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**BIOLOGICAL TREATMENT OF SALINE  
WASTEWATER BY USING HALOPHILIC  
ORGANISMS IN AN AERATED PERCOLATOR  
(IMMERSED FILTER) AND ROTATING BIOLOGICAL  
CONTACTOR (RBC) UNITS**

A Thesis Submitted to the  
Graduate School of Natural and Applied Sciences of  
Dokuz Eylül University  
In Partial Fulfillment of the Requirements for  
the Degree of Master of Science in Environmental Engineering, Environmental Technology  
Program

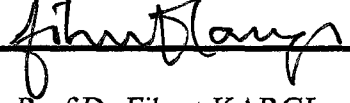
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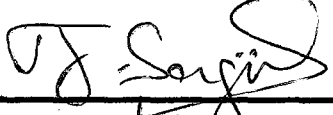
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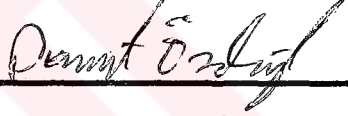
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## M.Sc THESIS EXAMINATION RESULT FORM


We certify that we have read this thesis and that in our opinion it is fully adequate, in scope and in quality, as a thesis for the degree of Master of Science.

  
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Ahmet **UYGUR**



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## ABSTRACT

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Efficiency of wastewater treatment operations near seashore areas is usually low because of high salt content of industrial and domestic wastewater in those areas (‰ 5 - ‰ 35). The reason for this is loss of activity of organisms in biological wastewater treatment operations due to plasmolysis of cells. Removal of salts from wastewater before treatment by reverse osmosis or ion exchange requires high capital and operating costs and is not practical. However, some halophilic organisms (e.g. **Halobacter halobium** or **Dunaliella salina**) can grow and remove carbon and nitrogen compounds from wastewater under saline conditions (‰1-‰10 NaCl). Utilization of these organisms for biological treatment of saline wastewaters has a great potential of improving the treatment efficiency. However, there are no extensive studies on this subject in literature.

Major objective of this study is to improve the treatment efficiency of saline wastewaters by using the **halophilic organisms** *Halobacterium halobium* in a percolator and in a RBC unit and to investigate the system's behavior. Synthetic wastewaters with different salt concentrations were treated in a continuous aerated submerged filter (**PERCOLATOR**) and rotating biodisc unit (**RBC**) by using a mixed culture of organisms containing *H. halobium*. Kinetic analysis of this system was accomplished. Variation of system performance with major process variables was investigated. A mathematical model describing the system behavior was developed and the model parameters were determined by using the experimental data.

These studies provided new technological approaches for biological treatment of saline wastewaters and also contributed to the understanding of behavior of such systems.

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## ÖZET

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Özellikle deniz kıyısındaki evsel ve endüstriyel atıksular tuz içerdikleri için (%o 5 - %o 35), arıtma tesislerinin verimleri düşük olmaktadır. Bunun nedeni yüksek tuz konsantrasyonlarında biyolojik arıtma ünitelerindeki organizmaların plazmoliz nedeniyle parçalanmaları ya da aktivitelerini kaybetmeleridir. Atıksulardan tuzun ters osmoz ya da iyon deęiştiriciler ile giderimi, işletme ve yatırım maliyetlerini artırdığından pratikte uygulanmamaktadır. Oysa bazı tuz tolere edebilen organizmalar (**Halobacterium Halobium** ya da **Dunaliella Salina**) yüksek tuz konsantrasyonlarında (% 1 - % 10 NaCl) ortamdaki karbon ve azot bileşiklerini giderip büyüebilmektedir. Bu organizmaların tuz içeren atıksuların biyolojik arıtımında kullanımı, arıtma verimini artırma potansiyeline sahiptir. Ancak bu konuda literatürde kapsamlı bir çalışma bulunmamaktadır.

Bu projenin amacı tuz içeren atıksuların arıtımında halophilic bir bakteri olan *Halobacterium halobium*'u kullanarak arıtma verimini yükseltmek ve sistemin davranışını bilimsel yöntemlerle incelemektir. Sentetik atıksular sürekli çalışan bir havalandırma percolatör ünitesi ve dönen biyodisk ünitelerinde *Halobacter halobium* içeren karma kültürlerle deęişik tuz konsantrasyonlarında arıtılarak sistemin kinetik analizi yapıldı ve kinetik sabitleri saptandı. Sistemin performansı üzerine önemli deęişkenlerin etkileri incelendi. Sistemin davranışını belirleyen bir matematiksel model geliştirilerek veriler deęerlendirildi ve model parametreleri tayin edildi.

Bu çalışmalar tuz içeren atıksuların biyolojik arıtımına yeni teknolojik yaklaşımlar getirmiş, ve bu tür sistemlerin daha iyi anlaşılmasına olanak sağlamıştır.

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## CHAPTER ONE

# INTRODUCTION

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Saline wastewaters are generated by some industrial establishments and also by domestic activities. Seawater is sometimes used for cooling and also processing purposes for industries located on coastal areas. Certain industries such as salt refineries, cheese and pickle manufacturers generate saline wastewaters.

Biological treatment of saline wastewaters have unique difficulties because of the salt content. High salt concentrations (larger than 1%) cause disintegration of cells because of loss of cellular water (*plasmolysis*) resulting in loss of microbial activity. Low microbial activity in saline waters are observed by many researchers (1-11). When organisms are adapted to saline environment by gradual increases in salt concentration loss of microbial activity is somewhat regained (1,2). However, when placed back to fresh water, organisms go through a severe loss of activity (1-2).

Salt concentrations of less than %1 may cause a slight increase in respiration rate (11). However, high salt concentrations (>1%) are reported to cause severe drops in respiration and BOD removal rates. As a result of cell disintegration and release of intracellular material into wastewater, BOD concentration was observed to increase in saline wastewaters (1). Classical activated sludge units (8, 9), fixed film biological systems (7) and rotating biological discs (4) were used for treatment of saline wastewaters. Adverse effects of salt concentration was observed in all these units with somewhat lesser extent in fixed film systems. Presence of salt is also reported to eliminate protozoa and filamentous organisms from activated sludge culture causing difficulties in sedimentation of the sludge (1,11).

Removal of salt content of wastewaters by reverse osmosis, ion exchange or electrodialysis before biological treatment are rather expensive. Utilization of salt tolerant organisms in biological treatment units seems to be a more reasonable approach for treatment of saline wastewaters.

In order to improve biological treatment performance of saline wastewaters a halophilic bacteria, *Halobacter halobium* was used along with activated sludge culture in these study. A rather novel type reactor namely "**Aerated Percolator Column**" and "**RBC Unit**" were used in order to keep high cell concentrations in the system. Percolator column is somewhat similar to immersed biological filters with some exceptions that liquid is recycled by pressurized air which provides aeration along with crushed ceramic particles (particle size  $D_p=4$  mm) on which microorganisms were immobilized in form of fixed film. Four perforated tubes were placed in the packed column and air was provided to the system through those tubes.

The objective of these study was to investigate biological treatment performance of saline wastewater by using halophilic bacteria along with activated sludge culture in the aerated percolator column reactor and in an RBC unit. Type of microbial culture and salt content of wastewater were the two major parameters investigated.

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## CHAPTER TWO

# LITERATURE SURVEY

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The effects of sodium chloride on bacteria have been subject to investigation in the microbial field. It was found that the endogenous respiration rate of *Bacillus cereus* increased by sodium chloride concentrations up to 0.2 M (1% salt); but decreased above this concentration (Kincannon, Gaudy, 1966).

Winslow and Haywood found that the growth of *Escherichia coli* was stimulated at sodium chloride concentrations of 0.005 M (0.3%) to 0.25 M (1.5%). Adaptation of *E. coli* to sodium chloride was studied and it was found that cells exhibited the greatest degree of adaptability to salt in the early stationary phase of growth. Younger and older cultures harvested some time after reaching the stationary phase exhibited a lower degree of adaptability (Kincannon, Gaudy, 1966).

In the pollution control field, Lawton and Eggert have studied the effects of salt on trickling filter slimes. Lower reduction in BOD was observed when salt concentrations greater than 20 g/L were applied (Kincannon, Gaudy, 1966)

Stewart, Ludwig, and Kearns have studied the effect of salt on the extended aeration process and the applicability of this process for the treatment of shipboard wastes. Temporary reductions in treatment efficiency were noted when abnormally severe changes in salinity were combined with heavy hydraulic and organic loading. It was found that the sludge concentration in the aerator decreased when salt water (ocean water) was replaced by fresh water (Kincannon, Gaudy, 1966).

Ludzack and Noran have investigated effects of salt concentrations up to 20 g/L on the activated sludge process. It was found that increasing salt concentrations led to decrease in flocculation and BOD removal efficiency. Nitrification also was reduced severely.

Kincannon and Gaudy found that sludges developed in waters with low salt content can withstand shock loadings of high NaCl concentration somewhat better than sludges developed in high-salts medium can withstand a rapid decrease in salt concentration. The results indicated an approximately 30 percent decrease in substrate removal efficiency when a "fresh-water" sludge was dosed 30 g/L of NaCl. However, when an activated sludge acclimated to 30 g/L NaCl was dosed with fresh-water medium, substrate removal efficiency was reduced by approximately 75 percent. At the 45 g/L salt level severe impairment of system efficiency was encountered when sludge acclimated to 45 g/L salt was subjected to fresh-water medium (Kincannon, Gaudy, 1966)

An interesting result of the aforementioned study is the change which occurred in the settling characteristics of the sludge. In the present study, sludge developed at either 30 g/L or 45 g/L salt were dispersed systems partially devoid of any flocculating tendency.

It is also important to note that sludge developed at high salt concentrations (45 g/L) generally removed COD at a slower rate than the sludge developed in fresh water. There was a greater tendency for the high salt sludge to release cellular intermediates or products during glucose metabolism than was found for sludge developed in fresh water. Another significant response to rapid changes in salt concentration was the immediate release of cellular constituents as evidenced by an increase in soluble COD and a decrease in biological solids. (Kincannon, Gaudy, 1966)

In studies specifically designed to assess the degree of lysis in response to rapid changes in salt concentration, it was found that cells grown in 45 g/L salt underwent some degree of lysis when placed in lower salt concentrations below 10 g/L. When cells grown in various levels of salt concentration were placed in salt-free medium, the degree of lysis was approximately linear with salt concentration in the original growth medium. From the data obtained it appears that, cells grown in salt concentrations between 5.00 and 10 g/L don't undergo a considerable amount of lysis when placed in fresh water. For example, when old cells grown in 45 g/L salt were placed in 10 g/L sodium chloride, representing a decrease of 35 g/L, there was an approximately 12 percent decrease in optical density of nearly 30 percent lysis. Sludges grown in high salt concentrations were characterized by low

carbohydrate and protein contents and abnormally high lipid and RNA contents (Kincannon, Gaudy, 1966)

Nancy E.Kinner, Paul L.Bishop, and M. Asce have studied the effects of salt on rotating biological disc performance. They used domestic wastewater containing up to full strength seawater salinity. The effects of salinity, loading rate and nutrient media composition on organic removal rate were examined. It was shown that RBC's can treat high salinity wastewater to the same degree as non-saline wastewater. Mean COD removals of 61% and 64% were achieved using hydraulic loading rates of 0.04 and 0.08 m<sup>3</sup>/m<sup>2</sup>.d respectively. (Nancy, Bishop, Asce, 1982).

Biological treatment of a combined wastewater stream of several chemical factories were tested by Belkin, Brenner and Abeliovich. The untreated wastewaters, rich in halogenated organic (1250 mg/L DOC), were also highly saline (32 g/L TDS) and toxic (microtox EC=1.5-2%). Biphasic (anaerobic/aerobic) laboratory bench, scale reactor systems yielded reduction of dissolved organic carbon by 70 to 84% in the absence and presence of powdered activated carbon, respectively. Similar efficiencies were obtained in either activated sludge or aerated lagoon type reactors; but in the later case, longer hydraulic retention times were required. DOC removal was found to decrease with increased salt concentration however, a 50% efficiency was achieved even at 90 g/L TDS. (Belkin, Brenner, Abeliovich, 1993)

Kargı, F. and Dinçer, A. R. have studied effects of salt on COD removal rate and efficiency by using Fed-Batch operation. Fed-batch operation of biological wastewater treatment reactors has attracted significant interest in the research community because of definite advantages over other conventional systems especially for toxic substance removal from wastewater. Salt content has affected COD removal efficiency especially for concentrations higher than 3%. (Kargı.F., and Dinçer, A. R., 1995)

## 2.1. Effects of Salt On Conventional Biological Treatment Cultures

Wastes of high salt concentration arise from the manufacture of cheese and pickle processing. Waters containing high concentrations of salt wasted from softening plants may be found occasionally in streams containing organic pollutants. Shipboard wastes contain high concentrations of salt. Also, in many areas, natural surface waters are highly saline. As the water supply becomes more critical, such water may be used in increasing amounts for certain industrial needs. If, in turn, these waters become contaminated with organic materials, biological treatment which may be necessary could be affected by the presence of salt. Many factors contribute to the recent interest in the treatment of organic wastes in saline waters.

One of the first experiments on the effects of the salt on wastewater treatment organisms was conducted by Zobel et al. (1957). In this research, dilutions of the water collected from the Great Salt Lake in Utah were used to prepare agar plates which were inoculated with organisms from domestic sewage, soil and other sources. None of the sewage organisms and less than 1% of the soil bacteria survived in full strength lake water (28% salt). Higher survival rates were recorded in dilutions containing less salt; however, only 7-18% of the sewage bacteria grew on medium containing 2.8 to 7% salt. Analogous studies conducted with seawater (Zobell, 1946) showed similar results. Only 13% of the sewage organisms grew on nutrient medium made with full strength seawater (approximately 3.5%). Similar results were obtained with soil organisms. A slight stimulatory effect was observed when soil and sewage organisms were grown in medium containing 10% seawater (0.35% salt).

Despite early evidence that freshwater and terrestrial organisms are not equipped for life in environments containing significant amounts of salt, repeated attempts have been made to utilize these organisms for saline wastewater treatment.

Studies conducted with conventional cultures indicate that the following four common difficulties exist when treating saline and hypersaline wastes with organisms derived from freshwater and soil ecosystems.



\* *Limited extent of adaptation:* The general consensus among practitioners is that conventional cultures cannot be effectively used to treat wastes containing greater than 3 to 5% salt (Tokuz and Eckenfelder, 1978; Wong, 1992; Hockenbury et al., 1978). It is also clear that any salt acclimation achieved is quickly lost when the organisms are exposed to dilute ionic conditions. (Stower, Obayashi, 1991).

\* *Sensitivity to changes in ionic strength:* In general, shifts in salt concentration from 0.5 to 2% salt will cause significant disruptions in overall system performance. Rapid shifts in salt concentration typically cause more problems than gradual shifts. In certain cases organisms acclimate to give satisfactory reactor performance under saline conditions. However, even with acclimated cultures adequate process performance requires constant ionic composition and rapid reductions in waste salt content cause additional upsets. Equalization to stable salt content has been identified as prime consideration in the design of facilities to treat saline wastewater. (Stower, Obayashi, 1991).

\* *Reduced degradation kinetics:* Degradation rates are sensitive to increased salt concentration. For example, Mahmoud and DAVIS observed a reduction in specific degradation rates of aliphatic ketons from 0.3 mg TOC/mg MLVSS.h in freshwater to 0.12 mg TOC/mg MLVSS/h in 3.5% salt solution. A reduction in organic degradation rates means that reactors must be operated at lower F/M ratios when treating saline wastewaters. (Stower, Obayashi, 1991).

\* *High effluent suspended solids concentrations:* Researchers have speculated that salt induces cell lysis and reduces the populations of protozoa and filamentous organisms required for proper flocculation (Ludzack and Noran, 1966; Wooland, 1993). Regardless of the mechanisms, effluent suspended solids concentrations of over 100 mg/L are commonly reported in the literature because of low sedimentation efficiencies (Witmayer et al., 1984; Stower and Obayashi, 1991; Hockenburn et al, 1978; Wore, 1992).

## 2.2. Characteristics And Utilization of Halophilic Bacteria

Studies on freshwater and terrestrially derived conventional treatment cultures indicated that these organisms are not genetically capable of surviving in hypersaline environments. Thus, biological removal of organics from hypersaline wastes without dilution will require specialized microbes. Natural hypersaline environments such as the Great salt lake and solar salterns contain complex and diverse communities of such organisms. These halotolerant and halophilic microbes are metabolically diverse and possess many of the same characteristics as conventional waste treatment cultures. Although nonhalophilic and marine bacteria can be found in these environments, it is true that halophiles are of particular interest for hypersaline wastewater treatment.

Distinguishing characteristics of halophilic bacteria is that they require salt (specifically NaCl) for growth. Two groups of halophiles exist. Moderate halophiles grow best in medium containing 3-15 % (0.5-2.5 M) NaCl. These organisms are predominantly Eubacteria and species from several genus can be classified as moderately halophilic.

Deleya, Pseudomonas, Vibrio, Flavobacterium and Alteromonas were found to be abundant taxonomic groups in a number of solar salterns. Extreme halophiles exhibit optimum growth in environments containing 15-30% (2.5-5.2M) NaCl. These organisms are Archaeobacter which typically contain red or orange pigments. Certain species possess retinal proteins which act as light driven proton pumps. Halophiles tolerate the desiccating osmotic forces present in hypersaline environments by accumulating compatible solutes within the cytoplasm. Moderate halophiles accumulate a mixture of inorganic cations (K, Na) and organic compounds (aminoacids, betaine, glycerol) for osmotic regulation whereas extreme halophiles selectively accumulate KCl. Although not required for osmotic equilibrium, both moderate and extreme halophiles have a specific requirement for Na. Nutrient uptake, regulation of cytoplasmic PH and potential maintenance all require Na.

Halophile enzymes are able to function in hypersaline environments because of unique adaptations in primary protein structure. In general, halophilic proteins have an excess of acidic amino acids and require cations to shield closely spaced negative charges and enhance hydrophobic interactions (Woolard, Irvine, 1994).

### 2.3. Percolator (Submerged Biofilter) Systems

Kargi, F. and Uygur, A. have studied biological treatment of saline wastewater in an aerated percolator unit utilizing halophilic bacteria, in this study. Biological treatment of saline wastewaters in conventional systems usually result in low BOD removal performances because of plasmolysis of organisms. In order to overcome this problem a salt tolerant organism (*Halobacter Halobium*) was used along with activated sludge culture in an aerated percolator unit containing immobilized cells on ceramic particles. Percolator unit filled with ceramic particles was aerated externally for liquid recycle and also internally through perforated tubes located inside the column. The system was operated continuously with different microbial cultures at different salt concentrations and effect of microbial culture and salt concentration on the rate and extent of COD removal were investigated. Inclusion of salt tolerant bacteria in activated sludge culture resulted in high COD removal efficiencies especially at high salt concentrations such as 5%. (20)

Simon Ganzalez and Javier Duque have researched aerobic submerged biofilm reactors for wastewater treatment. In this study, treatment of wastewater from the campus of the National University of Mexico in a four-stage aerobic submerged biofilm reactor containing Pall-rings resulted in effluent with BOD values of less than 10 mg/L. The nitrification started in the first compartment and the reactor nearly achieved complete nitrification. The resulting biomass was filamentous and had good settling characteristics. Analysis of data indicated that the Stover-Kincannon and the variable order models can be used for the description and design of the process. (23)

Pia Christiansen, Line Hallosen and Paul Harremoes have studied liquid film diffusion on reaction rate in submerged filters. In this study, experiments were carried out in order to investigate the influence of liquid film diffusion on reaction rate in a submerged filter with denitrification and in order to compare with a theoretical study of the mass transfer coefficient. The experiments were carried out with varied flow, identified by the empty bed velocity of inflow and recirculation, respectively 1.3, 2.8, 5.6 and 10.9 m/h. The filter material consisted of 3 mm polystyrene spheres. The results indicate that the influence of liquid film diffusion on reaction rate can be ignored. (24)

Çeçen, F. have studied nitrification-denitrification kinetics in submerged filters. The criteria for nitrification and denitrification are presented on the basis of biofilm kinetics in two upflow submerged filters in series. Nitrification followed first-half, and zero-order kinetics. For the half-order range the half order rate constants were about  $k_{1/2an}=0.4-0.9 \text{ g NH}_4\text{-N}^{1/2} \text{ m}^{-1/2} \text{ d}^{-1}$ . In the kinetic region where ammonia removal rate was independent of the ammonia concentration the zero-order rate constants for the DO ranges of 2-3 mg/L and 4-5 mg/L were found to be  $0.47 \text{ g NH}_4\text{-N m}^{-2} \text{ d}^{-1}$  and  $1.8 \text{ g NH}_4\text{-N m}^{-2} \text{ d}^{-1}$ , respectively. In the zero-order region ammonia removal proceeded as a half-order rate constants were about  $k_{1/2ao}=1,4-2,7 \text{ g O}_2^{1/2} \text{ m}^{-1/2} \text{ d}^{-1}$ . These results showed that nitrification is oxygen limited for practical purposes. Another observation was that nitrite accumulation reached a considerable degree at bulk oxygen to bulk ammonia ratios lower than 5 since the formation of nitrate was inhibited similar to nitrification half-and zero-order kinetic regions were also observed in denitrification. The half-and zero-order rate constants for COD unlimited cases ( $\text{COD/NO}_x\text{-N}>5$ ) were about  $k_{1/2ad}=0.27 \text{ g NO}_x\text{-N}^{1/2} \text{ m}^{1/2} \text{ d}^{-1}$  and  $k_{oad}=1,9 \text{ g NO}_x\text{-N m}^{-2} \text{ d}^{-1}$ , respectively. The transition from half-to zero-order region occurred approximately at a nitrogen concentration about 60 mg/L  $\text{NO}_x\text{-N}$ . The nitrite produced in the nitrification stage could be successfully reduced in denitrification without causing any inhibitory effect. (25)

## 2.4. Rotating Biological Contactor (RBC) Systems

Nancy E. Kinner and Paul L. Bishop have studied treatment of saline domestic wastewater using RBCs. In this study the treatment of high salinity wastewaters from small Island Communities by use of rotating biological contactors was evaluated. Domestic wastewaters containing up to full strength seawater salinity were utilized. The effects of salinity, loading rate and disc media composition on organic removal rate were examined. It was shown that RBCs can treat high salinity wastewaters to the same degree as non-saline wastewaters. The resulting effluent organic content was adequate for discharge into either marine or freshwater receiving waters. Mean COD removals of 61% and 64% were achieved using hydraulic loading rates of 0.04 and 0.08 m<sup>3</sup>/m<sup>2</sup>d respectively. At higher loading rates, up to 0.33 m<sup>3</sup>/m<sup>2</sup>d organic removal efficiency was reduced. No difference in treatment efficiency was observed using either plastic or masonite as the disc media. (4)

Alpaslan, N. and Okutucu, S. have evaluated the performance of an RBC system. In this study, the model established for the experimental investigations consists of three main units: feeding tank, biodisc reactor and sedimentation tank. The synthetic wastewater which is prepared as influent is seeded by domestic wastewater in order to provide a culture of microorganisms on the surface of the discs. At the beginning, the system has started up as a batch reactor. Within the first week the microbial activity has been very slow and a trace amount of biofilm has occurred on the discs and then system was started up as a continuous reactor. As a result of the rotating biological contactor process a wide variety of factors may be effective on treatment efficiency. Among them, hydraulic loading, volume to area ratio, organic loading and the rotational speed of the discs are encountered and their effects to the treatment efficiency were observed. (22)

Boshou Pan and L. Hartmann have studied activity of biomass in RBC system treating pulp industrial wastewater. In this study, the activity of biomass in a longitudinal laboratory-scale rotating biological contactor system used for the treatment of evaporator condensate from a sulfite pulping process was determined by chemical parameters (TKN:SS and VSS:SS) and biological kinetic parameters carried out in Warburg experiments. The results show that kinetic parameters are more effective in describing the activity of the biomass, both on the discs and in the basin of RBC, than chemical parameters. The biomass on the discs at the transition phase of carbonaceous removal to beginning of nitrification has a high

activity, which is comparable to that of activated sludge with high F/M ratio of 0.9 kg BOD<sub>5</sub>/kg MLSS/day. The biomass suspended in the basin of the RBC has only about one-tenth of the metabolic activity of the biomass. (26)

F. Wilson has attempted prediction of rotating biological contactor efficiency using TOC. In this study, the results of experiments treating high-strength vegetable-pickling waste in an experimental rotating biological contactor are described. The relationships between TOC, BOD and COD for the particular waste are presented. A high correlation was found between TOC and BOD. The use of TOC led to a mathematical model describing the effluent concentration in terms of the output TOC concentration and the hydraulic loading rate. It was found that a relationship involving these two independent variables alone explained more than 98% of the variation in the dependent variable. The model predicts that the effluent TOC concentration will be more sensitive to changes in influent TOC concentration than to changes in hydraulic loading rate and that it can be used to predict the effect of recirculation on the effluent substrate concentration. (27)

Charles I. Noss and Roy D. Miller have studied rotating biodiscs treating recarbonate alkaline wastewater. Lime addition to raw wastewater as an upgrading technique can significantly aid a rotating biological contactor plant in producing effluents in compliance with federal discharge permit limitations for biochemical oxygen demand, ammonia nitrogen and phosphorus. Low-level lime addition does not require recarbonation and does not produce the sludges typical of high pH lime treatment schemes. The pilot RBC process provided BOD<sub>5</sub> removal when subjected to an influent pH of 9.5 for hydraulic loading rates of 2.0, 3.0 and 4.0 gpd/sq ft (0.08, 0.12 and 0.6 m<sup>3</sup>/m<sup>2</sup> day). In addition to the removal of phosphorus, lime pretreatment reduced the organic loading on the RBC produced carbon dioxide, thereby forming carbonate alkalinity necessary for nitrification after the initial BOD<sub>5</sub> had been removed. The resultant pH after recarbonation was also in the optimal range for nitrification. (28)

Ching-san Huang has studied nitrification kinetics and its RBC application. In this study, the surface reaction model for the mechanism of mass transfer-metabolism in the fixed-film nitrification process was investigated and discussed for the design of trickling filter systems and rotating biological contactor systems. Two experimental studies were performed. The

first study, using a stationary fixed-film reactor to simulate the trickling filter process, revealed that the surface reaction kinetics followed a pseudo-homogeneous model. The second study, using a bench-scale RBC unit, indicated that the pseudo-homogeneous model was also applicable to the rotating fixed-film process. The effective slime thickness of an RBC system can be estimated from this model by locating the optimum  $\text{NH}_3\text{-N}$  removal rotating speed and finding the corresponding liquid film thickness at that rotating speed. (29)

Roland Leduc and Ian Buchanan have studied minimization of multistage RBC active disc area. In this study, an analytical approach to the minimization of total active disc area ( $A_T$ ), required for organic carbon removal by a multistage rotating biological contactor, was presented. RBC operation was simulated using semiempirical models based on a saturation-type (Monod) removal rate, and on a first-order kinetics removal rate. The parameters used in the predictive equations were considered to be different from one RBC stage to the next. The minimizations of  $A_T$ , based in turn on each of the two semiempirical models, lead to a relationship between the optimal soluble substrate concentration of any stage and those of the immediately preceding and succeeding stages. When used in design, these relationships allow the minimum  $A_T$ , as well as the optimal number of stages required for organic carbon removal. For the saturation model, two parametric ratios ( $\phi_p$  and  $\phi_{K_s}$ ) were introduced, which define the change in the model parameters with stage. Application of the relationships to the determination of the optimal number of stages and the minimum  $A_T$  is included. (30)

Edward J. Opatken has studied RBC nitrification design using zero-order kinetics. The zero-order kinetic design procedure was used to illustrate an alternative approach for specifying the number of RBC shafts required to achieve a final effluent  $\text{NH}_3\text{-N}$  concentration. The arrangement of RBC shafts into stages independent of parallel or series configuration when kinetics are zero-order. The effect of temperature on the  $\text{NH}_3\text{-N}$  reaction rate constant has a significant effect on the number of RBC shafts required to achieve a final effluent  $\text{NH}_3\text{-N}$  level. (31)

F. Wilson has studied kinetics and reaction order in rotating biological contactors using TOC. In this study, the result of experiments into the kinetics of the treatment of a high

strength vegetable pickling waste in an experimental RBC were described. A high correlation was found between TOC and BOD which justified the use of TOC in the investigation. The substrate loading rate removal was compared with predictions from the Stover-Kincannon model. The Stover-Kincannon relationship was found to satisfactorily predict the TOC loading removal rate and to be particularly useful in predicting the load removal rate at high applied loading rates. A linear regression analysis was used to determine the relationship between the output TOC in terms of the input TOC and the hydraulic loading rate. This enabled the prediction of an apparent half-order of reaction to be confirmed. Special features of this work are the use of TOC as the major analytical parameter and the very high input concentrations. (35)

Yeun C. Wu and Ed D. Smith have studied rotating biological contactor system design. In this study, practical application of Wu's model for the prediction of soluble BOD removal in RBC systems was discussed extensively for various operating conditions. The model was tested by using more than 80 data sets obtained from the operation of six full-scale RBC plants. Also, nomograms were constructed to demonstrate a method for selecting the design surface hydraulic loadings. More importantly, the hydraulic loading rates predicted/calculated on the basis of the criteria employed for four full-scale RBC plant designs were compared with the actual design hydraulic loadings. No significant difference between the predicted and the designed hydraulic loading was found. (36)

Chi-Yuan Lee has developed a model for biological reactors having suspended and attached growths. In this study, a hybrid reactor is characterized as having both suspended-growth and attached-growth bacteria. One important phenomenon of such a reactor is that a competition for substrates exists between these two growth-type bacteria, which can not be accurately predicted by single-growth models. This paper has presented a model that considers two growths competing for a single substrate in such a completely mixed reactor at steady-state condition. The critical advantage of this model stems from the fact that it maintains all essential concepts for single-growth kinetics but uses only two fundamental parameters of empty-bed hydraulic retention time and suspended biomass solid retention time to predict the competing results. These predictions are very useful for analyzing the design and performance of a variety of hybrid reactors that mix completely. (37)



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## CHAPTER THREE

# OBJECTIVES

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Major objective of this study is to improve the treatment efficiency of saline wastewaters by using the halophilic organism *Halobacterium halobium* along with activated sludge culture and to investigate the system's behavior. Synthetic wastewaters with different salt concentrations were treated in a continuous aerated submerged filters (**PERCOLATOR**) and rotating biodisc unit (**RBC**) by using a mixed culture of organisms containing *H. halobium*. Kinetic analysis of this system was accomplished. Variation of kinetic behavior with salt concentration was investigated. A mathematical model describing the system behavior was developed and the model parameters were determined by using the experimental data.

Major objectives of the presented thesis can be summarized as follows:

- a. To study effectiveness of various cultures in biological treatment of saline wastewater by using an aerated percolator unit.
- b. To study effects of various process parameters (COD loading rate, A/Q ratio, salt concentration, liquid phase aeration.) on biological treatment performance of saline wastewater in an RBC unit utilizing activated sludge and *Halobacter halobium* culture.
- c. To develop a mathematical model describing the system's behavior and to determine kinetic constants by using experimental data.

These studies provided a new technological approach for biological treatment of saline wastewater and also contributed to the understanding of behavior of such systems.

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## CHAPTER FOUR

# MATERIALS AND METHODS

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### 4.1. Experimental Set-up

A Schematic diagram of the percolator unit is depicted in Figure 4.1.1. Percolator reactor consisted of a plexiglass column of diameter  $D=16$  cm and height  $H=60$  cm. The reactor has a conical section of height  $H=6$  cm at the bottom and a external recirculation arm as shown in Figure 4.1.1. A perforated plexiglass plate was placed at the bottom of the column. The reactor was filled with crushed ceramic particles of diameter  $D_p=4$  mm. The height of the packed section was  $H_p=20$  cm. The reactor was filled with wastewater and the liquid height above the packing material was 10 cm resulting in an overall liquid height of  $HL=36$  cm including the conical section. Air was introduced to the external recirculation arm in order to circulate and aerate the liquid. Four perforated tubes were placed in the packed bed and air was introduced to the organisms on packing material through those perforated tubes. Additional aeration was provided to the liquid phase above packing material by using diffusers. With vigorous aeration at the different locations in the reactor D.O limitations were overcome.

The RBC unit used for the experimental investigations consists of two main units: feeding tank and biodisc reactor (See Figure 4.1.2.) The feed wastewater to the biodisc reactor was provided by a pump installed between feeding tank and the reactor. There are two subsequent stages in the biodisc reactor, each with twenty discs. The discs are made of polypropylene and each has a diameter of 0.20 m. The thickness of the disc and the spaces in between them are 2 mm and 4 mm respectively. Rubber supports are located between the discs in order to maintain stability during the rotation. The rotational speed of the motor unit is  $n=5$  rpm. Rotating biological contactor reactor (RBC) consisted of a stainless steel unit of diameter  $D=60$  cm and height  $H=20$  cm and width  $L=50$  cm. Total disc area was

2.512 m<sup>2</sup> and submerged disc area was 1.51 m<sup>2</sup>. Volume of the liquid in reactor was 9.5 L. Aeration was provided by perforated tubes placed in the RBC tank.

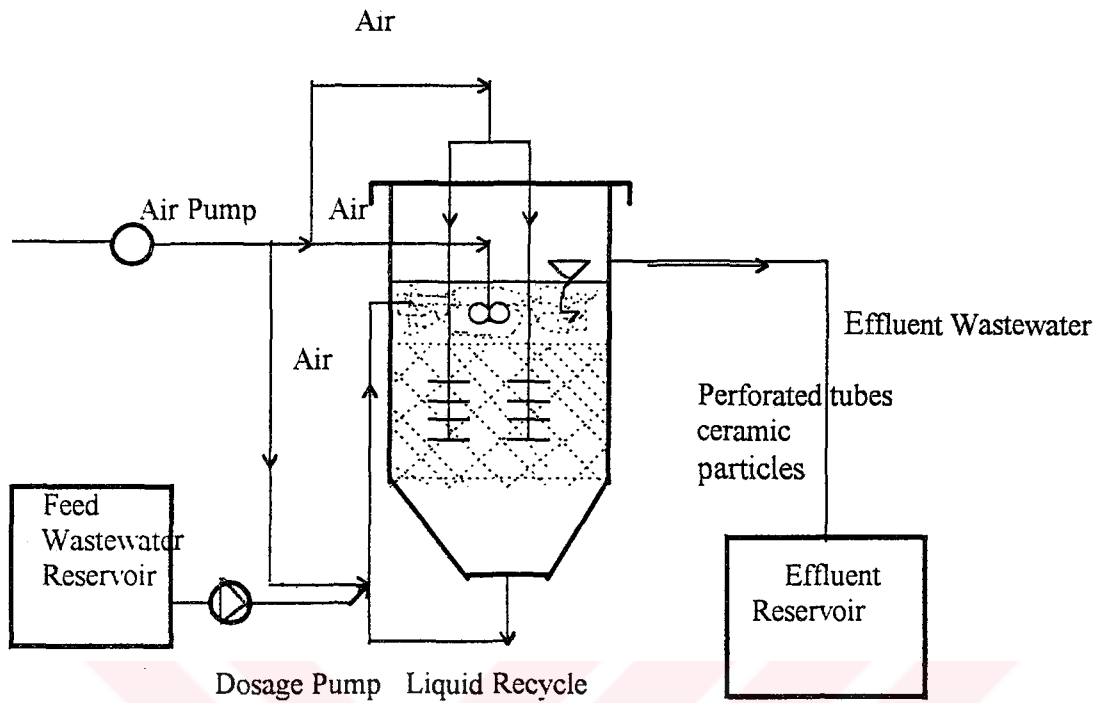


Figure 4.1.1. Schematic of Aerated Percolator Unit

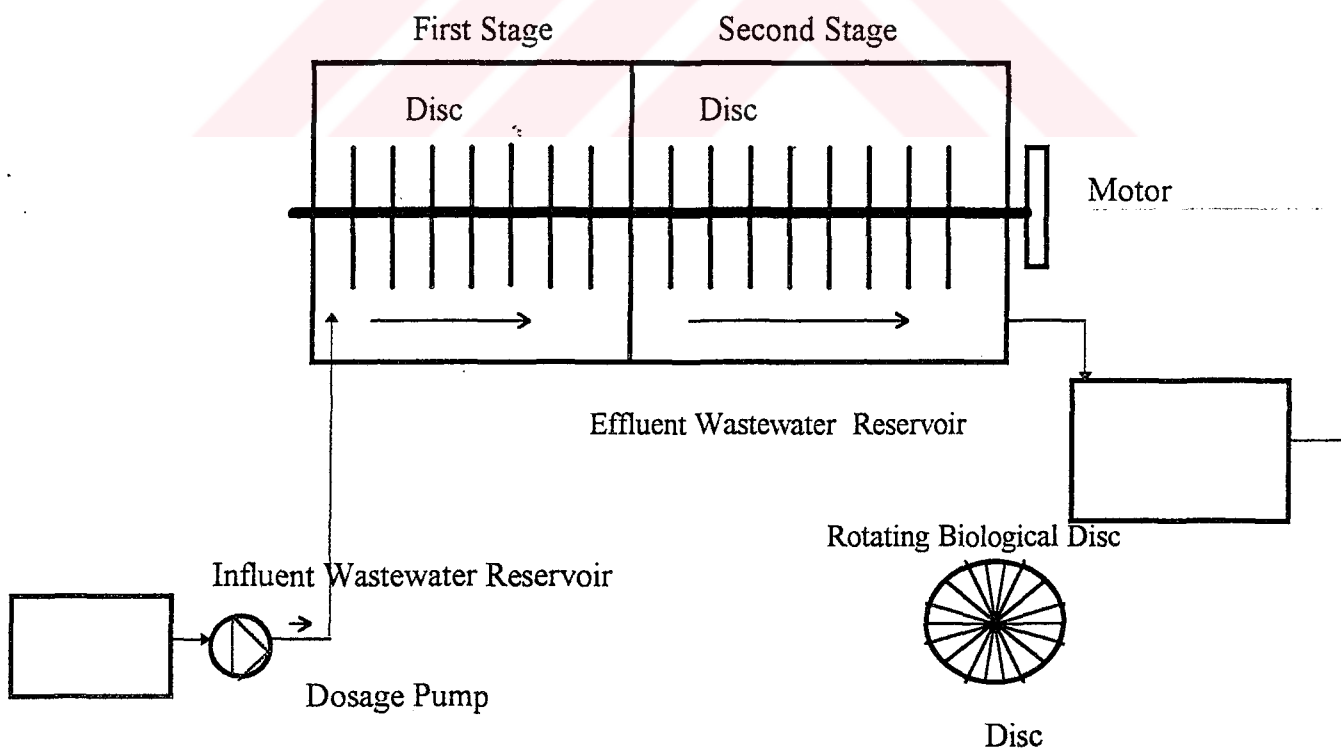


Figure 4.1.2. Schematic of Rotating Biological Contactor Unit

## 4.2. Operation

Before starting continuous experiments, percolator was filled with the synthetic wastewater, inoculated with a dense culture of microorganisms and was operated batchwise with recirculation and aeration for about a week. Wastewater media was replenished two times during batch operation. Continuous operation was started after a dense microbial flora was developed in the reactor at the end of batch operation. Feed wastewater was introduced to the recirculation arm and effluent was removed from the top of the reactor by gravity.

Synthetic wastewater was used to obtain constant wastewater quality. Synthetic wastewater was consisted of diluted molasses, urea,  $\text{KH}_2\text{PO}_4$  and  $\text{MgSO}_4$  to result in COD/N/P:100/10/1 ratio. Composition of influent wastewater was adjusted approximately COD=4500-5500 mg/L, TN=450-550 mg/L, P=45-55 mg/L and  $\text{MgSO}_4=0.05$  g/L. with this composition of wastewater the system was operated with COD limitations.

Biodiscs were placed on the shaft with 40% immersion in water. The system contained 40 discs with diameters of 20 cm. Total disc area was  $A_T=2.512$  m<sup>2</sup> and wet submerged of disc area was  $A_w=1.51$  m<sup>2</sup>. Volume of the tank was 9.5 L. Disc area per unit volume was  $a_w=264$  m<sup>2</sup>/m<sup>3</sup>. Discs were rotated with a constant speed of  $n=5$  rpm by a driving motor.

Aeration was provided by using an air pump and a perforated tube submerged in water. The system was operated with and without aeration in liquid phase and system performances were compared. Wastewater was fed to the system by a dosage pump with constant flow rates to result in  $\theta_H=2-10$  h. Effluent water was discharged to sewerage. Temperature of the system was kept constant between  $T=26.5-27.5$  °C by using a temperature control system. PH of water in the tank was controled approximately at PH=7-8. PH of influent water was approximately PH=6.5. During operation, pH of water in the tank increased to PH =8.5. PH control was accomplished by adding dilute  $\text{H}_2\text{SO}_4$  to the system twice a day.

Concentration of dissolved oxygen was measured everyday by using dissolved oxygen analyser and a DO probe to result in  $\text{DO}>3$  mg/L in liquid phase. Inoculation culture was a 50% mixture of activated sludge with *Halobacter halobium*, the system was operated

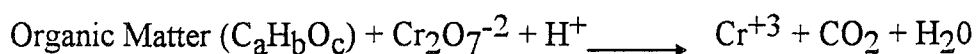
batchwise for about ten days to obtain a constant biofilm thickness on the discs. At the end of batch operation, biofilm thickness on the discs reached approximately, 1.5-2 mm. In continuous experiments, wastewater was fed to the system with a constant flow rate. Samples of liquid phase was taken twice a day and centrifuged. COD analysis were made on clear supernatant. Total suspended solids (TSS) concentrations in liquid phase were determined under steady-state conditions. Effects of the following parameters on system performance were investigated.

- a.) A/Q ratio (or hydraulic residence time,  $\theta_H$ )
- b.) COD Loading rate
- c.) Salt concentration
- d.) Feed COD concentration
- e.) Aeration in the liquid phase

### 4.3. Analytical Methods

#### 4.3.1. COD measurements

The COD test is used to measure the content of organic matter in both wastewater and natural water. The oxygen equivalent of the organic matter that can be oxidized is measured by using a strong chemical oxidizing agent in an acidic medium. Potassium dichromate is used as an oxidizing agent. The test must be performed at an elevated temperature. A catalyst (silver sulfate) is required to aid the oxidation of certain classes of organic compounds. Since some inorganic compounds interfere with the test, care must be taken to eliminate them. The principal reaction using dichromate as the oxidizing agent may be represented as follows:



The COD test is also used to measure the organic matter in industrial and municipal wastes that contain compounds which are toxic to biological life.

## Sampling and Storage:

Samples were with drawn from the reactor every day and were centrifuged at 6000 rpm for 1/2 hour. Clear supernatant was acidified and stored in a refrigerator. COD analysis were carried out on clear supernatant according to Standart Methods.

## Reagents:

a-) Standart Potassium Dichromate Solution (0.250 M): 12.259 g.  $K_2CrO_7$  primary standart grade; previously dried at 103 °C for 2 h is dissolved in distilled water and diluted to 1,000 ml.

b-) Silver Sulfate:  $Ag_2SO_4$  reagent or technical grade cristals or powder.

c-) Sulfuric Acid Reagent:  $Ag_2SO_4$  is added to concentrated  $H_2SO_4$  at the concentration of 22 g.  $Ag_2SO_4$ /4 kg bottle and is let to stand for 1 to 2 days to dissolve  $Ag_2SO_4$

d-) Sulfuric Acid: Concentrated  $H_2SO_4$

e-) Ferroin Indicator Solution: 1.485 g. 1.10 phenanthroline monohydrate and 695 mg  $FeSO_4 \cdot 7H_2O$  are dissolved in distilled water and diluted to 100 ml. This indicator solution may be purcashed and used as is.

f-) Standart Ferrous Ammonium Sulfate Titrant (0.25 N): 98 g.  $Fe(NH_4)_2(SO_4)_2 \cdot 6H_2O$  (FAS) is dissolved in distilled water. 20 ml concentrated  $H_2SO_4$  is added, cooled, and diluted to 1,000 ml. This solution is standardized daily against standart  $K_2Cr_2O_7$  solution, as follows:

10 ml standart  $K_2Cr_2O_7$  solution is diluted to about 100 ml. 30 ml concentrated  $H_2SO_4$  is added and cooled. This solution is titrated with FAS titrant by using 0.10 to 0.15 ml (2 to 3 drops) ferroin indicator.

Normality of FAS solution

$$\text{Volume } 0.25 \text{ N } \text{K}_2\text{CrO}_7 = \frac{\text{solution titrated, (ml)} * 0.25}{\text{volume FAS used in titration, (ml)}}$$

g-) Mercury Sulfate:  $\text{HgSO}_4$ , crystals or powder.

h-) Sulfonic Acid: Required only if the interference of nitrites is to be eliminated.

Potassium hydrogen phthalate standart: Potassium phthalate ( $\text{HOOC}_6\text{H}_4\text{COOK}$ ) is crushed and dried to constant weight at  $120^\circ\text{C}$ . 425 mg of K phthalate is dissolved in distilled water, and diluted to 1,000 ml potassium hydrogen phthalate has a theoretical COD of  $500 \text{ mg O}_2 / \text{L}$ . This solution should be prepared and used freshly each time.

### Procedure:

Depending on the strength of wastewater 1-20 ml of sample is taken and diluted to 20 ml with distilled water 5 ml sulfuric acid reagent is added to the diluted sample and then 0.4 g  $\text{H}_2\text{SO}_4$  is added while mixing to dissolve  $\text{HgSO}_4$ . Solution is cooled while mixing to avoid possible loss of volatile materials. 10 ml  $\text{K}_2\text{Cr}_2\text{O}_7$  solution is added and mixed. Flasks are attached to condenser and cooling water is turned on. Remaining sulfuric acid reagent (25 ml) is added through open end of condenser. Swirling and mixing is continued while adding sulfuric acid reagent. Mixture is refluxed for 2h. Reflux condenser is disconnected and the mixture is diluted to 140 ml volume with distilled water. Solution is cooled to room temperature and excess  $\text{K}_2\text{Cr}_2\text{O}_7$  is titrated with FAS, by using 0.10 ml to 0.15 ml (2 to 3 drops) ferroin indicator. The end point of the titration is taken as the first sharp color change from blue-green to reddish brown. The blue-green may reappear.

*In order to overcome adverse effects of salt in COD measurements, sufficient amounts of  $\text{HgSO}_4$  were added onto digested samples to precipitate chloride ions, before titration*

**Calculation:**

$$\text{mg COD / L} = \frac{(A - B) * N * 8000}{\text{ml sample}}$$

Where, A is volume FAS used for blank (ml), B is volume FAS used for sample (ml) and N is normality of FAS.

**4.3.2. Dissolved Oxygen Measurements**

The Hach Model 16046 portable Dissolved Oxygen Meter was used to measure dissolved oxygen in wastewater in the reactors. The instrument uses a Clark-type Polarographic electrode as the oxygen sensor and can measure dissolved oxygen (DO) in the ranges of 0-10 and 0-20 mg/L with the standard membrane. A rechargeable battery pack provides operating power. Automatic temperature and pressure compensation capabilities are incorporated. The instrument is fitted in a molded ABS carrying case with a hinged lid that is detachable for convenience in the laboratory.

**4.3.3. Suspended Solid Measurements**

Biomass in suspension was determined by filtering the sample through milipore filters (0.45  $\mu\text{m}$ ) and drying until constant weight in an oven at 105  $^{\circ}\text{C}$ . Immobilized biomass concentration on support particles were determined by washing cells from a disc into tap water and weighting the biomass after filtration and drying of the filter paper.

Calculations are made by using the following equation:

$$M = \frac{(A - B) * 1000}{V}$$

where, A is weight of filter + residue, (mg); B is weight of filter, (mg) and V is ml sample.



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## CHAPTER FIVE

# RESULTS AND DISCUSSION

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### 5.1. Experiments With Aerated Percolator System

Effects of two major parameters namely types of microbial flora and salt concentration on the rate and extent of COD removal were investigated. Total liquid volume in the reactor was  $V_L=6.5$  L with a feed flow rate of  $Q=1.5$  L/h resulting in a hydraulic residence time of  $\theta_H=4.3$  h. throughout experiments.

#### 5.1.1. Pre Steady-State Behaviour of The System

Continuous experiments were conducted until the system reached steady-state which was realized when three consecutive effluent COD measurements were approximately the same. Figure 5.1.1. depicts variation of the feed and effluent COD concentrations with time before steady-state for 5% salt concentration. It took about 72 h for the system to reach constant effluent COD level of  $S_e=1520$  mg/L. Feed COD level was about  $S_o=5500$  mg/L throughout the experiment. In other experiments, it took about 70-100 h for the system to reach quasi-steady-state. Effluent pH values were about  $Ph_e=7.5-8$  while feed pH values were  $pH_o=6$  throughout experiments.

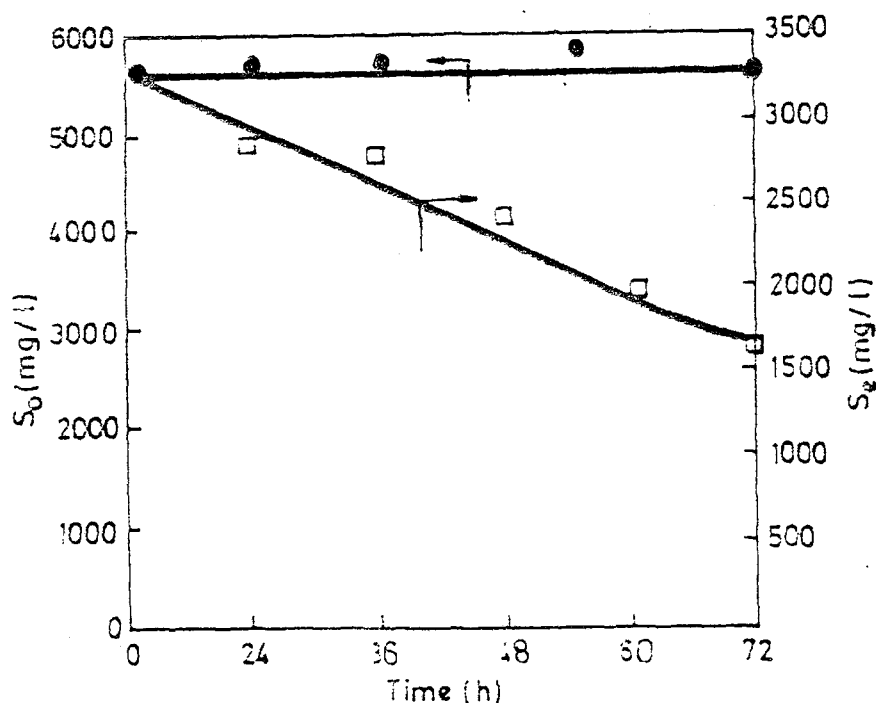


Figure 5.1.1. Presteady-state variation of feed and effluent COD Concentration

### 5.1.2. Performance of Different Types of Microbial Culture

In order to determine if inclusion of *H.halobium* in activated sludge culture has any advantageous effects in terms of COD removal rate and efficiency, experiments were conducted with different types of microbial flora at 1% salt concentration. Microbial flora used were a. Zoogleo Ramigera, b. Halobacter halobium, c. Zoogleo Ramigera and Halobacter halobium, d. Activated Sludge, e. Activated Sludge and Halobacter halobium.

Results of experiments at steady state are depicted in Figure 5.1.2. in form bar diagrams. Both COD removal efficiencies and rates were compared for different microbial flora. *H.halobium* alone resulted in the lowest efficiency since optimal salt concentration for Halobacter was nearly 15%. Performances of *Z.ramigera* and *A.sludge* culture were comparable. The best performance was obtained with a mixed culture of *A.sludge* and *H.halobium* resulting in nearly 90% COD removal efficiency and 1100 mg COD/L.h, COD removal rate. There may be some synergistic interaction among bacteria in activated sludge culture and *H.halobium* resulting in high COD removal efficiencies.

Considering the results of this experiment future experiments were conducted with a mixture of A.sludge and H.halobium in order to investigate the effects of salt concentration on performance of the system.

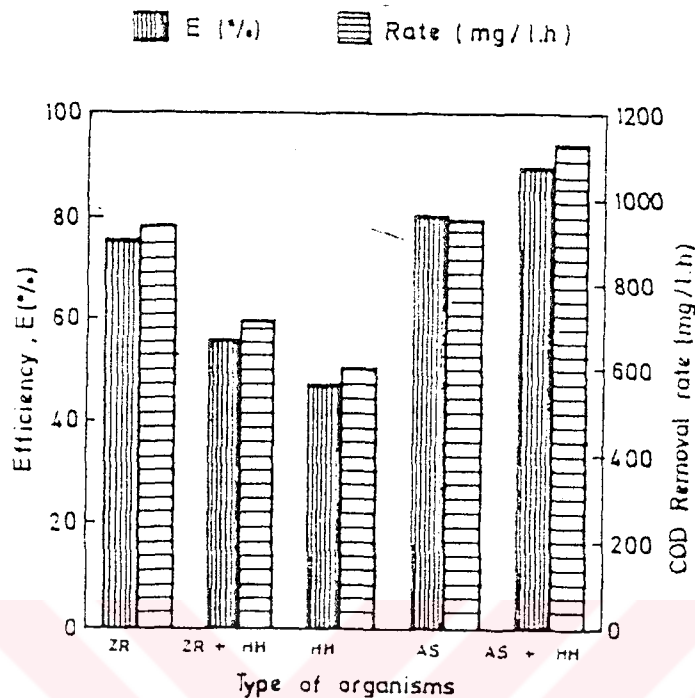


Figure 5.1.2. Comparison of performance of various microbial culture in the Percolator Unit (1% salt)

### 5.1.3. Effects of Salt Concentration

Percolator experiments were conducted with a mixed culture of activated sludge and H.halobium at different salt concentrations. Variation of the feed and steady-state effluent COD concentrations with salt concentration is depicted in Figure 5.1.3. Effluent COD concentration increased with salt concentration resulting in lower COD removal performances at high salt concentrations.

Variation of COD removal efficiency ( $E=1-S/S_0$ ) with salt content of the wastewater is depicted in Figure 5.1.4. COD removal efficiency was nearly 90% with 1% salt and dropped to 80% at 5% salt concentration.

The rate of COD removal also severely dropped with increasing salt concentration as shown in Figure 5.1.5. The rates of COD removal at 1% and 2% salt concentrations were

not significantly different. However, for salt concentrations above 3%, the rate dropped severely resulting in a value of  $r=620$  mg COD/L.h at 5% salt concentration.

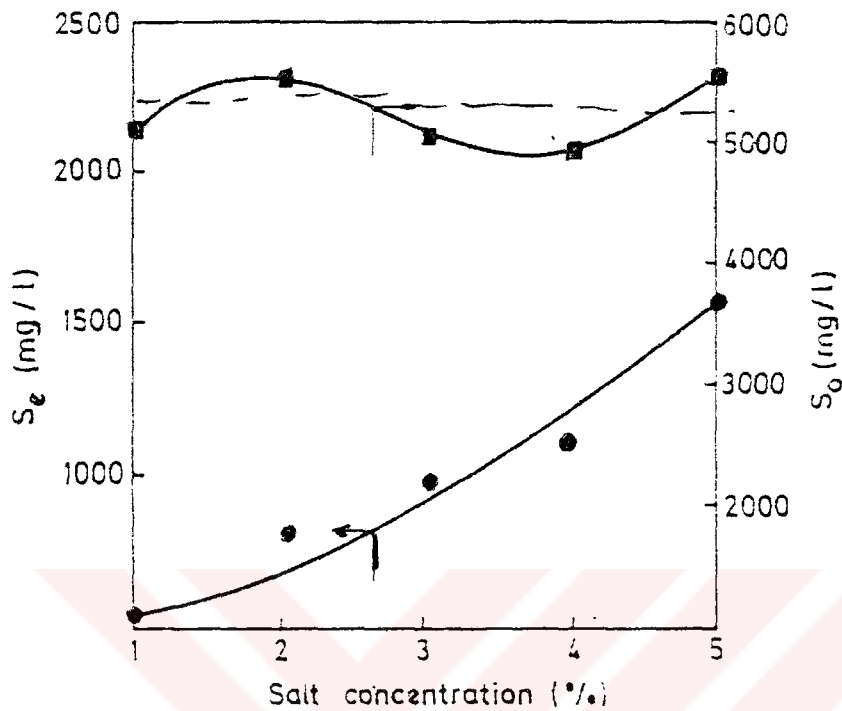


Figure 5.1.3. Variation of feed and effluent COD levels with salt concentration in the percolator unit (A.sludge and H.halobium)

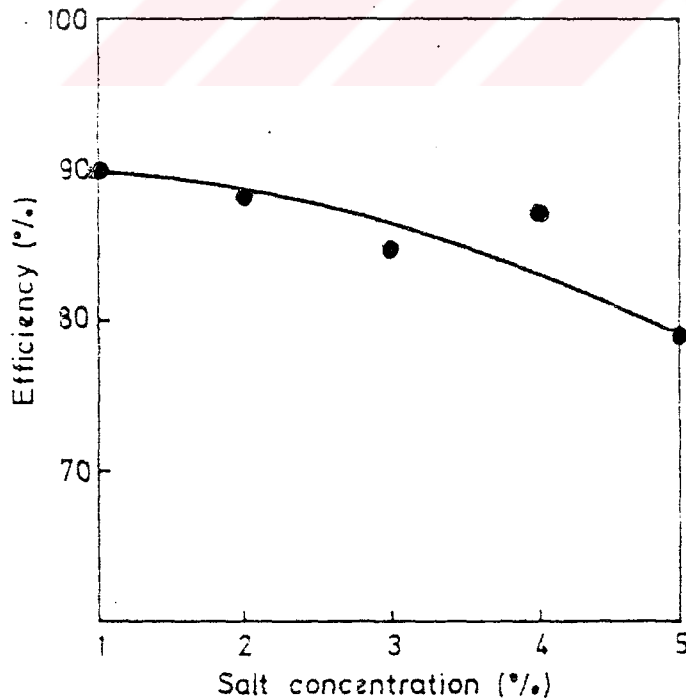


Figure 5.1.4. Effect of salt concentration on COD removal efficiency in the percolator unit.(A.sludge and H.halobium)

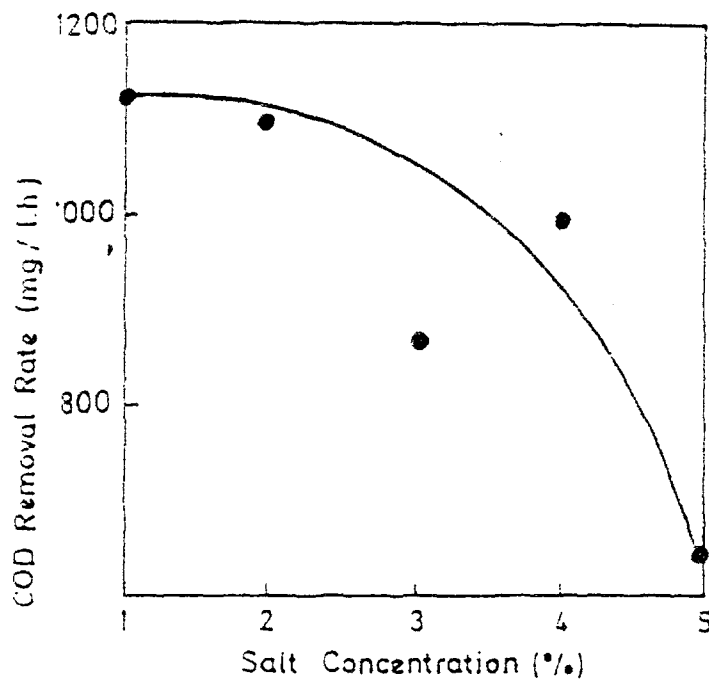


Figure 5.1.5. Effect of salt concentration on COD removal rate in the percolator unit.  
(A.sludge and H.halobium)

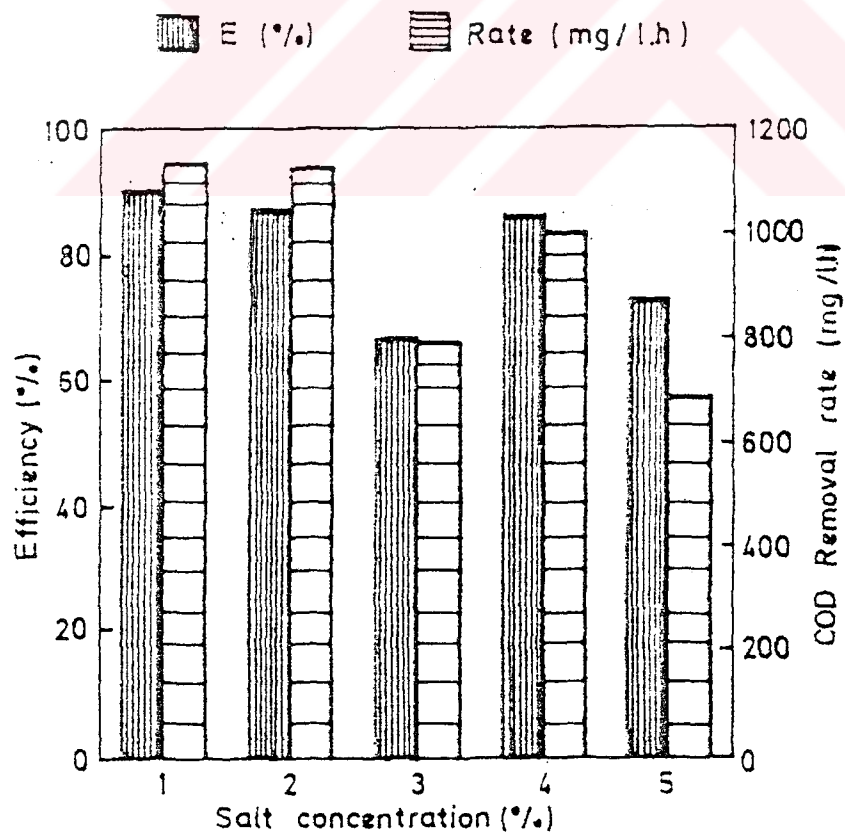


Figure 5.1.6. Effect of salt concentration on COD removal rate and efficiency in the percolator unit.(A.sludge and H.halobium)

Combined results of experiments conducted at different salt concentrations is depicted in Figure 5.1.6. in form of bar diagrams. COD removal efficiency is high at 1-2% salt concentrations because of the activity of activated sludge culture organisms. Halobacter is not reported to be very active at low salt concentrations. At 3% salt concentration neither activated sludge culture nor Halobacter are very active. 3% salt is probably inhibitory for A.sludge culture and is not high enough to be stimulatory for Halobacter. At higher salt concentrations such as 4-5%, probably because of high activity of Halobacter, COD removal efficiency increased. In other words, at low salt concentrations, A.sludge culture, and at high salt concentrations Halobacter actively removed COD; whereas, at medium salt concentrations neither culture were active enough to result in high COD removal efficiencies.



## 5.2. Experiments With Rotating Biological Contactor System

### 5.2.1. Effect of A/Q Ratio

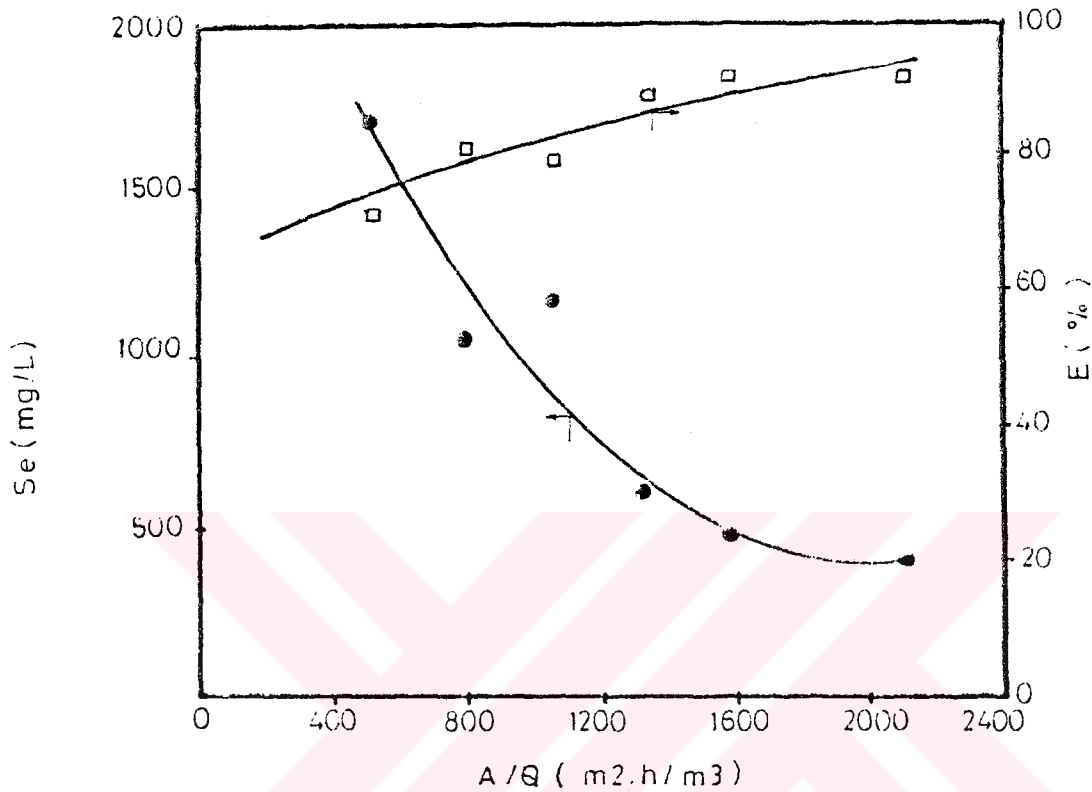


Figure 5.2.1. Variation of Effluent COD Concentration and COD Removal Efficiency with  $A/Q$  ratio in the RBC Unit

(Activated Sludge and Halobacter Halobium,  $\theta_H = 2, 3, 4, 5, 6$  and  $8$  h, 1% Salt Concentration, Aerated Liquid Phase,  $COD_o = 5000$  mg/L)

Activated Sludge and Halobacter halobium was used as microbial culture, in experiments conducted with the RBC unit.

Variation of effluent COD concentration with  $A/Q$  for 1% salt concentration is presented in Figure 5.2.1. with aerated liquid phase. COD concentration decreased and COD removal efficiency increased with increasing  $A/Q$  ratio, as a result of high biomass intensity on discs, at high  $A/Q$  ratios.

### 5.2.2. Effect of COD Loading Rate

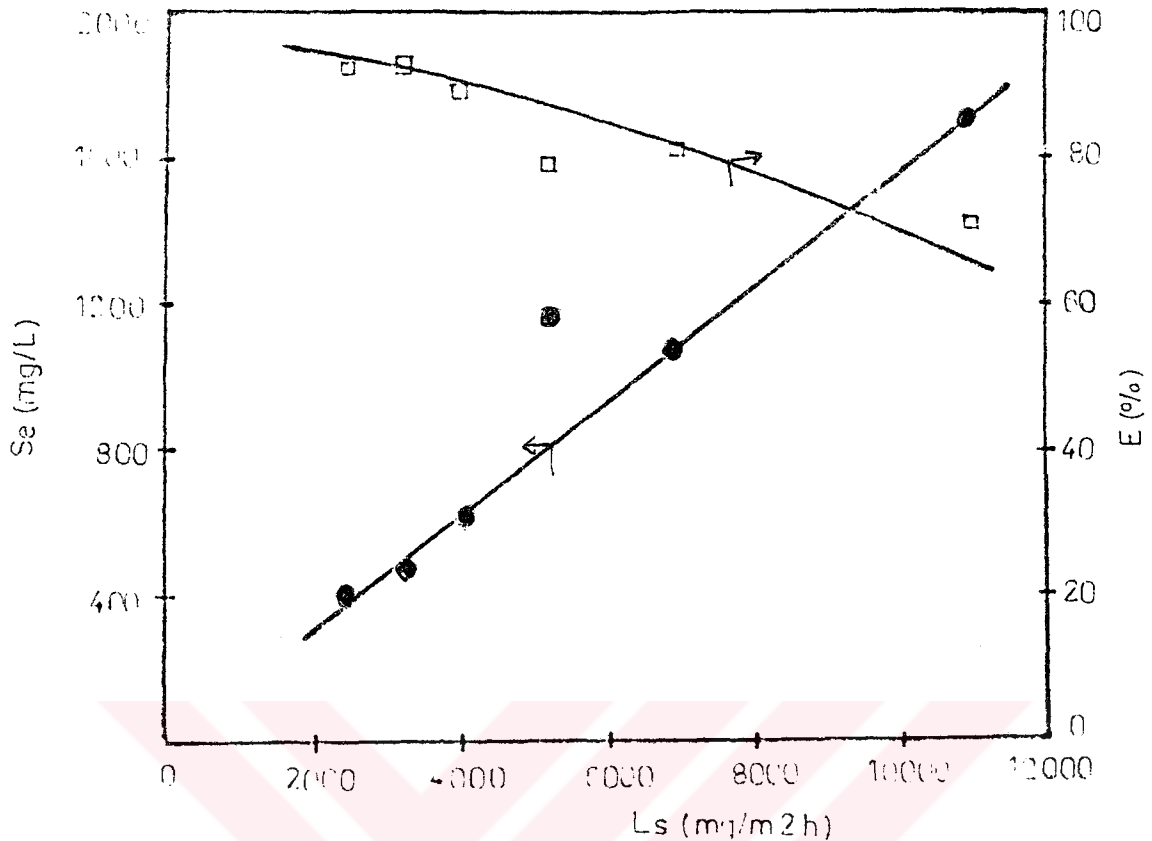


Figure 5.2.2. Variation of Effluent COD Concentration and COD Removal Rate with COD Loading Rate in the RBC Unit

(Activated Sludge and Halobacter Halobium.  $\theta_{Hf}$  = 2, 3, 4, 5, 6, and 8 h, 1% Salt Concentration, aerated liquid phase,  $COD_0$  = 5000 mg/L)

Variation of Effluent COD concentration with COD loading rate for 1% salt concentration is depicted in Figure 5.2.2. with aerated liquid phase. Effluent COD concentration increased; however, COD removal efficiency decreased significantly with COD loading rate ( $L_s$ )



### 5.2.3. Effect of Salt Concentration

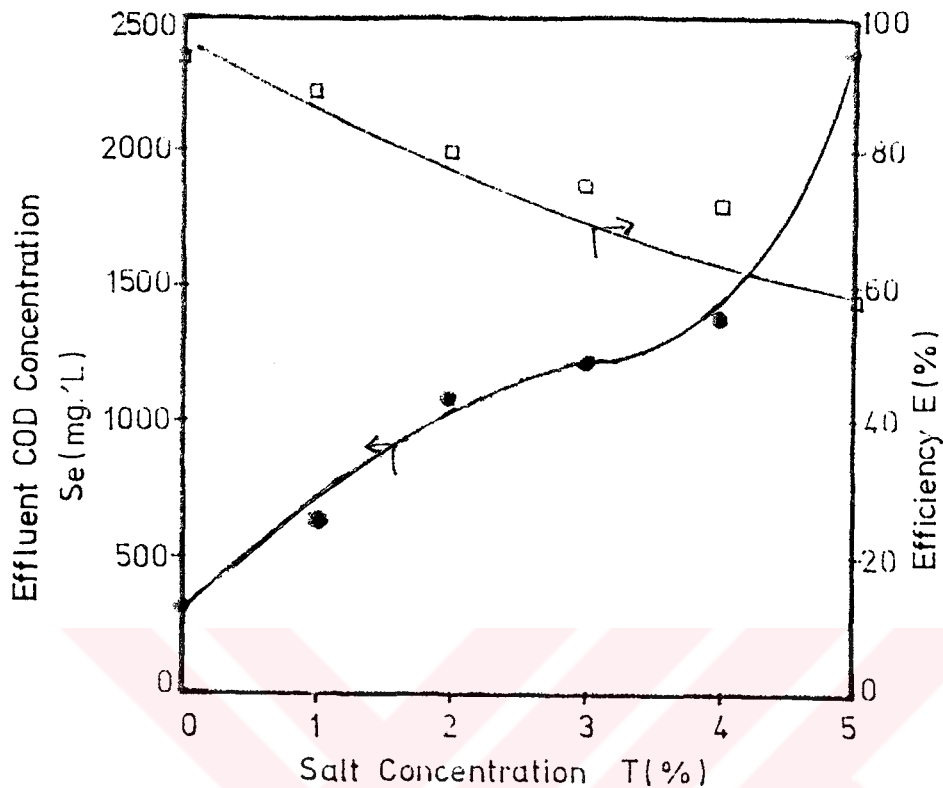


Figure 5.2.3. Variation of Effluent COD Concentration and COD Removal Efficiency with Salt Concentration in the RBC Unit

(Activated Sludge and Halobacter Halobium,  $\theta_H = 4$  h, 1%, 2%, 3%, 4% and 5% Salt Concentration, aerated liquid phase)

Variation of Effluent COD concentration with salt concentration for 4 h hydraulic residence time and different salt concentration (0%, 1%, 2%, 3%, 4% and 5%) is depicted in Figure 5.2.3. with aerated liquid phase. Effluent COD concentration increased and COD removal efficiency decreased with increasing salt concentration, as a result of adverse effects of salt on microbial culture.

### 5.2.4. Effect of Feed COD Concentration

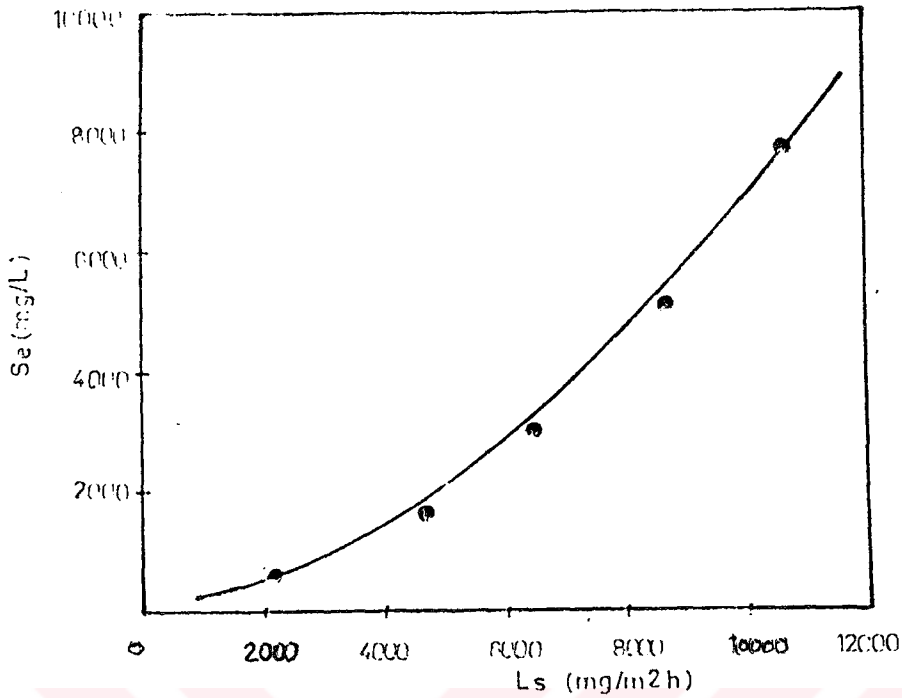


Figure 5.2.4.1. Variation of Effluent COD Concentration and COD Removal Efficiency with COD Loading Rate in the RBC Unit

(Activated Sludge and Halobacter Halobium,  $\theta_H = 4$  h, 1% Salt Concentration, Un-aerated liquid phase,  $COD_o = 2500, 5000, 7500, 10000$  and  $12500$  mg/L)

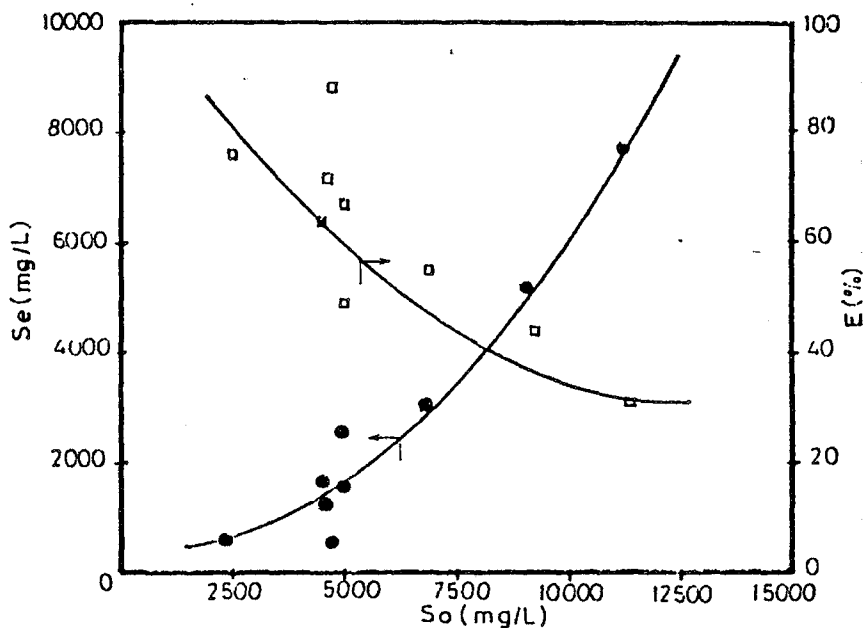


Figure 5.2.4.2. Variation of Effluent COD concentration and COD removal efficiency with Influent COD concentration

This set of experiments were performed with unaerated liquid phase and effects of feed COD concentration on system performance were investigated. Variation of Effluent COD concentration with COD loading rate for 1% salt concentration and  $\theta_H=4$  h, is depicted in Figure 5.2.4.1. Feed COD concentrations varied as 2500 mg/L, 5000 mg/L, 7500 mg/L, 10000 mg/L and 12500 mg/L (T=27 °C). Effluent COD concentration increased and COD removal efficiency decreased with increasing COD loading rate.

Variation of effluent COD concentration and COD removal efficiency with influent COD concentration is presented in Figure 5.2.4.2. for unaerated liquid phase. Effluent COD concentration increased and COD removal efficiency decreased with increasing influent COD concentration.



### 5.2.5. Effect of Liquid Phase Aeration

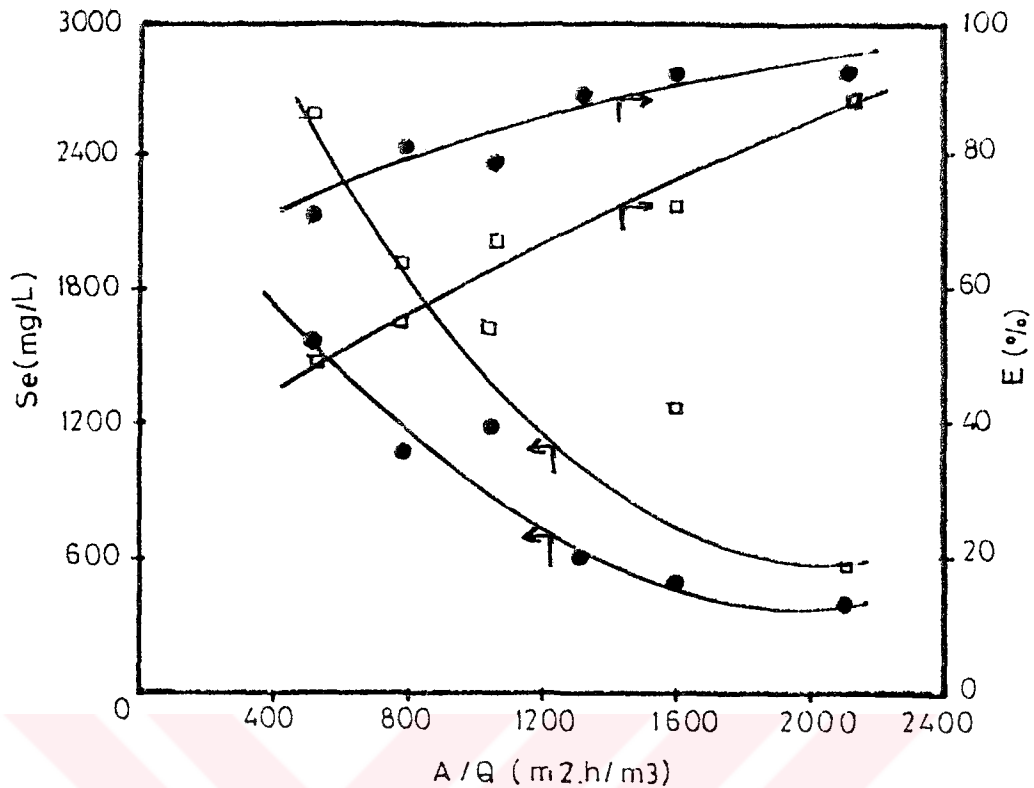


Figure 5.2.5. Variation of COD Concentration and COD Removal Efficiency with A/Q ratio in the RBC Unit  
(Activated Sludge and Halobacter Halobium,  $\theta_H = 2, 3, 4, 5, 6$  and  $8$  h, 1% Salt Concentration, ● Aerated Liquid Phase, □ Un-aerated liquid phase,  $COD_0 = 5000$  mg/L)

Variation of Effluent COD Concentration with A/Q for 1% salt concentration are compared in Figure 5.2.5. for aerated and un-aerated liquid phases in the RBC unit. COD concentration decreased and COD removal efficiency increased with increasing A/Q ratio. COD removal efficiencies obtained with un-aerated liquid phase were considerably lower than those of with aerated liquid phase for the same A/Q ratios.

### 5.3. Mathematical Modelling and Determination of Kinetic Constants

COD balance for the RBC unit with liquid phase aeration can be written as follows :

$$Q(S_o-S) = \frac{R_{mf} \cdot S}{K_s+S} \cdot A + \frac{R_{ms} \cdot S}{K_s+S} \cdot V \quad (1)$$

$$Q(S_o-S) = (R_{mf} \cdot A + R_{ms} \cdot V) \cdot \frac{S}{K_s+S} \quad (1a)$$

$$Q(S_o-S) = (a \cdot R_{mf} + R_{ms}) \cdot \frac{S}{K_s+S} \cdot V \quad (1b)$$

$$\text{or } Q(S_o-S) = (R_{mf} + R_{ms}/a) \cdot \frac{S}{K_s+S} \cdot A \quad (2)$$

$$\frac{S_o-S}{\theta_H} = (a \cdot R_{mf} + R_{ms}) \cdot \frac{S}{K_s+S} \quad (3)$$

$$\frac{S_o-S}{A/Q} = (R_{mf} + R_{ms}/a) \cdot \frac{S}{K_s+S} \quad (3a)$$

In reciprocal form,

$$\frac{\theta_H}{S_o-S} = \frac{1}{a \cdot R_{mf} + R_{ms}} + \frac{K_s}{a \cdot R_{mf} + R_{ms}} \cdot \frac{1}{S} \quad (4)$$

or

$$\frac{A/Q}{S_o-S} = \frac{1}{R_{mf} + R_{ms}/a} + \frac{K_s}{R_{mf} + R_{ms}/a} \cdot \frac{1}{S} \quad (5)$$

$$\frac{A/Q}{S_o-S} = \frac{1}{R_{mT}} + \frac{K_s}{R_{mT}} \cdot \frac{1}{S} = \frac{1}{R} \quad (5a)$$

In these equations,  $Q$  is wastewater flow rate ( $\text{m}^3/\text{h}$ );  $S_o$  and  $S$  are influent and effluent COD concentrations ( $\text{kg COD}/\text{m}^3$ );  $R_{mf}$  is COD removal rate for the biofilm ( $\text{kg COD}/\text{m}^2 \cdot \text{h}$ );  $R_{ms}$  is COD removal rate in liquid phase ( $\text{kg COD}/\text{m}^3 \cdot \text{h}$ );  $a$  is disc surface area for unit liquid volume ( $\text{m}^2 \text{ disc}/\text{m}^3 \text{ water}$ ),  $A$  is wet area of discs ( $\text{m}^2$ ),  $V$  is volume of the reactor ( $\text{m}^3$ ).

In Addition; following equations can be written for  $R_{mf}$  and  $R_{ms}$ ,

$$R_{mf} = k_f \cdot X_f = \frac{\mu_{mf}}{Y} \cdot X_f \quad (6)$$

$$R_{ms} = k_s \cdot X_s = \frac{\mu_{ms}}{Y} \cdot X_s \quad (6a)$$

$$A = 2 \cdot \pi \cdot N \cdot (r_o^2 - r_u^2) \quad (7)$$

In this equations,  $k_f$  and  $k_s$  are constants for COD removal rate for biofilm and liquid phase, respectively ( $1/\text{h}$ );  $X_f$  and  $X_s$  are concentrations of biomass for biofilm and liquid phase, respectively

( $\text{kg X}/\text{m}^2$ ), ( $\text{kg X}/\text{m}^3$ );  $\mu_{mf}$  and  $\mu_{ms}$  are specific growth rate of organisms for biofilm and liquid phase ( $1/\text{h}$ );  $Y$  is growth yield coefficient ( $\text{g X}/\text{g S}$ );  $N$  is number of discs,  $r_o$  is diameter of discs (m);  $r_u$  is diameter of dry part of disc (m)

One can plot  $(A/Q)/(S_o - S)$  versus  $1/S$  in order to determine kinetic constants, for salt free wastewater as shown in Figure 5.3.1.

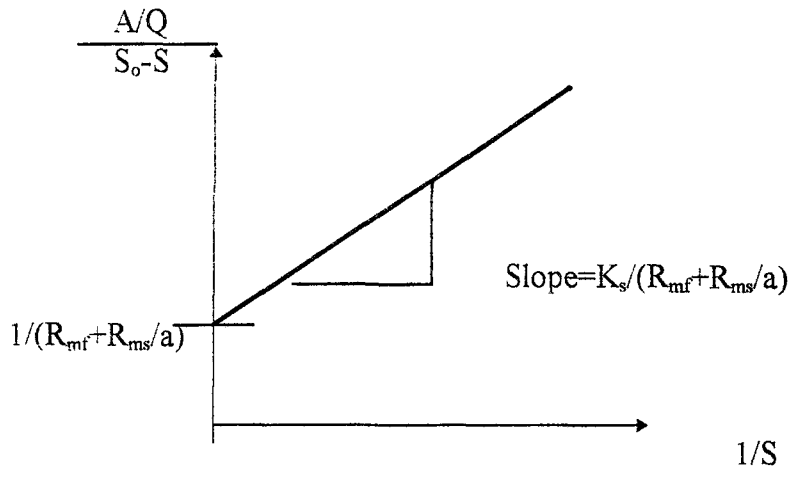


Figure 5.3.1.  $1/R_{mT}$  versus  $1/S$  for Determination of Kinetic Constants

## Effects of Salt Concentration

COD balance for biological treatment of saline wastewater in a continuously operating RBC unit can be written as follows:

$$Q.(S_o-S)=(R_{mf}+R_{ms}/a) \cdot \frac{S}{K_s+S} \cdot A \cdot \frac{K_T}{K_T+T} \quad (8)$$

or

$$R_s = \frac{Q}{A} (S_o-S) = \frac{R_{mT} \cdot S}{K_s+S} \cdot \frac{K_T}{K_T+T} = R_o \cdot \frac{K_T}{K_T+T} \quad (8a)$$

The first term on the right hand side of this equation represents effect of COD and the second term designates the effect of salt concentration on COD removal rate. T and  $K_T$  are salt concentration and salt inhibition constants, respectively.  $R_o$  is COD removal rate for salt free wastewater.

For salt free wastewater, ( $T=0$ ) equation 8a. takes the following form:

$$R_s = R_o = \frac{R_{mT} \cdot S}{K_s+S} \quad (9)$$

In double reciprocal form we can write the following:

$$\frac{1}{R_s} = \frac{1}{R_o} = \frac{1}{R_{mT}} + \frac{K_s}{R_{mT}} \cdot \frac{1}{S} \quad (9a)$$

With saline wastewater equation 8a. can be written as:

$$\frac{1}{R_s} = \frac{1}{R_o} + \frac{1}{R_o \cdot K_T} \cdot T \quad (10)$$

Experimental data can be plotted by using equation 9a. and 10. for salt free and saline wastewater in order to determine the kinetic coefficients.



### a. Aerated Liquid Phase

Experiments with aerated liquid phase were conducted with 1% to 5% salt concentrations. Experimental data were plotted as  $1/R_s$  versus  $T$  as shown in equation 10. Figure 5.3.2.  $R_o$  and  $K_T$  coefficients were determined as follows.

$$R_o = 5.4 \cdot 10^{-3} \text{ kg/m}^2 \cdot \text{h} = 5400 \text{ mg/m}^2 \cdot \text{h}$$

$$K_T = 65 \text{ kg/m}^3 = 65000 \text{ mg/L}$$

Therefore, equation 8a. takes the following form

$$R_s = 5.4 \cdot 10^{-3} \cdot \frac{65}{65+T} \quad (10a)$$

where  $R_s$  is in units of  $\text{kg COD/m}^2 \cdot \text{h}$

No experiments were conducted with saline free wastewater. Therefore, experimental results obtained with 1% salt concentration were used for determination of  $R_{mT}$  and  $K_s$ .

For 1% salt concentration equation 8a. takes the following form:

$$R_s = \frac{R_{mT} \cdot S}{K_s + S} \cdot \frac{65}{65+10} = 0.865 \cdot \frac{R_{mT} \cdot S}{K_s + S} \quad (11)$$

In double reciprocal form equation 11 can be written as:

$$\frac{0.865}{R_s} = \frac{1}{R_{mT}} + \frac{K_s}{R_{mT}} \cdot \frac{1}{S} \quad (11a)$$

Experimental data for 1% salt concentration was plotted as  $0.865/R_s$  versus  $1/S$  (Figure 5.3.3. From the intercept and the slope of the line one can find the following:

$$R_{mT} = 1.7 \cdot 10^{-2} \text{ kg/m}^2 \cdot \text{h} = 17000 \text{ mg COD/m}^2 \cdot \text{h}$$

$$K_s = 2.1 \text{ kg/m}^3 = 2100 \text{ mg/L}$$

Therefore, the rate expression for aerated liquid phase can be written as follows:

$$R_s = \frac{1.7 \cdot 10^{-2} \cdot S}{2.1+S} \cdot \frac{65}{65+T} \quad (12)$$

(kg/m<sup>2</sup>.h)

or

$$R_s = \frac{17000 \cdot S}{2100+S} \cdot \frac{65000}{65000+T} \quad (12a)$$

(mg/m<sup>2</sup>.h)

### b. Un-aerated Liquid Phase

Experiments with un-aerated liquid phase were only performed with 1% salt concentration at different A/Q ratios (or residence times). Since no experiments were performed with different salt concentrations, we were unable to determine  $K_T$ , by using experimental data. The  $K_T$  value was assumed the same as that of the aerated experiments ( $K_T=65 \text{ kg/m}^3=65000 \text{ mg/L}$ )

For 1% salt concentration, equation 8a. can be written as

$$R_s = \frac{R_{mT} \cdot S}{K_s+S} \cdot \frac{65}{65+10} = 0.865 \cdot \frac{R_{mT} \cdot S}{K_s+S} \quad (13)$$

In double reciprocal form equation 13. , becomes

$$\frac{0.865}{R_s} = \frac{1}{R_{mT}} + \frac{K_s}{R_{mT}} \cdot \frac{1}{S} \quad (13a)$$

Experimental data (obtained at different  $\theta_H$  or A/Q ratio) were plotted as  $0.865/R_s$  versus  $1/S$  (Figure 5.3.4.). From the intercept and slope of the line, one can obtain the following:

$$R_{mT}=5.65 \cdot 10^{-3} \text{ kg COD/m}^2 \cdot \text{h}=5650 \text{ mg COD/m}^3 \cdot \text{h}$$

$$K_s=0.88 \text{ kg/m}^3=880 \text{ mg/L}$$

Therefore, the rate equation for COD removal takes the following form for unaerated liquid phase.

$$R_s = \frac{5.65 \cdot 10^{-3} \cdot S}{0.88+S} \cdot \frac{65}{65+T} \quad (14)$$

(kg/m<sup>2</sup>·h)

or

$$R_s = \frac{5650 \cdot S}{880+S} \cdot \frac{65000}{65000+T} \quad (14a)$$

(mg/m<sup>2</sup>·h)

Kinetic coefficients obtained for aerated and unaerated liquid phases are compared in Tablo 5.3.1.

Table 5.3.1. Comparison of Kinetic Coefficients for Aerated and Unaerated Liquid Phase

	$R_{mT}$ (mg/m <sup>2</sup> ·h)	$K_s$ (mg/L)	$K_T$ (mg/L)
Aerated Liquid Phase	17000	2100	65000
Unaerated Liquid Phase	5650	880	65000

As one can see from the Table 5.3.1.; maximum rate of COD removal ( $R_{mT}$ ) for aerated liquid phase is about three times larger than that obtained for unaerated liquid phase.  $K_s$  values also increased upon aeration ( $K_{s,aer}=2100$  mg/L versus  $K_{s,unaer}=880$  mg/L) This increase indicates an adverse effect on COD removal rate with aeration. However, the effect of  $R_{mT}$  on COD removal rate is more pronounced than the effect of  $K_s$ , resulting in higher COD removal rates with liquid phase aeration (Figure 5.2.5.)

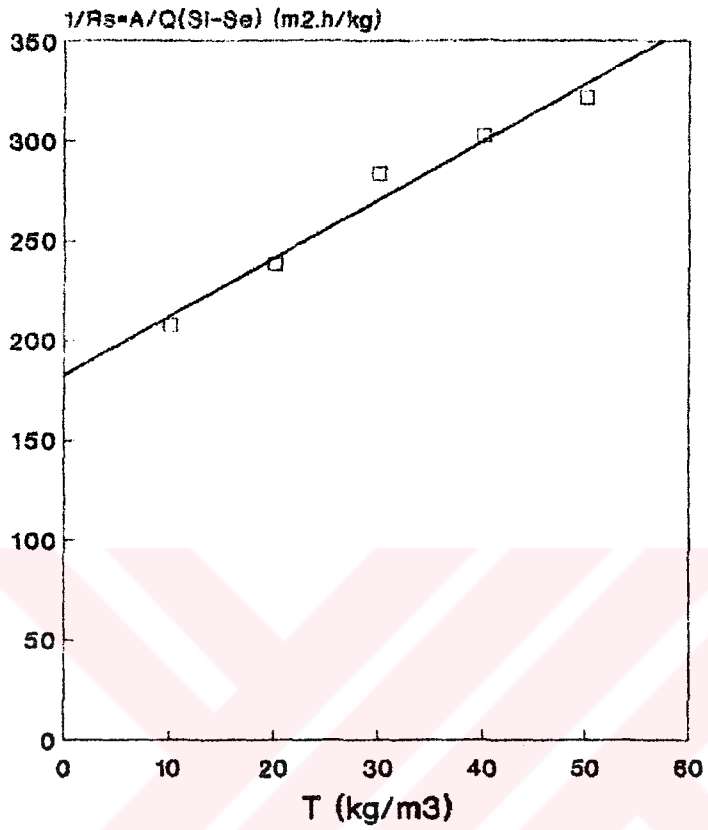


Figure 5.3.2.  $1/R_s = A/Q(S_o - S_e)$  versus Salt Concentration  $T$  (%) for Determination of Kinetic Constants (Aerated Liquid Phase)

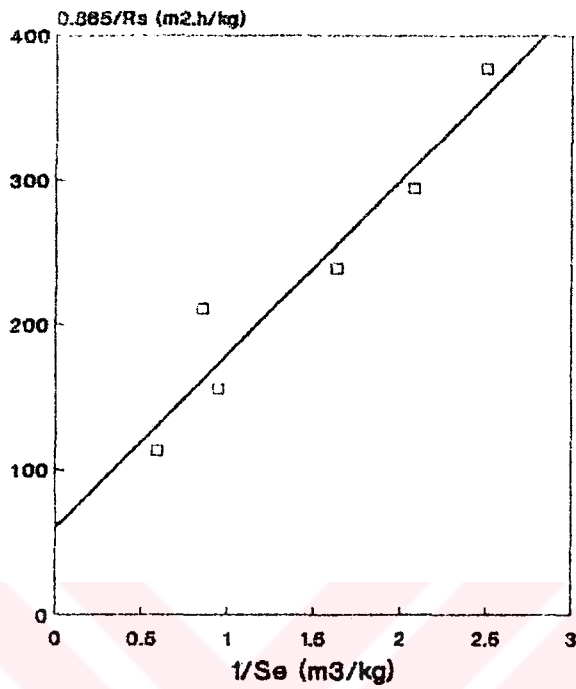


Figure 5.3.3.  $0.865/R_s$  versus  $1/S$  for Determination of Kinetic Constants  
(Aerated Liquid Phase, 1% Salt Concentration)

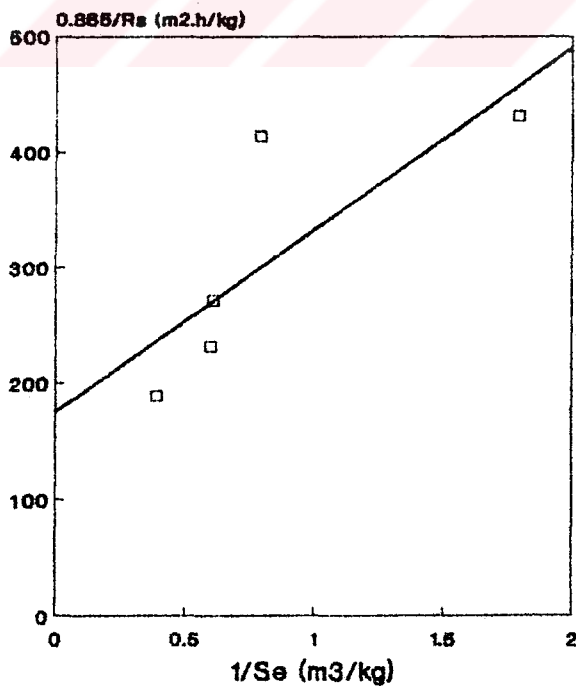


Figure 5.3.4.  $0.865/R_s$  versus  $1/S$  for Determination of Kinetic Constants  
(Unaerated Liquid Phase, 1% Salt Concentration)

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## CHAPTER SIX

# CONCLUSIONS AND RECOMMENDATIONS

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### Conclusions:

Among the cultures tested for biological treatment of saline wastewater in the percolator unit a mixed culture of activated sludge and Halobacter was found to be superior to the others.

Salt concentration adversely affected the performance of the mixed culture up to 3% salt concentration. High COD removal efficiencies were obtained at salt concentrations above 4% as a result of activity of Halobacter.

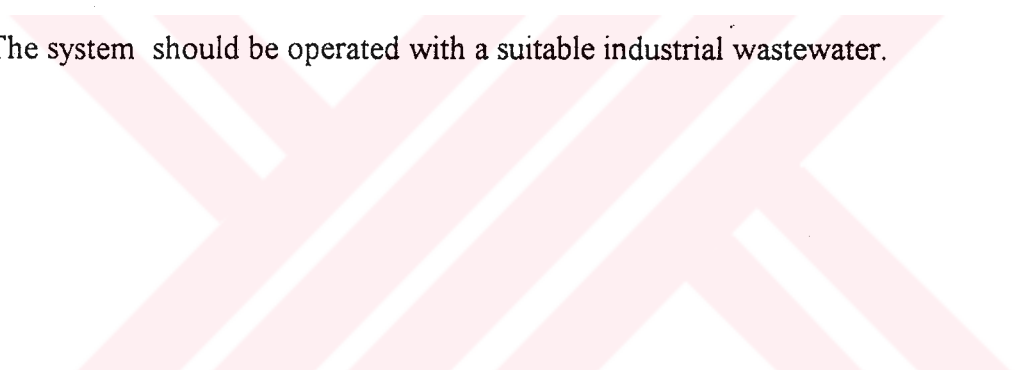
Therefore, inclusion of Halobacter into A.sludge culture was especially beneficial at high salt concentrations above 4%.

In experiments with RBC unit, COD removal efficiency increased with A/Q ratio. Effluent COD concentration increased and COD removal efficiency decreased with COD loading rate. Salt concentration adversely affected the system' s performance. Effluent COD concentration increased and COD removal efficiency decreased with salt concentration. Similarly, effluent COD concentration increased and COD removal efficiency decreased with increasing feed COD concentration. Liquid phase aeration favorably affected the system' s performance. COD removal efficiency significantly increased with liquid phase aeration.

A mathematical model was developed and kinetic constants were determined by using experimental data.

**Recommendations:**

The system performance must be investigated in more details, as specified below:

- 1.) Salt concentration should be varied in a larger range such as 0%-10%, in future experiments.
  - 2.) Effects of disc surface area on system performance should be investigated.
  - 3.) The system should be operated with sedimentation tank and liquid recycle.
  - 4.) Effect of Halobacter concentration in inoculum on the system's performance needs to be investigated.
  - 5.) The system should be operated with a suitable industrial wastewater.
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## CHAPTER SEVEN

## REFERENCES

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1. Kincannon, D. F., Gaudy, A.F. .Some Effect of High Salt Concentration on Activated Sludge. *J. WPCF*.38(7): 1148-1158, 1996.
2. Kincannon, D. F., Gaudy, A. F. .Response of Biological Waste Treatment Systems to Changes in Salt Concentrations. *Biotechnol.Bioeng* 10: 483-496, 1968.
3. Belkin, S., Brenner, A., Abeliovich, A. .Biological Treatment of a High Salinity Chemical Industrial Wastewater. *Wat. Sci. Technol.* 27: 105-112, 1993.
4. Kinner, N. E., Bishop, P. L., Asce, M. .Treatment of Saline Domestic Wastewater Using RBC's. *J. Environ. Eng., ASCE*.108: 650-663, 1982.
5. Woolard, C. R., Irvine, R. L. .Biological Treatment of Hypersaline Wastewater By a Biofilm of Halophilic Bacteria. *Wat. Env. Res.* 66(3): 230-235, 1994.
6. Woolard, C. R., Irvine, R. L. .Treatment of Hypersaline Wastewater in the Sequencing Batch Reactor. *Was. Res.* 29(4): 1159-1168, 1995.
7. Lawton, G. W., Eggert, C. V. .Effect of High Sodium Chloride Concentration on Trickling Filter. *Slimes.J. Wat. Pollut. Cont. Fed.* 29: 1228-1236, 1957.
8. Stewart, M. J., Ludwig, H. F., Kearnes, W. H. .Effects of Varying Salinity on the Extended Aeration Process. *J. Wat. Poll. Cont. Fed.* 34: 1161-1177, 1962.



9. Burnett, W. E. .The Effect of Salinity Variations on the Activated Sludge Process. Wat. Scw. Works. 121: 37-38, 1974.
10. Oren, A., Gurevich, P., Malkit, A., Henis, Y. .Microbial Degradation of Pollutants at High Salt Concentrations. Biodegradation.3: 387-398, 1992.
11. Ludzack, F. J., Noran, D. K. .Tolerance of High Salinities By Conventional Wastewater Treatment Processes. J. Wat. Poll. Cont. Fed.37(10): 1404-1416, 1965.
12. Standard Methods for the Examination of Water and Wastewater. 17 th Edn. APHA. Washington, D. C., 1989.
13. Kargı, F., Karaduman, A. .Tuz İçeren Atıksuların Biyolojik Arıtımında Halobacterium Halobium'un Kullanımı. Lisans Tezi, Haziran-1995.
14. Kargı,F., Dinçer,A. R. .Aerobic Biological Treatment of Saline Wastewater By Using Fed-Batch Operation and Halophilic Bacteria. <sup>DEÜ İFSE YALISIRCA</sup> Masters Thesis September, 1995.
15. Kargı, F. .Fundamentals of Environmental Biotechnology II.; lecture Notes, Dokuz Eylül University, Department of Environmental Engineering, 1992.
16. Shuler, ML and Kargı, F. .Bioprocess Engineering: Basic Concepts, Prentice Hall USA, 1992.
17. Hochstein, L. .The Physiology and Metabolizm of the Extremely Halophilic Bacteria, in Halophilic Bacteria, Vol. II, CRC Press. R. Radriguez.Valera, ed., BocaRaton, FL., 1988.
18. Kargı, Fikret. .Çevre Mühendisliğinde Biyoprosesler, DEÜ Mühendislik Fakültesi Yayınları, 1993.
19. Kushner, D. J., and Kamekura, M. .Phyiology of Halophilic Eubacteria.In Halophilic Bacteria. Vol., F. Rodriguez Valera (Ed.) CRC Press, BocaRaton, Fla., 1988.

20. Kargı, F. and Uygur, A. .Biological Treatment of Saline Wastewater in an Aerated Percolator Unit Utilizing Halophilic Bacteria., *Env. Tech.*, 1995.
21. George, T., Franklin, L. Burton, *Wastewater Engineering Treatment, Disposal and Reuse*, McGraw-Hill International Editions Civil Engineering Series, 1991.
22. Alpaslan, N., Okutucu, S. .Evoluation of the Performance of an RBC System, *Environment 88; Environmental Science and Technology Conference*, 5-9 June, İzmir/TÜRKİYE, 1988.
23. Simon Gonzalez-Martinez and Javier Duque. .Aerobic Submerged Biofilm Reactors for Wastewater Treatment, *Wat. Res. Vol. 26. No.6*, pp. 825-833, 1992.
24. Piachristiansen, Line Hollesen and Poul Harremoes. .Liquid Film Diffusion on Reaction Rate in Submerged Biofilters., *Wat. Res. Vol. 29. No.3*, pp. 947-952, 1995.
25. Çeçen, F. .Batık Filtrelerde Nitrifikasyon ve Denitrifikasyon Kinetiği, *SKKD Cilt 3, Sayı 2*, Sh. 113-120, 1993.
26. Boshou Pan, and L. Hartman. .Activity of Biomass in RBC System Treating Pulp Industrial Wastewater., *Journal of Env. Eng.*, Vol. 118, No.5, 1992.
27. Wilson, F. .Prediction of Rotating Biological Contactor Efficiency Using TOC., *Journal of the Env. Eng. Vol. 119, No.3*, 1993.
28. Charles I. Noss, Roy D. Miller, M.Asce, and Edgar D. Smith. .Rotating Biodisks Recarbonate Alkaline Wastewater., *Journal of the Env. Eng. Vol. 108, No. EE2*, 1982.
29. Ching-San Huang. .Nitrification Kinetics and Its RBC Application., *Journal of the Env. Eng.*, Vol. 108, No. EE3, 1982.

30. Roland Leduc and Ian Buchanan. .Minimization of Multistage RBC Active Disc Area., Journal of the Env. Eng. Vol. 119, No.2, 1993.
31. Edward J. Opatken. .RBC Nitrification Design Using Zero-Order Kinetics., Environmental Progress, Vol. 12, No.4, Page: 262-265, 1993.
32. Yeun C. Wu, E. D. Smith, and John Gratz. .Prediction of RBC Performance for Nitrification. Journal of the Env. Eng. Vol. 107. No.EE4, 1981.
33. E. B. Pike. .Rotating Biological Contactors. Wat. Res. Centre, Medmemhan, Marlow, Bucks, UK.
34. Andreas D., Andredakis, M. .Design of Multistage Rotating Biological Contactors. Journal of the Env. Eng. Vol. 113, No.1, 1987.
35. F. Wilson. .Kinetics and Reaction Order in Rotating Biological Contactors Using TOC., Wat. Res. Vol. 27, No.5, pp: 1423-1429, 1993.
36. Yeun C. Wu, and Ed D. Smith. .Rotating Biological Contactor System Design., Journal of the Env. Eng. Vol. 108, No.EE3, 1982.
37. Chi-Yuan Lee. .Model For Biological Reactors Having Suspended and Attached Growth., .Jornal of Environmental Engineering ,Vol. 118, No.6, pp: 982-987, 1992

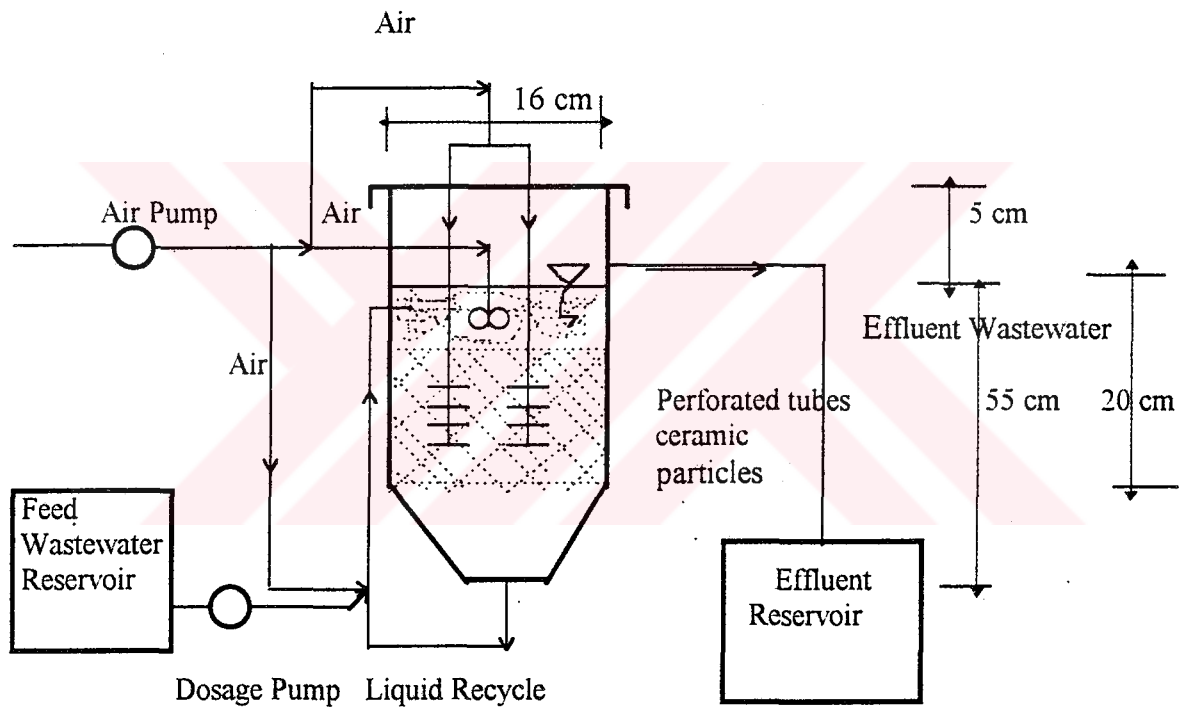
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## CHAPTER EIGHT APPENDICES

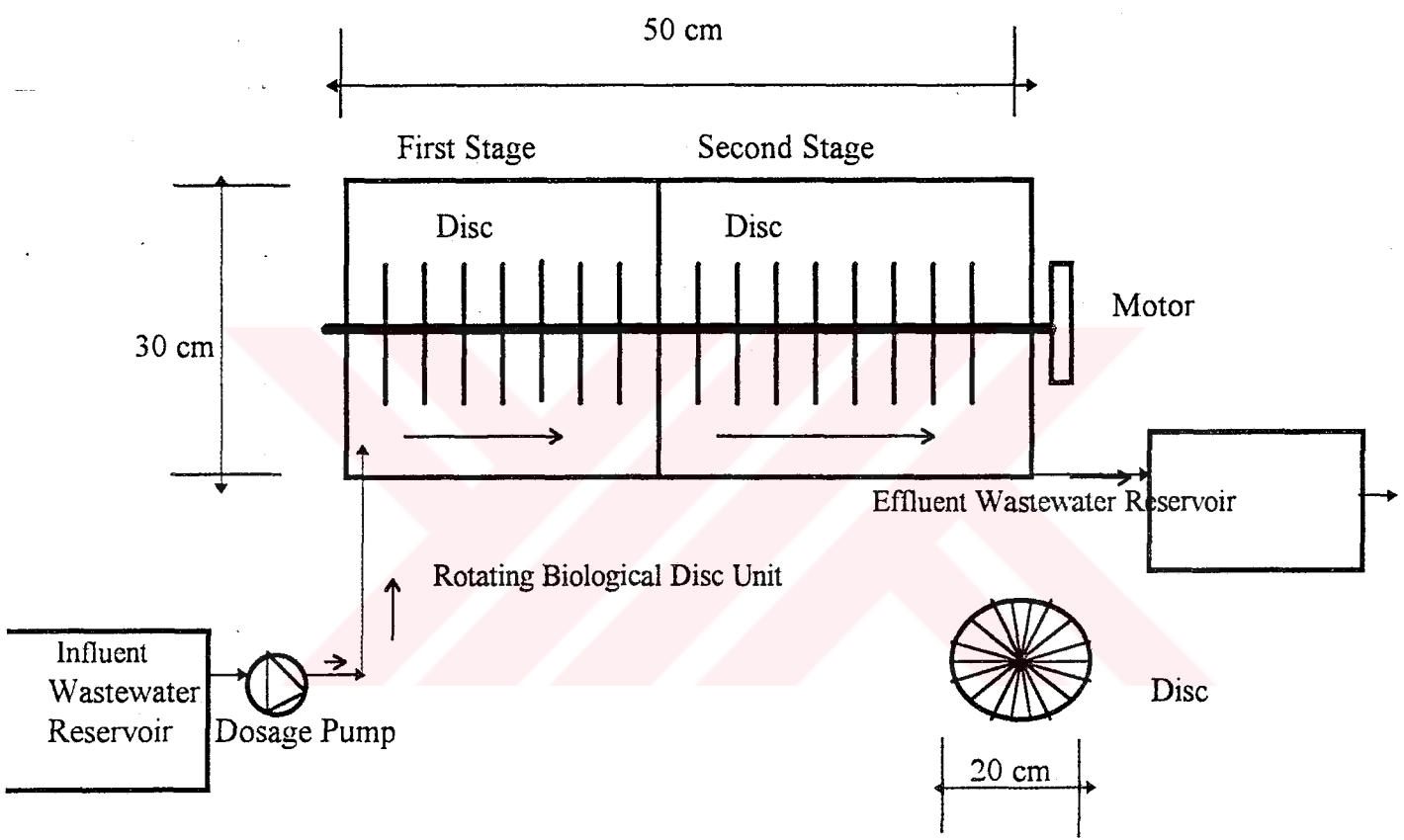
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### 8.1. Dimensions of Experimental Set-up

#### Aerated Percolator Unit



### Rotating Biological Contactor Unit



## 8.2. Raw Data of Experiments

TABLE 8.2.1. Average Raw Data for Continuous Operation of Aerated Perculator Experiments (1% Salt Concentration)

Effect of the microbial culture on system's performance

Types of Microorganism	$S_o$ (mg/L)	$S_e$ (mg/L)	Q (L/h)	$L_o$ (mg/L.h)	$R_s$ (mg/L.h)	E (%)
Zoogleo Ramigera	5327	3277	1.5	1229	473	38
Activated Sludge	4787	1880	1.5	1110	676	61
Halobacter Halobium	5476	3234	1.5	1275	529	41
Zoogleo Ramigera and Halobacter Halobium	5533	3233	1.5	1257	531	42
Activated Sludge and Halobacter Halobium	5160	592	1.5	1204	1067	89

$S_i=5000$  mg COD/l,  $\theta_H=4.3$  h,  $D_p=4.50$  mm (Ceramic),  $H=20$  cm,  $T=25$  °C, COD/N/P:100/10/1

TABLE 8.2.2. Average Raw Data for Continuous Operation of Aerated Percolator Experiments (Activated Sludge and Halobacter Halobium)

Effect of salt concentration on system' s performance

Salt Concentration (%)	$S_o$ (mg/L)	$S_e$ (mg/L)	Q (L/h)	$L_o$ (mg/L.h)	$R_s$ (mg/L.h)	E (%)
1	5160	592	1.5	1204	1067	89
2	5560	1480	1.5	1286	945	73
3	5160	2640	1.5	1172	563	49
4	5027	1960	1.5	1160	708	61
5	5660	2547	1.5	872	480	55

$S_i=5000$  mg COD/L,  $\theta_H=4.3$  h,  $D_p=4.50$  mm (Ceramic),  $H=20$  cm.  $T=25$  °C, COD/N/P:100/10/1

TABLE 8.2.3. Average Raw Data for Continuous Operation of RBC Experiments  
(Activated Sludge and Halobacter Halobium, 1% Salt Concentration)

Effect of A/Q ratio (or  $\theta_H$ ) on system's performance

$\theta_H$ (h)	$S_o$ (mg/L)	$S_e$ (mg/L)	Q (L/h)	$L_s$ (mg/m <sup>2</sup> .h)	$R_s$ (mg/m <sup>2</sup> .h)	X (mg/L)	E (%)	$\theta_H/(S_o-S_e)$ (m <sup>3</sup> .h/kg)	1/Se (m <sup>3</sup> /kg)	A/Q (m <sup>2</sup> .h/m <sup>3</sup> )	A/Q(Si-Se) (m <sup>2</sup> .h/kg)
2	5787	1707	4.75	10943	7715	2400	71	0.49	0.59	528.84	130
3	5467	1067	3.17	6892	5553	2902	81	0.68	0.94	792.43	180
4	5520	1173	2.38	5219	4119	2170	79	0.92	0.85	1055.46	243
5	5387	613	1.90	4075	3611	4000	83	1.05	1.63	1322.11	277
6	5173	480	1.58	3261	2952	3970	92	1.28	2.08	1589.87	339
8	5253	400	1.19	2483	2300	3880	92	1.65	2.50	2110.92	435

$S_o=5000$  mg COD/L, aerated liquid phase, COD/N/P:100/10/1



TABLE 8.2.4. Average Raw Data for Continuous Operation of RBC Experiments  
(Activated Sludge and Halobacter Halobium)

Effect of salt concentration on system's performance

Salt Concentration (%)	$S_o$ (mg/L)	$S_e$ (mg/L)	Q (L/h)	$L_s$ (mg/m <sup>2</sup> .h)	$R_s$ (mg/m <sup>2</sup> .h)	X (mg/L)	E (%)
0	5333	320	2.38	5042	4740	4070	94
1	5733	640	2.38	5420	4815	5000	89
2	5533	1093	2.38	5231	4198	4780	80
3	4960	1233	2.38	4689	3524	5500	75
4	4880	1387	2.38	4614	3302	7120	72
5	5653	2360	2.38	5345	3113	9940	58

$S_o=5000$  mg COD/L,  $\theta_H=4$  h, aerated liquid phase, COD/N/P:100/10/1

TABLE 8.2.5. Average Raw Data for Continuous Operation of RBC Experiment  
(Activated Sludge and Halobacter Halobium, 1% Salt Concentration)

Effect of A/Q ratio on system's performance

$\theta_{H}$ (h)	$S_o$ (mg/L)	$S_e$ (mg/L)	Q (L/h)	$L_s$ (mg/m <sup>2</sup> .h)	$R_s$ (mg/m <sup>2</sup> .h)	X (mg/L)	E (%)	$\theta_{H}/(S_o/S_e)$	1/Se (m <sup>3</sup> /kg)	A/Q (m <sup>2</sup> .h/m <sup>3</sup> )	A/Q(Si-Se) (m <sup>2</sup> .h/kg)
2	4987	2560	4.75	9430	4589	1940	49	0.82	0.39	528.84	218
3	4613	1653	3.17	5820	3731	2150	64	1.01	0.60	792.43	268
4	4987	1627	2.38	4720	3177	2230	67	1.19	0.61	1055.46	314
6	4587	1267	1.58	2890	2093	3290	72	1.81	0.79	1589.87	479
8	4800	560	1.19	2270	2004	4040	88	1.89	1.79	2110.92	498

$S_o=5000$  mg COD/L, unaerated liquid phase, COD/N/P:100/10/1

TABLE 8.2.6. Average Raw Data for Continuous Operation of RBC Experiment  
(Activated Sludge and Halobacter Halobium, 1% Salt Concentration)

Effect of feed COD concentration on system's performance

$S_o$ (mg/L)	$S_e$ (mg/L)	Q (L/h)	$L_s$ (mg/m <sup>2</sup> .h)	$R_s$ (mg/m <sup>2</sup> .h)	X (mg/L)	E (%)
2347	560	2.38	2219	1690	3000	76
4987	1627	2.38	4715	3177	2230	67
6907	3067	2.38	6530	3631	5500	55
9200	5173	2.38	8698	3807	4800	44
11307	7707	2.38	10690	3404	2300	31

$\theta_H=4$  h, unaerated liquid phase, COD/N/P:100/10/1

TÜRKÇE ABSTRAKT (en fazla 250 sözcük) \_\_\_\_\_ :

(TÜBİTAK / TÜRDOK'un Abstrakt Hazırlama Kılavuzunu kullanınız.)

## ÖZET

Özellikle deniz kıyısındaki evsel ve endüstriyel atıksular tuz içerdikleri için (%o 5 - %o 35), arıtma tesislerinin verimleri düşük olmaktadır. Bunun nedeni yüksek tuz konsantrasyonlarında biyolojik arıtma ünitelerindeki organizmaların plazmoliz nedeniyle parçalanmaları ya da aktivitelerini kaybetmeleridir. Atıksulardan tuzun ters osmoz ya da iyon değiştiriciler ile giderimi, işletme ve yatırım maliyetlerini artırdığından pratikte uygulanamamaktadır. Oysa bazı tuz tolere edebilen organizmalar (**Halobacterium Halobium ya da Dunaliella Salina**) yüksek tuz konsantrasyonlarında (% 1 - % 10 NaCl) ortamdan karbon ve azot bileşiklerini giderip büyüebilmektedir. Bu organizmaların tuz içeren atıksuların biyolojik arıtımında kullanımı, arıtma verimini artırma potansiyeline sahiptir. Ancak bu konuda literatürde kapsamlı bir çalışma bulunmamaktadır.

Bu projenin amacı tuz içeren atıksuların arıtımında halophilic bir bakteri olan Halobacterium halobium'u kullanarak arıtma verimini yükseltmek ve sistemin davranışını bilimsel yöntemlerle incelemektir. Sentetik atıksular sürekli çalışan bir havalandırılmalı percolatör ünitesi ve dönen biyodisk ünitelerinde Halobacter halobium içeren karma kültürlerle değişik tuz konsantrasyonlarında arıtılarak sistemin kinetik analizi yapıldı ve kinetik sabitleri saptandı. Sistemin performansı üzerine önemli değişkenlerin etkileri incelendi. Sistemin davranışını belirleyen bir matematiksel model geliştirilerek veriler değerlendirildi ve model parametreleri tayin edildi.

Bu çalışmalar tuz içeren atıksuların biyolojik arıtımına yeni teknolojik yaklaşımlar getirmiş, ve bu tür sistemlerin daha iyi anlaşılmasına olanak sağlamıştır.

## ABSTRACT

Efficiency of wastewater treatment operations near seashore areas is usually low because of high salt content of industrial and domestic wastewater in those areas (%o 5 - %o 35). The reason for this is loss of activity of organisms in biological wastewater treatment operations due to plasmolysis of cells. Removal of salts from wastewater before treatment by reverse osmosis or ion exchange requires high capital and operating costs and is not practical. However, some halophilic organisms (**e.g. Halobacter halobium or Dunaliella salina**) can grow and remove carbon and nitrogen compounds from wastewater under saline conditions (%1-%10 NaCl). Utilization of these organisms for biological treatment of saline wastewaters has a great potential of improving the treatment efficiency. However, there are no extensive studies on this subject in literature.

Major objective of this study is to improve the treatment efficiency of saline wastewaters by using the **halophilic organisms** Halobacterium halobium in a percolator and in a RBC unit and to investigate the system's behavior. Synthetic wastewaters with different salt concentrations were treated in a continuous aerated submerged filter (**PERCOLATOR**) and rotating biodisc unit (**RBC**) by using a mixed culture of organisms containing H.halobium. Kinetic analysis of this system was accomplished. Variation of system performance with major process variables was investigated. A mathematical model describing the system behavior was developed and the model parameters was determined by using the experimental data.

These studies provided new technological approaches for biological treatment of saline wastewaters and also contributed to the understanding of behavior of such systems.