


Fig. 6. Variation of effluent COD concentration ( $S_e$ ) and COD removal efficiency (E) with salt concentration. ( $A/Q = 1060$   $m^2 \cdot h/m^3$ )

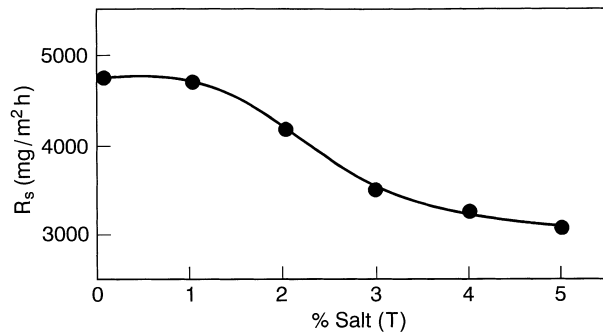


Fig. 7. Variation of COD removal rate ( $R_s$ ) with salt concentration ( $A/Q = 1060$   $m^2 \cdot h/m^3$ )

a result of adverse effects of salt on microbial population efficiency dropped almost linearly, down to  $E = 60\%$  ( $S_e = 2350$  mg/l) at 5% salt concentration.

Variation of COD removal rate ( $R_s$ ) with salt (NaCl) concentration is depicted in Fig. 7. The rate decreased with salt concentration quite significantly for salt concentrations larger than 2%. One percent salt concentration did not effect the rate significantly. However, the rate dropped down to  $R_s = 3100$  mg COD/ $m^2 \cdot h$  at 5% salt concentration from a value of  $R_s = 4750$  mg COD/ $m^2 \cdot h$  for salt free wastewater indicating a 35% reduction.

### 3.4 Effect of feed wastewater COD concentration

Feed wastewater COD concentration was varied between  $S_o = 2,500$  mg/l and  $S_o = 11,500$  mg/l and the effluent COD concentrations were determined at 1% salt concentration and  $A/Q = 1060$   $m^2 \cdot h/m^3$  ( $O_H = 4$  h) constant values. Fig. 8 depicts variation of effluent COD concentration and COD removal efficiency with feed COD concentration. As the feed COD concentration increased from  $S_o = 2,400$  mg/l to  $S_o = 11,500$  mg/l the effluent COD level increased from  $S_e = 560$  mg/l ( $E = \%76$ ) to  $S_e = 7,700$  mg/l ( $E = \%31$ ) resulting in a decrease in COD removal efficiency from  $E = 76\%$  to 31%.

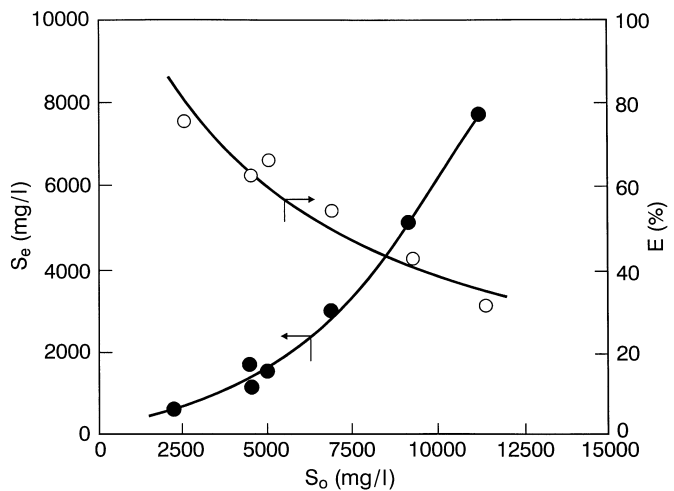


Fig. 8. Variation of effluent COD concentration ( $S_e$ ) and COD removal efficiency (E) with feed COD concentration ( $S_o$ ). (1% salt,  $A/Q = 1060$   $m^2 \cdot h/m^3$ , un aerated liquid phase)

Unlike other experiments performed with aerated liquid phase, this experiment was conducted with unaerated liquid phase. The reason for low COD removal efficiencies obtained in this set of experiments is a result of lack of forced aeration in liquid phase. With aerated liquid phase for  $S_o = 5000 \text{ mg/l}$  (1% salt,  $A/Q = 1060 \text{ m}^2 \cdot \text{h} / \text{m}^3$ ,  $O_H = 4 \text{ h}$ ) the COD removal efficiency was  $E = 90\%$  as compared to  $E = 67\%$  obtained with unaerated liquid phase. Therefore, liquid phase aeration improved the COD removal efficiency significantly (approximately 23% increase over the unaerated efficiency).

#### 4

##### Empirical mathematical model

Efficiency of COD removal with important process variables,  $A/Q$  and salt concentration can be described by the following empirical equation:

$$E = f(A/Q, T) = a_1 (A/Q)^b \cdot a_2 (T)^c = a (A/Q)^b \cdot (T)^c, \quad (1)$$

The following equation can be obtained by linearization of Eq. (1):

$$\ln E = \ln a + b \ln (A/Q) + c \ln (T), \quad (2)$$

Experimental data obtained at constant salt concentration ( $T = 10 \text{ kg/m}^3$ ) and different  $A/Q$  values ( $500 < A/Q < 2100 \text{ m}^2 \text{ h/m}^3$ ) were plotted in form of  $\ln E$  versus  $\ln(A/Q)$ . From the intercept and the slope of the best fit line, the following values were obtained for  $a_1$  and  $b$ :

$$a_1 = 0.22, \quad b = 0.21.$$

Similarly, experimental data obtained at constant  $A/Q$  ratio ( $A/Q = 1060 \text{ m}^2 \cdot \text{h/m}^3$ ) with different salt concentrations ( $T = 10\text{--}50 \text{ kg/m}^3$ ) were plotted in form of  $\ln E$  versus  $\ln(T)$ . From the slope and intercept of the best fit line, the following values were obtained for  $a_2$  and  $c$ :

$$a_2 = 1.55, \quad c = -0.23.$$

Therefore, Eq. 2 takes the following form:

$$E = 1 - \frac{S_e}{S_o} = 0.34 (A/Q)^{0.21} (T)^{-0.23}, \quad (3)$$

Eq. (3) can be used to predict the system's performance for variations of  $A/Q$  and salt concentrations within tested ranges of those parameters.

#### 5

##### Conclusions

A mixture of activated sludge culture and *Halobacter halobium* was found to be quite effective in treating saline wastewater in an RBC unit. COD removal efficiency and rate were effected by the  $A/Q$  ratio (or the surface biomass intensity), COD loading rate, feed COD concentration, salt concentration and liquid phase aeration. In order to obtain COD removal efficiencies of larger than  $E > 90\%$   $A/Q$  should be greater than  $A/Q > 1500 \text{ m}^2 \cdot \text{h/m}^3$ ; COD loading rate must be smaller than  $L_s < 4,000 \text{ mgCOD/m}^2 \cdot \text{h}$ ; salt concentration should be lower than  $(T) < 1\%$  and the liquid phase should be aerated.

An empirical model representing the effects of the aforementioned parameters on COD removal efficiency

was developed and the coefficients/exponents were determined by using the experimental data. This equation can be used to predict the system performance for  $500 < A/Q < 2100 \text{ m}^2 \cdot \text{h/m}^3$ ;  $2500 < L_s < 11000 \text{ mgCOD/m}^2 \cdot \text{h}$ ; and  $0\% < (T) < 5\%$ .

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