

**USE OF RHAMNOLIPID TO REMOVE HEAVY METALS  
FROM AQUEOUS STREAMS VIA MICELLAR  
ENHANCED ULTRAFILTRATION**

**Mohamed Abdel Monem El Zeftawy**

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## ABSTRACT

### USE OF RHAMNOLIPID TO REMOVE HEAVY METALS FROM AQUEOUS STREAMS VIA MICELLAR ENHANCED ULTRAFILTRATION OF IONS

M. A. Monem El Zeftawy, Ph. D.  
Concordia University, 2006

In this research, the rhamnolipid biosurfactant was utilized in micellar-enhanced ultrafiltration (MEUF) of heavy metals from contaminated waters. The effects of different major operating conditions on the (MEUF) system performance were investigated for copper, zinc, nickel and cadmium using two membranes. The optimal conditions were successfully applied to treat six contaminated wastewaters from metal refining industries and three contaminated groundwaters using the two membranes (> 99% rejection ratio). To efficiently choose the most influential factors to the MEUF system, optimization by the response surface methodology approach was utilized and data quality was examined.

Three studies were performed in order to investigate the mechanism and the interactions between the heavy metal ions and the rhamnolipid micelles in the rhamnolipid-based UF system. The first study successfully investigated the applicability of Gouy-Chapman approach for binding of heavy metal ions to rhamnolipid biosurfactant in the MEUF system and the mechanism via electrostatic attraction was suggested. The second study efficiently evaluated binding behavior of the four heavy metal ions individually and collectively onto the rhamnolipid. The order was  $\text{Cu}^{2+} > \text{Cd}^{2+} > \text{Zn}^{2+} > \text{Ni}^{2+}$ . The third study

analyzed the equilibrium data using Langmuir, Freundlich, and linear adsorption models. The Freundlich model fitted the data better and the  $K_F$  values were from 1.3 to 6.1.

In addition, solidification/stabilization (S/S) technology was successfully used to stabilize solid wastes contaminated with heavy metal. Furthermore, preliminary trials were performed to examine the recovery of the rhamnolipid from the retentates for possible reuse. The amounts recovered ranged from  $12 \pm 1 \%$  to  $18 \pm 1 \%$  of the rhamnolipid concentration initially present in the retentate. Similarly, preliminary trials to reuse water from the retentates were investigated. The total portions recovered ranged from about  $81.0 \pm 0.34 \%$  to  $82.3 \pm 0.46 \%$  of the total feed water. This work may be used as preliminary investigations for further detailed pilot and/or large-scale studies to treat contaminated groundwater and industrial wastewater.

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*I dedicate this work to my dear family.*

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## LIST OF SYMBOLS AND ABBREVIATIONS

$a_L$	adsorption intensity (Langmuir model)
$[C]_L$	bulk phase concentration (wt/vol.)
$C_{L,AD}$	Concentration of heavy metal ions at equilibrium (linear adsorption model)
COD	Chemical oxygen demand
CMC	Critical micelle concentration
k	Boltzmann constant
$K_L$	An indicator of adsorption capacity (Langmuir model)
$K_{L,AD}$	The distribution coefficient (linear adsorption model)
$K_F$	An indicator of adsorption capacity (Freundlich model)
MWCO	Molecular weight cut off
MEUF	Micellar Enhanced Ultrafiltration
n	Expresses adsorption intensity
$[Q]_L$	The adsorbed phase concentration
$[Q_F]$	Mass of adsorbate adsorbed per unit mass of adsorbent at equilibrium
$Q_{L,AD}$	The mass of heavy metal ion sorbed per unit mass of rhamnolipid
$R^2$	Square linear regression

## LIST OF SYMBOLS AND ABBREVIATIONS (CONT'D)

RS	Response surface
SOP	Second order polynomial (SOP)
T	Absolute temperature (degrees K)
[X <sub>F</sub> ]	Equilibrium concentration of adsorbate in solution at equilibrium (mass/volume)

# CHAPTER 1

## Introduction

### 1. Introduction

#### 1.1 General background and statement of the problem

The increasing pollution of industrial wastewater or groundwater by heavy metal ions has become a significant environmental problem in both industrialized and developing countries. In North America, some sites receive a continuous input of metal ions, specifically those near mining facilities and industrial waste discharge points. Heavy metals are responsible for much of the contamination found at hazardous waste sites in the United States, and have been detected in the soil and groundwater at approximately 65% of the U. S. Environmental Protection Agency Superfund sites (U. S. Environmental Protection Agency, 1997). Numerous developments to control pollution of heavy metal ions have been reported (Watmough and Hutchinson, 2004).

Heavy metals have many uses in industry. They are used to make batteries, to make pigments, they are used as catalysts in petroleum refining, as protective coatings for other metals so that they do not rust or corrode and as stabilizers in plastics. They are used in many other applications like chemical synthesis and metallurgical processes (Weiner, 2000; O'Neill, 1993). Because of human

activities and natural processes, heavy metals enter the air, water, and soil (Fergusson, 1990).

Pollution by heavy metals is of serious concern because they are non-biodegradable, and some of them are highly toxic and have a probable carcinogenic effect. Some heavy metals are known to be much more toxic than others. On the other hand, some heavy metals are essential elements for living beings in trace amounts. Mercury, lead and cadmium are known to be the 'big three' heavy metals poisons (Manahan, 2003). Potential short-term effects from exposure to cadmium are nausea, vomiting, shock, sensory disturbances, salivation convulsions, muscle cramps and diarrhea. Long-term exposure to toxic heavy metals can cause blood damage, fragile bones, liver damage, renal failure and lung damage (Garcia 2001; Weiner, 2000). Lead inhibits many enzymes and can cause paralysis of the limbs, neurological disorders, and comas (ATSDR, 1993).

Emphasis has been directed towards the removal of heavy metals from wastewaters and effluents. Traditionally, techniques such as sorption and chemical precipitation have been used for the removal of metal ions from aqueous effluents.

Removal of heavy metals from large volumes of wastewaters or groundwaters at low concentrations by traditional methods is not economical. This is because of

the large volumes of dilute polluted waters that need to be handled. In these cases, removal of heavy metal ions is more effective by ion-exchange resins or adsorption on solid surfaces. High capital and regeneration costs of activated carbon and ion-exchange resins are required in such cases. Low-cost adsorbents would be very good alternatives.

Membrane separation has become an attractive technique in the removal of heavy metals from large volumes of wastewaters or groundwaters at low concentrations. In order to separate metallic ions, reverse osmosis or nanofiltration should be used due to the size of the ions in aqueous phase. However, the usual permeate fluxes of reverse osmosis membranes are limited and require high transmembrane pressures, which makes the process expensive. The combination of surfactants and membranes in wastewater separations has led to the development of novel techniques or enhancement of existing techniques. Some examples of methods that apply surfactants are micellar-enhanced ultrafiltration (MEUF) methods (Huang et al., 2005).

The ability of synthetic surfactants to bind with heavy metal ions has been reported by researchers who have conducted bench-scale experiments involving water and wastewater treatment with different synthetic surfactants (Huang et al., 2005; Ahmadi et al., 1994). The results are in favor of using surfactants over other solvents or complexing agents for treatment ground water and wastewater contaminated with heavy metals (Akita et al., 1999; Yurlova et al., 2002).



The applications of synthetic surfactant-based separation methods possess a long-term risk of secondary pollution due to leakage. Principally in surfactant-enhanced ultrafiltration, surfactant molecules in concentrations above the CMC (critical micelle concentration) are mixed with polluted water. Smaller contaminants like heavy metal ions will be bound onto larger surfactant micelle complexes. As a consequence, heavy metal ions associated with surfactant macromolecules can be easily retained by an ultrafiltration membrane (Yurlova, et al., 2002). Thus, the permeate contains very low concentrations of metal ions and surfactant monomers. In surfactant-based ultrafiltration techniques, surfactant concentrations higher than the CMC are required. In many cases, monomeric surfactant molecules in the permeate can be discharged to the ecosystem or the environment if the surfactant is biodegradable. Biosurfactants can be used as suitable substitutes for chemical synthesized surfactants due to their biodegradability and renewable production sources (Lu et al., 2003). Neilson et al. (2003) used a biosurfactant as an environmentally compatible, not toxic compound. Thus, in biosurfactant-based ultrafiltration techniques, leakage into the permeate of biosurfactants cannot be considered another type of pollution due to their biodegradability, compatibility and non-toxic nature.

Biosurfactant-enhanced ultrafiltration membrane processes can be applied over a wide range of operating pressures, pH and temperatures. Also, it can minimize the waste to be disposed of. In addition, concentrated metals in the retentate can

be reused. Furthermore, the biosurfactant in the retentate may be regenerated. Biosurfactant regeneration is considered to be an important step to make the process economically feasible.

With regard to the limitations associated with the existing techniques, the use of biodegradable binding agents (like biosurfactants) might be a better alternative. Therefore, it would be highly desirable to investigate biosurfactant-based UF separation technique to remove heavy metal ions from wastewater and groundwater. A rhamnolipid biosurfactant was selected for this purpose.

## **1.2 Objectives**

The main purpose of this research is to develop a biosurfactant-based ultrafiltration technique for removal of five metal ions (cadmium, zinc, copper, lead and nickel) from metal refining wastewater and groundwaters and to develop an understanding of the binding mechanisms of the metals to the biosurfactant. A rhamnolipid biosurfactant produced by *Pseudomonas aeruginosa* was utilized in this work.

In order to meet these objectives, the following tasks were performed:

- Studying the influence of major operating conditions on the permeate flux and the removal efficiency in cases of all metal ions under investigation

- Applying the results produced from bench-scale MEUF experiments in order to provide an efficient method that can be used to treat the polluted groundwaters and to treat metal refining wastewaters.
- Investigating the applicability of the Gouy-Chapman approach for binding of heavy metal ions to rhamnolipid biosurfactant in the MEUF system.
- Studying the binding behaviors of the heavy metal ions individually and collectively to the rhamnolipid in MEUF system.
- Examining the applicability of the Langmuir binding model, the Freundlich binding and the linear isotherm model to the MEUF system.
- Investigating the possibility of regeneration for rhamnolipid biosurfactant
- Studying stabilization as a safe disposal of the wastes from the MEUF system.
- Examining the possibility of water reuse from the retentate.

### **1.3 Structure of the study**

The introduction and the aim of the work are presented in Chapter 1. Ultrafiltration (UF) enhanced by rhamnolipid biosurfactant was selected following an extensive literature search outlined in Chapter 2. Chapter 3 describes experimental methods and equipment utilized. Chapter 4 presents the experimental results and their extensive discussion. Conclusions from the results are drawn in Chapter 5. Chapter 6 presents the contributions of the reported

research. Chapter 7 provides recommendations designed for the direction of future research endeavors. Chapter 8 presents all the references.

# CHAPTER 2

## Background and Literature Review

### 2 Background and Literature Review

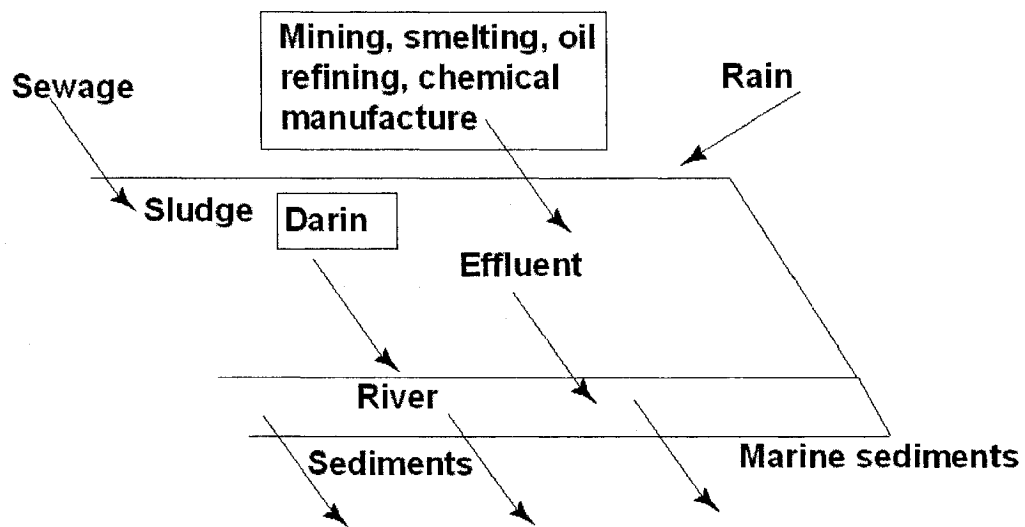
#### 2.1 Heavy metals

Heavy metals are very abundant in the earth's crust. In nature, heavy metals are most often present in many ores such as silicates, sulfides, and oxides (Harrison et al., 1996). Heavy metals have many uses in industry. They are used to make batteries and pigments, and in the manufacture of wires. Moreover, they are used as catalysts, in plating, in petroleum refining and in the hydrogenation of fats and oils (Weiner, 2000; O'Neill, 1993). They also can be mixed with other metals to form alloys, to serve as protective coatings for other metals so that they do not rust or corrode, to be mixed with other materials to produce viscose rayon yarns. They are used as stabilizers in plastics, and in the production of ground wood pulps and newspapers. They are used in many other applications in chemical synthesis and metallurgical processes (Weiner, 2000; O'Neill, 1993).

Because of industrial activities and natural processes, heavy metals enter the air, water, and soil. Most heavy metals enter the environment as the result of human

activities, such as mining, burning of wastes, metal production, chemical industries, domestic wastewater, purifying ores, coal burning, and metal manufacturing industries (Weiner, 2000; Fergusson, 1990). Heavy metals can also enter the environment as the result of smelting. Volcanoes also release some heavy metals to the environment. These releases can increase heavy metal levels in the atmosphere. In air, heavy metals present as fine dust particles. This dust can settle into soil and water. Also, rain and snow help in transporting heavy metals from air to soil and water (Lugon-Moulin et al., 2004; Fergusson, 1990).

Heavy metals can bioaccumulate into fish, plants, animals and humans (Harrison et al., 1996). Furthermore, it was reported that some heavy metals have potential to cause cancer (Weiner, 2000). Samples of blood can be collected and the level of heavy metals can be measured to determine whether high levels of toxic metals have accumulated in the body (Paasivirta, 1991). According to Zakrzewski (2002), cancer is known to be a common name for about 200 diseases characterized by abnormal cell growth. Cancer cases that can be attributed to environmental causes probably account for more than 60% of all cancers. The environment, in this case, involves not only air, water, and soil, but also food, drink, living habits, occupational exposure and drugs. It involves also all aspects of human interaction with the surroundings. As a consequence of their harmful effects on human health and living organisms, heavy metals concentrations in drinking water and wastewater are restricted by strict standards (Weiner, 2000).



**Figure 2.1: Sources and transport of heavy metal pollutants (Wright 2003; O'Neill, 1993).**

### **Cadmium**

Cadmium occurs naturally in the earth's crust. In nature, cadmium is most often present in zinc, lead, and copper ores and can be extracted as a by-product during the production of these metals (Weiner, 2000). The chlorides and sulfates

are the forms that most easily dissolve in water, but the oxide and sulfide are the forms that are relatively insoluble. Soluble cadmium compounds can be leached through soils to groundwater (Weiner, 2000; O'Neill, 1993).

Cadmium can bioaccumulate in fish, plants, animals and humans. Most of the cadmium in bodies of water, such as lakes or rivers, settles on the bottom. However, a small amount may remain either dissolved in water or as fine suspended particles. Some fish can accumulate cadmium in their bodies if they live in water polluted by cadmium. Cadmium may be taken up by animals that drink water or eat fish contaminated with cadmium. If other animals or humans eat these animals, they will also have increased amounts of cadmium in their bodies by bioaccumulation (Manahan, 1994; Harrison et al., 1996).

Itai-itai disease is one of the well-known diseases in Japan that is caused by the cadmium pollution. It was observed for the first time at the basin of the Ginzu river in Toyama prefecture in 1955 (Fergusson,1990). Itai-itai disease characterized by renal tubular dysfunctions and extreme pain (Hiyama et al., 2002; Fergusson,1990).

High doses of cadmium are known to be very toxic. A lethal ingestion dose is about 1 g. Acute exposure to cadmium can cause nausea and vomiting. It can also cause muscle cramps and diarrhea. Other potential short-term effects are sensory disturbances, liver injury, and salivation. Shock, renal failure, and



convulsions can result from short-term exposure to cadmium. Cadmium can bioaccumulate over many years of low exposure. Long-term exposure to cadmium can cause lung damage. It can also cause blood damage. Permanent liver damage can result from long-term exposure to cadmium as well. Other potential long-term effects are fragile bones (Weiner, 2000; Garcia, 2001). Urine test can be used to determine whether a human body contains high level of cadmium or not. Samples of blood can be collected and the level of cadmium can be measured (Paasivirta, 1991). Furthermore, it was reported that there is no adequate evidence to state whether or not cadmium has the potential to cause cancer from lifetime exposures in drinking water (Weiner, 2000). The US EPA drinking water standard for cadmium is 0.005 mg/L (Weiner, 2000) which is similar to the Quebec standard (Quebec environmental Quality Act, 2005).

## **Zinc**

In nature, the average zinc concentration in the earth's crust is estimated to be around 70 mg/kg (Weiner, 2000). Zinc is very chemically reactive. For this reason, it is not found free in nature but is generally associated with one of the known zinc minerals (mainly sulfides, oxides, carbonates, and silicates). It is reported that about ninety percent of industrial zinc production is from the minerals sphalerite (also called zincblende) and wurtzite (Vanloon and Duffy 2000; Weiner, 2000). Zinc can be found in natural waters in both dissolved and suspended forms. The dissolved form is the divalent cation,  $Zn^{2+}$ . In water of low

alkalinity and below pH 7,  $Zn^{2+}$  is the major form. Dissolved zinc can be rapidly sorbed to mineral clays (Weiner, 2000).

In trace amounts, zinc is an essential element needed by the human body because it plays an important role in metabolism of proteins and nucleic acids. It is a component of metalloenzymes and is considered as an activator of enzymes as well (Harrison et al., 1996). Too much zinc can be harmful to human health. Chronic exposures to zinc compounds (e.g. under occupational exposure conditions ) can cause “metal fume fever”. “Metal fume fever” is characterized by fever, nasal passage irritation, cough, headache, weakness, and reduced lung volume. Workers in zinc mines or processing facilities, usually breathe high levels of dust that contains zinc (Weiner, 2000; Garcia, 2001). A medical test can be used to determine whether a human body contains high level of zinc or not. Samples of blood can be collected and the level of zinc can be measured. Zinc is not known to cause cancer in humans (Paasivirta, 1991).

Without harmful effects, about 1 g/day of zinc may be ingested. Recommended dietary allowance is 15 mg/day for adults. The EPA has no primary drinking water standard for zinc. The EPA secondary drinking water standard (nonenforceable) is 5 mg/L (Weiner, 2000). The Canadian drinking water standard for zinc is 5.0 mg/L (Health Canada, 2006).

## Nickel

In nature, nickel is most often present in silicates, sulfides, arsenides, antimonides, and oxides. Its average concentration in the earth's crust is estimated to be around 75 mg/kg (Weiner, 2000). Nickel minerals tend to be associated with minerals of other metals (Vanloon and Duffy, 2000; Weiner, 2000).

Nickel is considered one of the most mobile heavy metals in the aquatic environment (Weiner, 2000). Its concentration in unpolluted water is controlled largely by co-precipitation and sorption with hydrous oxides of iron and manganese (Manahan, 1994; Weiner, 2000). Nickel enters waters with surface runoff or by percolation of dissolved nickel into groundwater (Fergusson, 1990; Harrison et al., 1996).

Nickel is considered as an essential trace nutrient in animals and humans. Daily intake of nickel, mainly from food, is around 150 µg/day; the adult requirement is between 5 and 50 µg/day. Although a few nickel compounds, e.g. nickel carbonyl, are known to be poisonous and carcinogenic, most nickel compounds are nontoxic. The toxicity of dissolved nickel that is ingested orally is low, compared to zinc, chromium, and manganese, perhaps because only 2-3% of ingested nickel is absorbed (Weiner, 2000). Currently in the U.S (Weiner, 2000) and Canada (Health Canada, 2006), there are no drinking water standards for nickel.

## **Copper**

Copper is an element known and used for a long time (from the bronze age) (Harrison et al., 1996). In some cases, copper occurs naturally as the pure metal. Usually, it occurs in the form of mineral ores that contain 2% or less of the metal. Copper-bearing ores like arsenites, sulfides, carbonates, and chlorides are the most abundant ones (Katz et al., 2005; Weiner, 2000).

Smelting operations and municipal incineration are considered sources of introduction of copper into the environment. Copper concentrations in acid mine drainage can reach several hundred mg/L, but, if the pH is raised to 7 or higher, most of the copper will precipitate (Kirjavainen et al., 2005; Weiner, 2000).

Copper occurs in drinking water mainly as a result of corrosion of copper pipes and fittings that are commonly used for interior plumbing in buildings because it is less toxic than lead. Consequently, the US EPA set an Action Level based on samples taken from distribution system taps. In the USA, the maximum contaminant level goal of copper is 1.3 mg/L (Weiner, 2000; O'Neill, 1993). The Canadian drinking water standard for copper is 1.0 mg/L (Health Canada, 2006).

Copper is known to be an essential element to life because it is a structural component of enzymes that catalyses oxidation reactions (Harrison et al., 1996). At high doses, it has been shown to cause stomach and intestinal distress, liver

and kidney damage, and anemia. Persons with Wilson's disease may be at a higher risk of health problems due to hyper-accumulation of copper than healthy individuals. There is inadequate evidence to state whether or not copper has the potential to cause cancer (Weiner, 2000; Garcia, 2001).

## **Lead**

Lead occurs naturally in the earth's crust. Lead oxides, carbonate, and sulfate minerals can be found naturally as well (Weiner, 2000). The main ore of lead is galena,  $PbS$ , from which lead can be obtained by roasting it with air in a furnace. Lead produced from the roasting process is called (pig lead) (Wright, 2003). Commercial ores have concentrations of lead in the range of 30-80 g/kg (Weiner, 2000).

In general, lead is present in the environment in different forms. Lead hydroxide, lead carbonate and lead sulfate are some common examples of insoluble lead compounds (Kalia et al., 2005; Fergusson, 1990). Insoluble lead compounds also include lead sulfides and lead oxides (Altorfer et al., 2003; Weiner, 2000).

Concentrations of dissolved lead in natural surface waters are generally low. Lead hydroxide, and lead carbonate are the most common lead forms that can occur in rivers, lakes and groundwater. (Leroy, 2004; Weiner, 2000).

Lead can bioaccumulate in fish, plants, animals and humans. In air, lead is present mostly as fine dust particles. High doses of lead are known to be very toxic. In the US, it was reported in 1988 that between 5.9 and 11.7 million children under the age of seven were at risk from lead-based paint. Other children were potentially exposed to lead in dust and soil (ATSDR, 1988). It has been shown that lead competes with iron in the intestine causing iron deficiency and anemia (Altorfer et al., 2003; Fergusson, 1990). Lead can accumulate in the bones, teeth, muscle and skin. Lead compounds are very hazardous. Acute exposure can cause paralysis of the limbs, neurological disorders, coma and even death (Altorfer et al., 2003; ATSDR, 1993). A report by American Association of Poison Control Centers cited more than 2,000 cases of lead poisoning (ATSDR, 1988). Tetraethyl lead and tetramethyl lead are known to be very hazardous. They can cause personality and behavioral problems, and also can cause mental illness (Wright, 2003). It has been reported that lead can inhibit many enzymes, such as  $\delta$ -aminolevulinic acid dehydrase (Leroy, 2004; Fergusson, 1990).

In the case of chronic exposure, lead can cause neurological disorders, increased blood pressure and kidney malfunction. In children, lead has caused growth problems (ATSDR, 1993). An increase in the erythrocyte protoporphyrin in blood is apparent in blood lead around 12-15  $\mu\text{g/l}$  (Kalia et al., 2005; Fergusson, 1990).

A medical test can be used to determine whether a human body contains high level of lead or not. Samples of blood can be collected and the level of lead can be measured. (Paasivirta, 1991). Lead (II) acetate and lead (II) phosphate have been identified as carcinogens as a result of animal experiments. The EPA classifies lead as a carcinogen (Kalia et al., 2005; Wright, 2003). The Canadian drinking water standard for lead 0.010 mg/L (Health Canada, 2006). Regarding US EPA drinking water standards lead, maximum contaminant level is 0.015 mg/L (Weiner, 2000).

### **Water treatment processes**

Several methods of wastewater and groundwater treatment are presently available. However, with the growth in the industries like computer chips and petroleum cracking, new problems have appeared that require innovative methods of treatment, or in some cases, the optimization of the existing processes to reduce or eliminate waste. In order to remove large particles and/or suspended solids from real industrial wastewaters like wastewater from metal refining plants, several methods can be utilized including sedimentation, filtration, pH adjustment, coagulation, adsorption, ion exchange, etc (Dotremont et al., 2004; Reynolds and Richards, 1996). Processes involving membrane separations are being utilized to remove small particles and/or dissolved solids for the purpose of waste treatment (Reynolds and Richards, 1996; Huang et al., 2005).

Removal of heavy metals from large volumes of wastewaters or groundwaters at low concentrations by traditional methods like pH adjustment and chemical precipitation are not economical. This is because of the large volumes of dilute polluted waters that need to be handled (Huang et al., 2005; Reynolds and Richards, 1996) In these cases, removal of heavy metal ions is more effective by ion-exchange resins or adsorption onto solid surfaces. Adsorption can be widely used in the biological treatment of organic industrial wastewater to adsorb organic compounds. One major limitation of adsorption method is the cost of the adsorbent used (Mohammad et al., 2004; Reynolds and Richards, 1996).

Ion exchange technology consists of a chemical reaction between ions in a liquid phase and ions in a solid phase. Certain ions in the solution are preferentially sorbed by the ion exchanger solid. Because electroneutrality must be maintained, the exchanger solid releases replacement ions into the solution (Reynolds and Richards, 1996). Some of the well known applications of this approach to water treatment are water softening (Ameskamp et al., 1999) and water demineralization (Shaposhnik et al., 2001) .

Water softening can be performed by exchange of sodium ions for calcium and magnesium ions (Ameskamp et al., 1999). In demineralization of water, the cationic resins are charged with the hydrogen ion and the anionic resins are charged with the hydroxyl ion. The cationic resins exchange hydrogen ions for cations, and the anionic resins exchange hydroxyl ions for anions (Shaposhnik et



al., 2001). Industries that utilize high-pressure boilers require demineralized water as boiler water (Michael, 2004).

One disadvantage of using an ion exchange system is that when scaling it up, the larger system will probably not achieve the same operating exchange capacity as a smaller laboratory scale (Hedstrom, 2001). In addition, if the regeneration consists of a combination of chemical and biological regeneration, the system will be even more complicated because both steps need to be optimized (Hedstrom, 2001; Curkovic, 1997). Also, one of the major problems of using ion exchange is that some large scale runs entailed a relatively high loss of time- and material-consuming regeneration processes for the exchanger (Ameskamp et al., 1999). When applying the ion exchange technique, the cost of adsorbent and regeneration should be taken into account (Reynolds and Richards, 1996; Hedstrom, 2001).

## **2.2 Membrane Separation Processes**

### **2.2.1 General Process Description**

Membrane separation has become an attractive technique in the removal of pollutants from wastewaters (Huang et al., 2005). Membrane separation processes are technologies used to separate dissolved substances and fine particles from water and wastewater (Reddy et al., 2006; Cardew and Le, 1998).

Different materials like polymers, ceramics and metals can be utilized to make membranes (Tansel et al., 2005; Cardew and Le, 1998). Table 2.1 summarizes the various materials that are used to manufacture membranes. In general, ultrafiltration membranes can be made from cellulose or non-cellulosic materials (synthetic polymers). Cellulosic membranes can be produced in different forms such as cellulose acetate or modified cellulose acetate. Non-cellulosic materials (synthetic polymers) can be synthesized in different forms as well like polysulfone or acrylates (Dotremont et al., 2004; Mallevalle, 1996). Different processes including casting, reactions, stretching, solvent phase inversion and coating are used to manufacture membranes. Table 2.2 summarizes the various processes that are used.

Membrane technologies are characterized in different ways. Membrane filtration processes are characterized in terms of pore size (Table 2.4) and molecular weight cut-off (MWCO) (Table 2.3). Size, charge and affinity are well known separation principles utilized in membrane separation processes (Table 2.4). Table 2.5 shows the relationship between thermodynamic driving forces and fluxes. The driving force for membrane separation comes from a pressure gradient. Commercial separation processes are mainly controlled by the differences in one or more factors like pressure, concentration, electric potential or temperature (Table 2.5).

### 2.2.2 Membrane Operations

Membrane filtration is a separation technology used to separate dissolved substances and fine particles from water and wastewater. These substances and particles may range in size from bacteria size to atom size (Reddy et al., 2006; Cardew and Le, 1998). A thin semipermeable membrane acts as a selective barrier that separates particles based on molecular or physical size (Mulder, 1991). In general, water and constituents which are smaller than the membrane pore size are able to pass through the membrane as "permeate" while particles and molecules larger than the membrane pore size are retained. The retained particles and molecules, known as "retentate" are returned to the feed tank to be concentrated further with time. "Permeate flux" is the term used to describe how fast permeate passes through a membrane. Flux is generally reported as volume/area-time. Permeate flux generally depends upon the individual membrane characteristics (e.g., membrane pore size and membrane surface charge), the characteristics of the feed stream (e.g., viscosity and solute particle size) and operating parameters (e.g., transmembrane pressure and feed temperature) (Reddy et al., 2006; Cardew and Le, 1998).

The utilization of membrane separation processes in a variety of industries is growing at a significant rate. Membranes can be used in production of high purity water for different uses, purification of coal gasification liquors, sludge

dewatering, landfill leachate treatment, food industry, purification of water, de-alcoholization of beer, water desalination, pervaporation (separation of water from organic solvents and solvent mixture), hazardous chemical removal from gas and liquid feed-streams, and nitrogen production for petroleum and gas facilities (Turner, 1991).

One of the attractive features of membranes is that the separation can be done under ambient conditions (Tansel et al., 2005; Cardew and Le, 1998).

### 2.2.3 Membrane Selection

The pore size of the membrane controls the size of the molecules that can pass through. In many cases membranes may become fouled by the some components of the feed solution.

**Table 2.1 Main processes that are used to manufacture membranes (adapted from Mallevalle, 1996; Cardew and Le, 1998).**

<b>Structure</b>	<b>Structure Type</b>	<b>Membrane Process</b>	<b>Manufacturing Process</b>
Dense	Homogenous	ED	Cast
Dense	Composite	ED	Reaction
Microporous	Homogenous	MF, UF	Stretching
Microporous	Asymmetric	MF, UF, RO	Solvent Phase Inversion
Microporous	Composite	RO	Coating

**Table 2.2 Rejected species and MWCO of membrane filtration processes**

(adapted from Cardew and Le, 1998; Reddy et al., 2006).

<b>Technology</b>	<b>MWCO</b>	<b>Material</b>	<b>Rejected Species</b>
Reverse osmosis	<200	Polymers	Salts, Sugars
Nanofiltration	200 - 1,000	Polymers	Sugar Pesticides
Ultrafiltration	> 1,000	Polymers, Ceramics,	Proteins, Viruses
Microfiltration	-	Polymers, Ceramics, Metals	Bacteria, Silts, Cysts, Spores

**Table 2.3 Separation principles and pore size ranges of membrane filtration**

processes (adapted from Cardew and Le, 1998; Dotremont et al., 2004).

<b>Technology</b>	<b>Separation Principle</b>	<b>Pore Size Range</b>
Reverse osmosis	Size, Charge, affinity	Less than 1 nm
Nanofiltration	Size, Charge, Affinity	About 1 nm
Ultrafiltration	Size, Charge	1 nm – 100 nm
Microfiltration	Size	0.1 $\mu\text{m}$ - 1 $\mu\text{m}$

**Table 2.4 List of main membrane technologies driving forces and fluxes used (Adapted from Mallevalle, 1996; Tansel et al., 2005).**

<b>Technology</b>	<b>Driving Force</b>	<b>Flows</b>
Reverse osmosis	Pressure	Volume flux
Nanofiltration	Pressure	Volume flux
Ultrafiltration	Pressure	Volume flux
Microfiltration	Pressure	Volume flux
Dialysis	Concentration	Solute flux
Thermodialysis	Temperature	Solute flux
Electrodialysis	Electric Potential	Solute flux

The chemical composition of the membrane material is significant because of possible interactions with the feed solution (Tansel et al., 2005; Mallevalle, 1996). As a consequent, the selection of the membrane is important for an efficient ultrafiltration process. Membranes are characterized by the diameter of a particle or molecular weight of a molecule that is retained (known as the molecular weight cutoff (MWCO) by the membrane (Dotremont et al., 2004; Mallevalle, 1996). MWCO and the membrane material affect the flux, the concentration of surfactants and heavy metals in the feed that passes through

the membrane (Reddy et al., 2006; Mallevalle, 1996). Selectivity comes through the interaction between the membrane and the surrounding phases (Mohammad et al., 2004; Cardew and Le, 1998).

Most membrane separation systems used in industrial applications are operated in a cross-flow feed configuration mode. In the cross-flow configuration, the concentrate passes parallel to the membrane surface. Membrane separation processes are gaining acceptance as alternatives to other treatment technologies (Dotremont et al., 2004; Cardew and Le, 1998).

## **2.3 General Types of Membrane Separation Processes**

Membrane processes can be differentiated by the size of particles they separate. The four principle membrane separation techniques used to treat wastewater and ground waters are: reverse osmosis (RO), nanofiltration (NF), ultrafiltration (UF), and microfiltration (MF).

### **Reverse osmosis (RO)**

Reverse osmosis (RO) is a membrane separation technique used to remove salts and ions from solutions. In RO, the pressure on the salt solution is raised above the osmotic pressure so the net movement of water is from the more concentrated saline side to the pure water side of the semi-permeable membrane. Reverse osmosis has been widely used in desalination of seawater,

and throughout the world (Mohammad et al., 2004; Mallevalle, 1996).

### **Nanofiltration (NF)**

Nanofiltration (NF) systems operate under the same basic principle as reverse osmosis. However, nanofiltration is generally used to remove particles in the range from 200 to 1,000 MWCO). Nanofiltration operates on the same basic principle as RO but is applied to larger particle sizes. Smaller constituents of a waste or process stream (e.g., dissolved salts) which can be removed using RO may pass through a nanofiltration membrane (Reddy et al., 2006; Mallevalle, 1996). A typical pore size range of NF membranes is about 1.0 nm. The types of contaminants rejected by NF membranes are sugars and pesticides (Dotremont et al., 2004; Cardew and Le, 1998).

### **Ultrafiltration and Microfiltration**

Ultrafiltration (UF) and microfiltration (MF) are pressure-driven membrane techniques based on a "sieving mechanism" in which particles are separated from solution based on size (Tansel et al., 2005; Mallevalle, 1996). The main distinction between ultrafiltration (UF) and microfiltration (MF) is the larger membrane pore size used in MF. Typical pore size range of MF membranes is from 0.1  $\mu\text{m}$  to 1.0  $\mu\text{m}$  while UF membranes have pore sizes from 1.0 nm to 100 nm. The types of contaminants rejected by MF membranes are large particles, bacteria, cysts and spores (Cardew and Le, 1998).



The main advantages of low-pressure UF membrane processes when they are compared with conventional clarification (direct filtration, settling/rapid sand filtration, or coagulation / sedimentation / filtration) and disinfection (postchlorination) processes are (1) no need for chemicals (coagulants, flocculants, disinfectants, pH adjustment) (2) size-exclusion filtration as opposed to media depth filtration, (3) good and constant quality of the treated water in terms of particle and microbial removal, regardless of the raw feedwater quality, (4) process and plant compactness, (5) simple automation (Reddy et al., 2006; Mallevalle, 1996).

Some of the basic differences that distinguish membrane separation from granular bed filtration are: (1) the size of the openings or pores in the membrane surface, (2) that separation takes place at the surface of the membrane, and (3) that feed flow in membrane filtration is across the face of the membrane compared with normal (dead-end) flow with granular bed filtration (Tansel et al., 2005; Mallevalle, 1996).

## **2.4 Conventional Ultrafiltration Modules**

The most common module configurations used in cross-flow ultrafiltration are tubular, hollow fiber, spiral wound, and plate and frame (Mallevalle, 1996).

### **Tubular Membrane Module**

Tubular membrane modules have the largest dimensions of all membrane

devices. A tubular membrane consists of a porous membrane material cast on a permeable support matrix housed inside a rigid hollow tube. Permeate is forced through the membrane under pressure and is collected through a port in the rigid outer casing. Tubular membranes are available in single or multi-tube configurations. Typical commercial tubular membranes can be connected in series when lower circulation velocities (1 m/s) are used. Due to large channel sizes, tubular membranes are commonly used to treat waste and process streams with high solids concentrations (Tansel et al., 2005; Mallevalle, 1996).

### **Hollow Fiber Membrane Modules**

Hollow fiber membrane module consists of a bundle of fine fibers made of the membrane material bound at each end of the module by an epoxy resin plug. The whole element is encased in a PVC tube. Unlike tubular membranes, hollow fiber membranes do not have an underlying support structure. Feed is forced into the membrane module under pressure through a perforated tube. The perforated tube is located in the center of the housing. Additionally, hollow fiber membrane modules have a higher packing density than tubular modules but are more subject to fouling effects than tubular systems (Reddy et al., 2006; Cardew and Le, 1998).

**Table 2.5 Advantages and disadvantages of UF membrane modules**

(Tansel et al., 2005; Cardew and Le, 1998).

MODULE	ADVANTAGES	DISADVANTAGES	TECHNOLOGY	Supplier
Plate-and-frame	<ul style="list-style-type: none"> <li>. Wide choice of membranes</li> <li>. Low energy requirement</li> </ul>	<ul style="list-style-type: none"> <li>. High cost</li> <li>. Replacing membrane is time Consuming</li> </ul>	MF,UF, RO	(1); (3)
Hollow-Fibre	<ul style="list-style-type: none"> <li>. Very compact system</li> <li>. Low capital cost</li> <li>. Backflushable</li> </ul>	<ul style="list-style-type: none"> <li>. Easily fouled</li> <li>. Not suitable for viscous systems</li> <li>. Limited range of products available</li> </ul>	MF,UF, NF, RO	(1); (2)
Spiral Wound	<ul style="list-style-type: none"> <li>. Compact system</li> <li>. Wide range of sizes</li> <li>. Low capital cost</li> </ul>	<ul style="list-style-type: none"> <li>. Can have dead spots</li> <li>. Cannot be backflushed</li> </ul>	UF, NF, RO	(1); (2) (3);
Tubular	<ul style="list-style-type: none"> <li>. Can tolerate feeds with high suspended solids</li> <li>. Easy to clean mechanically</li> </ul>	<ul style="list-style-type: none"> <li>. High energy requirement</li> <li>. High capital cost</li> <li>. Large space demand</li> <li>. High hold-up</li> </ul>	MF,UF, NF, RO	(1); (3)

(1) Koch Membranes

(2) Millipore

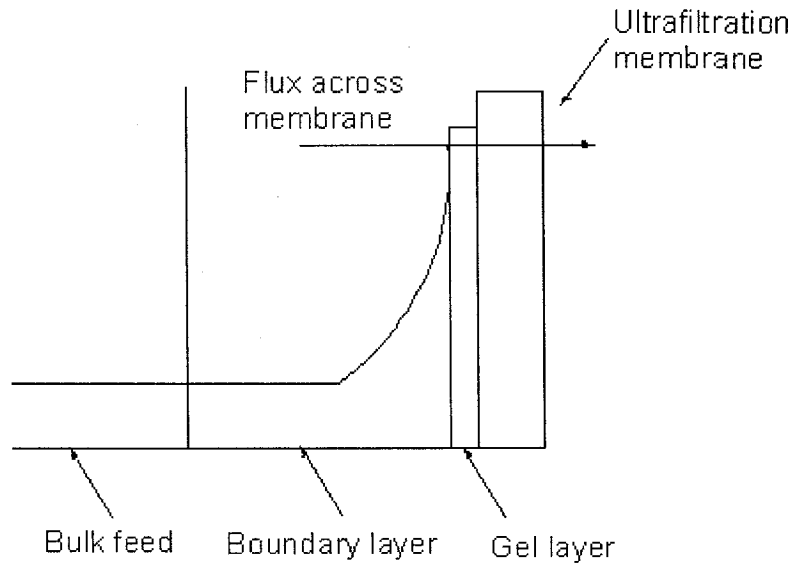
(3) Culligan Membranes

### **Spiral Wound Membrane Modules**

A spiral wound membrane module consists of alternating layers of porous membranes wrapped around a central hollow tube in a prescribed way into a Swiss roll configuration. The feed solution flows axially through the wound membrane module. The permeate is forced through the porous membrane and is collected through the hollow center tube while rejected feed solution exits at the end of the module. Spiral wound membranes foul more easily than tubular membranes due to smaller waste flows. This factor, coupled with the fact that spiral wound modules cannot be mechanically cleaned limits the application of these membrane configurations to waste and process streams with low solids concentrations (Dotremont et al., 2004; Mallevalle, 1996).

### **Plate-and-Frame Membrane Modules**

In these systems, flat-sheet membranes are utilized in plate-and-frame modules. The flat-sheet membranes are used to direct the feed along the module stacks (Reddy et al., 2006; Mallevalle, 1996). A stack of multiple membranes separated by spacers is placed in series or in parallel inside a cylindrical housing. Permeate is forced through the membrane under pressure and is collected at the outer edges of the membrane disc plate. Concentrated solute is collected at the top of the stack (Tansel et al., 2005; Cardew and Le, 1998). Table 2.6 shows the advantages and disadvantages of UF membrane modules.

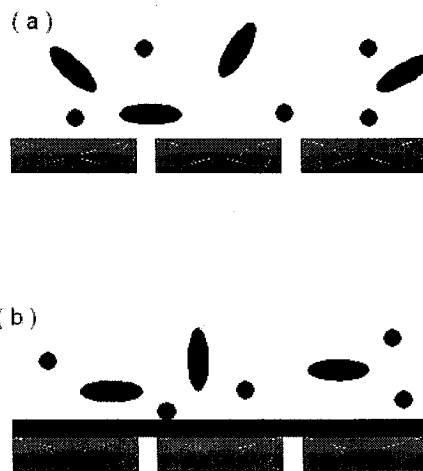


**Figure 2.2 Schematic of boundary layer and gel layer formation in ultrafiltration (Bouchard et al., 2003; Cardew and Le, 1998).**

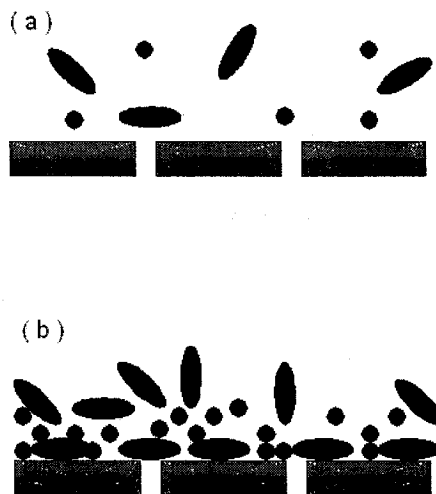
## **2.5 Factors Controlling Permeate Flux**

### **2.5.1 Concentration Polarization**

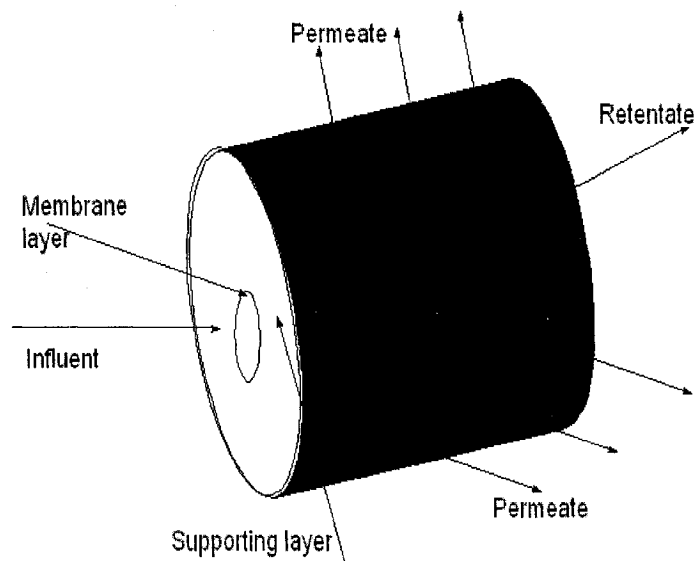
Polarization is the phenomenon that is of major significance in cross-flow membranes. As the productivity of the membrane increases, an inhomogeneous layer develops on the input side of the membrane (Choo et al., 2005; Cardew and Le, 1998). Figure 2.2 illustrates a schematic of boundary layer and gel layer formation in an ultrafiltration module. If the accumulated solute layer is sufficiently thick, a decrease in permeate flow rate will occur (Mallevalle, 1996).



**Figure 2.3 Schematic of concentration polarization in ultrafiltration, (a) normal- no effect, (b) materials at interface transform and form a secondary membrane (gel polarization) (Te Poele et al., 2004; Cardew and Le, 1998).**



**Figure 2.4 Schematic of fouling formation in ultrafiltration a) normal- no effect, (b) fouling increases the deposits on the ultrafiltration membrane (Te Poele, et al., 2004;Cardew and Le, 1998).**



**Figure 2.5 Schematic of an ultrafiltration module (Dotremont et al., 2004; Cardew and Le, 1998)**

Under conditions of low transmembrane pressure, low feed concentration, three competing solute transport mechanisms occur in ultrafiltration: (1) advective transport of solute to the membrane surface (2) back diffusion of accumulated solute to the bulk solution, and (3) solute permeation through the membrane (Te Poele et al., 2004; Mallevalle, 1996).

A schematic of concentration polarization in ultrafiltration is illustrated in Figure 2.3. As one descends down the particle size range, the sharp divide between a deposit and solution disappears and is replaced by concentration profile. This is

known as “concentration polarization” (Bouchard et al., 2003; Cardew and Le, 1998 ).

### **2.5.2 Membrane Fouling**

Fouling is considered the major difficulty encountered in the operation of membrane technology for water treatment. The term *fouling* includes the totality of phenomena responsible for decrease of permeate flux over a period of time. Consequently, these phenomena are dynamic and depend on such parameters as physicochemical properties of the membrane, characteristics of the fluid to be filtered, and hydraulic operating conditions of the system (Te Poele et al., 2004; Cardew and Le, 1998). Fouling increases the deposits on the ultrafiltration membrane. Figure 2.4 shows a schematic of fouling formation in ultrafiltration. The fouling of membranes will cause an increase in the feed pressure to maintain a constant output. Consequently, it will increase operating costs due to the increase in the energy spending. Fouling reduces the lifetime of membranes as well. Then, operating costs will increase. Thus, a good control of the deposits on the membrane will limit operating costs (Mohammadi et al., 2005; Mallevalle, 1996). Four main categories of fouling can be distinguished: biological, organic, colloidal and mineral (Plottu, 2003; Kimura et al., 2004). Figure 2.5 shows a schematic of an ultrafiltration module. The autopsy of membranes is a destructive analytical method but remains an excellent tool to characterize very precisely the nature of deposits (Plottu, 2003; (Mohammadi et al., 2005; Mallevalle, 1996).



Reversibility of fouling is therefore one of the important points to be resolved. Several techniques are available that make it possible to control fouling (Wright, 2001). These techniques include pretreatments, chemical washing, and operating pressure (Kimura et al., 2004; Mallevalle, 1996).

## **2.6 Surfactants**

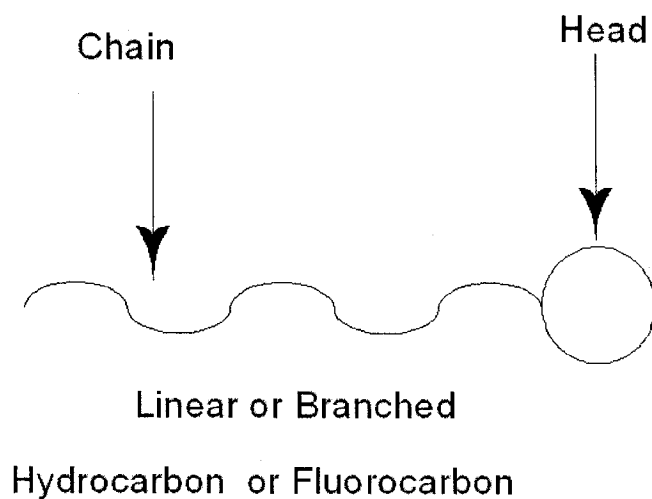
Amphiphiles or surface-active agents (surfactants), are molecules that have both hydrophilic (water-loving head) and hydrophobic (water-hating tail) portions. 'Surfactants' is an abbreviation for surface-active agents that literally means active at the surface (Tadros, 1984). The hydrophobic portion is generally characterized as a hydrocarbon or fluorocarbon chain that is relatively insoluble in water. The hydrophilic portion is relatively water soluble. Shown in Figure 2.6 is a schematic of a surfactant (SDS), and the associated hydrophobic and hydrophilic regions (Lang, 1999).

In the solution, surfactant molecules exist as individual and unassociated monomers below the critical micelle concentration (CMC). The first-formed aggregates are generally approximately spherical in shape. Such aggregates are called the critical micelle concentration (CMC). The critical micelle concentration (CMC) is known to be the minimum surfactant concentration at which surfactant monomers form micelles. Above the CMC, any additional surfactant added to the solution forms aggregates called micelles (Holmberg et al., 2003). Figure 2.7 shows an illustration of a spherical micelle composed of anionic surfactant in an

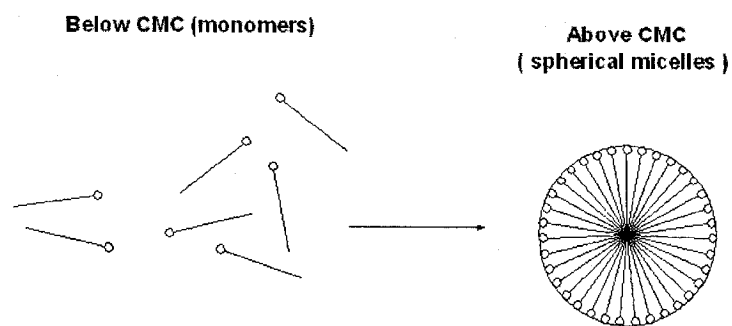
aqueous solution. The hydrophobic tails are oriented toward the center of the micelle and the head groups are oriented outward in contact with the polar solvent. The interior of the micelle is a hydrophobic core of intertwined tail groups (Lang, 1999).

Surfactants have been used industrially as adhesives, flocculating, wetting and foaming agents, de-emulsifiers and penetrants (Mulligan and Gibbs, 1993). In enhanced oil removal applications surfactants increase the solubility of petroleum components (Ishikawa et al., 2002). In addition, surfactants have been used extensively in the steel industry, textile industry, and agricultural industry. Furthermore, surfactants have many pharmaceutical applications (Lang, 1999). They have also been used for mineral flotation and in the pharmaceutical industries (Kujawinski et al., 2002 ).

Typical useful properties include critical micelle concentrations, wetting characteristics, foaming characteristics, pH stability, solubility enhancement, surface tension reduction (Tadros, 1984, Holmberg et al., 2003). Depending on the nature of the polar head-group, surfactants are classified as cationic, anionic, zwitterionic and nonionic (Figure 2.8).



**Figure 2.6 Schematic diagram of surface active molecule (a monomer).**  
(Adapted from Tadros, 1984; Lang, 1999).



**Figure 2.7 Schematic diagram of a spherical micelle (Lang, 1999).**

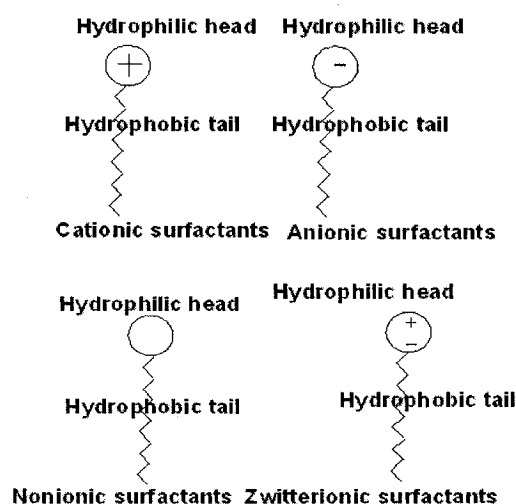
- Anionic: the surface-active portion of the molecule bears a negative charge.
- Cationic : the surface-active portion bears a positive charge.
- Zwitterionic : both positive and negative charges may be present in the surface-active portion.
- Nonionic: the surface-active portion bears no apparent ionic charge (Tadros, 1984) .

## **2.7 Biosurfactants**

Biosurfactants are natural, biodegradable, complex surface-active agents (surfactants) that are mainly produced by microorganisms (Banat, 2001). Like synthetic surfactants, they have a two-portion structure of hydrophilic and hydrophobic portions (Tsujii, 1998). Their compositions and yields depend on the fermentor design, pH, nutrient composition, substrate and temperature used (Mulligan and Gibbs, 1993).

### **2.7.1 Advantages of Biosurfactants**

Biosurfactants have several advantages over synthetic surfactants. Biosurfactants are more biodegradable and have a very low level of toxicity. Thus, they have been considered as a substitution for synthetic surfactants. Biosurfactants can find interesting applications in a number of industries including the petroleum industry, agriculture industry, food industry, textile industry, and paint industry (Kosaric and Cairns, 1987).



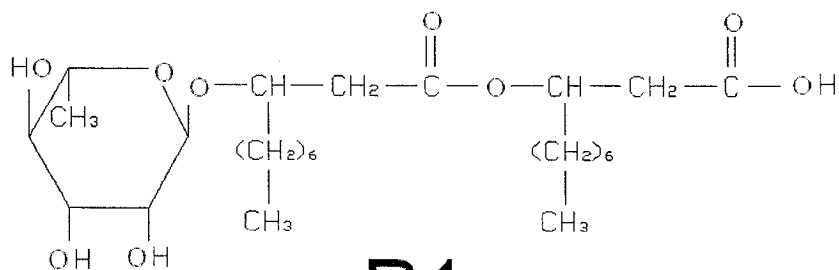
**Figure 2.8 Schematic diagrams of anionic, cationic, zwitterionic and nonionic surfactants (Lang, 1999).**

### 2.7.2 Types of biosurfactants

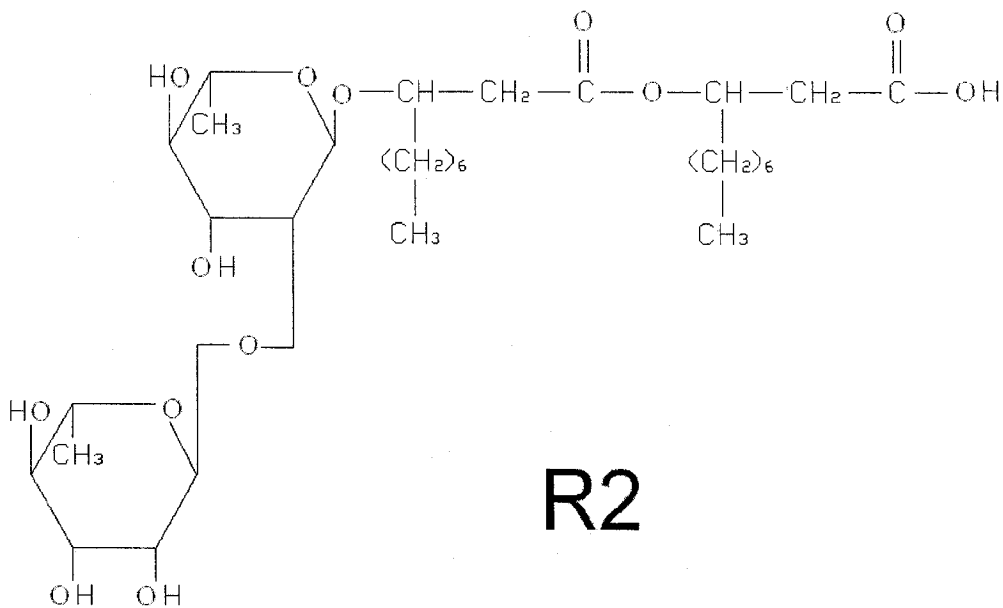
Biosurfactants include phospholipids, glycolipids, proteins and peptide derivatives, and other surface active natural substances like cholesterol, rosin, and saponin. The majority of biosurfactants are anionic. Rhamnolipids are biosurfactants from the glycolipid group made by *Pseudomonas aeruginosa* (Tsuji, 1998).

### 2.7.3 Synthesis of biosurfactants

Biosurfactant production has been observed in many different types of microorganisms, including bacteria (Yoon et al., 2003), yeast (Nielsen et al., 2003), and fungi (Paraszkiewicz et al., 2002).

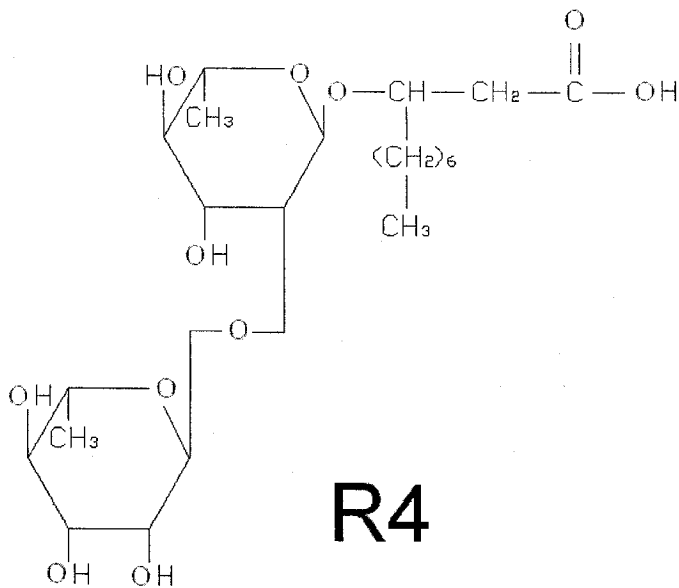
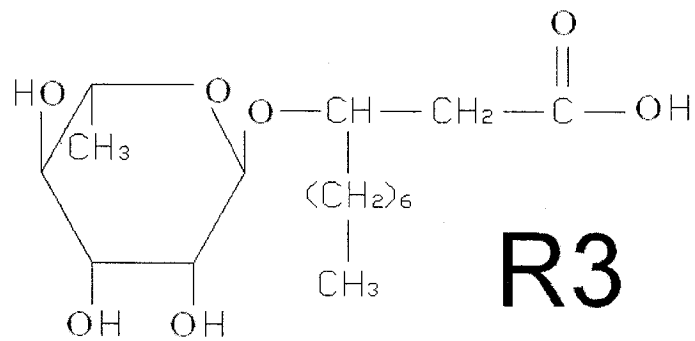


R1

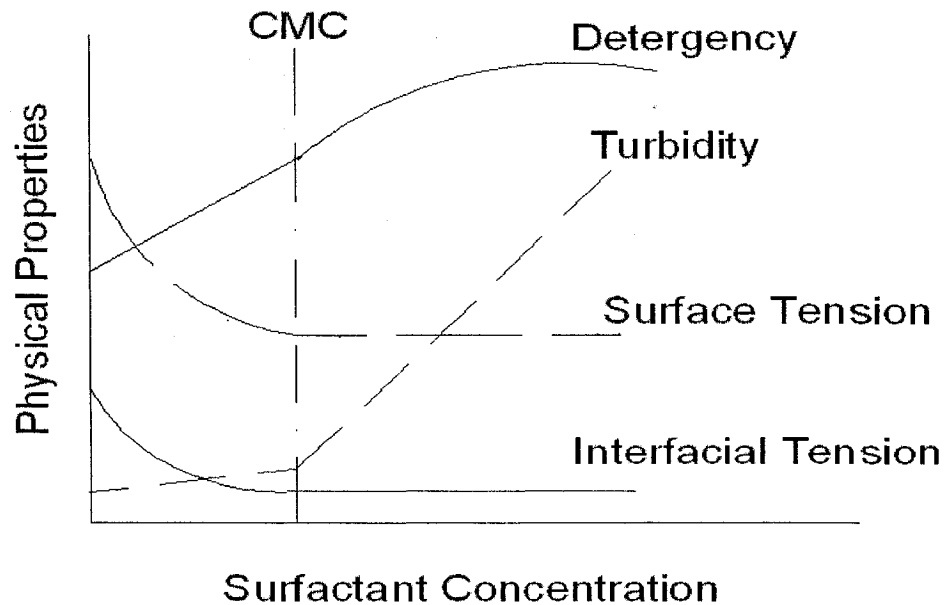


R2

**Figure 2.9 Structures of R1 and R2 rhamnolipids (adapted from Tsujii, 1998).**



**Figure 2.10 Structures of R3 and R4 rhamnolipids (adapted from Tsujii, 1998).**



**Figure 2.11** Variation of major physical properties as a function biosurfactant concentration (adapted from Myers, 1992).

Yoon et al. (2003) reported that biosurfactants can be produced by bacteria like *Pseudomonas fluorescens* PD101. Nielsen et al. (2003) investigated biosurfactant production by yeast like *Pseudomonas fluorescens* strains.

## **Biosurfactant production from low-cost sources**

Biosurfactant production has been observed in many different types of relatively inexpensive materials, including an oil-contaminated site (Lu et al., 2003), n-



alkanes (Philp et al., 2002; Yakimov et al., 1999; Kakugawa et al., 2002), oil-contaminated water (Banat et al. (2002), starch wastewater and starch wastewater sludge (Barnabe et al., 2003). The effect of biosurfactant concentration on variation of major physical properties is shown in Figure 2.11 (Myers, 1992).

## **2.8 Enhanced ultrafiltration**

The combination of surfactants and membranes in wastewater separations has led to the development of novel techniques or enhancement of existing techniques. Some examples of methods that apply surfactants are micellar-enhanced ultrafiltration (MEUF) methods (Huang et al., 2005; Kim et al., 2004). The technique of ultrafiltration and the measurement of octanol-water partitioning and  $\xi$ -potential were utilized to determine the mechanism of metal removal by surfactin. It was reported that surfactin was able to remove the metals by sorption at the soil interphase and metal complexation, followed by desorption of the metal through interfacial tension lowering and fluid forces and finally complexation of the metal with the micelles (Mulligan et al., 1999). Ultrafiltration was utilized in to purify and concentrate surfactin and rhamnolipids from culture supernatant fluids (Mulligan and Gibbs, 1990).

The ability of synthetic surfactants to bind with heavy metal ions is well reported many researchers have conducted bench-scale experiments involving water and wastewater treatment with different synthetic surfactants. Their results are in

favor of using surfactants over other solvents or complexing agents for treatment ground water and wastewater contaminated with heavy metals (Akita et al., 1999; Yurlova et al., 2002).

Generally in surfactant-enhanced ultrafiltration, large groups of surfactant molecules above the CMC (critical micelle concentration) are mixed with polluted water. Smaller contaminants like heavy metal ions will be bound onto larger surfactant micelles. As a consequence, heavy metal ions associated with surfactant macromolecules can be easily retained by the ultrafiltration membrane. Thus, the permeate contains very low concentrations of metal ions and surfactant monomers. In surfactant-based ultrafiltration techniques, surfactant concentrations higher than CMC are required. In many cases, monomeric surfactant molecules in the permeate can be discharged to the ecosystem or the environment if the surfactant is biodegradable.

Biosurfactants can be used as suitable substitutes for chemical synthesized surfactants due to their biodegradability and renewable production sources (Lu et al., 2003). Neilson et al. (2003) used a biosurfactant as an environmentally compatible, not toxic compound. Thus, in biosurfactant-based ultrafiltration techniques, leakage into the permeate of biosurfactants cannot be considered another type of pollution due to their biodegradability, compatibility and non-toxic nature. Biosurfactant-enhanced ultrafiltration membrane process can be applied over a wide range of operating pressure, pH and temperature. Also, it can

minimize the waste to be disposed of. In addition, concentrated metals in the retentate can be reused. Furthermore, biosurfactant in the retentate may be regenerated.

A process to remove the heavy metal ions from metal finishing industry effluents was developed. MEUF, with lecithin as a surfactant, has already been shown to remove heavy metals from aqueous (50 to 90%) and non-aqueous wastes (90 to 97%) (Koseoglu et al., 1993). The binding of various lecithins to cadmium, copper, lead, nickel, and zinc - in a mixture and individually - was studied using a continuous diafiltration method (Ahmadi et al., 1994; Ahmadi et al., 1995). Permeation data of cetyltrimethylammonium bromide (CTABr) in water by ultrafiltration (UF) through zircon membranes are reported as a first step in the knowledge of micellar enhanced ultrafiltration of heavy metals by cationic surfactants (Charbit et al., 1997). Removal of cadmium from wastewater by enhanced ultrafiltration was studied using ceramic membranes and ionic surfactants (Azoug, 1997). The micellar-enhanced ultrafiltration (MEUF) of gold (III) from dilute hydrochloric acid media has been examined using a nonionic surfactant (Akita, 1997). Ultrafiltration (UF) of cobalt(II) and nickel(II) in the presence of a micelle-solubilized hydrophobic ligand has been investigated (Akita et al. 1999). The feasibility of using of a sodium dodecyl sulfate anionic surfactant to purify contaminated waters of Ni(II) ions has been investigated (Yurlova, 2002).

Lai et al. (2004) used enhanced ultrafiltration for purification of protein. Xiarchos, et al. (2003) reported that by micellar-enhanced ultrafiltration, the separation of low molecular weight toxic substances and heavy metals is possible. (Zakrzewska-Trznadel, 2003) studied the application of enhanced ultrafiltration membranes for radioactive wastes processing purposes.

Currently worldwide surfactant production is at a very large scale, and the demand for them is increasing (Maier and Soberon-Chavez, 2000). However, the direction towards less environmentally damaging chemicals makes biosurfactants attractive as they have lower toxicity. With regard to the limitations associated with the existing techniques, the use of biodegradable binding agents (like biosurfactants) might be a better alternative. Therefore, it would be highly desirable to investigate a biosurfactant-based ultrafiltration technique to remove heavy metal ions from wastewater and groundwater.

One family of biosurfactants whose members have been studied extensively is the rhamnolipids of *Pseudomonas aeruginosa* (Perfumo et al., 2006; Lee et al, 2004). Rhamnolipids were first isolated from *P. aeruginosa* by Jarvis and Johnson, who described them as being primarily constructed from rhamnose structural units (Jarvis and Johnson, 1949). *P. aeruginosa* can produce rhamnolipids from many substrates including C11 and C12 alkanes, succinates, pyruvates, citrates, fructose, glycerol, glucose and mannitol (Lee et al, 2004;

Robert et al., 1989), soybean oil (Lang and Wullbrandt, 1999), corn oil (Linhard et al., 1989), and olive oil (Robert et al., 1989).

Rhamnolipids exhibit several promising industrial applications including the surface coatings, adhesives, flocculating, wetting and foaming agents, de-emulsifiers, penetrants. They are also used for the production of fine chemicals including as a source of stereospecific L-rhamnose that is used commercially in the production of high-quality flavor compounds (Maier and Soberon-Chavez, 2000; Banat et al., 2000; Desai and Banat, 1997). Their industrial uses are based on their abilities to lower surface tensions and increase solubility, and on their detergency power, wetting ability and foaming capacity (Banat et al., 2000; Mulligan and Gibbs, 1993). As well, rhamnolipids illustrate antifungal and antibacterial activities. This indicates the possibility of applications in the agricultural and medical fields (Desai and Banat, 1997). Similarly, applications for rhamnolipid as a biological control agent have been explored (Maier and Soberon-Chavez, 2000; Lebron-Paler and Pemberton, 2006).

Furthermore, rhamnolipids have potential for use in various pharmaceutical, health-care, and medical applications because they have low skin irritancy and emulsifying properties. They have also been shown to prohibit protein adsorption on medical implant surfaces (Lebron-Paler and Pemberton, 2006).

In addition, environmental applications of rhamnolipids have been extensively explored in the last two decades (Lebron-Paler and Pemberton, 2006; Mulligan, 2005 ; Maier and Soberon-Chavez, 2000). They can be used for biodegradation of organic contaminants including hexadecane, octadecane, n-paraffin, and phenanthrene in liquid systems, in addition to hexadecane, tetradecane, creosote, and hydrocarbon mixtures in soils. The two possible mechanisms for enhanced biodegradation suggested are: 1) enhanced solubility of the substrate for the microbial cells, and 2) interaction with the cell surface, which increases the hydrophobicity of the surface allowing hydrophobic substrates to associate more easily (Zhang and Miller, 1992; Maier and Soberon-Chavez, 2000). They are used for soil washing or flushing due to their ability to mobilize contaminants.

Realization of this wide variety of applications requires economical commercial-scale production of rhamnolipids (Banat et al., 2000; Maier and Soberon-Chavez, 2000). They are commercially produced in large-scale fermenters and in high yield, making them economically competitive with synthetic surfactants. They are being examined to meet US Environmental Protection Agency Guidelines (Maier, and Soberon-Chavez, 2000). Because of the reasons given above, the rhamnolipid seemed to be an excellent prospect for use in this research.

# CHAPTER 3

## Materials and Methods

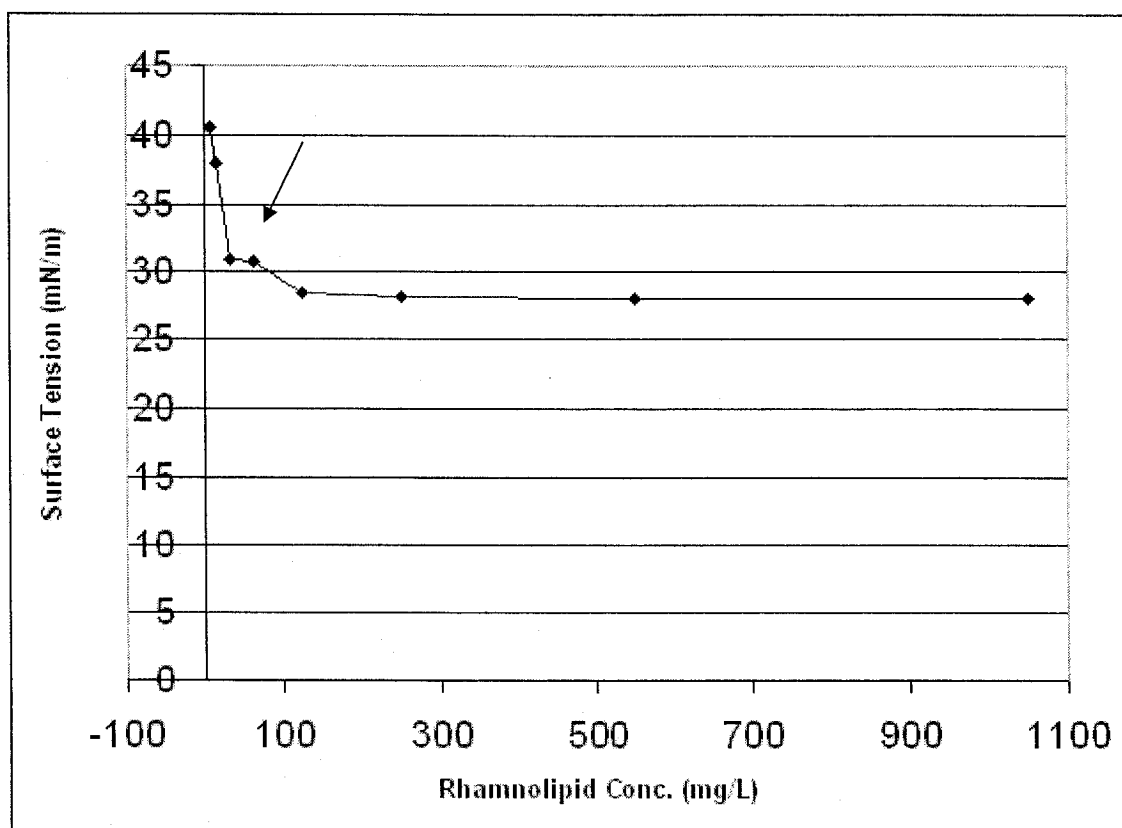
### 3.1 Chemicals

The rhamnolipid biosurfactant (JBR 425) was the surfactant used in this study. It was obtained from Jeneil Biosurfactant Co., USA. The main desirable properties of this surfactant are high molecular weight and low CMC. The surfactant was supplied as a 25% active liquid and used as received. The multivalent metal ions that were used in this study were limited to those of divalent charge. Cadmium nitrate, copper nitrate, nickel nitrate, lead nitrate and zinc nitrate of 99.5 % purity (analytical grade) were used. The salts of metal ions were supplied by Fisher Co. Double distilled water was used in preparation of all reagents and solutions. Wastewaters were collected from a metal refinery plant in Canada. pH analysis was performed using the AR25 Dual Channel pH/Ion Meter by Fisher Scientific. The pH was adjusted with NaOH (0.5N) and HNO<sub>3</sub> (0.5N).

### 3.2 CMC Determination

CMC was determined using the du Nouy ring method (ASTM D971 method). Surface tension measurements were conducted at different rhamnolipid biosurfactant concentrations. The CMC determination was performed by measuring the surface tension at various dilutions (Cooper et al., 1979). The CMC was defined as the point at which the surface tension abruptly increased. The reciprocal of CMC was used as an indication of relative concentrations.

Surface tension measurements were performed by the use of a Tensiomat 21 by Fisher Scientific.



**Figure 3.1. Surface tension as a function of the rhamnolipid concentration**  
What is the arrow? It is not indicated

As shown in Figure 3.1, the CMC for rhamnolipid biosurfactant was about 50 mg/L. The CMC of the rhamnolipid biosurfactant determined in this work is within the same range of the 18 mg/L determined by Thangamani and Shreve (1994) and 60 mg/L reported by Hisatsuka et al., (1981) for the rhamnolipid produced by *Pseudomonas aeruginosa*.



### 3.3 Membrane unit

Hollow-fiber ultrafiltration membrane cartridges from A/G Technology Corporation (MA, USA) were utilized. The effective membrane area was 140 cm<sup>2</sup>. Every cartridge contained polysulfone membrane tubules in a plastic housing. MWCO of 10,000 and 30,000 were utilized.

### 3.4 Experimental flow loop

The flow loop consisted of a feed reservoir, a peristaltic pump, pressure-relief valves, and permeate collection reservoir (A/G Technology Corporation). Permeate connections were of flexible tubing. The flow diagram of MEUF system that used is shown in Figure 3.2. The feed solution containing biosurfactants and metal ions were pumped through the inside (lumen) of the membrane tubes and recirculated as the retentate solution. Permeate solution was passed through the membrane walls, collected from the shell-side of the cartridge and measured to calculate the flux. The permeate flux was calculated as:

$$\text{Flux (l/m}^2\text{/h)} = [ 0.6 ] \times [ \text{permeate flow (ml/min.)} / \text{cartridge area (m}^2\text{)} ] \quad (3.1)$$

The rejection ratio R was calculated as :

$$R = [1 - C_p/C_r] 100\% \quad (3.2)$$

where  $C_r$  is the metal ion concentration in the permeate and  $C_p$  is the metal ion concentration in the retentate.

### **3.5 Experimental procedure for binding studies**

The experiments were run batchwise using an initial feed volume of 1000 mL, with continuous recycling of the retentate stream. A stirrer was utilized to agitate the solution in the feed tank. Before and after experiments, the water flux was measured at the optimized transmembrane pressure (TMP) of 69 kPa to observe the degree of membrane fouling. If the water flux was below 85-95% of the flux of new membrane (of 10,000 MWCO or 30,000 MWCO), the membrane was cleaned again (section 3.6).

The UF runs were performed utilizing the bench-scale cross-flow system. Cadmium nitrate, copper nitrate, nickel nitrate, lead nitrate and zinc nitrate of 99.5 % purity were used to prepare molar solutions in double distilled water for single system. Molar surfactant solutions were prepared using the same water. After preliminary studies, three hours was selected as the optimum contact time. The same salts were utilized to prepare equimolar solutions for binary and ternary solutions. The feed stream containing the metal ions and the surfactant were fed by the peristaltic pump to the inside of the membrane. The rejected solution leaving the cartridge was recycled to the feed reservoir. Samples from the feed, permeate and retentate were collected for further analysis of heavy metal concentrations by Atomic Absorption spectroscopy. Mass balances were done in order to examine the performance of MEUF process. All the experiments were carried out at  $25 \pm 1$  °C and pH  $6.9 \pm 0.1$ . After the run, double distilled water at room temperature was passed through the system in order to flush the

solution from the flow loop. The permeate and retentate were recycled in this case. Then, cleaning procedures for membrane system were followed.

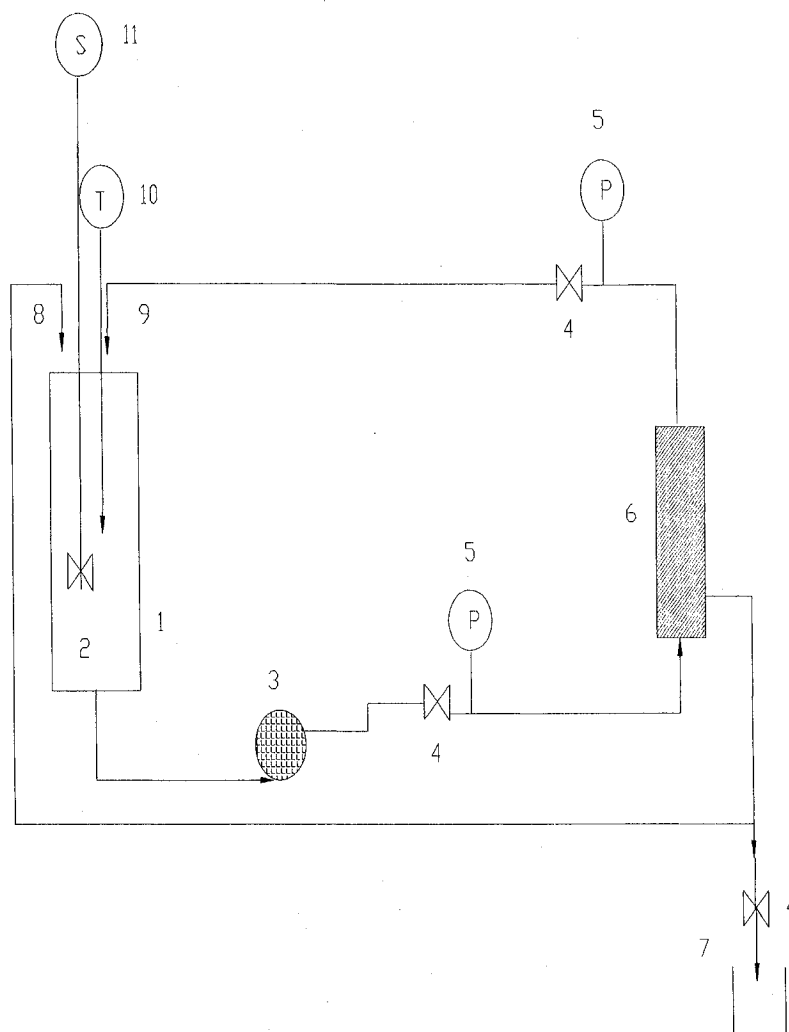
For quality purposes, metal-free and rhamnolipid-free blanks were utilized as controls. The atomic absorption spectrophotometer calibration was performed before each sample analysis session utilizing standard metal solutions prepared from standardized stock solutions. Duplicate samples were prepared and the average was reported. In order to minimize the effect of interferences, the composition of standards and sample solutions were matched as closely as possible.

### **3.6 Cleaning procedures for the UF membrane system**

The cleaning procedures for cross-flow bench-scale UF system are summarized in Appendix A. Also, a photograph of the UF system used in this study is shown in appendix A.

### **3.7 Analysis of Metal Concentrations**

The analysis of metal concentrations in the permeate, retentate and original samples was performed by atomic absorption spectrophotometer (AA) according to standard methods (APHA/AWWA/WPCF, 1998). The atomic absorption spectrophotometer (AA) utilized was a Model AAnalyst 100 by Perkin Elmer. Industrial wastewater samples were filtered using Whatman 42 filter papers and diluted as required.



**Figure 3.2 Flow diagram of MEUF system used in binding studies:**

- (1) Feed Reservoir;    (2) Feed Solution;
- (3) Peristaltic Pump;    (4) Pressure-Relief valve;
- (5) Pressure Gauge;    (6) Ultrafiltration Cartridge;
- (7) Permeate Solution;    (8) Permeate Recycle;
- (9) Retentate Recycle;    (10) Thermometer; (11) Stirrer

The samples were digested according to standard methods (APHA/AWWAWPCF, 1998). Table 3.1 illustrates the characteristic wavelengths utilized by AA for different metals studied.

**Table 3.1 Characteristic wavelengths utilized for AA analysis (flame).**

Metal	Wavelength (nm)
Cd (II)	228.8
Pb (II)	283.3
Cu (II)	327.4
Ni (II)	341.5
Zn (II)	213.5
Fe (II)	422.6
K (I)	248.3
Mg (II)	766.4
Ca (II)	285.2
Na (I)	588.9

### **3.8 Leaching Tests**

#### **Solidification/Stabilization (S/S) for Heavy metal-contaminated Solid Wastes**

In this work, the solidification/stabilization (S/S) technology was applied to stabilize solid wastes contaminated with heavy metals. The consolidated residual from rhamnolipid recovery and the water reuse was used as the basis for

solidification/stabilization (S/S) experiments. Before the solidification/stabilization (S/S), pH was adjusted to about  $12.4 \pm 0.15$  by dropwise addition of NaOH (0.5N). The upper aqueous layer was separated and the precipitate was collected to be utilized in solidification/stabilization (S/S) experiments. Using additives, cement-based solidification/stabilization (S/S) was investigated to stabilize heavy metal-contaminated solid wastes. The chemical composition of Portland cement is shown in Table 3.2. The additives evaluated were a combination of bentonite and epoxy resin. The epoxy used was Sika primer MB (Sika Inc., Canada). The mineral composition of the bentonite used is shown in Table 3.3.

The additives were mixed with type I Portland cement according to optimized ratios explained in section 4.7. The binder-to-solid waste (B/S) ratio was 0.2 to 0.45 (wt/wt) according to the nature of solid waste. The water-to-binder (W/B) ratio was from 0.2 to 0.6 (wt/wt) according to the nature of solid waste. After curing, samples were subjected to the TCLP test. Duplicate samples were prepared for each test, and the average was reported. The toxicity characteristics leaching procedure (TCLP) (EPA, 1992) has been selected as the main test for evaluating the leaching toxicity potential of the solid wastes.

In this work, the TCLP involved particle size of 9.5 mm or less (separated by sieving) in a leachate according to the Method 1311 (US EPA, 1992). Duplicate samples were prepared, and the average was reported. The toxicity characteristics leaching procedure (TCLP) was performed at  $23 \pm 2$  °C.

**Table 3.2 Chemical composition of Portland cement (Taylor, 1990; Frohnsdorff, 1986).**

Oxide	Weight %
CaO <sub>2</sub>	61 - 67
SiO <sub>3</sub>	17 - 24
Al <sub>2</sub> O <sub>3</sub>	3 - 8
Fe <sub>2</sub> O <sub>3</sub>	1 - 6
MgO <sub>3</sub>	0.1 - 4
Na <sub>2</sub> O+K <sub>2</sub> O	0.5 - 1.5
SO <sub>3</sub>	1 - 3

**Table 3.3 Mineral composition of the bentonite (personal communication and Janotka, et al., 2002)**

Material	Total bentonite (Weight %)
Quartz	4
Albite	2
Calcite	2
Pyrite	1
Cristobalite	1
Montmorillonite	90

The resultant TCLP leachate was analyzed for heavy metals using the atomic absorption spectrophotometer Model AAnalyst 100 by Perkin Elmer (Tables 3.1 and 3.4).

**Table 3.4 Water quality parameters measured and method used**

<b>Parameters</b>	<b>Method</b>
Calcium (II)	3111 B. Flame Atomic Absorption
Iron (II)	3111 B. Flame Atomic Absorption
Bicarbonates	2320 – B. (Bicarbonates)
Chemical oxygen demand (COD)	5220 D. (Closed Reflux, Colorimetric Method)
Magnesium (II)	3111 B. Flame Atomic Absorption
Sodium (I)	3111 B. Flame Atomic Absorption
Nitrates (N)	4500 – D. (Nitrate)
Zinc (II)	3111 B. Flame Atomic Absorption
Copper (II)	3111 B. Flame Atomic Absorption
Sulfates	4500 – E. (Sulfates)
Nickel (II)	3111 B. Flame Atomic Absorption
Potassium (I)	3111 B. Flame Atomic Absorption



## **Wastewater and groundwater analysis**

The samples of wastewaters and groundwaters were collected, prepared, filtered, preserved and analyzed according to standard methods (APHA/AWWAWPCF, 1998). Table 3.4 shows the water quality parameters measured and method used. The metal analysis and COD tests were performed at the Environmental Engineering lab, Concordia University. The values of other water quality parameters were provided by personal communications (Nov. 2004).

Metal binding was quantified from the following equation:

$$q = (C_{\text{init.}} - C_{\text{eq}}) V / M \quad (3.3)$$

where  $q$  is the mass of heavy metal ion bound per unit mass of rhamnolipid biosurfactant at equilibrium.  $C_{\text{init.}}$  is the initial concentration of metal ions in the solution,  $C_{\text{eq}}$  is the concentration of heavy metal ions at equilibrium,  $V$  is the volume of the solution, and  $M$  is the mass of rhamnolipid biosurfactant (Overdevest et al., 2000).

### **3.9 Preliminary trials for recovery of rhamnolipid**

Experiments were performed by adding nitric acid (0.5 N) to the retentate containing rhamnolipid biosurfactant, until the pH of the solution reached the optimum value. From preliminary trials, it was found that nitric acid gave better recovery than sulfuric or hydrochloric acids. The solution was then centrifuged at 2500 rpm with IEC HN-S II centrifuge. Then, the pH was raised again to  $\text{pH } 6.9 \pm$

0.1; finally, the concentration of rhamnolipid biosurfactant was determined by UV spectrophotometric measurement at 235 nm (Appendix B).

$$\% \text{ rhamnolipid regenerated} = 100 \left[ \left( \frac{[\text{rhamnolipid}] \text{ regenerated}}{[\text{rhamnolipid}] \text{ initially present in the retentate}} \right) \right] \quad (3.4)$$

### 3.10 Preliminary trials for water reuse

The top layer of the retentate from the MEUF process was collected. Erlenmeyer flasks of different sizes were utilized in these experiments. Each flask was shaken for 90 minutes at 120 rpm, and then left to precipitate for 9-12 hours. The water was separated to be reused. Temperature was maintained at  $25 \pm 1$  °C. The water recycle ratio (R) is the ratio of the volume of recycled water to the total volume of the water used in MEUF process (Appendix C).

$$\% \text{ reusable waters} = 100 \left[ \left( \frac{\text{volume of separated water}}{\text{volume of initial water}} \right) \right] \quad (3.5)$$

### 3.11 Methods of Mathematical Analysis

#### 3.11.1 Optimization Methodology

The response surface methodology was used for optimization studies (Madamba and Bekki, 2001; Mudahar et. al., 1990; Box and Behnken, 1960; Mudahar et.

al., 1989; SAS, 1985; Gilbert, 1987). Software packages used for data analysis were SPSS 10 (Campus Computer Store Inc. USA, 2002) and SAS (SAS Institute Inc, 1987). Second order polynomial (SOP) models as functions of the independent variables were utilized to generate response surface and contour plots. Some lack of fit tests were performed (Berthouex and Brown, 2002; Nete, et al., 1990; Drapper and Smith, 1998; Box and Jenkins, 1994; Gibbons, 1994). Separate validation experiments were performed under optimum conditions to verify predictions and adequacy of the second order polynomial (SOP) models.

### **3.11.2 Analysis of variance (ANOVA)**

Analysis of variance (ANOVA) is a parametric test used to measure the contribution of experimental factors (independent variables) to the prediction of a dependent variable and can be used to check for interactions between the independent variables (Berthouex and Brown, 2002; Drapper and Smith, 1998; Gilbert, 1987; Box and Jenkins, 1994; Gibbons, 1994).

In ANOVA evaluations, the F-test statistic was used to measure the contribution of each independent variable in predicting the dependent variable (Berthouex and Brown, 2002).

## 4. Results and Discussion

### 4.1. Effect of operating parameters on the performance of the MEUF system

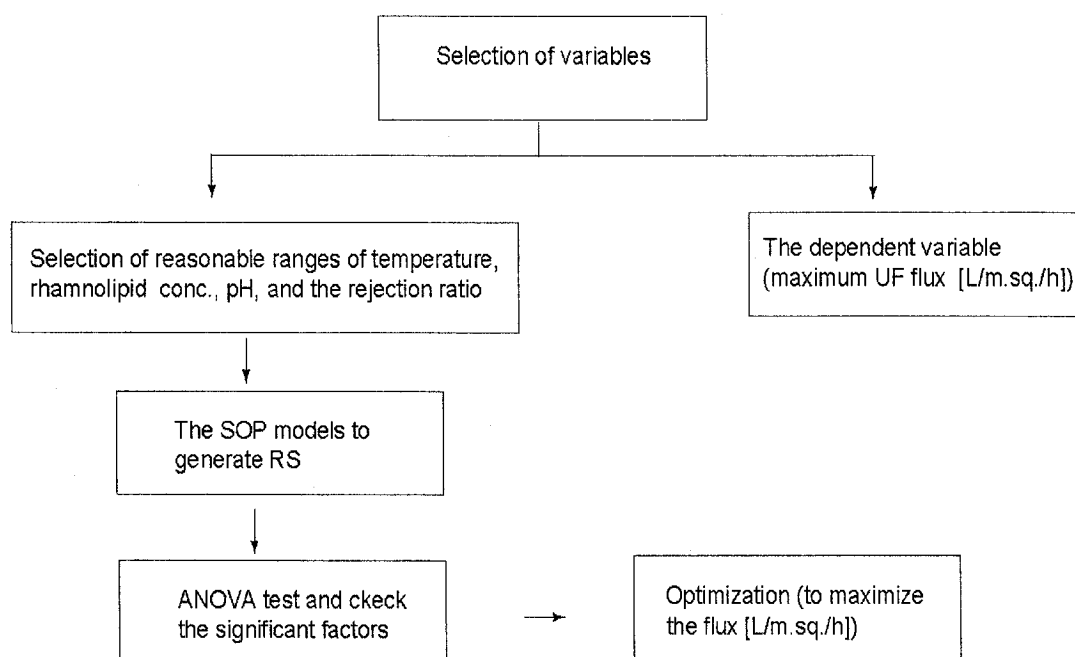
#### Optimized conditions for the MEUF system

In general, the ultrafiltration process is usually performed under moderate optimum conditions in order to give a relatively high flux. The goals of this work were to investigate the effects of different operating conditions on the system performance of cross-flow rhamnolipid-enhanced ultrafiltration. Several operating conditions for different metal and rhamnolipid biosurfactant concentrations were conducted to determine the optimal conditions for removal of metal ions from solution with the maximum flux.

In order to efficiently achieve the most influential factors to the MEUF system and the maximum flux, optimization by the response surface (RS) approach was utilized. The operating parameters including rhamnolipid concentration in the feed, pressure difference, temperature, and required rejection ratio were designated independent variables while the maximum flow rate was designated as a dependent variable.

Second order polynomial (SOP) models were utilized as functions of the independent variables. An analysis of variance (ANOVA) was performed to determine which of the parameters significantly affected the dependent variable.

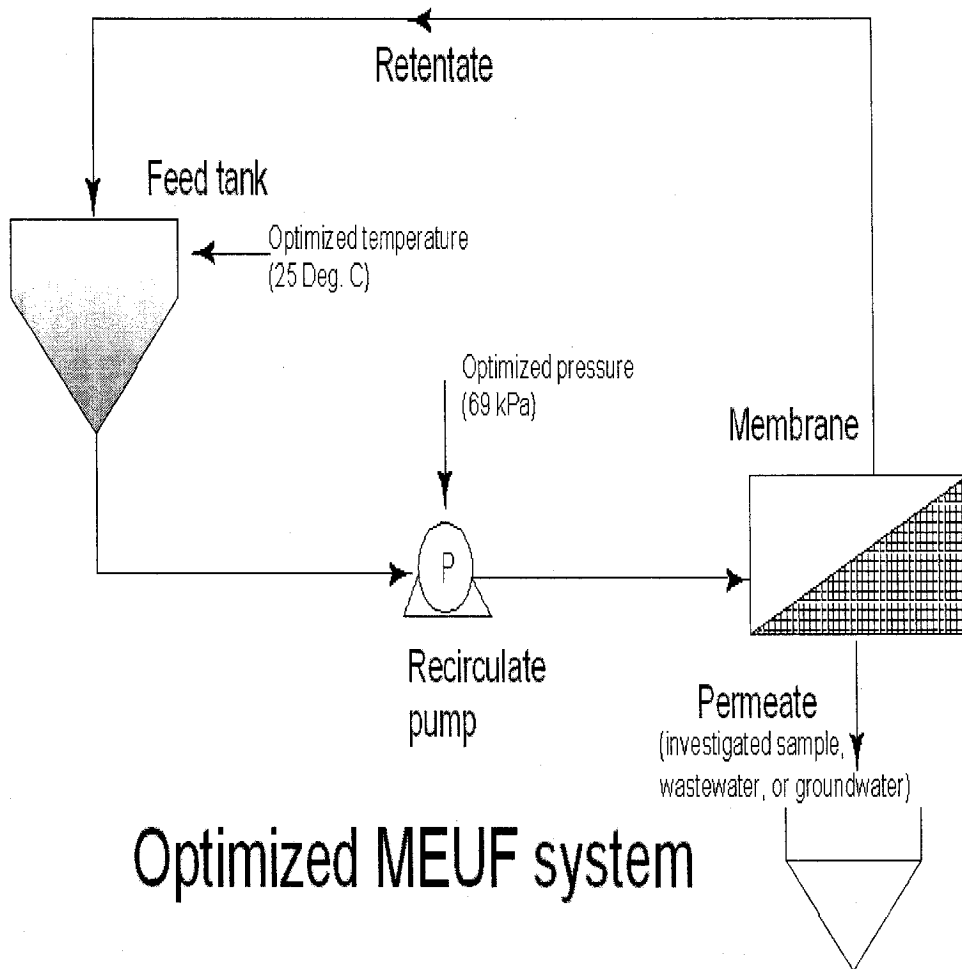
Response surface and contour plots were generated as shown in Figures 4.1.3, to 4.1.8 Optimum regions of independent and dependent variables were achieved.



SOP = Second order polynomial (SOP); RS = Response surface

Conc. = Concentration

**Figure 4.1.1 Experimental data flow to select the optimized conditions to the MEUF system and the maximum flux.**

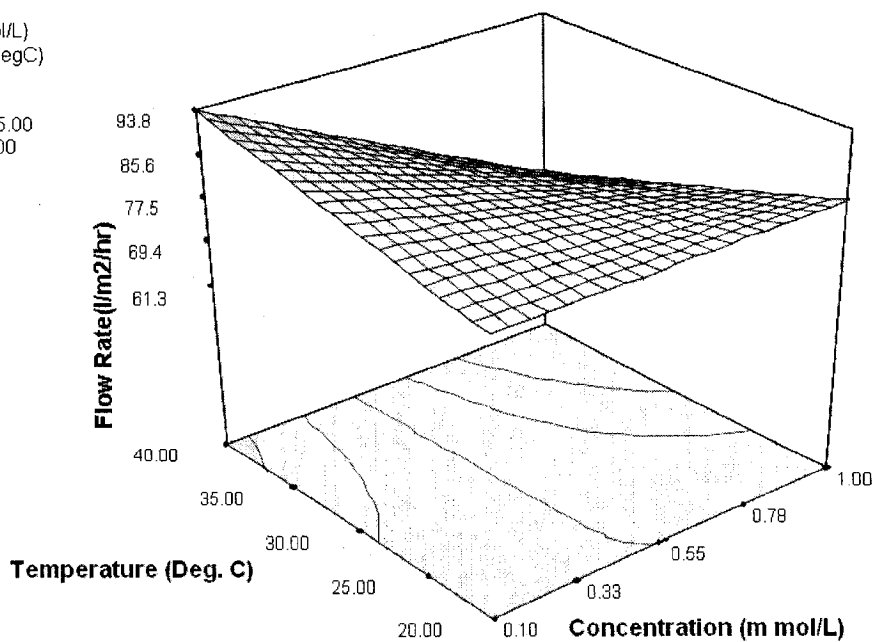


**Figure 4.1.2 Schematic illustration of the experimental MEUF system indicating optimum temperature and pressure.**

The major objective was to maximize the flux. Figure 4.1.1 shows the experimental data flow to achieve the maximum flux and optimum conditions. Figure 4.1.2 shows the ultrafiltration system utilized.

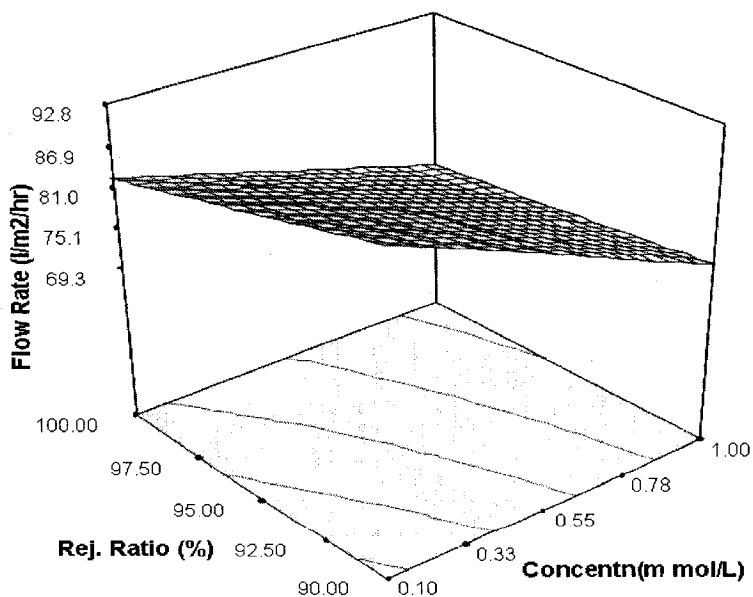
Flow Rate(l/m<sup>2</sup>/hr)  
 X = C: Concentn(m mol/L)  
 Y = A: Temperature (DegC)

Actual Factors  
 B: Pressure (Psig) = 65.00  
 D: Rej. Ratio(%) = 95.00



**Figure 4.1.3 Response surface obtained for rhamnolipid concentration and temperature.**

Flow Rate(l/m<sup>2</sup>/hr)  
 X = C: Concentn(m mol/L)  
 Y = D: Rej. Ratio(%)



**Figure 4.1.4 Response surface obtained for rejection ratio and rhamnolipid concentration.**

Flow Rate(l/m2/hr)

X = A: Temperature (DegC)

Y = D: Rej. Ratio(%)

Actual Factors

B: Pressure (Psig) = 65.00

C: Concentn(m.mol/L) = 0.55

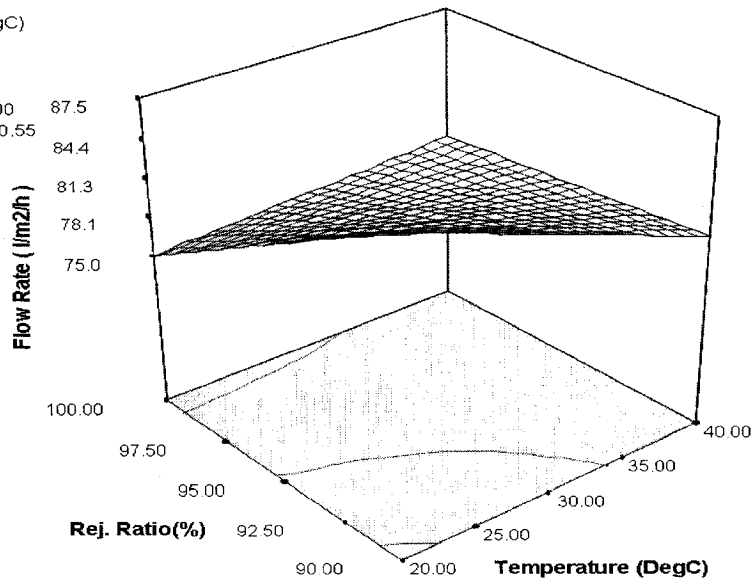


Figure 4.1.5 Response surface obtained for rejection ratio and temperature.

Flow Rate(l/m2/h )

X = B: Pressure

Y = C: Concentn

Actual Factors

A: Temperature (DegC) = 30.00

D: Rej. Ratio(%) = 95.00

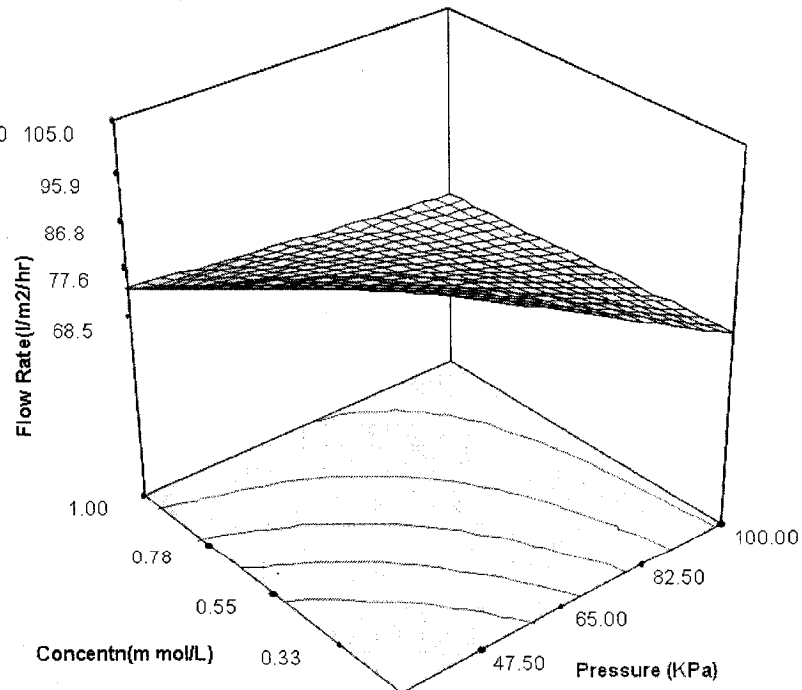


Figure 4.1.6 Response surface obtained for pressure and rhamnolipid concentration



Flow Rate(l/m<sup>2</sup>/hr)  
 X = A: Temperature (DegC)  
 Y = B: Pressure (Psig)

Actual Factors  
 C: Concentn(m.mol/L) = 0.55  
 D: Rej. Ratio(%) = 95.00

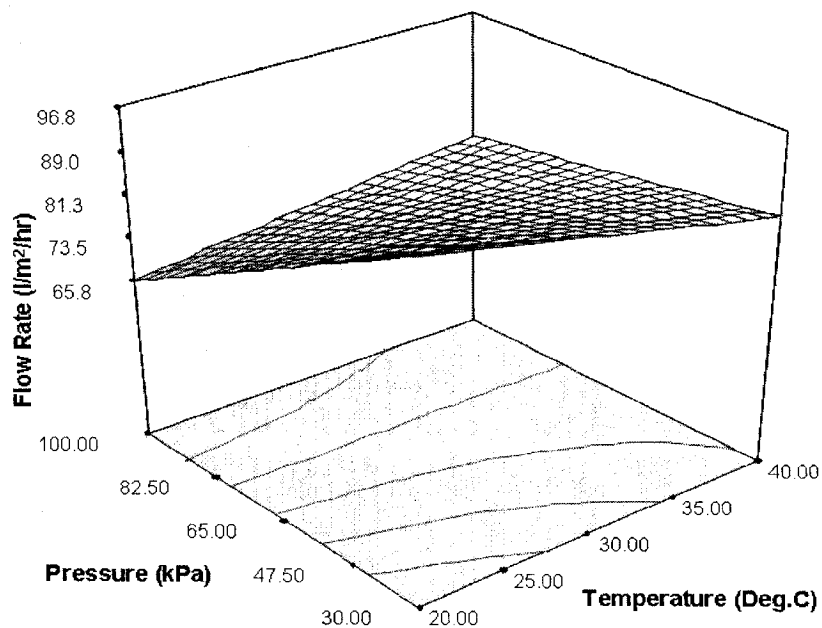


Figure 4.1.7 Response surface obtained for temperature and pressure.

Flow Rate(l/m<sup>2</sup>/hr)  
 X = C: Concentn(m mol/L)  
 Y = D: Rej. Ratio(%)

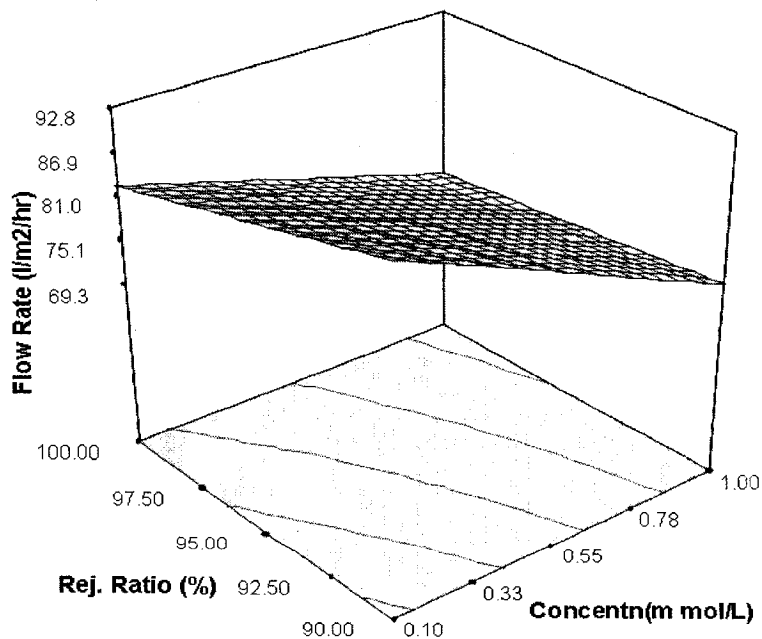


Figure 4.1.8 Response surface obtained for rejection ratio and rhamnolipid concentration.

From Figures 4.1.3, to 4.1.8, it can be seen that the suggested ranges of transmembrane pressures were from 30 to 100 kPa, biosurfactant-to-metal molar ratios of 2:1, temperature of 20 to 40 °C, and permeate flux of 300 to 100 (l/m<sup>2</sup>/h). Separate validation experiments using the lab-scale UF system were performed to verify these optimum conditions. The most applicable operating conditions were transmembrane pressure 69 ±2 kPa, biosurfactant-to-metal molar ratios of 2:1, temperature of 25 ±1°C, and pH of 6.9 ±0.1. These values are expected according the literature on UF treatment (Yoo et al., 2003; Akita et al., 1997).

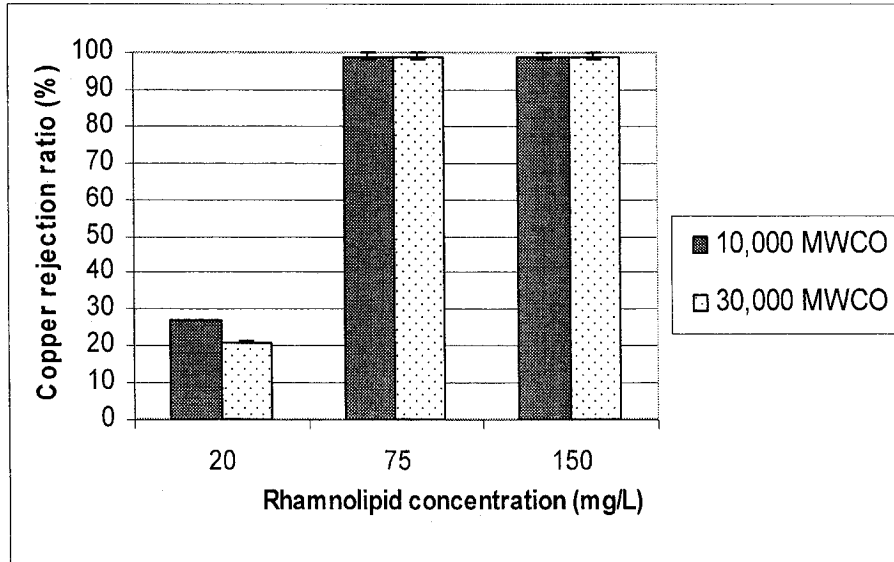
#### **4.1.2 Rhamnolipid concentration effect on metal ions**

The effect of initial rhamnolipid biosurfactant concentrations on rejection ratios of copper, zinc, nickel and cadmium ions in the permeate by membranes with molecular weight cut off (MWCO) of 30,000 Da and 10,000 Da at an initial metal ion concentration of 5 mg/L are illustrated in Figures 4.1.9 to 4.1.12, respectively. Increasing the relative concentration of the rhamnolipid biosurfactant in the feed will lead to an increase in the fraction of total rhamnolipid biosurfactant present in micellar form. As a consequence, the separation efficiency for copper, zinc, nickel and cadmium ions increased. The micellar-enhanced ultrafiltration (MEUF) experiments were conducted at a constant copper, zinc, nickel and cadmium ion concentration of 5 mg/L. These levels were chosen to reflect rejection ratios with

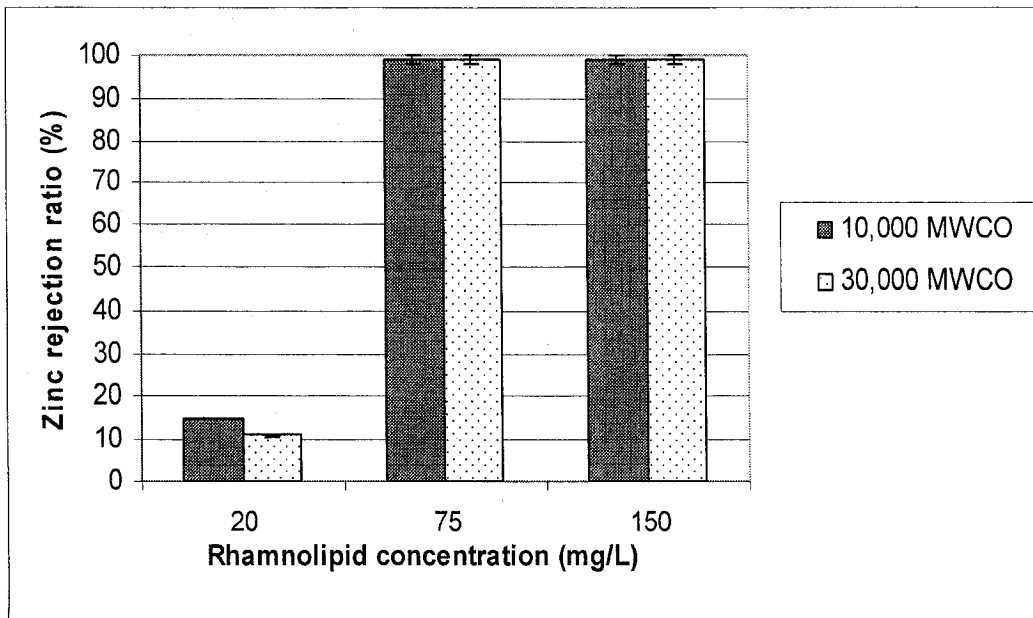
low metal ion concentrations. There is not much difference between the membranes with regard to the rejection ratio. This indicates that the size of rhamnolipid micelles is larger than the pore sizes of the two membranes.

Many surfactants show poor performance below the CMC's. However in this research, rejection of the metal ions with rhamnolipid was achieved even below the CMC using the UF system (Figures 4.1.9 to 4.1.12). This may be due to the formation of complexes between metal ions and the rhamnolipid biosurfactant even below the CMC. This may also be due to fouling on the membrane which retains some of the metals.

Regarding the rejection ratio of lead ions and rhamnolipid biosurfactants as a function of concentration of rhamnolipid biosurfactants in the feed at a constant lead ion concentration of 5 mg/L, it was found that the rejection ratio of lead ions is very high (>99.9%). Also, the lead concentrations of 50 mg/L or higher followed the same trend (data not shown). The concentration of lead ions in the permeate was very low (not detected by atomic absorption spectrophotometer which means that the metal concentration is less than 0.01 mg/L).



**Figure 4.1.9 Rejection ratio of copper ions at an initial copper ion concentration of 5 mg/L.**



**Figure 4.1.10 Rejection ratio of zinc ions at an initial zinc ion concentration of 5 mg/L.**

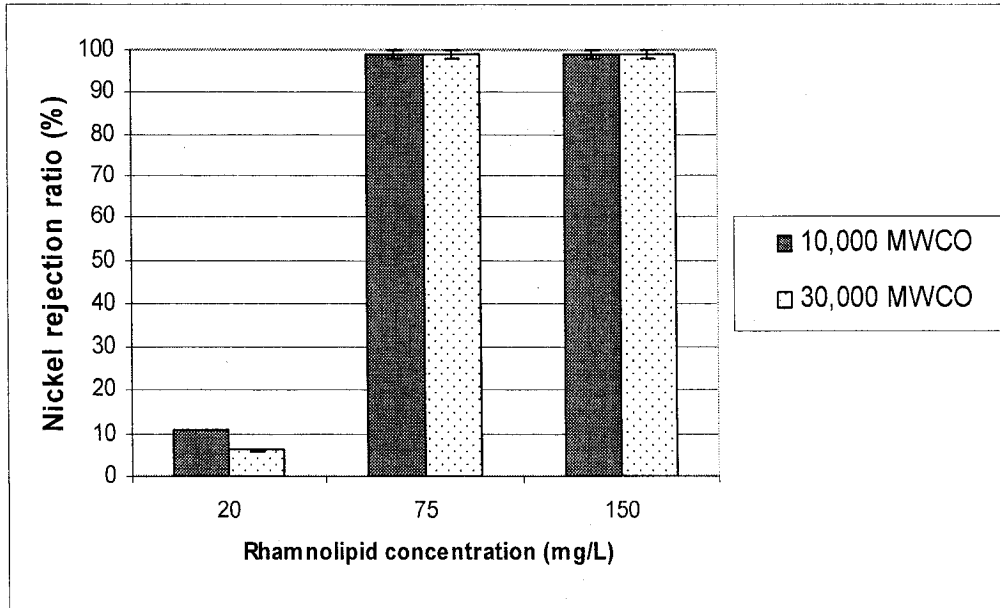


Figure 4.1.11 Rejection ratio of nickel ions at an initial nickel ion concentration of 5 mg/L.

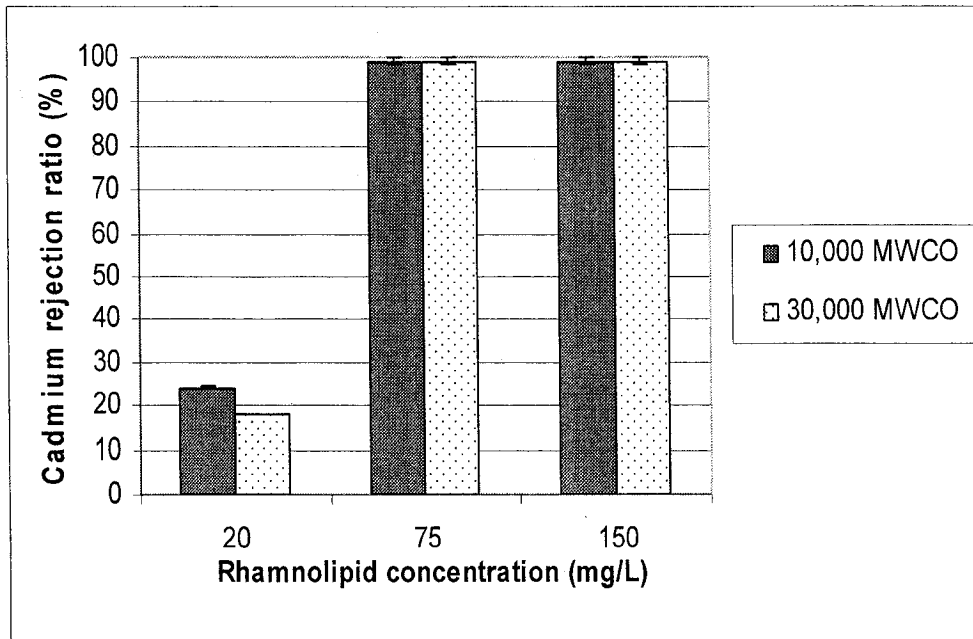


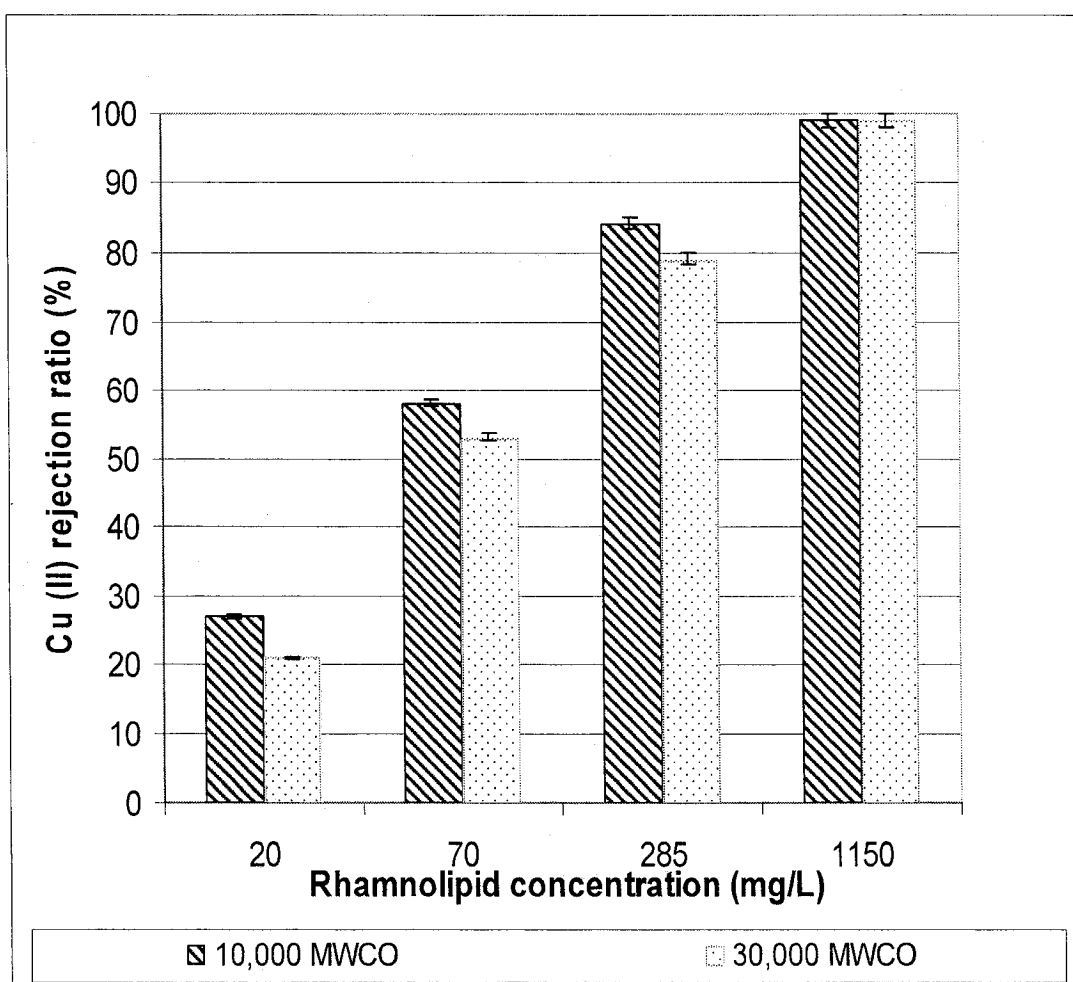
Figure 4.1.12 Rejection ratio of cadmium ions at an initial cadmium ion concentration of 5 mg/L.

The very high rejection ratio of lead ions as well as the very low concentration of lead ions in permeate may be due to the formation of lead-rhamnolipid complexes above and even below the CMC. The dark white complexes are precipitated and can be rejected by the UF membrane. This was observed upon subsequent examination. This lead ion behavior can, probably, be explained by taking into account the rhamnolipid precipitation in the gel layer of both membranes. Lead-rhamnolipid precipitates can be adsorbed on the surface of the membrane and/or plug the pores of the membranes. This, probably, explains why concentration polarization occurs at a lower concentration range compared to the other metal ions in this study.

To further investigate the effect of higher metal concentrations, metal ion levels of 50 mg/L were used. The effect of initial rhamnolipid concentrations on the rejection ratios of copper, zinc, nickel and cadmium ions in the permeate by membranes with molecular weight cut off (MWCO) of 30,000 Da and 10,000 Da at an initial metal ion concentration of 50 mg/L are shown in Figures 4.1.13 to 4.1.16, respectively. The micellar-enhanced ultrafiltration (MEUF) experiments were conducted at an initial copper, zinc, nickel and cadmium ion concentrations of 50 mg/L in the feed.

The rejection ratios of the metal ions in permeate followed the same trends as in the case of initial metal ion concentrations of 5 mg/L. Similarly, increasing the

relative concentration of the rhamnolipid biosurfactant in the feed will lead to an increase in the fraction of total rhamnolipid biosurfactant present in micellar form. Thus, the separation efficiency for the metal ions will be increased. It can be seen from Figures 4.1.9 to 4.1.16 that more than 99.0 % removal was achieved by applying biosurfactant-to-metal molar ratios of 2:1.



**Figure 4.1.13** Rejection ratio of copper ions at an initial copper ion concentration of 50 mg/L and pH 7.

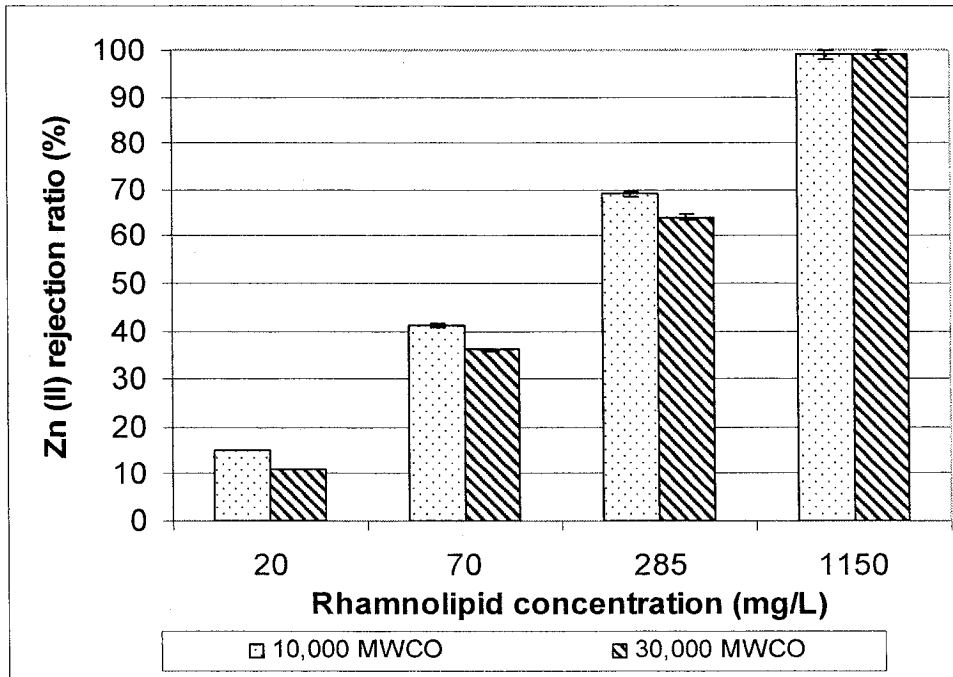


Figure 4.1.14 Rejection ratio of zinc ions at an initial zinc ion concentration of 50 mg/L and pH 7.

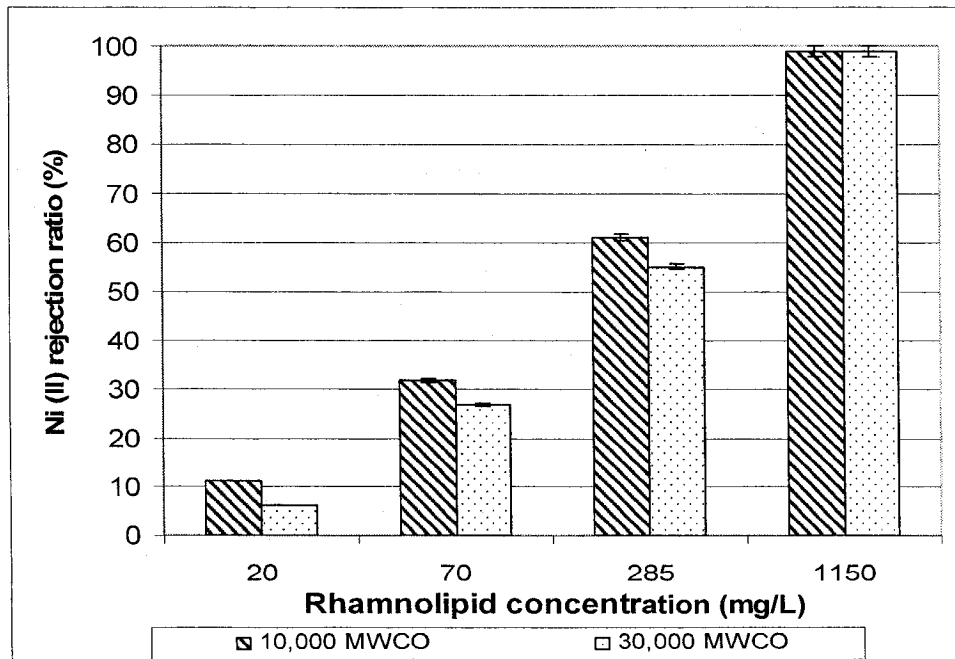
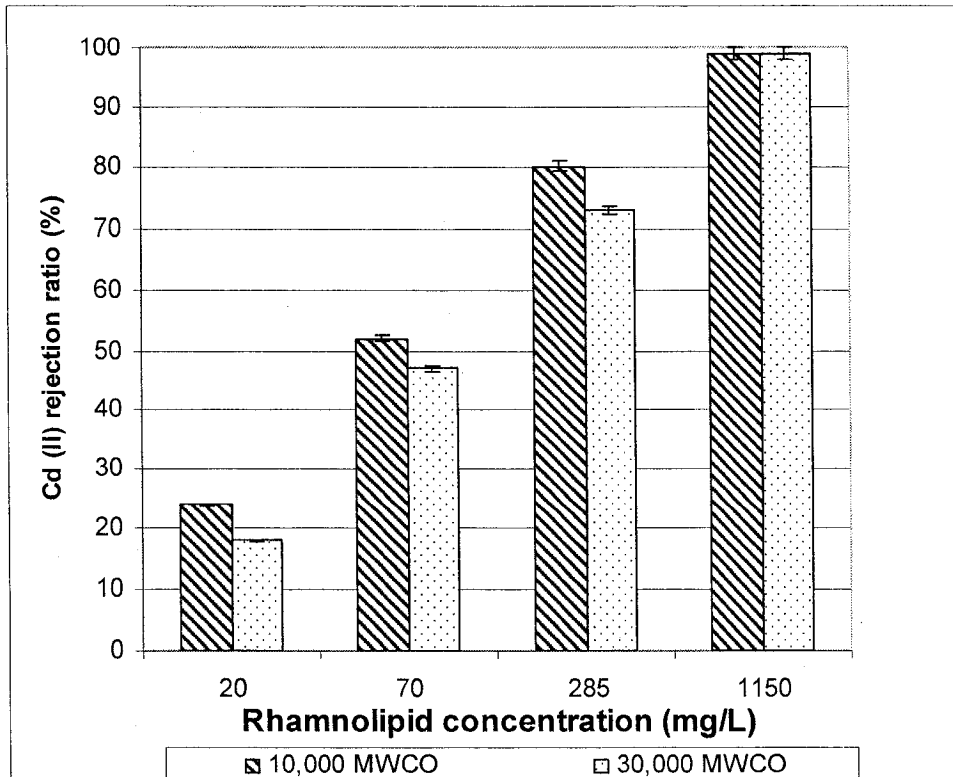


Figure 4.1.15 Rejection ratio of nickel ions at an initial nickel ion concentration of 50 mg/L and pH 7.



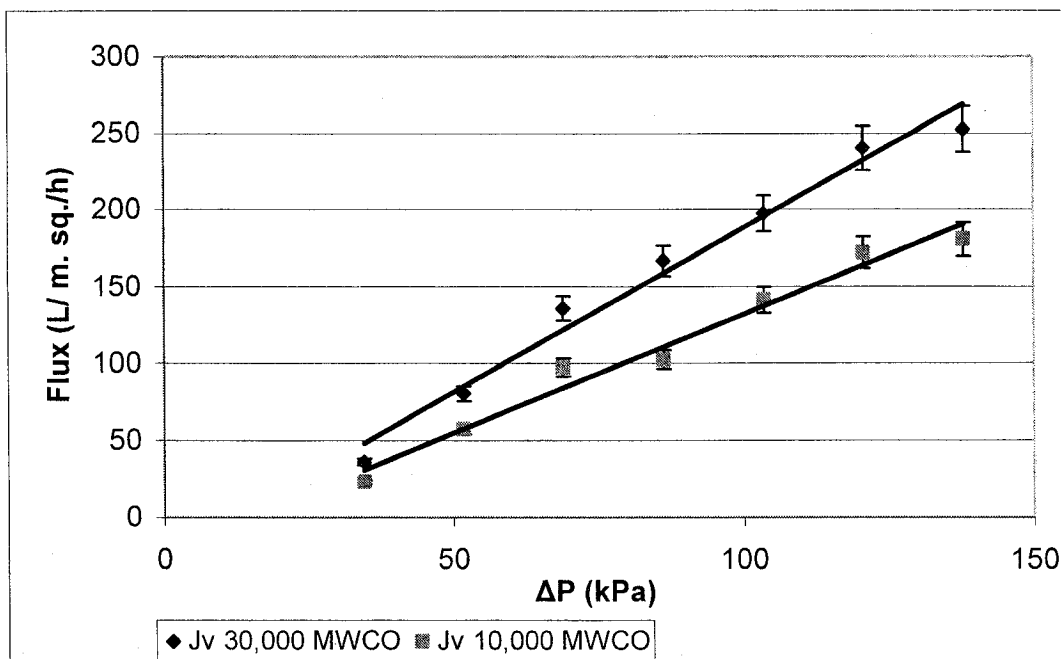


**Figure 4.1.16** Rejection ratio of cadmium ions at an initial cadmium ion concentration of 50 mg/L and pH 7.

#### 4.1.3 Effect of transmembrane pressure ( $\Delta P_{tm}$ ) on permeate flux

Figure 4.1.17 shows the changes of permeate fluxes of  $Zn^{2+}$  by membranes with molecular weight cut off (MWCO) of 30,000 Da and 10,000 Da as functions of transmembrane pressure ( $\Delta P_{tm}$ ) regarding zinc ions. Duplicate tests of five replications each were prepared, and the averages were reported. The transmembrane pressures ( $\Delta P_{tm}$ ) were found to have significant effects on the permeate fluxes. Within the ranges chosen for our experiments, the permeate fluxes are almost linear functions of

transmembrane pressure ( $\Delta P_{tm}$ ). The copper, cadmium, and nickel ions gave largely identical results, and therefore are not shown.



**Figure 4.1.17 Permeate flux of zinc ions as a function of transmembrane pressure ( $\Delta P_{tm}$ ) at an initial zinc ion of 50 mg/L and pH 7.**

Our results indicate that beyond these ranges, permeate fluxes reached plateau values that depend on the rhamnolipid biosurfactant concentrations and applied transmembrane pressures ( $\Delta P_{tm}$ ). It is important to mention that the tangential velocity of the feed solution has to be high enough to avoid any problem of

concentration polarization. For the permeate flux, an average of five readings was calculated and plotted.

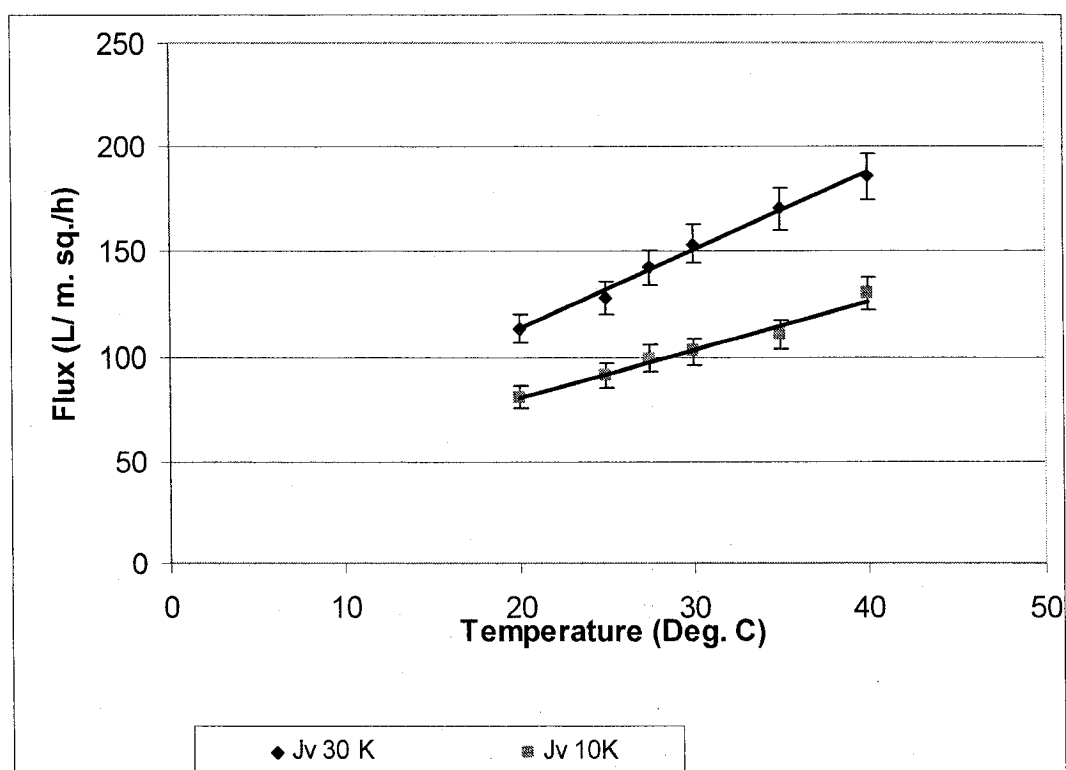
#### **4.1.4 Effect of transmembrane pressure ( $\Delta P_{tm}$ ) on rejection ratio**

Changes of rejection ratios (%) by membranes with molecular weight cut off (MWCO) of 30,000 Da and 10,000 Da were investigated as functions of transmembrane pressure ( $\Delta P_{tm}$ ). Optimal experimental conditions were found to be a metal concentration between 10 and 50 mg/L, a biosurfactant-to-metal molar ratio of 2:1, a temperature of  $25 \pm 1^\circ\text{C}$ , and a pH of  $7 \pm 0.1$ . The metal ions tested were nickel, copper, cadmium, and zinc ions (data not shown). It was found that the transmembrane pressure ( $\Delta P_{tm}$ ) does not have a significant influence on the rejection ratios (%). Within the ranges chosen for the experiments (transmembrane pressure from 30 – 140 kPa), the rejection ratios (%) did not change for all the metal tested by increasing the transmembrane pressure ( $\Delta P_{tm}$ ).

#### **4.1.5 Temperature effect on permeate flux**

The results presented in Figure 4.1.18 illustrate the changes in permeate fluxes of  $\text{Cu}^{2+}$  for the membranes of 10,000 and 30,000 as a function of the temperature at a pH of 7. It shows that increasing the temperature led to an increase in permeate flux for the two membranes. From a theoretical point of view, the permeate flux is known to be inversely proportional to viscosity (Cardew and Le,

1998). Increasing the temperature will lead to a decrease of viscosity of the solution and hence an increase in permeate flux. An average of three samples was calculated and plotted. The nickel, cadmium and zinc ions followed the same trend (data not shown).

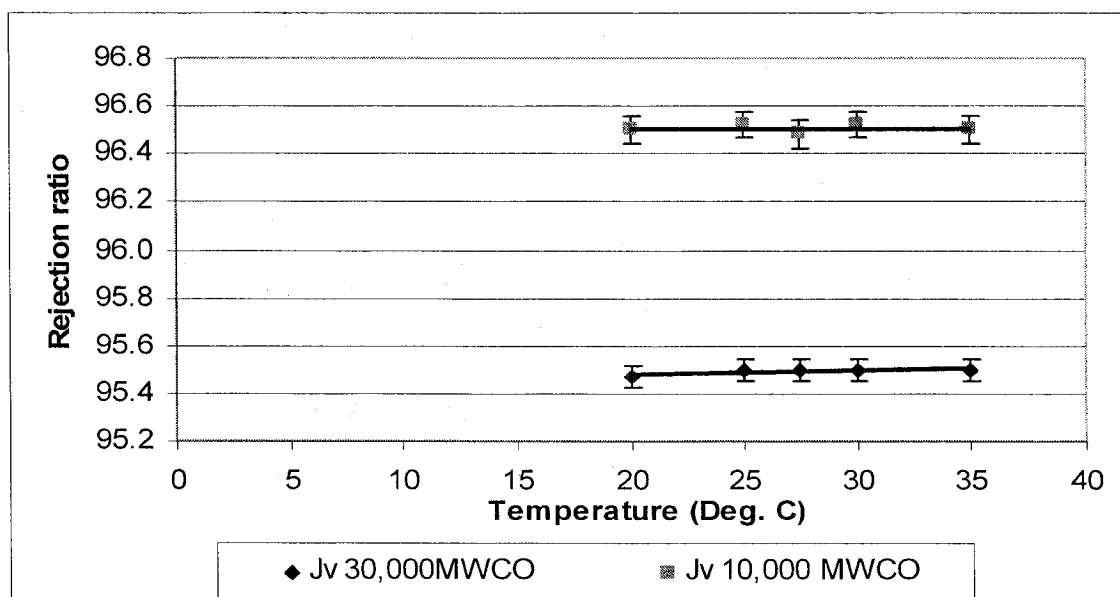


**Figure 4.1.18 Permeate flux of copper ions and rhamnolipid biosurfactants as a function of temperature.**

#### 4.1.6 Temperature effect on rejection ratio

Figure 4.1.19 shows the rejection ratios (%) by membranes with molecular

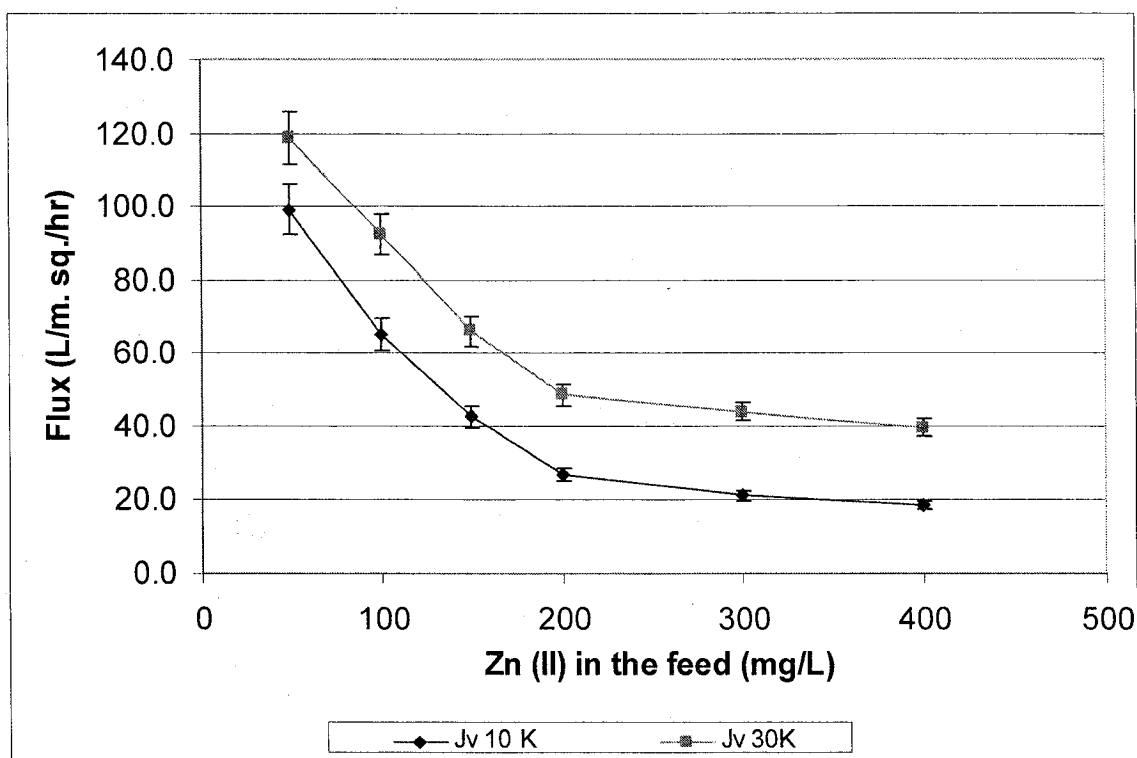
weight cut off (MWCO) of 30,000 Da and 10,000 Da as a function of the temperature regarding cadmium ions. The cadmium concentration was 0.5 mmol/L, the rhamnolipid concentration was 1.0 mmol/L, and the pH was 7. The temperature dependence of the metal separation using ultrafiltration has been the subject of experimental research. Canizares et al., (2005) reported that working at higher temperatures improved turbulence conditions over the membrane and made possible to increase the treatable concentration of metal ions in waste stream.



**Figure 4.1.19 Rejection ratio of cadmium ions and rhamnolipid biosurfactants as a function of temperature.**

In our MEUF system, increasing the temperature did not lead to a significant

influence on the rejection ratios (%). For the MEUF system, the temperature increase from 20 to 40 °C had a negligible effect on the rejection ratios (%) of copper, zinc, nickel, and cadmium ions. The nickel, copper and zinc ions followed the same trend (data not shown).



**Figure 4.1.20 Permeate flux of zinc ions and rhamnolipid biosurfactants as a function of concentration of metal ions in the feed.**

#### 4.1.7 Rhamnolipid biosurfactant concentration on permeate flux

Figure 4.1.20 shows the changes of permeate flux of zinc ions and rhamnolipid biosurfactants when filtered by membranes with molecular weight cut offs (MWCO) of 30,000 Da and 10,000 Da, as a function of initial zinc metal ion concentrations. Rhamnolipid concentration was held constant at 2 mmol/L, and pH was kept at 7. Here, for biosurfactant concentrations greater than the CMC, when the relative initial zinc metal ion concentrations increased (Figure 4.1.20), the solution viscosity of the retentate increased. Thus, permeate fluxes decreased.

Furthermore, Figure 4.1.20 shows that in this study, the ultrafiltration membrane with smaller pore size (MWCO 10,000) gave more decrease in permeate flux than did that of MWCO 30,000 using rhamnolipid-based UF for zinc ions. This flux behavior of zinc ions can, probably, be explained by taking into account that the increase in retentate viscosity of MWCO 10,000 was much higher than that of MWCO 30,000. Since the rhamnolipid micelles or zinc-rhamnolipid complexes were rejected by the ultrafiltration membranes, they usually form the highly viscous boundary layer adjacent to the membranes surface which is well-known as a gel layer. In addition to the hydrodynamic resistance of the membranes themselves, the gel layer can act as an additional resistance to flow through the membranes, resulting in the flux decline. The nickel, copper and cadmium ions

followed the same trend (data not shown). Mathematical optimization and lack of fit analysis on the data are presented in section 4.10.

### **Summary**

In this work, we examined how the rhamnolipid-enhanced ultrafiltration of different metal ions (lead, copper, zinc, nickel and cadmium) was affected by variation of different major operating conditions. It was found that the initial rhamnolipid biosurfactant concentration has a major effect on metal removal while transmembrane pressure ( $P_{tm}$ ) and temperature have no significant effects on metal removal using membranes with molecular weight cut off (MWCO) of 30,000 Da and 10,000 Da. The results indicated that the initial rhamnolipid biosurfactant concentration and transmembrane pressure ( $P_{tm}$ ) temperature have major effects on permeate flux. Additionally, the results indicated that the rhamnolipid-enhanced ultrafiltration system may not be suitable for the treatment of lead contaminated water (concentrations above 50 mg/L) due to the potential risk of membranes fouling. Optimization by the response surface methodology and validation experiments determined that the best operating conditions were a transmembrane pressure of  $69 \pm 2$  kPa, biosurfactant-to-metal molar ratios of 2:1, a temperature of  $25 \pm 1^\circ\text{C}$ , and pH of  $6.9 \pm 0.1$ .



## 4.2. Isotherm models

In an attempt to investigate the binding of the various metal ions onto the rhamnolipid biosurfactant, three isotherm models were employed for portions of metal ions bound to the rhamnolipid biosurfactant and in solution at equilibrium. The Langmuir, Freundlich, and linear isotherm models for each metal have been applied to determine which model fits the data better. The results obtained from bench-scale investigations for rhamnolipid enhanced UF under the optimum operating conditions were used (section 4.1). The same conditions of MEUF system, materials, reagents, methods and membranes were utilized.

### Langmuir isotherm model

The aim of the work was to determine the portions of metal ions bound to the rhamnolipid biosurfactant and in solution at equilibrium and to apply the Langmuir isotherm model to each metal ion. The two different UF membranes were used. The empirical Langmuir equation (equation 4.2.1) is known to be based on a monolayer adsorption by the adsorbent with a homogeneous energy distribution of active sites. The Langmuir adsorption isotherm is based on the following assumptions (Overdevest et al., 2000 and McCash, 2001).

1. adsorbate molecules are held at a fixed number of localized sites;
2. each site can accommodate one single adsorbate molecule;
3. adsorption energy is equal for all the sites; and
4. neighboring adsorbate-adsorbate interactions can be neglected.

$$[Q]_L = K_L [C]_L / ( 1 + a_L [C]_L ) \quad (4.2.1)$$

$[Q]_L$  is the adsorbed phase concentration, (mg/g). The constants  $K_L$  and  $a_L$  are the characteristics of the Langmuir isotherm equation.  $K_L$  is known to be an indicator of adsorption capacity and  $a_L$  is considered to express adsorption intensity.  $[C]_L$  is bulk phase concentration, (mg/L) (Overdevest et al., 2000). A linear form of Langmuir equation is (Moreno-Vellosalada et al, 2003; Moreno-Vellosalada et al, 2002) :

$$1 / [Q]_L = [ 1 / (K_L a_L ) ( 1 / [C]_L ) ] + 1 / [ a_L ] \quad (4.2.2)$$

The portions of the four metal ions bound to the rhamnolipid biosurfactant and in solution were determined. Langmuir isotherms were plotted for zinc, copper, nickel and cadmium binding onto rhamnolipid biosurfactant for the membranes with molecular weight cut-off (MWCO) of 10,000 and 30,000 Da. The square linear regression ( $R^2$ ) have been determined for each metal ion utilizing the least-square method. Table 4.2.1 presents the correlation regression coefficients ( $R^2$ ) for zinc, copper, nickel and cadmium . As shown in Table 4.2.1, the Langmuir isotherm model did not fit the data. Also as presented in same Table, the values of the correlation regression coefficients ( $R^2$ ) of Langmuir model are significantly low since they are within the range from 0.09 to 0.26.

**Table 4.2.1 Values of square linear regression ( $R^2$ ) for the Langmuir adsorption isotherm**

<b>Metal ion</b>	<b>Membrane</b>	<b>Square linear regression (<math>R^2</math>)</b>
<b>Zinc</b>	10,000 MWCO	0.0980
<b>Zinc</b>	30,000 MWCO	0.2353
<b>Copper</b>	10,000 MWCO	0.1743
<b>Copper</b>	30,000 MWCO	0.1156
<b>Nickel</b>	10,000 MWCO	0.2574
<b>Nickel</b>	30,000 MWCO	0.2041
<b>Cadmium</b>	10,000 MWCO	0.2642
<b>Cadmium</b>	30,000 MWCO	0.2354

Consequently it would appear that the Langmuir equation gives a poor representation of the experimental binding data.

#### **The linear isotherm model**

Because the Langmuir adsorption isotherm did not apply to our experimental equilibrium data, the linear isotherm model was investigated. The aim of the work was to determine the portions of metal ions bound to the rhamnolipid

biosurfactant and in solution at equilibrium and to apply the linear adsorption model to each metal ion. The two different UF membranes were used.

The linear isotherm model is commonly presented as :

$$Q_{L,AD} = K_{L,AD} C_{L,AD} \quad (4.2.2)$$

where

$Q_{L,AD}$  is the mass of heavy metal ion sorbed per unit mass of rhamnolipid biosurfactant at equilibrium

$K_{L,AD}$  is the distribution coefficient

$C_{L,AD}$  is the concentration of heavy metal ions at equilibrium (Eriksson, et al., 2004).

The experimental data were plotted for zinc, copper, nickel and cadmium binding onto rhamnolipid biosurfactant according to the linear isotherm model. The data for each metal were fitted by linear lines. The correlation regression coefficients ( $R^2$ ) have been calculated for each metal ion utilizing the least-squares method. Table 4.2.2 shows the correlation regression coefficients ( $R^2$ ) of linear adsorption model for zinc, copper, nickel and cadmium. As illustrated in Table 4.2.2, the linear adsorption model did not fit the data well. Also, the values of the correlation regression coefficients ( $R^2$ ) of the linear adsorption model are higher than

corresponding Langmuir correlation coefficients. Consequently it would seem that the linear adsorption model matches the data better than the Langmuir model but it does not match them well.

**Table 4.2.2 Values of square linear regression ( $R^2$ ) for the linear adsorption model**

<b>Metal ion</b>	<b>Membrane</b>	<b>Square linear regression (<math>R^2</math>)</b>
<b>Zinc</b>	10,000 MWCO	0.7272
<b>Zinc</b>	30,000 MWCO	0.6943
<b>Copper</b>	10,000 MWCO	0.6581
<b>Copper</b>	30,000 MWCO	0.7589
<b>Nickel</b>	10,000 MWCO	0.6508
<b>Nickel</b>	30,000 MWCO	0.7552
<b>Cadmium</b>	10,000 MWCO	0.6011
<b>Cadmium</b>	30,000 MWCO	0.6590

#### **Freundlich isotherm model**

The aim of the work was to determine the portions of metal ions bound to the rhamnolipid biosurfactant and in solution at equilibrium, and to apply the Freundlich isotherm model to each metal ions. The two different UF membranes were used.

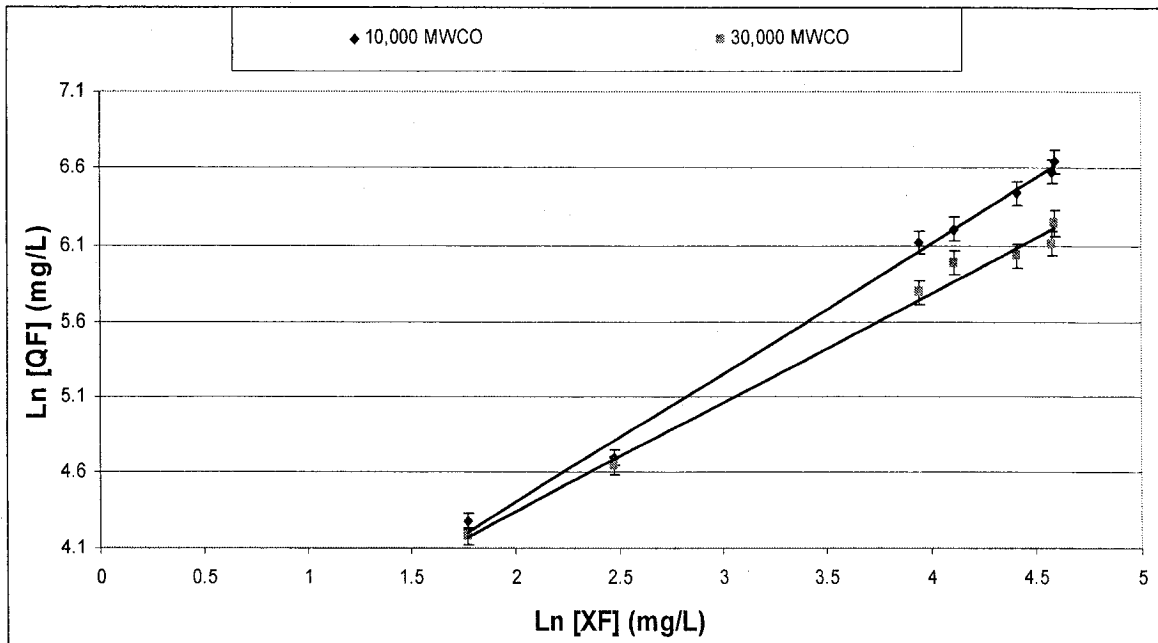
The Freundlich adsorption model is one of the well known models to describe an adsorption isotherm. It describes ideal chemisorption systems. It can be derived on the basis of kinetic, thermodynamic and/or statistical mechanical models (McCash, 2001).

There has been a growing interest in performing binding studies on the interactions of water-soluble polymers with metal ions by ultrafiltration using Freundlich adsorption isotherms (Moreno-Vellosalada and Rivas, 2003). Freundlich equation can be described by the formula

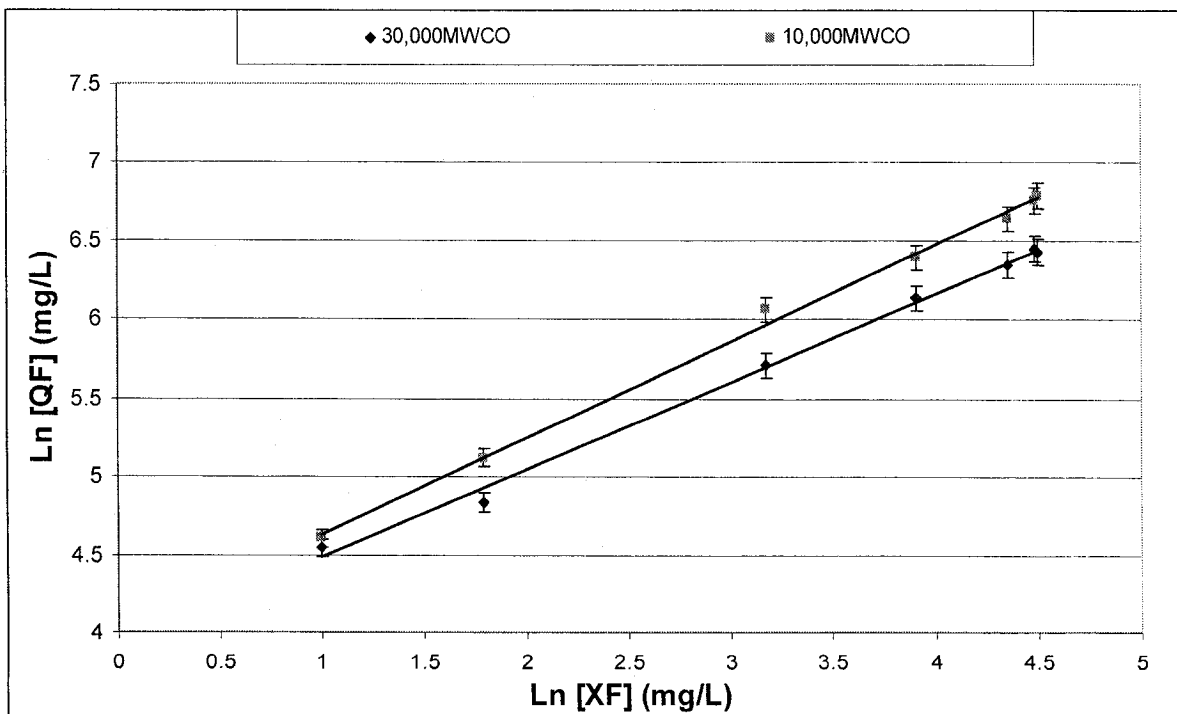
$$[Q_F] = K_F [X_F]^n \quad (4.2.4)$$

The constants  $K_F$  and  $n$  are the characteristics of the Freundlich isotherm equation.  $K_F$  is known to be an indicator of adsorption capacity and  $n$  is considered to express adsorption intensity.  $[Q_F]$  is mass of adsorbate adsorbed per unit mass of adsorbent at equilibrium.  $[X_F]$  is equilibrium concentration of adsorbate in solution at equilibrium (mass/volume) (Helminen et al., 2004). In order to make the fitting of the model to the experimental data easy, equation (4.2.4) can be converted into a linear form :

$$\ln [Q_F] = \ln K_F + (n) \ln [X_F] \quad (4.2.5)$$



**Figure 4.2.1** Freundlich isotherms for zinc onto rhamnolipid for the membranes of 10,000 and 30,000 MWCO, temperature =  $25 \pm 1$  °C and transmembrane pressure = 69 kPa.



**Figure 4.2.2** Freundlich isotherms for copper onto rhamnolipid for the membranes of 10,000 and 30,000 MWCO, temperature =  $25 \pm 1$  °C and transmembrane pressure = 69 kPa.

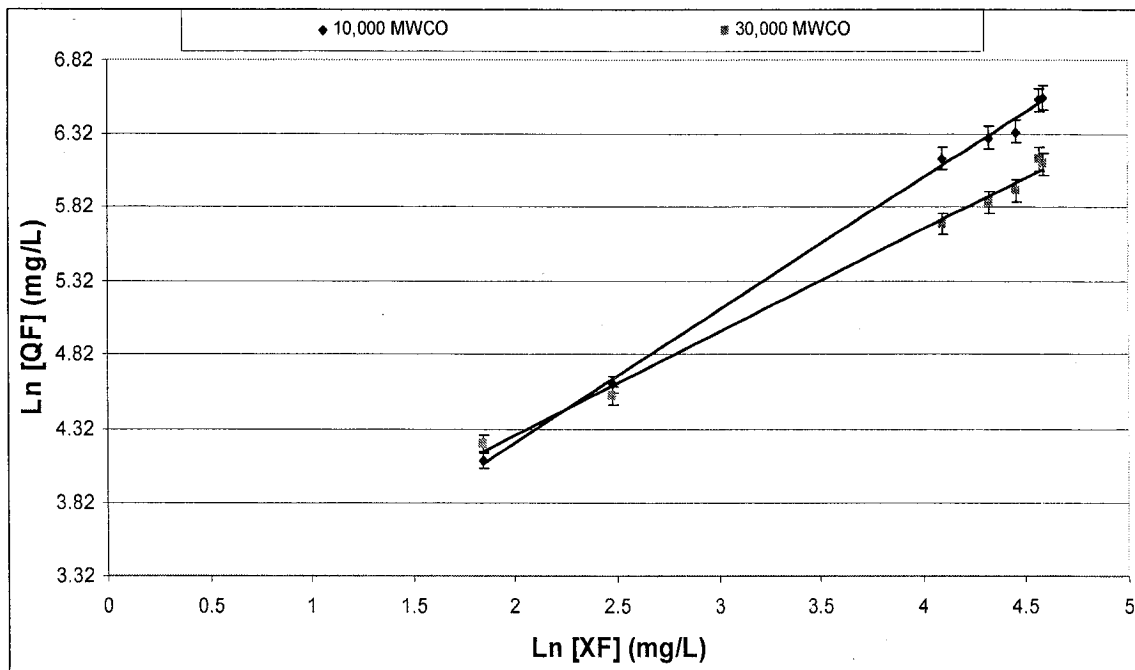


Figure 4.2.3 Freundlich isotherms for nickel onto rhamnolipid for the membranes of 10,000 and 30,000 MWCO, temperature =  $25 \pm 1$  °C and transmembrane pressure = 69 kPa.

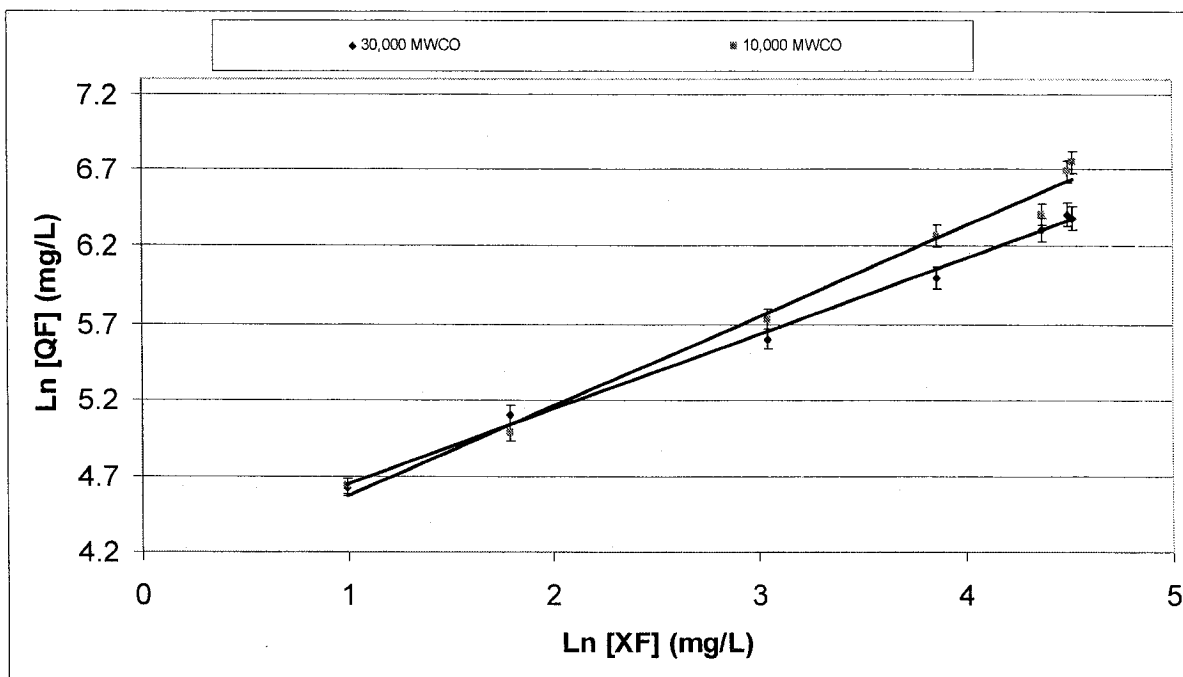


Figure 4.2.4 Freundlich isotherms for cadmium onto rhamnolipid for the membranes of 10,000 and 30,000 MWCO, temperature =  $25 \pm 1$  °C and transmembrane pressure = 69 kPa.



Figures 4.2.1 to 4.2.4 represent Freundlich plots for the experimental data for each metal using the membranes with molecular weight cut-off (MWCO) of 10,000 and 30,000 daltons. In the case of the Freundlich model, it can be seen that the data for each metal are best fitted by straight lines. Utilizing the least-squares method, the correlation regression coefficients ( $R^2$ ) have been quantified for each metal ion.

**Table 4.2.3 Values of  $K_F$  , b, and square linear regression ( $R^2$ ) for the Freundlich model**

<b>Metal ion</b>	<b>Membrane</b>	<b><math>K_F</math></b>	<b>b</b>	<b>Square linear regression (<math>R^2</math>)</b>
<b>Zinc</b>	10,000 MWCO	2.1	18.8	0.9773
<b>Zinc</b>	30,000 MWCO	2.3	9.5	0.9763
<b>Copper</b>	10,000 MWCO	6.1	22.8	0.9796
<b>Copper</b>	30,000 MWCO	4.2	13.9	0.9950
<b>Nickel</b>	10,000 MWCO	1.6	17.1	0.9829
<b>Nickel</b>	30,000 MWCO	1.3	8.6	0.9773
<b>Cadmium</b>	10,000 MWCO	4.6	21.9	0.9704
<b>Cadmium</b>	30,000 MWCO	2.3	12.6	0.9855

Table 4.2.3 presents values of  $K_F$ ,  $b$ , and the correlation regression coefficients ( $R^2$ ) of Freundlich isotherm model for zinc, copper, nickel and cadmium. As illustrated in Figures 4.2.1 to 4.2.4, the Freundlich isotherm model fits the data well. Additionally,  $K_F$ , and  $b$  values of Freundlich isotherm model in this study are consistent with the ones reported in other work (Moreno-Vellosalada and Rivas, 2003). It can be observed in Table 4.2.3 that the values of the correlation regression coefficients ( $R^2$ ) of Freundlich model are within the range from 0.9704 to 0.9950.

Similarly, it can be noticed in Tables 4.2.1, 4.2.2 and 4.2.3 that the values of the correlation regression coefficients ( $R^2$ ) of Freundlich model are significantly higher than the corresponding linear adsorption. Consequently it would appear that the Freundlich isotherm model fits the experimental isotherm data better. Furthermore, it can be seen from Table 4.2.3 that the highest correlation regression coefficients ( $R^2$ ) of Freundlich model are for copper by the membrane of 30,000 MWCO, while the lowest ones were for cadmium by the membrane of 10,000 MWCO. This may give an indication that better representation of the experimental data by Freundlich model were for copper.

### **Summary**

In this work, the equilibrium isotherms were determined for the single-component experimental system. The data have been analyzed for each metal ion using

langmuir, Freundlich, and linear adsorption model. The Freundlich isotherm model was shown to provide the best fit. The  $K_F$  values were found to be from 1.6 to 6.1 and from 1.3 to 4.2 for the membranes with molecular weight cut off (MWCO) of 10,000 Da and 30,000 Da, respectively. In addition, the  $b$  values were found to be from 17.1 to 22.8 and from 8.6 to 13.9 for the membranes with molecular weight cut off (MWCO) of 10,000 Da and 30,000 Da, respectively.

### **4.3 Applicability of Gouy-Chapman model on rhamnolipid enhanced UF**

The interaction of metals with colloids and electrolytes forming surfactant/counterion equilibrium, has been the subject of recent experimental research (Huang et al., 2005; Kim et al., 2004; Evans and Wennerstroem, 1999). Some surfactant types have been reported for their metal-sorption capacity (Huang et al., 2005; Evans and Wennerstroem, 1999). The promising use of MEUF technology in the treatment of metal-bearing solutions is mainly due to their low costs and high efficiency of removing metals present in low concentrations from polluted wastewater and groundwater contaminated with heavy metals (Huang et al., 2005; Kim et al., 2004). Despite this, applications of biosurfactants in metal-removal systems using MEUF are rare in the field of wastewater and ground water treatment for several reasons. First is to achieve demonstrations of product efficacy. Then, the new microbial product must be produced at an economical scale and yield. Further, it must meet regulatory requirements (O'Connor, 2002; Maier and Soberon-Chavez, 2000).

The Gouy-Chapman theory is known to be one of those most widely applicable on the association of metal ions with colloids and amphiphiles (Evans and Wennerstroem, 1999). An illustration of the basic Gouy-Chapman approach is shown in Figure 4.3.1. In general, all forms of the Gouy-Chapman approach have the common assumption that the charged micelle surface (the first layer) is planar and that no specific metal ion binding occurs. In addition, it is proposed

that all charges are assumed as point (i.e., infinitesimal) charges. The absolute value of the change in the potential gradient in the diffuse layer (the second layer) decreases exponentially, as does the metal ion concentration (Evans and Wennerstroem, 1999; Huang and Ruckenstein, 2004).

The potential and concentration profiles of ions in a double layer can be explained in different ways (Evans and Wennerstroem, 1999; Huang and Ruckenstein, 2004). The Poisson-Boltzmann method used in the Gouy-Chapman approach is one of the better-known possible explanations. The major advantage of the Gouy-Chapman approach is that it can be derived from basic electrochemical principles.

The goal of this work was to investigate the applicability of Gouy-Chapman approach for binding of heavy metal ions to rhamnolipid biosurfactant in a MEUF system based on a charge-to-charge interaction. The Poisson-Boltzmann equation was utilized for this purpose. The electrostatic approach was considered for this kind of interactions among the positive charges from some heavy metal cations and the negative charge of the carboxyl groups of rhamnolipid biosurfactants.

### **Applicability of Gouy-Chapman approach**

Assumptions that are made to apply the Poisson-Boltzmann method include: (1) the permittivity,  $\epsilon$ , is independent of position, (2) the charged surface is an

equipotential surface, and (3) at equilibrium, the electrochemical potential of ions is constant everywhere (Evans and Wennerstroem, 1999; Huang and Ruckenstein, 2004). The first two assumptions were applied mainly to Poisson's equation, and the last one was applied mainly to Boltzmann's equation. In addition, when the Poisson-Boltzmann equation is applied to rhamnolipid biosurfactants, the rhamnolipid biosurfactant concentration in solution is assumed to be small, such that the interactions between the charged particles can be neglected. Consequently, modeling a single micelle is adequate to model the collective group of micelles in solution as each one is proposed to be identical.

### **Applicability of Poisson-Boltzmann method**

If we take into consideration a portion of solution in which the counterions, rhamnolipid monomers and rhamnolipid micelles are distributed in a uniform density  $\rho^*$ , then the force between two charged particles  $\bar{F}_{1 \rightarrow 2}$  can be expressed as:

$$\bar{F}_{1 \rightarrow 2} = q_2 \bar{E}(r) = \frac{q_2 q_1 r}{4\pi \epsilon r^2} \quad (\text{Evans and Wennerstroem, 1999}) \quad (4.3.1)$$

where  $\bar{E}$  is electric field intensity,  $\epsilon$  is equal to  $(\epsilon_0 D)$ ,  $\epsilon_0$  is permittivity of free space ( $8.854 \times 10^{-14} \text{ C v}^{-1} \text{ cm}^{-1}$ ),  $D$  is dielectric constant (78.5 for water at 25 °C) and  $r$  is the radius of a rhamnolipid micelle.

Appendix D contains the derivations of equation 4.3.24 from equation 4.3.1.

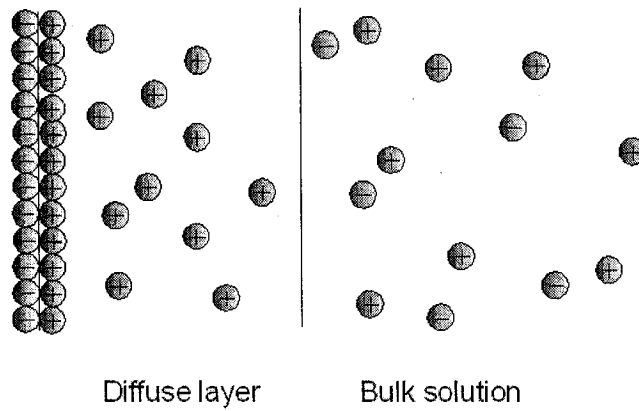
equation (4.3.24) is expressed as:

$$\frac{\sigma^2}{2\epsilon kT} = \frac{2KT}{\epsilon} \sum_i C_i N * 1000 [\exp(\frac{-z_i e \psi_o}{kT}) - 1] \quad (4.3.24)$$

where N is Avogadro's number ( $6.022 \times 10^{23}$  / mole), and  $z_i$  is the valence of ions i. If the surface charge density is known, equation (4.3.24) can be used to calculate the surface potential ( $\psi_o$ ).

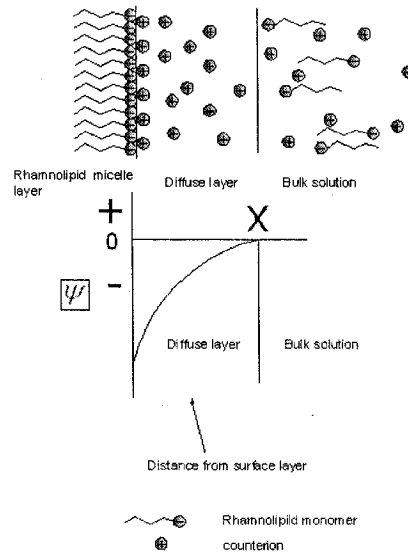
The Gouy-Chapman approach can provide an explanation of the metal-biosurfactant binding throughout the rhamnolipid micelles Figure 4.3.1 shows an illustration of the basic Gouy-Chapman approach (Evans and Wennerstroem, 1999). An illustration of the proposed Gouy-Chapman approach for the case of rhamnolipid biosurfactant micelles with metal-cations is shown in Figure 4.3.2. Considered as a specific instance of the general Gouy-Chapmann method, we can assume that the charged micelle surface (the first layer) is planar and no specific counterion binding occurs. All charges are assumed to be point charges. The absolute value of the change in the potential gradient in the diffuse layer (the second layer) decreases exponentially as does the counterion concentration.

# Gouy-Chapman theory



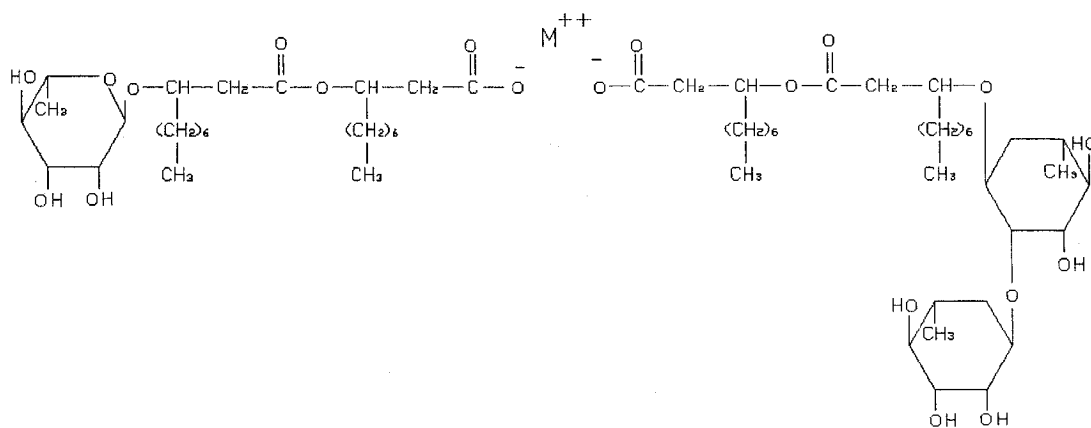
**Figure 4.3.1** Illustration of the basic Gouy-Chapman approach (Evans and Wennerstroem, 1999).

## Gouy-Chapman diffuse layer approach

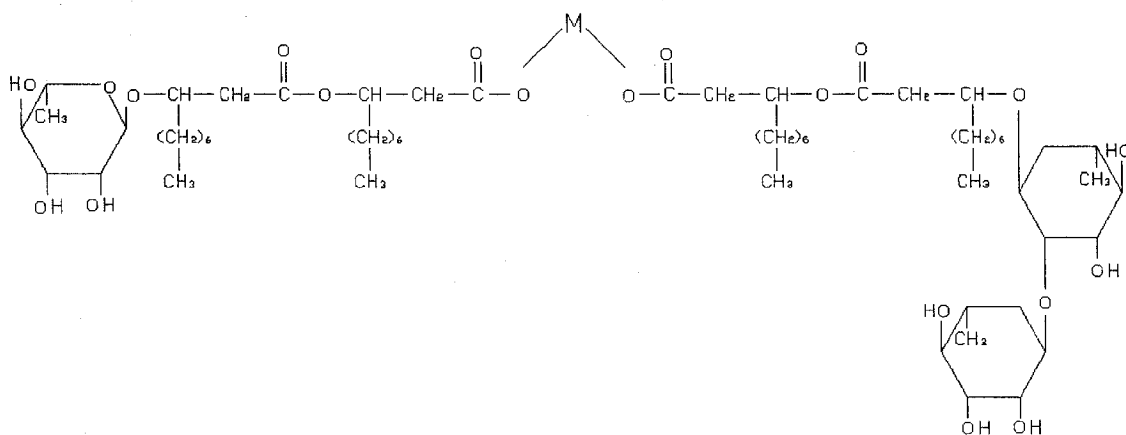


**Figure 4.3.2** Illustration of the proposed Gouy-Chapman approach for the case of rhamnolipid biosurfactant micelles with metal-cations.





**Figure 4.3.3 Structure of rhamnolipid biosurfactants which possess negatively charged carboxyl groups.**



**Figure 4.3.4 Illustration of the proposed interaction between two rhamnolipid biosurfactant molecules and one metal-cation.**

Principally, the relative binding affinity of some metal ions for characteristic chemical groups in surfactants play an important role in their electrostatic interactions (Takao et al., 2004). As seen in Figure 2.9 (Chapter 2), Figure 4.3.3 and Figure 4.3.4, the rhamnolipid has two carboxyl groups which allow it to form a dimer stabilized by the metal ions. It appears that the rhamnolipid biosurfactants which possess negatively charged carboxyl groups have affinities to cadmium, lead, zinc, copper and nickel ions utilized in this research. Thus, it is proposed that each one metal cation binds with two rhamnolipid molecules (Figure 4.3.3 and Figure 4.3.4). If the electrostatic approach for the interactions between the positive charges of the heavy-metal cations and the negative charges of the carboxyl groups of rhamnolipid biosurfactant was considered, then the Gouy-Chapman approach explains the metal-biosurfactant binding throughout the rhamnolipid micelles in the MEUF system by evoking a charge-to-charge interaction. This assumes that the charged rhamnolipid micelle surface (the first layer) is planar. This means that, according to the Gouy-Chapman approach, the biosurfactant-to-metal molar ratios of 2:1 should be applicable to the MEUF system. The results from lab-scale studies showed that this ratio, suggested by the Gouy-Chapman approach, was relevant. The removal of the metal ions present in contaminated industrial wastewater and groundwater were achieved (> 99% rejection ratio) by applying optimal conditions that include biosurfactant-to-metal molar ratios of 2:1.

## **Summary**

The heavy metal / rhamnolipid interaction was successfully investigated using the Poisson-Boltzmann equation according to the Gouy-Chapman approach. The electrostatic approach was considered for this kind of interactions among the positive charges from heavy metal cations and the negative charges of the carboxyl groups of rhamnolipid biosurfactants. The results from lab-scale investigations support the hypothesis that the Gouy-Chapman approach was valid.

## 4.4 Wastewater treatment by ultrafiltration

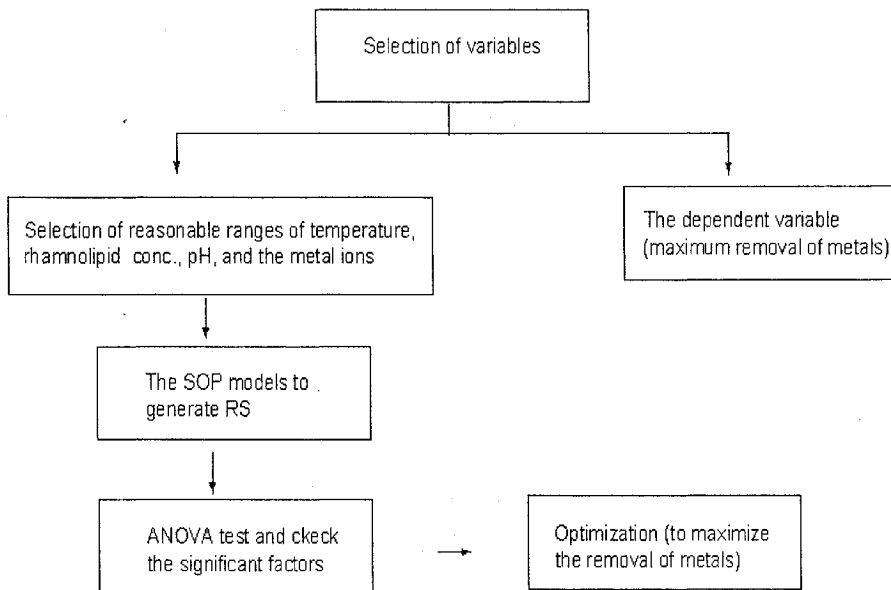
In this work, a rhamnolipid-based ultrafiltration technique was examined with the aim of determining its performance at the removal of heavy metal ions from wastewaters. Subsequently, the results from bench-scale MEUF experiments, which indicated optimized operating conditions, were extrapolated for use on industrial wastewater samples as filtered by two UF membranes (MWCO 10,000 and 30,000).

### **Treatment of heavy metal-contaminated industrial wastewaters**

A laboratory-scale UF system enhanced by rhamnolipid addition was designed and tested to assess its effectiveness at treating industrial wastewater from a metal refinery in Quebec, Canada. In order to efficiently achieve the maximum removal of metals from the industrial wastewaters, optimization by the response surface methodology approach was utilized. The pH, flux, and temperature were considered as independent variables while maximum removal of metals from the industrial wastewaters was designated as a dependent variable. Second order polynomial (SOP) models were utilized as functions of the independent variables. An analysis of variance (ANOVA) was performed to determine which of the parameters significantly affected the dependent variable.

Response surface and contour plots were generated as shown in Figures 4.4.2 and 4.4.3. Optimum regions of independent and dependent variables were achieved. The major objective was to maximize the removal of cadmium and

lead from the ground waters. From Figures 4.4.2 and 4.4.3, it can be seen that the suggested range of temperature was from about 24 to 26 °C, and the suggested range of pH values was within about 6.7 to 7.2. Also, the suggested flux range was from about 51 to 122 L/m<sup>2</sup>/h.



SOP = Second order polynomial (SOP); RS = Response surface

Conc. = Concentration

**Figure 4.4.1 Experimental data flow to achieve the maximum rejection of heavy metal ions from the industrial wastewaters.**

( Max. Rej. Cd and Pb )  
 X = C: pH  
 Y = D: Temperature (Deg. C.)

Actual Factors  
 A: Flux (L/M. sq./h) = 72.50

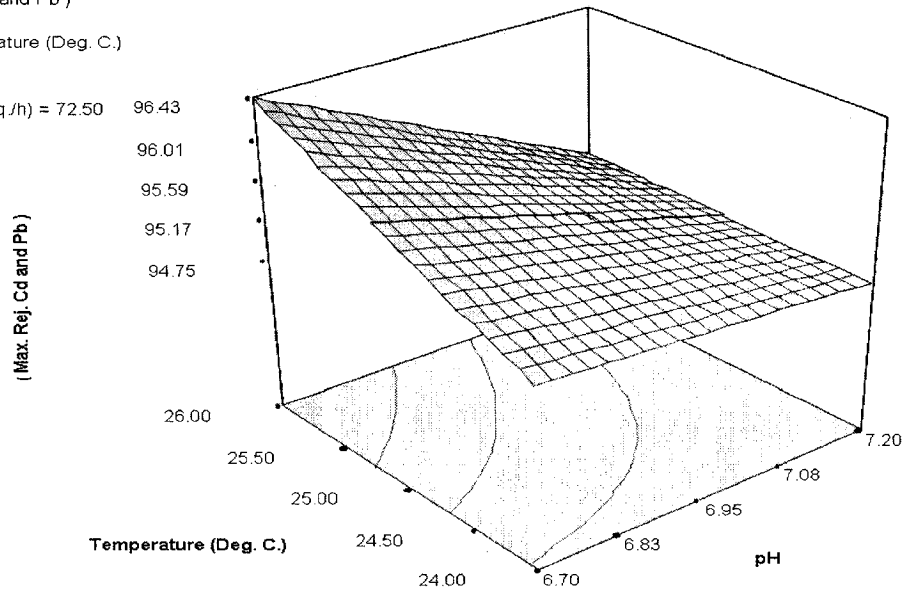


Figure 4.4.2 Response surface obtained for temperature and pH.

( Max. Rej. Cd and Pb )  
 X = D: Temperature (Deg. C.)  
 Y = A: Flux (L/M. sq./h)

Actual Factors  
 C: pH = 6.95

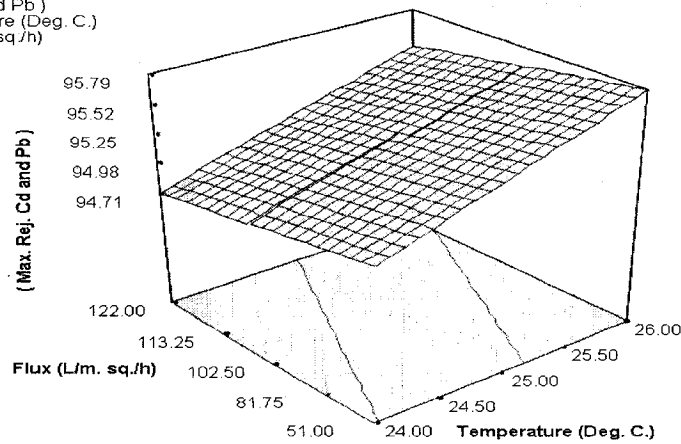
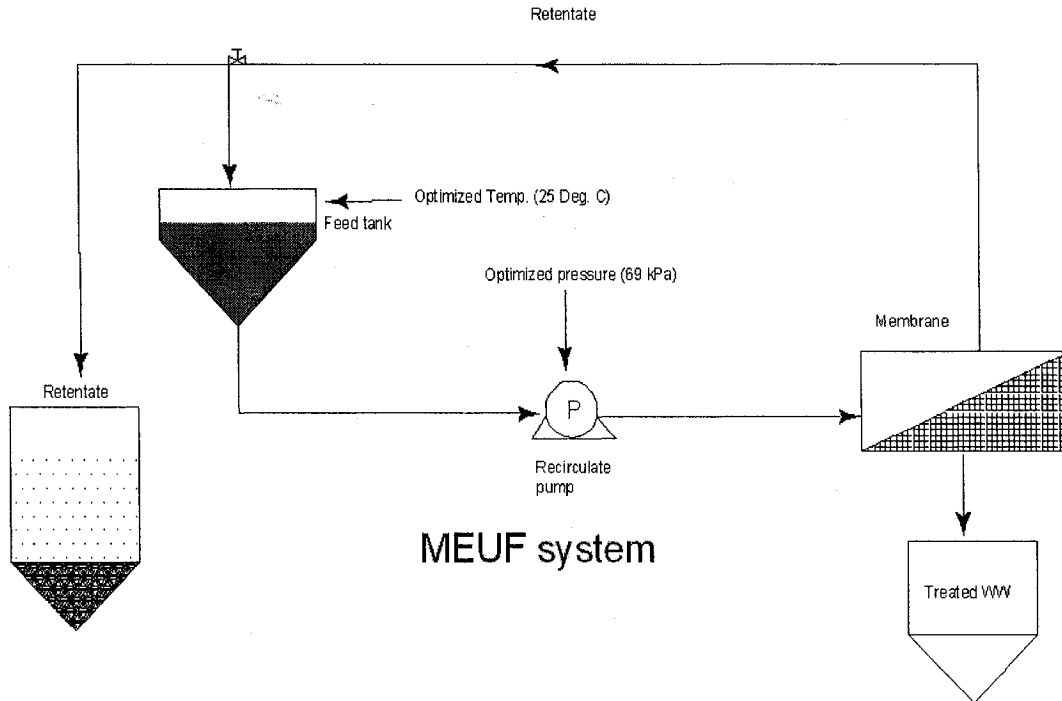


Figure 4.4.3. Response surface obtained for pH and flux.

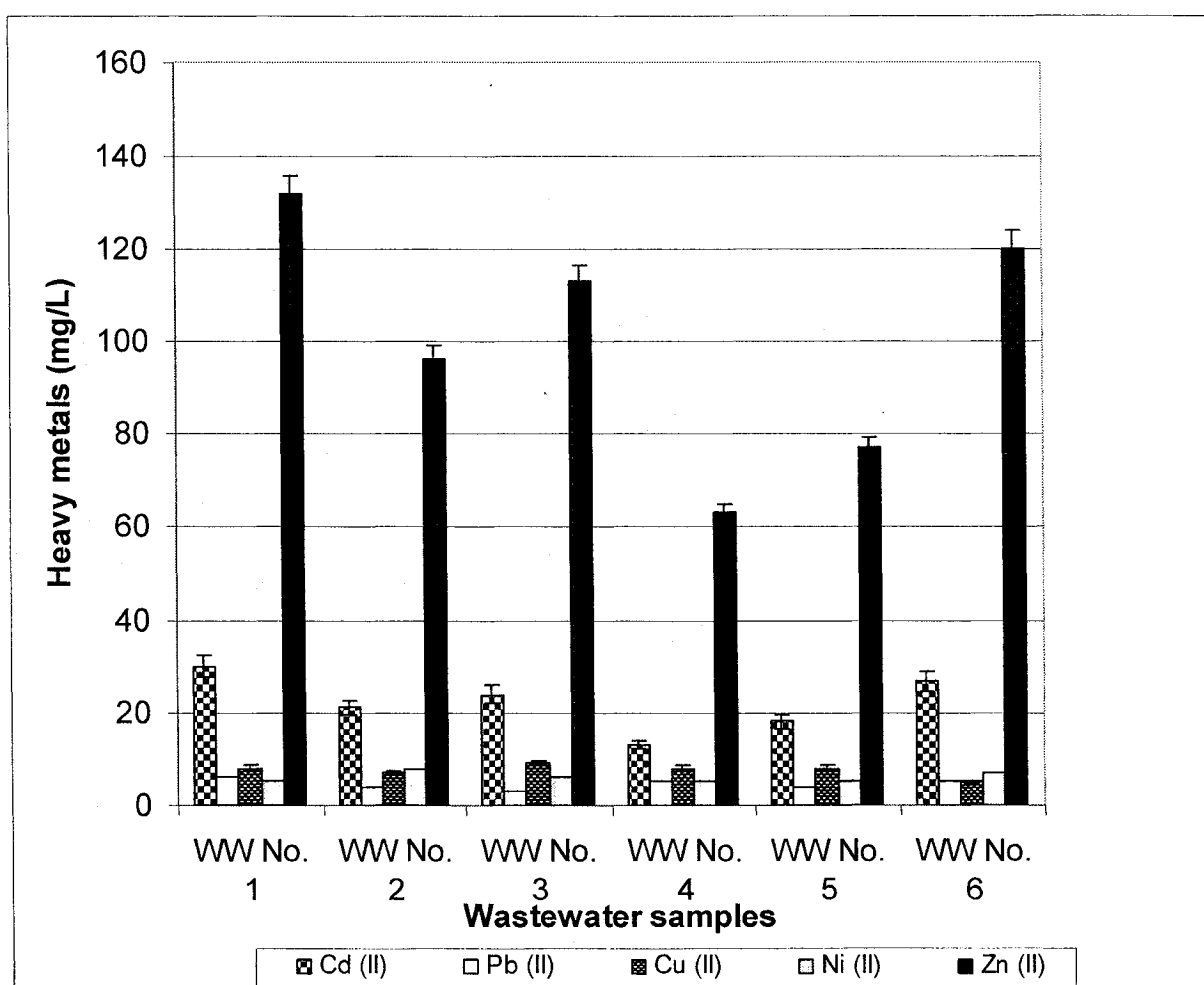


**Figure 4.4.4 Permeate as a safe wastewater from a MEUF system**

Separate lab-scale validation experiments using the lab-scale UF system were performed to verify these optimum conditions. The most applicable operating conditions were biosurfactant-to-metal molar ratios of 2:1, temperature of  $25 \pm 1^\circ\text{C}$ , and pH of  $6.9 \pm 0.1$ . The MEUF system shown in Figure 4.1.2 was used here.

Six wastewaters were collected from a metal refining in Quebec, Canada and utilized in this study. In the plant, wastewaters are generally treated by conventional wastewater treatment techniques such as sedimentation, filtration,

coagulation and pH adjustment in order to remove large suspended solids. Before performing UF runs and determination of heavy metal analysis at Concordia University, the suspended solids above 45  $\mu\text{m}$  in the samples were removed by filtration to remove suspended solids that could not be removed by sedimentation in the plant before collecting the samples. Figure 4.4.5 shows the typical characteristics of the raw feed wastewaters performed.



**Figure 4.4.5 Typical characteristics of the raw feed wastewater.**



A survey of Canadian surface finishing industries was performed in 1983 (EPS, 1987). It was reported that 275 companies (128 in Quebec) discharged a total of 1,364 m<sup>3</sup> of wastewater. Table 4.4.1 shows the federal discharge limits for heavy metals (EPS, 1987).

**Table 4.4.1 Federal discharge limits for heavy metals (EPS, 1987)**

<b>Metal</b>	<b>Canadian discharge limits (mg/L)</b>
Pb	1.5
Cd	1.5
Zn	2.0
Ni	2.0
Cu	1.0

Several trial runs with optimized ranges for the lab-scale technique were performed in order to provide the maximum rejection of heavy metal ions and maximum water recovery ratio as permeates. The membranes were with molecular weight cut-off (MWCO) of 10,000 and 30,000 Daltons. Table 4.4.2 shows some operating parameters of the six wastewaters. It shows also that the pH values of the feeds and retentates were monitored.

Figure 4.4.6 shows the permeate fluxes of membranes with molecular weight cut-off (MWCO) of 10,000 and 30,000 Daltons.

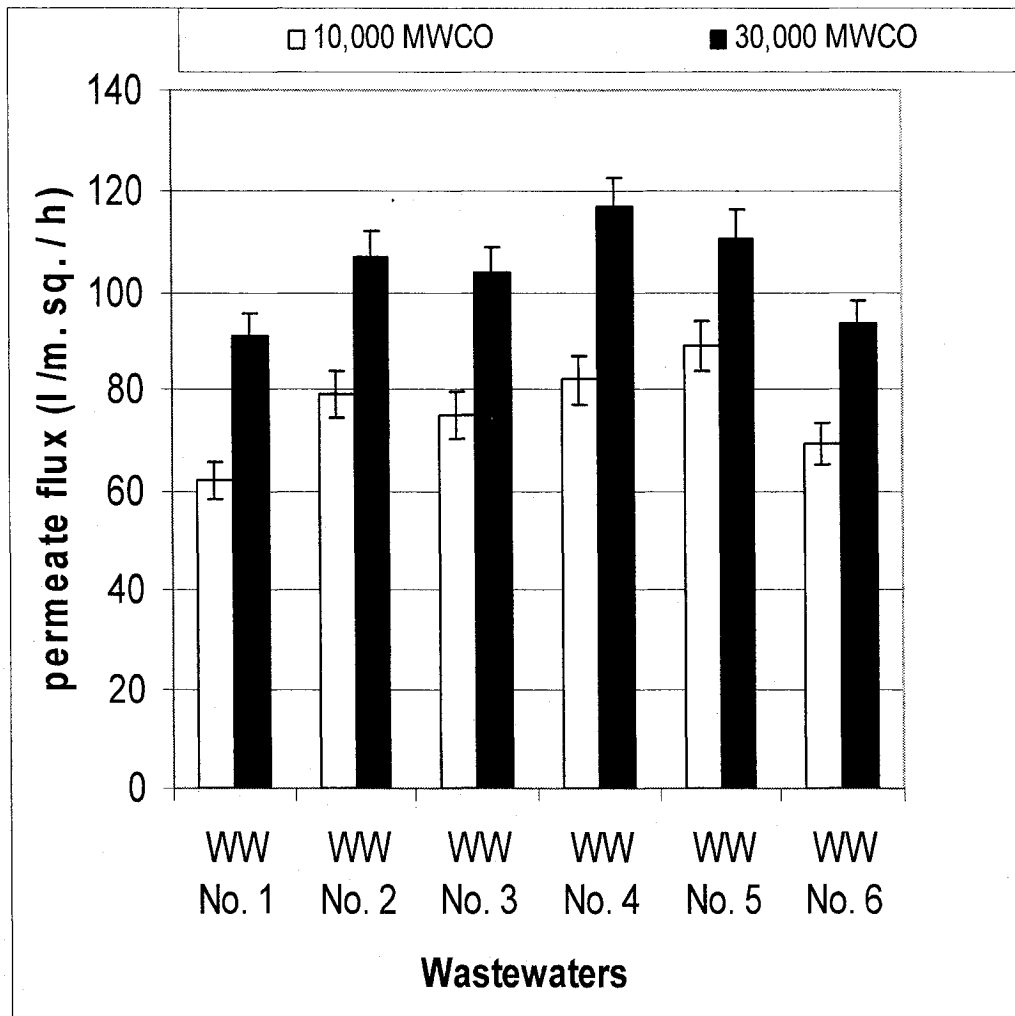
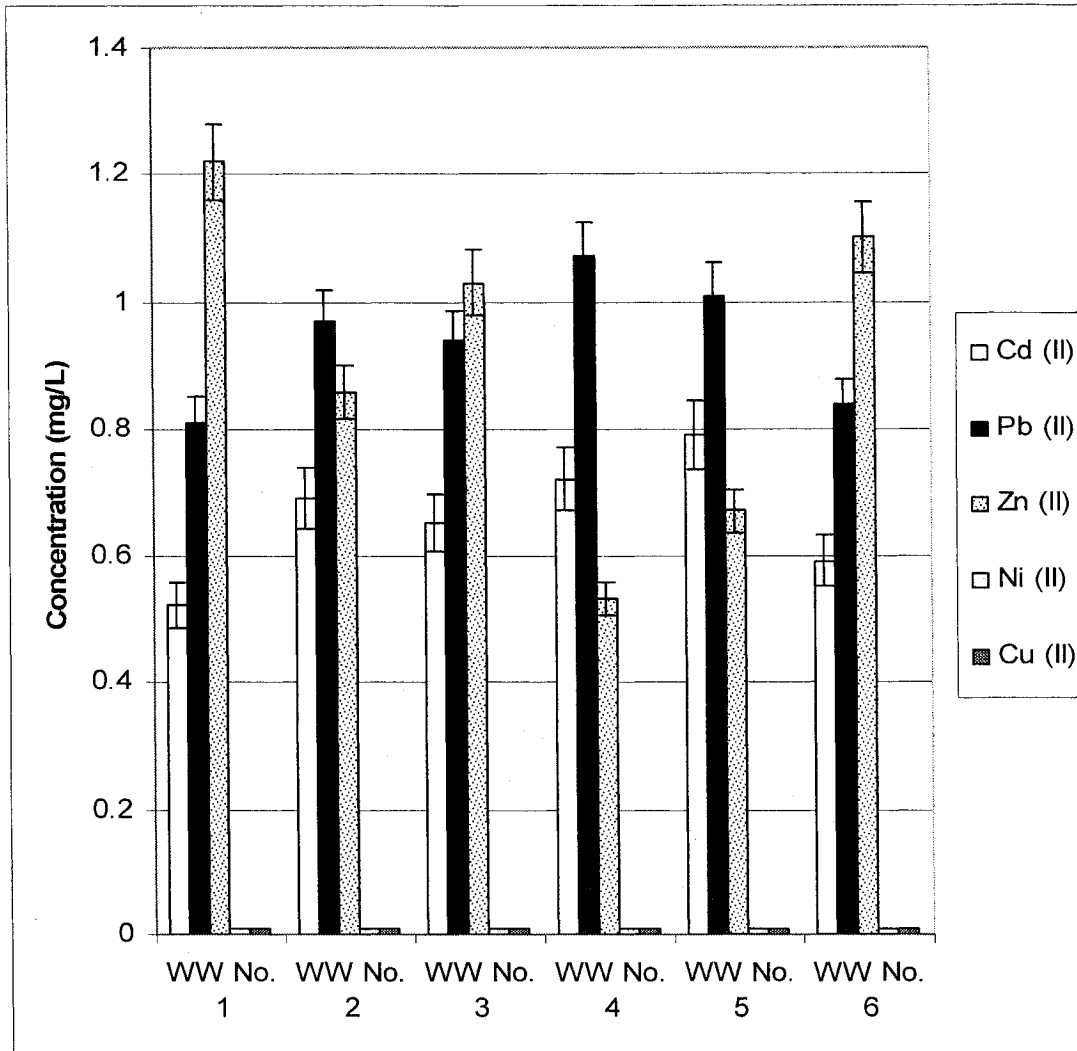


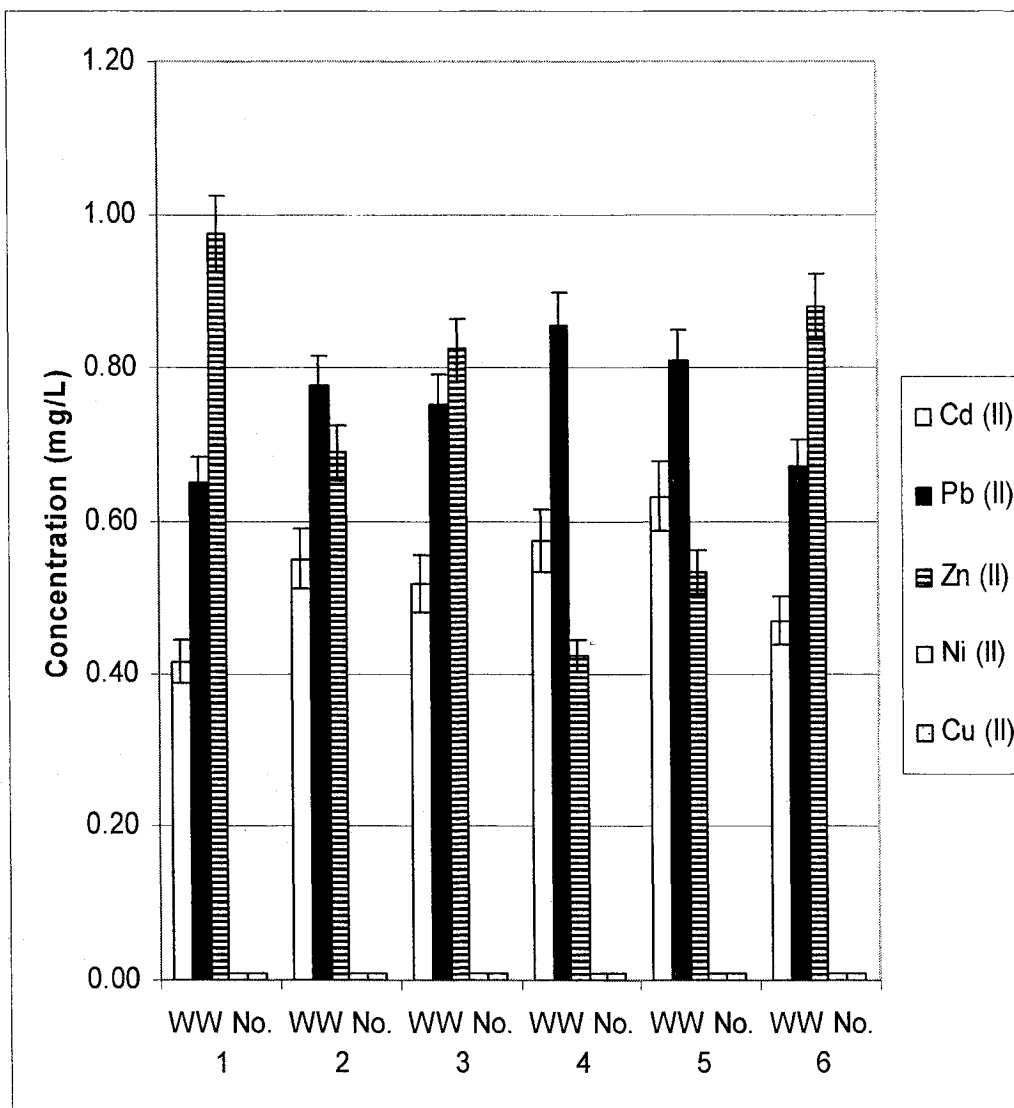
Figure 4.4.6 Permeate fluxes of membranes with MWCO of 10,000 and 30,000 Da.

It was confirmed by preliminary trials that when the rhamnolipid-based UF unit was operated at higher flux recovery 85% to 90% water recovery for the few hours (2-5 h), serious reduction in permeate fluxes of the two membranes resulted.



**Figure 4.4.7 Characteristics of the permeate quality of the wastewaters using the membrane with MWCO of 30,000 daltons.**

This may be due to the concentration polarization effect. When the micelles or metal-rhamnolipid complexes are rejected by the ultrafiltration membranes, they usually form the highly viscous boundary layer adjacent to the membrane surface which is called a gel layer causing the flux decline.



**Figure 4.4.8 Characteristics of the permeate quality of the wastewaters using the membrane with MWCO of 10,000 daltons.**

**Table 4.4.2 Effect of some operating parameters on the wastewaters**

<b>Parameter</b>	<b>WW 1</b>	<b>WW 2</b>	<b>WW 3</b>	<b>WW 4</b>	<b>WW 5</b>	<b>WW 6</b>
<b>Water recovery in permeate (%)</b>	73	73	73	73	73	73
<b>Transmembrane pressure (kPa)</b>	55	58	58	58	58	55
<b>Adjusted pH (feed)</b>	7.0	7.1	7.0	7.1	7.0	7.0
<b>pH (permeate)</b>	6.5	6.6	6.6	6.5	6.5	6.6

However, the permeate quality had to meet the Canadian discharge limits, at the same time, maximum permeate flux had to be produced. Thus, trial runs for continuous operation of the rhamnolipid-based UF unit at different pressures and permeate recoveries were conducted to discover the fouling tendency of the two membranes.

**Table 4.4.3 Some chemical parameters of the wastewater**

<b>Parameter</b>	<b>WW 1</b>	<b>WW 2</b>	<b>WW 3</b>	<b>WW 4</b>	<b>WW 5</b>	<b>WW 6</b>
<b>COD mg/L (feed)</b>	300 ±12.0	163 ±9.0	174 ±9.0	113 ±8.0	143 ±9.0	298 ±13.0
<b>pH (feed)</b>	6.1 ±0.1	5.8 ±0.1	5.4 ±0.1	5.3 ±0.1	5.6 ±0.1	5.9 ±0.1
<b>Adjusted pH (feed)</b>	7.0 ±0.1	7.1 ±0.1	7.0 ±0.1	7.1 ±0.1	7.0 ±0.1	7.0 ±0.1
<b>pH (10,000 MWCO, Permeate)</b>	6.5 ±0.1	6.6 ±0.1	6.6 ±0.1	6.5 ±0.1	6.5 ±0.1	6.6 ±0.1
<b>pH (30,000 MWCO, Permeate)</b>	6.6 ±0.1	6.5 ±0.1	6.4 ±0.1	6.6 ±0.1	6.6 ±0.1	6.5 ±0.1
<b>COD (30,000 MWCO, Permeate)</b>	36 ±2	21 ±2	23 ±1	19 ±2	18 ±1	37 ±2
<b>COD (10,000 MWCO, Permeate)</b>	32±2	17 ±2	18 ±2	16 ±1	16 ±2	30 ±2

The results are illustrated in Table 4.4.2. After several trial runs, it appeared that the UF membranes showed the lowest fouling tendency at 73% water recovery as permeate. Therefore, 73% water recovery as permeate was selected for the runs. Similarly, it can be seen from Figures 4.5.2 and 4.5.3 that the suggested flux range was from about 51 to 122 L/m<sup>2</sup>/h. According to the trial runs, the UF membranes showed the lowest fouling tendency at flux range from about 60 to 120 L/m<sup>2</sup>/h and at transmembrane pressure range from about 55 to 58 (kPa) . Therefore, these ranges were selected for the actual runs (Table 4.4.2). Table 4.4.3 shows that the COD in the permeates are much lower than those in the feed. This indicates, probably, that much of chemical organic matter in the feed precipitated and/or solubilized by the micelles as well and remain in the retentate, and then rejected by the two membranes (Appendix B).

Figures 5.4.7 and 5.4.8 shows the characteristics of the permeate quality of the wastewaters. It can be seen from the results in Table 4.4.1, Figure 4.5.7, and Figure 4.5.8 that the levels of heavy metal concentrations in the permeates are less than the Federal discharge limits for the metals under investigation. This can be explained by taking into account the high affinity of rhamnolipid biosurfactant to Cu<sup>+2</sup> , Zn<sup>+2</sup> , Ni<sup>+2</sup> ,Cd<sup>+2</sup> and Pb<sup>+2</sup> ions. Since it has been shown that rhamnolipid biosurfactant has a high affinity to Pb<sup>+2</sup> and Cd<sup>+2</sup> ions (Chapter 4, section 1). Thus, those metals in all contaminated industrial wastewaters under investigation can easily be bound onto the rhamnolipid micelles resulting in a

clean permeate from the two UF membranes. Consequently, it was concluded that the rhamnolipid-based ultrafiltration technique is an efficient technique for removal of cadmium (Cd) and lead (Pb) copper (Cu), zinc (Zn) and nickel (Ni) ions from the contaminated industrial wastewaters using the two membranes with (MWCO) of 10,000 and 30,000 daltons. The results produced from bench-scale MEUF experiments as well as the optimized operation conditions were helpful tools to improve the performance of the rhamnolipid-based ultrafiltration system regarding the contaminated industrial wastewaters from the metal refining. The results of these wastewaters were compatible with lab results. Using the lab-optimum conditions for MEUF system, the increased COD values did not reduce the ability of the heavy metals to be removed.

### **Summary**

The rhamnolipid-enhanced ultrafiltration system was utilized to treat samples of six actually contaminated wastewaters from metal refining industries using membranes with molecular weight cut off (MWCO) of 30,000 Da and 10,000 Da. The resulting heavy metal concentrations in the permeate were all significantly reduced to be in accordance with the federal regulations. The UF membranes showed the lowest fouling tendency at 73% water recovery as permeate.



## 4.5 Groundwater treatment by ultrafiltration

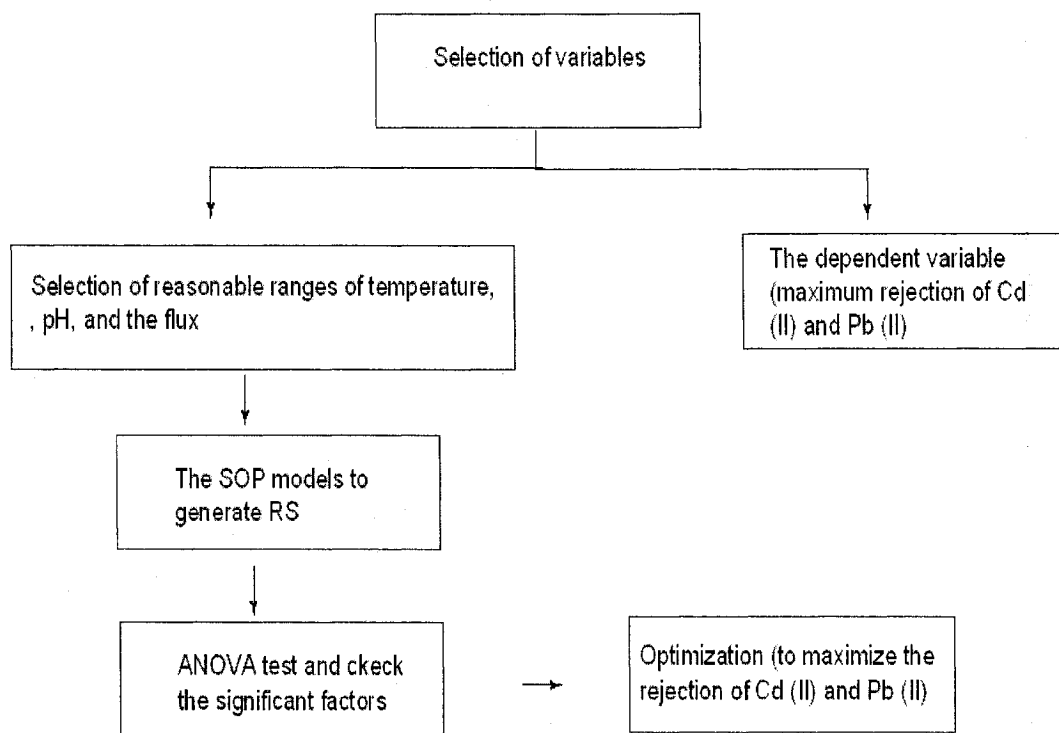
The main objective of this work was to investigate the capability and performance of a rhamnolipid-based ultrafiltration technique for removal of heavy metal ions from groundwater using two UF membranes (MWCO 10,000 and 30,000). Bench-scale MEUF experiments were used to confirm the optimized operating conditions (as shown in section 4.1).

### **Ultrafiltration (UF) for heavy metal-contaminated groundwaters**

Three samples of groundwater from Quebec and Ontario were utilized in this study. The results obtained from bench-scale investigations for rhamnolipid enhanced UF under the optimum operating conditions were used to investigate the treatment of heavy metal contaminated groundwater. The heavy metals were cadmium ( $\text{Cd}^{+2}$ ) and lead ( $\text{Pb}^{+2}$ ), individually and collectively. In order to investigate the possibility of interference from a competition effect with calcium ( $\text{Ca}^{+2}$ ), three different groundwaters with varying levels of calcium were selected. The optimum operating conditions were applied (section 4.1).

In order to efficiently achieve the maximum removal of cadmium and lead from the groundwaters, optimization by the response surface methodology approach was utilized. The pH, flux, and temperature were considered as independent variables while maximum removal of cadmium and lead from

the groundwater was designated as a dependent variable. Second order polynomial (SOP) models were utilized as functions of the independent variables. An analysis of variance (ANOVA) was performed to determine which of the parameters significantly affected the dependent variable. Figure 4.5.1 shows the experimental data flow to achieve the maximum removal of cadmium and lead from the groundwaters.



SOP = Second order polynomial (SOP); RS = Response surface

Conc. = Concentration

**Figure 4.5.1 Experimental data flow to achieve the maximum rejection of heavy metal ions from the groundwater.**

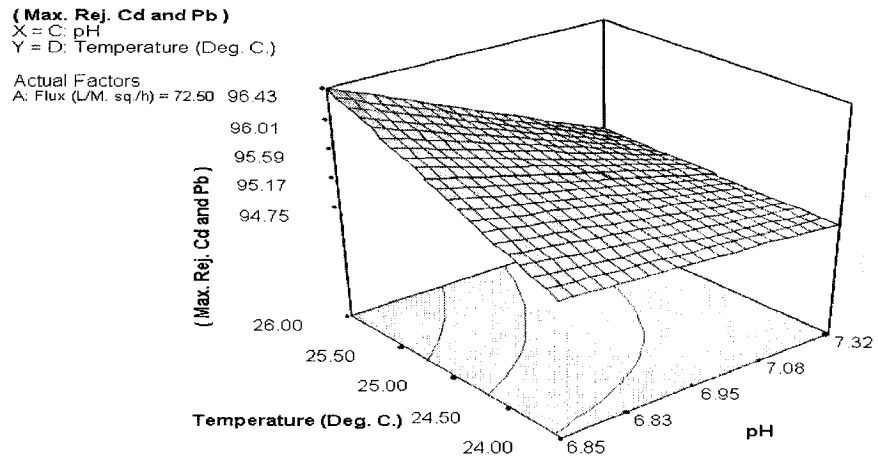


Figure 4.5.2 Response surface obtained for pH and temperature

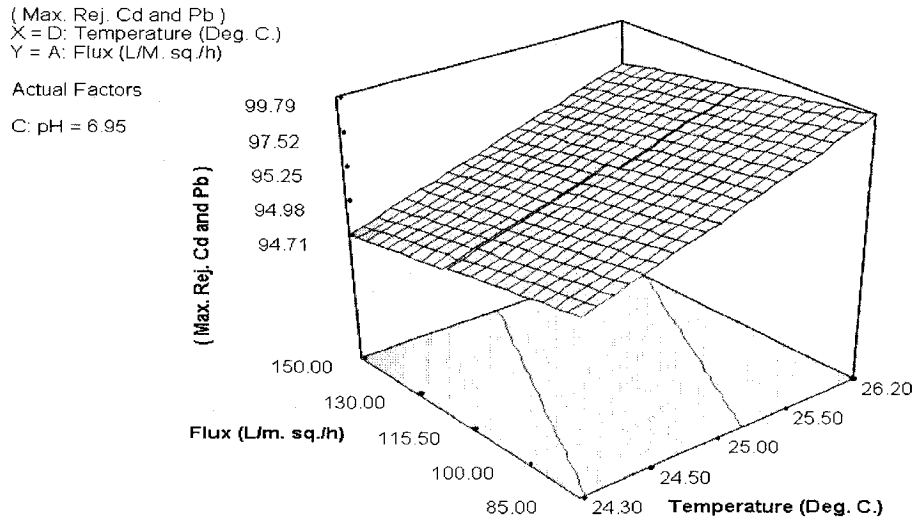


Figure 4.5.3 Response surface obtained for temperature and flux

Response surface and contour plots were generated as shown in Figures 4.5.2 and 4.5.3. Optimum regions of independent and dependent variables were achieved. The major objective was to maximize the removal of cadmium and lead from the groundwater. From Figures 4.5.2 and 4.5.3, it can be seen that the suggested temperature was 24.3 to 26.2 °C, and the suggested ranges of pH were within about 6.9 to 7.3.

Separate validation experiments using the lab-scale UF system were performed to verify these optimum conditions. The most applicable operating conditions were a transmembrane pressure of  $69 \pm 2$  kPa, biosurfactant-to-metal molar ratios of 2:1, temperature of  $25 \pm 1$  °C, and pH of  $7.0 \pm 0.1$ .

The first and the third groundwater samples were from Quebec (Canada). The second one was from Ontario (Canada). The calcium content was 25, 45 and 86 mg/L, respectively. Table 4.5.1 shows the characteristics of the first, second and third groundwater samples, respectively. Table 4.5.2 presents values of pH and COD for the three groundwaters. The metal analysis and COD tests were performed at the Environmental Engineering lab, Concordia University. The values of other water quality parameters were provided by personal communications (Nov. 2004).

**Table 4.5.1 Characteristics of the three groundwater samples that studied**

Parameters	Value (mg/L) in GW <sup>1</sup> 1	Value (mg/L) in GW2	Value (mg/L) in GW3
Calcium (II)	25 ± 1	48 ± 3.1	86 ± 3.0
Chloride (I)	53 ± 2	6 ± 0.5	82 ± 2.6
Iron (II)	N.D. <sup>2</sup>	N.D.	N.D.
Bicarbonates	68 ± 2.8	232 ± 13.0	250 ± 2.0
COD	23 ± 2.0	27 ± 3.0	20 ± 2.0
Magnesium (II)	8 ± 0.1	23 ± 1.8	23 ± 2.0
Sodium (I)	32 ± 1.2	4 ± 0.1	15 ± 2.0
Nitrates (N)	0.8 ± 0.1	0.6 ± 0.1	2 ± 2.0
Zinc (II)	N.D.	N.D.	N.D.
Copper (II)	N.D.	N.D.	N.D.
Sulfates	17 ± 0.9	22 ± 3.0	22 ± 2.0
Nickel (II)	N.D.	N.D.	N.D.
Potassium (I)	1.3 ± 0.1	1 ± 0.1	1.5 ± 0.1

1 = GW : groundwater.

2 = N.D. (not detected) means that the Fe<sup>2+</sup>, Zn<sup>2+</sup>, Cu<sup>2+</sup>, and Ni<sup>2+</sup> concentrations are less than 0.01 mg/L.

**Table 4.5.2 Values of pH and COD for the three groundwaters**

<b>Parameter</b>	<b>GW<sup>1</sup> 1</b>	<b>GW 2</b>	<b>GW 3</b>
<b>COD (feed) mg/L</b>	13 ± 2.0	17 ± 3.0	10 ± 2.0
<b>pH (feed)</b>	7.3 ± 0.1	7.2 ± 0.1	7.2 ± 0.1
<b>Adjusted pH (feed)</b>	7.0 ± 0.1	7.1 ± 0.1	7.0 ± 0.1
<b>pH (10,000 MWCO, Permeate)</b>	6.7 ± 0.1	6.6 ± 0.1	6.7 ± 0.1
<b>pH (30,000 MWCO, Permeate)</b>	6.6 ± 0.1	6.8 ± 0.1	6.7 ± 0.1

1 = GW : groundwater.

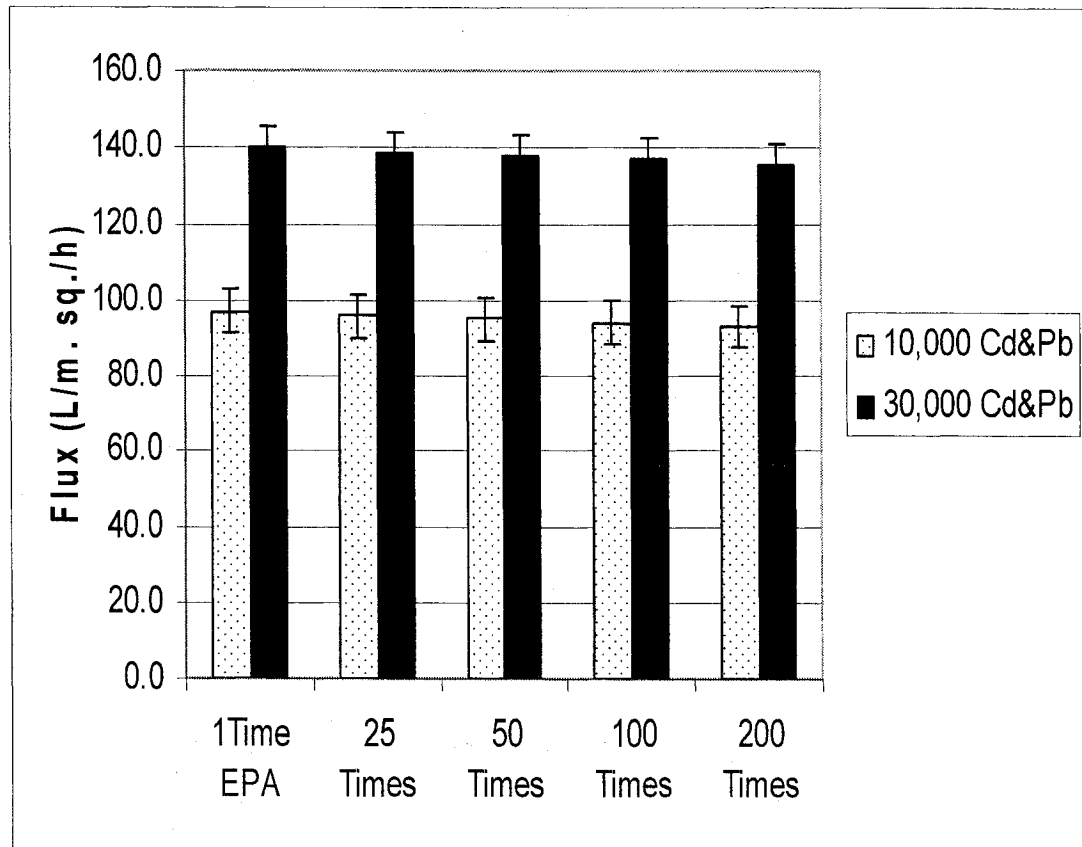
### **Contamination with cadmium (Cd) individually**

Using ultrafiltration (UF), the rhamnolipid biosurfactant was investigated to treat cadmium-contaminated groundwater. The first, second and third groundwaters were artificially contaminated by adding cadmium individually. The standard set by Quebec for drinking water regarding cadmium is 0.005 mg/L (Quebec Environment Quality Act, 2005). According to the USEPA, the maximum contaminant level goals (MCLG) and maximum contaminant levels (MCL) both are also 0.005 mg/L for cadmium in drinking water (Weiner, 2000).

Artificially contaminated groundwater solutions containing cadmium (Cd) individually were prepared by placing a predetermined amount of its metal salt (cadmium nitrate). A constant concentration of 0.2 mmol/L rhamnolipid was used. The solution concentrations were one, twenty five, fifty, hundred and two hundred times of the maximum contaminant level (MCL) set by Quebec and the US EPA for drinking water regarding cadmium.

The bench-scale MEUF system was used to investigate the capability and performance of the rhamnolipid biosurfactant for the removal of cadmium ions. The membranes used were those of 10,000 and 30,000 MWCO. According to the optimized operating conditions for the bench-scale MEUF

system in section 4.1, the transmembrane pressure ( $\Delta P_{tm}$ ) was set as 69 kPa and room temperature was applied ( $25 \pm 1^\circ\text{C}$ ).



**Figure 4.5.4 Averages of permeate fluxes of membranes with (MWCO) of 10,000 and 30,000 Da. (in case of  $\text{Cd}^{+2}$  and  $\text{Pb}^{+2}$  collectively).**

The values of cadmium (Cd) concentration in the artificially contaminated groundwater solutions were consistent with the level of contaminated groundwater in the USA (Paulson, 1997). Duplicate samples were prepared for each test, and the average was reported. Figure 4.5.4 illustrates the



averages of permeate fluxes for cadmium and lead ions regarding the first, second and third groundwater in case of  $\text{Cd}^{+2}$  and  $\text{Pb}^{+2}$ , collectively. No significant difference was observed in fluxes between these values and the ones in the cases of individual cadmium and lead. Cadmium concentrations in the permeates of the first, second and third groundwater were less than less than 0.01 mg/L using the membranes of 10,000 and 30,000 MWCO (Table 4.5.3). From Figure 4.5.4 and Table 4.5.3, since the initial concentrations of cadmium were reduced to less than 0.01 mg/L (more than 99.0 % removal efficiency), this indicates that the rhamnolipid-based ultrafiltration is an efficient technique for removal of cadmium (Cd) individually from the groundwater.

**Table 4.5.3 Different concentrations of cadmium ions individually and collectively in the permeate (for first, second and third groundwaters using 10,000 and 30,000 MWCO)**

Initial Concentration (mg/L)	Relative initial concentration to US EPA and Quebec Standards	Concentration in the permeate (mg/L)
0.005	1 times	N. D.
0.125	25 times	N. D.
0.25	50 times	N. D.
0.5	100 times	N. D.
1.0	200 times	N. D.
3.0	600 times	N. D.

N. D. (not detected) means that the concentration is less than 0.01 mg/L.

### Contamination with lead (Pb) individually

The first, second and third groundwaters were artificially contaminated with lead. The purpose was to investigate the capability and performance of the rhamnolipid biosurfactant enhanced by bench-scale UF system for removal of lead ions individually from the groundwater.

**Table 4.5.4 Concentrations of lead ions individually and collectively in the permeate (for first, second and third groundwaters using 10,000 and 30,000 MWCO)**

Initial concentration (mg/L)	Comparison of the initial concentration to US EPA Standard (ref. a)	Comparison of the initial concentration to Quebec Standard (ref b)	Concentration in the permeate (mg/L)
0.015	1 time	1.5 times	N. D.
0.375	25 times	37.5 times	N. D.
0.75	50 times	75 times	N. D.
1.5	100 times	150 times	N. D.
3.0	200 times	300 times	N. D.

N. D. (not detected) means that the lead concentration is less than 0.01 mg/L.

Reference a : (Quebec Environment Quality Act, 2005).

Reference b : (Weiner, 2000).

The standard set by Quebec for drinking water regarding lead was 0.010 mg/L (Quebec Environment Quality Act, 2005). According the USEPA, the maximum contaminant level goal (MCLG) and maximum contaminant level (MCL) both were 0.015 mg/L for lead in drinking water (Weiner, 2000).

Artificially-contaminated groundwater solutions containing lead (Pb) individually were prepared by placing a predetermined amount of its metal salt (lead nitrate) in the first, second and third groundwater.. Like the individual cadmium, the initial solution concentrations were one, twenty five, fifty, a hundred and two hundred times of the maximum contaminant level (MCL) set by Quebec and the US EPA for drinking water regarding lead.

The same bench-scale MEUF system as well the same compatible membranes of 10,000 and 30,000 MWCO were used. The optimized operating conditions for the bench-scale MEUF system in section 4.1, were applied. Duplicate samples were prepared for each test, and the average was reported. The lead metal concentrations in the artificially contaminated groundwater solutions were consistent with the level of contaminated groundwater in the USA (Paulson, 1997). Table 4.5.4 shows that the concentrations of lead in the permeate for all groundwater were below the levels of detection (less than 0.01 mg/L) using the membranes of 10,000 and 30,000 MWCO. Thus, the permeates were compatible with the standards of both Quebec and the USEPA.

## **Contamination with cadmium (Cd) and lead (Pb)**

Utilizing ultrafiltration (UF), the rhamnolipid biosurfactant was examined to treat contaminated groundwaters with cadmium (Cd) and lead (Pb) collectively. The first, second and third groundwaters were artificially contaminated with different predetermined amounts of cadmium (Cd) and lead (Pb) as mentioned in the individual metal cases. The lead and cadmium metal concentrations in the artificially contaminated groundwater solutions were consistent with the level of contaminated groundwater in the USA (Paulson, 1997). Also, the same bench-scale MEUF system, membranes and optimized operating conditions were applied.

Duplicate samples were prepared for each test, and the average was reported. Using the same membranes, the levels of cadmium and lead in the permeate for all tests were below the levels of detection (less than 0.01 mg/L).

Since the initial concentrations of cadmium and lead collectively was reduced to less than 0.01 mg/L (more than 99.0 % removal efficiency), it indicates that the rhamnolipid-based ultrafiltration is an efficient technique to treat groundwaters contaminated with a mixture of cadmium and lead. It can be seen in Figure (4.5.4) that the ultrafiltration membrane with MWCO

10,000 had a lower relative flux than that of MWCO 30,000 (with larger pore size) in the treatment of all groundwaters using rhamnolipid-based UF. This flux behavior can, probably, be explained by taking into account that the concentrations of rhamnolipid micelles and/or metal-rhamnolipid complexes rejected by the membrane of MWCO 10,000 was much higher than that of MWCO 30,000. Thus, those rejected higher concentrations caused a reduction of the flux. Also, the fluxes shown in Figure (4.5.4) are consistent with the ones in other UF studies (Sinclair et al., 2004).

Results of this study indicate that the presence of cations such as  $\text{Ca}^{+2}$  (from  $25 \pm 1$  to  $86 \pm 3.0$  mg/L),  $\text{Na}^+$  (from  $15 \pm 2.0$  to  $32 \pm 1.2$  mg/L), and  $\text{Mg}^{+2}$  (from  $8 \pm 0.1$  to  $23 \pm 2.0$  mg/L) did not affect the removal efficiency ( $> 99.0\%$ ) of  $\text{Pb}^{+2}$  (from 0.015 to 3.0 mg/L), or  $\text{Cd}^{+2}$  (from 0.005 to 3.0 mg/L), from the three groundwaters investigated. Regarding the removal of cadmium (Cd) and lead (Pb) ions individually or collectively from the three contaminated groundwaters using the two membranes, it was concluded that the rhamnolipid-based ultrafiltration technique is an efficient treatment method. The results produced from bench-scale MEUF experiments as well as the optimized operating conditions were helpful tools in improving the performance of the rhamnolipid-based ultrafiltration system regarding the contaminated groundwaters. Furthermore, the fluxes the rhamnolipid-based ultrafiltration technique experiments were reasonable compared to the UF studies. Unlike the case of wastewater treatment, there was no evidence of

serious fouling problems. This may be, explained by taking into account that the levels of both the metals and the organics in the groundwater were still too low compared to the ones of the wastewater.

### **Summary**

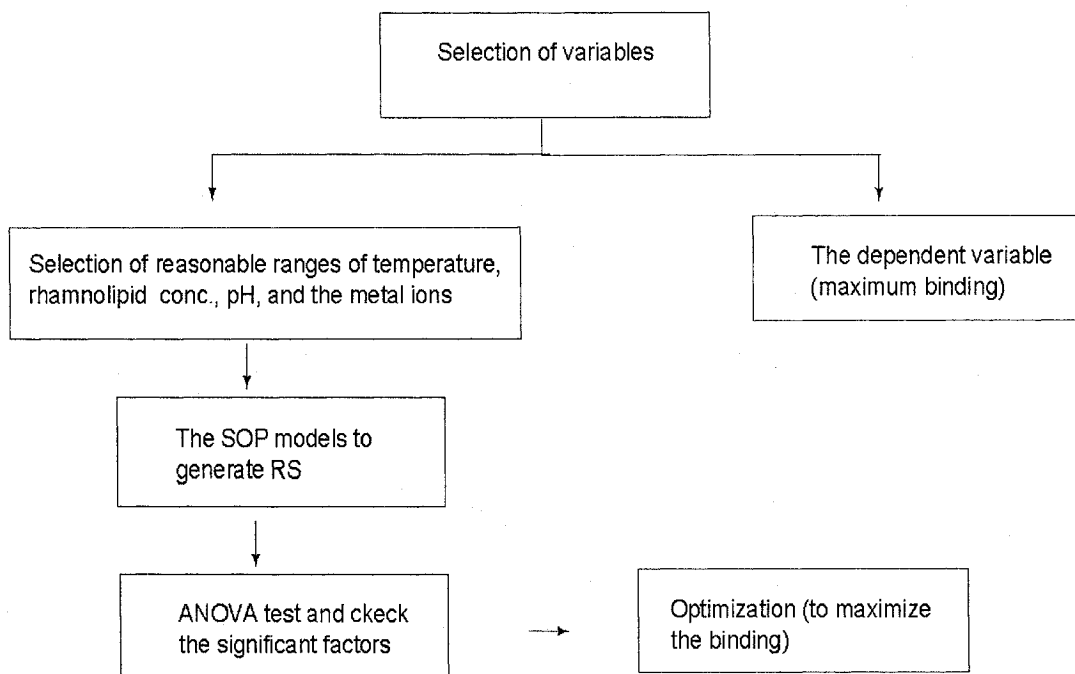
In this work, the rhamnolipid-enhanced ultrafiltration technique was utilized to treat samples of three artificially contaminated groundwater samples using membranes with molecular weight cut off (MWCO) of 30,000 Da and 10,000 Da. The resulting heavy metal concentrations in the permeate indicated that more than 99.9 % removal efficiency was achieved using both membranes. The permeates were compatible with the standards of both Quebec and the USEPA. In addition, the permeate fluxes were consistent with the ones in other UF studies. Furthermore, to investigate the possibility of interference from a competition effect with  $\text{Ca}^{+2}$ , three different groundwaters with varying levels of calcium were selected. It was found that  $\text{Ca}^{+2}$  did not affect the removal efficiency (> 99.0 %) of  $\text{Pb}^{+2}$  (from 0.015 to 3.0 mg/L), or  $\text{Cd}^{+2}$  (from 0.005 to 3.0 mg/L), from the three groundwaters investigated.

## 4.6 Competition systems

Industrial effluents usually contain more than one metal. Multicomponent systems are characterized by additional features from those of single components. There are possible interaction effects between different species in solution and potential interactions on the surface of rhamnolipid micelles in particular. Competition among the different metal ion species for the binding sites of the surface of rhamnolipid micelles usually took place and depended on several ionic chemical characteristics. The rhamnolipid biosurfactant showed very high capability for the removal of metal ions from industrial wastewaters and groundwaters. In this research, since the degree of metal removal from the industrial wastewaters depends on the multi-metal competition interactions with the rhamnolipid biosurfactant. Therefore, it would be desirable to study the binding from a mixture of metal ions onto the rhamnolipid biosurfactant. The objective of this work was to conduct binding experiments and to investigate the relative affinity of rhamnolipid for  $\text{Cu}^{2+}$ ,  $\text{Zn}^{2+}$ ,  $\text{Ni}^{2+}$ , and  $\text{Cd}^{2+}$  metal ions at equilibrium.  $\text{Pb}^{2+}$  proved to be problematic to the rhamnolipid-enhanced UF (Chapter 4, section 1), and so was excluded from further consideration.

In order to efficiently achieve the maximum binding of metal ions in binary system, optimization by the response surface methodology approach was utilized. The pH, temperature, and mixing of metal ions in the binary system were considered as independent variables while maximum binding of metal ions was designated as a dependent variable. Second order polynomial (SOP) models

were utilized as functions of the independent variables. An analysis of variance (ANOVA) was performed to determine which of the parameters significantly affected the dependent variable.



SOP = Second order polynomial ; RS = Response surface

**Figure 4.6.1. Experimental data flow to achieve the maximum binding of metal ions in the binary system.**



Max. adsorptn. (Binding Cap.)  
X = A: Cd (mmol/L)  
Y = B: Cu (mmol/L)

Actual Factors  
C: Ni (mmol/L) = 1.00  
D: Zn (mmol/L) = 1.03

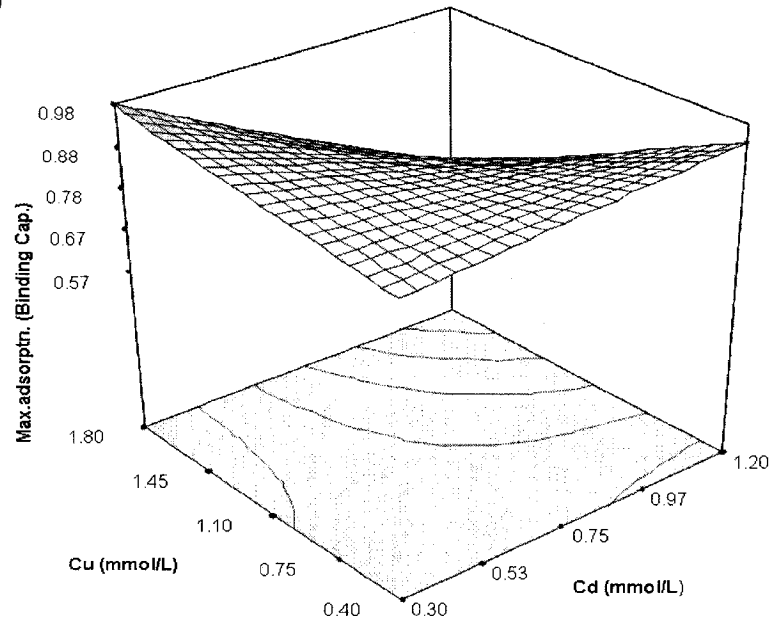


Figure 4.6.2. Response surface obtained for cadmium and copper

Max. adsorptn. (Binding Cap.)  
X = C: Ni (mmol/L)  
Y = A: Cd (mmol/L)

Actual Factors  
B: Cu (mmol/L) = 1.10  
D: Zn (mmol/L) = 1.03

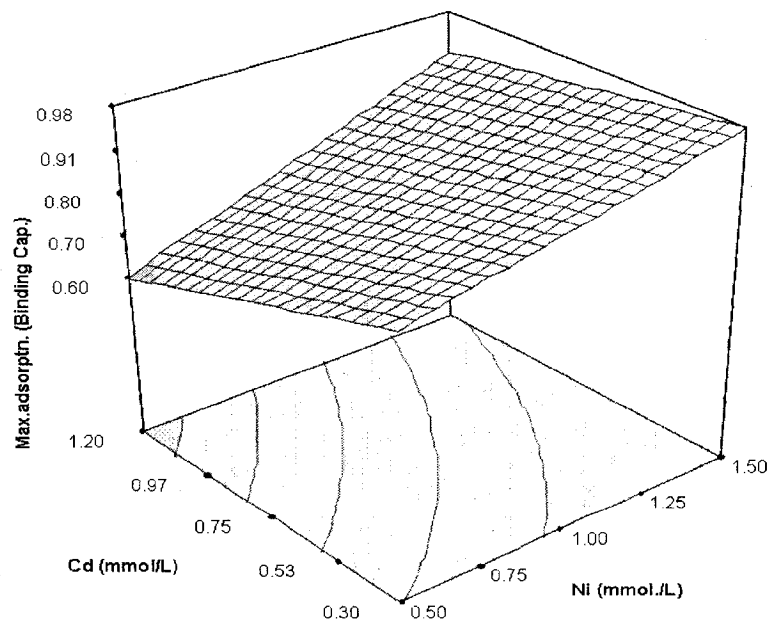
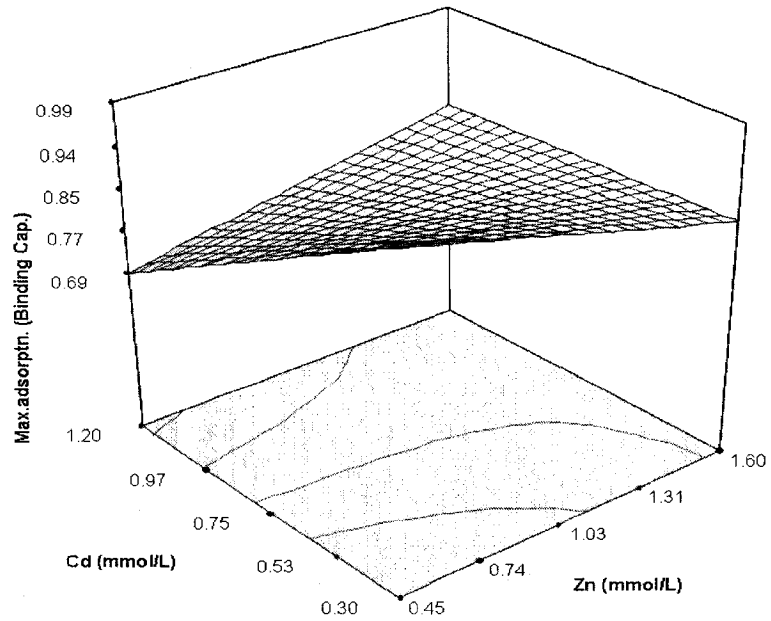


Figure 4.6.3. Response surface obtained for nickel and cadmium.

Max. adsorptn. (Binding Cap.)  
 X = D: Zn (mmol/L)  
 Y = A: Cd (mmol/L)

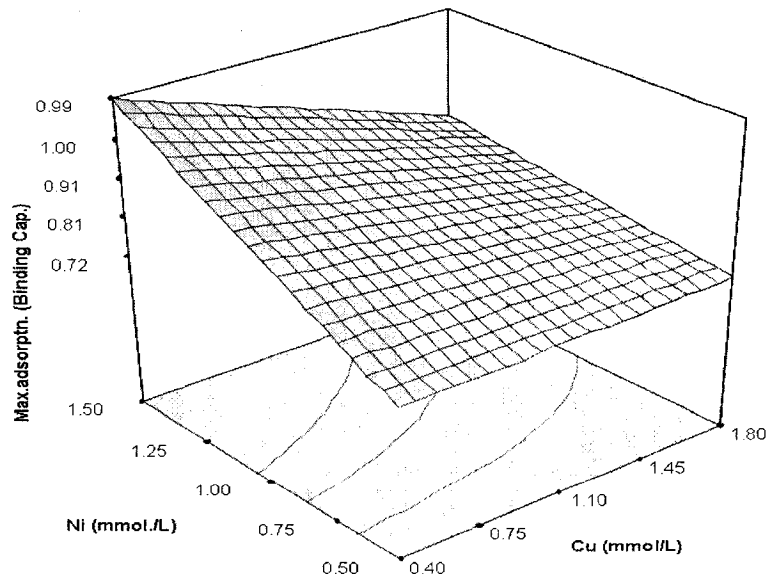
Actual Factors  
 B: Cu (mmol/L) = 1.10  
 C: Ni (mmol/L) = 1.00



**Figure 4.6.4. Response surface obtained for zinc and cadmium.**

Max. adsorptn. (Binding Cap.)  
 X = B: Cu (mmol/L)  
 Y = C: Ni (mmol/L)

Actual Factors  
 A: Cd (mmol/L) = 0.75  
 D: Zn (mmol/L) = 1.03



**Figure 4.6.5. Response surface obtained for copper and nickel.**

Max. adsorptn. (Binding Cap.)  
X = B: Cu (mmol/L)  
Y = D: Zn (mmol/L)

Actual Factors  
A: Cd (mmol/L) = 0.75  
C: Ni (mmol/L) = 1.00

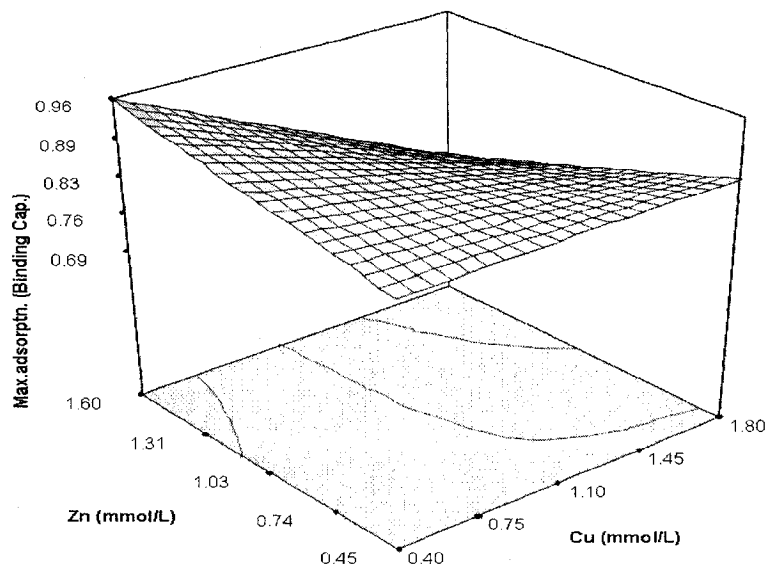


Figure 4.6.6. Response surface obtained for copper and zinc.

Max. adsorptn. (Binding Cap.)  
X = C: Ni (mmol/L)  
Y = D: Zn (mmol/L)

Actual Factors  
A: Cd (mmol/L) = 0.75  
B: Cu (mmol/L) = 1.10

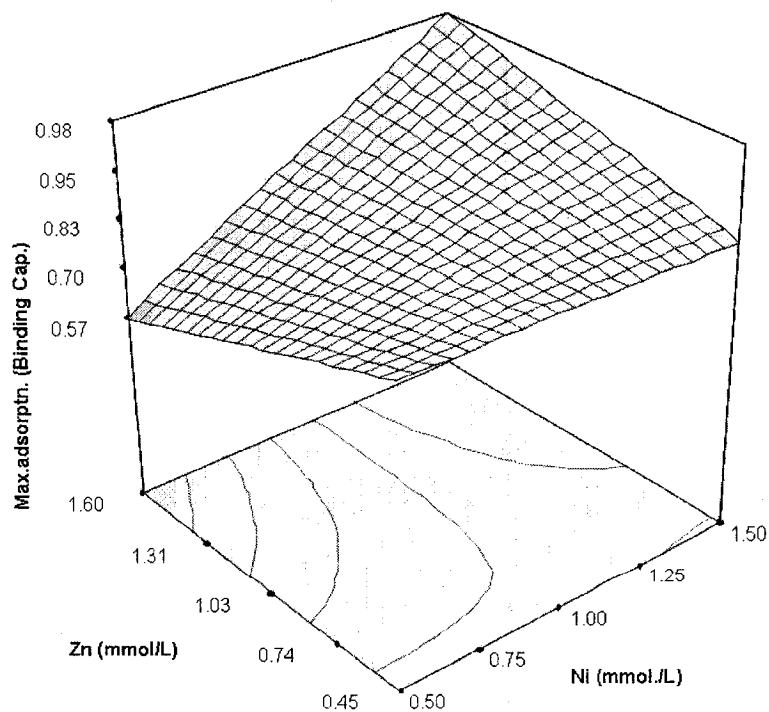


Figure 4.6.7. Response surface obtained for nickel and zinc.

Response surface and contour plots were generated as shown in Figures 4.6.2 to 4.6.7. Optimum regions of independent and dependent variables were achieved. The major objective was to maximize binding of metal ions in the binary system. Figure 4.6.1 shows the experimental data flow to achieve the maximum binding of metal ions in the binary system.

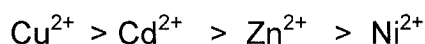
From Figures 4.6.2 to 4.6.7, it can be seen that the suggested ranges of heavy metal concentrations were within 0.45 to 1.6, 0.4 to 1.8, 0.5 to 1.5, and 0.3 to 1.2, mmol/L for zinc, copper, nickel, and cadmium, respectively. Separate validation experiments using the lab-scale UF system were performed to verify these optimum conditions. The most applicable operating conditions were transmembrane pressure  $69 \pm 2$  kPa, biosurfactant-to-metal molar ratios of 2:1, temperature of  $25 \pm 1^\circ\text{C}$ , and pH of  $6.9 \pm 0.1$ . The ranges of heavy metal concentrations were within about 0.05 to 2 mmol/L. The same ultrafiltration system was utilized.

### **Single, binary and ternary metal systems**

Binding of metals from solutions containing one, two, and three various metals were studied. The  $\text{Cu}^{2+}$ ,  $\text{Zn}^{2+}$ ,  $\text{Ni}^{2+}$ , and  $\text{Cd}^{2+}$  ions were selected and investigated in different combinations in single, binary and ternary systems for binding experiments. The samples were prepared with equimolar concentrations for the metals under investigation.

### **Single binding systems**

The equilibrium isotherms were quantified for the experimental systems regarding zinc, copper, nickel, and cadmium ions. Each metal ion was involved in a single component isotherm system using the membranes of 30,000 MWCO and 10,000 MWCO Da. Figures 4.6.8 to 4.6.11 show the individual single component isotherms for zinc, copper, nickel, and cadmium, respectively. The plateau for each metal was reached because the binding capacity of rhamnolipid biosurfactant is restricted by the fixed number of binding sites. It can be seen in regards to the individual single component isotherms, copper has the highest binding affinity followed by cadmium, zinc and nickel, respectively. The results of the single metal binding (molar basis) show the following order of affinity



### **Binary metal systems**

#### **Zinc ion system**

Figure 4.6.12 represents the binding isotherm of  $\text{Zn}^{2+}$  by the rhamnolipid biosurfactant in the two-metal system. The data of the single-metal system are also shown. The binding affinity of the rhamnolipid biosurfactant for  $\text{Zn}^{2+}$  was slightly suppressed in the presence of  $\text{Ni}^{2+}$  by about 30 % for the 30,000 MWCO membrane, and by about 32 % for the 10,000 MWCO Da membrane. These ratios are slightly lower than the case in the  $\text{Ni}^{2+}$ -system where nickel binding capacity was reduced in the presence of  $\text{Zn}^{2+}$  by about 34 % for the membrane of

30,000 MWCO, and by about 40 % for the membranes of 10,000 MWCO Da, (Table 4.6.1 and Figure 4.6.12). This could imply that the selectivity of the rhamnolipid biosurfactant for  $Zn^{2+}$  was slightly more than for  $Ni^{2+}$ .

The presence of  $Cu^{2+}$  in the two-metal system significantly suppressed the rhamnolipid binding affinity of  $Zn^{2+}$  using similar membranes. This indicates that binding affinity of the rhamnolipid biosurfactant for  $Cu^{2+}$  was more than for  $Zn^{2+}$ . Almost the same trend was found in the case of the presence of  $Cd^{2+}$  where the binding affinity of the rhamnolipid biosurfactant for  $Zn^{2+}$  was suppressed in the presence of  $Cd^{2+}$  by about 58 % for the membrane of 30,000 MWCO, and by about 63 % for the membranes of 10,000 MWCO Da. These ratios are slightly higher than the case in the  $Cd^{2+}$ -system where the affinity for  $Cd^{2+}$  was suppressed in the presence of  $Zn^{2+}$  by about 37 % for the membrane of 30,000 MWCO and by about 40 % for the membranes of 10,000 MWCO Da, (Table 4.6.1 and Figures 4.6.12 and 4.6.15). This could imply that the selectivity of the rhamnolipid biosurfactant for  $Cd^{2+}$  was more than for  $Zn^{2+}$ .

### **Copper ion system**

Figure 4.6.13 represents the binding isotherm of copper by the rhamnolipid biosurfactant in the two-metal system. The data of the single-metal system are also demonstrated. The figure illustrates that single-metal binding affinity of the rhamnolipid biosurfactant for copper was slightly suppressed in the presence of cadmium, zinc and nickel metal ions in the system using the membranes of

30,000 MWCO and 10,000 MWCO Da. This implies that the selectivity of the rhamnolipid biosurfactant for  $\text{Cu}^{2+}$  was more than for  $\text{Zn}^{2+}$ ,  $\text{Ni}^{2+}$ , and  $\text{Cd}^{2+}$ .

### **Nickel ion system**

When the binding of each metal in this binary system by the rhamnolipid biosurfactant was compared to the binding when each metal was bound from an individual solution, it was observed that the binding of  $\text{Ni}^{2+}$  decreased significantly in the presence of  $\text{Zn}^{2+}$ ,  $\text{Cu}^{2+}$ , and  $\text{Cd}^{2+}$  metal ions in the UF membrane systems. Also, it was noticed that the binding of  $\text{Zn}^{2+}$ ,  $\text{Cu}^{2+}$ , and  $\text{Cd}^{2+}$  in the binary system showed insignificant reduction by the presence of  $\text{Ni}^{2+}$  in the UF membrane systems. This implies that the selectivity of the rhamnolipid biosurfactant for  $\text{Ni}^{2+}$  was less than for  $\text{Zn}^{2+}$ ,  $\text{Cu}^{2+}$ , or  $\text{Cd}^{2+}$  (Table 4.6.1 and Figure 4.6.14).

### **Cadmium ion system**

Figure 4.6.15 represents the binding isotherm of cadmium by the rhamnolipid biosurfactant in the two-metal system. The data of the single-metal system are also demonstrated. The single-metal binding affinity of the rhamnolipid biosurfactant for  $\text{Cd}^{2+}$  was slightly suppressed in the presence of  $\text{Zn}^{2+}$  and  $\text{Ni}^{2+}$  in the binary system using the membranes of 30,000 MWCO and 10,000 MWCO Da. This may imply that the selectivity of the rhamnolipid biosurfactant for  $\text{Cd}^{2+}$  was more than for  $\text{Ni}^{2+}$  or  $\text{Zn}^{2+}$ .

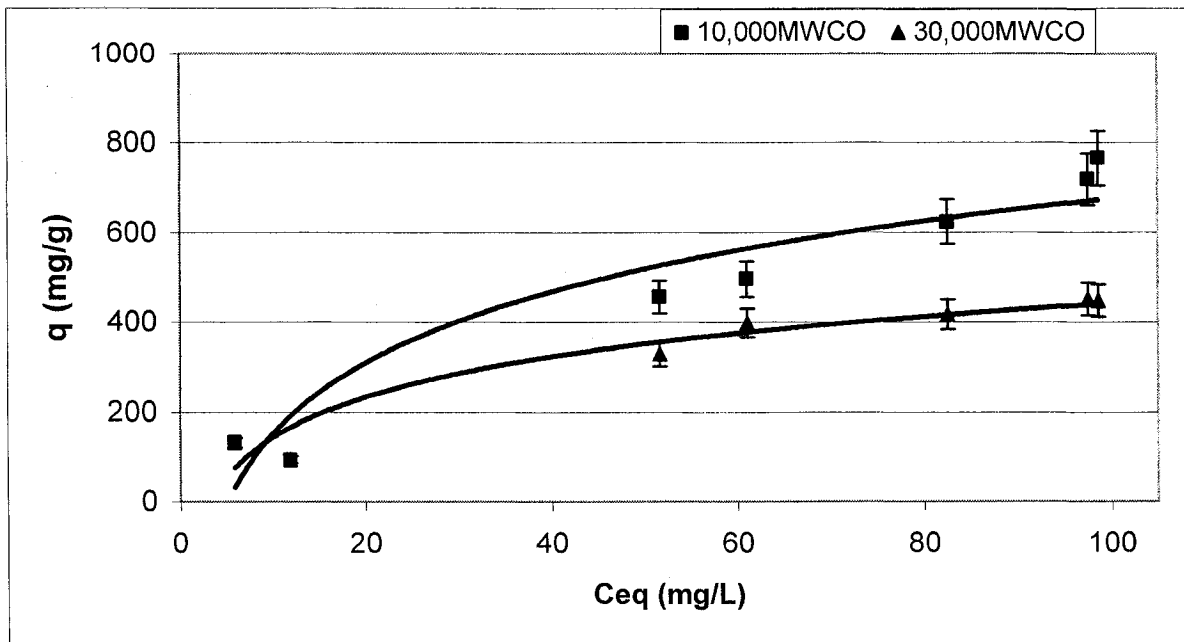


Figure 4.6.8. Binding isotherms of zinc for the two UF membranes

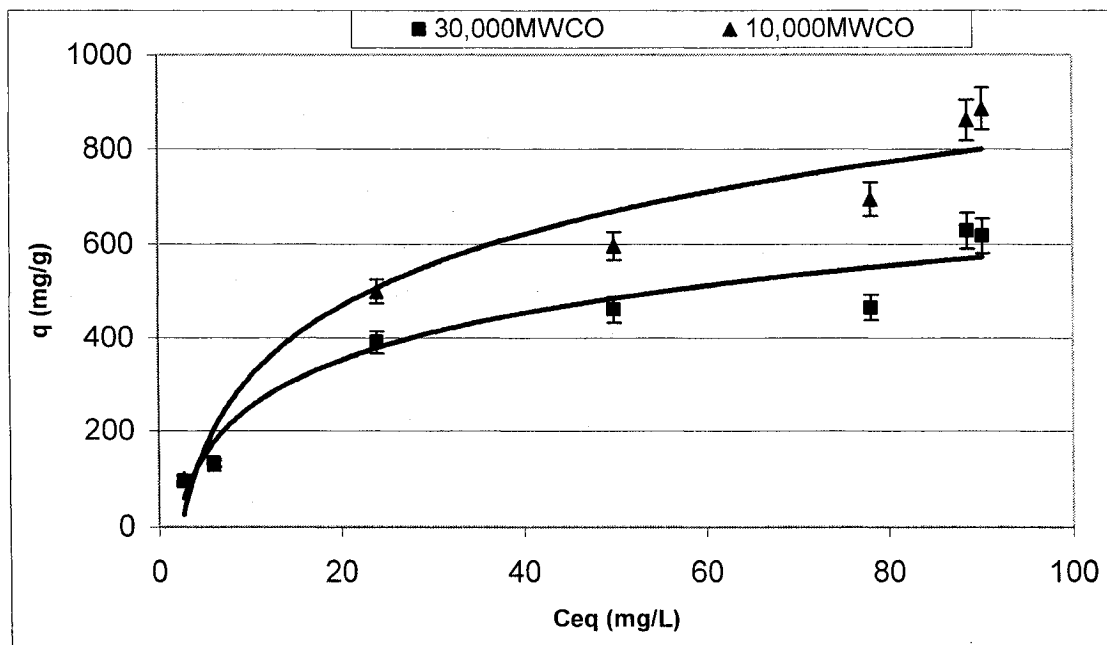


Figure 4.6.9. Binding isotherms of copper for the two UF membranes



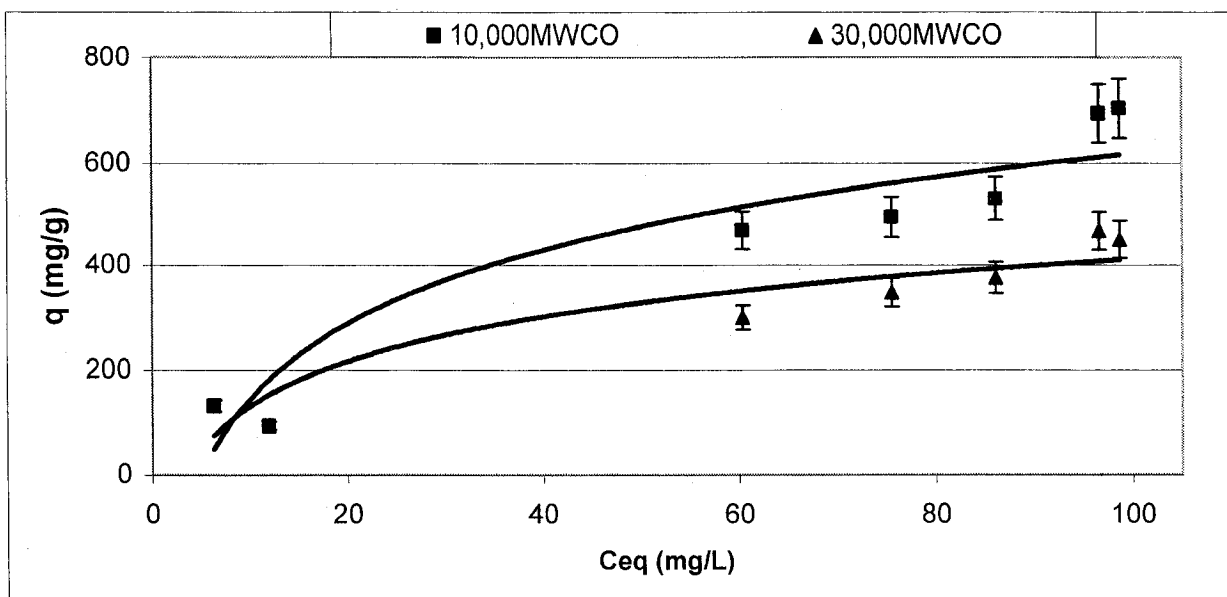


Figure 4.6.10. Binding isotherms of nickel for the two UF membranes

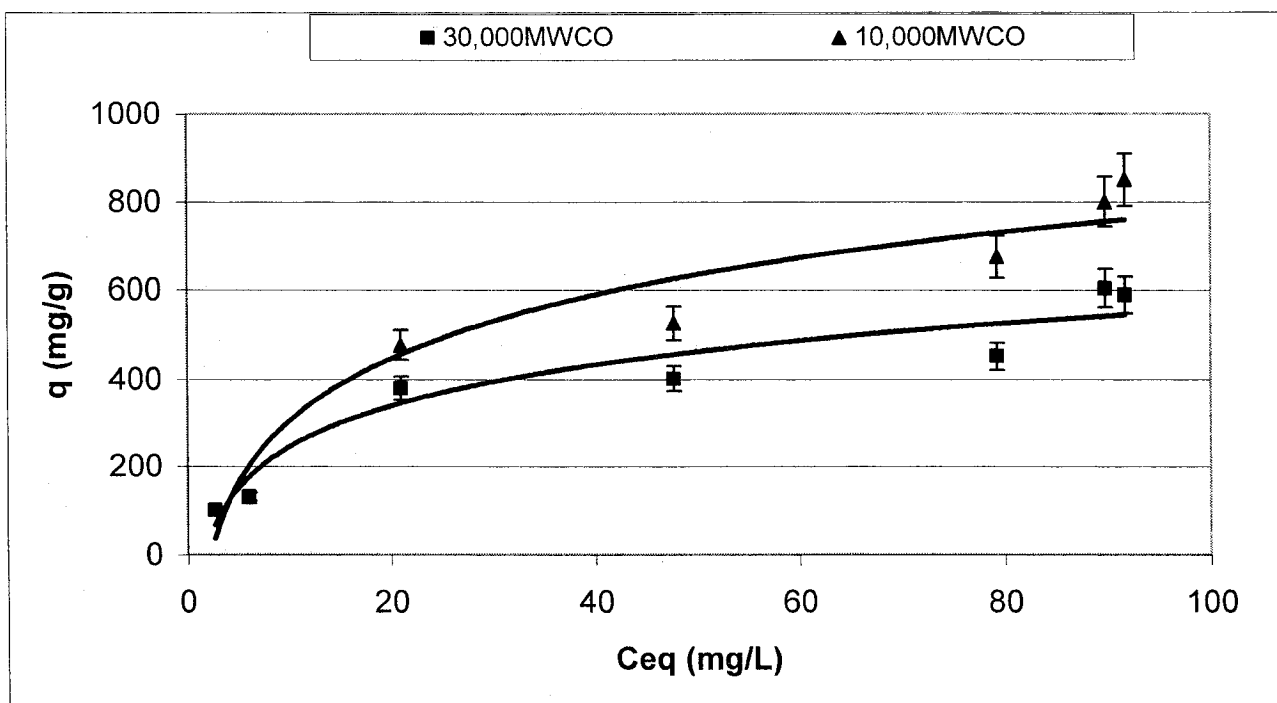


Figure 4.6.11. Binding isotherms of cadmium for the two UF membranes

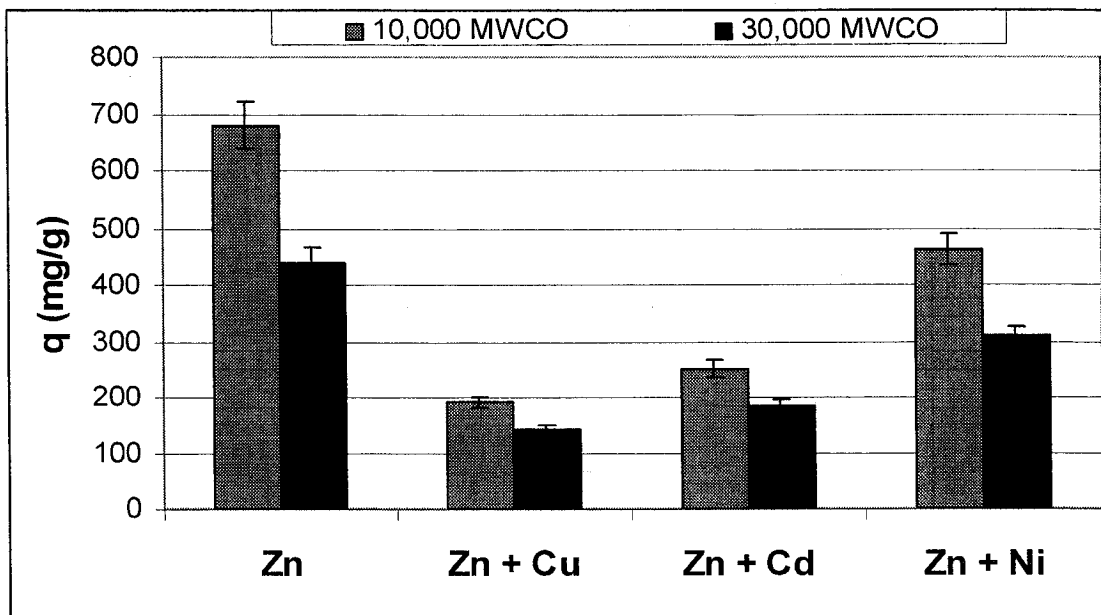


Figure 4.6.12. Binary system of zinc competition with  $\text{Cu}^{2+}$ ,  $\text{Ni}^{2+}$ , and  $\text{Cd}^{2+}$

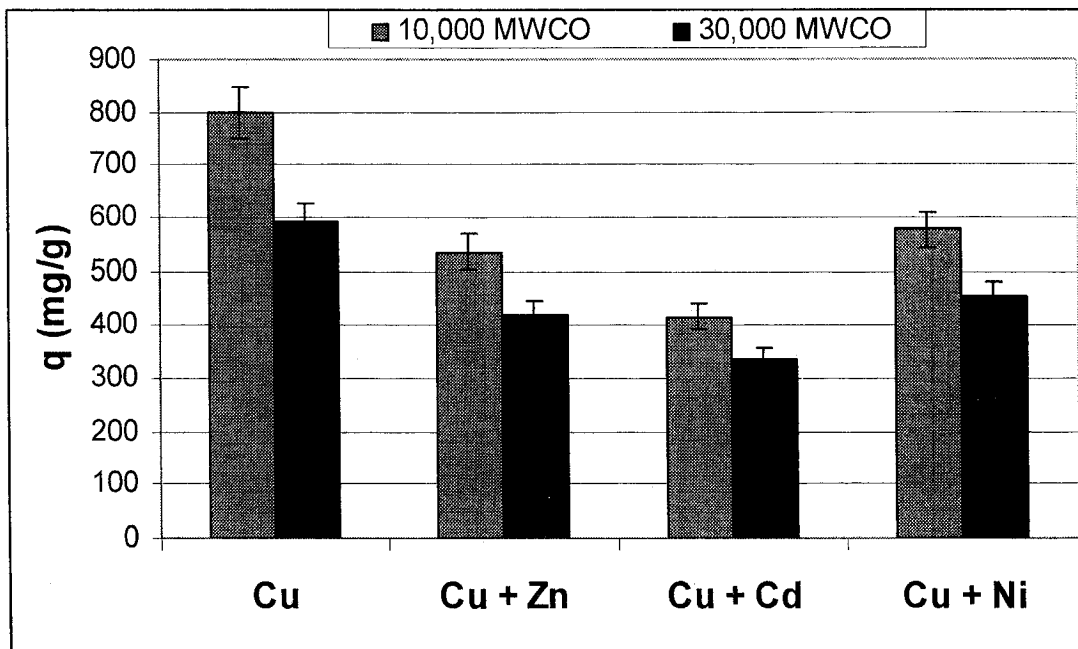


Figure 4.6.13. Binary system of copper competition with  $\text{Cd}^{2+}$ ,  $\text{Ni}^{2+}$ , and  $\text{Zn}^{2+}$

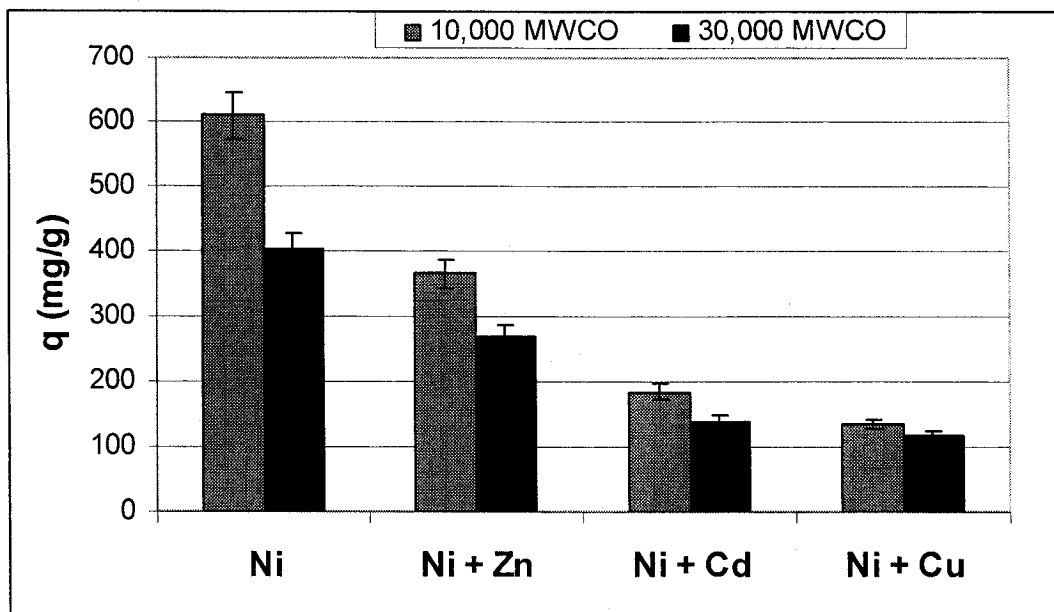


Figure 4.6.14. Binary system of nickel competition with  $\text{Cd}^{2+}$ ,  $\text{Cu}^{2+}$ , and  $\text{Zn}^{2+}$

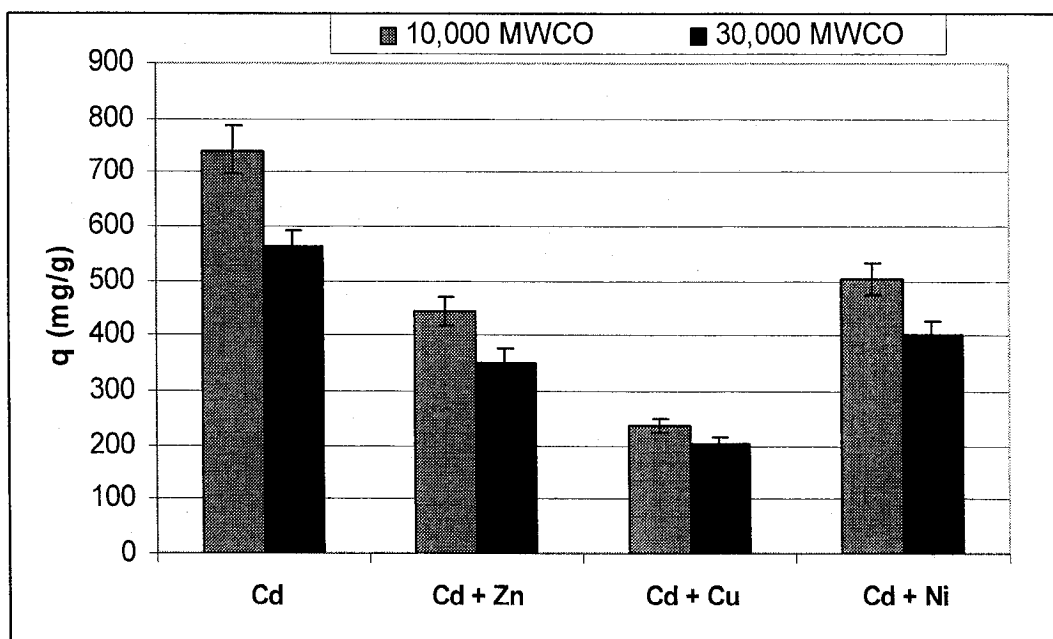


Figure 4.6.15. Binary system of cadmium competition with  $\text{Cu}^{2+}$ ,  $\text{Ni}^{2+}$ , and  $\text{Zn}^{2+}$

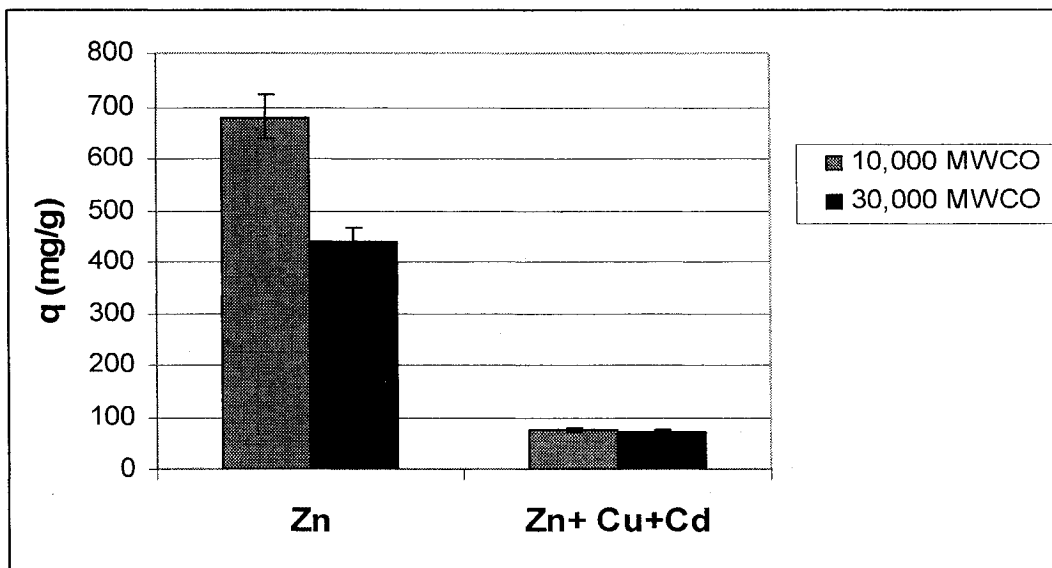


Figure 4.6.16. Ternary system of zinc competition with  $\text{Cu}^{2+}$  and  $\text{Cd}^{2+}$

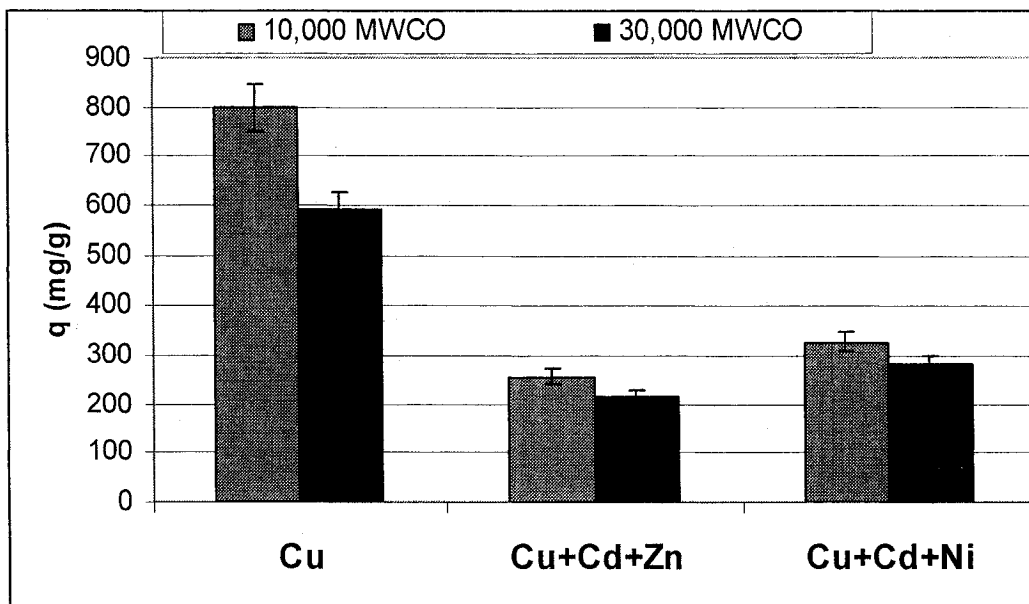


Figure 4.6.17. Ternary system of copper competition with  $\text{Cd}^{2+}$ ,  $\text{Ni}^{2+}$ , and  $\text{Zn}^{2+}$

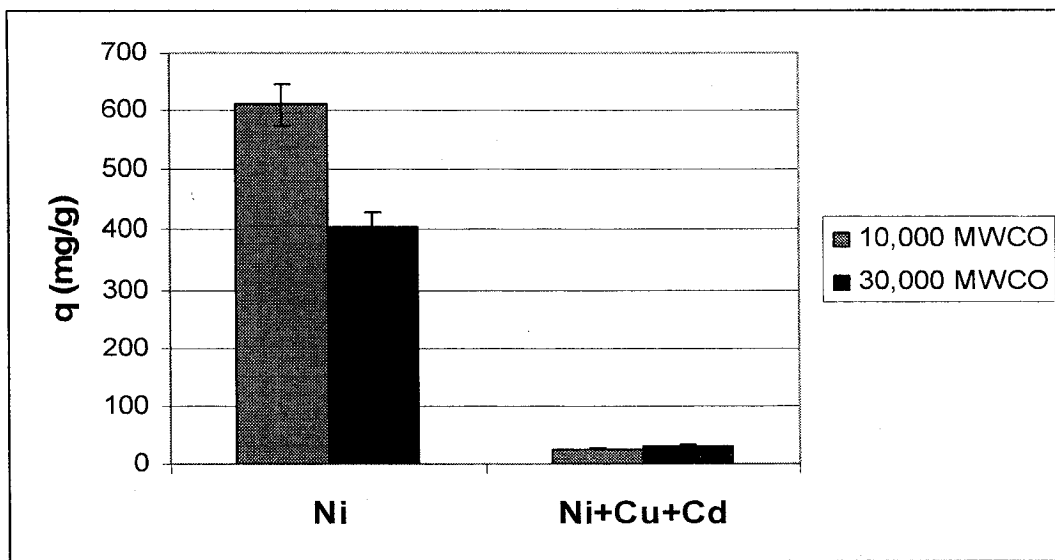


Figure 4.6.18. Ternary system of nickel competition with  $\text{Cd}^{2+}$ , and  $\text{Cu}^{2+}$

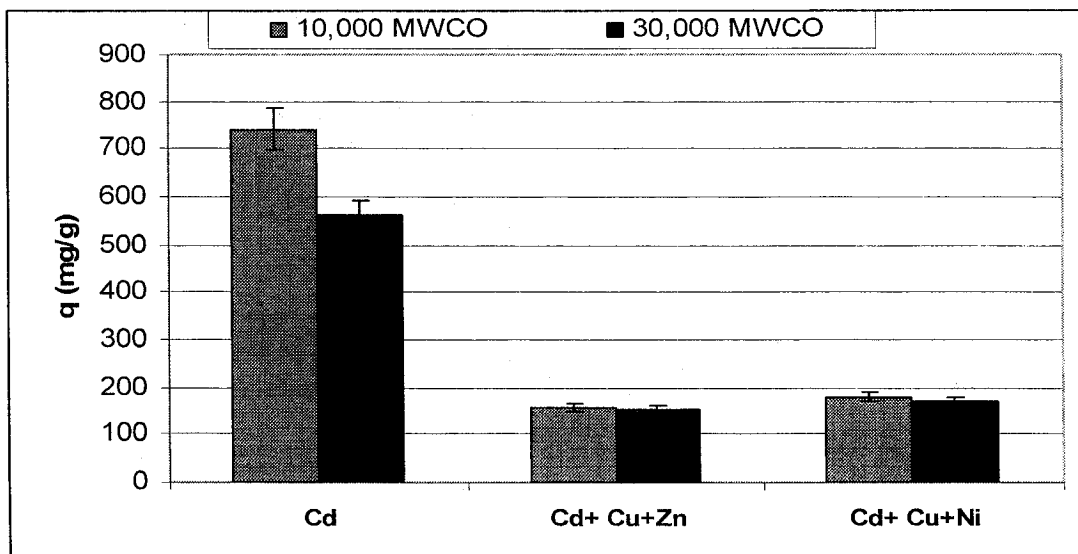


Figure 5.6.19. Ternary system of cadmium competition with  $\text{Cu}^{2+}$ ,  $\text{Ni}^{2+}$  and  $\text{Zn}^{2+}$

**Table 4.6.1 Metal systems and the reduction (%) in the single-metal binding affinity of the rhamnolipid biosurfactant for the two UF membranes**

<b>Metal ion (II)</b>	<b>Metal system</b>	<b>Reduction (%) in the single-metal binding affinity 30,000 MWCO</b>	<b>Reduction (%) in the single-metal binding affinity 10,000 MWCO</b>
Copper	Cu + Cd	43 ±2.1	48 ±2.2
Copper	Cu + Zn	29 ±2.0	33 ±2.1
Copper	Cu + Ni	23 ±1.1	28 ±1.3
Copper	Cu + Cd + Zn	64 ±2.8	68 ±3.1
Copper	Cu + Cd+ Ni	52 ±2.3	59 ±2.2
Cadmium	Cd + Cu	64 ±2.9	68 ±3.1
Cadmium	Cd + Zn	37 ±1.4	40 ±1.6
Cadmium	Cd + Ni	28 ±1.1	32 ±1.3
Cadmium	Cu + Cd + Zn	73 ±3.1	79 ±3.5
Cadmium	Cu + Cd + Ni	70 ±4.1	76 ±3.9
Zinc	Zn + Cu	70 ±4.1	76 ±3.9
Zinc	Zn + Cd	58 ±2.3	63 ±3.1
Zinc	Zn + Ni	30 ±2.2	32 ±2.1
Zinc	Cu + Cd + Zn	84 ±5.1	89 ±4.9
Nickel	Ni + Cu	71 ±5.1	78 ±6.4
Nickel	Ni + Cd	66 ±4.6	70 ±2.9
Nickel	Ni + Zn	34 ±2.1	40 ±2.5
Nickel	Cu + Cd + Ni	92 ±5.3	96 ±3.8

The presence of  $\text{Cu}^{2+}$  ions in the two-metal system suppressed the rhamnolipid binding affinity of  $\text{Cd}^{2+}$  by 64% and 68% for the membranes of 30,000 MWCO and 10,000 MWCO Da, respectively. These ratios are higher than in the  $\text{Cu}^{2+}$  system, where copper binding capacity was reduced in the presence of  $\text{Cd}^{2+}$  by about 34 % for the membrane of 30,000 MWCO and by about 40 % for the membranes of 10,000 MWCO Da (Table 4.6.1 and Figure 4.6.15). This indicates that binding affinity of the rhamnolipid biosurfactant for  $\text{Cu}^{2+}$  was more than for  $\text{Cd}^{2+}$ .

### **Ternary metal systems**

#### **$\text{Cu}^{2+}$ - $\text{Cd}^{2+}$ - $\text{Ni}^{2+}$ system:**

When comparing the binding of each metal in this ternary system by the rhamnolipid biosurfactant to the individual solutions, it was observed that the binding of  $\text{Ni}^{2+}$  decreased significantly, while the binding of  $\text{Cu}^{2+}$  decreased only slightly (Table 4.6.1 and Figure 4.6.17). Also, it was seen that the binding of  $\text{Cd}^{2+}$  decreased more than  $\text{Cu}^{2+}$  for both UF membranes.

### **Cu<sup>2+</sup> - Cd<sup>2+</sup> - Zn<sup>2+</sup> system:**

It can be seen from Table 4.6.1 and Figure 4.6.17 that the suppression of Zn<sup>2+</sup> was significant while the binding of Cu<sup>2+</sup> decreased only slightly in this ternary system. However, the suppression of Zn<sup>2+</sup> was not as much as that of Ni<sup>2+</sup>. Also when the binding of each metal in this ternary system by the rhamnolipid biosurfactant was compared to the binding of each from their individual solutions, it was noticed that the binding of Cd<sup>2+</sup> decreased more than Cu<sup>2+</sup> but not as much as that of Ni<sup>2+</sup> or Zn<sup>2+</sup>. This may imply that the selectivity of the rhamnolipid biosurfactant for Cd<sup>2+</sup> was more than for Ni<sup>2+</sup> or Zn<sup>2+</sup>. Thus, the order of metal binding (molar basis) in the ternary systems was: Cu<sup>2+</sup> > Cd<sup>2+</sup> > Zn<sup>2+</sup> > Ni<sup>2+</sup> which is consistent with the orders of the single and binary systems.

In order to evaluate other factor that might be significant in the binding process of the four heavy metal cations onto the rhamnolipid biosurfactant, a number of chemical properties will be assessed. The binding process of the four heavy metal cations onto the rhamnolipid biosurfactant can be explained in terms of Pauling's electronegativity (Cotton et al., 1999; Chien et al., 2005). Pauling electronegativity is a measure of the ability of an atom or molecule to attract electrons in the context of a chemical bond. The type of bond formed is principally determined by the difference in electronegativity between the atoms involved. Each element has a characteristic electronegativity on the Pauling scale (Cotton et al., 1999; Chien et al., 2005). According to Pauling, the higher



the electronegativity, the greater the capacity for binding. The data in Table 4.6.2 show that the more electronegative metal ions will be more strongly attracted onto the rhamnolipid biosurfactant. Copper has the highest Pauling electronegativity and the greatest binding capacity. The Pauling electronegativity of cadmium is slightly higher than zinc or nickel. The same trend is shown for their respective binding capacities, but nickel was the only exception. Again regarding the standard reduction potential, this trend is followed by these metals with the exception of nickel.

Alternately, the binding of the four heavy metal cations onto the rhamnolipid can be explained in terms of the " Hard Soft Acid Base Principle " (HSAB) (Koch, 2005b; Cotton et al., 1999;). According to the " Hard Soft Acid Base Principle " (HSAB), the facility for an acid–base reaction to take place depends mainly on the strength of both acid and base. Also, it depends on their hardness or softness (Koch, 2005b; Pearson, 1987). In the case of soft bases, the donor atoms have a low electronegativity and a high polarizability and they are easy to oxidize. They hold their valence electrons loosely. In the case of hard bases- the donor atoms have a high electronegativity and a low polarizability and they are difficult to oxidize. They hold their valence electrons tightly. With regard to the case of soft acids, the acceptor atoms are large, have a low positive charge density, and contain unshared pairs of electrons (p or d) in their valence shells. They have a high polarizability and a low electronegativity.

**Table 4.6.2 Selected chemical properties of some metal ions**

<b>Ion (II)</b>	<b>Coordination Number<sup>1</sup></b>	<b>Valency<sup>1,2</sup></b>	<b>Pauling Electronegativity<sup>1,2</sup></b>	<b>Standard Reduction Potential (V) <sup>2</sup></b>
<b>Cu</b>	6	2	1.90	+ 0.35
<b>Cd</b>	6	2	1.69	- 0.40
<b>Zn</b>	6	2	1.65	- 0.76
<b>Ni</b>	6	2	1.80	- 23

**1 :** (Cotton et al. 1999)

**2:** (Lee, 1991)

Regarding the case of hard acids, the acceptor atoms are small, have a high positive charge density, and do not contain unshared pairs in their valence shells. They have a low polarizability and a low electronegativity (Koch, 2005b; Pearson, 1987).

“ Hard Soft Acid Base Principle ” (HSAB) indicates that hard acids prefer binding to hard bases while soft acids prefer binding to soft bases. (Ayer, 2005; Koch,

2005b). Thus, the binding process of the four heavy metal cations onto the rhamnolipid biosurfactant is controlled by the relative softness or hardness of the two species. Table 4.6.3 shows the relative hardness and softness of some acids and bases.

**Table 4.6.3 Hard and soft acids and bases**

Acids or bases	Examples
Hard base	COOH
Intermediate base	NO <sup>-2</sup>
Soft base	C <sub>6</sub> H <sub>6</sub>
Soft Acid	Cd <sup>+2</sup> , Cu <sup>+2</sup>
Intermediate acid	Ni <sup>+2</sup> , Zn <sup>+2</sup>

Adapted from Koch, (2005a); Raize et al., (2004); Alfarra et.al., (2004).

According to the HSAB principle, cadmium and copper are soft acids, zinc and nickel are intermediate acids (Table 4.6.3). Thus, cadmium and copper (soft acids) can interact, most likely, with oxygen-containing carboxylate groups (hard base) to produce markedly ionic bonds. Zinc and nickel (intermediate acids) can produce fewer ionic bonds than can cadmium and copper. Since the binding

capacities for cadmium and copper to rhamnolipid are more than that for zinc and nickel, this supports the experimental trend.

Similarly, zinc is softer than nickel. This is probably because the *d* electrons of zinc do not participate in bonding (Cotton et al., 1999). These observations are supported by order of the binding capacities of metals ( $Zn^{+2} > Ni^{+2}$ ). Thus, the results are compatible with the “ Hard Soft Acid Base Principle ” (HSAB).

The crystal field principle can be applied to interpret the binding properties of copper, cadmium, zinc and nickel metal ions onto the rhamnolipid biosurfactant. This principle focuses mainly on how the energies of the *d* orbitals are affected by complex ion formation (Cotton et al., 1999; Chien et al., 2005). This is important because the energies of the *d* orbital of copper complex ions and the ones of cadmium, nickel and zinc ions can be explained by this principle.

According to the crystal field model (Cotton et al., 1999; Chien et al., 2005), the following assumptions are made:

- 1- Heavy metal ions are considered as point charges.
- 2- There is no interaction between metal orbital and rhamnolipid orbital.
- 3- The *d* orbitals on the metal all have the same energy in the free atom (Cotton et al., 1999; Yang et al., 2005).

According to this model, the difference in the binding behaviour of Cu (II) ,compared to Cd (II) and Zn (II), result from the change in the energies and the metal ion  $d$  orbital caused by the metal-rhamnolipid interactions. When a metal-rhamnolipid complex is formed, the degeneracy of these orbitals will be destroyed (Cotton et al., 1999; Stoilova, 2004). This means that the orbitals of each metal have different energies after the formation of a metal-rhamnolipid complex. In addition, this model considers that the crystal field stabilization energy (CFSE) for Cd (II) complex with  $d^{10}$  structure is equal to zero, and the (CFSE) of Cu(II) complex with  $d^9$  structure is higher than zero. Taking this into account, the complexation of Cu (II) is expected to be more stable than that of Cd(II) or Zn(II). Consequently, Cu(II) ions were preferred by the rhamnolipid biosurfactant relative to Cd(II), Ni (II), or Zn(II).

The Zeeman effect (Liboff, 2002), electron paramagnetic resonance studies (Griffiths, 2004; Cotton et al., 1999), and quantum mechanics (Liboff, 2002; Griffiths, 2004) can be applied to interpret the binding properties of copper, cadmium and zinc and nickel metal ions onto rhamnolipid biosurfactant. In many atoms, there exist several electronic configurations which have the same energy, so that transitions between different pairs of configurations correspond to a single line (Griffiths, 2004; Cotton et al., 1999). The presence of a magnetic field breaks the degeneracy, since it interacts in a different way with electrons with different quantum numbers, slightly modifying their energies. Thus, there are different

energies that give rise to several very close spectral lines. The Zeeman effect explains the possibility of the splitting of a spectral line into several components in the presence of a magnetic field (Liboff, 2002; Griffiths, 2004).

From a quantum mechanical point of view, an electron has a magnetic moment (Cotton et al., 1999; Liboff, 2002). When this magnetic moment placed in an external magnetic field, the electron can align itself parallel or antiparallel to the external field. The former is a lower energy state than the latter. To move between the two energy levels, the electron can absorb electromagnetic radiation (Liboff, 2002; Griffiths, 2004). Copper (II) has one unpaired electron and is paramagnetic (Cotton et al., 1999; Williams et al., 2006). Consequently, it can be attracted by a magnetic field possibly originating from the carboxyl groups of rhamnolipid biosurfactant (Cotton et al., 1999; Zvyagin et al., 2006). The Cd(II), Ni (II), and Zn(II) ions are much more stable since they do not have unpaired electrons. Consequently, they are diamagnetic; slightly repelled by a magnetic field (Cotton et al., Zvyagin et al., 2006). That may explain why the binding of Cu(II) was superior compared to Cd(II), Ni (II), and Zn(II) ions.

### **Summary**

Binding experiments were conducted to investigate the relative affinity of rhamnolipid for  $\text{Cu}^{2+}$ ,  $\text{Zn}^{2+}$ ,  $\text{Ni}^{2+}$ , and  $\text{Cd}^{2+}$  metal ions at equilibrium. The order of metal binding (molar basis) in the single, binary, and ternary systems was:  $\text{Cu}^{2+} > \text{Cd}^{2+} > \text{Zn}^{2+} > \text{Ni}^{2+}$ . Some chemical properties were assessed in order to

evaluate the binding order of the four heavy metal cations onto the rhamnolipid biosurfactant. The binding order were explained in terms of Pauling's electronegativity, the " Hard Soft Acid Base Principle " , the crystal field principle, the Zeeman effect, electron paramagnetic resonance studies, and quantum mechanics. In general, these chemical properties were complementary to interpret the binding order.

## 4.7 Waste Stabilization

Handling of contaminated solid waste with heavy metals while meeting the stringent regulations is considered to be a challenge. This is due to the lack of knowledge on the effectiveness and limitations of existing stabilization technologies. Among the many contamination groups, lead and cadmium are considered two of the most common contaminants found in contaminated sites (U. S. Environmental Protection Agency, 1997). The purpose of this work was to evaluate an existing technology, solidification/stabilization (S/S), in terms of its effectiveness and limitations to stabilize cadmium, lead and mixtures of heavy metal solid wastes.

There is a great concern about population problems related to hazardous wastes. The safety and acceptability of numerous widely used solid waste management practices are of serious concern from the public health point of view. The term pollution includes both the act of polluting and the consequences of that act. Waste describes unwanted residues that are usually perceived to be of negative value. Pollution may be defined as the introduction into the natural environment by humans of substances, materials or energy that cause hazards to human health, harm to living resources and ecological systems, damage to structures and amenities or that interfere with the legitimate uses of the environment. It is implicit in the definition that pollution only describes situations where unwanted effects occur (Royal Commission on Environmental Pollution, 1984).



Making health assessment of exposure to waste-site chemicals is a more complex task (OSHA, 1993; ATSDR, 1995). Exposures such as those found in occupational settings can involve exposures to relatively high chemical concentrations for a long duration and can be well-defined and quantified (NIOSH, 1996; ATSDR, 2002a). Exposures from waste sites can be infrequent, of unknown duration, and at low concentrations making them difficult to characterize and quantify. ATSDR's findings have pointed to health threats faced by people living near hazardous waste sites who drink contaminated water, eat contaminated fish, breath toxic fumes, or are otherwise exposed to hazardous substances (ATSDR, 2003; ATSDR, 2002b). ATSDR identifies the need for (1) health studies to be conducted, or (2) issuance of a public health advisory to recommend immediate actions to reduce, prevent, and eliminate exposure (ATSDR, 2003; ATSDR, 2002c).

The great concern about population problems related to hazardous wastes led to the development and implementation of some stringent regulations. Subtitle C of RCRA (Solid Waste Disposal Act, 1976) establishes 'a 'cradle to grave' regulatory scheme governing the treatment, storage, and disposal of hazardous waste' . The US Environmental Protection Agency (EPA) is authorized to delegate regulatory authority to individual states, provided the state's hazardous waste regulatory program is at least as stringent as RCRA Subtitle C. In the USA, most states have received approval from EPA to implement and enforce their own hazardous waste regulatory scheme. Pursuant to its rule-making authority under RCRA, EPA has also adopted literally hundreds of pages of

regulations, which classify wastes as either hazardous or non-hazardous (Solid Waste Disposal Act, 1984; US Environmental Protection Agency, 1994).

**Table 4.7.1 Summary of available technologies for waste treatment (Ragaini, 1994)**

<b>Technology</b>	<b>Sludge Media</b>	<b>Organic Waste</b>	<b>Inorganic Waste</b>
<b>Thermal</b>	Yes	Yes	No <sup>1</sup>
<b>Physical/Chemical</b>	Yes	Yes	Yes
<b>Biological</b>	Yes	Yes	No <sup>2</sup>
<b>S/S</b>	Yes	Limited	Yes

1: Thermal can be used but only for mercury and other volatile metals (Hospido, et al., 2005).

2: It can be used in special cases such as the conversion to Cr(III) which is more stable, less mobile and toxic than Cr(VI) but may be not good for large volumes of waste (Agrawal et al, 2006).

The most common available technologies for solid waste treatment are physical/Chemical, thermal, biological and solidification/stabilization (S/S) (Ragaini, 1994). The potential of using the above technologies for waste handling is summarized in Table 4.7.1. It illustrates that solidification/stabilization (S/S) treatment can be widely used for handling inorganic wastes. As a consequent, this technology is evaluated for its effectiveness in treating cadmium, lead and mixtures of heavy metal solid wastes in this study. The main objective of solidification/stabilization (S/S) processes is to improve physical and chemical properties of some wastes and contain hazardous constituents in a monolithic to prevent leaching of toxic materials into the environment. They have to be flexible enough to accommodate mixtures of contaminants and economical enough to be utilized for large volumes of wastes (Ragaini, 1994).

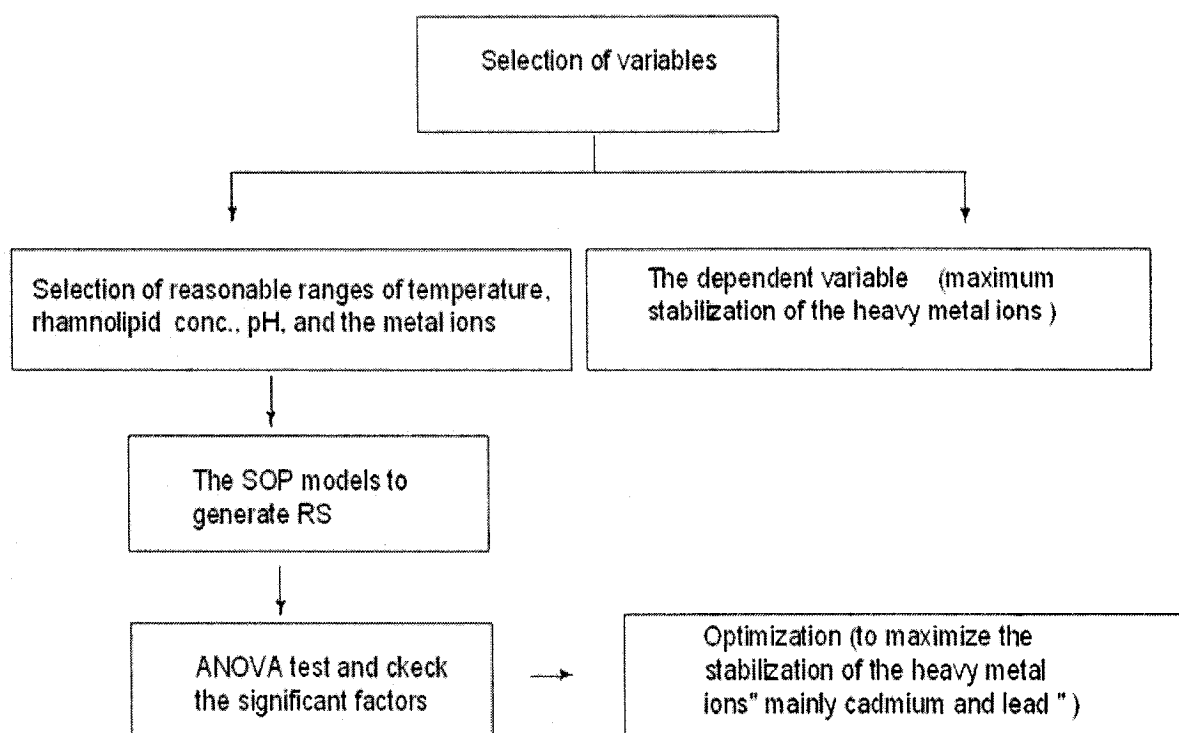
Alkaline matrices such as  $\text{Ca}(\text{OH})_2$ , and Portland cement are commonly used in waste conditioning because they are inexpensive, have an extensively documented history of use and draw upon readily-accessible technology (Carmalin et al., 2005). Their alkalinity greatly reduces the solubility of many inorganic toxic or hazardous inorganics and inhibits microbiological processes. Moreover, since these matrices require water for hydration, they readily incorporate wet wastes (Connor, 1990; Al-Ansary et al., 2004).

If water is not present in the waste material itself, it is added to the mixture in order to initiate the hydration reactions. These kinds of reactions take place in

order to ensure a proper bonding between the wastes and cement. Water is essential to initiate the hydration reactions but a broad range of water/cement ratios can be sustained. Water in excess of the minimum required for hydration increases fluidity (Taylor, 1990; Frohnsdorff, 1986). Also, it increases the porosity and the permeability of the hardened product. As a consequence, if low permeability barrier materials are required, w/c ratios need to be kept as low as possible consistent with other requirements. On the other hand, it may be desirable to maximize the ratio of waste to cement and, if the waste is wet or in solution, high w/c ratios may be more economic (Taylor, 1990; Frohnsdorff, 1986)

A variety of binders and/or additives have been utilized to improve the performance characteristics of cement (Glasser, 1987). Bentonite is the main component in many environmental areas. Bentonite is a unique clay mineral with very high swelling potential and water adsorption capacity. When wet, bentonite is the least permeable of all naturally occurring soil-like minerals (Alexiew, 2000; CETCO, 1994). A variety of polymers and epoxy materials and/or additives have been utilized to improve the handling of contaminated solid wastes (William 2003). Unconfined compressive strength is considered to be one of the commonly utilized physical tests in order to evaluate the solidification/stabilization (S/S) processes. Also, the leaching test is one of the famous chemical examinations for the solidification/stabilization (S/S) processes (EPA, 1989).

In order to efficiently achieve the ratios of binders, optimization by the response surface methodology approach was utilized. The operating parameters like initial rhamnolipid biosurfactant concentration, pressure difference, temperature, and required rejection ratio as independent variables while the maximum flow rate was designated as a dependent variable.

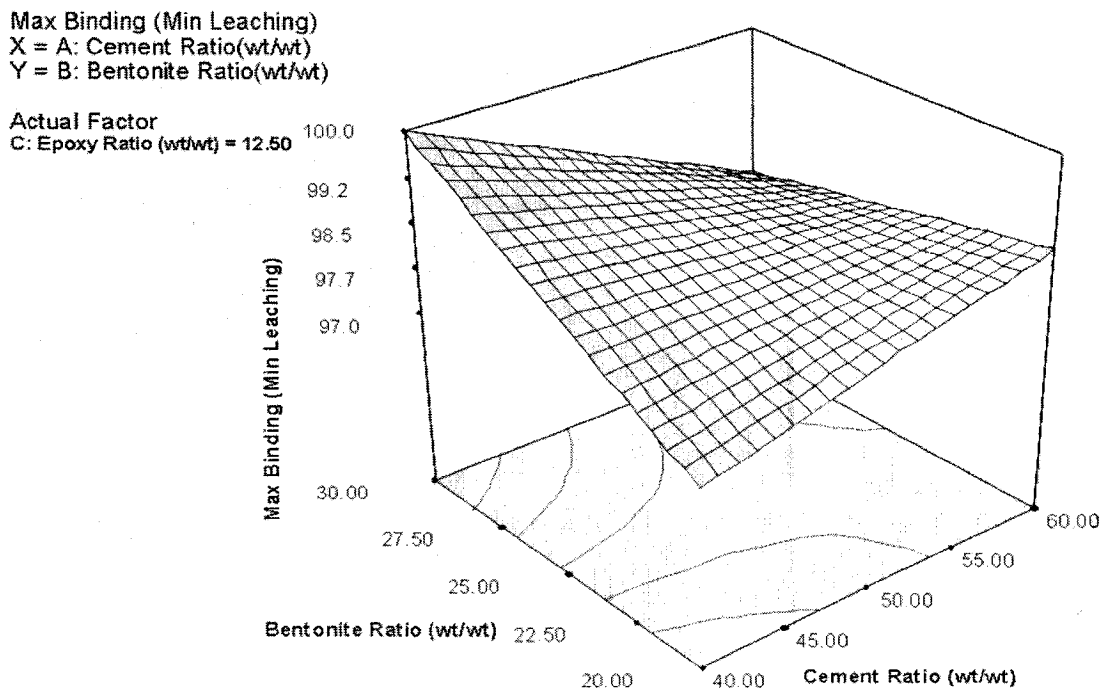


SOP = Second order polynomial ; RS = Response surface

Conc. = Concentration

**Figure 4.7.1. Experimental data flow to select the optimized ratios of binders.**

Second order polynomial (SOP) models were utilized as functions of the independent variables. An analysis of variance (ANOVA) was performed to determine which of the parameters significantly affected the dependent variable. Response surface (RS) and contour plots were generated as shown in Figures 4.7.2, to 4.7.5. Optimum regions of independent and dependent variables were achieved.



**Figure 4.7.2** Response surface obtained for bentonite ratios and cement ratios.

Max Binding (Min Leaching)  
 X = C: Epoxy Ratio (wt/wt)  
 Y = A: Cement Ratio (wt/wt)

Actual Factor  
 B: Bentonite Ratio (wt/wt) = 25.00

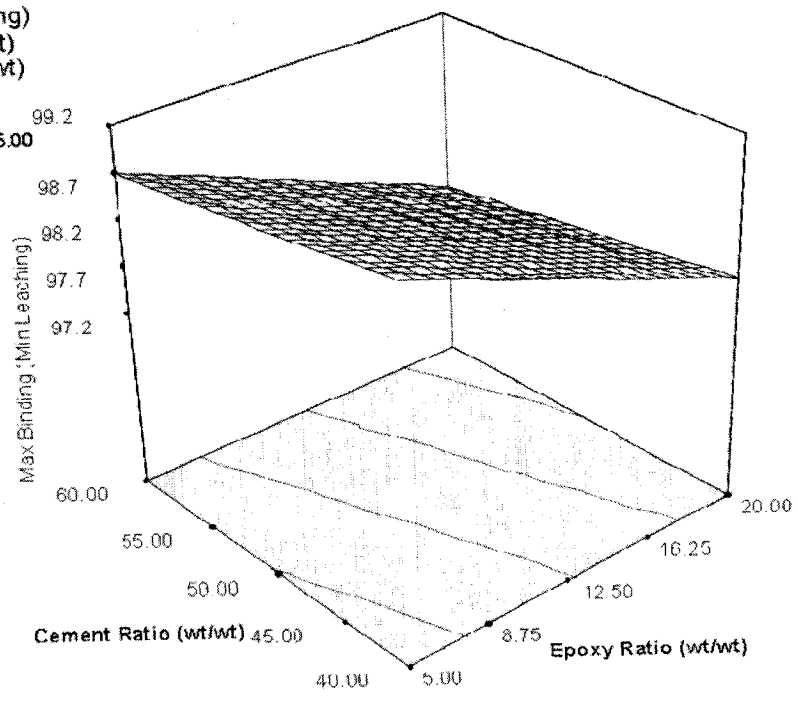


Figure 4.7.3 Response surface obtained for cement ratios and epoxy ratios.

Max Binding (Min Leaching)  
 X = B: Bentonite Ratio (wt/wt)  
 Y = A: Cement Ratio (wt/wt)

Actual Factor  
 C: Epoxy Ratio (wt/wt) = 12.50

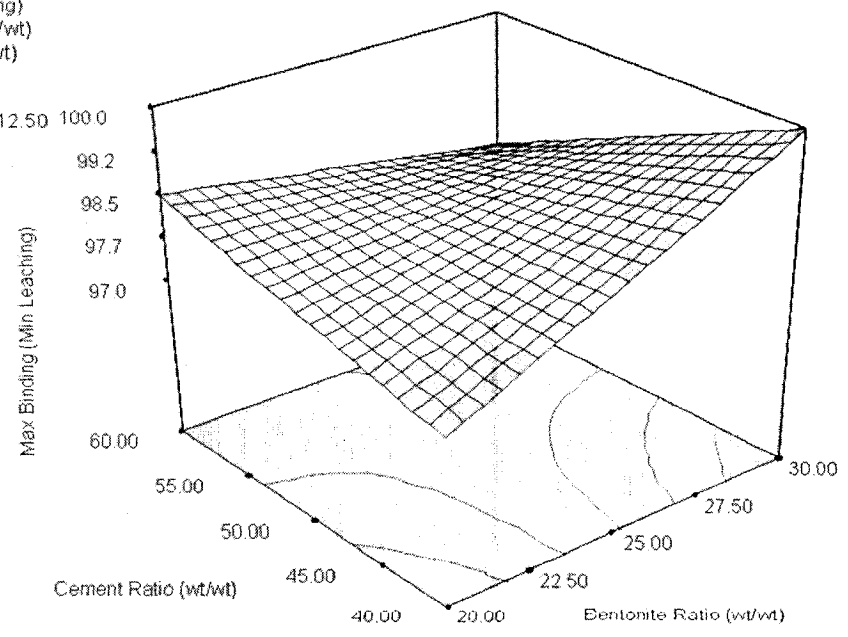
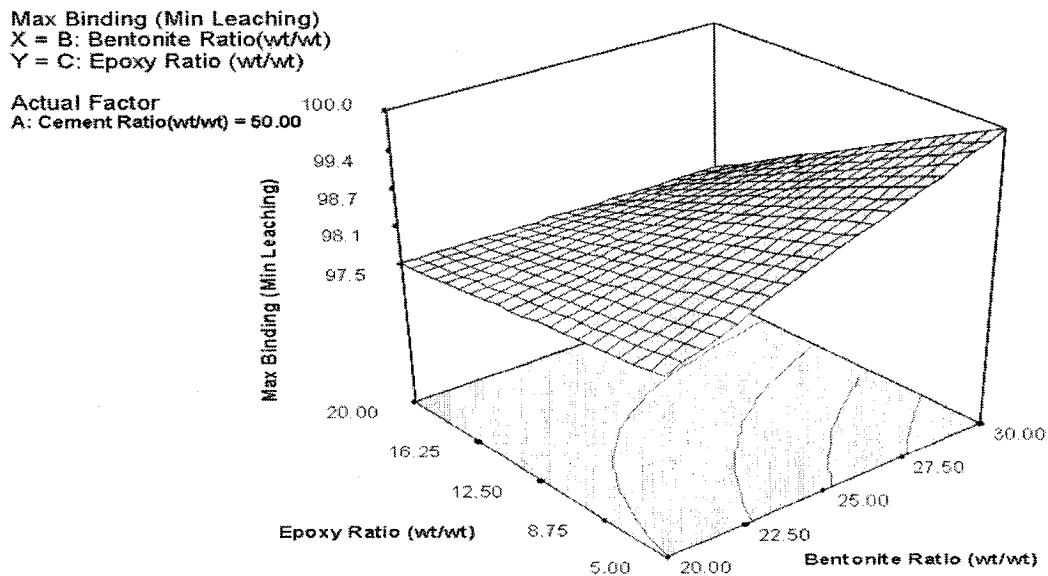


Figure 4.7.4 Response surface obtained for bentonite ratios and cement ratios.



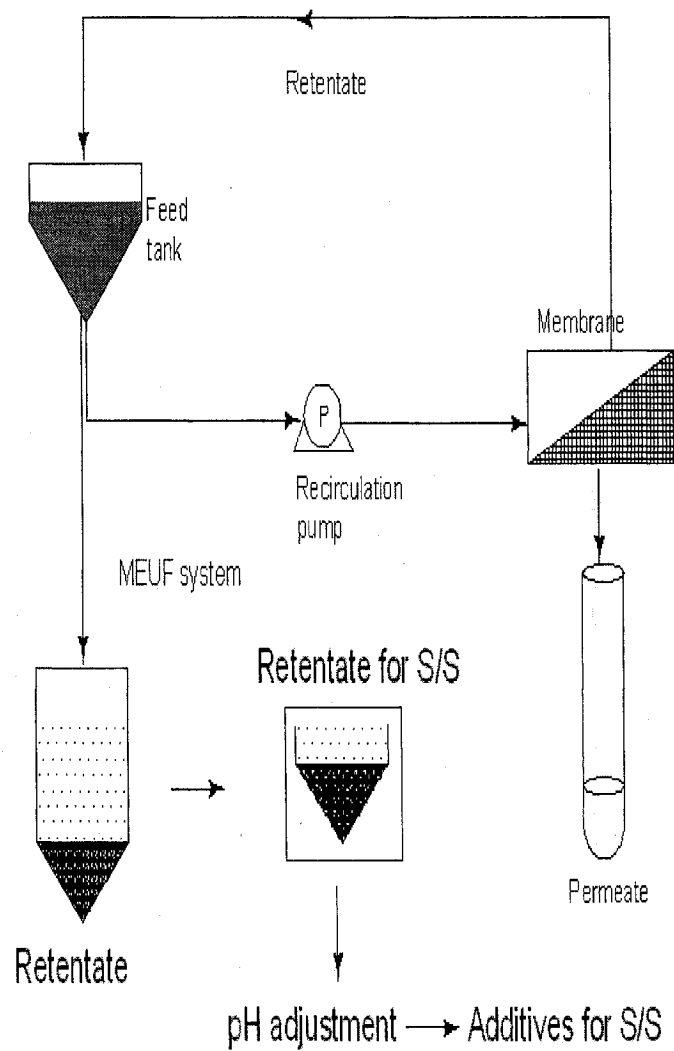
**Figure 4.7.5 Response surface obtained for epoxy ratios and bentonite ratios.**

The major objective was to select the optimized ratios of binders to achieve minimum leaching as a result of solidification/stabilization (S/S) processes. Figure 4.7.1 shows the experimental data flowsheet to select the optimized ratios of binders.

From Figures 4.7.2, to 4.7.5, it can be seen that the suggested ranges of binders were within 0.40 to 0.60, 0.20 to 0.30, and 0.05 to 0.20 (by weight) for cement, bentonite, and epoxy respectively. Separate validation experiments using the lab-scale UF system were performed at room temperature (temperature of  $25\pm 1^{\circ}\text{C}$ ) to verify these optimum conditions. The ultrafiltration system shown in Figure



4.7.6 was utilized. The consolidated residuum from rhamnolipid recovery and the water reuse was used as the basis for solidification/stabilization (S/S) process.



**Figure 4.7.6. Retentate for S/S and water recycle from MEUF system.**

Regarding cement, the most applicable cement/total binder ratios to solidification/stabilization (S/S) processes were within the range from 0.45 to 0.65 (by weight) in the case of solids from contaminated ground waters and within the range from 0.35 to 0.5 (by weight) in the case of solids from contaminated industrial wastewaters.

**Table 4.7.2 Leachate Quality Criteria for cadmium and lead, and their concentrations in TCLP leachates.**

<b>Element</b>	<b>Leachate Quality Criteria (mg/L)</b>	<b>Concentration in TCLP leachates (mg/L)</b>
Lead	0.5	N.D.
Cadmium	5.0	N.D.

N.D. : (not detected) means that lead and cadmium concentrations were less than 0.01 mg/L.

In the case of bentonite, the most applicable bentonite/total binder ratios to solidification/stabilization (S/S) processes were within the range from 0.20 to 0.25 (by weight) in case of solids from contaminated ground waters and within the

range from 0.3 to 0.35 (by weight) in the case of solids from contaminated industrial wastewaters. Regarding epoxy resin, the most applicable epoxy/total binders ratios to solidification/stabilization (S/S) processes were within the range from 0.07 to 15 (by weight) in the case of solids from contaminated ground waters and from 0.15 to 0.25 (by weight) in the case of solids from contaminated industrial wastewaters.

Using additives in cement-based solidification/stabilization (S/S) was investigated to stabilize heavy metal-contaminated solid wastes. The additives evaluated here were a combination of bentonite and epoxy resin as explained in Chapter 3 section 3.8. The toxicity characteristics leaching procedure (TCLP) (EPA, 1986) was selected as the main test for evaluating the leaching toxicity potential of the solid wastes. Table 4.7.2 shows the Leachate Quality Criteria for cadmium and lead (US EPA, 1990).

#### **TCLP Results of contaminated Solid Wastes**

The cadmium and lead concentrations in the contaminated solid wastes were within  $23.3 \pm 1$  to  $52.8 \pm 1.5$  and  $5.4 \pm 1$  to  $11.7 \pm 1$  mg/g for cadmium and lead, respectively. With regards to the results of TCLP of contaminated solid wastes, it can be seen from Table 4.7.2 that the leached cadmium and lead concentrations were much lower than USEPA's TCLP regulatory limits. Consequently, it was concluded that the solid wastes were not toxic and could be classified as non-

hazardous wastes. Thus, they could be disposed of in sanitary landfills without further treatment.

### **Summary**

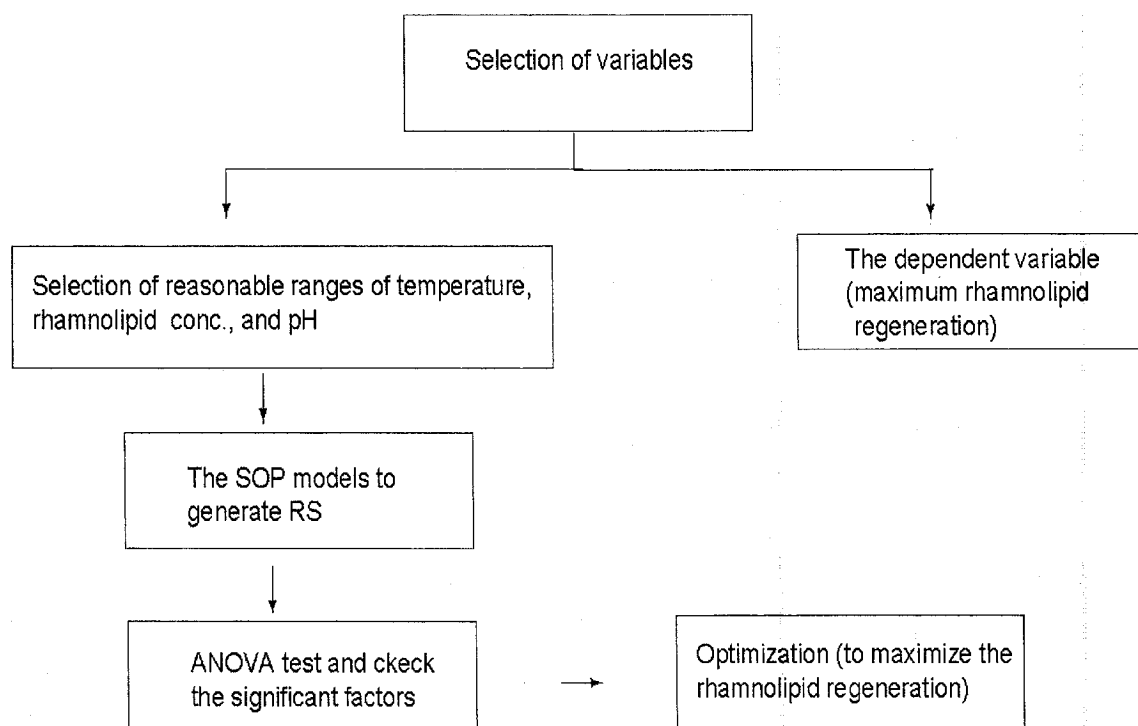
In this work, the solidification/stabilization (S/S) technology was successfully applied to stabilize solid wastes contaminated with heavy metals. The results indicated that the cadmium and lead concentrations in the leachates were much lower than USEPA's TCLP regulatory limits. Consequently, the wastes could be disposed of in sanitary landfills without further treatment.

## **4.8. Preliminary trials for recovery of rhamnolipid**

In order to make rhamnolipid-enhanced UF processes more economically feasible in wastewater treatment, methods have to be developed to regenerate and recycle the rhamnolipid. In this part of the study, following the MEUF lab-scale separation technique, separation of the heavy metal ions and biosurfactant in the retentate was performed. The retentates from rhamnolipid-enhanced UF of polluted wastewaters were containing rhamnolipid-metal ion complexes. The main objective of preliminary trials was to examine the efficiency of pH precipitation for separating the rhamnolipid biosurfactant from the retentates of MEUF technique for possible reuse.

In order to efficiently achieve the maximum rhamnolipid biosurfactant regeneration from the retentates of treated industrial wastewaters, optimization by the response surface methodology approach was utilized. The pH and temperature were considered as independent variables while maximum rhamnolipid biosurfactant regeneration was designated as a dependent variable. Based on these data, second order polynomial (SOP) models were utilized as functions of the independent variables. An analysis of variance (ANOVA) was performed to determine which of the parameters significantly affected the dependent variable. Response surface and contour plots were generated as shown in Figures 4.8.2, to 4.8.4. Optimum regions of independent and dependent variables were achieved.

The major objective was to maximize rhamnolipid biosurfactant regeneration. Figure 4.8.1 shows the experimental data flow to achieve the maximum rhamnolipid biosurfactant regeneration.



SOP = Second order polynomial ; RS = Response surface

**Figure 4.8.1. Experimental data flowsheet to achieve the maximum rhamnolipid biosurfactant regeneration.**

Max. Rhamnolipid Regtn. (%)  
 X = A: Temp. ( Deg. C )  
 Y = B: pH

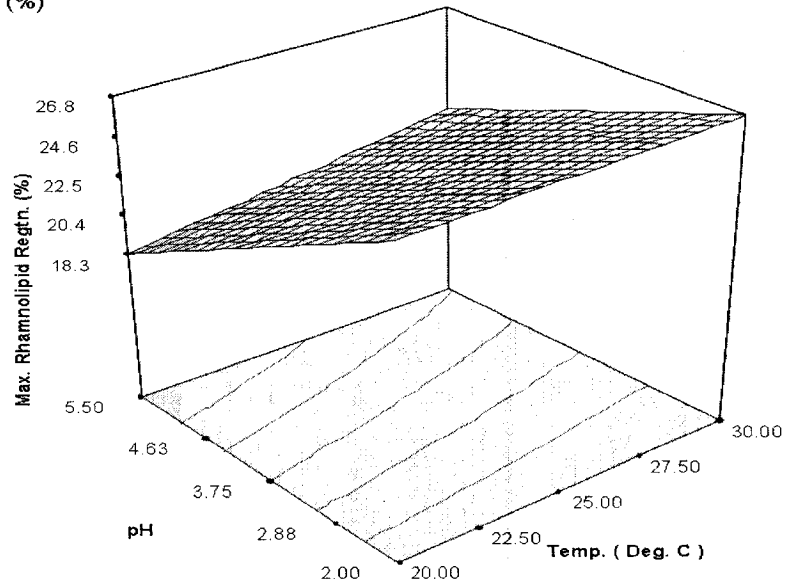


Figure 4.8.2. Response surface obtained for temperature and pH.

Max. Pptrn. for Regtn.  
 X = A: Rhamnol. C. ( m. mol. / L )  
 Y = B: Temp. (Deg. C.)

Actual Factor  
 C: pH = 3.75

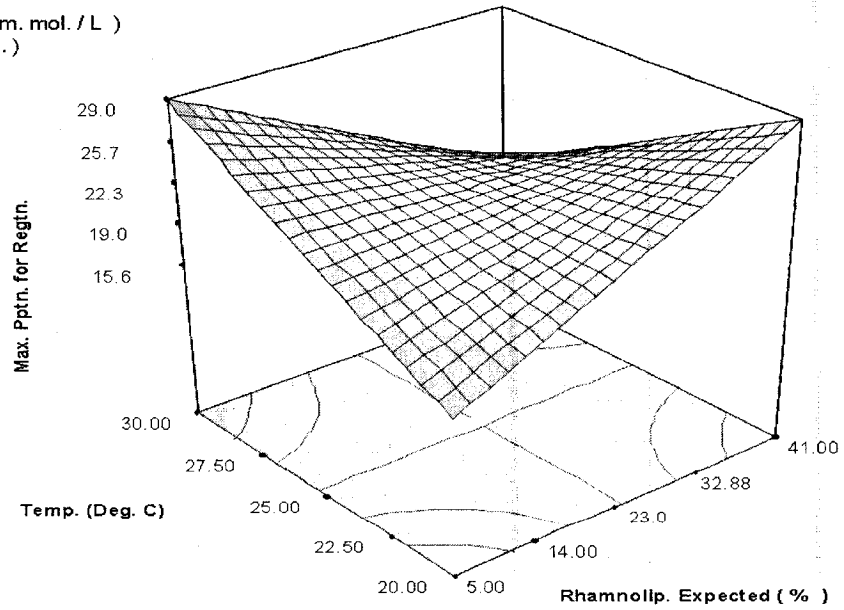
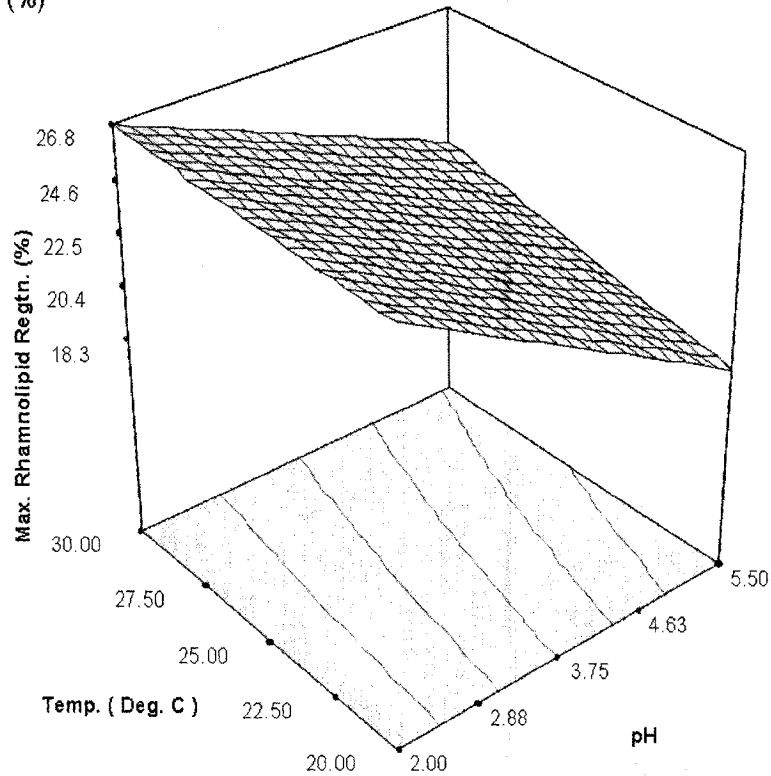


Figure 4.8.3. Response surface obtained for expected percentage of regeneration and temperature .

Max. Rhamnolipid Regtn. (%)  
X = B: pH  
Y = A: Temp. ( Deg. C )

Actual Factor  
C: Rhamnolip. Expcd ( % ) = 23.00

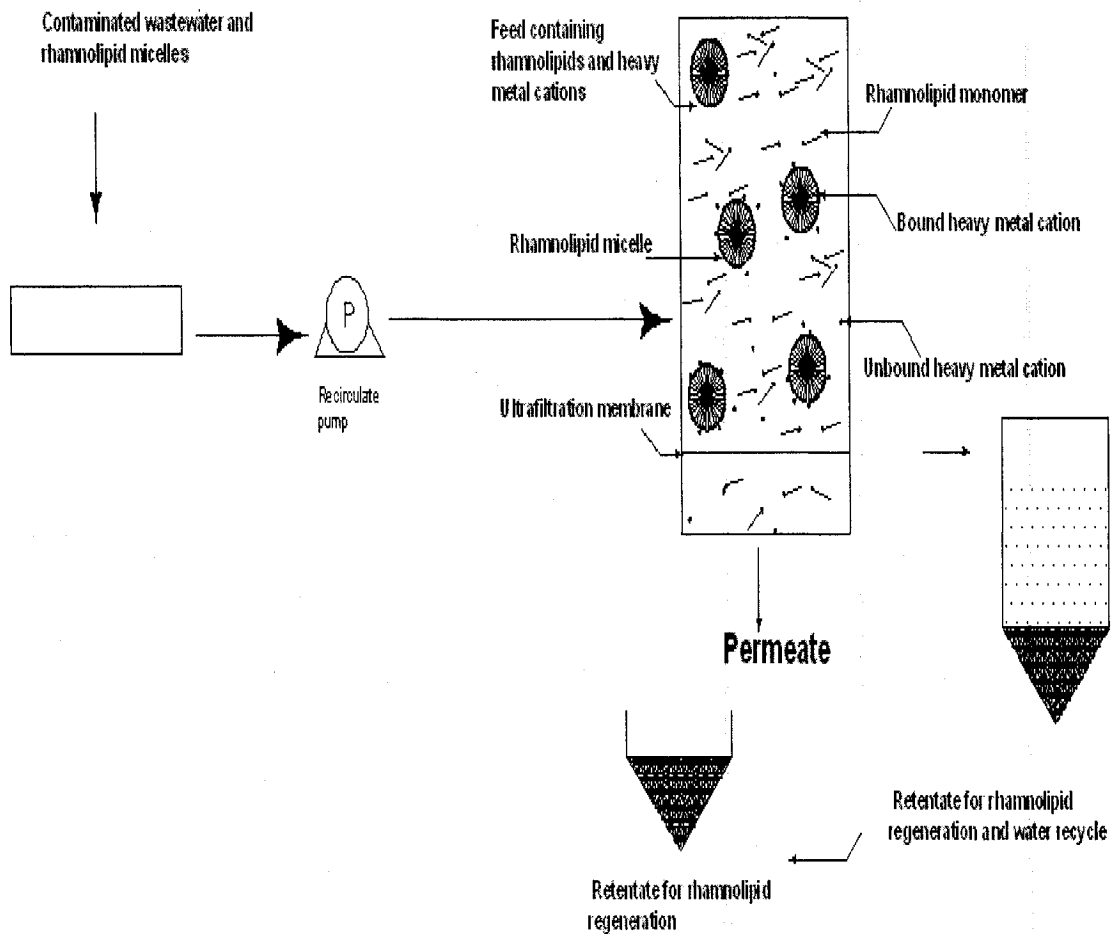


**Figure 4.8.4. Response surface obtained for pH and temperature.**

From Figures 4.8.2, to 4.8.4, it can be seen that the suggested ranges of pH from preliminary tests were within 2.0 to 5.5. The suggested ranges for the temperatures were from 20 °C to 30 °C and the suggested ranges for rhamnolipid biosurfactant recovery (%) were within about 18.3 to about 26.8 % of rhamnolipid present in the retentate initially before recovery. The ultrafiltration system shown in Figure 4.8.5 was utilized.



Separate validation lab-scale experiments were performed to verify these optimum conditions. The most applicable temperature was  $25 \pm 1$  °C, and pH values were about  $2.2 \pm 0.1$  to  $4.2 \pm 0.1$ .



**Figure 4.8.5. Schematic of the rhamnolipid-based UF process and the recovery step.**

The retentates from rhamnolipid enhanced UF of polluted wastewaters contained rhamnolipid-metal ion complexes with cadmium, zinc, copper and nickel. Figure 4.8.5 presents a schematic of the rhamnolipid-based UF process and the recovery step utilized in this work. The rhamnolipid rich retentate stream containing rhamnolipid-metal ion complexes went to a recovery step where it is mixed with an acidic solution. The upper aqueous layer with some small amounts of white suspension was taken for water reuse trials (Appendix B).

Regeneration trials were investigated that consisted of lowering the solution pH by nitric acid (0.5 N) to increase the extent of disassociation of metal-rhamnolipid complexes. By lowering the pH, the carboxylate groups can be protonated. This “breaks-up” the metal-rhamnolipid complexes (Cotton et al., 1999). Once most of the metal-rhamnolipid complexes were disassociated, the solution was then centrifuged in order to remove the precipitated rhamnolipid biosurfactant. The precipitating portion of the rhamnolipid biosurfactant could be seen visually (Appendix B). In order to verify the separated material, the pH was raised again to  $\text{pH } 6.9 \pm 0.1$  and then the concentration of rhamnolipid biosurfactant was determined by UV spectrophotometer at 235 nm (Appendix B).

The range of pH examined was from pH 2 to pH 5.5 (Figures 4.8.3 and 4.8.5). According to the separate validation experiments performed to verify the optimum conditions, the most pH values were about  $2.2 \pm 0.15$  to  $4.2 \pm 0.15$ . Within pH about 4.8 to 5.3, the rhamnolipid started to precipitate. The optimum

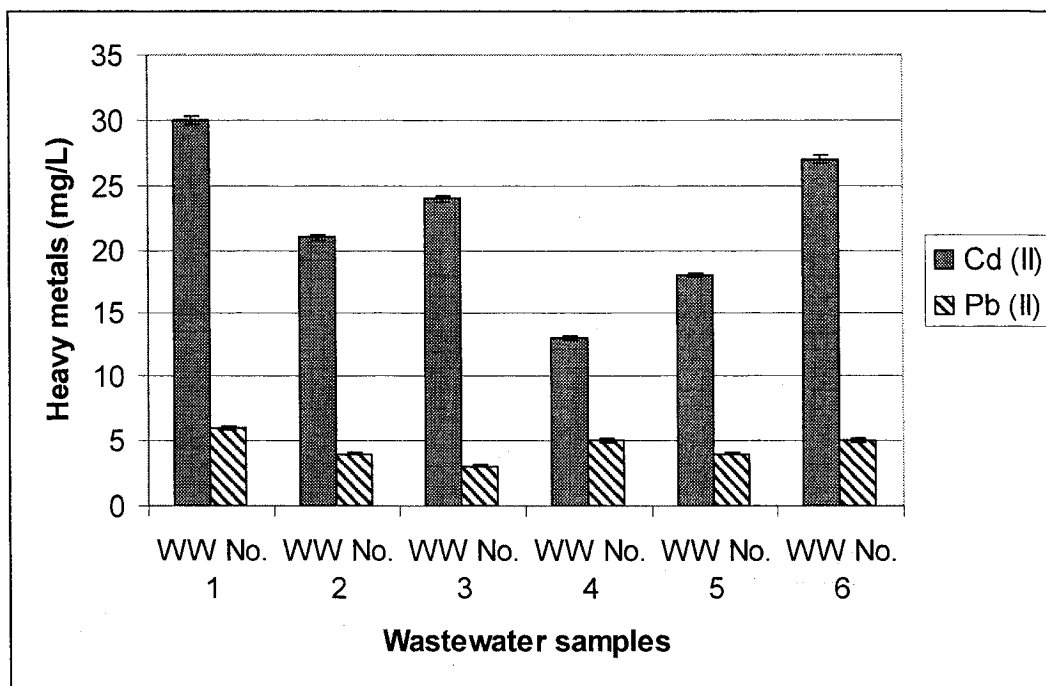
pH values for the regeneration method with rhamnolipid are shown in Table 4.8.1, where a sufficient fraction of nitric acid was able to precipitate most of the rhamnolipid available. The results for rhamnolipid biosurfactant, shown in Table 4.8.1, indicate that rhamnolipid recoveries from the retentates could be realized by nitric acid within a range of pH values of  $2.6 \pm 0.15$ .

**Table 4.8.1. Rhamnolipid biosurfactant regeneration results using nitric acid for precipitation.**

Wastewater	Optimum pH	Fraction of rhamnolipid recovered (%)
Wastewater 1	$2.62 \pm 0.1$	$12 \pm 1$
Wastewater 2	$2.54 \pm 0.1$	$15 \pm 1$
Wastewater 3	$2.70 \pm 0.1$	$15 \pm 1$
Wastewater 4	$2.49 \pm 0.1$	$18 \pm 1$
Wastewater 5	$2.57 \pm 0.1$	$16 \pm 1$
Wastewater 6	$2.41 \pm 0.1$	$13 \pm 1$

The initial rhamnolipid biosurfactant concentrations in the retentates were from  $14.3 \pm 1.1$  mmol/L about to  $31.7 \pm 1.3$  mmol/L. From the preliminary experiments and the predicted values by optimization, it can be seen that the predicted values (%) were within about 18.3 to 26.8 (Figures 4.8.2 to 4.8.4). Thus, the predicted values were higher than the experimental ones. This may be due to the formation

of stable metal-rhamnolipid complexes and metal-rhamnolipid precipitates in the UF permeates (Appendix B).



**Figure 4.8.6. Cadmium (II) and lead (II) concentrations in the raw feed wastewaters.**

It can be seen from Figure 4.8.6 and Table 4.8.1 that the optimum percentage for recovery was, probably, dependent upon the level of cadmium and /or lead in the raw feed wastewaters. In general, higher cadmium and lead concentrations caused a decrease in the percent of rhamnolipid recovered and an increase in the cost of rhamnolipid-based UF process.

Separate validation experiments were performed for the rhamnolipid, lead and/or cadmium alone and together. The results confirmed this observation. Lead and/or cadmium individually and together formed stable white complexes with the rhamnolipid. Higher cadmium and/or lead concentrations caused a reduction in the percentage of rhamnolipid recovered (Appendix B). These lab-scale trials of rhamnolipid regeneration may be utilized as preliminary studies for further detailed investigations. However, the limited rhamnolipid recovery percentage indicates that its large-scale use may not yet be economically feasible.

## **Summary**

In this work, preliminary trials for rhamnolipid recovery were examined. The optimum pH values for the regeneration recoveries from the retentates could be realized by nitric acid within a range of pH values of  $2.6 \pm 0.15$ . In order to verify the separated material, the pH was immediately raised again to  $6.9 \pm 0.1$  and the same UV method was utilized. The amounts recovered were within the range from  $12 \pm 1 \%$  to  $18 \pm 1 \%$  of the rhamnolipid concentration initially present in the retentate.

## 4.9. Preliminary trials for water reuse

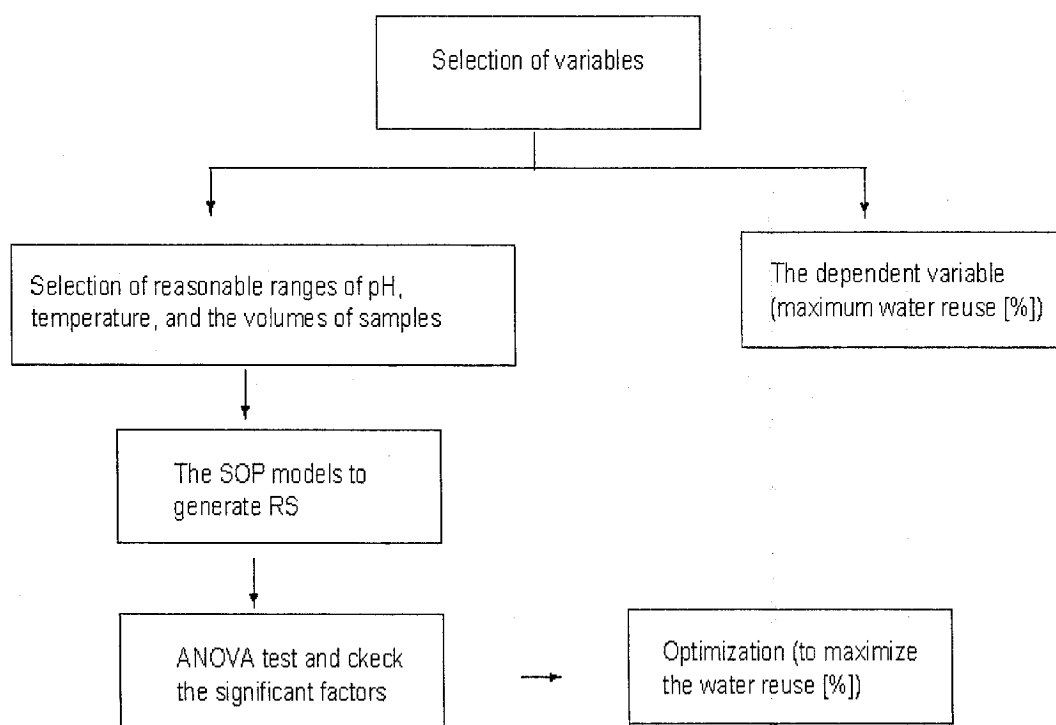
In order to minimize the liquid waste generated from metal refining, the rhamnolipid enhanced by UF was selected to treat diluted contaminated wastewaters (sections 4.4). Treatment of a large amount of the retentate as a liquid waste is costly. In order to adapt to tightening restrictions on effluents, rising costs of waste handling, and reduce water consumption, it is desirable to minimize the discharge of retentates from the process of rhamnolipid enhanced by UF. The goal of this study was to reduce water consumption in metal refining by reusing a fraction of the retentate of the rhamnolipid-based UF process as a good water management practice.

Water availability per capita has been steadily decreasing since the 1960s in Africa, Asia, and Latin America. In North America and Europe, this same parameter has remained fairly constant. Nevertheless, several regions that suffer chronic drought periods (e.g. Southern California in the United States and Andalusia in Spain) can undergo periodic water shortages (World Resources 1996-1997, FAO, 1993).

Industrial applications of wastewater is considered to be a marginal activity, on the average accounting for less than 6-15 of the overall reclaimed wastewater used in the industrial sector per year (World Resources 1996 -1997). China, Taiwan and Denmark essential industrial reuse programs are mainly greater towards industrial wastewater recycling (Renaud et al., 1997). Thus, it is

desirable to reduce water consumption in metal refining by reusing a fraction of the retentate of the MEUF process as a useful application of industrial wastewater recycling.

In order to efficiently choose the optimum percentage of reusable water from the retentate of treated wastewaters, optimization by the response surface methodology approach was utilized.



SOP = Second order polynomial ; RS = Response surface

**Figure 4.9.1. Experimental data flow to select the optimized ratios of water reuse.**

The pH, percentage of water available in the retentate, and temperature were considered as independent variables while maximum rhamnolipid biosurfactant regeneration was designated as a dependent variable.

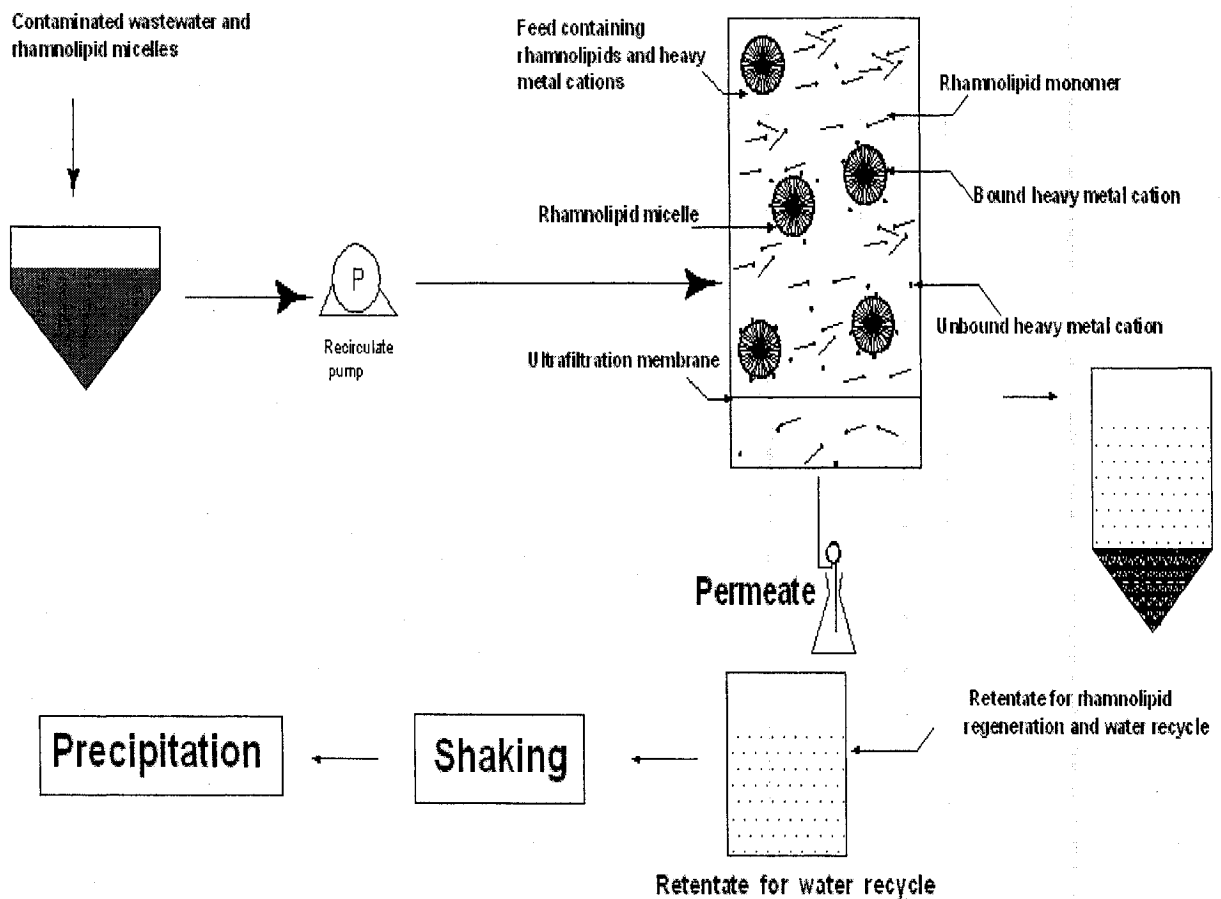
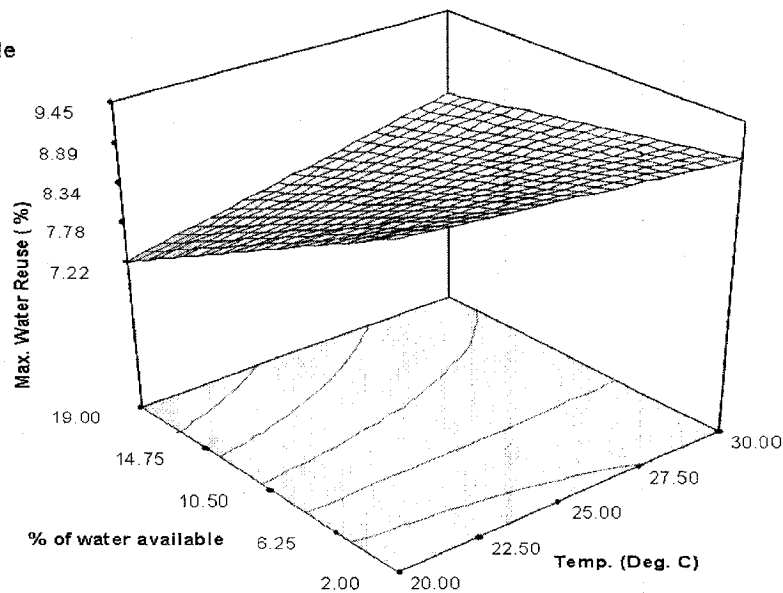


Figure 4.9.2. Schematic of the rhamnolipid-based UF process and the water reuse step.



Second order polynomial (SOP) models were utilized as functions of the independent variables. An analysis of variance (ANOVA) was performed to determine which of the parameters significantly affected the dependent variable. Response surface and contour plots were generated as shown in Figures 4.9.3 to 4.9.5. Optimum regions of independent and dependent variables were achieved. The major objective was to maximize water reuse. Figure 4.9.1 shows the experimental data flow to achieve the maximum water reuse.

Max. Water Reuse (%)  
 X = A: Temp. (Deg. C)  
 Y = B: % of water available  
 Actual Factor  
 C: pH = 6.81



**Figure 4.9.3. Response surface obtained for temperature and percentage of water available in the retentates.**

Max. Water Reuse (%)  
 X = C: pH  
 Y = A: Temp. (Deg. C)

Actual Factor

B: % of water available = 10.50

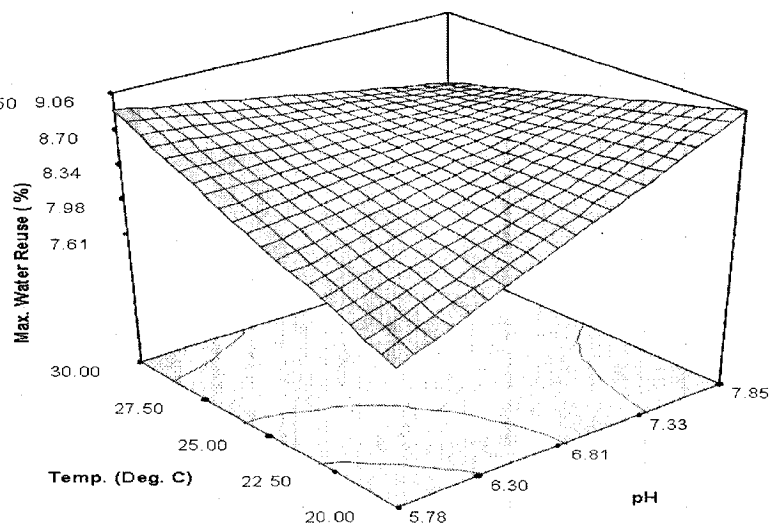


Figure 4.9.4. Response surface obtained for pH and temperature.

Max. Water Reuse (%)  
 X = B: % of water available  
 Y = A: Temp. (Deg. C)

Actual Factor

C: pH = 6.81

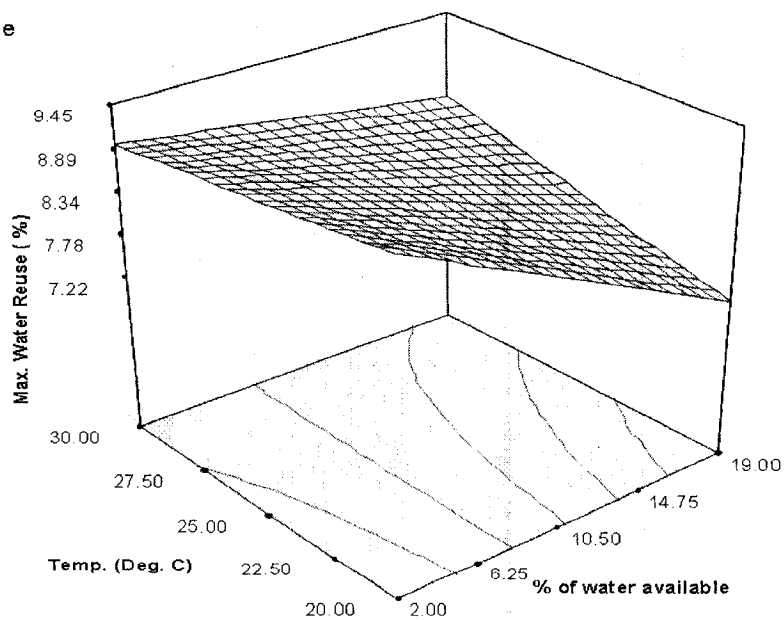


Figure 4.9.5. Response surface obtained for percentage of water available in the retentates and temperature.

Figure 4.9.2 illustrates a schematic of the rhamnolipid-based UF process and the water reuse step utilized in this work. The rhamnolipid rich retentate stream containing rhamnolipid-metal ion complexes went to a recovery step where it was mixed with an acidic solution. The upper aqueous layer with some small amounts of white suspension was taken for water reuse trials (Appendix B). From Figures 4.9.3. to 4.9.5, it can be seen that the suggested ranges of optimum temperature were from about 23 to 26 °C.

**Table 4.9.1. Fractions of recovered or reused water from different industrial wastewaters.**

Wastewater	Fraction of water recovered as a permeate (%)	Fraction of water reused (%)	Total recovered and reused water (%)
Wastewater 1	73 ± 3.2	4.7 ± 0.2	77.7 ± 3.4
Wastewater 2	73 ± 3.1	4.9 ± 0.2	77.9 ± 3.3
Wastewater 3	73 ± 3.2	5.1 ± 0.1	78.1 ± 3.3
Wastewater 4	73 ± 3.1	5.2 ± 0.3	78.2 ± 3.4
Wastewater 5	73 ± 3.1	5.4 ± 0.1	78.4 ± 3.2
Wastewater 6	73 ± 3.2	4.8 ± 0.1	77.8 ± 3.3

The suggested ranges of pH were within 5.78 to 7.85. In addition, the suggested ranges for water available in the retentate (%) were from about 2 to 19, and the suggested ranges for water reuse (%) were within about 7.22 to 8.90 based on the original water volume. Separate validation lab-scale experiments were performed to verify these optimum conditions.

The most applicable temperature was  $25 \pm 1$  °C. The upper aqueous layer with some small amounts of white suspension was taken for water reuse trials (Appendix B). Water reuse trials were investigated that consisted of separation and shaking the upper aqueous layer with some small amounts of white suspension of the retentate. Leaving the solution to precipitate enabled the supernatant water to be separated and to be reused. The most possible portions of the reusable waters were illustrated in Table 4.9.1.

The fraction of water recovered as a permeate (%) was about 73 (Chapter 4, section 4, Table 4.4.2 ). The data indicate that possible portions of the reusable waters from the retentates could be realized within the range from  $4.7 \pm 0.2$  (%) to  $5.4 \pm 0.10$  (%) based on the original water volume in the feed. The percentages of the retentate were from  $16.3 \pm 2$  % to  $18.4 \pm 3$  % based on the total feed water. It can be seen that the possible portions of total reusable waters from the permeates and retentates could be realized within the range from  $77.7 \pm 3.4$  (%) to  $78.4 \pm 3.2$  (%) based on the original water volume. From the

preliminary experiments and the predicted values by optimization, it can be seen that the predicted values (%) based on the original water volume for the reusable waters from the retentates were within about 7.2 (Figure 4.9.3) to 8.9 (Figures 4.9.4 and 4.9.5). Thus, the predicted values were higher than the experimental ones. This may be due to the formation of stable metal-rhamnolipid complexes and metal-rhamnolipid precipitates in the UF retentates. In most cases, there were suspended stable white complexes that reduced the portion for water reuse (Appendix C).

After the treatment to remove the biosurfactant by centrifugation, some preliminary trials indicated that the only available portions for water reuse were within the range from  $3.3 \pm 0.2$  (%) to  $3.9 \pm 0.2$  (%) based on the original water volume. This, probably, was due to the stable complexes that were suspended reduced the chance for water reuse (Appendix C).

## Summary

In this work, preliminary trials for water reuse were examined. The main idea was to separate and shake the top layer of the retentate. The percentages of reusable waters that could be achieved from the retentates were within the range from  $4.7 \pm 0.10$  % to  $5.4 \pm 0.15$  % of the total feed water. The percentages of the retentate were from  $16.3 \pm 2$  % to  $18.4 \pm 3$  % of the total feed water. The portions of the water recovery in permeates were about  $73 \pm 3.1$  %. Alternative

tests for water recovery from the surfactant recovery process were investigated. However, it gave only 3.3 to 3.9 (%) recovery based on the original water volume.

## 4.10 Data quality and the lack of fit analysis

The aim of this work was to apply advanced mathematical approaches in order to examine data quality from the rhamnolipid-based UF. To achieve this aim, the objectives of this study were to examine the normality assumption for the data sets and the lack of fit. To investigate the need for mathematical transformation was one of the objectives as well.

Appendix E shows the examination of data quality, the normality assumption for the data sets, the lack of fit, and the need for mathematical transformation. The 95 % confidence interval level was applied. From the data presented and their analysis presented in appendix E, it can be seen that there was no need for the mathematical transformation, there were no outliers detected for the data sets, and the normality assumption was satisfied for the data sets.

### Summary

In this work, advanced mathematical analysis was applied in order to examine data quality, the normality assumption for the data sets, the lack of fit, and the need for mathematical transformation. The 95 % confidence interval level was utilized. The results indicated that the normality assumption was satisfied for the data sets and there were no outliers detected for the data sets.

## CHAPTER 5

### Conclusions

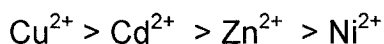
The following conclusions can be drawn from this work:

- The effects of different major operating conditions on the system performance of rhamnolipid-enhanced ultrafiltration were investigated for different metal ions (lead, copper, zinc, nickel and cadmium). Various rhamnolipid biosurfactant concentrations were examined to determine the optimal conditions for removal of metal ions from solution with the maximum flux using membranes with molecular weight cut off (MWCO) of 30,000 Da and 10,000 Da. The results indicated that that the initial rhamnolipid biosurfactant concentration has a major effect on metal removal. Also, the initial rhamnolipid biosurfactant concentration and transmembrane pressure ( $P_{tm}$ ) temperature have major effects on permeate flux. The response surface methodology (RSM) was very useful to determine the optimal factors that have significant effects on rhamnolipid-based UF system.
- The removal of the metal ions present in contaminated industrial wastewaters and groundwaters can be best achieved by applying optimal conditions, including surfactant-to-metal (S/M) ratio. In this research, the optimal surfactant-to-metal (S/M) ratio for heavy metal removal (> 99% rejection ratio) from lab-scale study was found to be around  $2 \pm 0.1$  for the rhamnolipid-based UF system.



- The Gouy-Chapman model was successfully applied to the binding of heavy metal ions to rhamnolipid biosurfactant in a MEUF system based on a charge-to-charge interaction. The Poisson-Boltzmann equation was utilized for this purpose. The electrostatic approach was relevant for this kind of interaction between the positive charges from heavy metal cations, and the negative charges of the carboxyl groups of rhamnolipid biosurfactants.
- The optimal operating conditions were utilized to treat polluted industrial wastewaters using membranes with molecular weight cut off (MWCO) of 30,000 Da and 10,000 Da. The rhamnolipid-enhanced ultrafiltration system was shown to be effective in treating six actually contaminated wastewaters from metal refining industries. The levels of heavy metal concentrations in the permeates were less than that of the federal discharge limits for the metals under investigation.
- The results produced from bench-scale MEUF experiments and the optimized operation conditions were investigated in order to provide an efficient method that can be used to perform metal ions separation from polluted ground waters. Using membranes with a molecular weight cut off (MWCO) of 30,000 Da and 10,000 Da the rhamnolipid biosurfactant were effective in treating three contaminated ground waters. More than 99.0 % removal efficiency was achieved using both membranes. The permeates were compatible with the standards of both Quebec and the USEPA.

- The binding behavior of the four heavy metal ions individually and collectively at pH  $6.9 \pm 0.1$  onto the rhamnolipid biosurfactant (> 99% rejection ratio) using the two membranes was evaluated. Regarding the individual single component systems, two-component systems and three-component systems the rhamnolipid biosurfactant was able to bind the heavy metal ions with the order (molar ratio) copper followed by cadmium, zinc and nickel, respectively.



- Solidification/stabilization (S/S) technology was used to stabilize of solid wastes contaminated with heavy metals. It was found that the leached cadmium and lead concentrations were much lower than USEPA's TCLP regulatory limits (0.5 and 5.0 mg/L for cadmium and lead, respectively). Consequently, it was concluded that the solid wastes were not toxic and could be classified as non-hazardous wastes. Thus, they could be disposed of in sanitary landfills without further treatment.

- The equilibrium isotherms were determined for the single-component experimental system. The data have been analyzed for each metal ion using langmuir, Freundlich, and linear adsorption model. The Freundlich isotherm model was demonstrated to provide the best correlation for the binding of the metal ions onto the rhamnolipid biosurfactant. The characteristic parameters for the Freundlich model were determined. The  $K_F$  values were found to be from 1.6

to 6.1 and from 1.3 to 4.2 for the membranes with molecular weight cut off (MWCO) of 10,000 Da and 30,000 Da, respectively. Also, the b values were found to be from 17.1 to 22.8 and from 8.6 to 13.9 for the membranes with molecular weight cut off (MWCO) of 10,000 Da and 30,000 Da, respectively.

- Preliminary trials for rhamnolipid recovery and water reuse were examined. The optimum pH values for the regeneration recoveries from the retentates could be realized by nitric acid within a range of pH values of  $2.6 \pm 0.15$ . The amounts recovered were within the range from  $12 \pm 1 \%$  to  $18 \pm 1 \%$  of the rhamnolipid concentration initially present in the retentate. Similarly, the percentages of reusable waters that could be achieved from the retentates were within the range from  $4.7 \pm 0.10 \%$  to  $5.4 \pm 0.15 \%$  of the total feed water. The percentages of the retentate were from  $16.3 \pm 2 \%$  to  $18.4 \pm 3 \%$  of the total feed water. The portions of the water recovery in permeates were about  $73 \pm 3.1 \%$ .

# CHAPTER 6

## Original contributions

The key contributions to scientific knowledge may be summarized as follows:

- This work proposes a novel methodology for the treatment of groundwaters and industrial wastewaters which have been contaminated by heavy metals, using rhamnolipid-based ultrafiltration. The operating conditions for the methodology have been mathematically optimized by response surface (RS). This is a wholly new approach to the optimization of techniques of water treatment via UF, and opens up many possibilities for future improvements to other UF methodologies. It is unlikely that other traditional approaches to process development, which vary one factor at a time, would be as successful in finding the optimum set of operating conditions as quickly or effectively as RS.
- The electrostatic mechanism was introduced by evoking a charge-to-charge interaction between the positive charges from heavy metal cations and the negative charges of the carboxyl groups of rhamnolipid biosurfactants in the MEUF system.
- The Gouy-Chapman model was used to explain the rhamnolipid/heavy metal interactions, as described by the Poisson-Boltzmann equation.

- The order of relative binding in the single, binary, and ternary systems of  $\text{Cu}^{2+}$ ,  $\text{Zn}^{2+}$ ,  $\text{Ni}^{2+}$ , and  $\text{Cd}^{2+}$  ions (molar ratio) onto the rhamnolipid at equilibrium using the two UF membranes was evaluated, and explained in terms of chemical properties.
- The equilibrium isotherm data were analyzed for  $\text{Cu}^{2+}$ ,  $\text{Zn}^{2+}$ ,  $\text{Ni}^{2+}$ , and  $\text{Cd}^{2+}$  ions using the Langmuir, Freundlich, and linear adsorption models. The Freundlich constants for rhamnolipid-based UF systems were determined, based on experimental data using the two UF membranes.
- A complete system of contaminated water treatment, stabilization of contaminated solid wastes, rhamnolipid recovery for possible reuse, and reduction of water consumption was established. The lab-scale work reported here can be utilized as preliminary studies for further pilot/large-scale detailed investigations.

## CHAPTER 7

### Recommendations for future research

1. Further investigate the feasibility of pilot-scale rhamnolipid-enhanced UF process for groundwaters and wastewaters.
2. Evaluate the rhamnolipid-based ultrafiltration process for removal of other contaminants, such as mixtures of heavy metals and organic contaminants, from groundwaters and wastewaters and determine the effect of organic contaminants on the removal heavy metals (in other words, to determine if there is competition between the metals and organic compounds).
3. Examine the rhamnolipid-based ultrafiltration technology for removal of other heavy metals from groundwaters and wastewaters.
4. Further study the UF modeling of heavy metal removal with rhamnolipid-based ultrafiltration.
5. Further investigate the economic feasibility of industrial-scale rhamnolipid-enhanced UF process.
6. An all-atom molecular dynamics simulation of a water-solvated micelle containing rhamnolipid biosurfactant is suggested for further investigation. Structural properties of the micelle and heavy metal ions could be evaluated.

## CHAPTER 8

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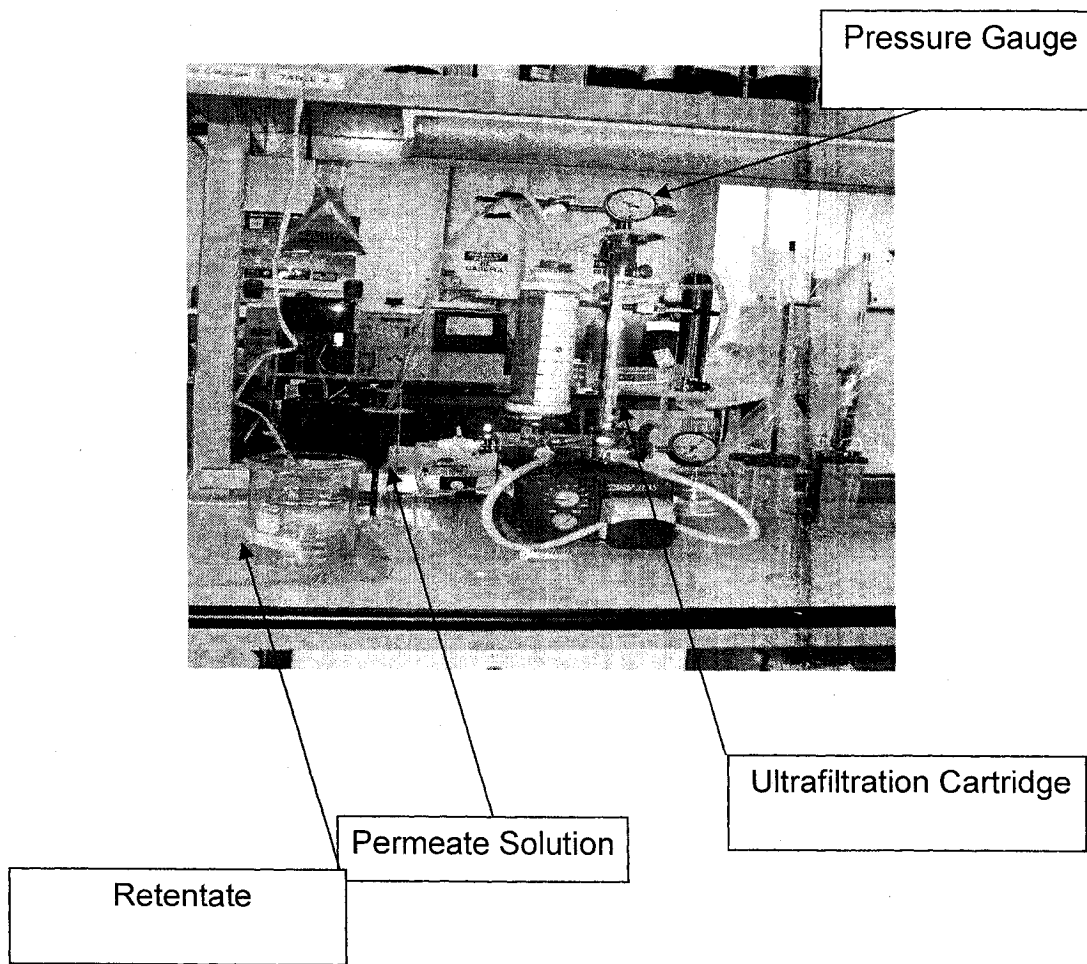
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**APPENDIX A: A Photograph of the UF membrane system used; cleaning procedures for the UF membrane system, and Isotherms of the Langmuir model and the linear isotherm model for zinc, copper, nickel, and cadmium.**



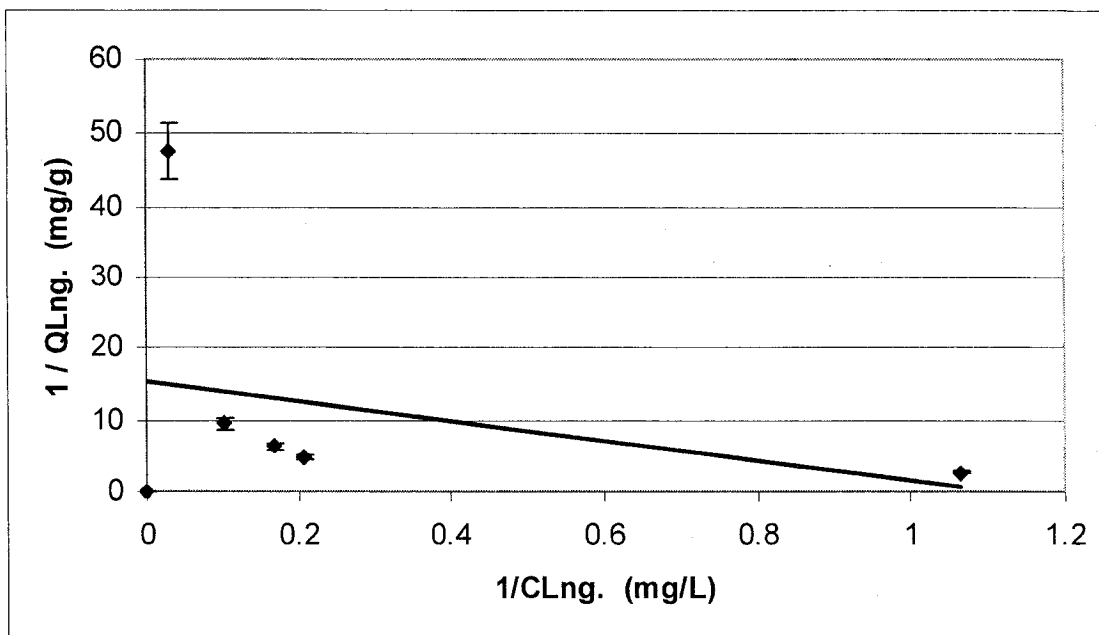
A Photograph of the UF membrane system used.

### **Cleaning procedures for the UF membrane system**

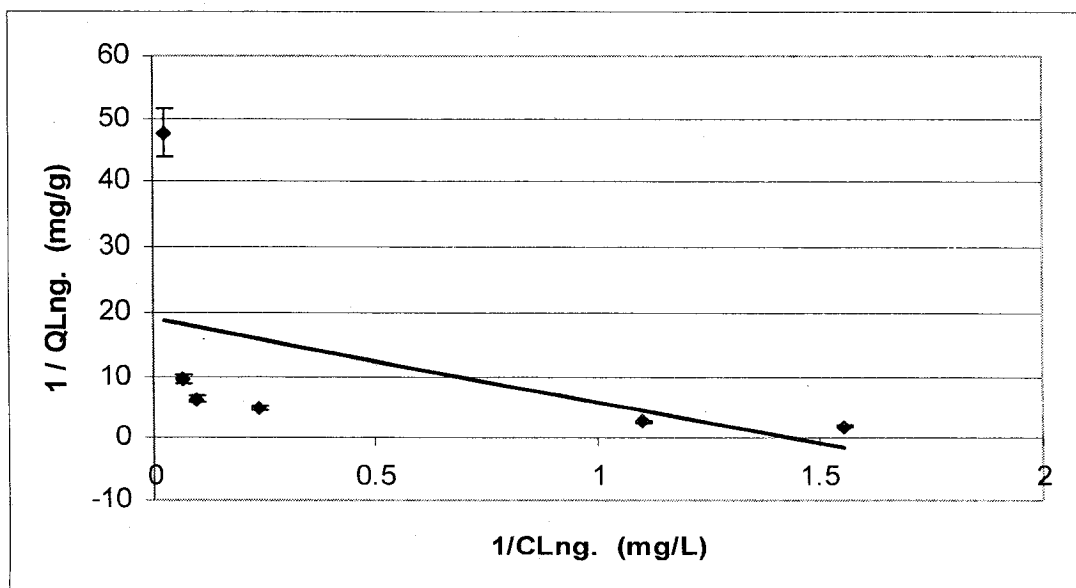
The cleaning procedures for cross-flow bench-scale system are summarized as follows:

- (1) Pump the retentate out,
- (2) Flush the system with double-distilled water,
- (3) Recirculate 0.5N NaOH at 50°C for one hour,
- (4) Flush with double-distilled water,
- (5) Recirculate NaOCl at 50°C (pH 10 -11) for one hour,
- (6) Flush the system with double distilled water, and
- (7) Analyze the collected permeate sample for residual metal ions.

**A- 3 Isotherms of the Langmuir model and The linear isotherm model for zinc, copper, nickel, and cadmium.**



**Figure A.3.1a Langmuir isotherms for zinc onto rhamnolipid for the membranes of 10,000 MWCO, temperature = 25±1 °C and transmembrane pressure = 69 kPa.**



**Figure A.3.1b Langmuir isotherms for zinc onto rhamnolipid for the membranes of 30,000 MWCO, temperature = 25±1 °C and transmembrane pressure = 69 kPa.**



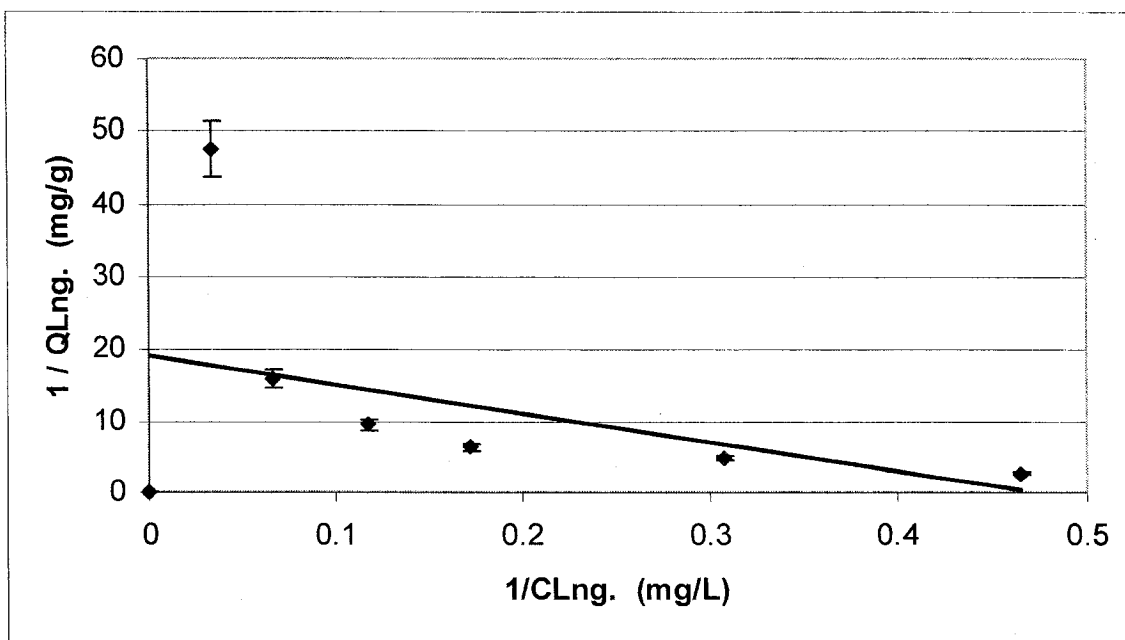


Figure A.3.2a Langmuir isotherms for copper onto rhamnolipid for the membranes of 10,000 MWCO, temperature =  $25 \pm 1$  °C and transmembrane pressure = 69 kPa.

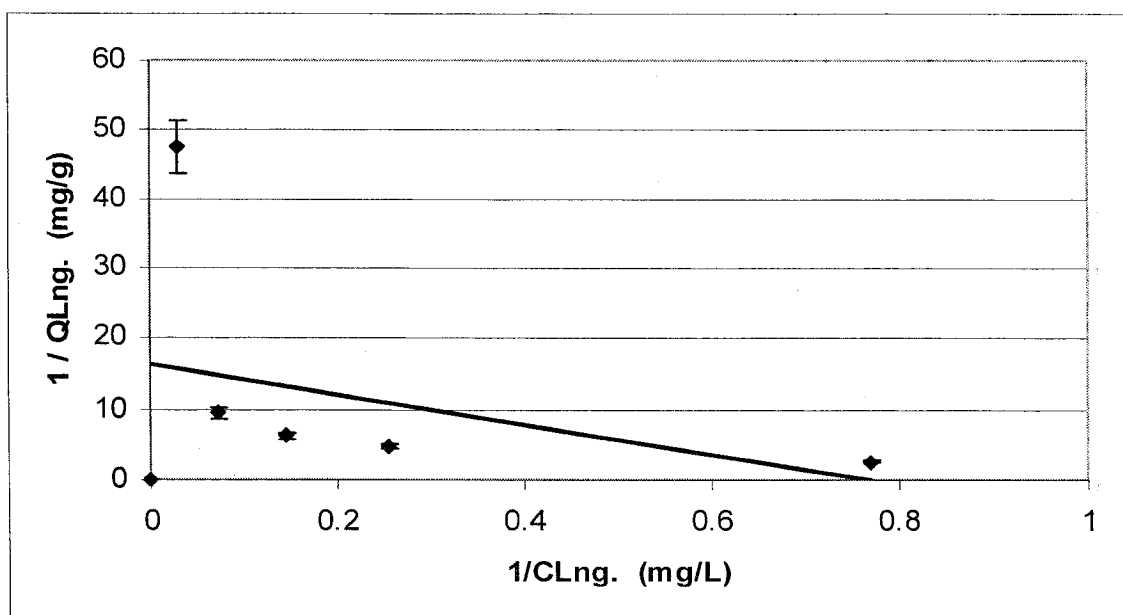


Figure A.3.2b Langmuir isotherms for copper onto rhamnolipid for the membranes of 30,000 MWCO, temperature =  $25 \pm 1$  °C and transmembrane pressure = 69 kPa.

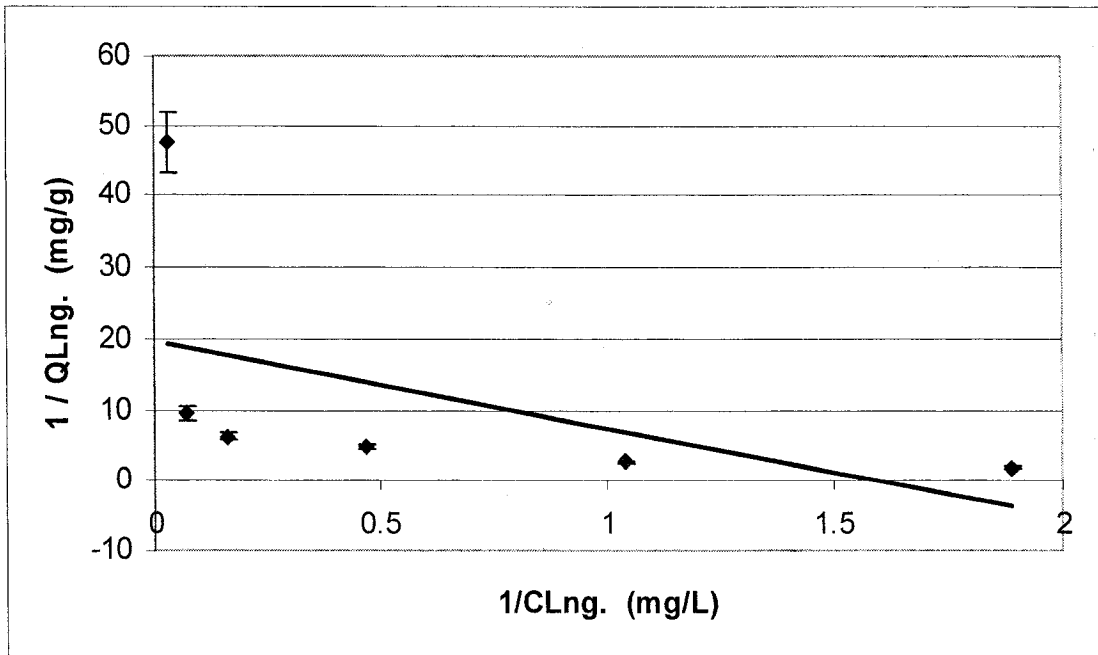


Figure A.3.3a Langmuir isotherms for nickel onto rhamnolipid for the membranes of 10,000 MWCO, temperature =  $25 \pm 1$  °C and transmembrane pressure = 69 kPa.

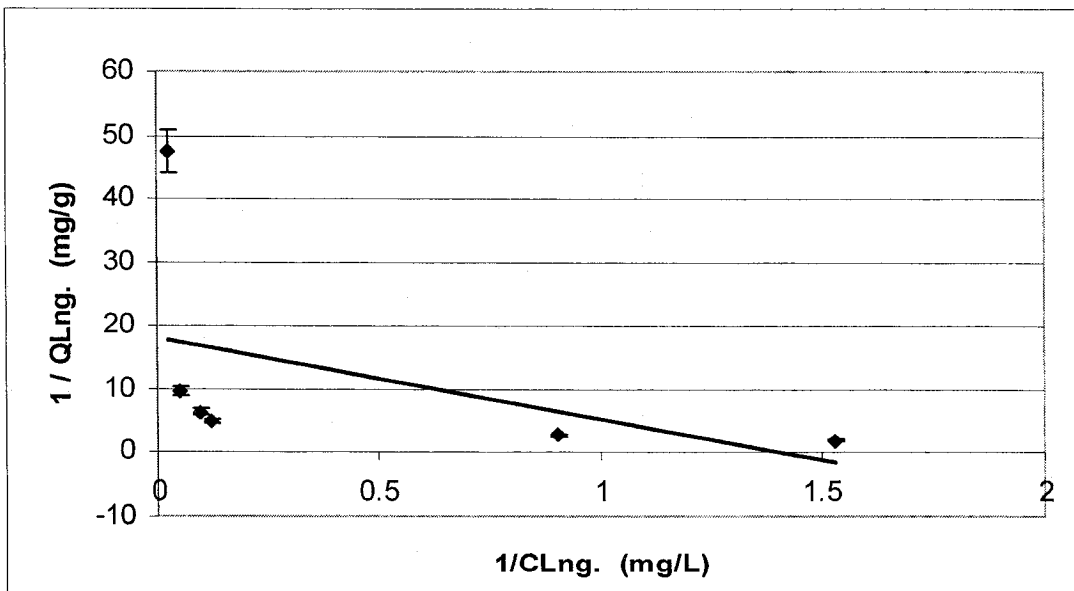


Figure A.3.3b Langmuir isotherms for nickel onto rhamnolipid for the membranes of 30,000 MWCO, temperature =  $25 \pm 1$  °C and transmembrane pressure = 69 kPa.

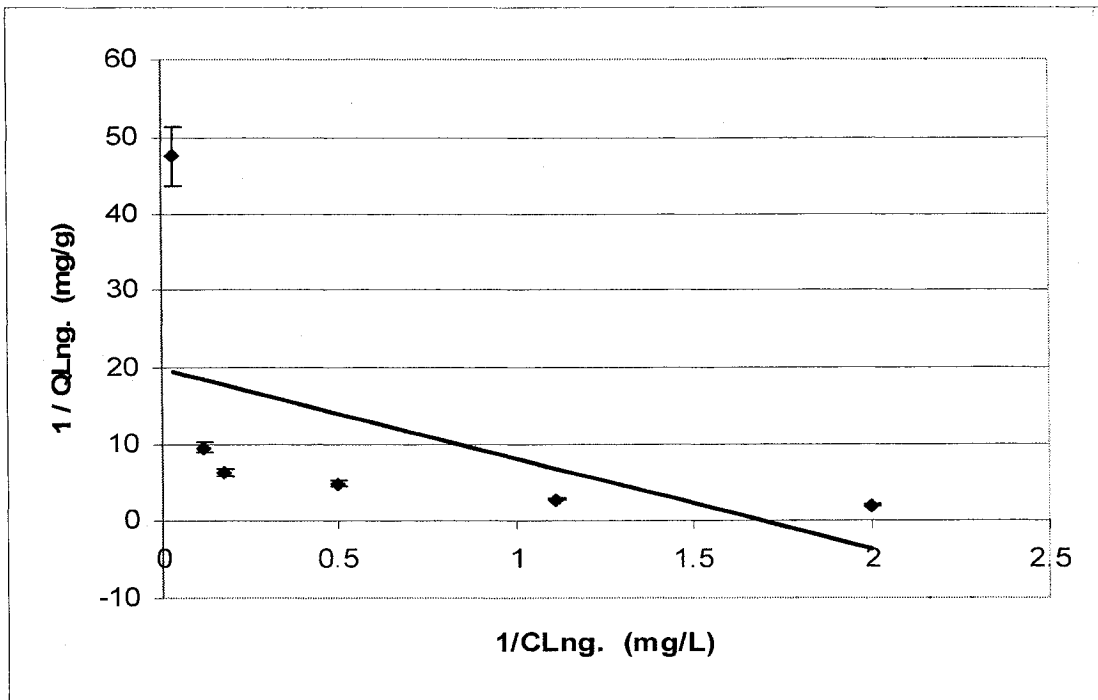


Figure A.3.4a Langmuir isotherms for cadmium onto rhamnolipid for the membranes of 10,000 MWCO, temperature =  $25 \pm 1$  °C and transmembrane pressure = 69 kPa.

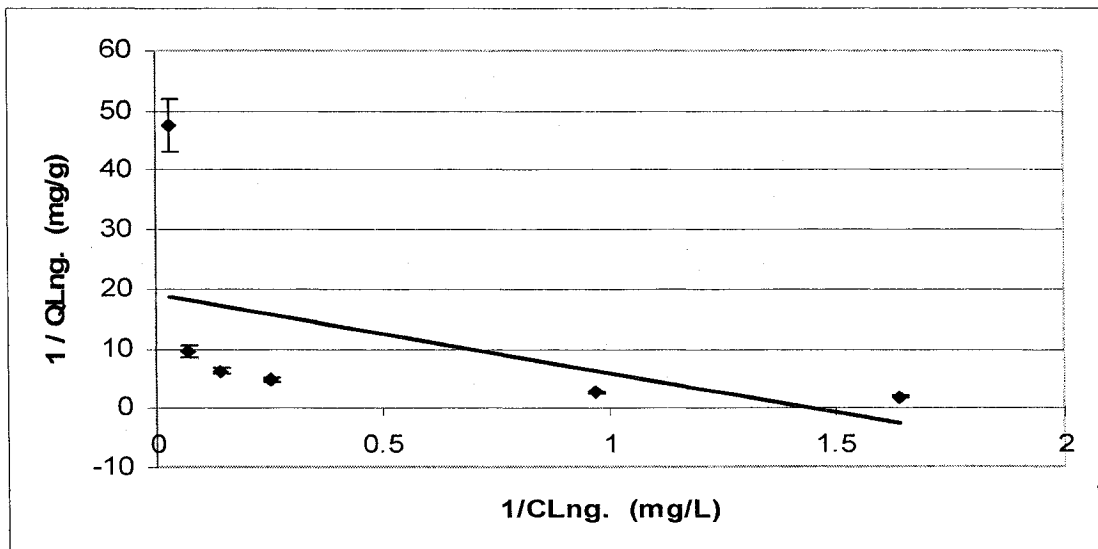
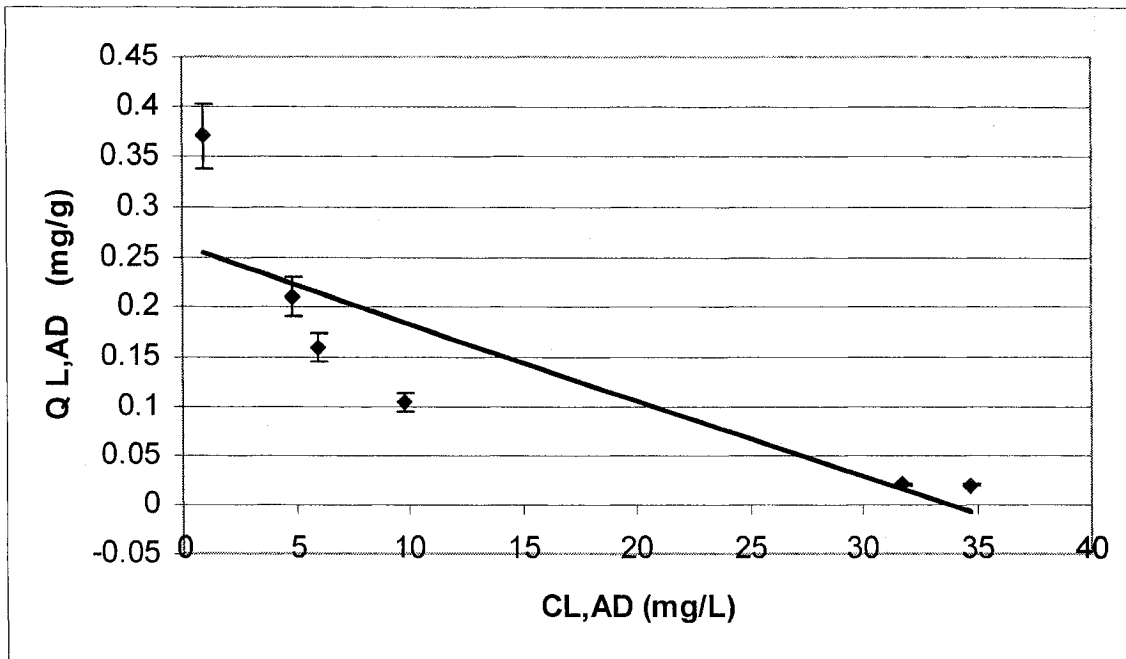
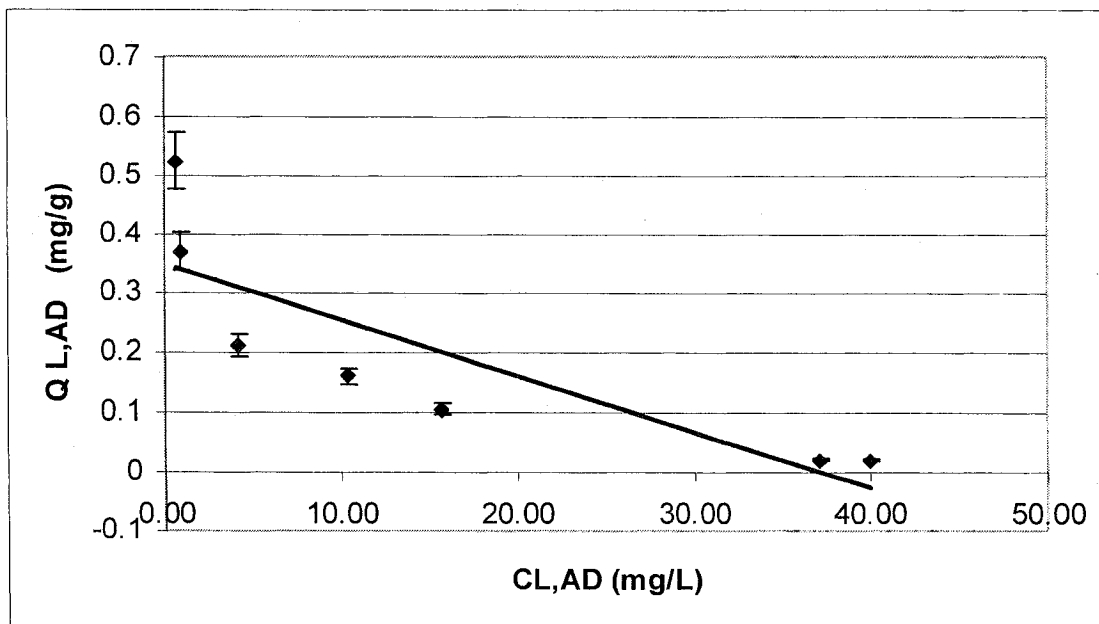


Figure A.3.4b Langmuir isotherms for cadmium onto rhamnolipid for the membranes of 30,000 MWCO, temperature =  $25 \pm 1$  °C and transmembrane pressure = 69 kPa.



**Figure A.3.5a** Linear adsorption model isotherms for zinc onto rhamnolipid for the membranes of 10,000 MWCO, temperature =  $25 \pm 1$  °C and transmembrane pressure = 69 kPa.



**Figure A.3.5b** Linear adsorption model isotherms for zinc onto rhamnolipid for the membranes of 30,000 MWCO, temperature =  $25 \pm 1$  °C and transmembrane pressure = 69 kPa.

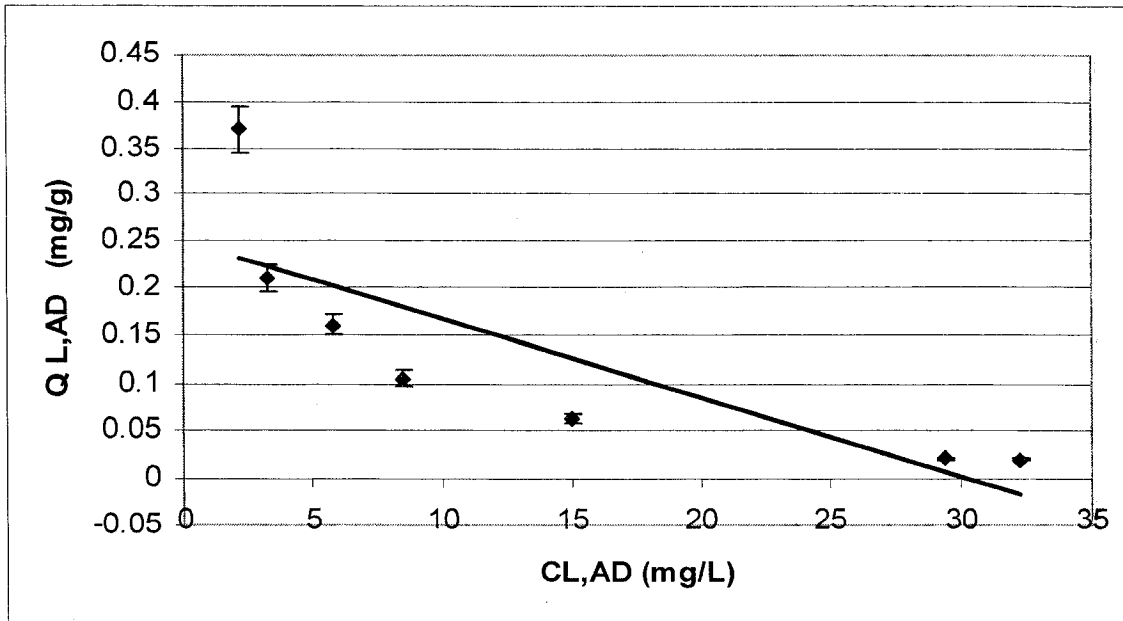


Figure A.3.6a Linear adsorption model isotherms for copper onto rhamnolipid for the membranes of 10,000 MWCO, temperature =  $25 \pm 1$  °C and transmembrane pressure = 69 kPa.

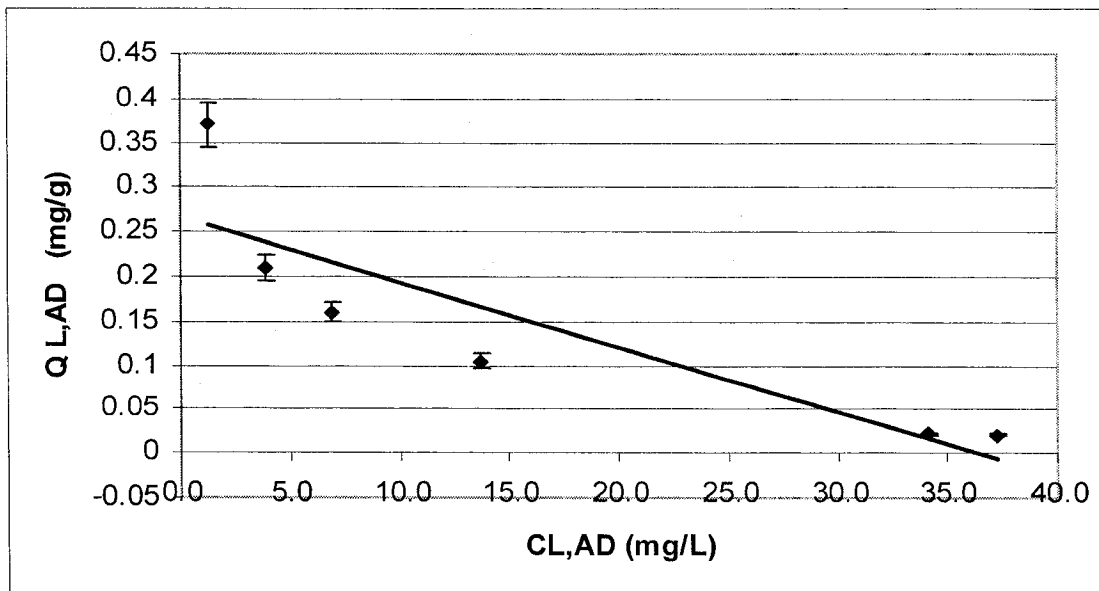


Figure A.3.6b Linear adsorption model isotherms for copper onto rhamnolipid for the membranes of 30,000 MWCO, temperature =  $25 \pm 1$  °C and transmembrane pressure = 69 kPa.

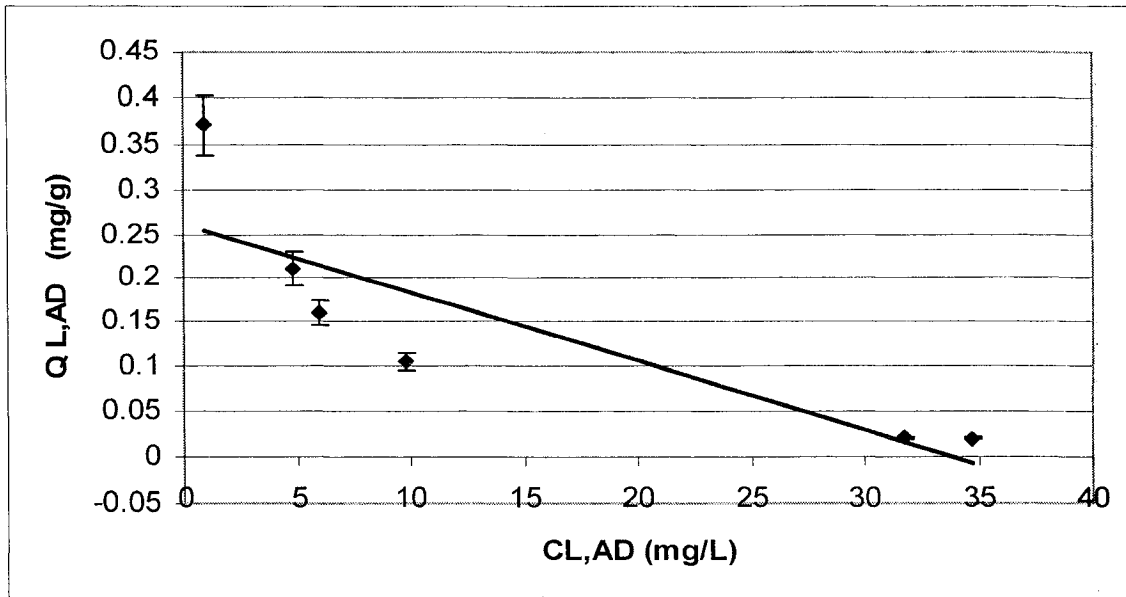


Figure A.3.7a Linear adsorption model isotherms for nickel onto rhamnolipid for the membranes of 10,000 MWCO, temperature =  $25 \pm 1$  °C and transmembrane pressure = 69 kPa.

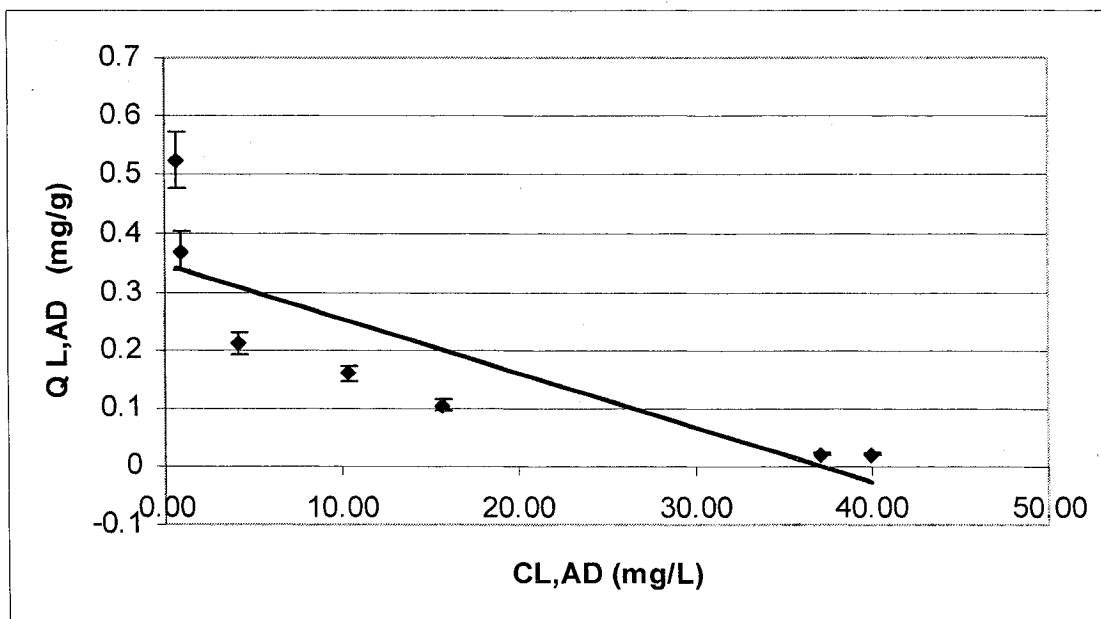


Figure A.3.7b Linear adsorption model isotherms for nickel onto rhamnolipid for the membranes of 30,000 MWCO, temperature =  $25 \pm 1$  °C and transmembrane pressure = 69 kPa.

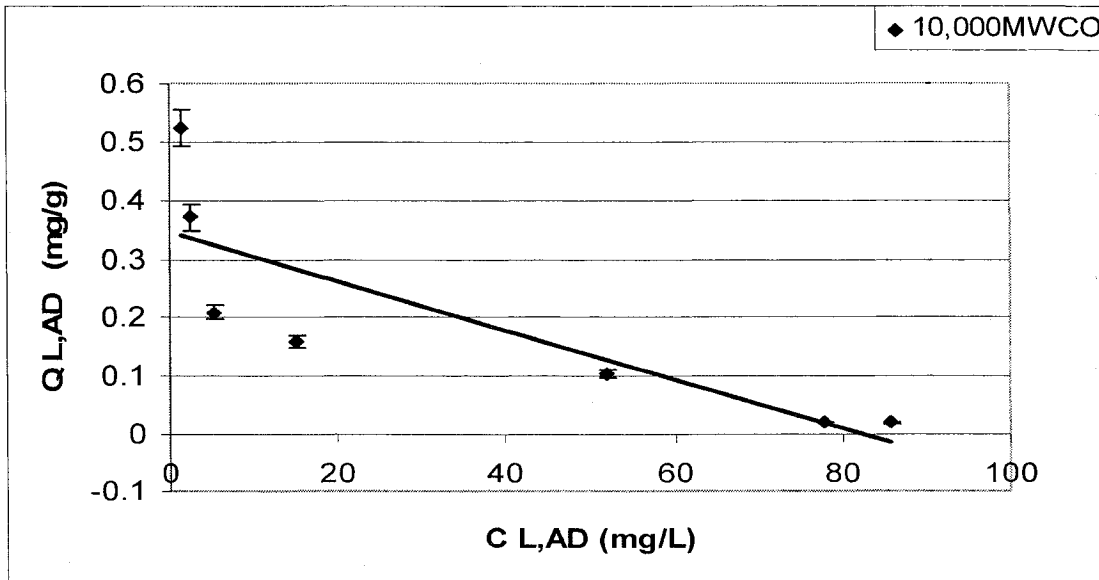


Figure A.3.8a Linear adsorption model isotherms for cadmium onto rhamnolipid for the membranes of 10,000 MWCO, temperature =  $25 \pm 1$  °C and transmembrane pressure = 69 kPa.

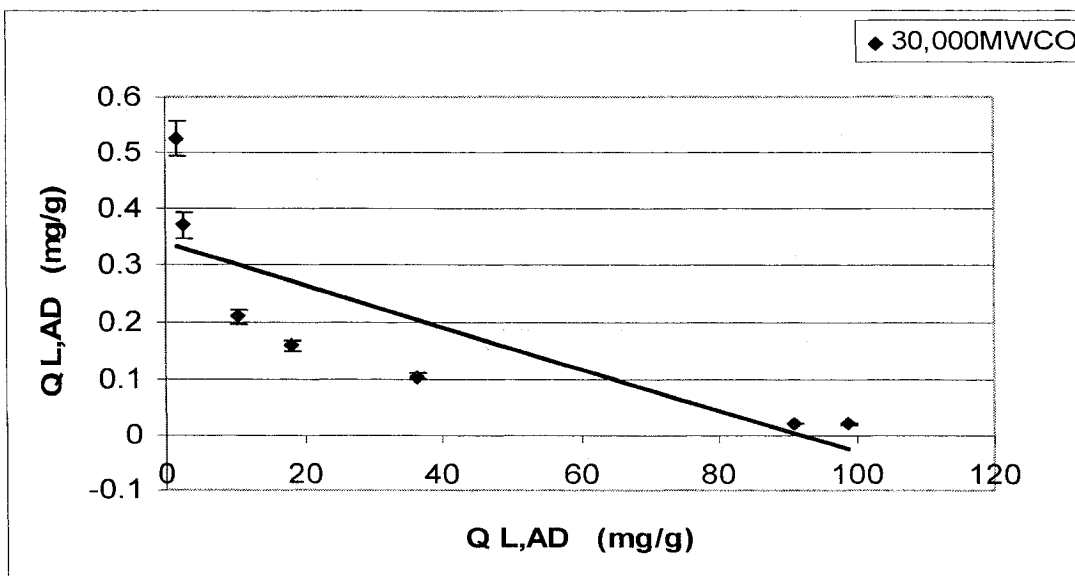
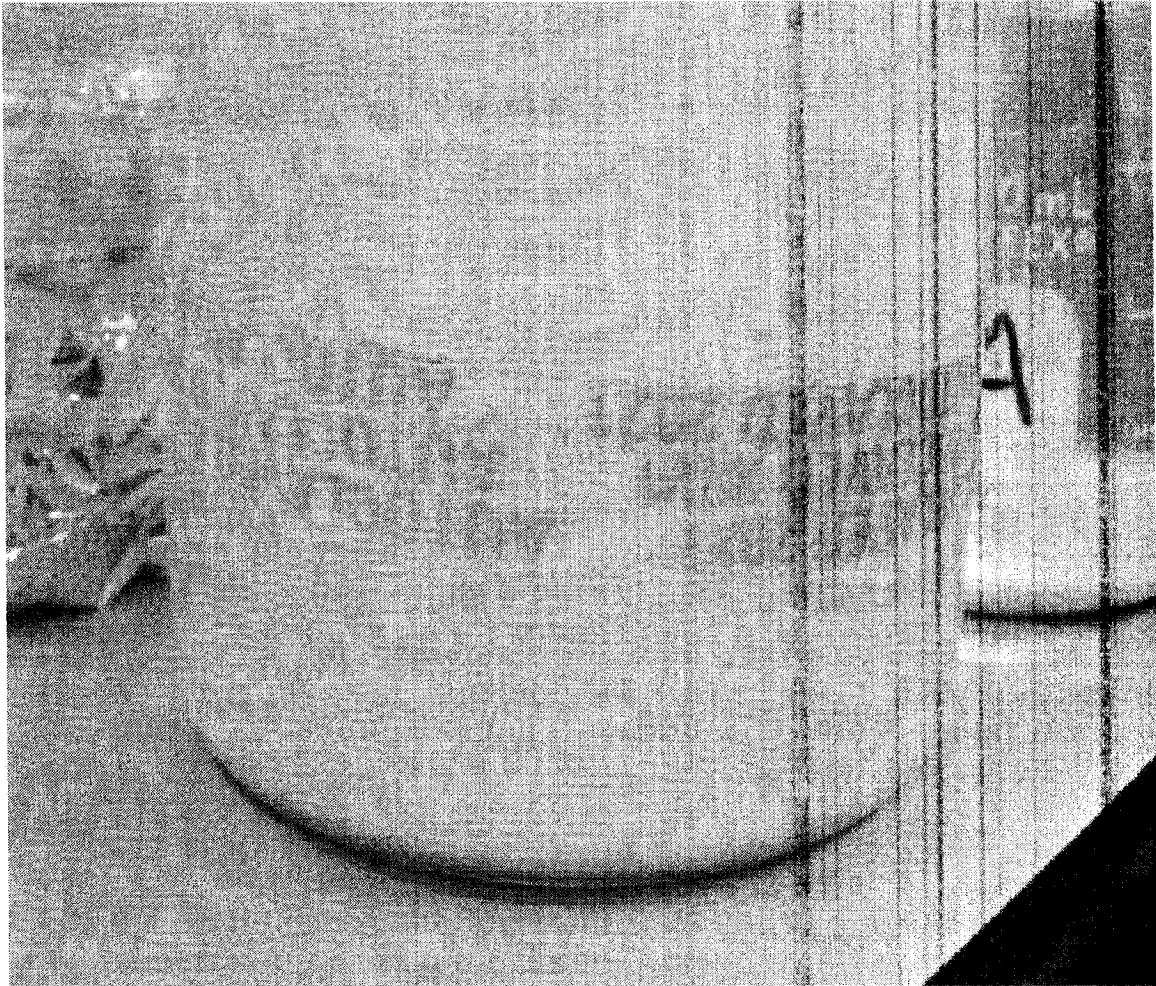


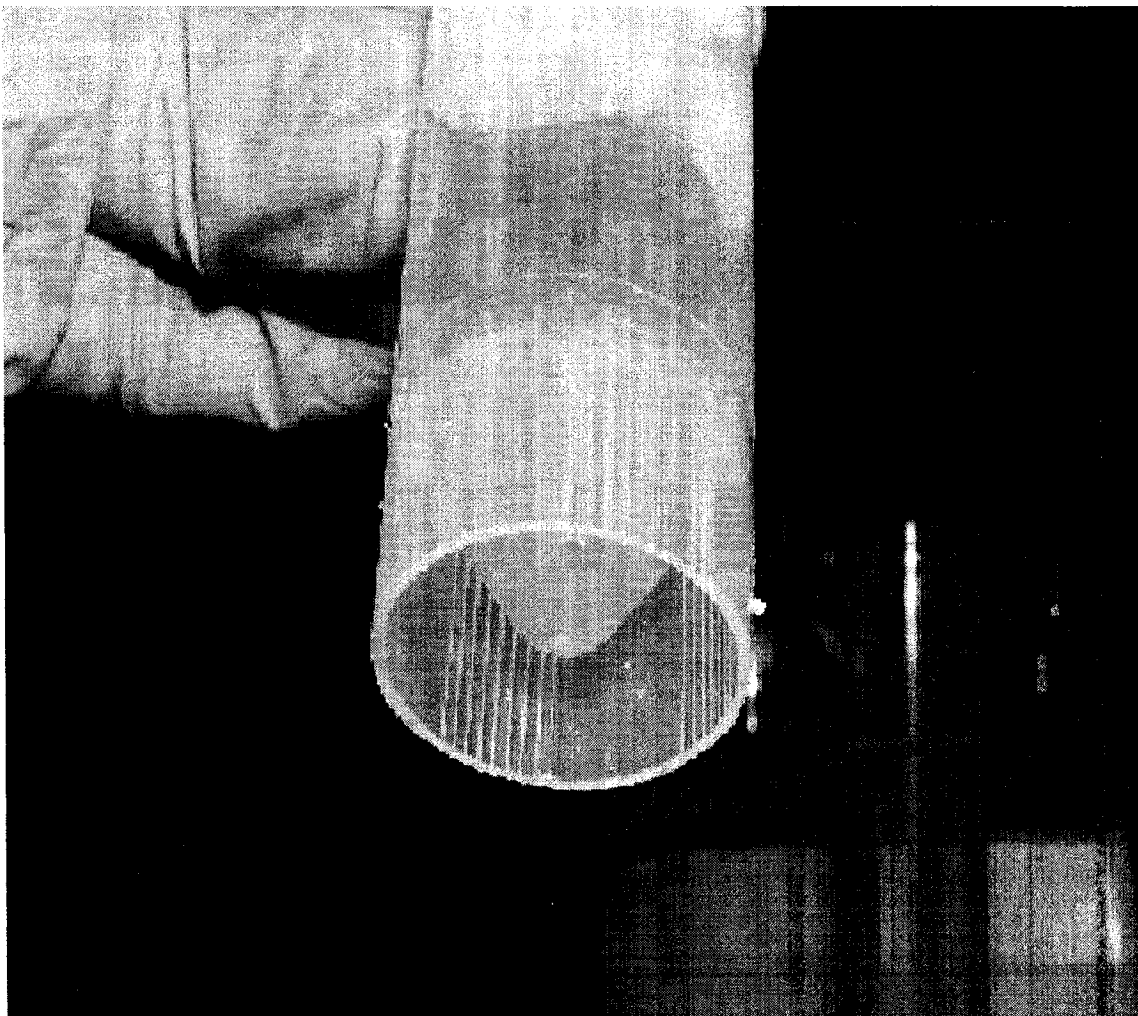
Figure A.3.8b Linear adsorption model isotherms for cadmium onto rhamnolipid for the membranes of 30,000 MWCO, temperature =  $25 \pm 1$  °C and transmembrane pressure = 69 kPa.

**APPENDIX B:**  
**Photographs of rhamnolipid recovery trials**





**Retentate from wastewater treatment by UF.**

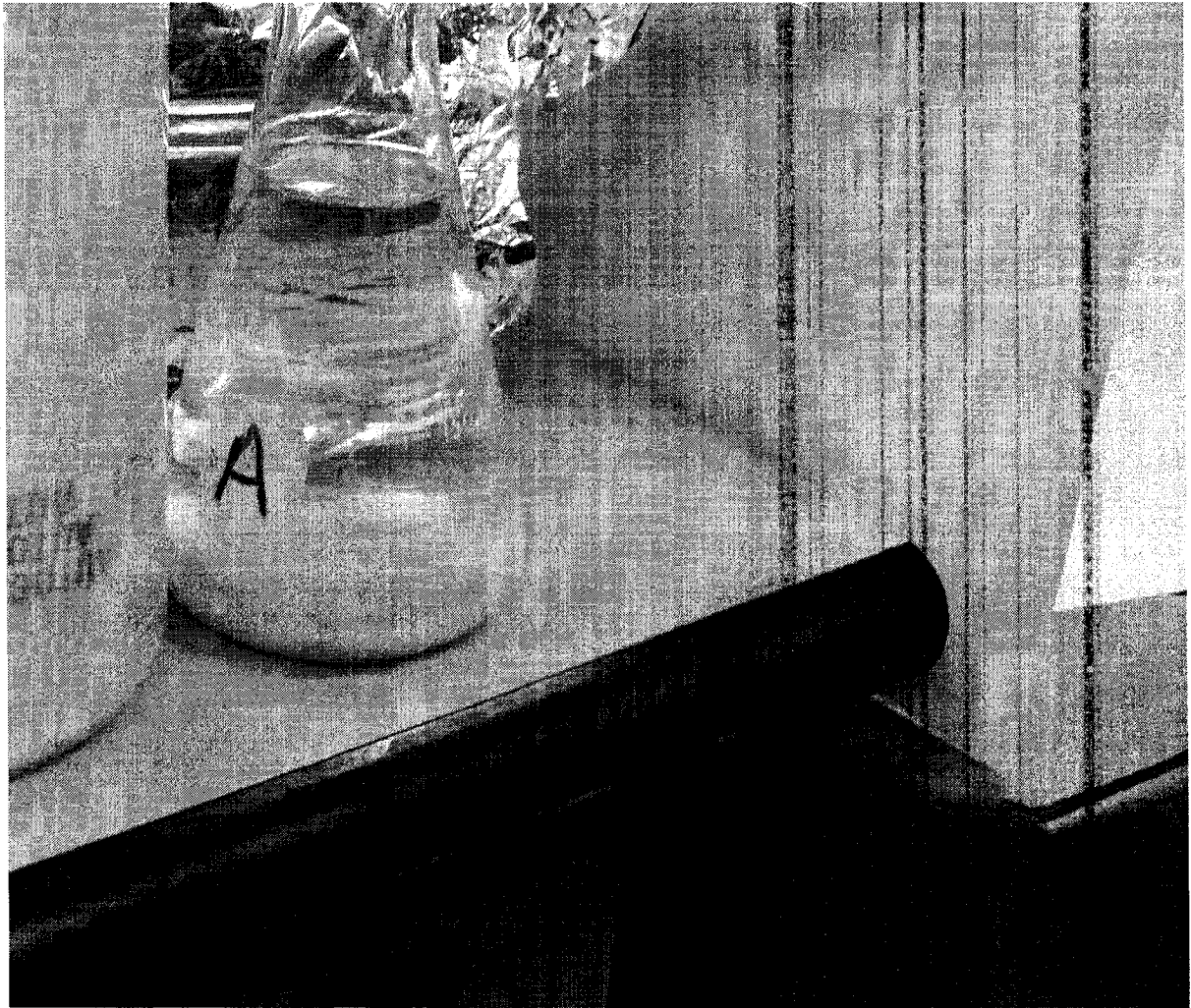


**Precipitated rhamnolipid after centrifuge.**



**Lead and cadmium formed stable white complexes and precipitates with the rhamnolipid (right). Water was used for comparison (left).**

**APPENDIX C:**  
**Photographs of water reuse trials**



**The lab-scale trial to reuse a fraction of the retentate of the rhamnolipid-based UF process as a good water management practice.**

**APPENDIX D:**  
**Simple treatment of the double layer based on  
the Poisson-Boltzmann equation and Gouy-Chapman model**

This work introduces a simple treatment of the electrical double layer that is based on the Poisson-Boltzmann equation and Gouy-Chapman model. The approaches that were taken into consideration were the modified Boltzmann equation for dependence of the ion-wall correlation functions on the diffuse layer potential (Smagala et al., 2006), the planar-symmetry Gouy-Chapman approximation (Tamashiro et al., 2005), the hypernetted chain/mean spherical approximation (HNC/MSA) for description of electrolyte around a spherical macroparticle (Guerrero-Garcia, et al., 2005), and application of the functional analysis method (Wang et al., 2001).

### Applicability of Poisson-Boltzmann equation

If we take into consideration a portion of solution in which the counterions, rhamnolipid monomers and rhamnolipid micelles are distributed in a uniform density  $\rho^*$ , then the force between two charged particles  $\bar{F}_{1 \rightarrow 2}$  can be expressed as:

$$\bar{F}_{1 \rightarrow 2} = q_2 \bar{E}(r) = \frac{q_2 q_1 r}{4\pi \epsilon r^2} \quad (\text{Evans and Wennerstroem, 1999}) \quad (4.3.1)$$

where  $\bar{E}$  is electric field intensity,  $\epsilon$  is equal to  $(\epsilon_o D)$ ,  $\epsilon_o$  is permittivity of free space ( $8.854 \times 10^{-14} \text{ C v}^{-1} \text{ cm}^{-1}$ ),  $D$  is dielectric constant (78.5 for water at 25 °C) and  $r$  is the radius of a rhamnolipid micelle.

Since  $\epsilon = \epsilon_o D$  (Evans and Wennerstroem, 1999)

$$\bar{F}_{1 \rightarrow 2} = q_2 \bar{E}(r) = \frac{q_2 q_1 r}{4\pi \epsilon_0 D r^2} \quad (4.3.2)$$

The units of force F:  $C^2 / ((C^2/N.m^2).m^2) = \text{Newtons}$

Electric field intensity  $\bar{E}$  can be expressed as

$$\bar{E} = -\nabla \psi \quad (\text{Evans and Wennerstroem 1999})$$

where  $\psi$  is the electrical potential (volts). For the case of spherical; symmetry in

which  $\psi$  is a function of r only,;  $\bar{E}(r)$  can be shown as

$$\bar{E}(r) = -\nabla \psi(r)$$

This equation can be rewritten as

$$E = -\frac{d\psi}{dr} \quad (4.3.3)$$

Equation (4.3.3) can then be expressed as:

$$E(r) = \frac{q_1}{4\pi \epsilon_0 D r^2} \quad (4.3.4)$$

For the case when the potential of a charged sphere (e.g. a charged rhamnolipid micelle) is immersed in a dielectric,  $q_1$  is equal to the total charge on the sphere

Q. Considering



$$\psi(r) = \frac{q_1}{4\pi\epsilon_0 D r} \quad (4.3.5)$$

where the units of potential  $\psi$  are  $C / ((C^2/N.m^2) . m^2) = N/C = J/C =$  (energy/charge). If a portion of solution in which the charge is distributed with a uniform density  $\rho$  ( $r$ ) is considered,  $q_1$  can be written as:

$$q_1 = \frac{4}{3} \pi r^3 \rho \quad (4.3.6)$$

where  $r$  is the radius of a micelle. Substituting (4.3.6) in (4.3.4), this gives:

$$E = \frac{r \rho}{3\epsilon} \quad (4.3.7)$$

By multiplying both sides of equation (4.3.7) by  $r^2$  and differentiating with respect to  $r$ , we get:

$$\frac{d}{dr}(r^2 E) = \frac{r^2 \rho}{\epsilon} \quad (4.3.8)$$

From there, we substitute equation (4.3.3) into equation (4.3.8), which gives:

$$\frac{1}{r^2} \frac{\partial}{\partial r} \left( r^2 \frac{\partial \psi}{\partial r} \right) = \frac{-\rho}{\epsilon_0 D} \quad (4.3.9)$$

In spherical coordinates, the most general form of equation (4.3.9) is

$$\frac{1}{r^2} \frac{\partial}{\partial r} \left( r^2 \frac{\partial \psi}{\partial r} \right) + \frac{1}{r^2 \sin \theta} \frac{\partial}{\partial \theta} \left( \sin \theta \frac{\partial \psi}{\partial \theta} \right) + \frac{1}{r^2 \sin^2 \theta} \left( \frac{\partial^2 \psi}{\partial \phi^2} \right) = \frac{-\rho}{\epsilon_0 D} \quad (4.3.10)$$

In Cartesian coordinates the Poisson equation can be expressed as :

$$\nabla \cdot \nabla \psi = \nabla^2 \psi = \frac{\partial^2 \psi}{\partial x^2} + \frac{\partial^2 \psi}{\partial y^2} + \frac{\partial^2 \psi}{\partial z^2} = \frac{-\rho}{\epsilon_0 D} \quad (4.3.11)$$

where  $\nabla \cdot \nabla = \nabla^2$  is known to be Laplace operator, that means div grad. Also, it gives the impression that Cartesian coordinates are utilized.

At equilibrium, the electrochemical potential of the ions must be constant everywhere, thusly:

$$\bar{\mu}_i = \mu_i + z_i e \psi = \text{constant} \quad (4.3.12)$$

where  $\bar{\mu}_i$  is the chemical potential for species  $i$ ,  $\psi$  is the electrical potential (volts) and  $z_i$  is the valence of ions  $i$ , then:

$$\nabla \bar{\mu}_i = 0 \quad (4.3.13)$$

Since the electrical and the diffusional forces on the ions must to balance out, then:

$$\nabla\mu_i = -z_i e \nabla\psi \quad (4.3.14)$$

For a planar double layer, the electrostatic potential and the chemical potential are constant in planes parallel to the wall so that equation (4.3.14) can be expressed as:

$$\frac{d\mu_i}{dx} = -z_i e \frac{d\psi}{dx} \quad (4.3.15)$$

By the use of the definition of the chemical potential for type i ion (joules per ion) in the form:

$$\mu_i = \mu_i^0 + kT \ln(n_i \gamma_i) \quad (4.3.16)$$

where  $\mu_i^0$  is the standard chemical potential,  $n_i$  is the number of ions per unit volume,  $k$  is the Boltzmann constant ( $1.38 \times 10^{-23}$  J/K),  $T$  is the absolute temperature (K)

and  $\gamma_i$  is an activity coefficient. By taking the derivative of equation (4.3.16) and combining it with equation (4.3.15), the following expression can be obtained:

$$\frac{d\mu_i}{dx} = kT \frac{d \ln(n_i \gamma_i)}{dx} = kT \frac{d \ln(n_i \gamma_i) d(n_i \gamma_i)}{d(n_i \gamma_i) dx} = kT \frac{1}{(n_i \gamma_i)} \frac{d(n_i \gamma_i)}{dx} = -\frac{z_i e}{kT} \frac{d\psi}{dx} \quad (4.3.17)$$

Integrating from the bulk aqueous solution (where  $n_i\gamma_i = n_i^0\gamma_i^0$  and  $\psi = 0$ ) to any point in the diffuse layer (where  $n_i\gamma_i =$  ion number of species  $i$  per volume and  $\psi$  is the electrical potential ) gives:

$$\int_{n_i^0\gamma_i^0}^{n_i\gamma_i} \frac{d(n_i\gamma_i)}{(n_i\gamma_i)} = -\frac{z_i e}{kT} \int_0^\psi d\psi \quad (4.3.18)$$

$$n_i\gamma_i = n_i^0\gamma_i^0 * \exp\left(\frac{-z_i e \psi}{kT}\right) \quad (4.3.19)$$

In the case of utilizing very low concentrations (i.e. very low ionic strength), activity coefficients are close to 1.0. Thus, the Boltzmann equation can be simplified to the form:

$$n_i = n_i^0 * \exp\left(\frac{-z_i e \psi}{kT}\right) \quad (4.3.20)$$

where  $n_i$  is the number of ions  $i$  per unit volume diffuse layer solution ( $\text{cm}^{-3}$ ),  $n_i^0$  is number of ions,  $i$ , per unit volume bulk phase solution ( $1/\text{cm}^3$ ),  $z_i$  is the valence of ions  $i$ ,  $e$  is the charge of an electron ( $1.602 \times 10^{-19}$  C/electron),  $\psi$  is the electrical potential (volts),  $k$  is the Boltzmann constant ( $1.38 \times 10^{-23}$  J/K) and  $T$  is the absolute temperature (K).

Utilizing equations results in the Poisson-Boltzmann equation that can be simplified to the form:

$$\nabla^2\psi = \left(\frac{-1}{\epsilon_0 D}\right) * \sum_i n_i^0 z_i e * \exp\left(\frac{-z_i e\psi}{kT}\right) \quad (4.3.21)$$

The Gouy-Chapman model assumes that the surface is planar and of uniform potential along the plane (i.e., the potential gradients with respect to y and z are both zero), and results in the following:

$$\nabla^2\psi = \frac{d^2\psi}{dx^2} = \left(\frac{-1}{\epsilon_0 D}\right) \sum_i n_i^0 z_i e * \exp\left(\frac{-z_i e\psi}{kT}\right) \quad (4.3.22)$$

Equation (4.3.22) is a nonlinear, second-order ordinary differential equation. This equation has no known analytical solution. Consequently, some simplifications and additional assumptions are needed. In order to solve this equation, boundary conditions are required: (1) at  $x = \infty$  (at infinite distance from the surface of the rhamnolipid micelle in the bulk solution where the charge surface has no electrical effect),  $\psi = d\psi / dx = 0$ ; and (2) at  $x=0$  (at the plane of the surface),  $\psi_0$  can be calculated from:

$$\frac{\sigma^2}{\varepsilon^2} = \frac{2KT}{\varepsilon} \sum_i n_i^0 \left[ \exp\left(\frac{-z_i e \psi_o}{kT}\right) - 1 \right] \quad (4.3.23)$$

where  $\sigma$  (C/m<sup>2</sup>) is the surface charge density. By utilizing the molar concentration,  $C_i$ , for each ion,  $i$ , instead of ion numbers: ( $C_i$  (moles/L) x  $N$  (number of ions / mole) x 1000 (1/m<sup>3</sup>) =  $n_i^0$  (number of ions / m<sup>3</sup>), equation (4.3.23) can be expressed as:

$$\frac{\sigma^2}{2\varepsilon kT} = \frac{2KT}{\varepsilon} \sum_i C_i N * 1000 \left[ \exp\left(\frac{-z_i e \psi_o}{kT}\right) - 1 \right] \quad (4.3.24)$$

where  $N$  is Avogadro's number ( $6.022 \times 10^{23}$  / mole), and  $z_i$  is the valence of ions  $i$ . If the surface charge density is known, equation (4.3.24) can be used to calculate the surface potential ( $\psi_o$ ).

**APPENDIX E:**  
**Data quality and the lack of fit analysis**

## Data quality and the lack of fit analysis

### Box-Cox method

In order to investigate the need for mathematical transformation, the Box-Cox method was used to determine the transformation parameter lambda and to evaluate the data sets. The Box Cox method is a method to help in determination the most appropriate power transformation to apply to response data. Most data transformations may be described by the power function,

$$\sigma = \text{fn}(\mu^\alpha)$$

where sigma ( $\sigma$ ) is the standard deviation, mu ( $\mu$ ) is the mean and alpha ( $\alpha$ ) is the power. Generally, the Box Cox plot illustrates the minimum lambda values, as well as lambdas at a confidence range.

In this work, the 95 % confidence interval level was applied. Lambda ( $\lambda$ ) is expressed as  $(1-\alpha)$ . Generally, if the standard deviation associated with an observation is proportional to the mean raised to the  $\alpha$  power, then transforming the observation by the  $\lambda$  power gives a scale satisfying the equal variance requirement of the statistical model. Plotting the logarithm of residual sums of squares (residual SS) against lambda to obtain the Box-Cox plot was performed (Madamba and Bekki, 2001; Mudahar, et. al., 1990; Box and Behnken 1960). Figures E.1, to E.7 show the outputs when the Box-Cox method was applied to the data of the operating parameters, industrial wastewater treatment data, ground water treatment data, the data of metal competition systems, waste

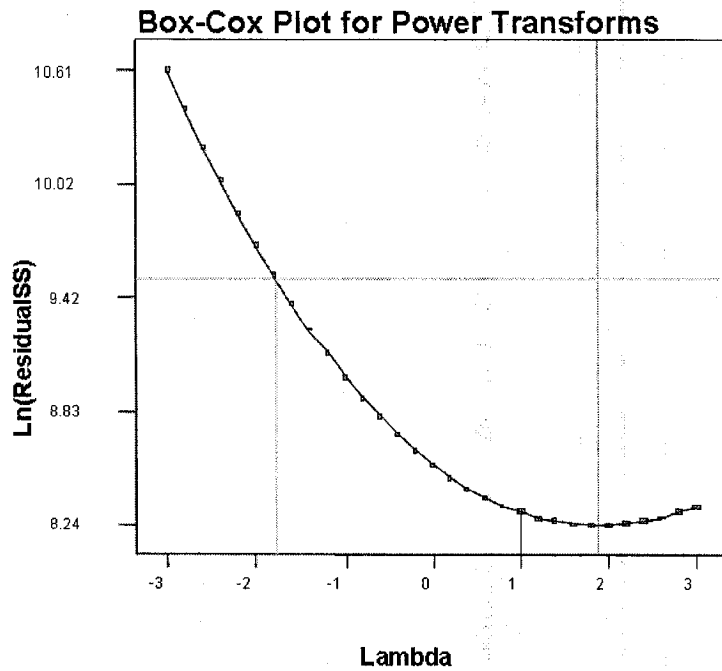


stabilization data, rhamnolipid regeneration data and water recycle data, respectively. Each graph includes the current value of lambda and the best value of lambda.

Flow Rate(l/m<sup>2</sup>/hr)

Lambda  
Current = 1  
Best = 1.88442  
Low C.I. = -1.77158  
High C.I. = 5.95004

Recommend transform:  
None  
(Lambda = 1)



**Figure E.1** Box-Cox method applied to the data of the operating parameters.

( Max. Rej. Cd and Pb )

Lambda  
Current = 1  
Best = -3  
Low C.I. =  
High C.I. =

Recommend transform:  
None  
(Lambda = 1)

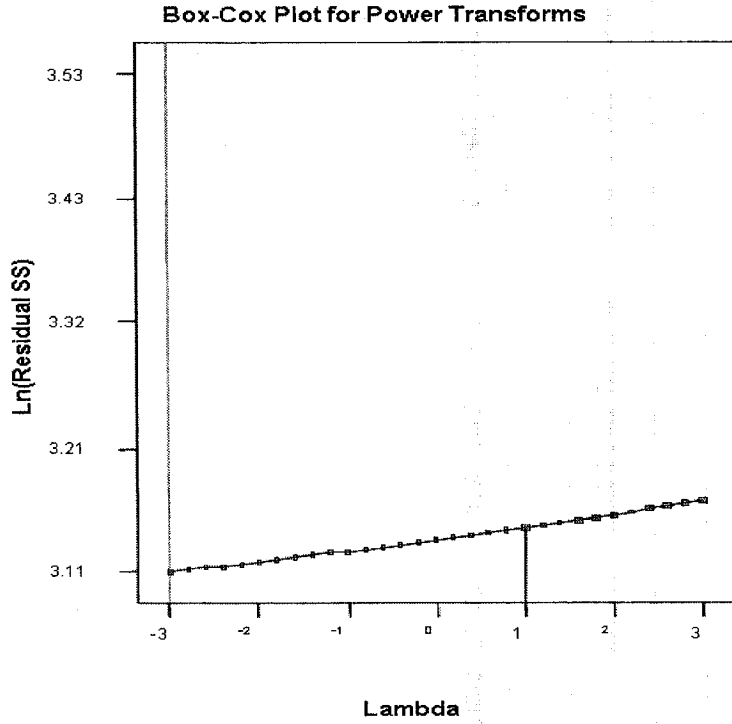


Figure E.2 Box-Cox method applied to the industrial wastewater treatment data

Max. Rejectn ( Cd and Pb )

Lambda  
Current = 1  
Best = 3  
Low C.I. =  
High C.I. =

Recommend transform:  
None  
(Lambda = 1)

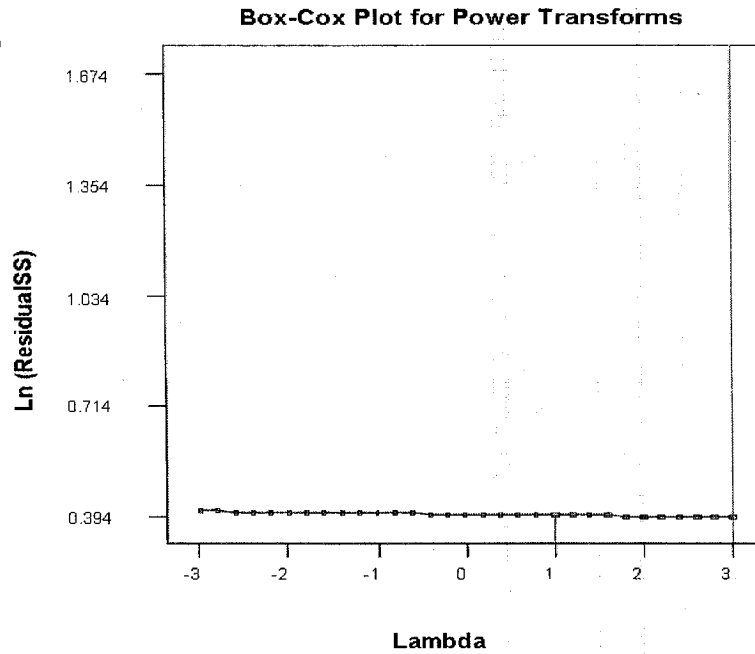


Figure E.3 Box-Cox method applied to the data of the ground water treatment data.

Max. adsorptn. (Binding Cap.)

Lambda  
Current = 1  
Best = 0.644458  
Low C.I. = -1.12137  
High C.I. = 2.5722

Recommend transform:  
None  
(Lambda = 1)

Box-Cox Plot for Power Transforms

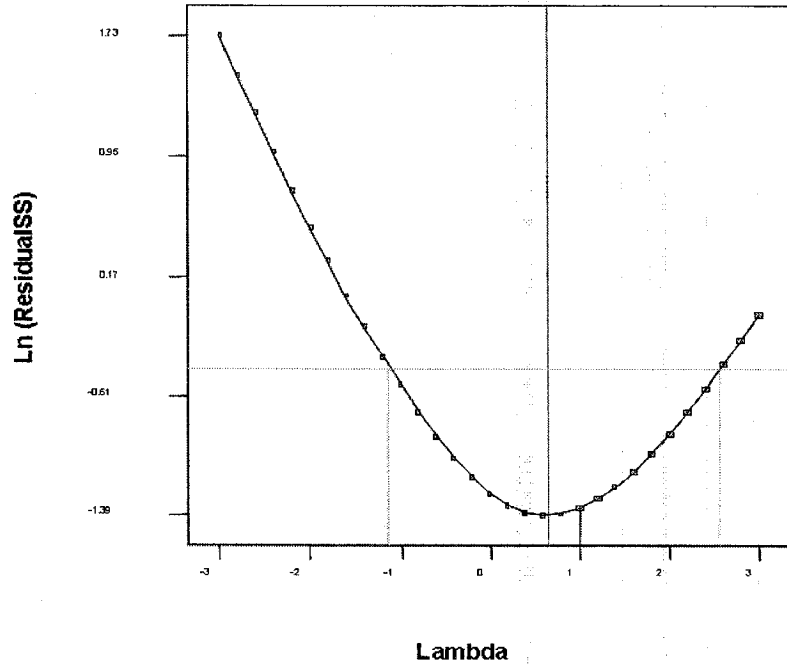


Figure E.4 Box-Cox method applied to the data of metal competition systems.

Max Binding (Min Leaching)

Lambda  
Current = 1  
Best = 3  
Low C.I. =  
High C.I. =

Recommend transform:  
None  
(Lambda = 1)

Box-Cox Plot for Power Transforms

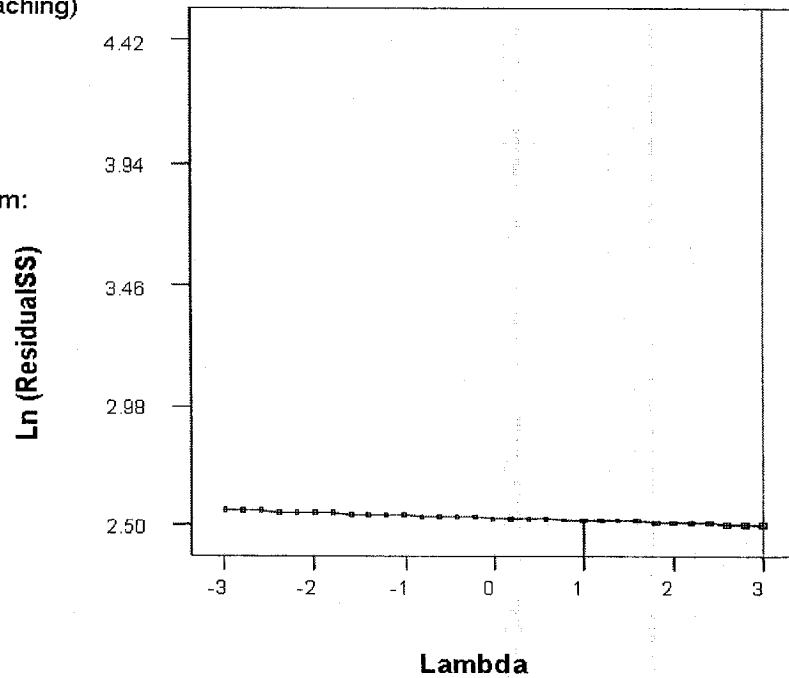


Figure E.5 Box-Cox method applied to the waste stabilization data.

Max. Pptn. for Regtn.

Lambda  
Current = 1  
Best = 3  
Low C.I. =  
High C.I. =

Recommend transform:  
None  
(Lambda = 1)

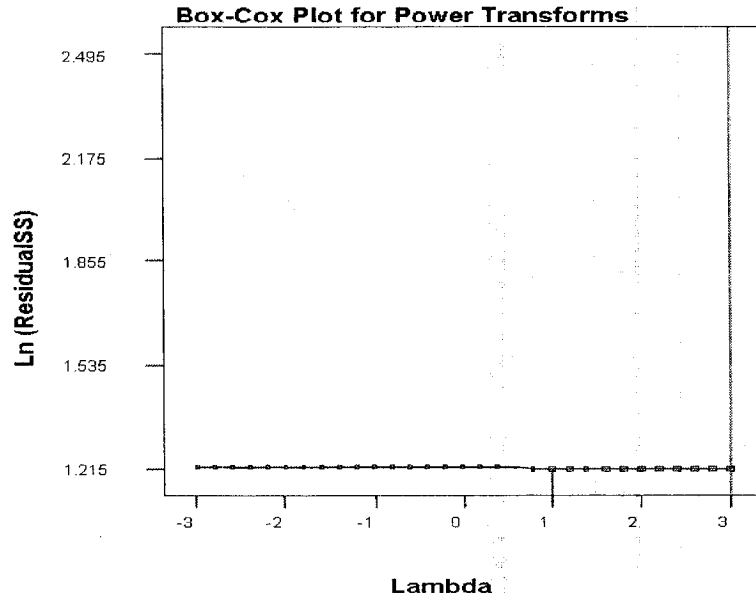


Figure E.6 Box-Cox method applied to the rhamnolipid regeneration data.

Max. Water Reuse (%)

Lambda  
Current = 1  
Best = 1.47729  
Low C.I. = -0.801976  
High C.I. = 4.0922

Recommend transform:  
None  
(Lambda = 1)

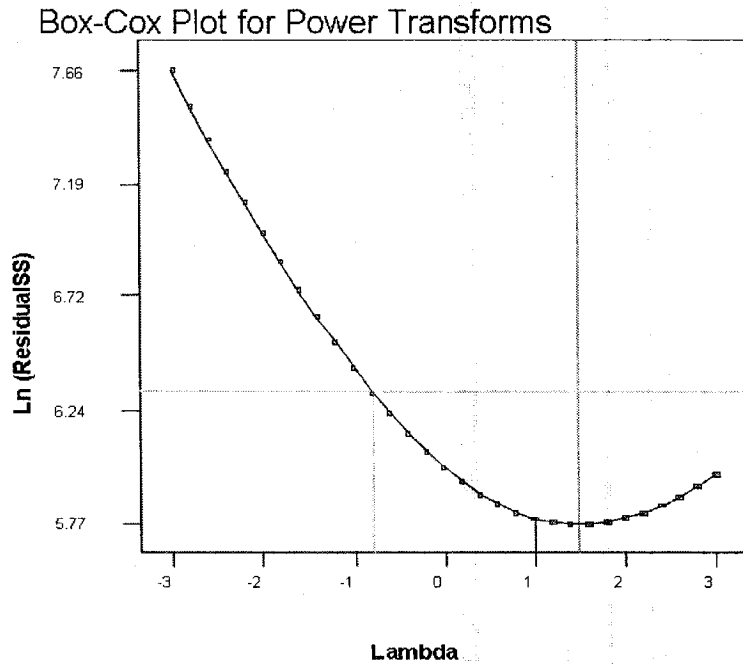


Figure E.7 Box-Cox method applied to the water reuse data.

In Figures E.2, E.3, E.5 and E.6, there is still an indication of a not good response of the logarithm of residual sums of squares (residual SS) and Lambda. However, the confidence intervals include all the current values of lambda. Therefore, this gives an indication that response values are acceptable within 95 % confidence interval level. It implies also that the current values of lambda do not support the need for the mathematical transformation.

### **Investigation of the normality assumption**

A check of the normality assumption for the data may be performed by establishing a normal probability plot of the residuals. Figures E.8 to E.13 show the outputs when the normal probability plot of the residuals was applied to the data of the operating parameters, ground water treatment data, the data of metal competition systems, waste stabilization data, rhamnolipid regeneration data and water recycle data, respectively. In Figures E.8 and E.11, there is still an indication of variability of original data. However, it can be seen that most of the residual plots were approximately along straight lines. Consequently, the Figures E.8 to E.13 indicate no apparent problem with normality. Thus the normality assumption is satisfied for the data sets.

Flow Rate(l/m<sup>2</sup>/hr)

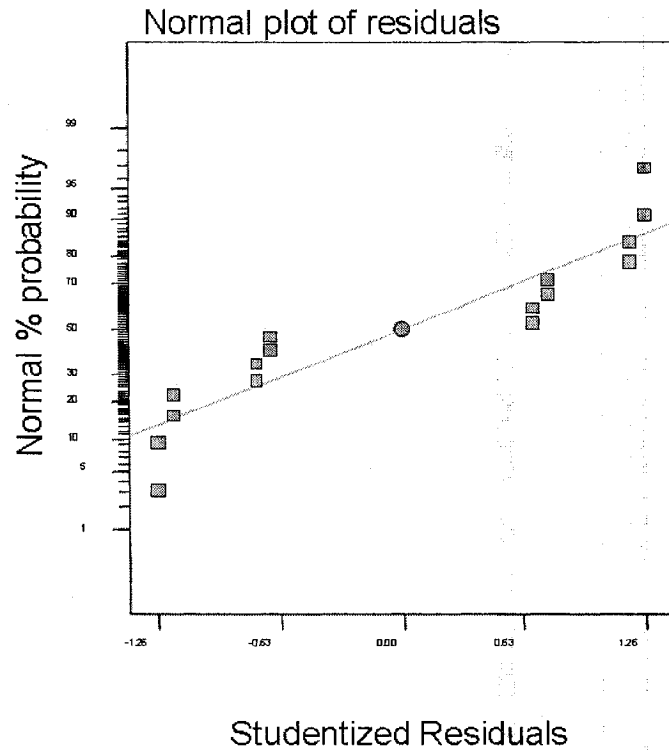


Figure E.8. Normal probability plot of residuals for the operating parameters.

Max. Rejectn ( Cd and Pb )

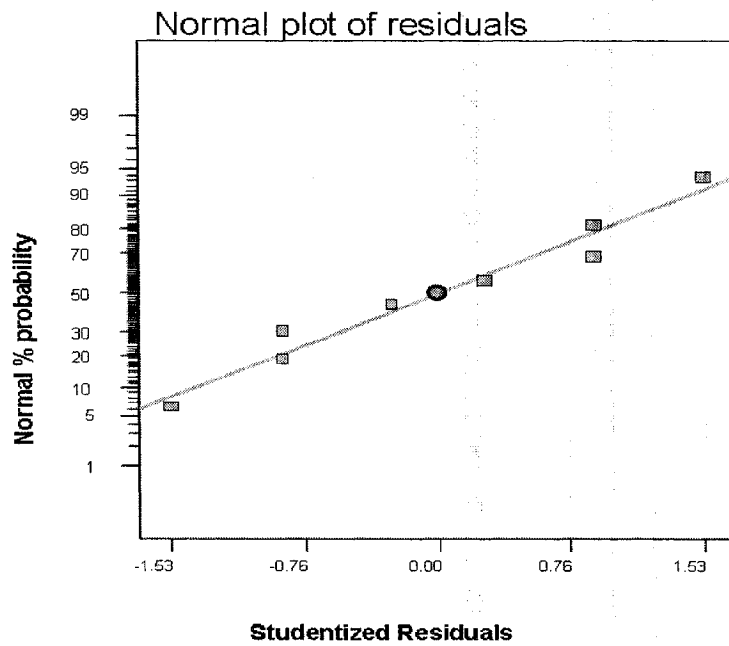


Figure E.9 Normal probability plot of residuals for the ground water treatment data.

Max.adsorptn. (Binding Cap.)

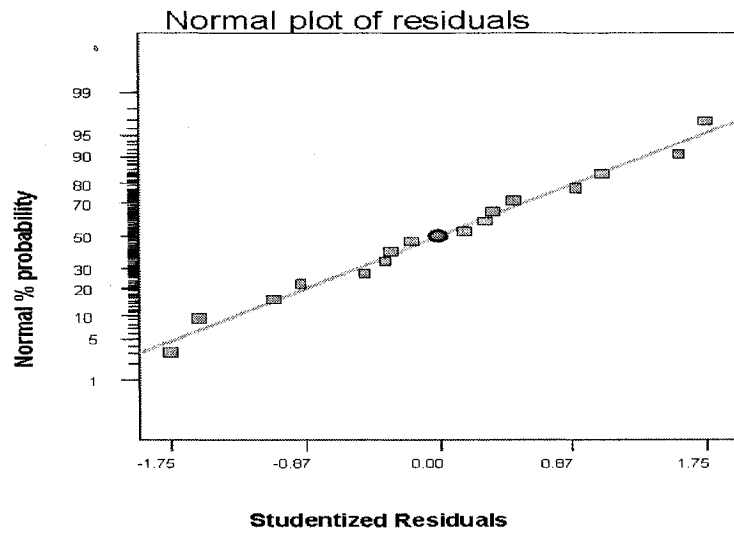


Figure E.10 Normal probability plot of residuals for the data of metal competition systems.

Max Binding (Min Leaching)

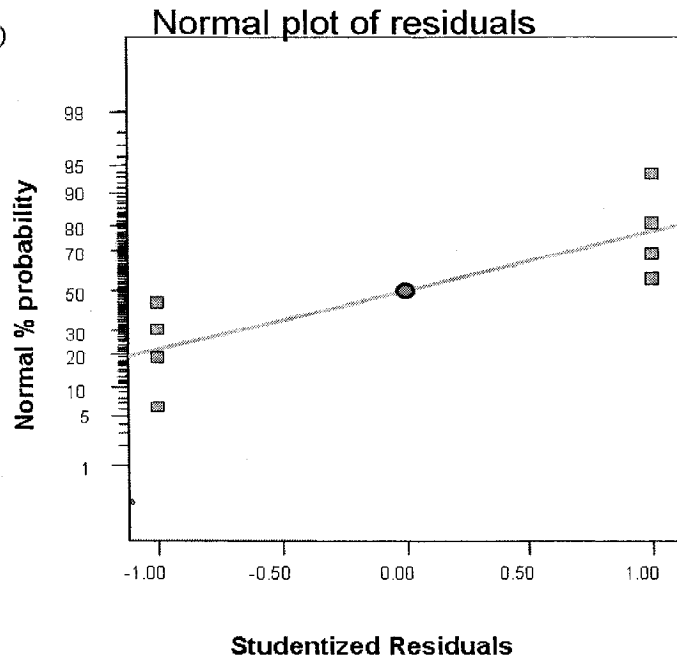


Figure E.11 Normal probability plot of residuals for the waste stabilization data.

Max. Pptn. for Regtn.

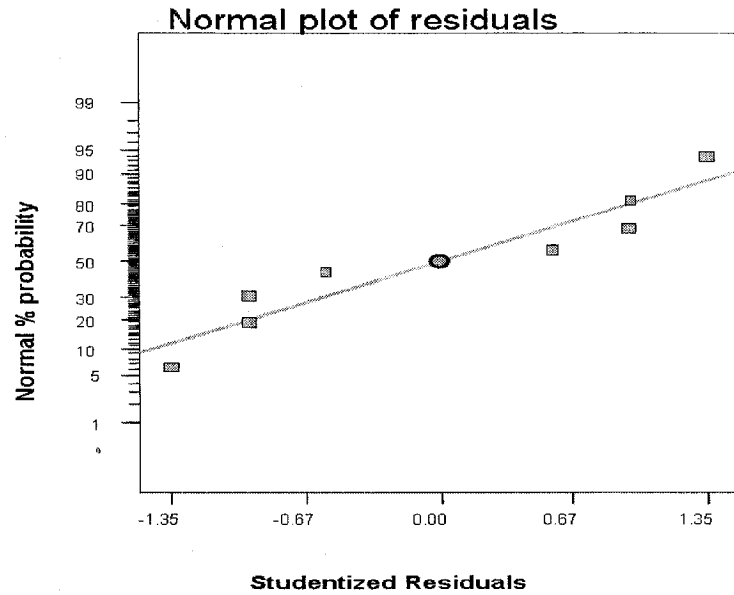


Figure E.12 Normal probability plot of residuals for the rhamnolipid regeneration data.

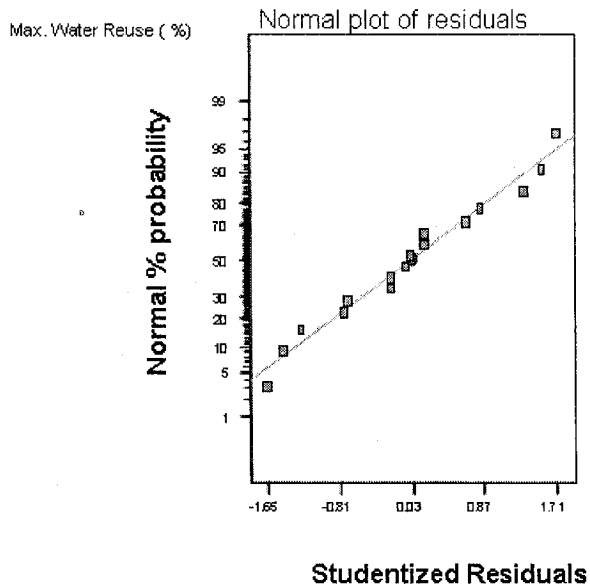
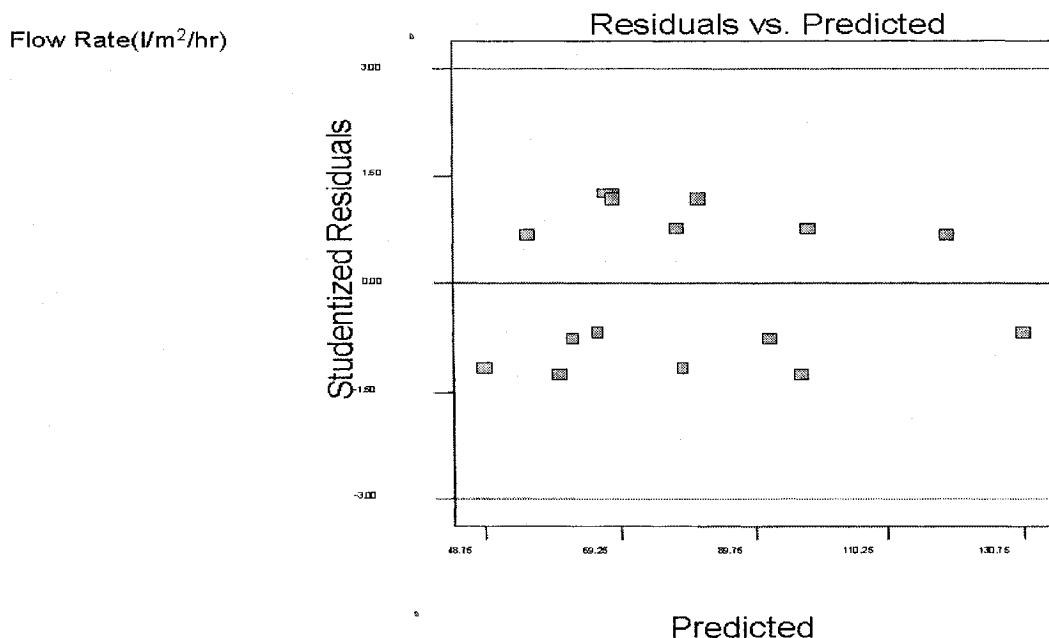


Figure E.13 Normal probability plot of residuals for the water reuse data.



## Investigation of the outliers

Figures E.14, to E.20 show the plots of studentized residuals versus the predicted response for data from the operating parameters, industrial wastewater treatment data, ground water treatment data, the data of metal competition systems, waste sbstabilization data, rhamnolipid regeneration data and water recycle data, respectively. In figures E.15, E.17 and E.20, there is still an indication of inequality of residual distributions on the display.



**Figure E.14 Studentized residuals as a function of predicted data of the operating parameters.**

(Max. Rej. Cd and Pb)

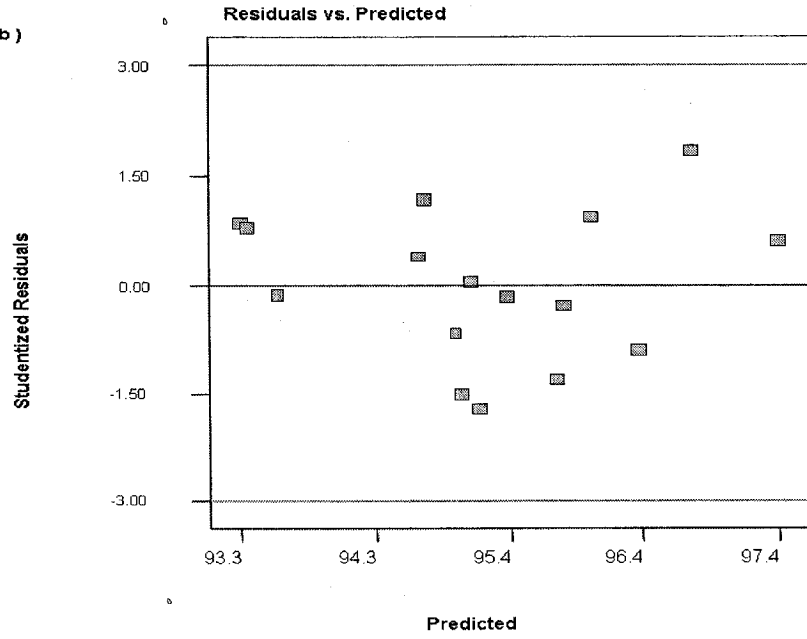


Figure E.15 Studentized residuals as a function of predicted industrial wastewater treatment data.

Max. Rejectn (Cd and Pb)

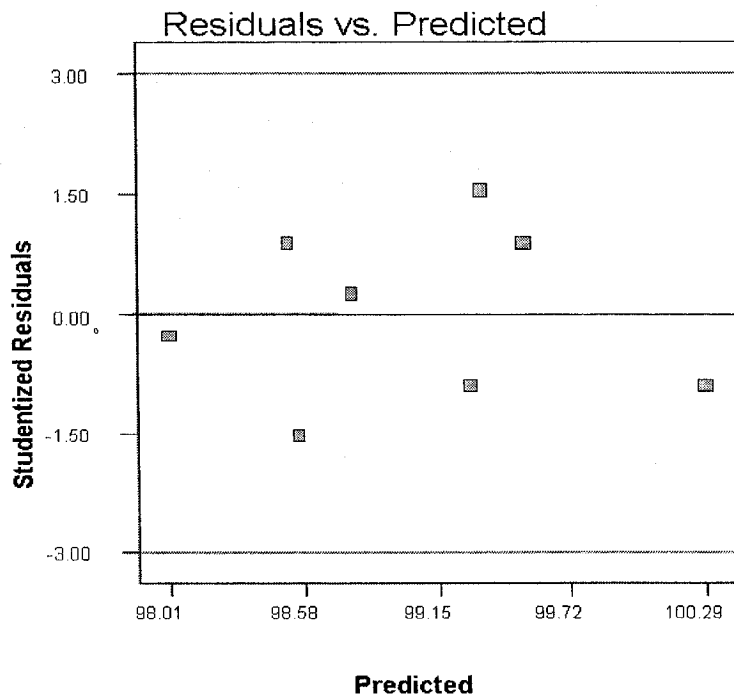


Figure E.16 Studentized residuals as a function of predicted ground water treatment data.

Max. adsorptn. (Binding Cap.)

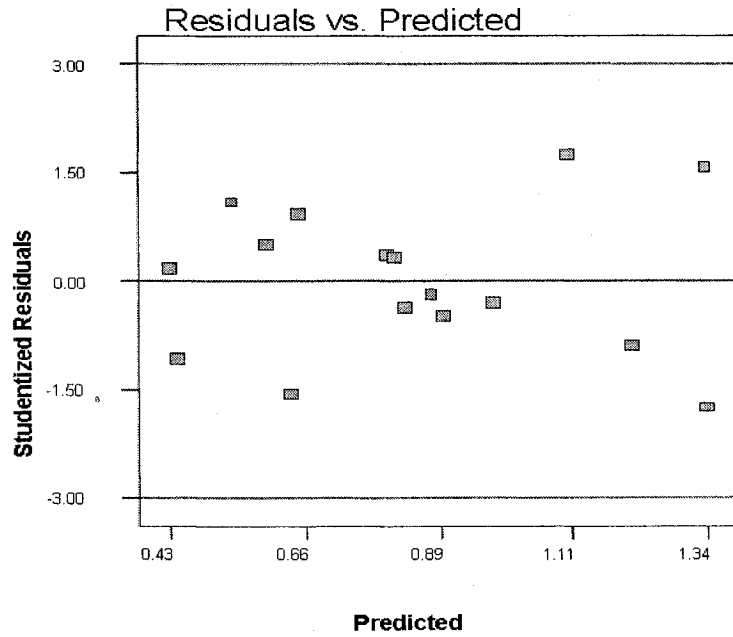


Figure E.17 Studentized residuals as a function of predicted for the data of metal competition systems.

Max Binding (Min Leaching)

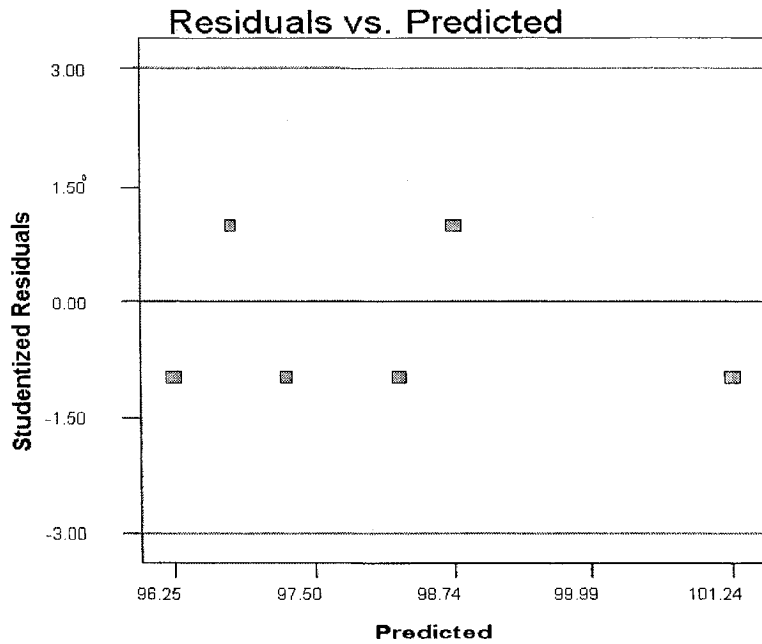


Figure E.18 Studentized residuals as a function of predicted waste stabilization data.

Max. Pptrn. for Regtn.

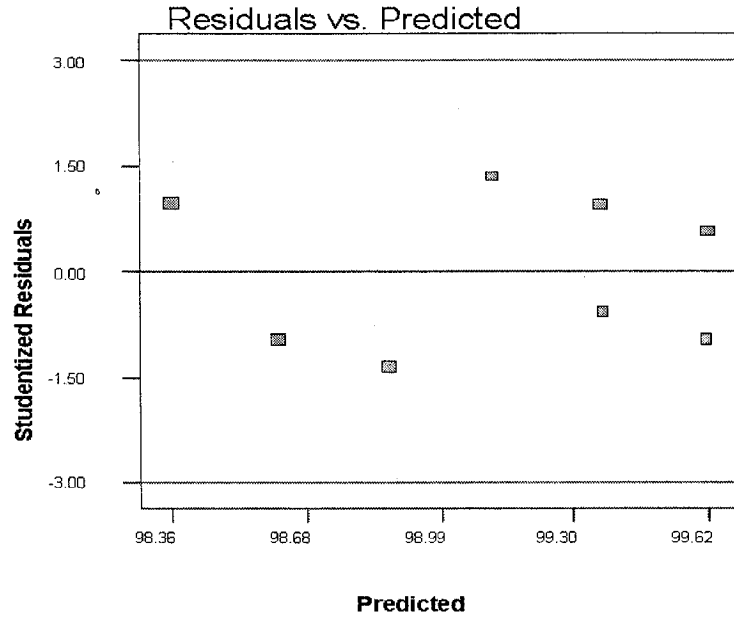


Figure E.19 Studentized residuals as a function of predicted rhamnolipid regeneration data.

Max. Water Reuse (%)

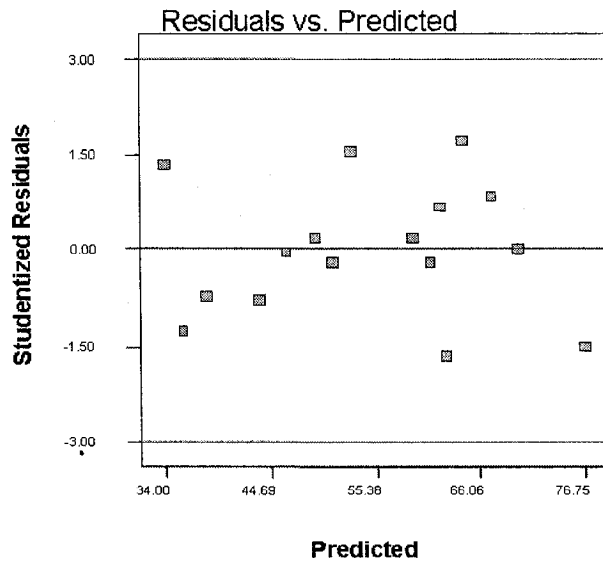
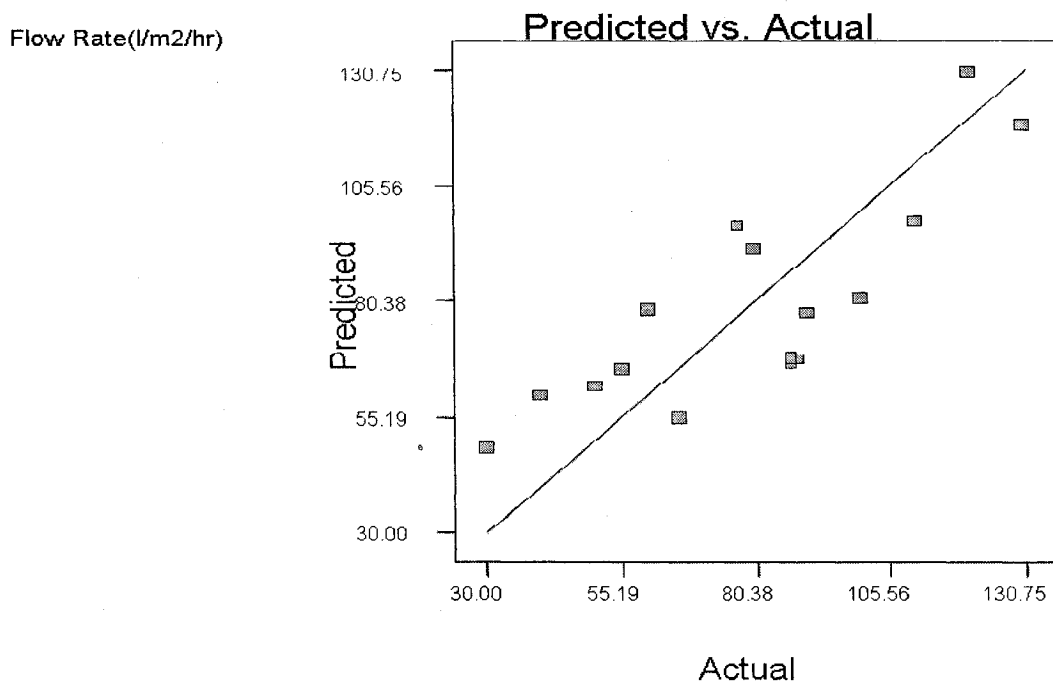


Figure E.20 Studentized residuals as a function of predicted water reuse data.

However, the general impression is that in Figures E.14 to E.20 there was no observation outside the interval of the standardized residual. This means that all the points fall within the redlines set at plus-or-minus 3.0. Consequently, there were no outliers detected for the data sets. Thus, by investigation of the need for mathematical transformation, investigation of the normality assumption and investigation of the outliers in the data sets the overall indication was that there was no potentially unusual value.



**Figure E.21 Plot of predicted versus actual response for the operating parameters.**

Max. Rejectn ( Cd and Pb )

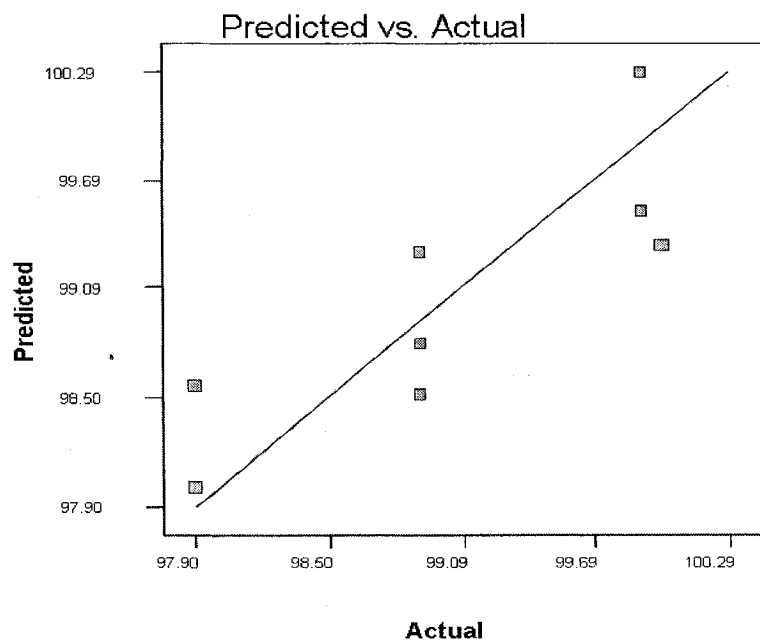


Figure E.22 Plot of predicted versus actual response for the ground water treatment data.

Max.adsorptn. (Binding Cap.)

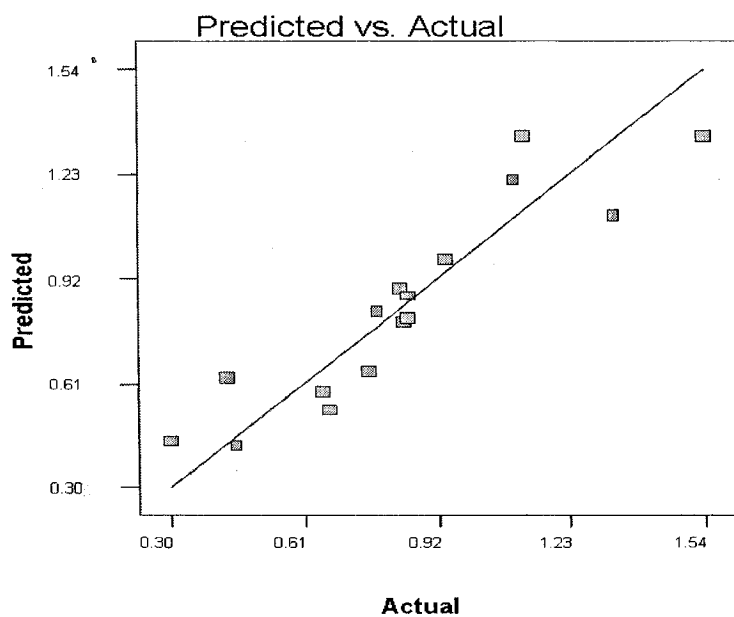
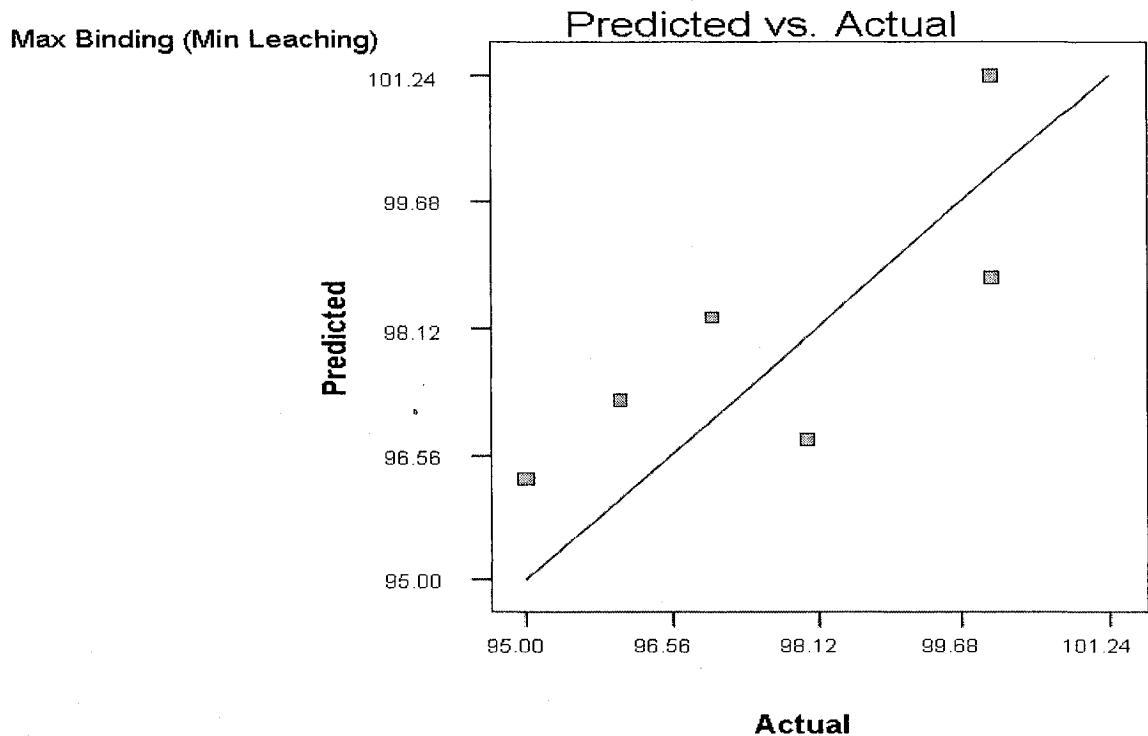
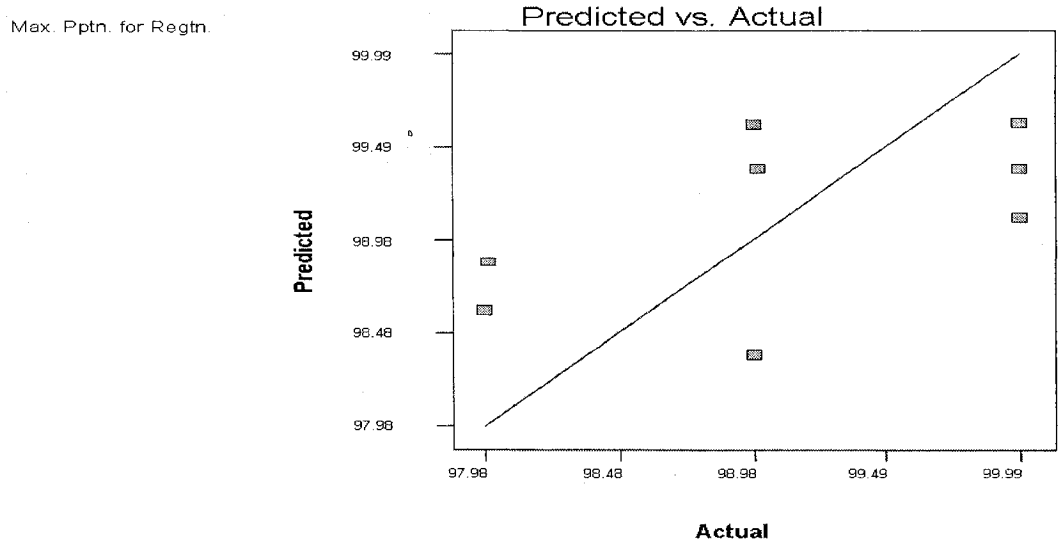


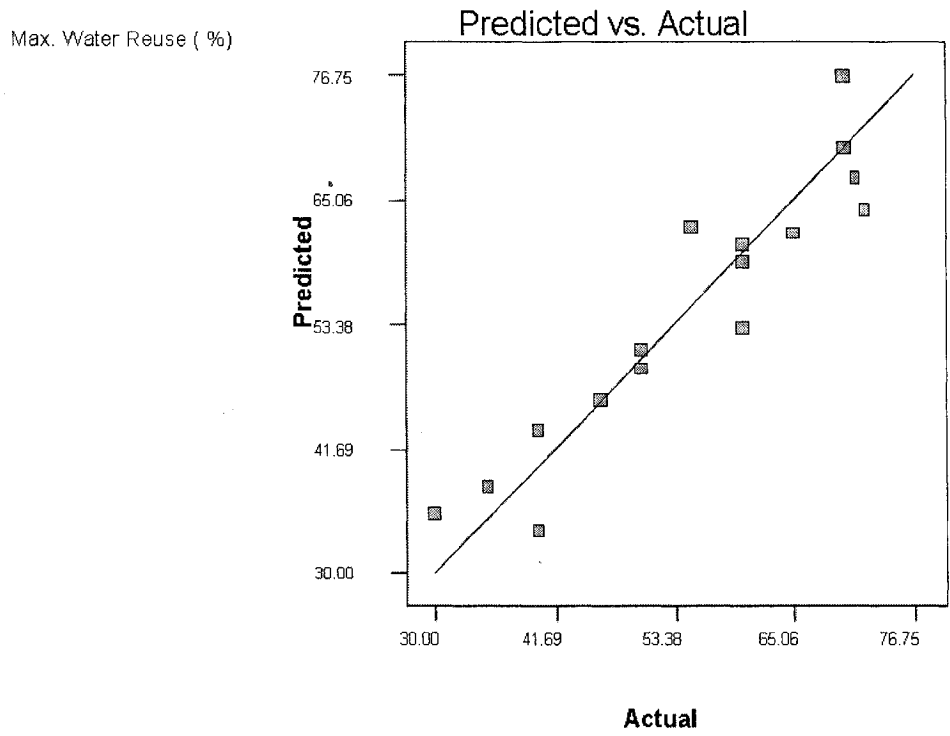
Figure E.23 Plot of predicted versus actual response for the data of metal competition systems.



**Figure E.24** Plot of predicted versus actual response for the waste stabilization data.



**Figure E.25** Plot of predicted versus actual response for the rhamnolipid regeneration data.



**Figure E.26 Plot of predicted versus actual response for the water reuse data.**

Another way for investigation of the outliers may be performed by plotting predicted versus residual responses using the sum of squares method. Figures E.21 to E.26 show the outputs when this method was applied to the data of the operating parameters, ground water treatment data, the data of metal competition systems, waste stabilization data, rhamnolipid regeneration data and water recycle data, respectively. In Figures E.22 and E.26, there is still an indication of variability of original data. However, it can be seen that most of the predicted values plot approximately along straight lines. Consequently, the Figures E.21 to E.26 indicate that there is no apparent problem with normality.