

**BIOLOGICAL TREATMENT OF INDUSTRIAL WASTEWATER CONTAINING
HIGH CONCENTRATIONS OF LINEAR ALKYL BENZENE SULFONATE
(LAS)**

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ABSTRACT

Lauryl Alkylbenzene Sulfonate (LAS) is the major anionic surfactant used worldwide in detergent and household cleaning product formulations. Its biodegradation and removal has been extensively studied in wastewater treatment facilities and laboratory-scale tests at low concentrations (<10 mg/L) typical of those encountered in municipal wastewater treatment plants. Less effort, however, has been expended investigating degradation of higher concentrations of LAS representative of those expected in wastewater generated at LAS manufacturing operations.

The research described in this thesis was conducted to study biological processes for treating wastewaters containing high concentrations (e.g., 400 mg/L) of LAS. Initial experiments were carried out using a respirometry technique, and subsequently, three different laboratory-scale bioreactor systems. The three systems studied were a Sequencing Batch Reactor (SBR), a Sequencing Batch Biofilm Reactor (SBBR), and an Intermittent Cycle Extension Aeration System (ICEAS). The SBR and ICEAS were operated on a five-day cycle basis with a hydraulic retention time of four days. The SBBR was operated mainly in a two-day cycle having a hydraulic retention time of ten days as well, and polyurethane foam cubes were used as a support medium for attached biomass growth.

The three systems were compared on their ability to remove LAS measured in terms of total organic carbon (TOC) and methylene blue active substances (MBAS). The reactors were also compared on the basis of foam production. The ICEAS showed the best performance in terms of controllable foam production while exhibiting a capacity for effectively dealing with transient periods of elevated loading. When a short Fill period was used, the SBR and SBBR had the disadvantage of producing excessive foaming, and an intermittent aeration strategy was required avoid overflow.

CHAPTER 1 INTRODUCTION

Linear alkylbenzene sulfonates are widely used as surfactants in formulated detergent products. Because of their use in household and industrial detergents, LAS is discharged into wastewater collection systems worldwide. A substantial body of literature suggests that when present at relatively low concentrations, LAS can be removed from wastewater by sorption to biomass, or alternately, LAS can be biodegraded, in which case it loses its tensioactive properties. Although many studies reported in the literature have examined the fate of LAS in municipal wastewater treatment plants and the fate of LAS discharged to the environment, there have been relatively few reports on biodegradation of LAS at high concentrations such as those present in wastewaters generated from LAS manufacturing.

The goal of the research described was to investigate the removal of high concentrations of LAS (400 mg/L) from synthetic industrial wastewater using biological processes. This objective was inspired by the desire to effectively treat an industrial wastewater stream at a manufacturing facility in Honduras that contains high concentrations of LAS. At the facility of concern, wastewater with an average LAS concentration of approximately 350 mg/L is currently treated through a dissolved air flotation system combined with the addition of cationic polymers for the removal of LAS. At the facility where the wastewater is generated, there are currently four aerated lagoons with a hydraulic residence time of five days according to the flow rate of the wastewater produced in the facility. Operational costs may be reduced if the polymer usage during the DAF stage could be reduced if the wastewater containing the LAS could be treated using the available lagoon system. Thus, research described in this thesis was conducted to investigate alternatives for biological treatment for wastewaters with high LAS concentrations. Specifically, research was carried out to compare biodegradation, LAS removal efficiency,

foaming, and required hydraulic retention times in bioreactors subjected to three different operating strategies. The reactor configurations tested included a sequencing batch reactor (SBR), an Intermittent Cycle Extension Aeration System (ICEAS), and a Sequencing Batch Biofilm Reactor (SBBR) fed with a synthetic wastewater inflow containing 400 mg/L of LAS.

To achieve the objective listed above, the research was broken down into several tasks, which are summarized below.

1.1 Selection and Enrichment of Initial Microbial Populations

Laboratory studies employed a respirometry technique to enrich for a population of microorganisms capable of biodegrading LAS. This action was undertaken for two main reasons: 1) to select for bacterial cultures capable of using it as a primary carbon source due to the similar chemical structure to LAS, and 2) to verify if high surfactant concentration would have any inhibitory or bactericide effect due to potential interactions with lipids in the cell wall. A Comput-OX Respirometer (model OO-244SC from N-CON Systems) was used to develop the LAS degrading culture. The Comput-OX Respirometer is generally used to assess the ability of a bacterial population to remove substances from wastewater (treatability or biodegradability) and to determine the effect of substances in the bacteria (e.g., inhibition or toxicity). Cultures developed using the respirometer were used as an inoculum in subsequent bioreactor operation experiments described in Section 3.3.

1.2 Development and Refinement of Operating Strategies

Experiments employed three laboratory-scale reactors. One of the reactors was operated as a Sequencing Batch Reactor (SBR), the second reactor as an Intermittent Cycle Extension Aeration System (ICEAS), and the third reactor as a Sequencing Batch Biofilm Reactor (SBBR). All three reactors were inoculated with an identical mixture of the culture produced using the

respirometer technique described in Section 1.1, and all three reactors were fed with a synthetic wastewater influent containing an LAS concentration of 400 mg/L. The length of the various operating periods (i.e., feed, react, settle, draw, and idle), as well the fraction of the reactor volume decanted per cycle were adjusted as necessary to achieve acceptable wastewater treatment. Treatment performance was evaluated on the basis of surfactant removal (e.g., reduction in soluble Total Organic Carbon (TOC), and methylene blue active substances), foam production, and settling capacity.

Based on its promising performance in terms of minimal foam production and higher LAS removal, the ICEAS was selected for testing under additional loading conditions consisting of even higher influent LAS concentrations (800 mg/L) to assess its performance under transient loading conditions which may occur in a full-scale treatment system.

1.3 Sorption Tests

The overall removal of LAS from the systems was quantified, and sorption of LAS to biomass was experimentally measured in an attempt to assess whether LAS removal from the synthetic wastewater was due to biodegradation, sorption to biomass, or some combination of the two. Also, batch sorption tests were performed with biomass collected from the SBR and ICEAS to obtain a more clear relationship between the mass of LAS that can be sorbed per mass of biomass present.

1.4 Anti-foaming Agents Testing

A potential problem of great practical concern in aerobic biodegradation of surfactants is that the aeration supply, whether surface aerators or bubble diffusers, can cause excessive foam production. Excessive foam production is an aesthetic concern and it can also cause excessive loss of biomass. To assess the potential for chemical addition to minimize foam production

should it be necessary in the biological treatment process, a screening study was conducted to evaluate the potential for several different anti-foaming agents to minimize foaming in solutions containing LAS. Based on the results of the screening study, one of the anti-foaming agents was selected for further testing in the SBR and SBBR. The ICEAS was not submitted to anti-foam addition due to the low and controllable quantities of foam produced during the operating cycles.

1.5 Thesis Organization

Chapter two of this thesis contains a literature review summarizing previous studies on biodegradation of LAS, its fate in the environment, and biological waste treatment processes that may be applicable for its removal. Chapter three contains a description of the materials and methods used in experiments. Chapter four contains results and discussion. Chapter five presents overall conclusions as well as recommendations for future research. References cited throughout the thesis can be found in the reference section.

CHAPTER 2 LITERATURE REVIEW

2.1 Overview of Linear Alkylbenzene Sulfonate (LAS)

Linear alkylbenzene sulfonate (LAS) has become the major cleaning agent for laundry detergents in most parts of the world (Leon *et al.*, 1990). LAS surfactants were introduced in the 1960's because they were found to be fully biodegradable, in contrast to the branched chain products like tetrapropylbenzenesulfonates (Leon *et al.*, 1990). About 2.5 million tons per year of LAS are produced worldwide (Schulze, 1996), accounting for an estimated 28% of all synthetic surfactants.

Due their high volume use in consumer products, detergent chemicals have the potential for broad-scale release into aquatic and terrestrial environments. Following its widespread use as a household product, LAS is typically disposed of in wastewater. Two primary routes exist for LAS to enter the environment, (i) effluents from sewage treatment facilities which discharge to rivers, lakes, and estuaries, and (ii) municipal sludge, which is applied to agricultural lands as a soil conditioner. The concentration of LAS in municipal wastewater is variable depending on its use in industrial processes in addition to domestic activities. Average influent concentrations of 1 – 10 mg/L have been reported for municipal wastewater treatment facilities receiving only municipal wastewaters (Metcalf and Eddy, 1993).

Commercial LAS is composed of a linear alkyl chain consisting of 10-14 carbon atoms, a benzene ring, and a sulfonate group (see Figure 2.1). The alkyl chain includes, on average, 11.7 carbon atoms (Swisher, 1987). The benzene ring is randomly distributed in all positional isomers except the 1-phenyl and the sulfonate group in *para* position. This mixture is the synthetic chemical product whose biodegradation has been examined to the greatest extent by regulatory agencies (OECD, 1981).

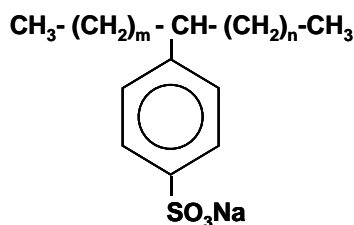


Figure 2.1: Structure of 3-(4-sulphophenyl) dodecane (3-C12-LAS) (Swisher, 1987)

2.2 Biodegradability of LAS

Numerous studies have been conducted by industrial and academic scientists during the past 30 years on the fate, environmental effects, and relative environmental safety of LAS (e.g., Larson *et al.*, 1981; Lee *et al.*, 1997). Much of the biodegradability database has been developed in standard laboratory tests, which are routinely used in North America and Europe to determine the biodegradation potential of organic substances prior to their introduction as consumer products (OECD, 1981).

In much of the work conducted to date regarding biodegradation of surfactants, the Dieaway test has been used to quantify biodegradation. In such cases, the surfactant being tested is exposed to microbes in an isolated system, and the progress of biodegradation is observed by analysis conducted over time as the surfactant “dies away” through biodegradation using the MBAS assay (Swisher, 1987). The dieaway rate of the surfactant is expressed as a half-life, the time taken for the concentration to drop to half its initial value.

Studies on biodegradation of LAS have also included research on biodegradation of dialkyltetralin sulfonate (DATS), (Trehy *et al.*, 1995), and sodium dodecylsulfonate (SDS), (Zhang *et al.*, 1999). The chemical structures of DATS and SDS are depicted in Figure 2.2.

DATS can be produced during the synthesis of Lauryl Alkylbenzene (LAB), followed by sulfonation, where LAB is converted in LAS. The mixture of alkyl chain lengths used to prepare homologs for LAB results in varying chainlength DATS. DATS structures are more complex

than LAS due the formation of cis/trans isomers. DATS biodegrades to tetralinsulfonate carboxylates intermediates (DATSI). Primary biodegradation of DATS and LAS has been determined by following the reduction of methylene blue active substances (MBAS) over time. LAS levels are generally reduced more rapidly than DATS (Trehy *et al.*, 1995). Testing of a 100 µg/L DATS blend for several weeks in a semi-continuous activated sludge system with natural sewage sludge populations resulted in nearly complete (99%) primary biodegradation, based on MBAS results under steady-state conditions (Trehy *et al.*, 1995).

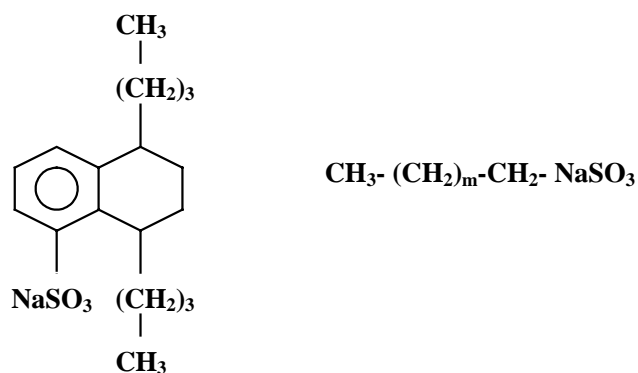


Figure 2.2: Chemical structure of C₈- DATS (left) and SDS (right) (Trehy *et al.*, 1995)

SDS is comprised of straight carbon chains, usually 12 to 14, and is incorporated into household products together with LAS. Due to its more simple structure (i.e., no benzene ring), SDS can undergo rapid complete mineralization in wastewater treatment facilities within 48 hours (Fendinger *et al.*, 1994). Experiments performed by Zhang *et al.* (1999) demonstrated that for a mixed culture of activated sludge from a municipal wastewater treatment plant, incubation with an SDS concentration between 500 -2500 mg/L increased microbial specific growth rates in comparison to cultures incubated with lower SDS concentrations (in the range of 0.0379 to 0.0567 h⁻¹), indicating not inhibitory effects.

Half-life values determined for LAS in natural environmental compartments have shown that biodegradation is a significant removal mechanism for LAS (Larson *et al.*, 1995).

According to Larson *et al.*, for low concentrations of LAS (i.e., in the range of 1 to 100 µg/g), half-lives in aquatic and benthic compartments, where the residence time can vary from days to weeks, have been observed to be one day or less. Meanwhile, in terrestrial and subsurface compartments where the residence time can vary from months to years, half-lives in the range from less than one day to a few weeks have been observed (Takada *et al.*, 1987).

Laboratory-scale tests showed that the rate of LAS removal is biphasic based on kinetic analysis of radiolabeled LAS using acclimated sludge (Nielsen *et al.*, 1997). Porous Pot Biodegradation Test System, which assesses biodegradation of the test compound simulating a wastewater activated sludge treatment, showed that the first-order initial rates for LAS removal were at least twice as fast as the apparent zero-order final rates. This study claims that the initial rates correspond to both mineralization and incorporation of the radiolabeled LAS into cell components. The slower, final rate reflects the turnover of the incorporated carbon, or just the formation of more biologically stable metabolites. However, explanations on the mechanisms or significance of the final rate are lacking. Overall results showed that 98.4% of the parent LAS were removed after 45 days. From this, 86.1% suffered ultimate biodegradation conformed by 57.5% of mineralization, measured as % ¹⁴CO₂, and 28.6% incorporated into cell biomass. The rest, 13.9%, remained as residual in the liquid portion. Half-life ranged between 3.4 and 4.6 days.

A generalization of the LAS biodegradation pathway has been established (Swisher, 1987). As shown in Figure 2.3, biodegradation begins with oxygenation at the end of the alkyl chain, yielding a carboxylic acid, which is subject to β-oxidation, followed by opening of the ring with conversion of the sulfonate group to inorganic sulfate.

For LAS to be biodegraded following the pathway depicted in Figure 2.3, it is understood that aerobic systems will have an advantage over anaerobic systems due to the fact that the first stage of degradation, oxygenation at the end of the alkyl chain, requires oxygen.

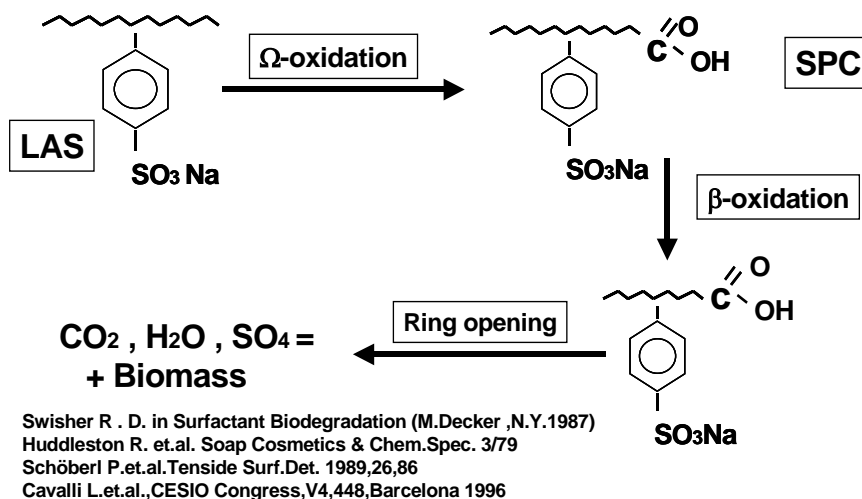


Figure 2.3: Biodegradation pathway of LAS

Although most of the literature describing experiments to examine degradation of LAS under aerobic conditions has reached the common conclusion that this compound is biodegradable (see, for example, Pérez et al., 1996, Lung et al., 1990, Trehy et al., 1996), there have been some conflicting reports. For example, in a study conducted to evaluate surfactants for use in enhancing pump-and-treat processes for clean-up of contaminated soils and aquifers, Zhang *et al.* (1999) reported that the sodium salt of LAS was not biodegraded even after three weeks of incubation with activated sludge from a municipal wastewater treatment facility; LAS did not induce microorganism's growth at the tested concentration, 500 mg/L. The authors' conclusion from this study was that the compound resisted biodegradation likely due to the presence of the benzene ring.

Few studies have directly examined the biodegradability of LAS in anaerobic environments. Some previously reported studies indicate that LAS degradation can proceed under anaerobic conditions if preceded by a period of aerobic exposure (Larson *et al.*, 1995). In

these studies, a series of ^{14}C -LAS homologs (C10-C14) in concentrations of 100 $\mu\text{g/g}$ were incubated aerobically for 5-6 hours in activated sludge and then transferred to digester sludge and incubated under strictly anaerobic (methanogenic) conditions. After a period of aerobic exposure, mineralization of individual LAS homologs in anaerobic sludge was comparable to that observed in aerobic sludge. Half-life values for anaerobic degradation ranged from 2.1 to 2.6 days and showed no significant difference as a function of alkyl chain length. The researchers hypothesized that aerobic exposure allows ω -oxidation of the terminal carbon of the alkyl side chain. The initial oxidative attack is the only step that requires molecular oxygen. Once formed, the sulfophenyl carboxylates can be biodegraded via beta-oxidation and ring hydroxylation/cleavage under strictly anaerobic conditions.

Indirect support for the lack of LAS degradation under anaerobic conditions comes mainly from studies reporting that high concentrations of LAS are commonly found in anaerobic digester sludges at municipal wastewater treatment facilities (McEvoy *et al.*, 1986). Studies have indicated that the inhibition of LAS degradation during anaerobic digestion is due to LAS toxicity and the exposure time as a factor of influence (Mösche *et al.*, 2002). However, anaerobic transformation of LAS during digestion of sewage sludge in a CSTR has been achieved when the surfactant concentration was below 200 mg/L (Angelidaki *et al.*, 2000). It was found that 14-25% of LAS fed to the anaerobic digester was transformed, and that the transformation was limited by bioavailability due to sorption and toxic effects as the surfactant concentration increased.

2.3 Biodegradation of LAS and Other Surfactants in Wastewater Treatment Plants

Although a number of studies (see previous section) have demonstrated that LAS is biodegradable, that does not mean that it is necessarily biodegraded under conditions imposed in

wastewater treatment facilities. To address this latter issue, several studies have been conducted to determine the fate of LAS during wastewater treatment. One such study encompassed a total of 50 municipal wastewater sites and included 15 activated sludge systems, 12 trickling filters, 6 oxidation ditches, 8 lagoons, and 9 rotating biological contactor, (RBC), treatment facilities in the United States (McAvoy *et al.*, 1993). Influent concentrations of LAS for all treatment plants showed a normal distribution with a mean of 5 mg/L. Average effluent LAS concentrations ranged from 0.04 mg/L for activated sludge plants to about 1 mg/L for trickling filter plants. A range of removal rates over 99% for activated sludge treatment and an average of 77% for trickling filter plants were observed. Average removal of LAS in other treatment plant types ranged from 96% to 98%.

Results from McAvoy *et al.*, 1993 show close agreement to LAS concentrations measured from 1973-1986 that were reported by Rapaport and Eckhoff (1990). There is close agreement in the two studies supporting almost constant influents and effluents over the past 15-20 years. The more recent study indicates improved activated sludge plant performance over the 1973-1986 results, where an average of only 0.7% versus 0.2% of influent LAS remaining in the sewage effluent. Similar results were reported in a study performed in Torino, Italy (Cavalli *et al.*, 1992). The wastewater treatment plant presented a hydraulic retention time (HRT) of 4 days with a sludge retention time of 12 days, and a MLSS concentration of 2500 mg/L. Although these studies suggest that low concentrations of LAS can be readily removed from municipal wastewater using biological processes, some difficulties related to foaming and long retention times have been found in the treatment of wastewater with high (in the range of 20 to 50 mg/L) LAS concentrations (McAvoy *et al.*, 1993).

Although LAS has been successfully removed from wastewater by aerobic processes in full-scale municipal wastewater treatment plants, much of the surfactant load into a treatment

facility may be removed by sorption to suspended solids (e.g., MLSS), rather than by direct biodegradation by aerobic microorganisms. In such cases, at least of a portion of the LAS removed from the influent wastewater is directed via primary or secondary sedimentation into sludge management processes. Mösche *et al.* 2002, suggested that when surfactant-containing wastewater is fed to a bioreactor, the surfactant concentration will decrease initially due to adsorption by biomass. The theory postulated by Mösche is supported by studies reported by Rittmann *et al.*, (2001) where a nonsteady-state model was used to calculate the effects of community adaptation and sorption kinetics on the fate of LAS in batch experiments with activated sludge that was fed with different concentrations of LAS. They found that when LAS stays in solution it will rapidly degrade, but slow desorption of LAS initially sorbed to the sludge occurs may limit biodegradation rates due to limited bioavailability. Also, it has been found that concentration of LAS in dewatered sludge ranged between 11 and 16,000 mg/Kg-total solids depending on the way the sludge is stabilized (Madsen *et al.*, 1999), giving more support to the concept of sorption of LAS in systems where activated sludges are used.

Aerobic composting of anaerobic sludge, where the highest concentrations of LAS is normally found in municipal wastewater treatment plants, helps to remove the LAS, but it is not a common practice done by wastewater treatment facilities (Federle, *et al.*, 1990). LAS has been reported to be removed in large quantities (>97%) during composting of anaerobic sludge in a very short period of time. Prats, *et al.* (2000), reported that the concentration of LAS was reduced by 50 and 90% of the initial level after 3 and 9 hours, respectively, in the systems that they studied. LAS removal occurred even during poor composting conditions like poor aeration and sub-optimal temperatures. In cases where sludge is applied on agricultural soil, where the nutrient content is utilized for plant growth, it will generally contain LAS. However, LAS is not

expected to accumulate in the top layers of the soil, which is supported by studies of vertical distribution of microbial biomass, activity and biodegradation of LAS in the subsurface of two soil profiles (Federle, *et al.*, 1990), and biodegradation will occur in the upper soil layers once the microorganisms present are previously exposed to LAS.

Following secondary treatment, the majority of wastewater effluents are discharged into freshwater receiving bodies. In such cases, LAS not removed by the wastewater treatment process can remain in solution or be sorbed by sediments. Half-lives of LAS in river water in the range from 21 to 31 hours have been reported (Larson *et al.*, 1995). This indicates that microbial communities in streams in contact with LAS may be well acclimated and in-stream biodegradation can play a major control when secondary wastewater treatment is minimal and direct exposure of wastewater to receiving water occurs (Romano *et al.*, 1992). Takada *et al.* (1987), reported the presence of LAS in freshwater sediments where the average concentrations are quite variable, ranging from less than 1 to greater than 100 $\mu\text{g/g}$. In general, the authors found that degradation of LAS was comparable in river water and sediments and proceeded at similar rates when sediments were exposed to gentle agitation.

Coastal estuarine environments receive approximately 780 million cubic meters (3 trillion gallons) of domestic wastewater a year, representing 20–30% of the total domestic wastewater flow in the United States (Lung *et al.*, 1990). Studies conducted in the United States (Larson *et al.*, 1995) and Europe (Leon *et al.*, 2001) conclude that the kinetic patterns observed for LAS degradation in estuarine water and sediments were comparable to those observed in the freshwater systems. The rate and extent of biodegradation were most extensive in sediment samples collected from acclimated sites, those with long exposure histories), and less extensive in water samples collected from control sites having little or no prior LAS exposure, indicating

the importance of prior exposure on rapid degradation. Close work with bacterial communities selected from coastal seawater continuously polluted by urban sewage showed that complete surfactant biodegradation was achieved by the biodegradation pathway described by Swisher (Sigoillot *et al.*, 1992).

2.4 Sequencing Batch Reactor Technology

Sequencing batch reactor (SBR) systems have been used to remove specific organic compounds present in industrial effluents (Irvine *et al.*, 1989). SBRs are a specific fill-and-draw version of the activated-sludge process. Metabolic reactions and solid-liquid separation are carried out in one tank in a well-defined and continuously repeated time sequence (Wilderer *et al.*, 1993). Sequencing batch reactors have been used to manipulate both the organisms' distribution established in the reactor and the physiological state of the organisms developed (Morgenroth *et al.*, 1998). Physically, the SBR system is a set of tanks that operate on a fill-and-draw basis. Each tank is filled during a discrete period of time and then operated as a batch reactor. After treatment of the target compounds, the mixed liquor is allowed to settle and clarified supernatant is drawn from the tank.

The complete SBR cycle consist in four discrete steps: (1) reactor filling; (2) reaction; (3) biomass settling; and (4) effluent decanting and discharge. During the *fill* period, the influent wastewater containing the target compounds is added to biomass retained in the system after the previous cycle. The influent volume added can be as little as 25% of the total volume of the reservoir or as great as 70% of it (Irvine *et al.*, 1989). Degradation of the target compounds, which may be initiated during the fill period, is completed during the *react* stage. The duration of this period is usually dictated by the time where the target compound reaches a desire concentration. Time dedicated to react can vary from a low of zero to more than 50% of the total cycle time (Irvine *et al.*, 1989).

One of the advantages of sequencing batch reactors is that the same tank serves as clarifier during quiescent settling conditions. Under these conditions the *settle* period takes place and biomass separation is the desired result. After the settle period is complete, the treated supernatant is removed and discharged during the *draw* period. The volume to be discharge is the same as in the fill period, and the time to accomplish it can range from 5 to more than 30% of the total cycle time. If no wastewater is available (e.g. on industrial application sites), the SBR can rest in an *idle* phase. The sum of the phases make up a process cycle that is progressively repeated. During each cycle, unsteady-state conditions prevail. In the long term, control and periodic repetition of the short-term unsteady state allows the enhancement of certain effects such as (a) enzymatic activity, (b) accumulation of metabolic products, and (c) selection and enrichment of specific groups of microorganisms (Morgenroth *et al.*, 1998).

In a study on biodegradation of alternate types of surfactants, the non-ionic surfactants Neodol 91-8 and Makon 12, at concentrations between 100 and 500 mg/L, using sequencing batch reactors (SBRs), Figueroa *et al.* (1997) demonstrated that biodegradation could be achieved when acclimated microorganisms were present and an appropriate system operation was employed. The total times for one SBR cycle were 2, 3, and 10 days for initial surfactant concentrations of 100, 250, and 500 mg/L respectively. The authors used a mechanical mixer instead of diffused air as an oxygen supply mechanism to avoid production of foam. Relatively low (in the range of 5 to 35 mg/L measured as TOC) concentrations of the surfactants at the end of the treatment where accomplished (Figueroa *et al.*, 1997).

Although the term SBR was originally introduce to describe a specific type of activated sludge periodic process characterized by continuous repetition of periods of fill, react, settle, draw, and idle, it is also used to describe various alternate versions of variable volume systems.

Variants of the SBR include ICEAS (Intermittent Cycle Extension Aeration System) and SBBR (Sequencing Batch Biofilm Reactor). The ICEAS process is a variant of an SBR system where the processes of biological oxidation, nitrification, phosphorous removal and liquids/solids separation can be achieved continuously in a single tank. What makes the ICEAS process different is a continuous inflow, even during the settle and decant phases of the operating cycle (Khararjian *et al.*, 1990). In a conventional SBR, wastewater enters each tank during only a portion of the operating cycle (i.e., the fill period). An ICEAS process combines activated sludge and extended aeration principles in a fill-and-draw basis which accommodates continuous inflow to the tank. The ICEAS relies on a timed sequence of events, whereas traditional SBR systems may rely on liquid level controls to sequence events. A conventional SBR has required periods to allow settling and decant, at least two tanks are needed to treat a continuous flow of wastewater. In an ICEAS system, a single tank may be employed.

The SBBR (Sequencing Batch Biofilm Reactor) combines the benefit of attached growth process and batch system. The organisms responsible for treatment are attached to the surfaces of media such as rock, sand, plastic or other materials (Chemical Engineering World, 2001). Biofilm processes maintain high cell densities and retain cultures of slow growing or poorly settling microbes that would be washed out in suspended growth systems (Bryers and Characklis, 1990). An SBBR cycle usually consists of three phases: fill, react, and draw, no time for settling is required. Suspended solids and detached biomass may be retained in the reactor during the drain phase as a result of filtration processes or they may be discharged. During the fill period, the flow conditions in the packaging may be laminar, plug-flow conditions; and mixing of wastewater constituents in flow direction remains limited (Arnz *et al.*, 2000). Periodic operation of biofilm reactors and the length of the fill period can result in an even distribution of the

biomass throughout the reactor and as a result, the entire system is better able to treat large shock loads compared to a continuous flow process (Wilderer *et al.*, 1993).

CHAPTER 3 MATERIALS AND METHODS

3.1 Overview

Initial laboratory studies employed a respirometry technique to enrich for a population of microorganisms capable of biodegrading LAS. The experimental procedures employed for these initial experiments are described in Section 3.2. The cultures developed in the initial respirometer studies were then used as a seed culture to inoculate a series of sequencing batch reactors (SBRs) as described in Section 3.3. Next, the cultures developed in the initial SBR experiments were used as a seed culture to inoculate subsequent SBR, ICEAS, and SBBR experiments during which time a more detailed analysis of operational performance was conducted, as described in Section 3.4. Analytical techniques employed in the experiments are described in Section 3.5.

3.2 Development of LAS Degrading Microbial Population Using Respirometry

The respirometry technique for developing an LAS degrading culture employed a model 244SC Comput-OX Respirometer (N-CON Systems, Crawford, GA). The Comput-OX was calibrated following the manufacturer's recommended protocol, and two water baths were used to hold four 500 mL borosilicate glass respirometer reactors each (total of eight reactors). Temperature was maintained a constant 25°C. The total liquid volume for each of the reactors was 350 mL. Oxygen uptake rate readings were recorded at 15-minute intervals.

For initial tests, three different surfactants were used: Sodium Dodecylsulfate (SDS) (Aldrich 43,614-3), Sodium Dodecylbenzene Sulphonic Acid (SDBS) (Aldrich D-2525), and Alkylate 225 Sulphonic Acid (Huntsman). For the purposes of clarity, the acronym LAS is used throughout the remainder of this thesis to refer to the Sodium Dodecylbenzene Sulphonic Acid (SDBS) obtained from Aldrich to clearly differentiate it from the SDS. Stock solutions of each

of the surfactants, each containing 10,000 mg/L surfactant, were prepared using deionized water. Calculations accounted for the fact that the surfactants, as obtained from the vendors, were not 100% purity. For example, the LAS (Aldrich D-2525) was approximately 80% surfactant including all homologues [main homologues are C10 - C13 with homologue C12 comprising approximately 20%. The remainder is sodium sulfate and sodium chloride (approximately 17%) and water (approximately 3%) – data provided by the manufacturer]. Thus, to make a 10,000 mg/L surfactant solution containing LAS, 12,500 mg of Aldrich D-2525 was added per 1.0 L of solution.

Separate nutrient stock solutions were also prepared for each of the constituents listed in Table 3.1, using tap water.

Table 3.1. Nutrients solutions concentrations

Component	Nutrient stock solution concentration (g/L)	Volume delivered into glass reactor (mL)	Final nutrient concentration in reactors (mg/L)
Iron	1.2 as FeSO ₄	1.31	4.5 as FeSO ₄
Magnesium	15.0 as MgSO ₄	0.70	30 as MgSO ₄
Calcium	3.0 as CaCl ₂	1.30	10.8 as CaCl ₂
Manganese	1.8 as MnSO ₄	1.30	6.6 as MnSO ₄
Inorganic nitrogen	30.0 as NH ₄ Cl	0.92	78.6 as NH ₄ Cl
Phosphorus	9.0 as Na ₂ HPO ₄	1.56	40 as Na ₂ HPO ₄
	9.0 as KH ₂ PO ₄	1.56	40 as KH ₂ PO ₄
Buffer	60.0 as NaHCO ₃	5.60	960 as NaHCO ₃

Total organic carbon (TOC) and Chemical oxygen demand (COD) equivalents for SDS and LAS were calculated and measured to help in construction of TOC standards curves , stock solution preparation , and determine size samples for accurate and reliable results. Table 3.2, shows the calculated and measured TOC and COD equivalents for SDS and LAS. Stoichiometric

equations (3.a for LAS and 3.b for SDS) were used to calculate the TOC and COD equivalents. Direct COD analysis was performed in samples of standard solutions of SDS and LAS.

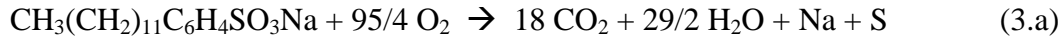


Table 3.2: TOC and COD equivalents for SDS and LAS

	Calculated, mg/mg	Measured, mg/mg
SDS	0.528 (TOC)	0.648 ± 0.05 (TOC)
	1.96 (COD)	---
LAS	0.528 (TOC)	0.571 ± 0.03 (TOC)
	2.20 (COD)	2.78 (COD)

Each of the eight 500 mL glass respirometer reactors was cleaned and prepared according to the instructions enclosed in the user's manual, and 12 pellets of KOH (Fisher Scientific, Cat. No. P250-500) were introduced into the CO₂ trap attached to each cap. The general procedure for preparation of the reactors for every test was the same. First, 150 mL of deionized water was added to each reactor followed by the delivery of stock nutrient solution (quantities in milliliters specified in Table 3.1). Then, a volume of activated sludge was added to attain 100 mg/L TSS in a final liquid volume of 350 mL. Reactors were submerged in the water baths and equilibrated for one hour to reach the working temperature of 25°C. Finally, surfactant was added by pipetting a sufficient volume of the stock surfactant solution to obtain the desired test concentration. Deionized water was then added as necessary to obtain a final liquid volume of 350 mL. This resulted in a final nutrient concentration in each reactor as listed in Table 3.1. Finally, the reactors caps containing the CO₂ traps were put in place to seal the reactors and operation of the Comput-OX was started to provide automated O₂ addition and consumption measurements.

Table 3.3 summarizes the various experiments (ordered chronologically, arbitrarily referred to as Experiments I-IX) that were carried out in the respirometer. As shown in the table, activated sludge from a municipal wastewater treatment facility (Baton Rouge Central Treatment Plant) was used as the inoculum in Experiment I, and activated sludge from a petroleum refinery wastewater treatment facility (ExxonMobil, Baton Rouge, LA) was used as the inoculum in Experiment V. All other experiments used activated sludge derived from these reactors. For Experiments II-IV and VI-XI, activated sludge from a recently completed experiment was collected by filtering (Whatman 42), washed with nutrient solution, and then re-suspended in deionized water. The Total Suspended Solids (TSS) concentration for each of the sludge samples was performed in duplicate following standard method 2540C (APHA, 1998). Reactors were inoculated the same day samples of activated sludge were collected. The surfactants employed and their initial concentrations in the various reactors varied between the different experiments as summarized in Table 3.3.

At regular time intervals, duplicate 4-mL samples were drawn from each reactor through a sampling port located in the reactor cap using plastic 5 mL syringes. The samples were then filtered through a 0.45 μm syringe filter (25 mm diameter, Whatman, Cat. No.6874-2504), and collected in 5 mL glass vials. Total Organic Carbon (TOC) was measured immediately after collection. In some experiments, surfactant concentrations were also measured in terms of methylene blue active substances (MBAS) as described in Section 3.5.

3.3 Biomass Production

Three 4.0 L glass kettle reactors, each 35 cm in height and 13 cm in internal diameter (Pyrex®, Corning, NY), were used to further enrich and increase the quantity of biomass containing microbial populations capable of using LAS as a carbon source. A schematic diagram of the reactor configuration is shown below in Figure 3.1.

Table 3.3. Summary of experiments performed using the Comput-OX Respirometer.

Experiment No.	Duration (hours)	Reactor Inoculum	Inoculum concentration (mg/L MLSS)	Surfactant type	Surfactant concentration (mg/L)
I (June 23 – July 8, 2002)	360	Activated sludge from municipal wastewater treatment plant	100	SDS	30 60 150 300
II (July 11 – July 26, 2002)	360	Biomass collected from Experiment I	100	SDS	300 400 500 600
III (Aug. 31 – Sept. 20, 2002)	480	Biomass collected from Experiment II	100	SDS	100 400 1000 2000
IV (Sept. 21 – Oct. 6, 2002)	264	Biomass collected from Experiment III	100	LAS	30 60 100 200 300 400
V (Oct. 10 – Oct. 31, 2002)	266	Activated sludge from petroleum refinery wastewater treatment facility	100	LAS	30 60 100 200 300 400
VI (Oct. 31 – Nov. 20, 2002)	266	Biomass collected from Experiment V	100	LAS	100 200 300 600
VII (Nov. 21 – Dec. 10, 2002)	288	Biomass collected from Experiment VI	100	LAS	500 (All reactors)
VIII (Dec. 10 – Dec. 21, 2002)	288	Biomass collected from Experiment VII	100	LAS	500 (All reactors)
IX (Jan. 06- Jan. 22, 2003)	384	Biomass collected from Experiment VIII	100	LAS Alkylate 225 Huntsman	500 (3 reactors) 500 (4 reactors)

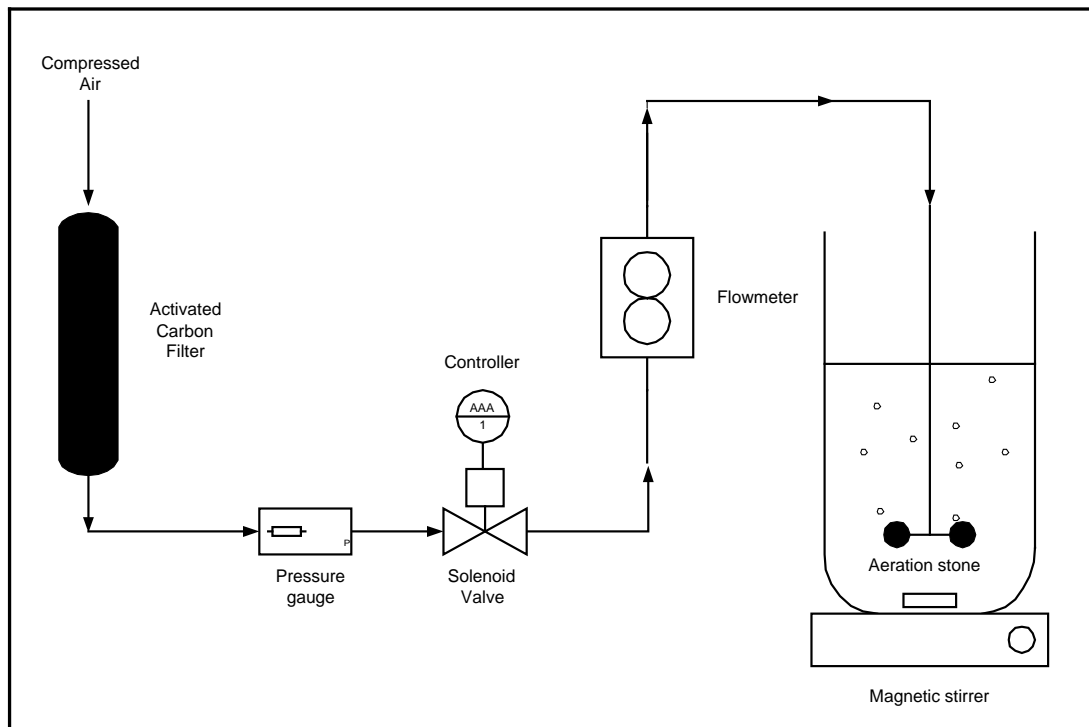


Figure 3.1. Schematic diagram of laboratory-scale reactor configuration.

Laboratory air passed through an activated carbon trap (type F-300, Calgon Carbon Corporation, Pittsburgh, PA) to removed unwanted contaminants, and pressure was controlled by a regulator (series R35, Arrow Pneumatics, Inc., Broadview, IL). The air flow rate, set to 500 mL/min, was measured and regulated with a rotameter (Gilmont Instruments, 150 mm scale Accucal flow meter Cole-Palmer Instruments Co., Vernon Hills, IL). Air entered the reactor via two fine bubble diffuser stones (Fisher Scientific, Sewanee, GA) connected to the influent airline by Viton™ tubing.

An intermittent aeration strategy was implemented to avoid excessive foaming due the presence of surfactant. A programmable microprocessor (Model XT, Chron-Trol Corp., San Diego, CA) controlled the opening and closing of a stainless steel solenoid valve connected to the air stream (Automatic Switch Company, NJ) to provide aeration for 30 seconds every 10

minutes. This aeration frequency was sufficient to maintain a dissolved oxygen concentration between 3.5 and 6.5 mg/L during the reactors' operation. Reactors were placed on magnetic stir plates to allow mixing using a Teflon-coated stir bar.

The first reactor (arbitrarily named R1) was inoculated using activated sludge collected from the eight respirometer reactors at the end of Experiment III (September 20, 2002 – see Table 3.3). The second reactor, R2, used biomass collected from respirometer experiment VI (November 23, 2002– see Table 3.3), and the third (R3) was inoculated with biomass collected from respirometer Experiment IX (January 22, 2003 – see Table 3.3). To collect biomass from the respirometer bottles to inoculate the reactors, the contents of the respirometer bottles were filtered (Whatman 42) and then washed using a nutrient solution consisting of the following compounds added to tap water: FeSO₄ (4.5 mg/L), MgSO₄ (30 mg/L), CaCl₂ (10.8 mg/L), MnSO₄ (6.6 mg/L), NH₄Cl (78.6 mg/L), Na₂HPO₄ (40 mg/L), KH₂PO₄ (40 mg/L), and NaHCO₃ (960 mg/L). The washed sludge was then re-suspended in nutrient solution, and the TSS concentration was measured in duplicate.

An identical start-up procedure was used for each reactor (R1, R2, and R3). The process consisted of adding a sufficient volume of tap water to the washed activated sludge to result in a mixed liquor suspended solids (MLSS) concentration of 2000 mg/L in a volume of 1.25 L. The reactor was then aerated continuously for 30 minutes before aeration was temporarily stopped and 1.25 L of synthetic wastewater was added to bring the total reactor volume to 2.5 L with an MLSS concentration of 1000 mg/L. The synthetic wastewater consisted of the surfactant type and concentration listed in Table 3.3 along with the nutrient concentrations listed in Table 3.1 added to tap water. Once the reactors were filled, the intermittent aeration cycle was initiated and controlled by the programmable microprocessor. Table 3.4 summarizes the characteristics, operation times, and surfactant types to which each reactor was subjected.

R1 was operated using a sequencing batch strategy. The complete SBR cycle consisted of four steps: (A) reactor filling, (B) reaction, (C) solids settling; and (D) decanting and discharge. After decanting, the sequence was repeated. The reactor was filled to a total liquid volume of 2.5 L and were decanted to 1.25 L of liquid volume (i.e., the fill and draw volume was equal to one half of the total working volume) using a Cole Palmer Materflex® console drive peristaltic pump (model 7521-40, Bernant Co, Barrington; IL). The reactor was filled with a synthetic wastewater influent containing 400 mg/L of surfactant and nutrients in composition and concentrations as shown in Table 3.4. The first four SBR cycles were five days in length each and SDS was the surfactant used. The following eight cycles were ten days in length each and the surfactant type was changed from SDS to LAS.

Total organic carbon, methylene blue active substances, total suspended solids, and oxygen uptake rate were measured. Two 5 mL samples were taken every day from each reactor and total organic carbon (TOC) and methylene blue active substances (MBAS) were measured. Oxygen uptake rate (OUR) was measured at regular intervals. Total suspended solids (TSS) concentrations were measured in duplicate at the end of the reactor cycles.

R2 and R3 were intended to be an SBR but due to the poor settling quality of the biomass the process of decanting was ceased. The proper amount of nutrients and LAS was added every five days to maintain a concentration of 400 mg/L. Mixing of biomass from reactors R1 and R2 was carried out in an attempt to improve the sludge's settling characteristics and decrease time needed for degradation of LAS. Before proceeding to mix the reactors, they were left to settle for two hours at the end of their operation cycle and 1.875 L was pumped out of each one.

The remaining 0.625 L in reactor R1 were transferred to reactor R2 to get 1.25 L. The reactor (named R1-NEW) was then filled with 1.25 L of synthetic wastewater to a total volume

Table 3.4. R1, R2, and R3 Reactor operation.

Reactor	Testing period.	Reactor Inoculum	Initial TSS concentration (mg/L)	Surfactant type and concentration	SBR cycle times (days)
R1	Sept. 30 – Oct. 20, 2002 4 Cycles 5 days each	Biomass collected from experiment III.	1000	SDS 400 mg/L	0.002 (A) 4.96 (B) 0.044 (C+D)
	October. 21, 2002 – Jan.11, 2003 8 Cycles 10 days each	Biomass collected from experiment III	1100	LAS 400 mg/L	0.002 (A) 9.96 (B) 0.044 (C+D)
R2	November 23, 2002 – Jan. 11, 2003 10 Cycles 5 days each	Biomass collected from experiment VI	1000	LAS 400 mg/L	
R1-NEW	Jan. 11 - February 20, 2003 8 Cycles 5 days each	Biomass collected from the mixture of R1 and R2	1000	LAS 400 mg/L	0.002 (A) 4.96 (B) 0.044 (C+D)
R3	Jan. 20 - February 20, 2003 6 Cycles 5 days each	Biomass collected from experiment IX	1000	LAS 400 mg/L	

of 2.5 L and a new operation cycle was initiated. The R1-NEW SBR was operated for 8 cycles, each having 5 days. Then, R3 was mixed with R1-NEW following a procedure similar to that described above; however, only 0.625 L was decanted from each reactor prior to mixing the remaining liquid volume (a total of 3.75 L) in a 7 L plastic container. From this, 1.25 L was transferred to each of three new reactors (arbitrarily named K1, K2, and K3) that were operated as described in the following sections. Then, reactor volumes were filled to 2.5 L with synthetic wastewater and operated as sequencing batch reactors; K1 and K2 for 60 days completing 12

cycles of 5 days each, and K3 for 15 days (3 cycles of 5 days each). Upon completion, K1 was continued as an SBR while K2 was transformed into an Intermittent Cycle Extension Aeration System. K3 was set as a Sequencing Batch Biofilm Reactor with the introduction of polyurethane foam cubes (Zander, Germany).

3.4 Comparison of Bioreactor Operating Strategies for LAS Biodegradation and Foam Production: SBR, ICEAS, and SBBR

3.4.1 Reactor Set-up

Once the enrichment cultures were developed using SBR operation as described in Section 3.3, K1, K2, and K3, were operated using different control strategies. Reactor K1 was operated as an SBR, reactor K2 was operated as an Intermittent Cycle Extension Aeration System (ICEAS), and reactor K3 was operated as a Sequencing Batch Biofilm Reactor (SBBR).

3.4.1.1 K1

The SBR operation for K1 is the same as that described in Section 3.3. The cycle length of 5 days, a hydraulic retention time of 10 days, and having a 400 mg/L LAS synthetic wastewater as a feed. The reactor was filled to 2.5 L and decanted to 1.25 L of liquid volume (i.e., the fill volume was one half of the total volume). The hydraulic retention time (HRT), is defined as follows (Metcalf and Eddy, 1991).

$$HRT = V/Q \quad (3.1)$$

Where:

V = volume of the reactor (L)

Q = influent flowrate (L/d , 1.25/5)

Operation times for fill, react, solid settling; and decanting and discharge stages were 0.002, 4.96, 0.042 and 0.002 days, respectively. No intentional biomass wasting was performed during K1 operation other than sampling conducted for analysis of TSS. This decision was made

to increase the TSS concentration. Aeration was provided for 30 seconds every 10 minutes as described in section 3.3.

3.4.1.2 K2

K2 was operated as a sequencing batch reactor for 30 days before its transition to become an ICEAS. Once the reactor was started to operate as an ICEAS, the system had three distinct phases in each operating cycle: fill/react, settle; and draw. Even when the system was in the settling and draw phases, synthetic wastewater was continuously flowing into the bottom of the reactor. The operation times were 4.956, 0.042, and 0.002 days for fill/react, settling, and draw, respectively. Like the SBR, the ICEAS was filled to 2.5 L and decanted to 1.25 L of liquid volume with an HRT of 10 days (i.e., the cycle length was 5 days).

During the operation period (June 28 – October 22, 2003), the reactor was fed continuously using a Cole Palmer Materflex® L/S digital standard drive peristaltic pump (model 7523-40, Bernant Co, Barrington; IL) which pumped synthetic wastewater containing 400 mg/L of LAS from a 1 L graduated glass reservoir. The influent was introduced at a flow rate of 0.175 mL/min at the bottom to the reactor by 1/16” Masterflex® tubing (Cole Palmer Cat. No.6412-14). During the draw period, the treated wastewater was removed from the top of the liquid volume through a 2-mL plastic pipette attached to a Masterflex® tubing, and a peristaltic pump. The pipette was manipulated in a way that the tip was always at the surface of the liquid as the level decreased. This procedure was utilized to minimize withdrawal of settled sludge and minimize short-circuiting (i.e., removal of untreated influent wastewater), and mimicked performance of a floating decanter in a full-scale system.

Intermittent aeration was employed during the first 30 days of operation, controlled by a programmable microprocessor as described in section 3.3. During subsequent operation, the

system was aerated continuously. During the period when the system underwent continuous aeration, a peristaltic pump was used to deliver 100 mL/min of air during the fill/react period. The air flow rate was monitored by a rotameter. The reactor was placed on a magnetic stir plate to allow mixing using a Teflon coated stir bar. A schematic diagram for the ICEAS cycle is presented in Figure 3.2.

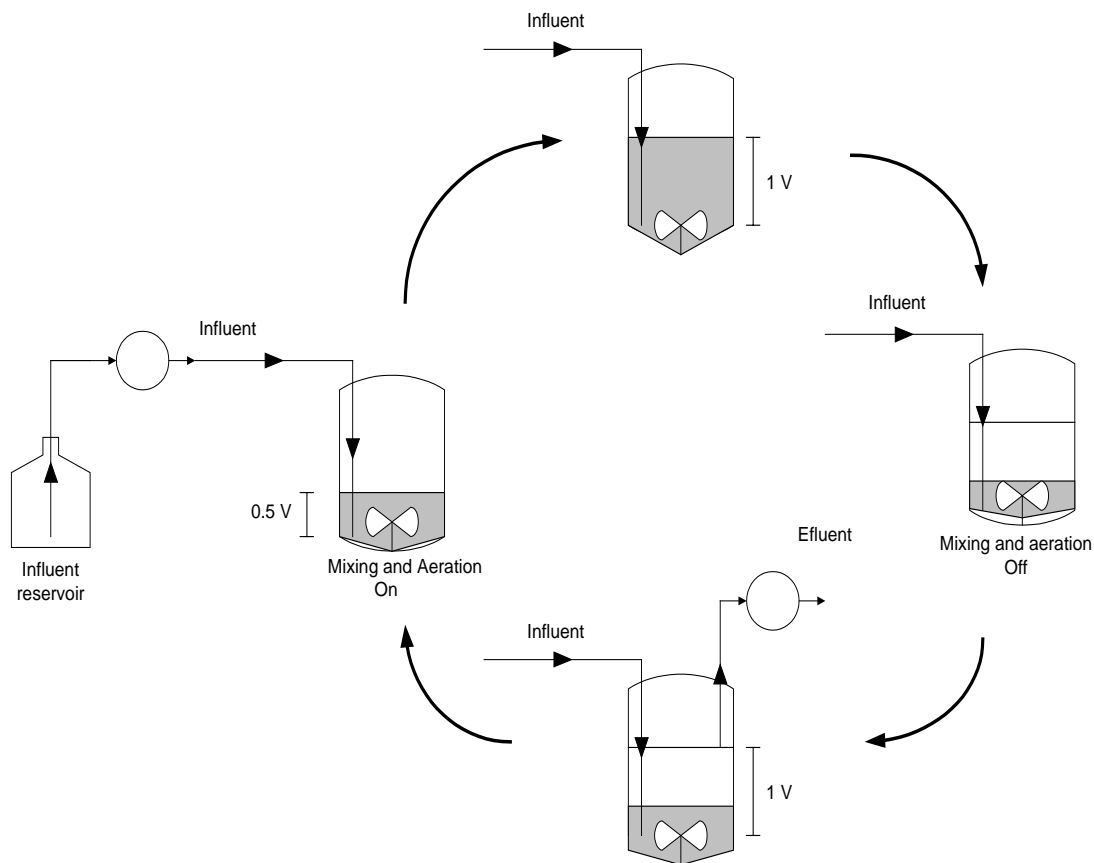


Figure 3.2. ICEAS laboratory scale schematic cycle diagram.

A solution containing sodium hydroxide (10 M) was added as necessary to control pH in the K2 reactor. Using a plastic transfer pipette, the sodium hydroxide solution was added drop wise until a steady pH value of 7.5 was achieved as monitored by an Orion pH-meter (Orion model 720A) equipped with an epoxy body combination electrode (Thermo-Orion, Fisher Cat. No.13-641-259). Regulation of pH was performed manually every 6 hours during the React phase.

3.4.1.3 K3

To operate K3 as an SBBR, 12 g of 1.5-cm per side polyurethane foam cubes (Zander, Germany) were introduced into the reactor. The foam cubes were held in a wire cage submerged into the liquid volume of 2.5 L. Foam-free spaces of 2.0 cm were left between the cage and the surface of the liquid, and 4 cm between the cage and the bottom of the reactor.

Influent and treated wastewaters were fed and drawn at the bottom of the reactor using a 2 mL plastic pipette attached to a Masterflex® tubing, and a Cole Palmer Materflex® console drive peristaltic pump (model 7521-40, Bernant Co, Barrington; IL). To perform these activities, the pipette was carefully inserted through the foam media in one of the sidewalls of the reactor until the tip reached the bottom. Aeration was controlled to avoid excessive production of foam during the reactor's operation. Like the ICEAS, the SBBR needed constant pH regulation and the same procedure was performed to maintain the pH between 6.5 and 7.5 during the React period.

The SBBR operative cycle consisted of three phases: (1) reactor filling, (2) reaction, and (3) draw. After discharging the treated water, the sequence was repeated. The reactor was filled to 2.5 L with synthetic wastewater containing 400 mg/L of LAS, and decanted to 1.25 L of liquid volume using a Cole Palmer Materflex® console drive peristaltic pump (model 7521-40, Bernant Co, Barrington; IL). Reactor's fill and draw phases were 0.002 days each (2.88 minutes), while three different reaction times 4.99, 2.99, and 1.99 days were tested. Aeration was provided for 30 seconds every 10 minutes as described in Section 3.3.

3.4.2 LAS Removal

The removal efficiency associated with each operating cycle was calculated using the mass of each LAS measured as TOC entering the reactor, and the mass of LAS at the end of the SETTLE period.

$$\text{Percent Removal} = \frac{C_{in} - C_{out}}{C_{in}} \times 100 \quad (3.2)$$

Where:

C_{in} = concentration of TOC of MBAS entering the reactor (mg/L).

C_{out} = concentration of TOC of MBAS at the end of the settling cycle (mg).

To compare the LAS removal between the reactors, two 5 mL samples were taken every day during the react phase from each reactor. A 5-mL plastic syringe was used to filter samples through a syringe filter 25 mm diameter and 0.45 μm pore size (Whatman Cat. No.6874-2504). Filtered samples were collected in 5 mL glass vials and sealed with paraffilm, and total organic carbon (TOC) and methylene blue active substances (MBAS) were measured. TOC was measured using a TOC analyzer model 5050A from Shimadzu, and MBAS was carried on following Standard Methods for the Examination of Water and Wastewater (APHA 1998).

3.4.3 Foam Production

Foam height was measured in each of the reactors during their cycle operation. Readings were averaged from 5 cycles in the SBR and ICEAS, and from 8 cycles in the SBBR. Using a ruler, the foam was measured from the surface of the liquid. In the SBR and ICEAS, recordings were made every 30 minutes during the first three hours, then every hour during the next 4 hours, and finally every 12 hours until the end of the react period. For the SBBR, foam height was measured and recorded at 30 minutes intervals during the first 4 hours, 90 minute intervals in the next 6 hours, and then every 12 hours until completion of the cycle. Aeration conditions were similar for the SBR and SBBR. The solenoid valve opened 30 seconds every 10 minutes and air flowed at a rate of 500 mL/min through the aeration stones. Aeration in the ICEAS was continuous at a flow rate of 100 mL/min.

3.4.4 Sorption of LAS on Sludge and Polyurethane Foam – Reactor Scale

Simple experiments were conducted to determine the sorption characteristics of LAS in the activated sludge and the polyurethane foam under normal operation conditions in the reactors. The mass of LAS sorbed was estimated by performing a mass balance on TOC using equation 3.3 below.

$$TOC\ sorbed = [C_r V_{r-meas} + C_{in} V_{in}] - C_{final} (V_r + V_{in}) \quad (3.3)$$

Where: V_{r-meas} = measured volume of liquid remaining in the decanted reactor at end of previous cycle (L); V_{in} = measured volume of liquid entering the reactor during the FILL period (L); C_r = measured concentration of soluble TOC remaining in the reactor at the end of the previous cycle (mg/L); C_{in} = measured concentration of TOC in the influent wastewater (mg/L); and, C_{final} = measured concentration of TOC in the reactor after the end of the FILL period (mg/L).

After filling the reactors, samples were collected for measurement of TOC and MBAS to perform the LAS mass balance and measure sorption under the assumption that the fill period was sufficiently short so that no biodegradation occurred. The number of sorption tests performed in each of the reactors is summarized in Table 3.5.

Table 3.5. Amount of LAS degradation, foam production, and sorption tests performed in reactors.

	LAS removal	Foaming	Sorption
SBR	26	5	7
ICEAS	22	5	***
SBBR	66	8	16

These experiments were done at the end of the operation cycles, after the draw phase using the remaining liquid volume in the reactors. 1.28 L of mixture of settled sludge and treated wastewater were measured using two glass cylinders (1000 mL and 500 mL). Two samples of 10 mL were taken (with a 10-mL plastic pipette) to measure total suspended solids, and other two samples of 5 mL for TOC and MBAS. The remaining 1.25 L was returned to the reactor. The same analyses were performed to 1.28 L of influent synthetic wastewater prior mixing.

3.4.5 Special Tests

3.4.5.1 Variation in Hydraulic Retention Time (HRT) and the Effect on LAS Degradation and Foam Production

LAS biodegradation and foam production were measured at values of hydraulic retention times of 6.25, and 12.4 days. The length of the operation cycle was left to be 5 days for the SBR and ICEAS, and 2 days for SBBR. With an HRT of 6.25 days, the volume decanted during the draw period was 2.0 L, and 1.0 L for a hydraulic retention time of 12.4 days. Then the reactor was filled to 2.5 L. Two samples of 5 mL were taken every day from each reactor to measure TOC and MBAS.

Foam height in the reactors was measured following the same procedure performed during the 10 days HRT.

3.4.5.2 ICEAS - Transient Loading Experiments

Three transient loading experiments were performed in the ICEAS to assess reactor response to higher influent LAS concentrations. The reactor was subjected to a loading condition during which the influent LAS concentration was increased to two times that of the normal. During the first two experiments the system was fed with synthetic wastewater containing 800 mg/L of LAS (513 mg/L as TOC) during two 5-day cycles. The influent was pumped in at a flow rate of 0.175 mL/min beginning at SETTLE and DRAW periods of the previous cycle.

A different approach was taken during the third transient loading experiment. Influent wastewater containing the higher LAS concentration was started to entering the system during the second day of the cycle. The feed continued until completion of the 5-day cycle.

During the transient loading experiments conducted in the ICEAS, airflow was set to 100 mL/min, and foam height, OUR, TOC, and MBAS were measured. Two samples of 5 mL were taken every day for analysis, and foam height readings were recorded 10 minutes during the first two hours of the react phase, and then every six hours until the completion of the cycle.

3.4.5.3 Testing Anti-foaming Agents in Its Addition to SBR and SBBR

Twelve anti-foaming agents (laboratory preparations from Vulcan Performance Chemicals, Columbus, GA) were tested to be use in the SBR and SBBR systems. Different concentrations of each of the anti-foaming agents ranging from 30 to 2000 mg/L were added to synthetic wastewater containing 200 mg/L of LAS (113 mg/L as TOC). The principal characteristics of the tested anti-foaming agents are presented in Table 3.6.

The testing procedure consisted of placing 100 mL of the wastewater containing the anti-foaming agent in a 1000-mL glass cylinder. The cylinder was graduated in centimeter scale using a ruler and masking tape. A peristaltic pump (Cole Palmer Materflex® console drive model 7521-40, Bernant Co, Barrington; IL) was used to provide a continuous flow of air at a flow rate of 100 mL/min as measured by a Cole-Palmer Rotameter (Gilmont Instruments, 250 mL/min scale Accucal flow meter Cole-Palmer Instruments Co., Vernon Hills, IL).

Air flow was delivered continuously through an aeration stone (Fisher Scientific, Sewanee, GA) connected to ¼” Tygon tubing (Cole Palmer Cat. No. A-06408-47). Measurement of the resulting height of foam in the graduated cylinder began as soon the stone reached the bottom of the cylinder, and readings were recorded at 30 second intervals for duration of 10 minutes. Results from the anti-foaming agent test evaluations are shown in Appendix 1.

Table 3.6. Antifoaming agents evaluated in this study.

Anti-foaming agent	Color / Appearance	Solubility in water
3112A	Gold-brown oil like liquid	Completely soluble
3112	Bright yellow oil like liquid	Completely soluble
3131A	Light brown oil like liquid	Only with surfactant present
3132A	Light brown oil like liquid	Completely soluble
3126	White thick liquid	Only with surfactant present
3249	Light gold-yellow oil like	Completely soluble
3249A	White thick liquid	Completely soluble
3102A	White thick liquid	Completely soluble
3377	Light white thick liquid	Completely soluble
3379	White thick liquid	Partially, but soluble when surfactant is present.
3142	Light brown pearled liquid	Completely soluble
E-10	White thick liquid	Partially, but soluble when surfactant is present

Of the twelve anti-foaming agents tested, Callaway 3142 was selected to decrease foaming on the SBR and SBBR systems. The anti-foaming agent was added to the influent synthetic wastewater. Two concentrations of anti-foam agent Callaway 3142 were tested, 25 mg/L and 50 mg/L. The SBR operated under a cycle length of 5 days and HRT of 10 days. The SBBR had a cycle length of two days and the same HRT of 10 days.

Foam height was measured and recorded at 10-minute intervals during the first two hours of the REACT period, and then at six hour intervals during the remainder of the REACT period. Also, two 5-mL samples were taken daily for TOC and MBAS, and OUR was measured in duplicate using 5-mL sample volumes.

3.4.5.4 Batch Sorption Experiments

Experiments were conducted to measure the sorption characteristics of the biomass from reactors K1 and K2, and the polyurethane foam used as attaching media in K3. K1 and K2 were let to settle and biomass was removed from the bottom using a 10 mL plastic pipette. Once the collection was completed, samples were submitted to continuous washing and centrifuging to remove LAS. This process was performed as follows: 35-mL plastic centrifuge tubes were filled with biomass samples and submitted to 10,000 rpm for 10 minutes. After centrifuging, supernatant was discharged and deionized water was added to the 35 mL mark. Tubes were shaken manually for 30 seconds and centrifuged again. This process was repeated 20 times. Between washes 15 to 20, TOC was measured in the supernatant to assess the effectiveness of the washing. Biomass samples were collected from every tube and mixed into two beakers, one for biomass from K1 and the other for K2.

Different amounts of biomass were placed in separate 120 mL narrow mouth amber glass bottles (I-Chem, New castle, DE). Three LAS solutions differing in concentration were prepared to fill the bottles (50, 400, and 1000 mg/L). The LAS solutions were amended with 1000 mg/L of NaN_3 . Three bottles were filled with 50 mg/L LAS solution, another three with 400 mg/L, and another with 1000 mg/L. Also, blanks for each of the solution were prepared containing only the solution itself. Then bottles were covered with Teflon caps and placed in a rotary tumbler for 36 hours. To measure the moisture content of the biomass placed into the bottles, two 0.5 g

samples of biomass from each reactor were dried at 105°C for one hour. Amounts of biomass placed into bottles and the average moisture content are showed in Table 3.7.

Table 3.7: Amounts of biomass from K1 and K2 used on batch sorption test.

Working LAS solution	K1			K2		
	mass (mg)	moisture,%	dry mass (mg)	mass (mg)	moisture,%	dry mass (m, mg)
50 mg/L	515.7	95.3	24.13	1.1721	94.54	64.00
	1231.4		57.63	1.448		79.06
	1387.9		64.95	1.6591		90.59
500 mg/L	477.8	95.3	22.36	1.448	94.54	79.06
	1145.6		53.61	1.4558		79.49
	1448.4		67.79	2.8646		156.41
1000 mg/L	651.8	95.3	30.50	1.2181	94.54	66.51
	1313		61.45	1.7185		93.83
	1216.8		56.95	1.8542		101.24

After the 36-hour equilibration period, aqueous samples were removed, filtered through 0.45 µm PTFE syringe filters, and analyzed for TOC to determine the equilibrium concentration of LAS measured as total organic carbon. The sorption characteristics of the biomass samples were modeled using Freundlich isotherms. The empirically derived Freundlich isotherm equation is defined as follows (Metcalf and Eddy, 1991).

$$x/m = K_f C_e^{1/n} \quad (3.4)$$

Where;

x/m = mass of adsorbate adsorbed per unit mass of adsorbent (mg/mg)

C_e = equilibrium concentration of adsorbate in solution after adsorption (mg/L)

K_f and n = empirical constants.

The same procedure described above was used for measuring sorption of LAS to the polyurethane foam used as a solid support in the K3 reactor. Different amounts of dry foam were placed into the glass bottles instead of biomass. Two solutions of LAS were prepared for this test, 50 and 400 mg/L. Three bottles for the 50mg/L solution and 11 for 400 mg/L were prepared.

3.4.5.5 Sludge Volume Index (SVI)

The settling capacity of the activated sludges from the SBR and ICEAS were measured in terms of the sludge volume index (SVI) as described in Section 3.5.5. The SVI was determined from each reactor at the end of the React period during three consecutive operation cycles.

3.4.5.6 Chemical Oxygen Demand (COD) Balance in the SBR and ICEAS

COD measurements were performed during 1 cycle on K1 and K2 to verify the overall performance of the reactors. COD analysis was performed in samples collected every 24 hours during the REACT period on each reactor. Also, COD was measured in the influent. OUR was measured every 6 hours. The TSS from every reactor was measured at the end of the REACT cycle.

3.5 Analytical Techniques

3.5.1 Total Organic Carbon (TOC)

Samples collected for measurement of total organic carbon (TOC) were analyzed the same day they were collected. Samples were collected using a 5-mL plastic syringe to withdraw samples from the reactor, and then the samples were filtered through a syringe filter 25 mm diameter and 0.45 μm pore size (Whatman Cat. No.6874-2504). Filtered samples were collected in 5 mL glass vials and sealed with parafilm prior to analysis.

TOC measurements were performed using a Shimadzu Scientific Instruments Inc. model 5050 TOC analyzer (Japan). The equipment was calibrated following standard method for water and wastewater 5310B, two stock solutions were prepared following the same method, one for organic carbon (1.0 mL = 1.0 mg carbon), and the second for inorganic carbon (1.0 mL = 1.0 mg carbon).

One calibration curve was prepared for total carbon (TC) covering a range of 0 – 400 mg/L. Standard solutions of 0, 50, 100, 200, and 400 mg/L were prepared pipetting the

appropriate volumes of organic carbon stock solution into 100 mL volumetric flasks and then filling with deionized water to create the various dilutions. The inorganic carbon (IC) calibration curve covered a range of 0 – 200 mg/L. Standard solutions of 0, 25, 50, 100, and 200 mg/L were prepared by pipetting the appropriate volumes of inorganic stock solution into 100 mL volumetric flasks and then filling with deionized water to create the various dilutions. The values of each point in the standard curves corresponded to the average of three injections, and the least squares option in the 5050 TOC analyzer was selected for linear fit analysis.

The 5050 TOC analyzer was equipped with a 250 μ L syringe, set to have a standard injection speed, and perform four washes before sampling for measurement. The injection volumes were set to 40 and 16 μ L for total carbon and inorganic carbon analysis respectively. Reported concentrations of total carbon and inorganic carbon obtained from the analysis corresponded to the average of two injections from the same sample. Finally, the total organic carbon value was calculated subtracting the inorganic carbon from the total carbon values ($TOC = TC - IC$). Calibration curves were checked at least once per month using standard solutions prepared from fresh stock solutions.

3.5.2 Methylene Blue Active Substances (MBAS)

Lauryl Alkylbenzene Sulfonate (LAS) was measured directly by methylene blue active substances analysis (Standard Methods for the Examination of Water and Wastewater 5540C, APHA, 1998). Samples were filtered through a syringe filter 25 mm diameter and 0.45 μ m pore size (Whatman Cat. No.6874-2504). MBAS analysis was performed in duplicate the same day samples were collected or else samples were acidified and refrigerated until analysis.

A calibration curve covering a range from 0 to 1000 mg/L was constructed using Sodium Dodecylsulfate (SDS, 99%, Aldrich 43,614-3). A sample size of 1 mL was used to perform the

analysis following method 5540C. Absorbance was measured at a wavelength of 652 nm using a UV-VIS Spectrophotometer (model UV-1201, Shimadzu Scientific Instruments, Inc., Japan) equipped with 10 mm rectangular UV quartz cells (Hach Cat. No. 48228-00, Hach Co., Loveland, CO).

3.5.3 Oxygen Uptake Rate (OUR)

For respirometer experiments, the oxygen uptake rate (OUR) was measured directly by the model OO-244SC Comput-OX Respirometer (N-CON Systems, Crawford, GA). For other experiments, OUR was measured using a YSI model 5300 Biological Oxygen Monitor (Yellow Springs, OH). OUR measurements were conducted in duplicate using a 5 mL sample volume, and dissolved oxygen concentrations were recorded every 30 seconds over a five minute period. OUR values were calculated from a linear regression analysis of the dissolved oxygen concentrations over time.

3.5.4 Total Suspended Solids (TSS)

Total Suspended Solids (TSS) concentrations were measured in duplicate following standard method 2540C (APHA, 1998).

3.5.5 Chemical Oxygen Demand (COD)

Chemical Oxygen Demand was measured following the USEPA approved reactor digestion method for reporting wastewater analysis (HACH method 8000, 0-1500 mg/L range). This method is an adaptation of Standard Methods for the Examination of Water and Wastewater 5220D, APHA, 1998. Samples were filtered through a syringe filter 25 mm diameter and 0.45 μm pore size (Whatman Cat. No.6874-2504). From this, 1.0 mL was introduced into the COD vial followed by 1.0 mL of deionized water to complete a total volume sample of 2.0 mL. Then were put into the COD reactor and heated for two hours at 150°C. COD concentrations were measured

in HACH spectrophotometer DR/4000 using the 435 COD High Range program. COD concentrations in samples were then calculated using the appropriate dilution factor.

3.5.6 Sludge Volume Index (SVI)

The settling capacity of the activated sludges from the SBR and ICEAS were measured using the Sludge Volume Index. A variation of Standard Method for the Examination of Water and Wastewater 2710D (APHA 1998) was used to perform the analysis. An Imhoff settling cone graduated from 0 to 1000 mL (Nalgene, Fisher Cat. No. 15-438) was used instead of the settling vessel described in the method. Duplicate 10 mL samples were analyzed for total suspended solids to complete the test.

3.5.7 Ammonia, Nitrate, and Nitrite Analysis

During the first operation cycles of K3 (cycles 3-8), ammonia, nitrite and nitrite analysis were performed to determine if nitrification was taking place in the reactor. Samples were taken at the beginning and end of the React cycle and diluted as necessary for the ammonia, nitrite and nitrite to fall into the range of the analytical kit used. HACH tests kits (HACH Company , Loveland CO) were used to measure $\text{NH}_3\text{-N}$, $\text{NO}_3\text{-N}$, and $\text{NO}_2\text{-N}$ in the samples taken during the operation of the reactor. Ammonia was measured using HACH method 8038 (EPA approved Nessler method). Nitrate was measured using HACH method 8039 (NitraVer5 Nitrate AccuVac, high range), and nitrite by HACH method 8192 (NitriVer3 Nitrite reagent AccuVac, low range). Samples were analyzed following procedures specified in every kit's booklet.

CHAPTER FOUR RESULTS AND DISCUSSION

4.1 Microbial Population Development

4.1.1 Respirometer Results Using Sodium Dodecyl Sulfate (SDS) as Carbon Source

During Experiment I, the respirometer bottles were seeded with activated sludge from a municipal wastewater treatment facility, and various concentrations of sodium dodecyl sulfonate (SDS) were provided as a carbon source. The initial SDS concentrations ranged from 30 to 300 mg/L SDS (corresponding to initial TOC concentrations ranging from 12.9 to 133 mg/L). Respirometer data showed a lag phase of 35 hours before any measurable oxygen uptake occurred. After that period of time, respirometry activity increased, reaching highest oxygen uptake rates between days 3 and 6 as depicted in Figure 4.1 (top). Oxygen uptake then continued but at a lower rate until the end of the test period (a total of 360 hours).

As depicted in Figure 4.1 (bottom), the TOC concentration decreased by an average of 37% (average of all eight respirometer reactors) during the first 24 hours. The lack of oxygen consumption during this period of time when soluble TOC decreased suggests that a portion of the SDS added to each respirometer reactor may have been removed from solution via sorption to the activated sludge. Initial and final TOC concentrations for Experiment I are presented in Table 4.2. The initial TOC concentrations listed in the table are values measured using samples collected approximately one minute after surfactant was added and the sample was mixed. In the same table, the total mass of oxygen consumption is presented, calculated from data obtained from the respirometer. The oxygen uptake in a control respirometer reactor, prepared exactly like the other bottles but without surfactant addition, was subtracted from oxygen readings for each of the bottles containing surfactant. At the end of the experiment, TOC removal was 84.4%, 75.2%, 70.6%, and 46.0% for samples initially containing 30, 60, 150, and 300 mg/L SDS.

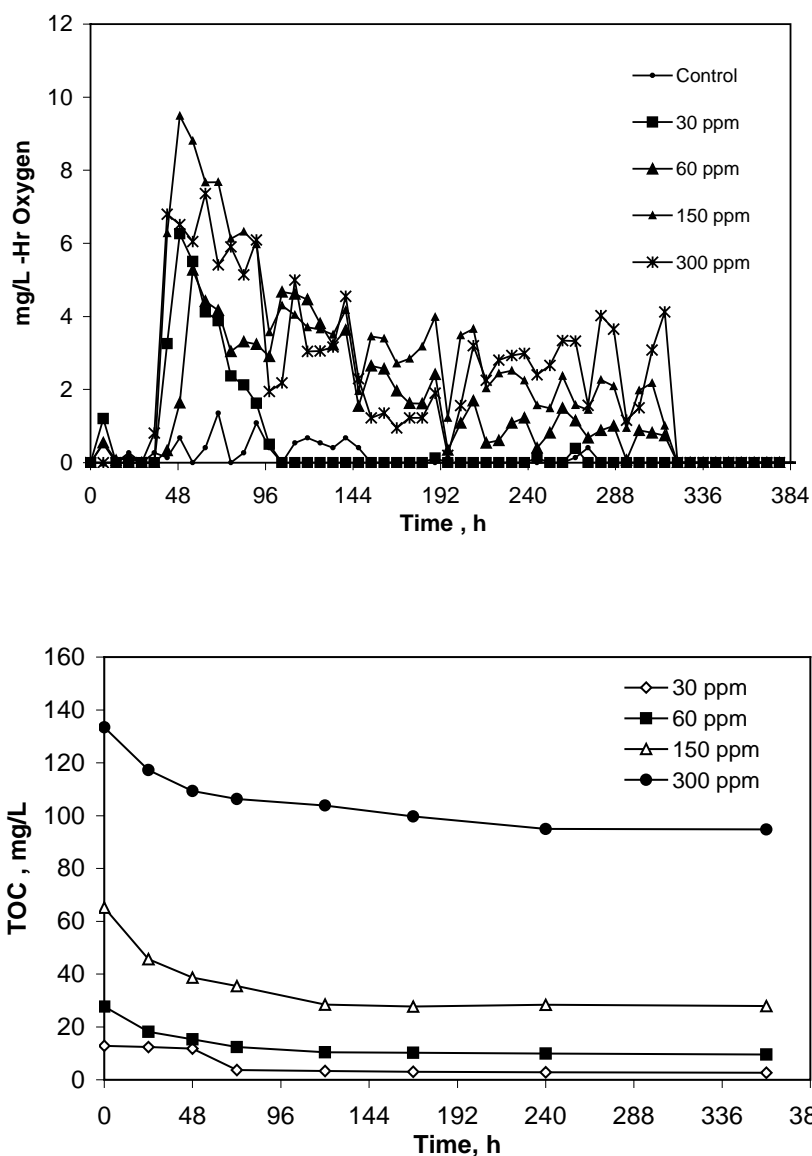


Figure 4.1: Oxygen uptake rates (top) and TOC concentrations (bottom) as a function of time for Experiment I using SDS. For SDS concentrations for which there were duplicate respirometer bottles, the data depicted are the average.

Based on the TOC concentrations measured as a function of time, the first order kinetic rate constant, k , was calculated for each of the respirometer bottles. Data for each individual bottle can be found in Appendix 2, and average parameter values considering all surfactant-containing samples from Experiment I are summarized in Table 4.1. As shown in the table, the average first order rate constant, k (hr^{-1}), for Experiment I was 0.0023 hr^{-1} . This corresponds to

a half-life of 438 hours. R^2 values for the first order model ranged from 0.578 to 0.716 and averaged 0.6554.

Biomass recovered at the end of Experiment I was used as the inoculum for Experiment II. In Experiment II, the initial SDS concentrations in the respirometer bottles was higher than for Experiment I, ranging from 300 to 600 mg/L SDS (corresponding to initial TOC concentrations ranging from 129 to 268 mg/L). As depicted in Figure 4.2, during Experiments II, there was a markedly shorter lag period, higher OUR, and higher TOC removal than was observed in Experiment I. As shown in the figure, the highest OURs occurred between days 0 and 5 (0 – 120 hours).

Overall TOC removal averaged 85.2% in Experiment II, where 74.0% of it was consumed by the fourth day. Adaptation and enrichment of a microbial community capable of using SDS as a carbon source could be the cause why results from trial to trial improved. Individual values for TOC removal and oxygen consumption for Experiment II are presented in Table 4.2. Data for each individual bottle can be found in Appendix 2, and average parameter values considering all surfactant-containing samples from Experiment II are summarized in Table 4.1. As shown in the table, the average first order rate constant, k (hr^{-1}), for Experiment II was 0.0046 hr^{-1} , twice that of Experiment I. This corresponds to a half-life of 154 hours. R^2 values for the first order model ranged from 0.584 to 0.940 and averaged 0.814.

Table 4.1. First order rate constants for SDS degradation in Experiments I, II, and III.

Experiment No	k (h^{-1}) average \pm std dev	Half Life (h) average \pm std dev	R^2 Range	R^2 Average
I	0.0023 ± 0.00133	438.28 ± 299.39	0.5783 – 0.7162	0.6554
II	0.0046 ± 0.00061	153.74 ± 22.92	0.5845 – 0.9400	0.8138
III	0.0137 ± 0.00366	54.36 ± 16.18	0.7485 – 0.8783	0.8003

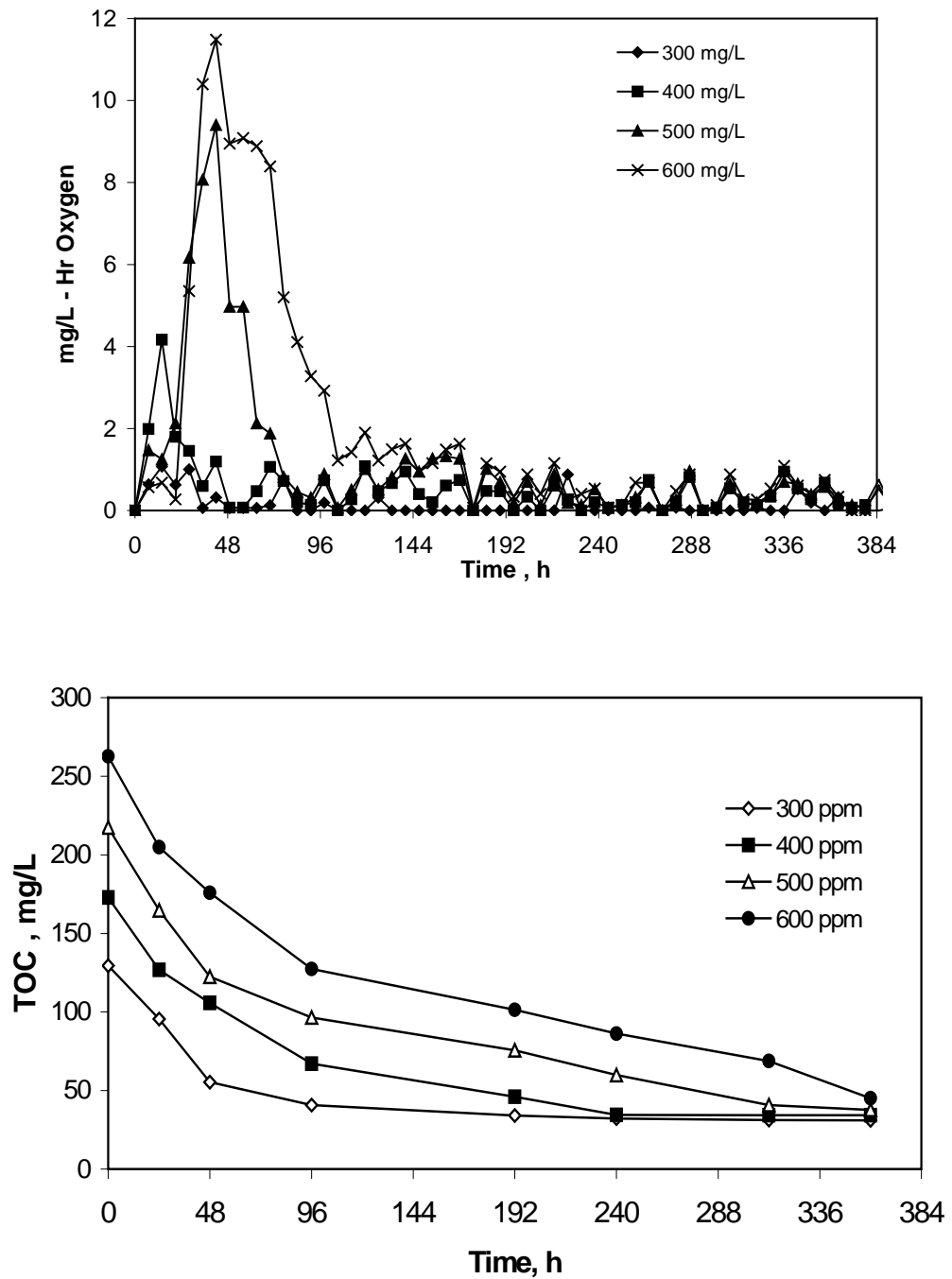


Figure 4.2: Oxygen uptake rates (top) and TOC concentrations (bottom) as a function of time for Experiment II using SDS.

Biomass recovered from respirometer bottles at the end of Experiment II was used as the inoculum for Experiment III. In Experiment III, the initial SDS concentrations in the

respirometer bottles were higher than for Experiments I and II, ranging from 100 to 1000 mg/L SDS (corresponding to initial TOC concentrations ranging from 44.2 to 428.6 mg/L). Oxygen uptake rates and TOC concentrations as a function of time during Experiment III are shown in Figure 4.3.

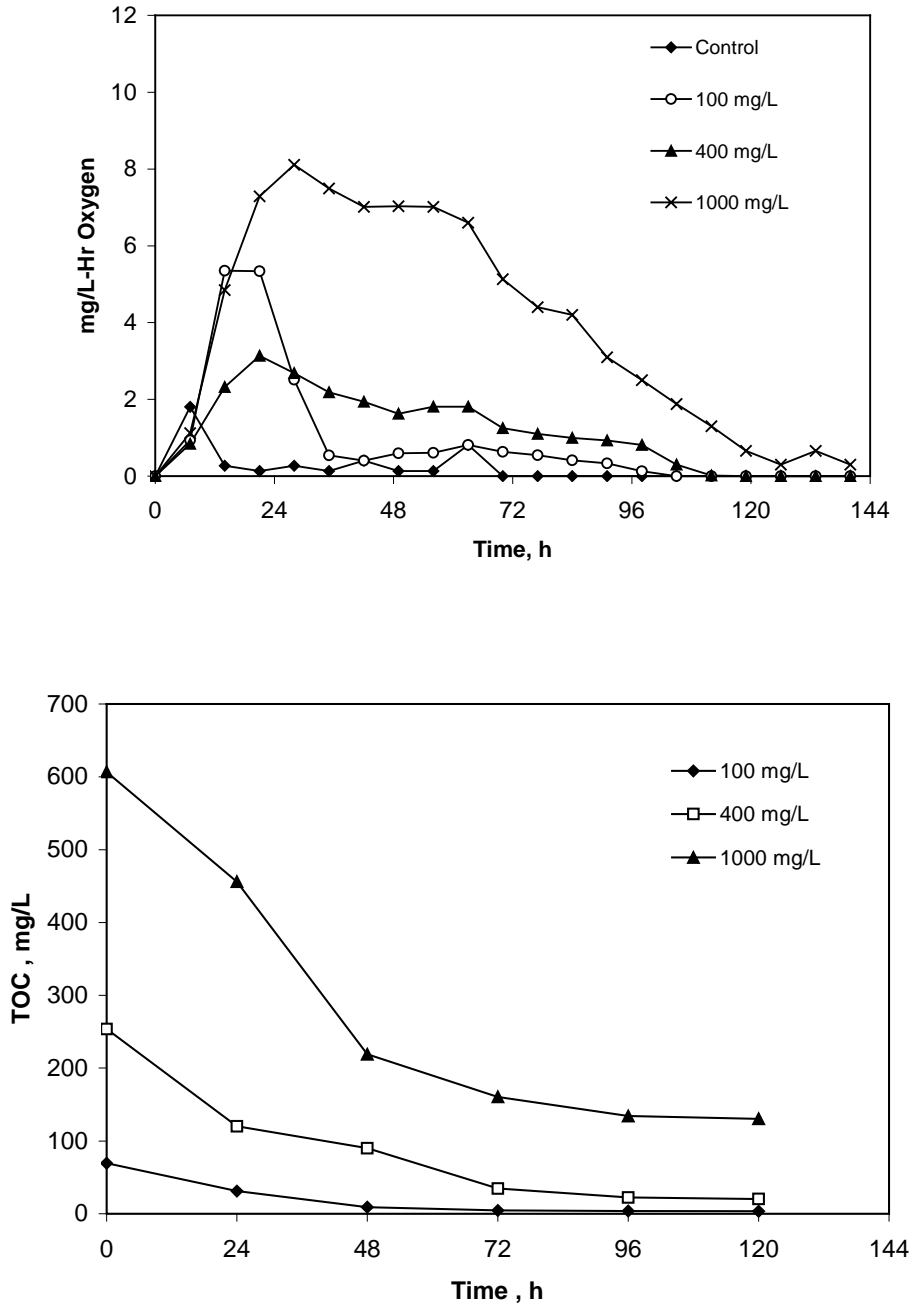


Figure 4.3: Oxygen uptake rates (top) and TOC concentrations (bottom) as a function of time during Experiments III using SDS.

The high concentrations of SDS used in Experiment III (e.g., 1000 mg/L) did not take more time to be consumed or show a different shape in the oxygen uptake rate data plot compared to lower concentrations. This suggests that microorganisms were not affected, and high concentrations of the surfactant did not produce a toxic effect. Furthermore, in Experiment III, TOC removal averaged 93.6% at the end of the experiment (a total of 480 hr), with 95.4% of the TOC removal occurring by the end of the third day (72 hours).

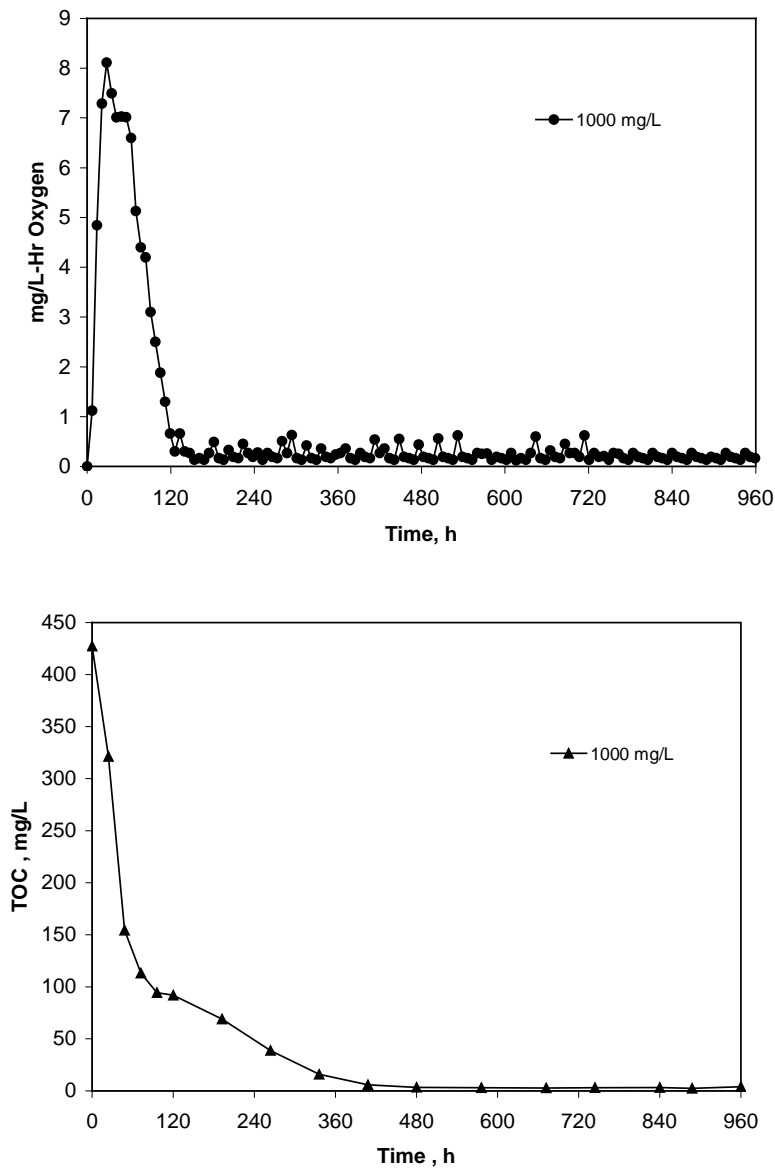


Figure 4.4: Results for 1000 mg/L SDS sample monitored for 40 days. Oxygen uptake rate (top), and TOC consumption (bottom).

One of the Experiment III respirometer reactors, with an initial SDS concentration of 1000 mg/L, was monitored for a total of 40 days. As shown in Figure 4.4 and Table 4.2, essentially complete TOC removal was achieved (the final TOC concentration was 3.38 mg/L). This indicates that the microbial population was capable of mineralizing the SDS.

As with data from Experiments I and II, the first order kinetic rate constant, k , was calculated for each of the respirometer bottles based on the TOC concentrations measured as a function of time. Data for each individual bottle can be found in Appendix 2, and average parameter values considering all surfactant-containing samples from Experiment III are summarized in Table 4.1. As shown in the table, the average first order rate constant, k (hr^{-1}), for Experiment III was 0.0137 hr^{-1} . This corresponds to a half-life of 54.4 hours. R^2 values for the first order model ranged from 0.749 to 0.878 and averaged 0.800.

The average first order rate constant and corresponding half-life, calculated as the average of all respirometer bottles within each of the three experiments (i.e., I, II, and III) are summarized in Table 4.1. As also shown in the table, average R^2 values for the first order model were 0.655, 0.814, and 0.800 in Experiments I, II, and III, respectively. The relatively low R^2 values indicate that a first order model does not perfectly fit the experimental data. This was not particularly surprising in this case, given the fact that this simple model does not account for differences in biomass concentration as a function of time within the various respirometer bottles. Nevertheless, because biomass concentrations were not measured as a function of time in the experiments described herein (and therefore data are not available to allow a more sophisticated modeling approach that includes changes in biomass), the first order model was used as a basis of comparison. The increase in k values provide evidence that the microbial population was enriched for microorganisms capable of SDS over time.

Table 4.2: Total organic carbon (TOC) removal and oxygen consumption in Experiments I, II, and III using sodium dodecylsulfate (SDS).

Experiment No.	Concentration as SDS (mg/L)	Concentration as TOC (mg/L)		Soluble TOC Removed m/m (%)	Total mass oxygen consumed (mg)	Mass Oxygen consumed per mass TOC consumed (mg/mg)
		Initial	Final			
I	30	12.87	2.66	84.35	11.78	3.10
	60 (1)	27.72	9.59	73.81	51.09	7.14
	60 (2)	25.89	8.03	76.52	64.77	9.34
	150 (1)	65.2	27.94	67.55	120.28	7.80
	150 (2)	71.07	24.82	73.56	104.85	5.73
	300 (1)	133.4	94.71	46.25	201.59	9.34
	300 (2)	120.06	86.16	45.66	235.19	12.26
	II	300	129.45	31.05	81.84	200.22
400 (1)		172.88	34.29	84.98	203.76	3.96
400 (2)		148.68	25.49	87.02	185.17	4.09
500 (1)		217.4	37.75	86.85	326.78	4.94
500 (2)		228.27	49.64	83.54	358.77	5.38
600 (1)		262.6	45.19	86.97	393.64	4.92
600 (2)		273.1	42.99	88.08	418.98	4.98
III		100 (1)	45.82	2.38	96.07	46.35
	100 (2)	43.02	2.03	96.43	57.96	3.99
	400 (1)	173.51	13.95	93.91	176.02	3.09
	400 (2)	197.97	19.92	92.38	205.61	3.21
	1000 (1) *	427.3	3.38	99.40	527.33	3.55
	1000 (2)	429.86	92.48	83.71	472.28	3.75

1000 (1)* = Sample monitored for 40 days.

4.1.2 Respirometer Results Using Sodium Dodecylbenzenesulfonate (LAS)

After positive results in the development of a microbial culture capable of using high concentrations of SDS as a carbon source (i.e., Experiments I-III), Experiment IV was performed to determine if the SDS-degrading culture was able to biodegrade sodium dodecylbenzene sulfonate (LAS). During Experiment IV, oxygen uptake rates and TOC concentrations measured as a function of time indicated that surfactant removal was very low compared to previous tests.

As depicted in Figure 4.5 (top), the pattern of oxygen consumption was a different from that observed with tests conducted using SDS, and the final oxygen consumption for every

sample was almost the same. As depicted in Figure 4.5 (bottom), TOC concentrations did not decrease at a rapid rate.

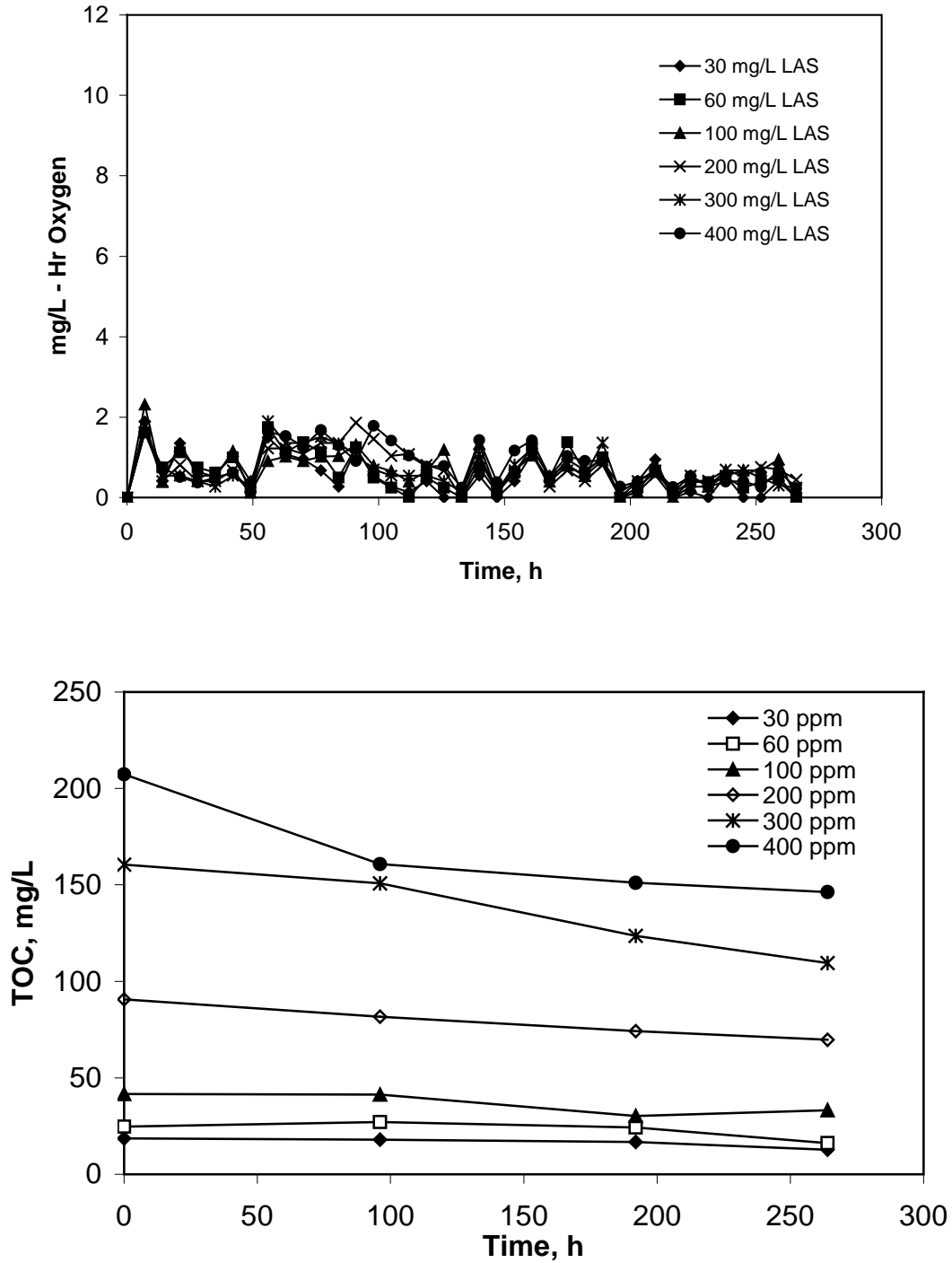


Figure 4.5: Oxygen uptake rate for Experiment IV using LAS (top) and TOC consumption over time (bottom).

Even when some oxygen uptake was measured during the time of the test, most of these values are assumed to belong to endogenous respiration based on oxygen consumption for the control reactor and calculations of oxygen needed for total oxidation for LAS using stoichiometric equations. At the end of the 11-day test period (264 hours), the average TOC removal was 47.5%. The presence of the benzene ring in the LAS chemical structure is believed to be the cause of the low TOC removal from samples during Experiment IV. In the LAS degradation pathway proposed by previous researchers (see Swisher, 1987), ring cleavage is an important step. If the culture developed during Experiments I-III was unable to open the ring, the process of degradation would not proceed to completion.

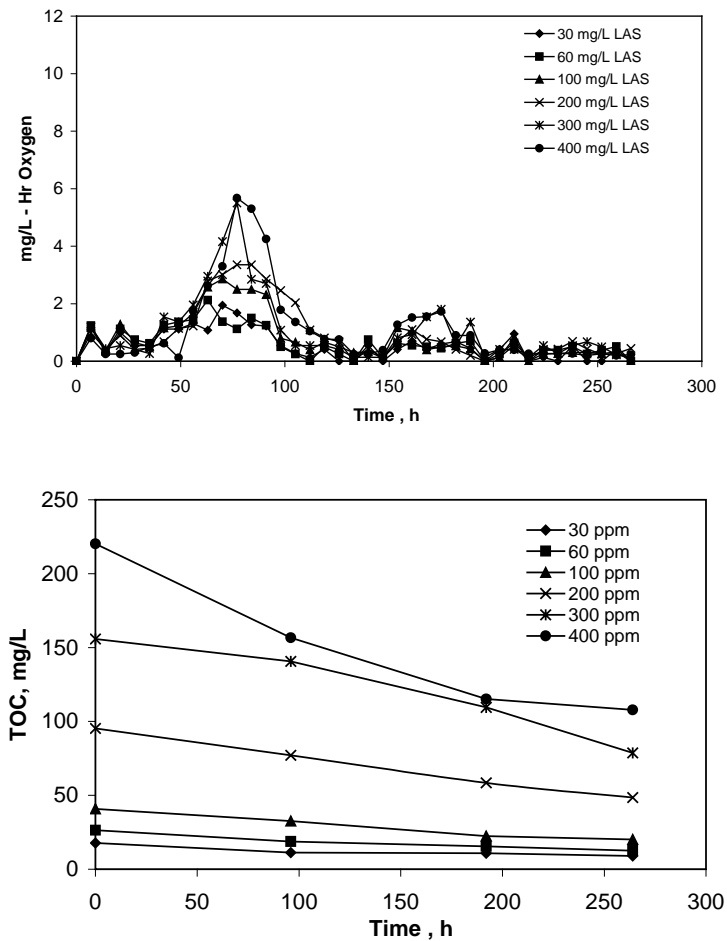


Figure 4.6: Oxygen uptake rate for Experiment V using LAS (top) and TOC consumption over time (bottom).

Experiment V was conducted with the same surfactant type (SDBS), same initial surfactant concentrations (ranging from 30 to 400 mg/L), and same inoculum size (initial concentration of 100 mg/L as TSS) as were used in Experiment IV; however, the source of the microbial inoculum was activated sludge from an industrial wastewater treatment facility. OUR and TOC concentrations measured as a function of time are depicted in Figure 4.6. In Experiment V, the average TOC removal was 62.4% at the end of the test period (11 days).

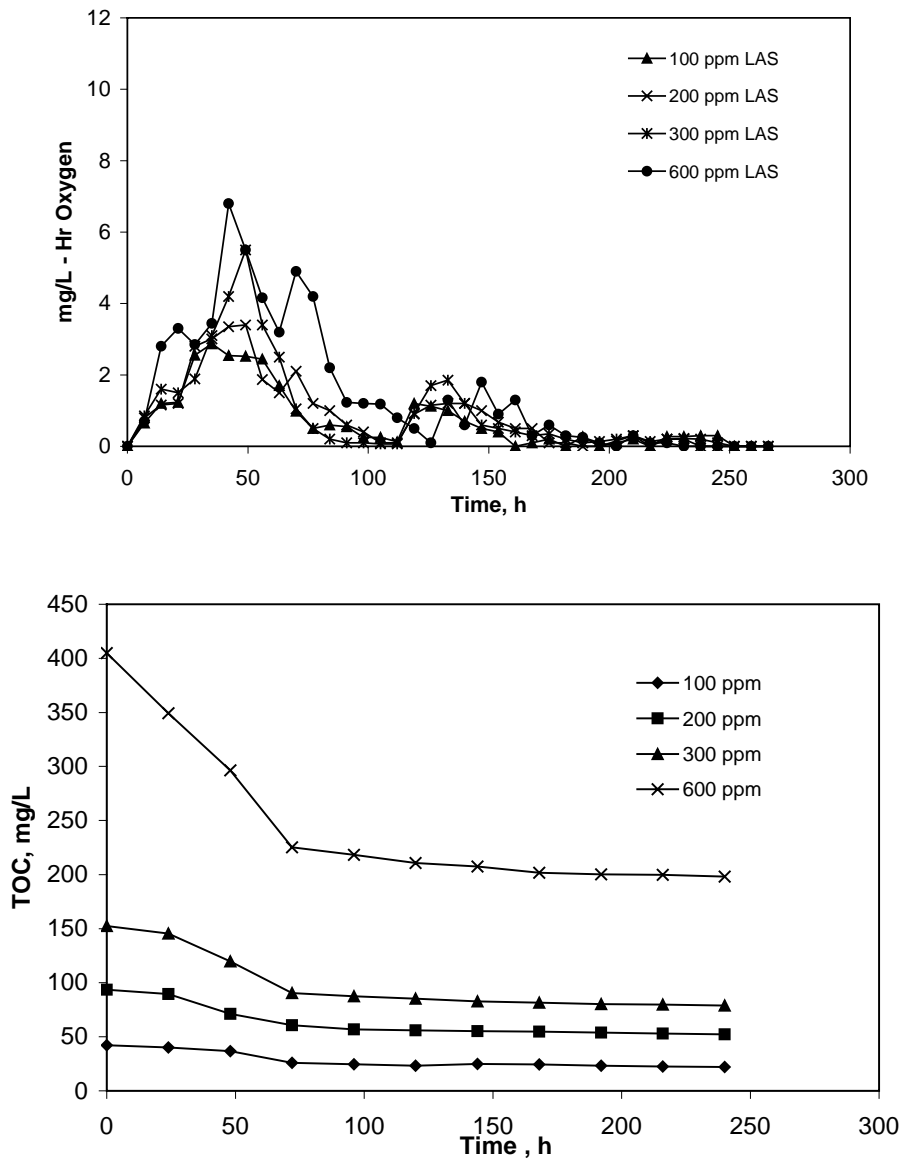


Figure 4.7: Oxygen uptake rate for Experiment VI using LAS (top) and TOC consumption over time (bottom).

Experiment VI was conducted using activated sludge recovered at the end of Experiment V as the inoculum. As shown in Figure 4.7 (top), the maximum OUR observed during Experiment VI was markedly higher than that observed during Experiment V. This provides a general indication that there was enrichment for microorganisms capable of LAS biodegradation. This is further supported by the fact that, as shown in Figure 4.7 (bottom), more rapid consumption of TOC occurred during the first three days of the test. After the third day, TOC concentrations continued to decrease but at a lower rate. The average TOC removal was 60.4% in experiment VI (12 days test period) as shown in Table 4.3.

Table 4.3: Total organic carbon removal in Experiments IV, V, and VI using sodium dodecylbenzene sulfonate (i.e., LAS).

Experiment No.	Concentration as SDS (mg/L)	Concentration as TOC (mg/L)		Soluble TOC Removed m/m (%)	Total mass oxygen consumed (mg)	Mass Oxygen consumed per mass TOC consumed (mg/mg)
		Initial	Final			
IV	30	18.52	12.65	48.28	4.37	1.40
	60	27.15	16.12	55.05	10.01	1.91
	100	41.52	30.18	44.96	12.08	1.85
	200	90.7	69.77	41.76	20.42	1.54
	300	160.29	109.55	48.25	28.19	1.04
	400	207.35	146.32	46.57	31.10	0.92
V	30	17.8	9.02	61.63	9.46	2.46
	60	26.43	12.63	63.82	19.12	3.24
	100	40.9	20.17	62.66	31.93	3.56
	200	95.2	48.55	61.39	62.74	3.07
	300	155.78	78.7	61.75	101.97	3.03
	400	220.3	107.9	62.92	143.40	2.96
VI	100	42.06	21.99	60.41	25.71	2.89
	200	93.45	52.33	57.60	43.45	2.31
	300	152.5	78.89	60.83	79.72	2.46
	600	404.81	198.27	62.92	169.08	1.90

In Experiments VII and VIII, all of the replicates contained the same initial concentration of LAS (500 mg/L of LAS and 319 mg/L as TOC) at the start of the experiment. Figure 4.8 shows the average oxygen uptake rate (top) and TOC and MBAS concentrations as a function of time (bottom). Table 4.4 summarizes the overall results. Average TOC removal was 57.6% and 58.4% for Experiments VII and VIII respectively. The action of averaging data from

Experiments VII and VII was decided due to the similarities in mass of oxygen consumption and TOC removal.

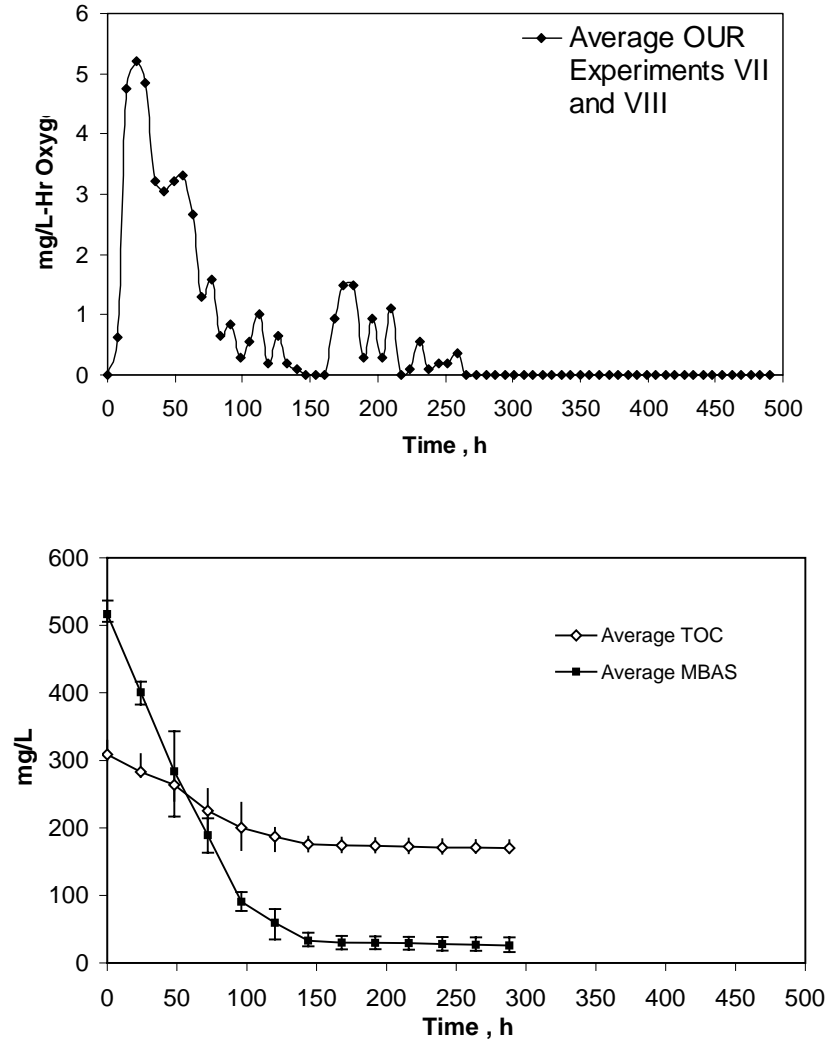


Figure 4.8: Average oxygen uptake rate (OUR) from experiments VII and VIII (top) and TOC consumption (bottom). Error bars denote the range of concentrations measured

There were some differences between the tests using SDS and LAS (i.e., Experiments I-III for SDS and Experiments V-IX for LAS). One difference was the pattern of oxygen uptake rate during the test periods. Samples using LAS exhibited the highest OUR during the first 3.5 days, decreasing during the fourth and fifth days, and then, between the sixth and ninth days, the OUR increased again. TOC data, however, did not show any increase in TOC removal rate

during these days. Another difference between the tests using SDS and LAS was that higher TOC concentrations remained in solution at the end of the test period when LAS was supplied as a carbon source. In tests where SDS was used (Experiments I- III), the remaining TOC concentration was less than 10% when initial SDS concentration was lower than 500 mg/L, and less than 15% for initial concentrations higher than 500 mg/L. However, continuous monitoring from one of the respirometer reactors with an initial SDS concentration of 1000 mg/L during Experiment III showed that a total consumption of TOC was achieved after 40 days. Complete TOC removal was not achieved in any of the experiments using LAS. It should be noted, however, that the maximum incubation period was longer for some SDS experiments than for LAS experiments.

Table 4.4: Summary of results from Experiments VII, VIII, and IX using LAS.

Experiment No.	Concentration as SDS (mg/L)	Concentration as TOC (mg/L)		Concentration as MBAS (mg/L)		Soluble TOC Removed m/m (%)	MBAS Removed m/m (%)	Total mass oxygen consumed (mg)	Mass Oxygen consumed per mass TOC consumed (mg/mg)
		Initial	Final	Initial	Final				
VII	500 (1)	308.49	179.67	505.04	37.6	55.90	92.86	107.28	1.78
	500 (2)	301.5	168.03	513.37	31.5	57.80	93.43	86.35	1.42
	500 (3)	307.52	174.11	512.84	30.8	57.13	93.19	91.59	1.49
	500 (4)	304.87	168.37	518.32	26.5	58.19	93.48	112.51	1.81
	500 (5)	304.38	165.59	522.2	23.1	58.81	93.64	105.19	1.68
	500 (6)	303.9	162.81	526.12	19.7	59.44	93.79	110.42	1.75
VIII	500 (1)	329.26	160.84	505.15	19.34	63.01	93.61	98.57	1.36
	500 (2)	303.86	179.98	509.08	37.9	55.15	92.91	102.56	1.75
	500 (3)	310.29	175.67	518.29	21.78	57.13	93.20	85.57	1.38
	500 (4)	325.62	181.47	536.64	16.09	57.80	93.22	108.56	1.65
	500 (5)	300.29	165.85	522.98	21.74	58.18	93.64	80.57	1.32
	500 (6)	306.63	164.27	507.97	25.15	59.44	93.51	107.56	1.69
IX	500 (LAS)	360.5	107.65	486.24	18.81	77.39	95.56	192.47	1.97
	500 (LAS)	347.9	84.93	477.06	21.16	81.52	96.43	201.88	2.03
	500 (LAS)	360.2	106	492.17	26.34	77.72	95.68	186.05	1.90
	500 (Huntsman)	317.7	133.5	500.12	23.56	68.18	94.64	139.27	1.84
	500 (Huntsman)	322.1	99.5	505.5	17.35	76.61	96.05	154.11	1.78
	500 (Huntsman)	321.8	89.7	507.34	26.15	78.90	96.45	142.83	1.61

The methylene blue active substances (MBAS) analysis was incorporated in Experiments VII and VIII to assess the actual concentration of the LAS parent compound at the end of the test period. The results shown that more than 90% of the parent LAS was no longer available in solution (Table 4.4). From this, it was concluded that the remaining TOC concentration probably

was produced by co-products from degradation reactions which could not be used as carbon source by the microorganisms present.

Experiment IX, the last test using the respirometer, was conducted to determine whether the culture developed in previous respirometer tests was able to degrade a LAS sample provided for Huntsman Co. that has the same characteristics as the LAS produced at the manufacturing facility of concern in Honduras. Figure 4.9 depicts average OUR, TOC, and MBAS consumption for Experiment IX.

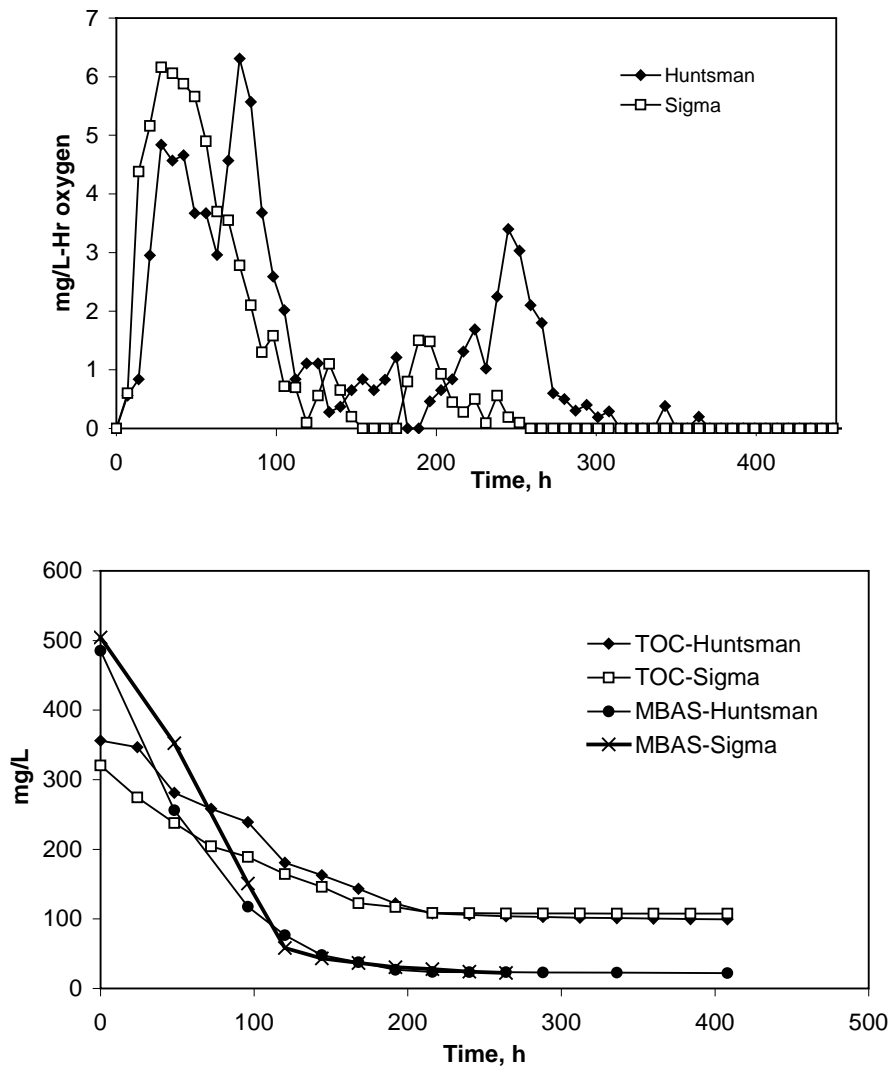


Figure 4.9: Average oxygen uptake rate (OUR) from Experiment IX (top) and TOC and MBAS consumption (bottom).

At the end of the 15-day test period, results showed average of TOC and MBAS removal of 74.6% and 95.7% respectively (Table 4.4), which were very close to those obtained from samples with LAS from Sigma at the same initial concentration. As shown in the top graph, the same biphasic oxygen consumption pattern observed in Experiments VII and VII was observed in Experiment IX. As shown in the bottom graph, TOC and MBAS removal were practically identical for the LAS surfactants provided by both suppliers.

The average first order rate constant for TOC removal and corresponding half-life, calculated as the average of all respirometer bottles within each of the five experiments utilizing LAS (i.e., IV to IX) are summarized in Table 4.5.

Table 4.5. First order rate constants for LAS degradation in Experiments IV to IX.

Experiment No	k (h ⁻¹) average ± std dev	Half Life (h) average ± std dev	R ² Range	R ² Average
IV	0.0014 ± 0.00024	527.09 ± 96.08	72.66 – 99.80	86.37
V	0.0026 ± 0.00019	266.17 ± 20.71	86.93 – 99.71	95.31
VI	0.0026 ± 0.0002	267.88 ± 22.32	74.34 – 75.71	75.34
VII TOC	0.0022 ± 0.00023	313.31 ± 33.81	72.05 – 85.31	81.18
MBAS	0.0113 ± 0.00101	61.56 ± 5.45	83.77 – 85.41	84.31
VIII TOC	0.00186 ± 0.0003	377.38 ± 51.34	53.09 – 77.81	69.24
MBAS	0.0114 ± 0.00102	61.21 ± 5.50	82.58 – 88.86	85.00
IX Huntsman TOC	0.0034 ± 0.00052	206.83 ± 29.05	81.98 – 84.20	84.07
MBAS	0.0078 ± 0.00075	89.03 ± 8.52	68.22 – 84.33	76.65
Sigma TOC	0.0026 ± 0.00061	278.29 ± 75.03	68.22 – 84.33	76.65
MBAS	0.0080 ± 0.00062	86.98 ± 6.58	67.21 – 75.69	71.61

The relatively low R^2 values indicate that a first order model does not perfectly fit the experimental data. As was the case for experiments conducted with SDS, this was not particularly surprising given the fact that the simple first-order degradation model does not account for differences in biomass concentration as a function of time within the various respirometer bottles. Nevertheless, because biomass concentrations were not measured as a function of time in the experiments described herein (and therefore data are not available to allow a more sophisticated modeling approach that includes changes in biomass), the first order model was used as a basis of comparison.

As shown in the table, the first order rate constant was markedly higher when calculated based on MBAS than for TOC. This likely reflects the fact that, as discussed previously, a fraction of the LAS was converted to intermediates that were measurable in terms of TOC but which were not measurable in terms of MBAS, and the overall kinetics of MBAS removal (representing consumption of parent compound) was more rapid than soluble TOC removal (representing mineralization to CO_2 or production of insoluble biomass) in the systems studied.

4.2 Biomass Production

Initial experiments conducted to treat surfactant-containing wastewater in laboratory-scale bioreactors (as opposed to respirometer-bottles) were conducted in reactors arbitrarily designated as R1, R2, and R3. These reactors, operated over a period spanning almost seven months (September 30, 2002 – April 20, 2003), were used to develop the activated sludge which was used in final batch reactor systems described in the following section (Section 4.3).

The first reactor, R1, was started up using biomass collected from Experiment III. R1 was supplied with a synthetic wastewater containing 400 mg/L of SDS during the first four cycles (five days each). During these cycles TOC removal averaged 81%. The situation

changed when the reactor was fed with LAS instead of SDS at the same feeding concentration. Expecting the enrichment of the microorganisms capable of using LAS with every cycle, the reactor (R1) performed eight cycles (ten days each). Each cycle was able to remove an average of only 36% of the initial TOC within cycles. Figure 4.10 shows the overall performance of R1. Clearly, the TOC removal decreased when the surfactant transition occurred from SDS to LAS.

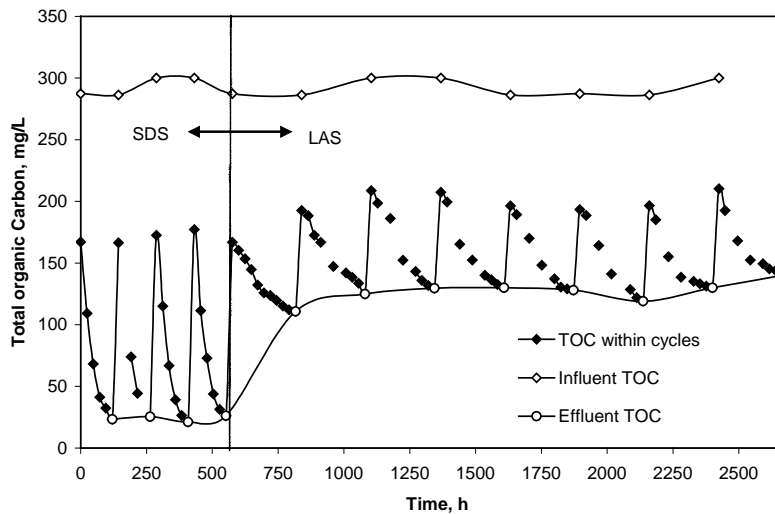


Figure 4.10: Overall performance of R1 (dotted line shows the transition of surfactant type in the influent from SDS to LAS).

After receiving positive results from respirometer Experiment VI (using activated sludge from the petrochemical refinery), reactor R2 was set up and operated for ten cycles (five days each). R2 was supplied with synthetic wastewater containing 400 mg/L of LAS. TOC concentrations as a function of time are shown in Figure 4.11. As shown in the figure, overall TOC removal in each cycle averaged 53%.

Even though R2 performed better than R1, there was a noticeable problem with sludge settling (TSS at the end of the cycles was 160 mg/L). R1 did not produce the settling problem; the TSS concentration in the clarified supernatant after settling at the end of each cycle in R1 was only 10 mg/L. In an attempt to produce an activated sludge with better settling

characteristics while retaining the potential for TOC removal, biomass from reactors R1 and R2 were mixed together and placed in a reactor designated as “R1- NEW.” This new reactor was operated for eight SBR cycles lasting five days each. Mixing accomplished the desired outcome of improving settling characteristics; however, TOC removal during the 5-day cycle time remained essentially unchanged (36% removal), as shown in Figure 4.12.

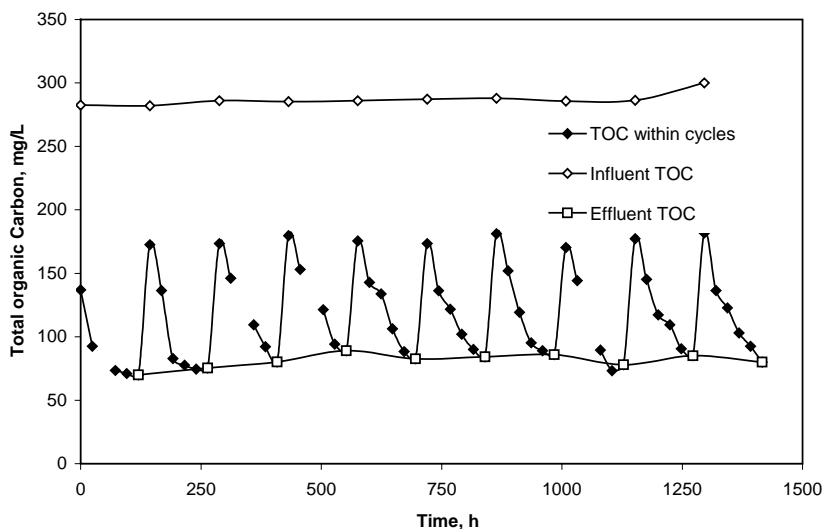


Figure 4.11: Overall performance of R2 in terms of TOC removal

During one cycle, the duration of the REACT period was extended to a total cycle length of ten days. As shown in Figure 4.12, extending the duration of the REACT period in cycle 7 did not improve TOC removal. The cause of the failure to improve TOC removal is unknown. One potential explanation is that one or more of the LAS homologues present in the LAS mixture was not biodegraded; however, because individual homologues were not quantified in the experiments described herein, such a conclusion cannot be drawn from the data collected.

A decision was made to examine the effects of reactor operating strategy on LAS biodegradation. In order to obtain the goal of biomass production to seed the reactors used to study various operating strategies, reactor R3 was set up using activated sludge from Experiment

IX. R3 performed six SBR cycles (five days each) achieving an average of 45.2% TOC removal. Overall performance of R3 is depicted in Figure 4.13.

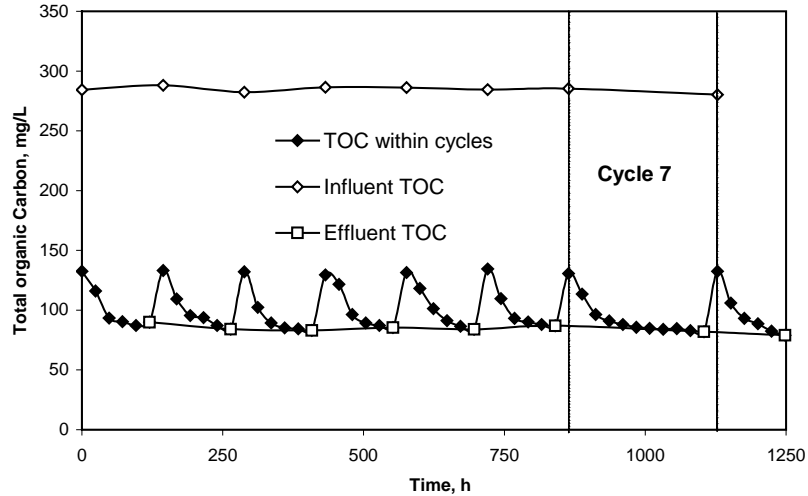


Figure 4.12: Overall performance of R1-NEW in terms of TOC removal

As was observed with R2, poor settling quality of the biomass was observed for R3. TSS measured at the end of SETTLE ranged from 120 to 180 mg/L. Even when the time for SETTLE was increased from one to three hours, there was only a 15% reduction from the initial TSS value measured at the end of the SETTLE period.

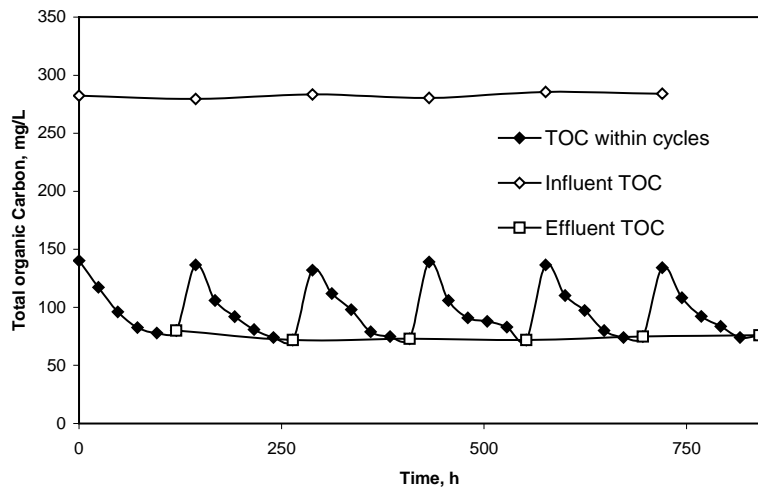


Figure 4.13: Overall performance of R3 in terms of TOC removal

Activated sludge from R3 was then mixed with R1-NEW, and the resulting mix was divided into three kettle reactors arbitrarily named K1, K2, and K3. As described in the subsequent section, these new reactors were used to assess the impact of various operating strategies on LAS biodegradation.

4.3 Comparison of Bioreactor Operating Strategies for LAS Biodegradation and Foam Production: SBR, ICEAS, and SBBR

4.3.1 LAS Removal

4.3.1.1 LAS Removal in K1- Sequencing Batch Reactor

Reactor K1 was operated as an SBR for 185 days, completing 37 cycles lasting five days each. Influent and effluent TOC and MBAS concentrations are shown in Figure 4.14 (top and bottom, respectively). As shown in the figure, over the long-term operation of the bioreactor, a gradual improvement in reactor performance was observed in terms of both TOC and MBAS removal. TOC removal increased from 56.0% during cycle 1 to 77.8% during cycle 37. Following a similar trend, MBAS analysis showed a gradual increase in LAS removal from 82.1% in cycle 1 to 93.6% in cycle 37. Considering all cycles, average TOC removal was 70.1%, and average MBAS removal was 91.2%. This corresponds to average effluent TOC and MBAS concentrations of 80.5 and 38.5 mg/L, respectively.

A considerable amount of the initial TOC was consumed during the first three days of every cycle. Of the fraction of TOC that was removed, approximately 95.0% of the TOC removal occurred during the first three days of each cycle with markedly lower TOC removal rates observed during the fourth and fifth days. This phenomenon can be seen in Figure 4.14 (top) as well as Figure 4.15 (top), which depicts average TOC concentrations as a function of time within several cycles (further data are available in Appendix 3).

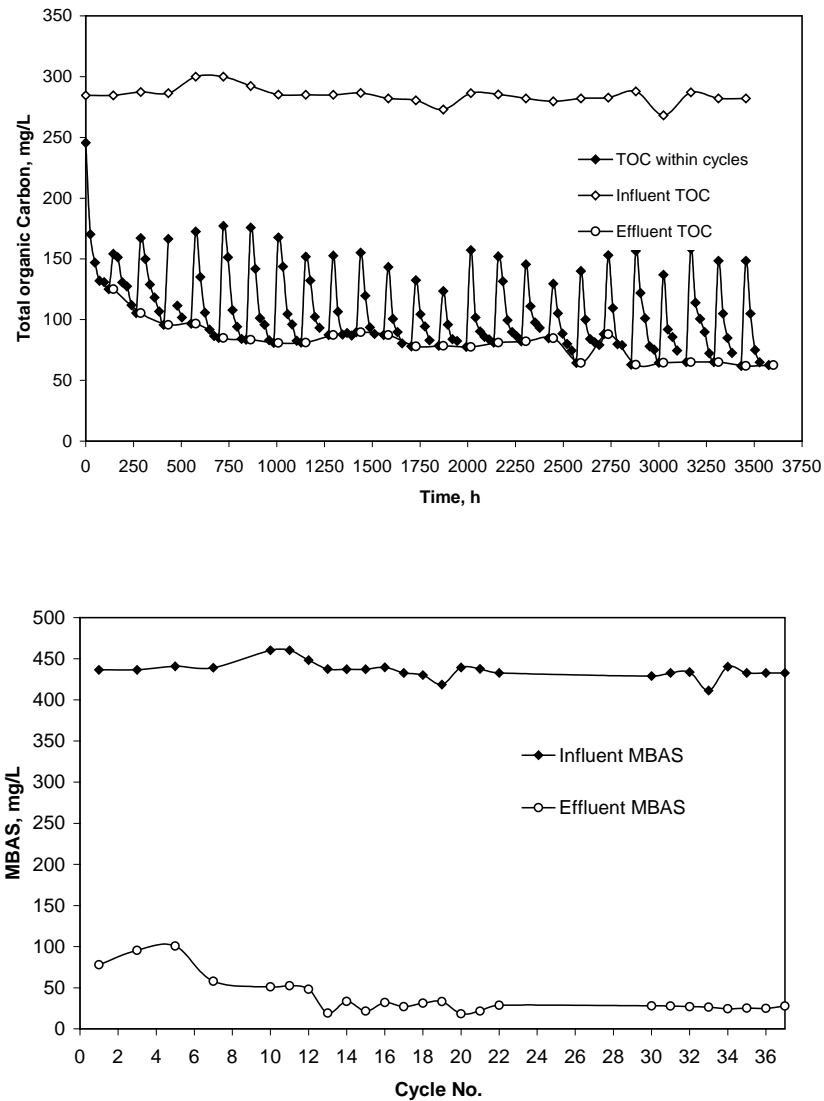


Figure 4.14: TOC concentration in influent, within cycles, and effluent (top) and MBAS concentration in the influent and effluent (bottom) during K1 operation (SBR).

A similar trend was observed for MBAS removal (see Figure 4.15, middle). The MBAS concentration decreased relatively quickly during the first 72 hours of the cycle (3 days), and then remained relatively constant for the remainder of the cycle duration (total of 120 hours, 5 days). Consistent with the pattern observed for TOC and MBAS removal, OUR was relatively high during the first 72 hours and then decreased (see Figure 4.15, bottom). All of these data are consistent with Experiments VII and VIII conducted using the respirometer and demonstrate that aerobic biodegradation of LAS in an SBR is a feasible process for LAS removal.

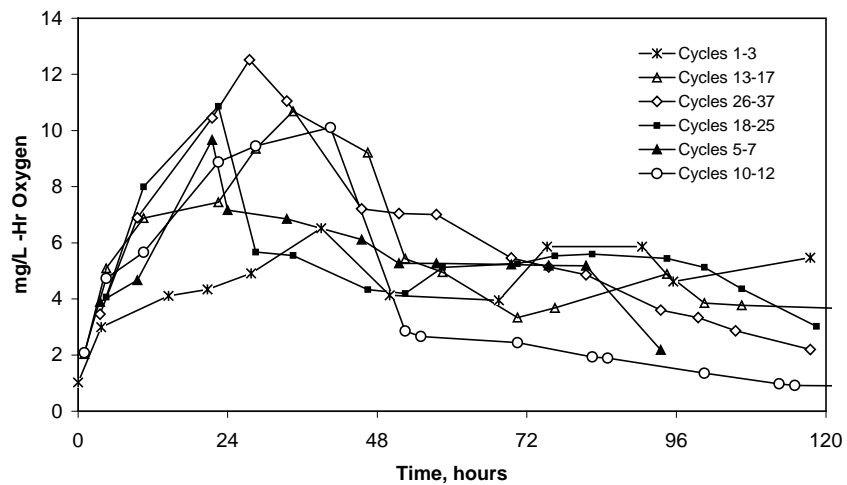
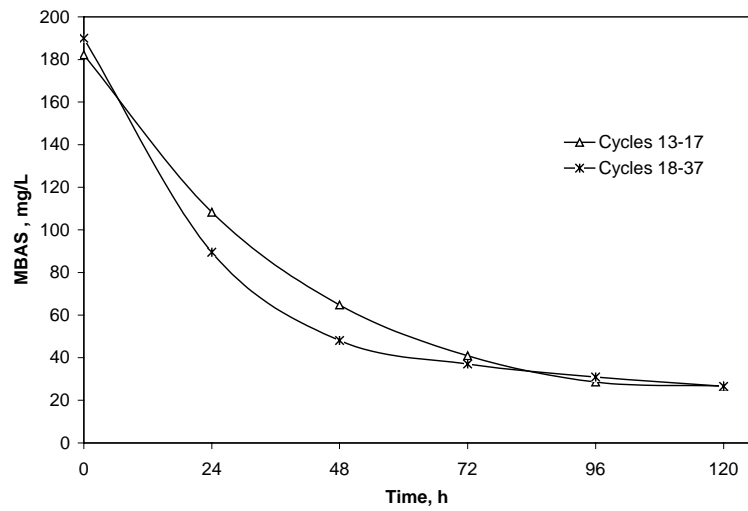
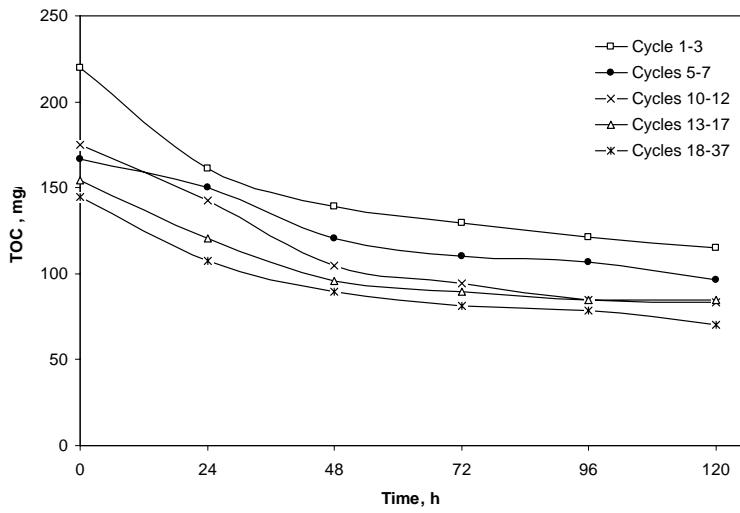


Figure 4.15: Average TOC values within the five-day cycle in SBR (top), average MBAS (middle), and OUR (bottom).

Because the K1 reactor utilized a short (2.88 minutes) un-aerated FILL period, the surfactant concentration in the reactor at the start of the REACT period was relatively high, averaging 158 mg/L considering all SBR cycles. Consequently, when air was supplied via the diffuser stone at the start of the REACT period, foam was produced. The quantity of foam produced in the system was measured and recorded during selected cycles. Foam production followed almost the same trend from cycles 15 (the first cycle in which foam production was measured) through 37 (the last cycle for which foam production was measured, see Appendix 7 for a complete listing of data). Figure 4.16 shows the typical foam height as a function of time during an SBR cycle (average data from cycles 15, 16, 21, 22, and 36). Figure 4.17 depicts the foam height observed in K1 during the 10-minute interval between air supply to the reactor (average data collected during cycles 15, 16, 21, and 22). Time zero in the graph corresponds to the start of the 30-second interval when aeration was supplied.

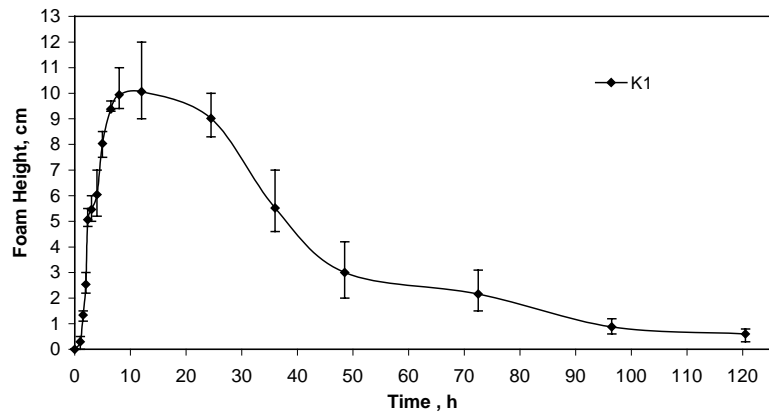


Figure 4.16: Foam production as a function of time during the REACT period in the SBR (K1). Data shown are the average of measurements collected during cycles 15, 16, 21, 22, and 36. Error bars represent the range of foam heights measured during the testing cycles.

As shown in Figure 4.17, the foam height did not change appreciably as a function of time within the short-term (i.e., 9.5 minute) interval between when air was supplied. As shown in Figure 4.16, however, there was a marked difference in the amount of foaming over the longer-

term (i.e., 5-day) cycle length. Rapid foam development was observed during the first three hours even though an intermittent aeration strategy was employed (air supplied for a 30 second interval during each 10 minute period – see Section 3.4.1.1). Peak foam height (approximately 10 cm) was observed approximately ten hours into the REACT period, and then a gradual decrease followed during the next 40 hours. During the last two days of the REACT period (i.e., after 72 hours), the quantity of foam was relatively small (e.g., one centimeter in height).

Visual observation revealed that the physical appearance of the foam changed as a function of time during the length of the cycle. During the first two days, the foam was comprised of tiny bubbles that did not readily coalesce or collapse. By the third day (i.e., 72 hours) the appearance of the foam changed. The foam was much less stable, with bubbles coalescing and bursting after a short period of time. These characteristics coincide with the fact that by the third day, most of the degradable TOC had already been consumed.

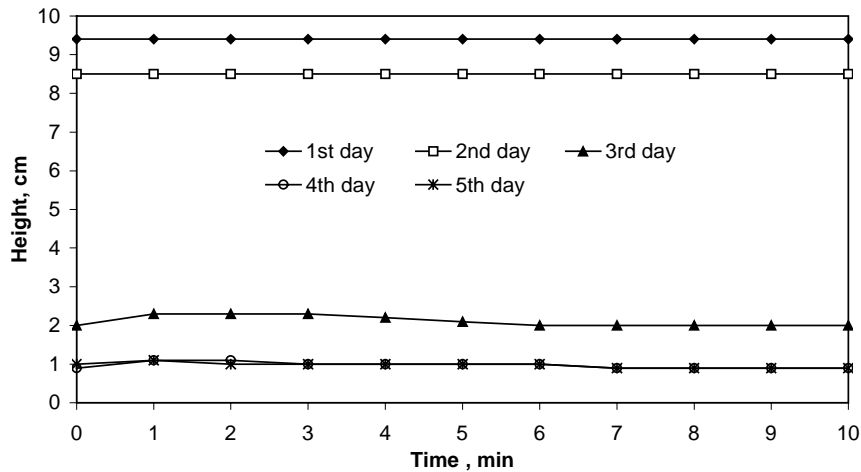


Figure 4.17. Foam height observed in K1 during the 10-minute interval between air supply to the reactor (average data collected during cycles 15, 16, 21, and 22). Time zero in the graph corresponds to the start of the 30-second interval when aeration was supplied.

Foam production in the K1 reactor led to some operational difficulties. Once aeration began, a fraction of the suspended solids became entrained in the foam that formed. The foaming deposited biomass onto the interior surfaces of the glass reactor. Daily scraping of the

reactor's walls was necessary, using a rubber spatula, to reincorporate the biomass into the liquid suspension.

Figure 4.18 (top) depicts the Mixed Liqueur Suspended Solids (MLSS) concentration in the K1 reactor measured as TSS. The MLSS concentration was 1,200 mg/L at the beginning of cycle 1. As shown in Figure 4.18, during the early stages of reactor operation (i.e., cycles 1 to 7), the MLSS concentration decreased.

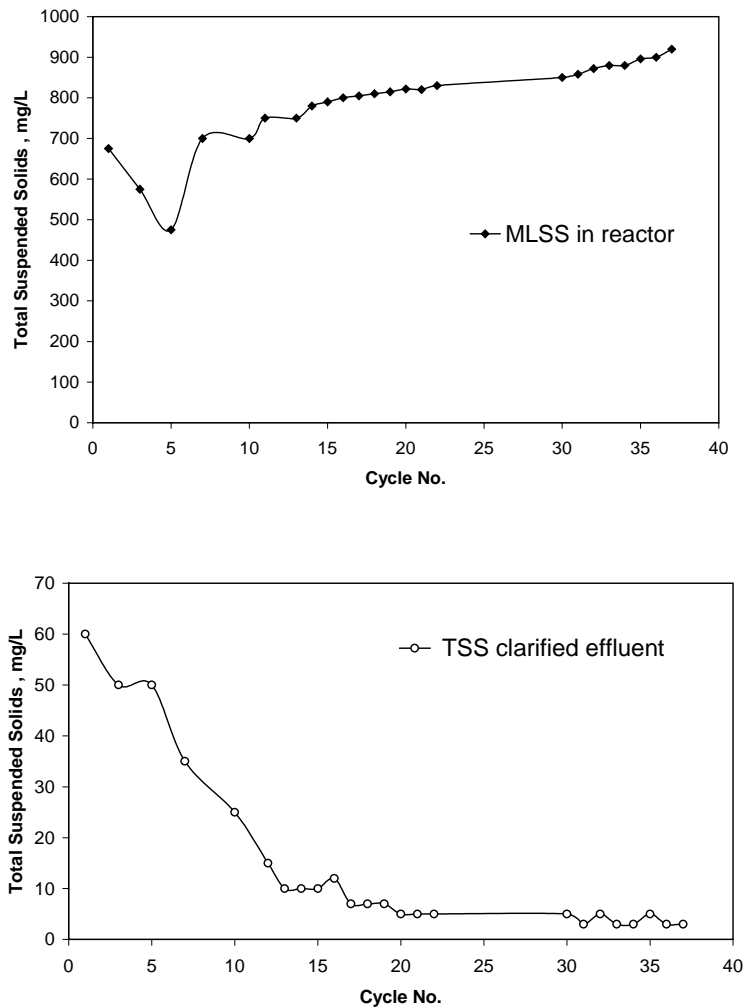


Figure 4.18: Mixed Liqueur Suspended Solids (MLSS) in the reactor measured at the end of every cycle (top), and TSS of the clarified effluent from the SBR (K1)

The decrease in MLSS concentration was likely due to two separate effects. First, the TSS concentration in the clarified supernatant (reactor effluent) was relatively high,

approximately 60 mg/L during cycle 1, and averaging 53.3 mg/L for cycles 1 – 7. This led to discharge of suspended solids in the treated effluent during the decant procedure. Second, some fraction of the sludge was likely lost because it was expelled from the reactor due to excessive foaming.

The settling capacity of the activated sludge in K1 gradually improved with time. After cycle 19, TSS measured in the clarified supernatant remained almost constant at 5 mg/L (see Figure 4.18, bottom). After the effluent TSS concentration decreased, more solids were retained in the system, and the MLSS concentration gradually increased over time, reaching 920 mg/L TSS at the end of cycle 37.

The SVI was measured as an indicator of sludge settling capacity; however, SVI measurements were only performed during three cycles, 20, 21, and 22. An average SVI of 51.9 mL/g was calculated from measurements performed at the end of cycles 20, 21, and 22 in the SBR. Table 4.6 presents values of TSS and settled sludge volume recorded in the Imhoff settling cone for each of the trials. The low SVI (i.e., less than 50 mL/g) is indicative of excellent settling and compaction characteristics (Grady *et al.*, 1999).

Table 4.6: Sludge volume index values for the SBR (K1).

Cycle No.	TSS (mg/L)	Settled sludge volume (mL/L)	SVI (mL/g)
Cycle 20	822	43.1	52.4
Cycle 21	820	41.9	51.2
Cycle 22	830	43.2	52.1

Throughout the operation of K1, there was not any intentional wasting of biomass. Biomass was removed from the system, however, through sample collection as well as via discharge of TSS in the clarified effluent. Because biomass loss from the system from these two mechanisms was expected to be relatively small, the solids residence time (SRT) was rather

large. Nevertheless, the net increase in biomass (measured as MLSS) was rather small (see Figure 4.18, top), indicating that the net yield was rather small.

Figure 4.19 depicts a COD balance performed on the K1 reactor during cycle 37. The total COD mass at the beginning of the cycle was 5,034 mg. It was comprised of residual soluble COD remaining at the end of cycle 36 (391 mg), COD contributed from the influent synthetic wastewater containing LAS (1469 mg), and COD equivalent of biomass in the system (3,174 mg). The mass of residual and influent COD was calculated from direct measurements of COD concentrations and measured liquid volumes. The COD mass from the activated sludge was calculated using an assumed COD-to-biomass ratio of 1.42 (Metcalf and Eddy, 1993). Total COD mass accounted for at the end of cycle 37 was 5,707 mg. This was comprised of residual soluble COD (measured at the end of cycle 37), COD from the activated sludge (calculated from the MLSS concentration measured at the end of the cycle and the assumed COD equivalent of biomass), and the mass of oxygen consumed. The latter was calculated using the OUR measured during the entire cycle (see Appendix 6). COD removed from the system during sampling was also accounted for in the calculations.

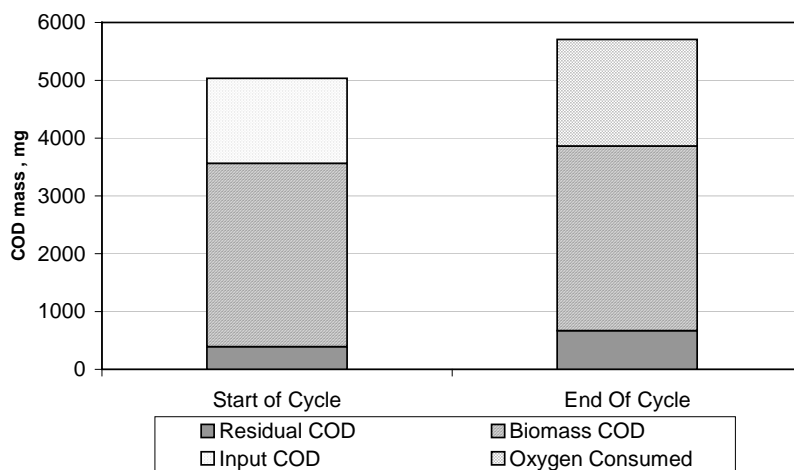


Figure 4.19: COD mass balance on cycle 37 for the SBR.

The difference between the initial COD mass and that accounted for as COD remaining at the end of the cycle or COD consumed or removed during the cycle was 673 mg, 13.4% of the total initial COD. Thus, there was a reasonable closure on the COD balance within the system.

The pH in the K1 reactor was measured and recorded at various time intervals during cycles 20 to 28 and 32-36. Average pH at the beginning and end of the React periods were 7.7 and 6.8, respectively. Consequently no additional pH adjustment or control was deemed necessary. From TOC analysis, it was observed that the average inorganic carbon concentration ranged between 140 to 77 mg/L from the beginning to the end of the React period.

4.3.1.2 LAS removal in K2 – Intermittent Cycle Extended Aeration System (ICEAS)

For the first 12 cycles of operation (lasting 5 days each, total of 60 days), reactor K2 was operated as an SBR, and then the transition was made to operation as an ICEAS. K2 had an initial TSS concentration of 1200 mg/L at the start of operation as an SBR. K2 completed 22 cycles as an ICEAS (total operation time as an ICEAS of 125 days). For purposes of clarity, in the remainder of this thesis section, the cycle number refers to time in operation as an ICEAS (i.e., cycle 1 refers to the first cycle in which K2 was operated as an ICEAS).

Figure 4.20 depicts the overall performance of K2 in terms of TOC and MBAS removal. In this figure, data for cycles 12, 17, and 20 are not represented in the graphs because transient-loading tests were performed during those cycles (results from the transient loading tests are described in section 4.3.3.2). For the data depicted in Figure 4.20, the average influent concentration of LAS was 282 mg/L as TOC and 432 mg/L as MBAS (see Appendix 4 for data in table format). In the beginning cycle (cycle 1), TOC removal was 62.2%. Over the long-term operation of the bioreactor, performance in terms of TOC and MBAS removal gradually improved. By the end of cycle 22, removal improved to 78.7% for TOC and 95.6% for MBAS.

This corresponds to effluent TOC and MBAS concentrations of 61.0 and 19.5 mg/L respectively. Considering all 19 cycles of operation as an ICEAS that are depicted in Figure 4.20, average TOC removal was 77.4% and 95.6% as MBAS, corresponding to effluent TOC and MBAS concentrations of 72 and 26 mg/L, respectively.

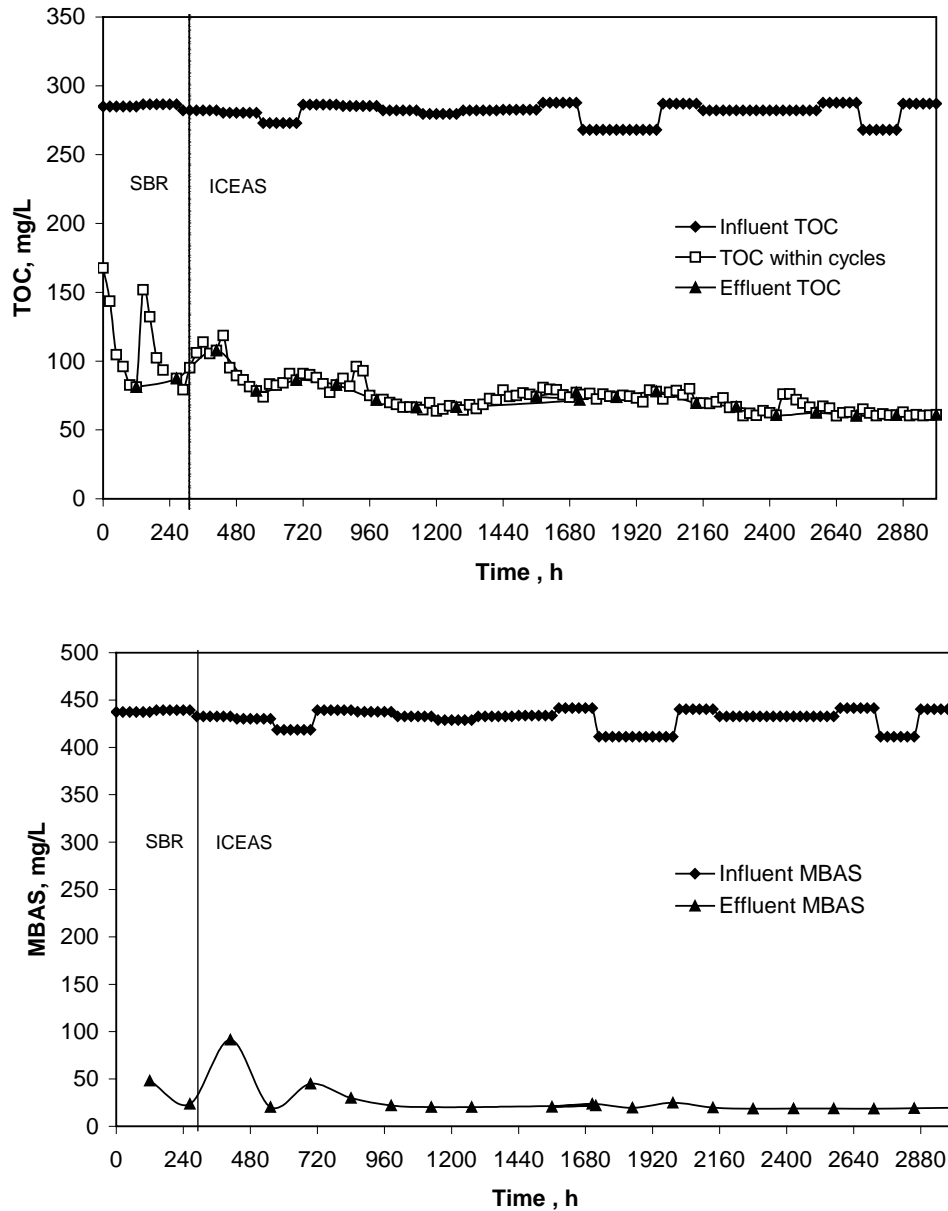


Figure 4.20: Overall performance of K2. TOC concentrations in the influent, effluent, and within cycles (top), and MBAS concentration of the influent and effluent (bottom) during ICEAS operation.

From Figure 4.20, it can be seen that the TOC within the cycles remained almost constant with concentrations being similar to those of the effluent. This can be further observed from data depicted in Figure 4.21 which shows the TOC concentration (top), MBAS concentration (middle) and OUR (bottom) as a function of time within the reactor cycles. Because mixing and aeration were supplied to the reactor during the entire duration that synthetic wastewater continuously entered the reactor over the 5-day cycle length, LAS was degraded as it entered the reactor. This prevented the degree of surfactant accumulation within the reactor that was observed for K1 where the Fill period was quite short (2.88 minutes).

After the end of cycle 6 (1,128 hr), when performance had stabilized, the pattern of OUR as a function of time within the reactor cycle consistently followed a trend of an initial increase at the start of the cycle followed by a steady decrease during the remainder of the cycle length. As further described in the following pages, the initial increase in OUR can be readily attributed to degradation of surfactant accumulated in the system during the unaerated settle and draw periods. The decrease in OUR as a function of cycle length can be readily attributed to the fact that the MLSS concentration decreased throughout the cycle length. The mass of biomass in the reactor was relatively constant (see subsequent discussion of net yield) while the reactor's liquid volume doubled from 1.25 to 2.5 L during the 4.95-day Fill period (thereby decreasing the MLSS concentration by approximately half). As shown in Figure 4.21 (bottom), the OUR at the end of the Fill/React period was roughly half of that observed at the start of the cycle, indicating that the specific oxygen uptake rate (SOUR, mass of oxygen consumed per mass of suspended solids per time) was relatively constant during most of the cycle length. This supports the notion that after the first 24 hours of the cycle, LAS was degraded as it entered the system and little accumulation occurred.

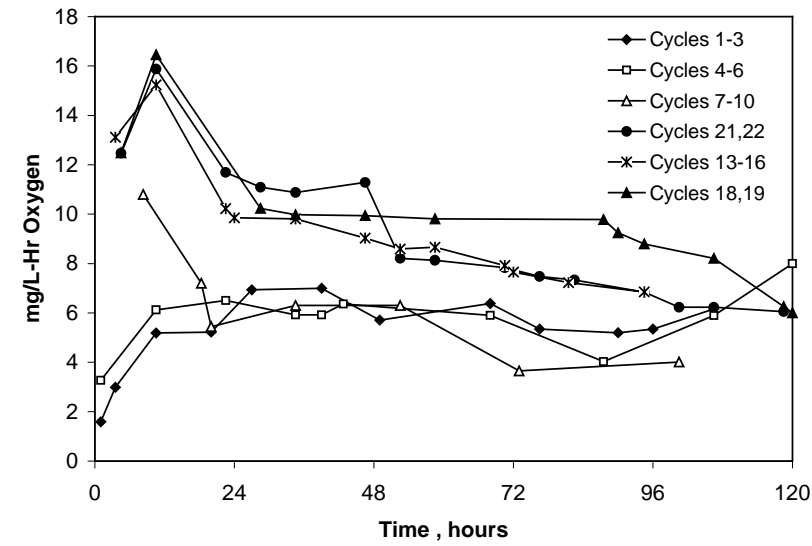
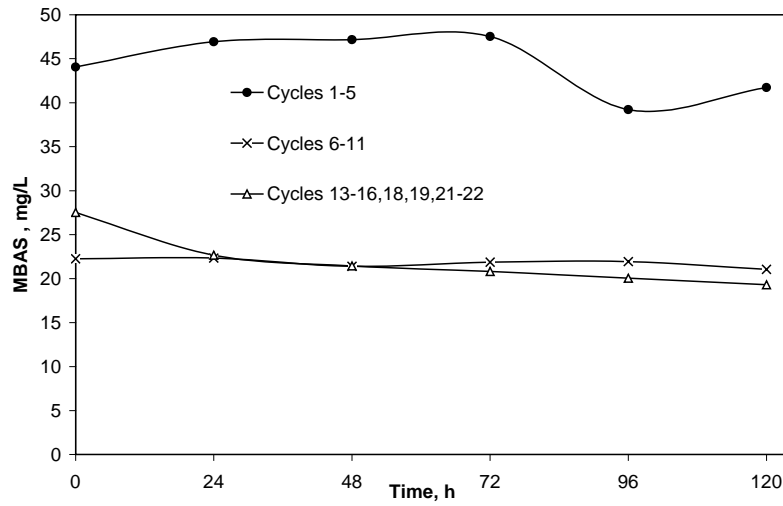
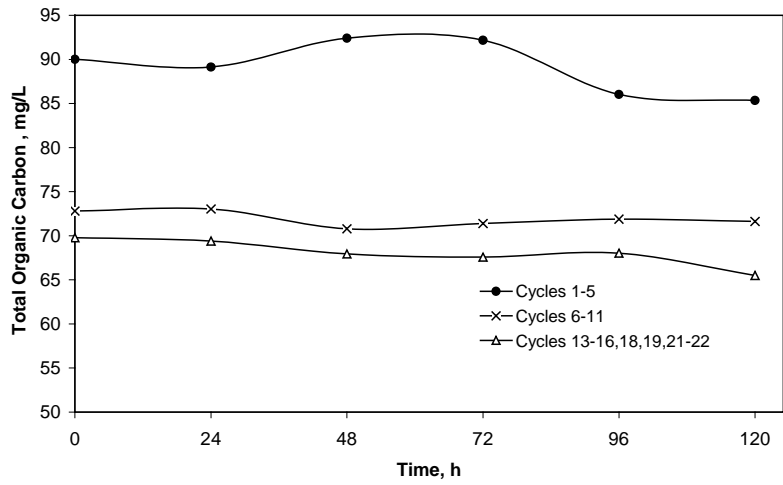


Figure 4.21: Performance of K2 within cycles. TOC (top), MBAS (middle), and OUR (bottom).

During the first two cycles after the transition was made from operation as an SBR to operation as an ICEAS, the amount of foam observed in the system was relatively constant with a height between 1.5 and 2.0 cm during the entire cycle length. For cycles 3 to 8 (see Figure 4.22), a small amount foaming (approximately 0.2 cm of height) was briefly observed during the first two hours at the beginning of the cycle, and then foaming ceased until approximately 24 hours when it was observed again. Peak foam height was observed at a time 36 hours into the cycle, and then it decreased. Final foam height at the end of cycle 8 was small but measurable, averaging 0.3 cm.

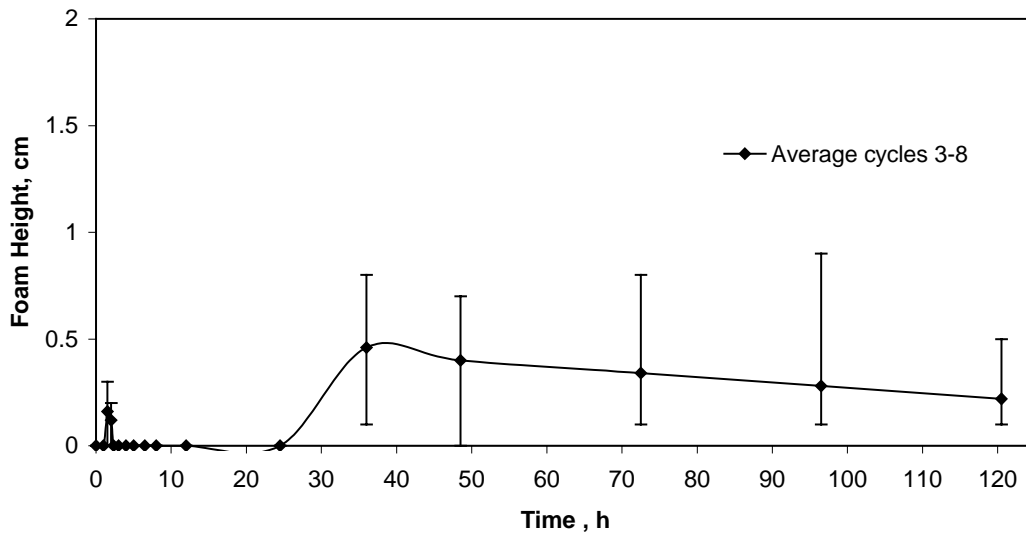


Figure 4.22 Foam production as a function of time in the ICEAS. Data shown are the average measurements collected during cycles 3 to 8.

From cycles 9 to 22, foam was transiently observed only at the beginning of the cycle. This is consistent with the fact that there was some accumulation of influent LAS at the bottom of the reactor during the 1.12 hour interval when settle and draw periods were taking place. Because the surfactant was then removed at approximately the same rate that it entered during the remainder of the operating cycle (see figure 4.21), there was no appreciable foam production during the rest of the operating cycle. In fact, most of the cycles had no measurable amount of

foam production. This is in marked contrast to the foam production observed in K1 where substantial foam production was observed for the first two days of every cycle (see Figure 4.16).

From analysis of TOC data collected during the first few cycles of ICEAS operation, it was noticed that the concentration of total inorganic carbon in the reactor remained almost constant at 6 mg/L throughout the cycle duration, substantially lower than that observed in K1. Starting during cycle 4, the pH was regulated through regular addition of 10 M NaOH solution. This maintained the reactor's pH between 6.6 and 7.3 during the cycle length.

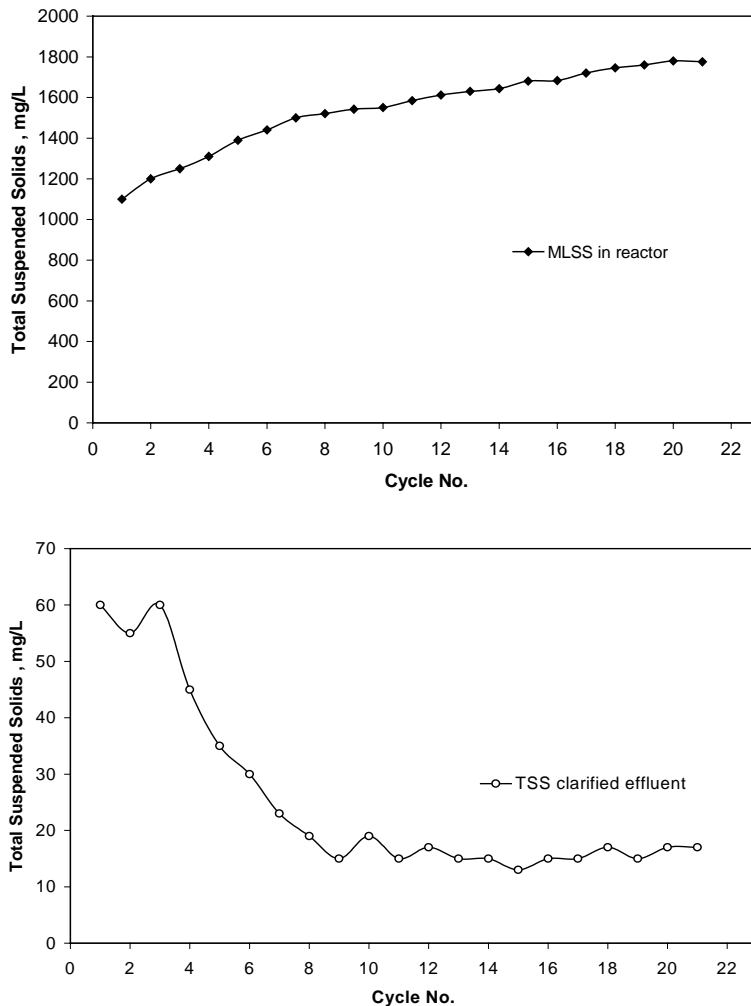


Figure 4.23: Mixed Liqueur Suspended Solids (MLSS) in the reactor measured at the end of every cycle (top), and TSS of the clarified effluent from the ICEAS (K2) (bottom)

Figure 4.23 (top) depicts the MLSS concentration (measured as TSS) in the K2 reactor at the end of REACT periods, and Figure 4.23 (bottom) depicts the TSS concentration in the decanted discharge. As shown in the figure, the TSS concentration in the clarified effluent was relatively high at the start of operation as an ICEAS (averaging 55.0 mg/L during cycles 1 – 4). Performance gradually improved over time, however, and after cycle 13, the effluent TSS concentration was relatively stable, ranging between 15 and 17 mg/L. Because there was no intentional wasting from the system other than from removal of samples for analysis, the MLSS concentration in K2 increased over time, reaching 1,775 mg/L at the end of cycle 22.

At the end of cycle 6, visual observation revealed a light green color in the reactor and in samples taken to measure TSS. It was suspected that algal growth was responsible for this observation. To minimize the potential for algal growth, all three of the bioreactors (i.e., K1, K2, and K3) were covered with aluminum foil to avoid light penetration. The green color disappeared by cycle 8. To further assess sludge settling in the ICEAS system, the SVI was measured during cycles 13, 14, and 15. Table 4.7 presents TSS concentrations, settled sludge volume recorded in the Imhoff settling cone, and SVI for each of the three measurements. The average SVI, 135 mL/g, calculated for the ICEAS (K2) was higher than that observed for the SBR (K1); however, it was still within the generally acceptable range of less than 150 mL/g (Grady *et al.*, 1999).

Table 4.7: Sludge volume index values for the ICEAS (K2).

Cycle No.	TSS (mg/L)	Settled sludge volume (mL/L)	SVI (mL/g)
Cycle 13	1612	214.87	133
Cycle 14	1630	222.3	136
Cycle 15	1643	222.5	135

Figure 4.24 depicts results from a COD mass balance performed on the K2 reactor during cycle 22, following the same procedure and using the same assumptions as were used for the COD mass balance on the SBR (K1). The total mass of COD present in the K2 reactor at the beginning of the cycle (residual soluble COD remaining from the end of cycle 21 and the COD equivalent of biomass) plus the mass of COD entering the reactor as LAS-containing synthetic wastewater was 7,703 mg. Total COD mass accounted for at the end of cycle 22 was 8,332 mg. The latter was comprised of residual soluble COD measured at the end of the cycle, COD equivalent of the activated sludge, the mass of oxygen consumed (calculated using OUR measured during the cycle – see Appendix 6), and COD associated with samples removed from the system. The difference between the initial and final, 629 mg COD, represents 8.2% of the initial COD and indicates that there was a reasonable closure on the COD balance in the system.

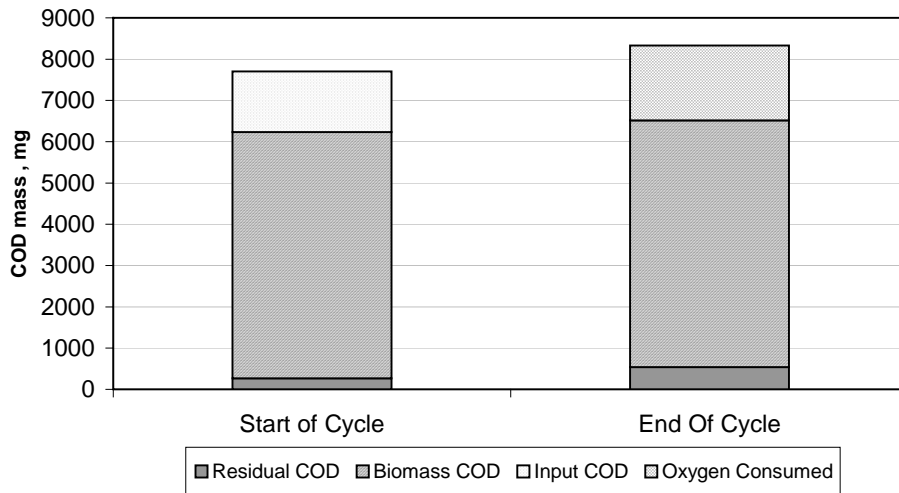


Figure 4.24: COD mass balance on cycle 22 for the ICEAS.

4.3.1.3 LAS removal in K3 – Sequencing Batch Biofilm Reactor (SBBR)

Reactor K3 was operated as an SBBR for 66 cycles (lasting from 1 to 5 days each) over a period lasting a total of 122 days. The initial TSS in K3 was 1200 mg/L, the same as for K1 and K2. Visual observation of the K3 reactor at various time intervals during the first cycle of

operation revealed that a portion of the biomass initially introduced into the reactor as suspended solids attached to the packing medium and there was a noticeable decrease in turbidity over time. A portion of the biomass, however, did not attach to the foam packing medium and remained in the reactor as suspended solids during the initial cycles. Because there was no distinct settle period (i.e., mixing was provided during the draw period), suspended solids were removed with the decanted water at the end of the treatment cycle. As shown in Figure 4.25, the effluent TSS concentration was approximately 75 mg/L at the end of cycle 1 and then decreased during the next nine cycles until reaching a low stable value for the remainder of reactor operation.

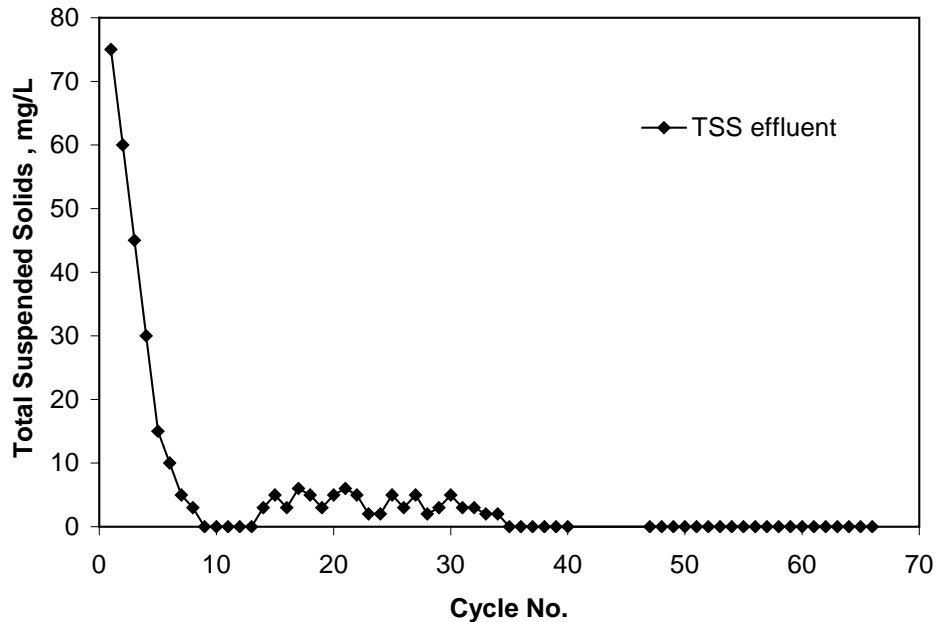


Figure 4.25. Effluent Total Suspended Solids (TSS) for K3.

During the first 31 cycles of operation, a number of operational difficulties were encountered. Figure 4.26 shows the performance of K3 during the first 31 cycles in terms of TOC and MBAS concentrations. Table 4.8 summarizes the cycle lengths employed in reactor operation during this time interval. As shown in Figure 4.26, TOC and MBAS removal in the K3 reactor was lower and more variable than that observed in the K1 and K2 reactors.

Table 4.8: Cycle length description for the SBBR during the first 31 cycles

Length of the cycle, hours	Cycles
24	6-8, 13-30
48	2-5,12,31
72	1,9-11

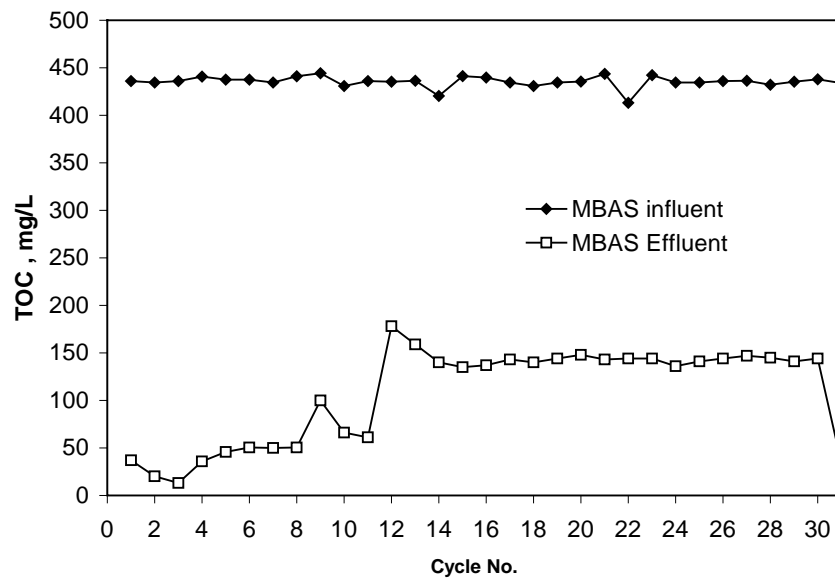
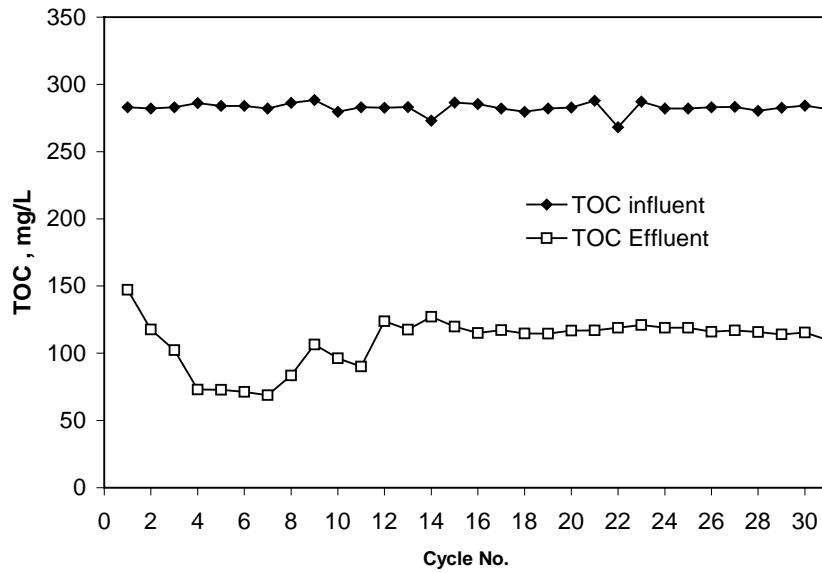


Figure 4.26: Performance of K3 during the first 31 cycles in terms of TOC (top) and MBAS (bottom) concentrations

A likely explanation for the poor performance during cycles 1 to 13 is variability and decrease in pH over time during each cycle. As shown in Figure 4.27 (average data from cycles 9, 10, and 11), the pH decreased from approximately 7.4 at the start of the React period (the end of the Fill period) to approximately 4.9 at the end of a three-day (72-hour) cycle. Furthermore, analysis from TOC measurements from cycles 1 to 13 revealed that the concentration of inorganic carbon in the reactor markedly decreased during the React period (from an average initial concentration of 371 mg/L to an average final concentration of 2.2 mg/L after 72 hours).

It was hypothesized that nitrifying bacteria may be consuming the inorganic carbon and caused the drop in pH (ammonia was supplied as a nitrogen source in the synthetic wastewater, and the process of nitrification consumes both inorganic carbon and alkalinity). To assess whether nitrification was taking place, ammonia, nitrite, and nitrate concentrations were measured at various intervals during cycles 3 and 8. If consumption of inorganic carbon was caused by nitrifying bacteria, then it was expected that the ammonia concentration would decrease at a rate proportional to the rate that inorganic carbon was consumed and nitrite or nitrate were formed. Results from the ammonia analysis showed that an average of 89 mg of NH_3 was consumed per cycle. There was no measurable concentration of nitrite or nitrate. Using the measured change in TOC concentration during cycles 3 to 8 and making several assumptions, the stoichiometric requirement for nitrogen associated with microbial growth was calculated to be 116.2 mg. The assumptions used in making this calculation were that: 1) the COD to TOC ratio was 3.37 mg/mg (calculated based on the chemical formula for LAS); 2) the net microbial yield on a COD basis was 0.6 mg dry biomass produced per mg COD consumed; and 3) dry biomass is 12% nitrogen on a mass basis. The fact that the ammonia consumption was less than the rough estimate of the stoichiometric requirement for heterotrophic growth

combined with the fact that no nitrite or nitrate was detected collectively suggest that nitrification was not taking place in the reactor. An alternate explanation for the decrease in pH over time is that acidic intermediates were produced as a result of LAS degradation (Perez *et al.*, 1996, Swisher, 1987); however, because no attempt was made to identify or quantify intermediates, a general conclusion regarding whether that was in fact the case cannot be made.

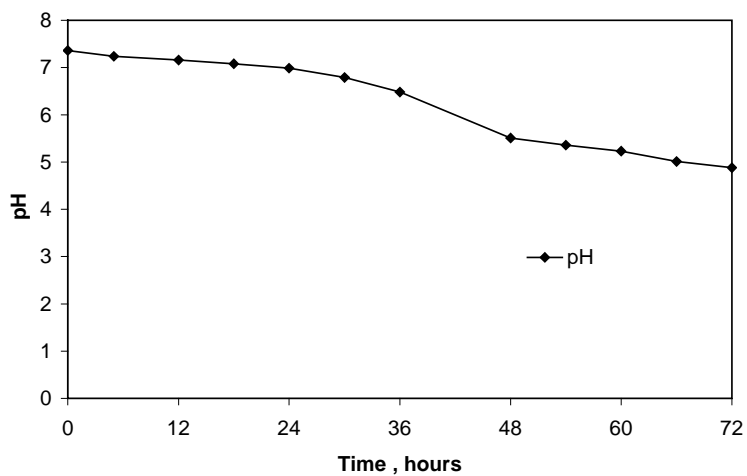


Figure 4.27. Average pH values in a three-day cycle in the SBBR (data from cycles 9-11)

Regardless of the cause of the pH decrease over time, three techniques were employed in an attempt to regulate pH in the reactor. During cycles 14-16, the amount of NaHCO_3 in the nutrient solution was increased. This caused the pH in the synthetic wastewater to be high (pH 8 – 10). An ammonia-like odor was detected, and analysis of the synthetic wastewater feed revealed that the ammonia concentration in the feed solution was much lower than that of the freshly prepared media. Consequently, in subsequent cycles, the NaHCO_3 in the synthetic wastewater was returned to its original level (see Table 3.1).

From cycles 17 to 31, NaHCO_3 was added separately to the reactor at 12 hours intervals as necessary to maintain a pH between 7.5 and 8.0. Increased turbidity started to appear in the reactor during cycle 19. Two potential reasons for the increased turbidity are that: 1) the

NaHCO₃ used to regulate pH may have caused precipitation of mineral salts present in the nutrient solution, and 2) growth of suspended biomass that did not readily attach to the polyurethane foam packing medium. Neither of these potential explanations was confirmed by experimental analysis, and the addition of NaHCO₃ continued until cycle 31.

Starting in cycle 32 and continuing until the end of the experiment, a small volume (usually 0.6 mL) of solution containing 10 M NaOH was added directly to the reactor as necessary (at approximately every 6 hours) to maintain the pH at a level between pH 6.5 and 7.5 during the entire length of the cycle. Once the pH was stabilized, reactor performance began to improve. K3 performed a total of 34 cycles during which pH was controlled by NaOH addition. Of these, cycles 52, 56, and 58 ran for 3 days each. Cycle 64 ran for 4 days. The remainder of the cycles ran for 2 days each. At the end of cycle 66, biomass had covered the $\frac{3}{4}$ of the surface of the foam. Most of the biomass observed was present at the bottom of the foam and gradually decreased towards the top.

Figure 4.28 shows the overall performance of K3 starting from cycle 32. Average influent concentration of LAS was 284 mg/L as TOC and 435 mg/L as MBAS. In cycle 32, TOC removal was 65.6% and MBAS removal was 77.8%, corresponding to effluent TOC and MBAS concentration of 96.8 and 45.8 mg/L, respectively. Over the long-term operation, performance gradually improved. By the end of cycle 66, the removal improved to 80.5% for TOC and 95.8 % for MBAS. The average removal within the 34 cycles during which pH was controlled by NaOH addition was 74.6% for TOC and 96.7% for MBAS (corresponding to average effluent TOC and MBAS concentrations of 65 and 12 mg/L respectively). Influent and effluent total organic carbon (TOC) and methylene blue active substances (MBAS) concentrations for every cycle are shown in table format in Appendix 4.

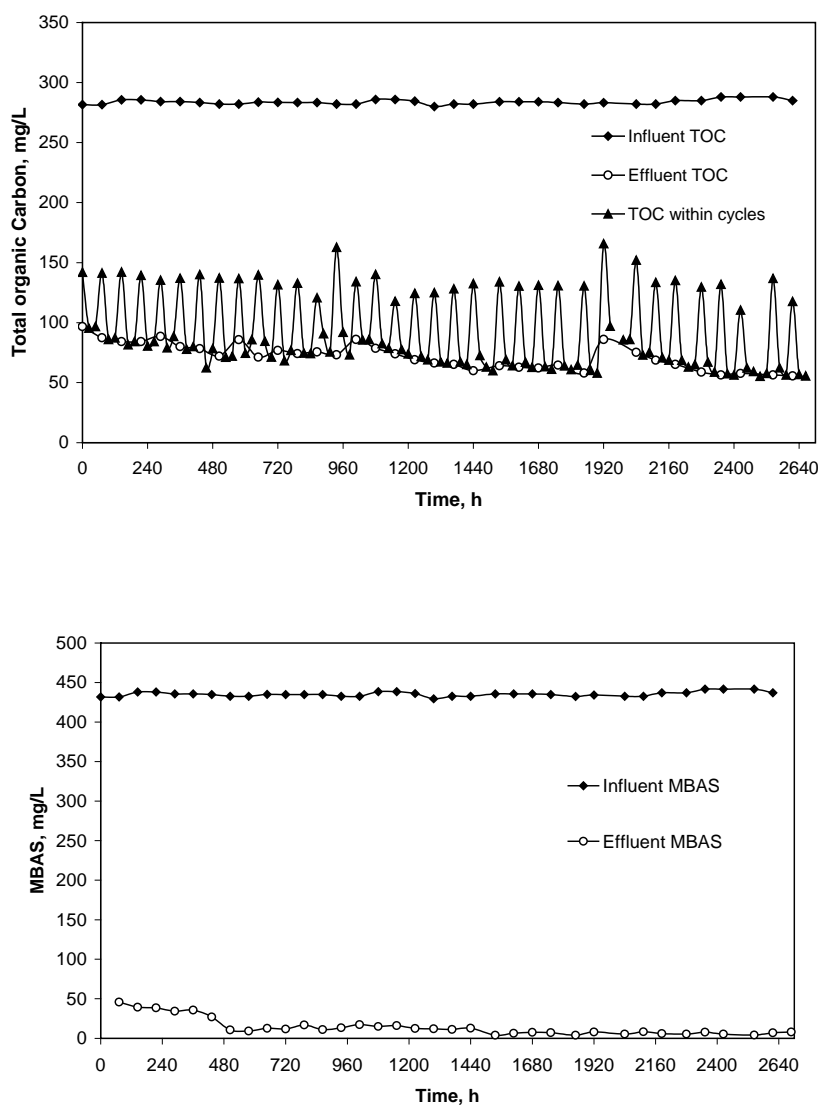


Figure 4.28: TOC concentrations in the influent, effluent, and within cycles (top), and influent-effluent MBAS concentration (bottom) during SBBR operation.

As shown in Figure 4.28 (top), during the last 34 cycles of operation, of the TOC that was removed from solution, approximately 81% was removed during the first 30 minutes after a new cycle was initiated. 89% was removed after 2 hours, and 99% by the end of the fifth hour. After the fifth hour, TOC concentrations in the reactor remained almost constant until the end of the React portion of the cycle. It is interesting to note, however, that a further decrease in concentration in MBAS took place even when the TOC remained constant. During cycles 52,

56, and 58, the duration of the React period was extended for 24 hours (to a total cycle length of 3 days (72 hours)) in order to determine whether MBAS would continue to decrease.

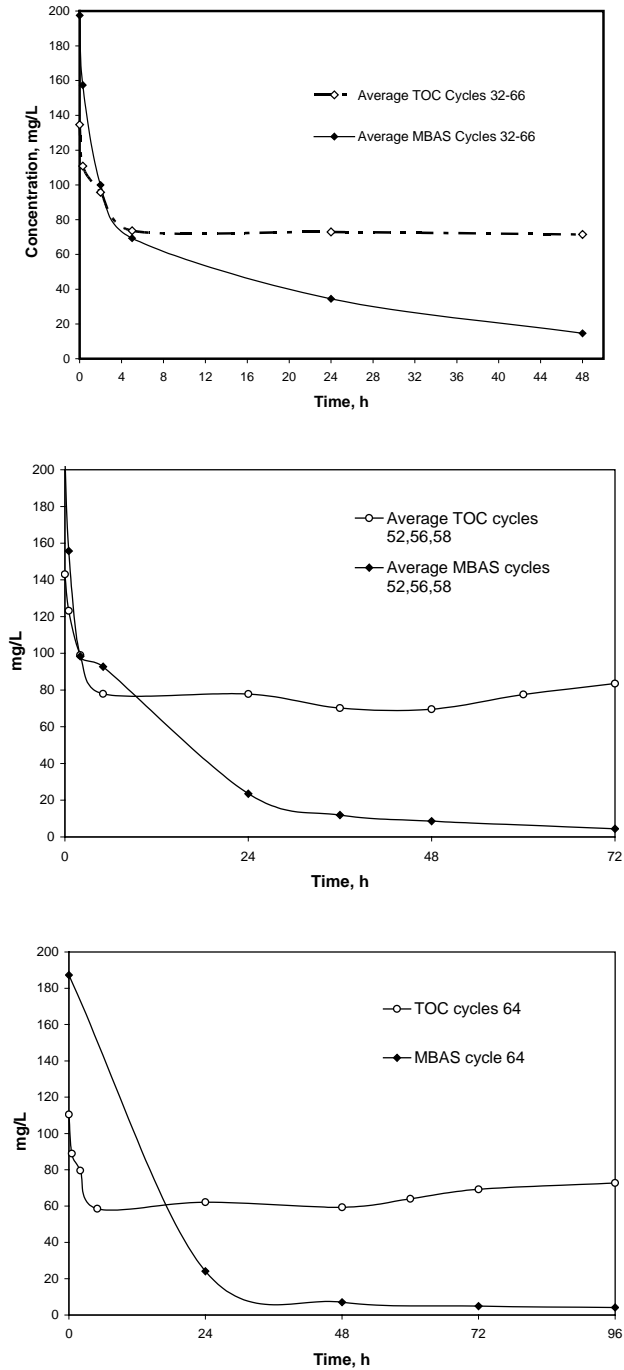


Figure 4.29: Average TOC and MBAS values within two days in SBBR (top), three days (middle), and four days (bottom)

During cycle 64, the cycle length was extended to a total of 96 hours (4 days) for the same reasons. As shown in Figure 4.29, MBAS concentrations did not appreciably decrease further when the cycle length was extended to 72 hours (middle) or 96 hours (bottom).

This rapid removal of TOC lead to perform sorption test using the polyurethane foam to verify its participation during the LAS disappearing. Results are showed and explained in further sections.

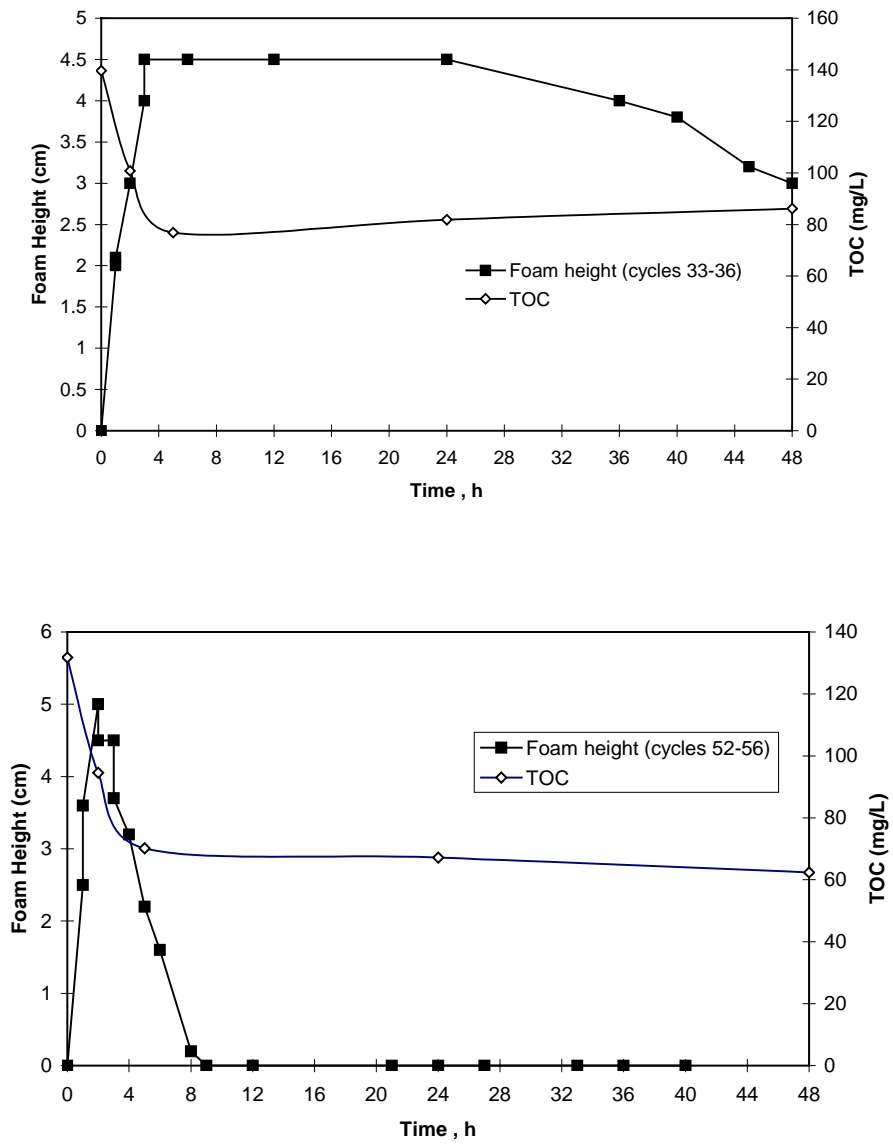


Figure 4.30: Comparison of foam production during cycles 33-36 (top) and 52-56 (bottom)

Foam production in the SBBR (i.e., K3) was measured during two periods. The first period was from cycles 33 thru 36, shortly after the start of pH regulation using NaOH. The second period of foam measurement was during cycles 52 thru 56. As shown in the top graph of Figure 4.30, during the first interval of foam measurements (cycles 33 thru 36), the foam depth increased to approximately 4.5 cm after 4 hours into the React period, remained relatively constant for the next 20 hours, and the subsequently decreased to approximately 3 cm at the end of the React period. During the second interval during which foam was measured (cycles 52 through 56), a markedly different trend was observed. Foam was only transiently produced during the first eight hours of the React period. As in the SBR, the decrease in foam production coincided with removal of TOC (see Figure 4.30).

Because the majority of biomass in K3 grew attached to the packing media and the packing medium would not fit into the chamber used for OUR measurements, OUR was not measured in K3. Instead, the only oxygen measurements collected for K3 was periodic measurement of dissolved oxygen in the bulk liquid to verify that the aeration supply was maintaining a minimum D.O. of 2 mg/L.

4.3.2 Reactor Sorption Measurements

4.3.2.1 Sorption of LAS in the SBR (K1)

Previous studies reported in the literature regarding removal of LAS in full-scale treatment plants (Madsen *et al.*, 1999), and specialized test using activated sludge (Mösche *et al.* 2002, Rittmann *et al.*, 2001), suggest that sorption of LAS to biomass can play an important role in the overall removal of LAS. To quantify the extent of LAS sorption to biomass contained in the SBR (K1), mass balances based on TOC were performed for eight different cycles as described in Section 3.4.4. Values for individual trials are presented in Table 4.9. The percentage difference between the measured concentration of soluble TOC at the end of the fill

period and the concentration of soluble TOC calculated based on a mass balance (i.e., using the measured TOC of the synthetic wastewater entering the reactor, the measured residual soluble TOC remaining in the reactor after the end of the previous cycle, and the volume of each) averaged 20.9%. The TOC that was “missing” was assumed to be sorbed to biomass.

Table 4.9: LAS sorption in the SBR.

Cycle	TSS	Feed		Reactor (end of previous cycle)			Calculated concentration (after mixing)			Measured concentration (after mixing)			Sorbed	%
		TOC, mg/L	Volume, L	TOC, mg/L	Volume, L	TOC, mg	TOC, mg/L	Volume, L	TOC, mg	TOC, mg/L	Volume, L	TOC, mg		
16	770	286.5	1.25	89.7	1.25	112.13	188.10	250	470.25	155.1	25	387.75	82.50	17.54
17	777.5	282.1	1.25	87.5	1.25	109.38	184.80	250	462.00	143.3	25	388.25	103.75	22.46
18	782.5	280.4	1.25	78	1.25	97.50	179.20	250	448.00	132.5	25	381.25	116.75	26.06
19	785	272.9	1.25	78.5	1.25	98.13	175.70	250	439.25	123.5	25	387.75	130.50	29.71
20	792.5	286.4	1.25	77.6	1.25	97.00	182.00	250	455.00	157.3	25	388.25	61.75	13.57
21	802	285.4	1.25	81.12	1.25	101.40	183.26	250	458.15	152	25	380.00	78.15	17.06
22	817.5	282.1	1.25	82.19	1.25	102.74	182.15	250	455.36	145.4	25	363.50	91.86	20.17

To further assess the role of biomass (measured in terms of TSS) in the SBR on sorption of LAS, data summarized in Table 4.9 were fit to Freundlich (top) and Langmuir (bottom) isotherm models. As shown in Figure 4.31, the experimental data could not be well-described by either of these models. The poor model fit may be due to the fact that the measured TSS concentration may not have been representative of the biomass content inside the reactor. As mentioned in section 4.3.1.1, foaming and attachment of biomass on tubing and aeration stones made accurate measurement of TSS difficult. In spite of the poor fit of the Freundlich and Langmuir isotherm models, data presented in Table 4.9, suggest that sorption likely played a role in TOC removal during the period immediately after Fill. It should be noted, however, that surfactant sorbed at the end of the Fill period may have subsequently desorbed and biodegraded during the subsequent React period when biodegradation decreased the concentration of LAS in the bulk liquid, thereby creating a driving force for desorption.

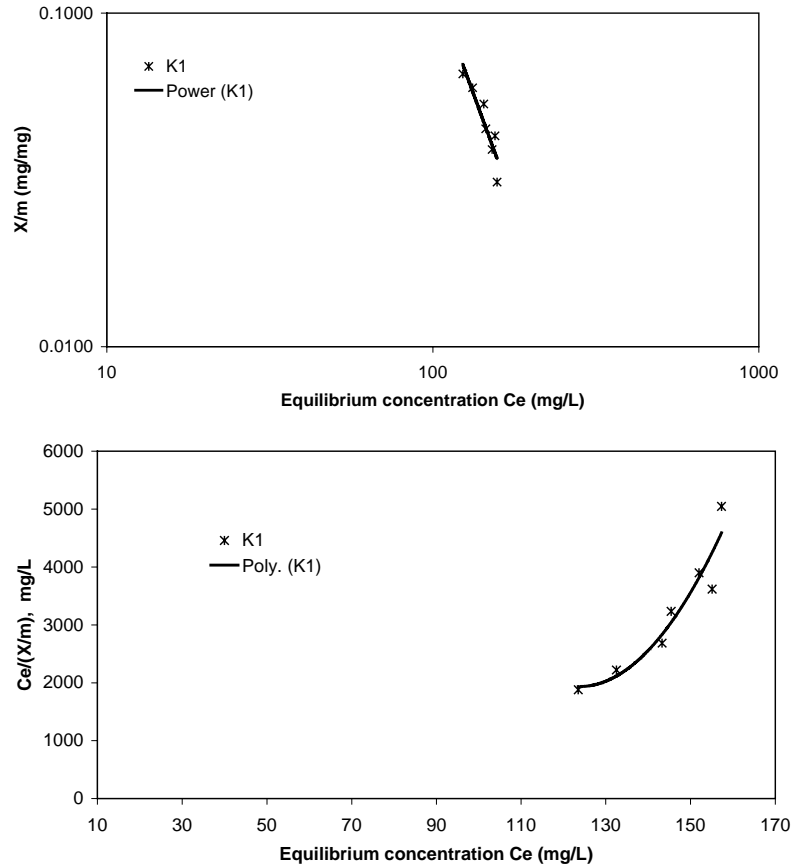


Figure 4.31: Freundlich adsorption isotherm for LAS (top) and Langmuir isotherm (bottom) sorption on to K1 biomass (measured as TOC)

4.3.2.2 Sorption of LAS in the ICEAS (K2)

Due to the continuous feed characteristic of the ICEAS, the approach used to quantify sorption of LAS in the SBR could not be performed in the ICEAS. Instead, a batch sorption test using biomass collected from K2 at the end of cycle 22 was performed in an attempt to assess the extent of LAS sorption. Results from this test appear in Appendix 8. As described in the appendix, results from this test were inconclusive.

4.3.2.3 Sorption of LAS in the SBBR (K3)

Using the same procedure that was used to assess sorption in the K1 reactor (described in section 3.4.4), sixteen sorption measurements were performed in the SBBR. Data for individual tests are presented in Table 4.10. On average, of 26.7% of the LAS (measured as TOC) could

not be accounted for right after the FILL period was finished. Although this provided a general indication that sorption was likely playing at least a temporary role in TOC removal, a more complete analysis of these data using Freundlich or Langmuir isotherm models is not possible because the mass of biomass attached to the polyurethane foam packing medium was not measured.

The slightly higher TOC removal during the Fill period in the SBBR (K3) compared to the SBR (K1) may have been due to the fact that there was a larger quantity of biomass in the SBBR, or it may have been due to LAS adsorption by the polyurethane foam packing medium.

Table 4.10: Percentages of LAS sorbed in the SBBR based on calculation on mass balance

Cycle	Feed		Reactor (end of previous cycle)		Calculated concentration (after mixing)			Measured concentration (after mixing)			Sorbed, mg	%	
	TOC, mg/L	Volume, L	TOC, mg/L	Volume, L	TOC, mg	TOC, mg/L	Volume, L	TOC, mg	TOC, mg/L	Volume, L			TOC, mg
46	282.1	1.25	73.20	1.25	91.50	177.65	2.50	444.13	134.20	2.50	335.50	108.63	24.46
47	285.9	1.25	86.06	1.25	107.58	185.98	2.50	464.95	140.27	2.50	350.68	114.28	24.58
48	285.9	1.25	78.64	1.25	98.30	182.27	2.50	455.68	117.90	2.50	294.75	160.93	35.32
49	284.4	1.25	74.04	1.25	92.55	179.22	2.50	448.05	124.37	2.50	310.93	137.13	30.60
50	280.0	1.25	69.01	1.25	86.26	174.51	2.50	436.26	125.00	2.50	312.50	123.76	28.37
51	282.1	1.25	66.42	1.25	83.03	174.26	2.50	435.65	128.22	2.50	320.55	115.10	26.42
52	282.1	1.25	65.21	1.25	81.51	173.66	2.50	434.14	132.60	2.50	331.50	102.64	23.64
53	284.0	1.25	59.89	1.25	74.86	171.95	2.50	429.86	133.91	2.50	334.78	95.09	22.12
54	284.0	1.25	64.07	1.25	80.09	174.04	2.50	435.09	130.55	2.50	326.38	108.71	24.99
55	284.0	1.25	62.79	1.25	78.4875	173.40	2.50	433.49	131.06	2.50	327.65	105.84	24.42
56	283.4	1.25	61.23	1.25	76.5375	172.32	2.50	430.79	130.80	2.50	327.00	103.79	24.09
62	285.0	1.25	65.17	1.25	81.46	175.09	2.50	437.71	129.63	2.50	324.08	113.64	25.96
63	288.0	1.25	58.86	1.25	73.58	173.43	2.50	433.58	131.86	2.50	329.65	103.93	23.97
64	288.0	1.25	56.45	1.25	70.56	172.23	2.50	430.56	110.43	2.50	276.08	154.49	35.88
65	288.0	1.25	57.70	1.25	72.13	172.85	2.50	432.13	136.80	2.50	342.00	90.13	20.86
66	285.0	1.25	56.41	1.25	70.51	170.71	2.50	426.76	117.60	2.50	294.00	132.76	31.11

To assess whether sorption to the polyurethane foam packing medium may have occurred, an isotherm experiment was conducted using virgin foam (i.e., foam that had not been placed in the bioreactor) as described in Section 3.4.5.4. Figure 4.32 depicts the Freundlich adsorption isotherm for LAS (measured as TOC) built from results obtained during the batch sorption test. Freundlich constants (K_f and $1/n$) for the polyurethane foam tested were calculated along with the correlation coefficient. These values are presented in Table 4.11.

From the Freundlich adsorption isotherm built from the batch sorption tests, it was found that mass of TOC adsorbed per unit mass of foam increased as the concentration of TOC in solution increased. The model fit the experimental data quite well ($R^2 = 0.9879$).

Table 4.11: Summary of Freundlich parameters for LAS (as TOC) sorption to polyurethane foam.

Media Tested	K_f	$1/n$	R^2
Polyurethane foam (Zander)	9 E -10	3.2928	0.9879

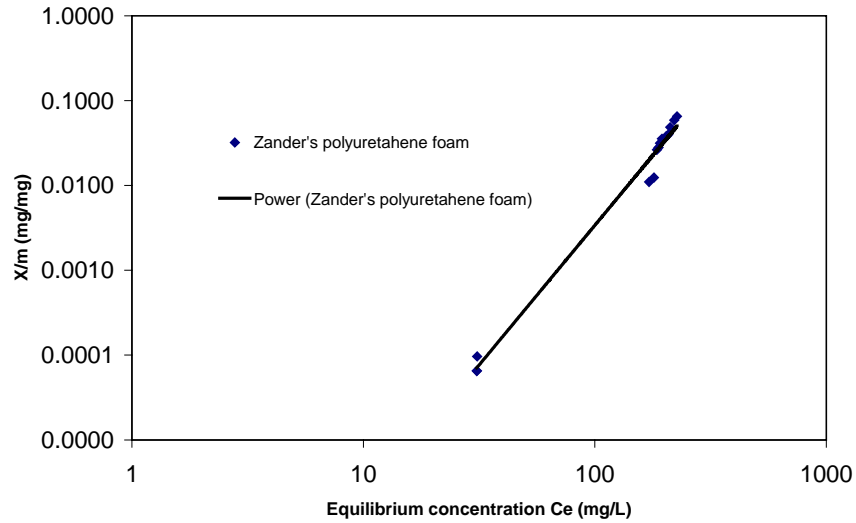


Figure 4.32: Freundlich adsorption isotherm for LAS (measured as TOC) sorption on to Zander polyurethane foam.

The Freundlich adsorption isotherm was used to calculate the mass sorbed under two different equilibrium concentrations. The first concentration is that from the end of the previous cycle (an average of 65.95 mg/L from Table 4.10). The second is the measured equilibrium concentration at the end of Fill (after mixing) which is 128.45 mg/L. Subtracting the second value from the first one will account for the mass of TOC sorbed. Then using the known quantity of polyurethane foam inside the reactor, 12,000 mg, the mass of TOC that can be sorbed was calculated to be 84.3 mg. This corresponds to 19% of the total TOC inside the reactor. This value is less than the overall average 26.7% from data shown in Table 4.8. This result lead to the conclusion that at the moment of sampling (just right after the fill period finished), the amount of

TOC “missing” was mainly sorbed by the biomass in the reactor and less likely onto the polyurethane foam. It can be concluded that the overall removal of LAS in reactor K3 was produced by a combination of sorption and biodegradation, but the contribution of each is difficult to ascertain.

4.3.3 Special Tests

4.3.3.1 Variation in Hydraulic Retention Time (HRT) and the Effect on LAS Degradation and Foam Production

During cycles 23 and 26, tests were conducted in the SBR (K1) to assess the role of HRT on bioreactor performance. During cycles 23 and 26, the HRTs were adjusted to 6.25 and 12.4 days, respectively, by changing the fraction of the reactor volume that was replaced during the fill and draw procedure (see section 3.4.5.1). During the test using a 6.25-day HRT, the LAS concentration measured as MBAS at the end of the FILL period was 292 mg/L compared to 192 mg/L average from cycles 16 to 22 when the HRT was 10 days (Appendix 2). The higher LAS concentration, expected because a larger fraction of the reactor volume was decanted and filled at the start of the cycle, caused a greater amount of foam production. To control the foaming to avoid overflowing the reactor, the aeration cycle had to be changed. Instead of supply air via the diffuser stones for 30 seconds every 10 minutes, the solenoid valve was programmed to open only 10 seconds every 10 minutes for the remaining time of the cycle. As a consequence, lower values of dissolved oxygen (2.3 to 3.5 mg/L) were measured in the reactor compared to those measured during a regular 10-day HRT cycle (3.5 – 6.5 mg/L). A constant foam height of 15 cm was measured during the entire length of the cycle. At the conclusion of the cycle (five days total cycle length), the MBAS concentration had decreased to only 105 mg/L (compared to 27 mg/L average from cycles 16 to 22 when the HRT was 10 days). The original aeration cycle was restored once the LAS concentration in the reactor was decreased by adjusting the LAS

concentration in the reactor adding de-ionized water prior to the start of cycle 24 during with the HRT was returned to 10 days.

During cycle 26, when the 12.4-day HRT test was performed in the SBR (K1), the MBAS concentration measured in the reactor at the end of the Fill period was 162.2 mg/L. This concentration, lower than that observed during experiments with the 10-day HRT, is consistent with the fact that a smaller fraction of the reactor volume was decanted and filled. With a 12.5-hour HRT, there was no appreciable difference in foam production or MBAS removal compared to previous cycles with an HRT of 10-days.

A different scenario was observed for the ICEAS (K2). With both 6.25 and 12.4 day HRTs, the overall performance of the reactor did not change appreciably. The only noticeable difference took place during the 6.25 day HRT test. Foam production at the beginning of the cycle was higher due the LAS influent accumulation at the bottom of the reactor. The lower volume of liquid at the beginning of the cycle (0.5L) compared to the regular 1.25 L produced a higher LAS concentration due to a lower dilution factor. The biomass concentration; however, was increased. Normal foam levels were reached after two hours. The 12.4 HRT test did not produce any significant difference in MBAS concentration along the cycle and foam production. The SBBR (K3) exhibited similar behavior to the SBR (K1) during these tests when the HRT was adjusted to either 6.25 or 12.4 days. The aeration interval had to be changed to avoid foam overflowing the reactor during the 6.25 HRT test. After three hours the aeration cycle was restored due to decrease of foam production. Final values of LAS were the same as in the end of regular cycles.

4.3.3.2 Transient Loading Experiments in ICEAS

Transient-loading experiments were conducted in the ICEAS (K2) during cycles 12, 17, and 20. During the transient loading condition, the influent LAS concentration was increased to

two times that of the normal loading (approximately 800 mg/L LAS during transient loading vs. approximately 400 mg/L during “normal” loading). The higher influent LAS concentration wastewater was pumped into the reactor for the entire cycle length during cycles 12 and 17, and for three days of the 5-day cycle during cycle 20. The influent LAS concentration during these tests, measured as TOC, corresponded to 567, 571, and 563 mg/L for cycles 12, 17, and 20, respectively.

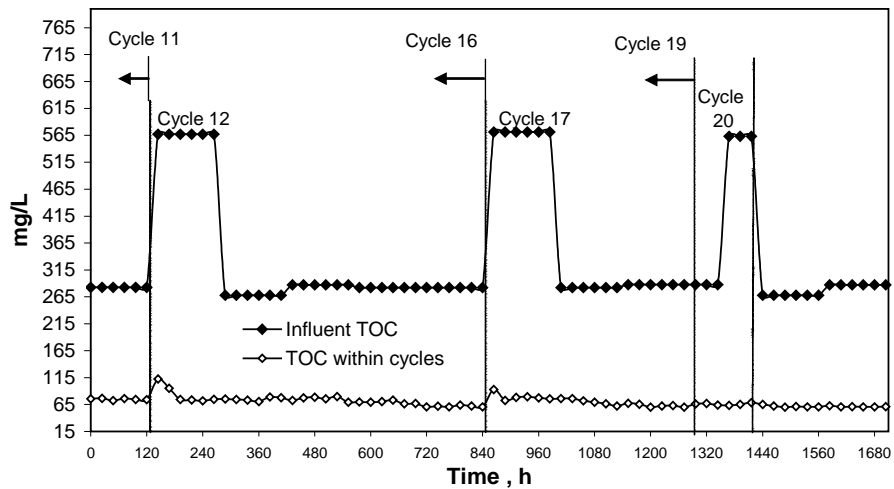


Figure 4.33: Overall behavior of ICEAS during transient loading experiments. Time zero is the start of the React period during cycle 11

Figure 4.33 depicts the performance of the ICEAS in terms of TOC in the influent and TOC within cycles. The effect of the high LAS influent concentration can be observed during cycles 12 and 17. Accumulation of influent LAS (measured as TOC) in the reactor during SETTLE and DRAW stages increased the overall LAS concentration in the reactor at the start of the Fill/React period. Average TOC concentrations before the transient loading began (i.e., at the start of the settle period) were 74.8 mg/L during cycle 11, and 61.7 mg/L in cycle 16. Once the reactor started to be fed with the higher concentration of LAS in the influent solution, the TOC concentrations increased up to maximum concentrations of 112.4 mg/L in cycle 12 and 92.6 mg/L in cycle 17 (measured at the beginning of the React period on cycles 12 and 17).

However, TOC concentrations decreased and remained almost constant after 48 hours in both experiments.

Foam height during the first day of the REACT period during cycles 12 and 17 ranged from 8 to 10 cm (see Figure 4.34). During the second day, foam production decreased, and by the third day, no measurable foam was present. Although foam height was considerable at the beginning of cycles 12 and 17, continuous aeration was maintained in the reactor during the entire cycle length.

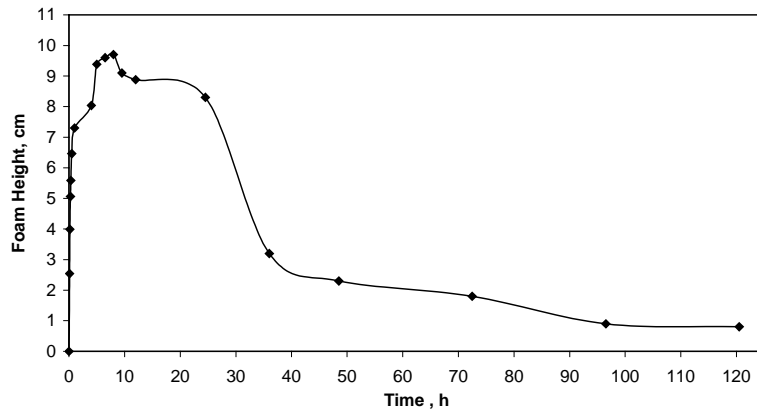


Figure 4.34: Average foam height measured during transient loading experiments in cycles 12 and 17 in the ICEAS.

During the third transient loading experiment in cycle 20, the higher loading rate condition was initiated after three days of normal operation and lasted for two days, until the end of the cycle. Because accumulation did not take place during the unaerated SETTLE and DRAW periods as in the previous transient loading tests, the reactor's operation was not affected as, can be seen in Figure 4.33. It should be noted, however, that a constant 0.5 cm foam was present for the remaining duration of the test.

4.3.3.3 Addition of Anti-foaming Agents in the SBR (K1) and SBBR (K3)

Two experiments in the SBR (K1) were conducted using anti-foam agent Callaway 3142. In the first experiment (cycle 32), antifoaming agent was added to the feed to make up a

concentration of 25 mg/L in the reactor at the end of Fill. During the second experiment (cycle 33), the desired anti-foaming agent concentration in the reactor was 50 mg/L. Aeration was constant during the entire cycle.

Noticeable reduction in foam production was obtained during the first experiment. As can be seen in Figure 4.35, even though aeration was supplied on a continuous basis, almost a 50% reduction in foam was achieved compared to foam measurements from cycles where anti-foam was not added. The LAS removal was 77.7% as TOC and 93.7% as MBAS during this experiment. These values are similar to values obtained during regular SBR operation.

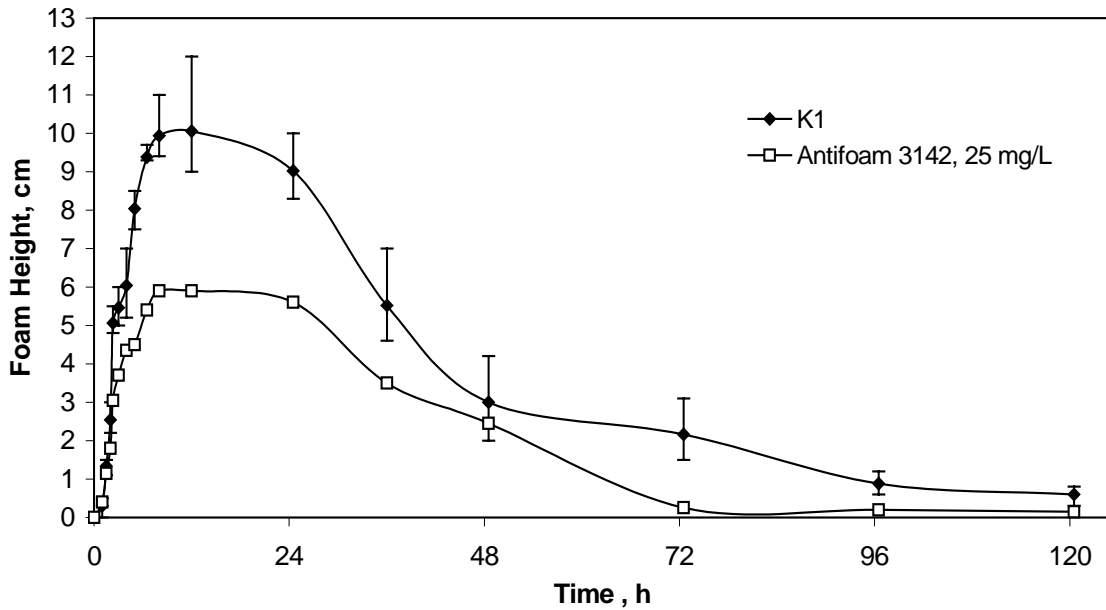


Figure 4.35: Comparison between foam height measured during regular cycles and using anti-foaming agent Callaway 3142 at 25 mg/L in cycle 32.

Foam produced during the 25 mg/L antifoaming test was different in appearance. The foam appeared to be not as compact and light as the one present during regular cycles. It tended to collapse as it was rising, and as a consequence, build up was not possible.

Reduction of foam was more dramatic in the experiment using 50 mg/L of antifoam agent. A white precipitate was present at the surface of the liquid a few minutes after aeration

was started in the reactor. Foam was similar in appearance as mentioned above, but in less quantity. Higher values of foam reached between 2.5 and 3.0 cm. No measurable foam was present by the second day of the cycle.

MBAS analysis showed an LAS concentration of 158 mg/L at the beginning of the cycle. This value is low compared to regular values from previous and following cycles (cycles 30 to 37, Appendix 3), which ranged between 175 and 208 mg/L. Although LAS concentration measured as MBAS was low, TOC measurements did not show any significant difference as can be seen in Figure 4.36. Also, overall LAS removal as TOC and MBAS were close to the ones from previous and following cycles, 75.9% and 93.6% respectively.

Experiments using anti-foam agent 3142 were conducted in the SBBR (K3) during cycles 45 and 58. The influent synthetic wastewater was amended with 25 mg/L of antifoaming agent in cycle 45, and 50 mg/L during cycle 58. Continuous aeration could be maintained during the entire cycle in both experiments due to low production of foam. Foam production was the same in both experiments. Even when the concentration of antifoaming agent was double in cycle 58, the recordings of foam height were similar to those observed during cycle 45. The maximum foam height recorded, 6 cm, took place 5 minutes after the start of the cycle. Visual observation of the foam produced in the reactor revealed that the foam had similar physical characteristics presented during experiments performed in the SBR with anti-foam chemicals.

The 5 cm foam height was almost constant during two hours due to collapsing of the foam as it rose. After two hours, no measurable foam was produced in the reactor. TOC and MBAS concentrations did not show any remarkable difference from others recorded from previous and following tests as shown in Figure 4.37. Partial sorption of the antifoaming agent either onto the polyurethane foam or biomass is one potential explanation for why both experiments showed similar outcomes in terms of foam production.

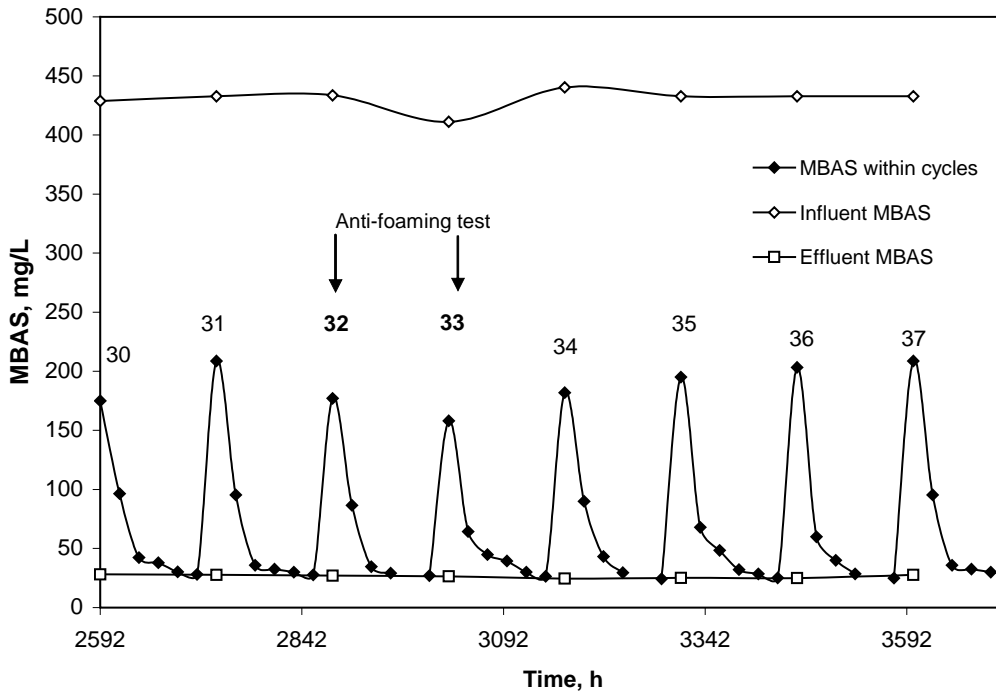
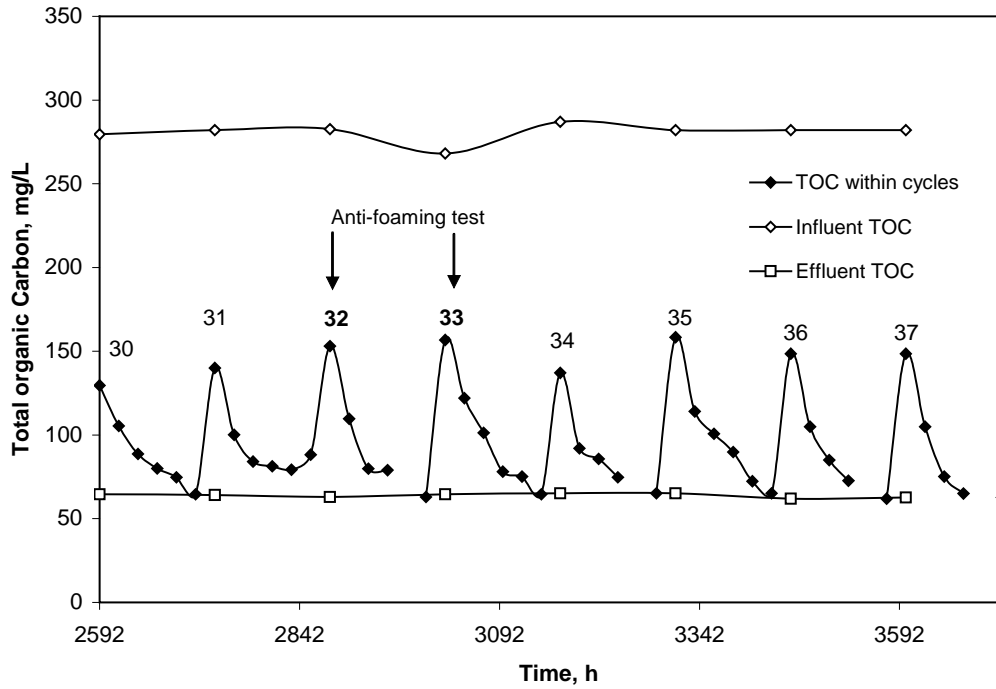


Figure 4.36. TOC (top) and MBAS (bottom) concentrations from cycles 30 to 37 in the SBR. Anti-foaming agent 3142 was tested in cycles 32 (25mg/L) and 33(50 mg/L) in the SBR (K1)

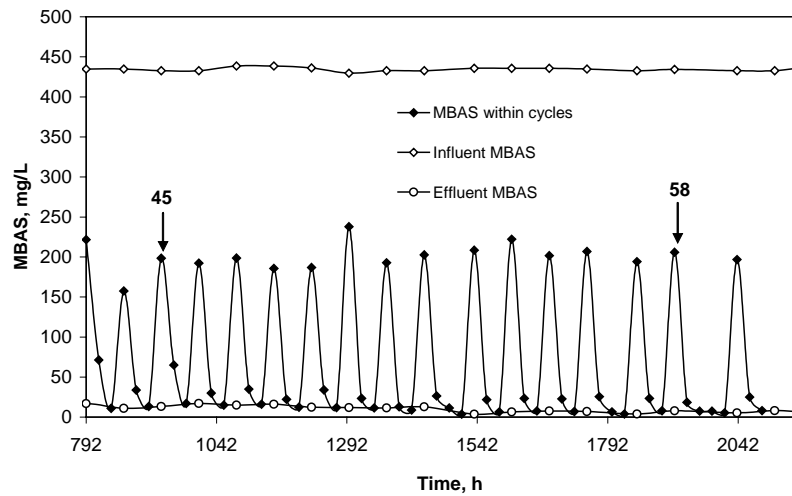
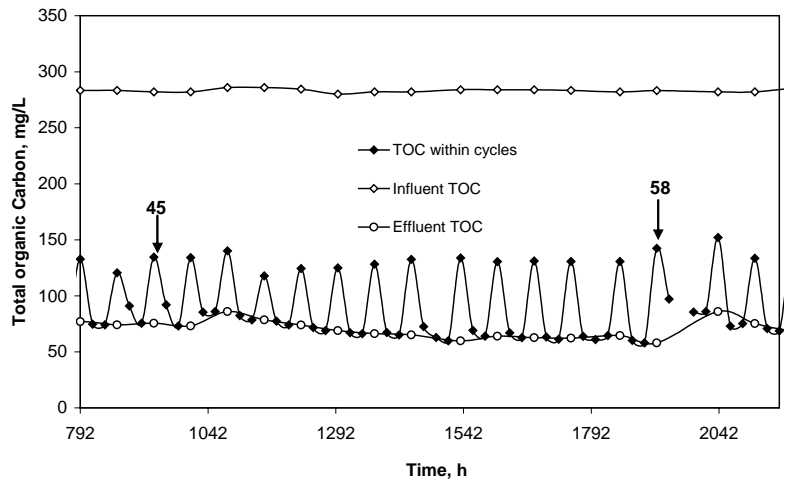


Figure 4.37. TOC and MBAS concentrations from cycles 43 to 59 in the SBBR. Anti-foaming agent 3142 was tested in cycles 45 (25mg/L) and 58 (50 mg/L)

Even when the quantity of foam was decreased by using antifoaming agents in the K1 and K3 reactors, foam production was still a problem. Biomass was still been expelled from solution due the foaming in K1, and special care was taken in the feed preparation so the concentration of LAS would not be high so foam overflow could occur due the continuous aeration. Furthermore, there is the restriction in using low concentrations of anti-foaming agent due precipitation of the LAS. High concentrations of anti-foaming agents will control foam production, but a true biological removal process will not be taking place.

CHAPTER 5 OVERALL CONCLUSIONS AND RECOMMENDATIONS FOR FUTURE RESEARCH

Development of a culture capable of using LAS in high concentrations was accomplished using the respirometer. This issue was very important because during literature investigation there was a lack of information regarding degradation of LAS in concentration greater than 10 mg/L. Analysis using first-order kinetics revealed that average half-lives were 12.1 days (from TOC data) and 3.1 days (from MBAS data) for a solution containing an initial LAS concentration of 500 mg/L seeded with a biomass concentration of 100 mg/L TSS. The average half-life of 3.1 days calculated from MBAS is close to values reported by Nielsen *et al.* (1997) for LAS in low concentrations from a Porous Pot Biodegradation Test System.

For laboratory scale reactors treating a synthetic wastewater containing 400 mg/L LAS, removal was achieved in all three of the process configurations tested (SBR, ICEAS, and SBBR). Results from this research showed that the SBR produced an effluent with an average of 75 mg/L in TOC and 32 mg/L as MBAS from an influent containing an average of 282 mg/L TOC and 430 mg/L as MBAS. Although results indicate that the SBR operating cycle reported in this thesis is a feasible process for treating wastewater with high concentrations of LAS, excessive foam production was exhibited for the system when a short Fill period was utilized. This resulted from the fact that high LAS concentrations were present in the reactor at the start of the React period. Similarly, excessive foam production was also observed in the SBBR when a short Fill period was utilized. For full scale implementation, it is anticipated that continuous aeration will not be possible and that controllable addition of antifoaming agents will be necessary in cases where short Fill periods or very high influent LAS concentrations result in high LAS concentrations in the reactor at the start of aeration.

In terms of foam production, the ICEAS showed the best performance in treating an influent with high concentrations in LAS. Minimal foaming was observed even when continuous

aeration was supplied even during transient loading tests where the influent wastewater contained LAS concentrations of 800 mg/L. Treated effluent from the ICEAS had an average of 60 mg/L of TOC and 20 mg/L as MBAS.

Of the three systems evaluated in the research reported in this thesis, the SBBR (i.e., K3) consistently produced the lowest TOC and MBAS concentrations in the effluent after a pH control strategy was implemented. Effluent TOC and MBAS concentrations produced by this system had an average of 60 mg/L TOC and 7 mg/L as MBAS. Furthermore, the SBBR accomplished this treatment with a cycle time of only two days (HRT of 4 days) compared to a cycle length of five days (HRT of 10 days) in the ICEAS and SBR.

The removal of LAS achieved in the SBR and ICEAS was concluded to be predominantly due to biodegradation. Oxygen Uptake Rates measured during the operational cycles in both the SBR and ICEAS verified microbial activity in the presence of LAS. TOC and MBAS measurements support the fact that LAS was being consumed while OUR was taking place. The activated sludge developed in the reactors was able to use LAS as sole source of carbon.

Because biomass in the SBBR grew attached to solid packing medium, OUR was not measured using the procedure used to confirm biodegradation as in the case of the SBR and ICEAS. Nevertheless, experimental data support the notion that biodegradation was the main cause for LAS removal in the SBBR system based on the following facts: 1) the mass of LAS removed during the 66 operational cycles was quite large compared to the expected abiotic sorption capacity of the polyurethane foam packing medium or biomass; 2) production of a considerable amount of biomass growth could be visually observed over time; 3) there was a need for continuous pH regulation, a phenomenon expected from production of acidic intermediates reported by other researchers, and 4) the dissolved oxygen measured in the reactor

vessel was much lower than saturation indicating that it was being consumed by the microbial population. Although the amount of biomass in the SBBR was not measured at the end of the test, a crude estimate based on visual observation indicates that it was at least two times more than that present in the ICEAS. The higher amount of biomass present in K3 could be the explanation why the LAS removal in K3 occurred faster compared to K1 and K2 when treating a wastewater with the same LAS concentration.

Based on the number of cycles performed and the shorter cycle length, the SBBR removed approximately three times more LAS than the ICEAS and two times more than the SBR. If sorption onto the polyurethane foam was the main cause of the removal, it have to be assumed that it had a much large sorption capacity than was determined from batch sorption tests performed using virgin foam.

The need for almost continuous pH regulation in the SBBR (K3) can be explained by the results obtained by Perez *et al.*, 1996. In their study, it was established that LAS influenced the pH self-regulation capacity in aerobic degradation of organic matter when its concentration is greater than 20 mg/L and external neutralization was required. The drop in pH was attributed to the production of acidic intermediates as a result of LAS degradation following the degradation pathway proposed by Swisher (1987). This last observation provides further credibility to the notion that biodegradation rather than sorption was primarily responsible for to LAS removal in K3 (i.e., the SBBR).

Complete removal of LAS was not achieved in either the SBR or ICEAS. There was an average of 20% of the incoming TOC and 6% of the influent MBAS present at the end of the last cycles performed in each system. From the data collected, it cannot be determined whether the remaining TOC consisted of partially transformed degradation products (i.e., intermediates that

could not be used as carbon source by the microorganisms present), remaining LAS homologues that could not be transformed by the microbial population, or if the residual TOC was comprised of relatively inert soluble microbial products (i.e., dead biomass). The fact that the concentration of MBAS was not zero suggests that residual surfactant accounted for at least a portion of the residual TOC.

Foaming was of particular concern during the operation of the reactors. The ICEAS was the only system capable of operating under continuous aeration during its REACT period. Implementation of an intermittent aeration strategy in the SBR and SBBR was necessary to avoid excessive foaming due to the presence of high concentrations of surfactant at the start of the REACT period. Using anti-foaming agents made it possible to aerate the reactors continuously;; however, it did not completely eliminate the foaming problem. The amount of anti-foaming agent was limited to 25 mg/L because higher concentrations precipitate the LAS in solution making it unavailable to bacteria. There was no significant difference in LAS removal between cycles aerated continuously and those under the intermittent aeration strategy.

The SVI and TSS content on the clarified effluent from the SBR and ICEAS showed that even when the ICEAS had a better performance in LAS removal and foam production, activated sludge in this system did not have as good settling capacity as the SBR. Also, visual observations from each of the activated sludge revealed differences in the color and physical appearance of the activated sludge. Activated sludge in the SBR had a light brown color with a granular appearance while activated sludge from the ICEAS presented a light yellow color and fluffy appearance. These characteristics suggest that there were at least some differences in the microbial communities developed in each of the systems. If this is the case, it can be said that different operation strategies lead to development of different bacterial cultures even when the same departure activated sludge is used.

Based on the results from this research, a system consisting of an SBR or SBBR with a long aerated Fill period would likely to be the best alternative in a full-scale treatment plant to treat an influent wastewater containing high concentrations of LAS. It would venture to speculate that an aerated Fill period (i.e., 3 days) instead of the short static fill performed, followed by a shorter React period (i.e., two days) would avoid high LAS concentrations and will not produced excessive foaming. Furthermore, using an SBR or SBBR would not present the disadvantage observed in the ICEAS of having accumulation influent with high concentrations of surfactant during the SETTLE and DECANT/DRAW periods that caused production of foam as soon the aeration was started. Additional experiments could be conducted to verify the above stated.

Although the research reported in this thesis demonstrated that biological treatment is a technically feasible option for treating wastewaters containing high concentrations of LAS, that does not mean that it would be the most cost effective alternative for full-scale systems. It is recommended that future research efforts include an economic analysis to determine if biological treatment would offer a cost advantage in comparison to the DAF-polymer treatment used at the industrial facility in Honduras.

From a research point of view, characterization of the microbial population in each of the reactors would be of academic interest because it would allow further insights into whether the various operating strategies influenced the composition of the microbial community structure. Denaturant Gradient Gel Electrophoresis (DGGE) on the activated sludge from the SBR, SBBR, and ICEAS will show if the populations are the same or different due to the selection and enrichment strategies.

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APPENDIX 1: TESTING ANTI-FOAMING AGENTS

A.1 Objective

Compare 12 different antifoaming agents (laboratory preparations from Vulcan Performance Chemicals, Columbus, GA). Different concentrations of each anti-foaming agent were tested to measure foam production using a 200 mg/L LAS solution.

A.2 Materials and Methods

A.2.1 LAS Stock Solution.

2 liters of LAS (Aldrich D-2525) stock solution containing 4.35 mg-LAS/mL was used to prepare the working solutions.

A.2.2 Working Solutions

The working solutions were prepared pipetting 23 mL of LAS stock solution into a 500 mL flask. The final LAS concentration was 200 mg/L.

The amounts of antifoaming agents were weighed in a small beaker on an analytical scale with 0.0001 g precision. The beaker containing the antifoaming agent was washed with 10 mL of LAS stock solution and transferred into the 500 mL flask, then the rest 13 mL of stock solution were added and then the volume completed to 500 mL. The beaker was washed with stock solution due that some of the testing antifoaming agents were soluble in water only when the LAS was present. Every antifoaming agent tested was added into different working solutions to make up concentrations ranging from 10 to 2000 mg/L.

A.2.3 Procedure

The testing procedure consisted of placing 100 mL of the wastewater containing the anti-foaming agent in a 1000-mL glass cylinder. The cylinder was graduated in centimeter scale using

a ruler and masking tape. A peristaltic pump (Cole Palmer Materflex® console drive model 7521-40, Bernant Co, Barrington, IL) was used to provide a continuous flow of air at a flow rate of 100 mL/min as measured by a Cole-Palmer Rotameter (Gilmont Instruments, 250 mL/min scale Accucal flow meter Cole-Palmer Instruments Co., Vernon Hills, IL). Air flow was delivered continuously through an aeration stone (Fisher Scientific, Sewanee, GA) connected to ¼” Tygon tubing (Cole Palmer Cat. No. A-06408-47). Measurement of the resulting height of foam in the graduated cylinder began as soon the stone reached the bottom of the cylinder, and readings were recorded at 30 second intervals for duration of 10 minutes.

A.3 Results

Table A1 shows the principal characteristics presented for the antifoaming agents and their foaming behavior during testing.

Table A.1: Characteristics of anti-foaming agents and foaming characteristics presented during testing

Anti-Foaming Agent	Color / Appearance	Precipitate formation	Foaming
3112A	Gold-brown oil like	no	steady and stable
3112	bright yellow oil like	yes	steady and stable
3131A	light brown oil like liquid	no	steady and stable
3132A	light brown oil like liquid	no	steady and stable
3126	White thick liquid	yes	foam breaks and falls down
3249	light gold-yellow oil like	no	steady and stable
3249A	White thick liquid	no	steady and stable
3102A	White thick liquid	no	foam breaks but does not falls down (air pockets formation)
3377	light white thick liquid	yes	foam breaks and falls down
3379	White thick liquid	yes	foam breaks and falls down
3142	light brown/white thick	*	foam breaks and falls down
E-10	White thick liquid	yes	foam breaks and falls down

* formation of precipitate only when concentration is over 100 mg/L
 steady and stable = homogeneous foaming , no air pockets formation

No significant differences in foam production were found for the antifoaming agents 3131A, 3249, 3249A, 3132A, 3112, and 3112A in concentrations ranging from 10 to 2000 mg/L. Foam height measured of each concentration from every anti-foaming agent was averaged and presented in Figure A1. The foam produced was stable and homogeneous presenting small size bubbles.

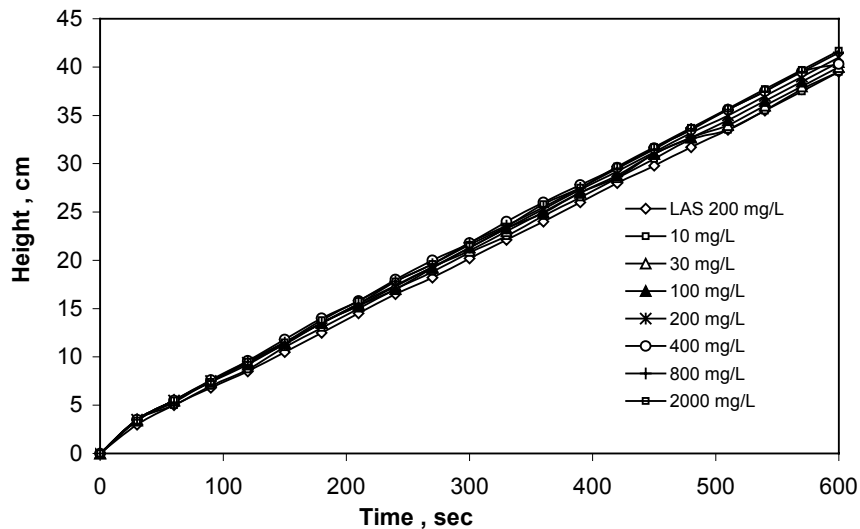


Figure A.1: Foam produced by antifoaming agents 3131A, 3249, 3249A, 3132A, 3112, and 3112A

Four samples, 3102A, 3126, 3377, and 3379 presented a different type of foaming. The foam was not homogeneous with formation of air pockets. These air pockets made the foam to collapse when they burst. Also, the foam produced had bigger bubbles compared to the first samples mentioned above. Figures A2, A3, A4, and A5 show the foam production of samples 3379, 3377, 3126, and 3102A respectively.

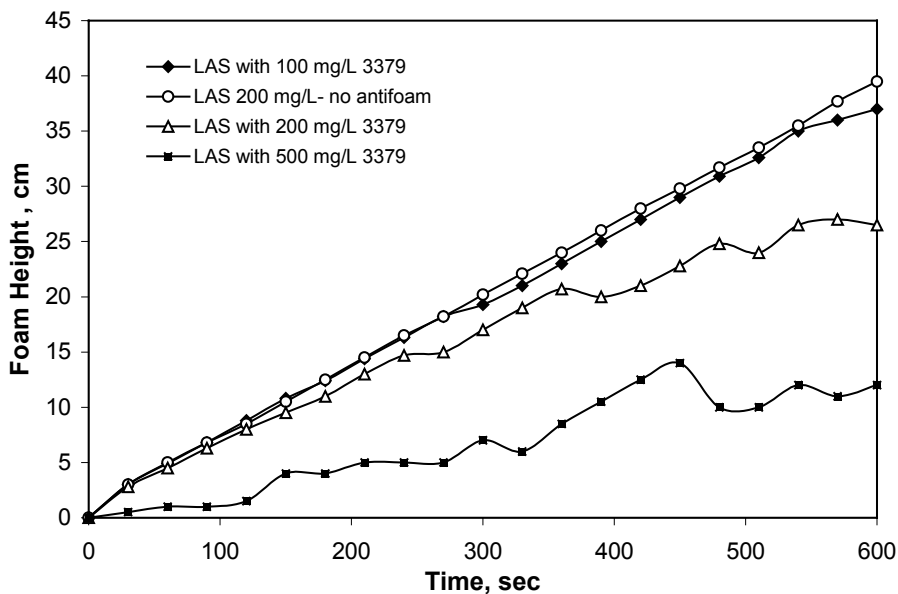


Figure A2: Foam production with antifoaming agent 3379

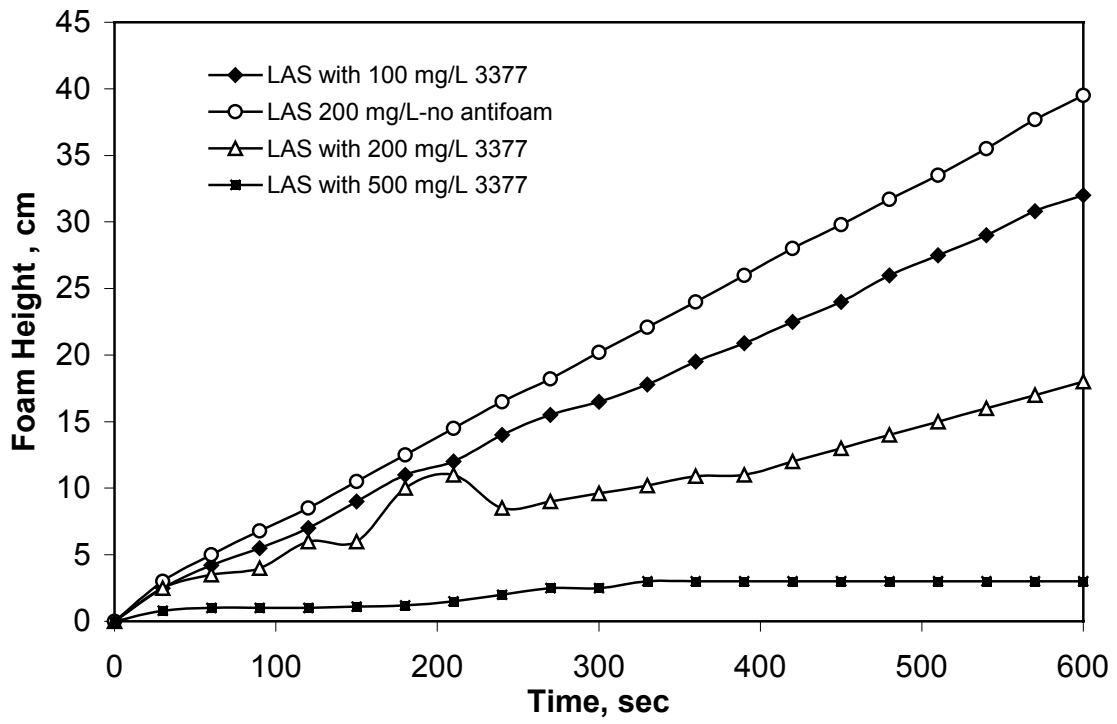


Figure A3: Foam production with antifoaming agent 3377

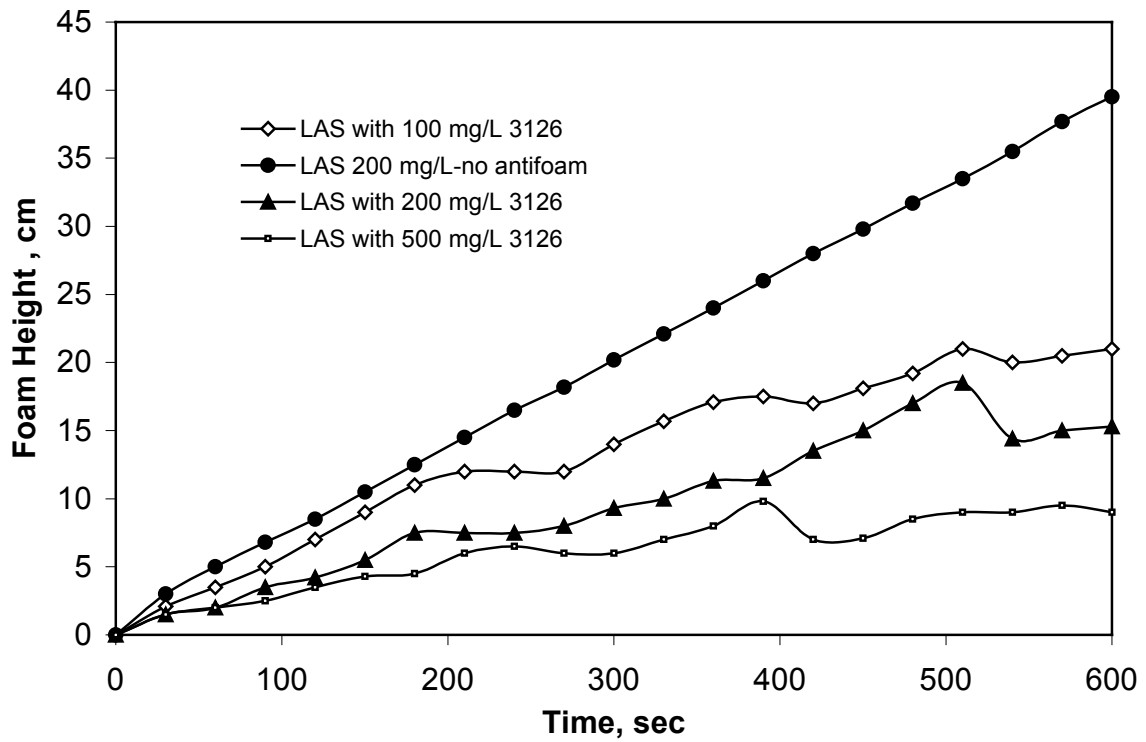


Figure A4: Foam production with antifoaming agent 3126

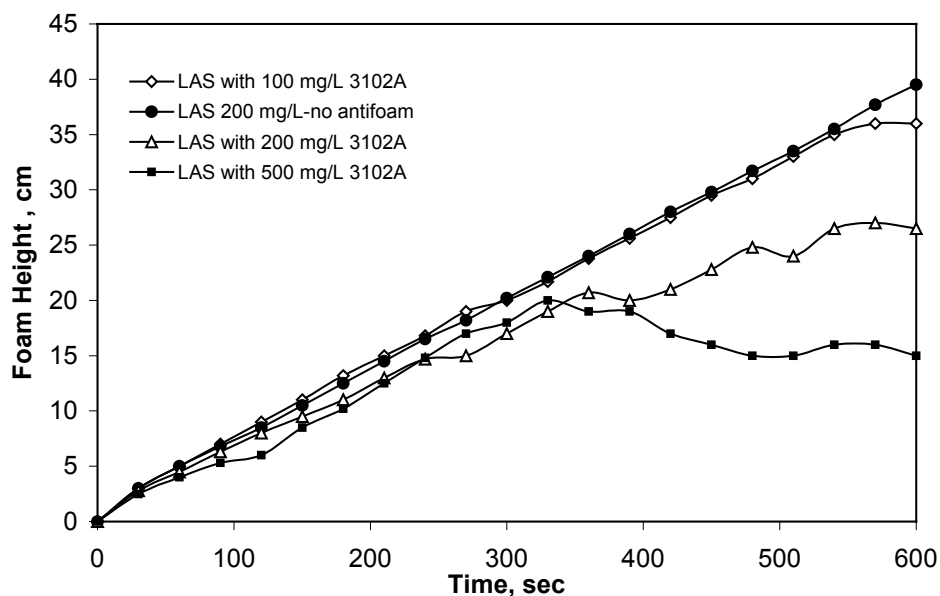


Figure A5: Foam production with antifoaming agent 3102A

Two samples, E-10 and 3142 presented the best results, but for sample E-10 to produce the same or closer results as sample 3142 the antifoaming agent needed to be present in concentrations above 100 mg/L. Figures A6 and A7 show the foaming production of these samples.

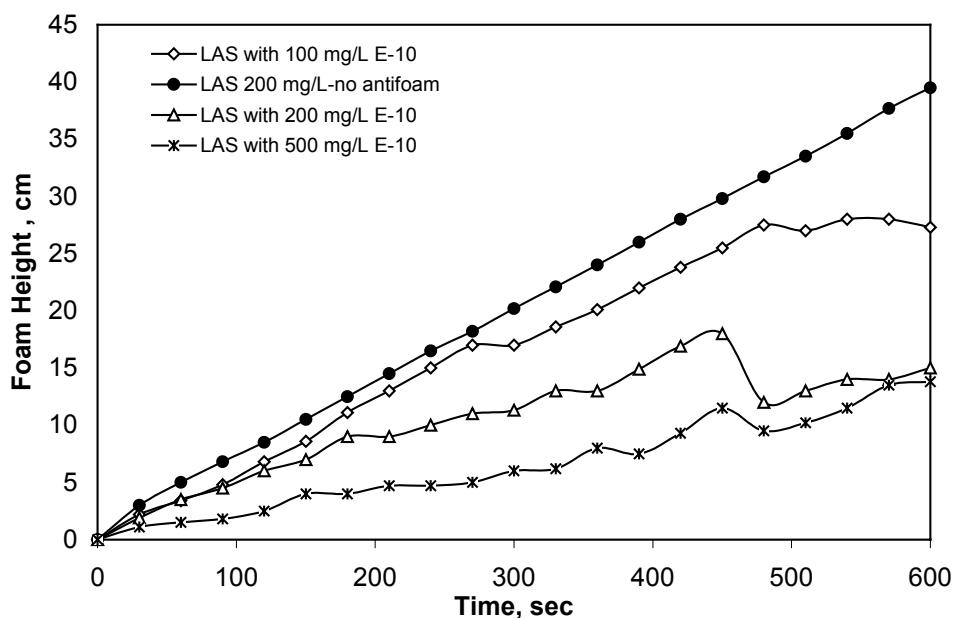


Figure A6: Foam production with antifoaming agent E-10

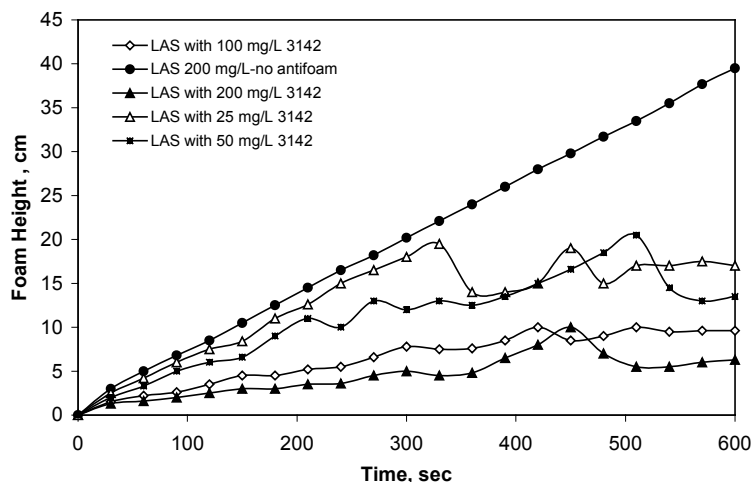


Figure A7: Foam production with antifoaming agent 3142

Sample 3142 had a non-homogeneous foam starting at concentrations of 25 mg/L. A white precipitate was produced when the antifoaming agent 3142 concentration was above 50 mg/L. Total Organic Carbon, TOC, and Methylene Blue Active Substances, MBAS, analysis were performed to verify and measure the contribution of the antifoaming agent. It was found that when the antifoaming 3142 was present in a concentrations of 25 mg/L its contribution, in both TOC and MBAS, was not considerable. Above 50 mg/L, a white precipitate was formed and further TOC and MBAS analysis showed that the surfactant (LAS) was removed from solution. Table A2 shows the results of these analyses.

Table A2: TOC and MBAS analysis of LAS samples containing antifoaming agents E-10 and 3142

Sample	TOC ,mg/L	MBAS, mg/L
LAS (200 mg/L)	113.01	200.1
LAS with 25 mg/L 3142	114.2	204.2
LAS with 50 mg/L 3142	120.31	207.5
LAS with 100 mg/L 3142	75.93	108.71
LAS with 100 mg/L E-10	139.2	215.6
LAS with 200 mg/L E-10	117.51	104.2

Based on these results, anti-foaming agent 3142, was used on further tests to decrease foaming in reactors K1 and K3.

APPENDIX 2: TOC CONCENTRATIONS AS FUNCTION OF TIME FOR RESPIROMETER BOTTLES

Experiments I-IX.

Experiment I

days	Time, hours	30	60(1)	60(2)	150(1)	150(2)	300(1)	300(2)
0	0	12.87	27.72	25.89	65.20	71.07	133.40	120.06
1	24	12.42	18.20	15.00	45.65	49.76	117.28	105.55
2	48	11.80	15.33	14.32	38.70	42.18	109.30	98.37
3	72	3.65	12.38	11.56	35.46	35.65	106.26	95.63
5	120	3.32	10.37	9.68	28.45	29.01	103.87	93.48
7	168	3.05	10.21	8.95	27.74	26.23	99.63	89.67
10	240	2.85	9.93	8.27	28.36	25.91	94.89	86.40
15	360	2.66	9.59	8.03	27.94	24.82	94.71	86.16

Experiment II

days	Time, hours	300	400(1)	400(2)	500(1)	500(2)	600(1)	600(2)
0	0	129.45	172.88	148.68	217.40	228.27	262.60	273.10
1	24	95.45	126.80	109.05	164.71	172.95	204.87	213.07
2	48	55.40	105.68	90.88	122.35	128.47	175.96	183.00
4	96	40.74	67.07	57.68	96.40	101.22	127.29	132.38
8	192	34.10	46.12	39.66	75.70	79.49	101.37	102.42
10	240	32.19	34.40	29.58	59.88	75.87	86.29	79.74
13	312	31.22	34.35	26.54	40.72	52.76	68.80	51.55
15	360	31.05	34.29	25.49	37.75	49.64	45.19	42.99

Experiment III

days	Time, hours	100(1)	100(2)	400(1)	400(2)	1000(1)	1000(2)
0	0	45.82	43.02	173.51	197.97	427.30	429.86
1	24	20.64	19.38	82.15	93.74	321.36	323.28
2	48	6.01	5.64	61.60	70.29	154.34	155.26
3	72	2.91	2.73	23.80	27.16	113.13	113.81
5	96	2.64	2.48	15.46	22.64	94.46	95.03
7	120	2.38	2.03	13.95	19.92	91.93	92.48

Experiment IV

days	Time, hours	30	60	100	200	300	400
0	0	18.58	27.15	41.52	90.7	160.29	207.35
4	96	17.91	24.74	41.29	81.72	150.78	160.78
8	192	16.83	24.2	33.25	74.27	123.58	151.08
11	264	12.65	16.12	30.18	69.77	109.55	146.32

Experiment V

days	Time, hours	30	60	100	200	300	400
0	0	17.8	26.43	40.9	95.2	155.78	220.3
4	96	11.2	18.72	32.55	77.07	140.7	156.7
8	192	10.84	15.4	22.36	58.48	109.5	115.3
11	264	9.02	12.63	20.17	48.55	78.7	107.9

Experiment VI

days	Time, hours	100	200	300	600
0	0	42.06	93.45	152.5	404.81
1	24	40.22	89.45	145.44	349.17
2	48	36.81	71.22	119.67	296.55
3	72	26.01	60.72	90.35	225.12
4	96	24.55	56.91	87.43	218.13
5	120	23.16	55.91	85.20	210.70
6	144	24.70	55.27	82.76	207.44
7	168	24.32	54.75	81.55	201.70
8	192	23.18	53.96	80.3	200.12
9	216	22.57	52.98	79.7	199.78
10	240	21.99	52.33	78.89	198.27

Experiment VII												
Time, hours	TOC						MBAS					
	1	2	3	4	5	6	1	2	3	4	5	6
0	308.49	301.50	307.52	304.87	304.38	303.90	505.04	513.67	512.84	518.32	522.22	526.12
24	296.46	275.36	298.27	291.84	292.75	293.65	413.40	409.30	400.80	395.23	388.93	382.63
48	265.27	270.57	273.29	277.72	281.73	285.74	309.77	320.34	322.54	330.32	336.71	343.09
72	247.73	246.09	252.12	253.04	255.24	257.43	212.33	196.43	193.60	182.06	172.69	163.33
96	207.65	195.19	184.40	237.20	225.58	213.95	104.30	92.75	94.27	87.08	82.06	77.05
120	200.05	182.45	177.61	197.46	198.69	199.92	79.25	76.05	77.12	75.34	74.28	73.21
144	184.52	173.20	179.48	174.02	171.50	168.97	44.59	36.30	33.01	26.39	30.60	24.81
168	183.17	171.71	177.54	171.84	169.03	166.21	39.96	32.67	32.44	27.50	23.74	19.98
192	182.84	171.26	176.35	170.32	167.07	163.82	39.01	32.41	31.97	27.42	23.90	20.38
216	182.32	170.42	175.60	169.38	166.02	162.66	38.40	32.17	31.88	27.63	24.37	21.11
240	181.55	169.58	174.82	168.59	165.22	161.86	38.16	31.83	31.60	27.30	24.02	20.74
264	180.12	168.48	174.56	168.82	166.04	163.26	37.70	31.70	31.12	26.93	23.64	20.35
288	179.67	168.03	174.11	168.37	165.59	162.81	37.60	31.50	30.80	26.50	23.10	19.70

Experiment VIII												
Time, hours	TOC						MBAS					
	1	2	3	4	5	6	1	2	3	4	5	6
0	329.26	303.86	310.29	325.62	300.29	306.63	505.15	509.08	518.29	536.64	522.98	507.97
24	309.03	266.13	268.95	271.51	270.12	261.50	394.79	416.71	412.98	390.29	398.79	404.76
48	240.53	231.29	262.93	253.40	262.63	257.34	217.70	312.25	223.22	249.95	223.39	216.78
72	203.40	204.01	164.67	229.25	205.53	186.18	190.70	214.03	198.20	166.59	183.70	194.25
96	196.17	197.05	186.06	210.80	183.64	167.15	92.86	105.13	93.58	78.59	87.86	91.72
120	185.69	188.05	179.21	197.05	170.45	165.64	45.96	59.88	46.73	34.68	36.02	35.21
144	174.21	181.76	181.09	187.06	169.30	165.07	32.51	44.95	36.63	25.10	26.62	35.90
168	173.85	180.42	179.13	185.45	168.16	164.23	31.95	40.28	32.96	20.38	27.75	32.31
192	172.00	180.10	177.93	184.96	168.02	165.30	30.49	39.32	32.70	20.79	26.67	32.05
216	168.74	179.59	177.18	184.05	166.84	164.12	28.40	38.71	30.46	19.53	25.88	31.81
240	162.07	178.82	176.39	183.14	166.06	163.31	25.13	38.47	28.12	18.16	24.55	28.48
264	161.33	177.42	176.13	181.96	166.29	164.73	20.65	38.00	25.99	17.75	23.17	26.35
288	160.84	176.98	175.67	181.47	165.85	164.27	19.34	37.90	21.78	16.09	21.74	25.15

Total Organic Carbon TOC (mg/L); Methylene Blue Active Substances

**Experiment IX
HUNTSMAN**

Time, hours	TOC 500 mg/L LAS in solution			MBAS mg/L		
	1	2	3	1	2	3
0	360.5	347.9	360.2	486.24	477.06	492.17
24	350.2	340.4	349.7			
48	280.9	280.5	282.16	267.8	244.78	255.87
72	257.5	261.5	255.6			
96	240	239.9	237.6	125.2	107.6	119.8
120	154	228.3	160.35	86.25	67.4	75.92
144	155.45	179.6	153.2	72.5	39.12	32.56
168	149.85	134.2	145.78	44.95	27.06	41.56
192	129.02	104.4	133.5	24.77	23.85	32.44
216	114.12	89.6	120.32	21.15	21.98	28.4
240	112.95	88.78	115.87	21	21.5	27.8
264	111.35	88.01	112.66	21	21.5	27.6
288	110.32	87.35	110.54	20.93	21.5	27.4
312	109.54	86.32	109.12			
336	108.99	85.98	108.45	19.88	21.5	26.72
360	108.67	85.66	107.3			
384	107.87	85.01	106.3			
408	107.65	84.93	106	18.81	21.16	26.34

Total Organic Carbon TOC (mg/L); Methylene Blue Active Substances

**Experiment IX
SIGMA**

Time, hours	TOC 500 mg/L LAS in solution			MBAS mg/L		
	1	2	3	1	2	3
0	317.7	322.1	321.8	500.12	505.5	507.34
24	289	278.1	256.5			
48	255.8	245.2	211.8	361.25	350.6	345.6
72	209.5	227.2	176.4			
96	191.9	209.3	165.5	156.4	150.44	145.87
120	151.9	192.6	148.9	57.12	62.2	55.04
144	142.26	152.4	143.21	45.74	41.1	42.5
168	140.53	132.2	95.3	40.88	34.5	35.17
192	138.27	118.7	94.6	32.79	27.1	32.04
216	135.2	100.3	90.78	30.82	23.44	29.82
240	134.6	100.1	90.4			
264	134	99.8	90			
288	133.8	100.2	89.56			
312	133.9	99.8	90	25.5	20.1	26.87
336	133.6	99.7	89.6			
360	133.4	99.6	89.6			
384	133.8	99.4	89.5			
408	133.5	99.5	89.7	23.56	17.35	26.15

APPENDIX 3: SBR DATA

TSS and TOC within cycles, and influent TOC

TSS	Initial, mg/L	595	625	515	410		565	680	740	760
	Final, mg/L	675	575	475	700		700	750	770	780
	Feed (TOC) mg/L	284.6	284.6	287.4	286.3	300	300	292.3	285.2	285.1
	Time	Cycle 1	Cycle 3	Cycle 5	Cycle 7	Cycle 10	Cycle 11	Cycle 12	Cycle 13	Cycle 14
	0	245.6	194.1	167.1	166.4	172.5	177.104	175.8	167.56	151.9
	24	170.3	151.4	149.9		135.12	151.4	141.9	143.6	132.2
	48	147	130.7	128.9	111.48	105.77	107.9	101.4	104.6	102.3
	72	132	127.5	118.2	101.92	91.83	94.2	96.02	96.1	93.4
	96	130.87	112.1	106.8		86.62	84.1	83.16	82.64	
	120	125.1	105.42	95.85	96.83	84.91	83.4	80.9	81.16	87.4
	Total Removal	49.06	45.69	42.64	41.81	50.78	52.91	53.98	51.56	42.46
TSS	Initial, mg/L	770	770	777.5	782.5	795	792.5	802	817.5	
	Final, mg/L	790	800	805	810	815	822	820	830	
	Feed (TOC) mg/L	285.1	286.5	282.1	280.4	272.9	286.4	285.4	282.1	
	Time	Cycle 15	Cycle 16	Cycle 17	Cycle 18	Cycle 19	Cycle 20	Cycle 21	Cycle 22	
	0	152.7	155.1	143.3	132.5	123.5	157.3	152	145.4	
	24	106.6	119.7	100.7	104.4	95.9	101.8	131.5	111.1	
	48	87.7	93.6	89.8	94.4	84	90.3	99.6	97.82	
	72	89	88.3	80.6	83	82.5	85.86	89.82	93.4	
	96	87					83.86	86.51		
	120	89.7	87.5	78	78.5	77.6	81.12	82.19	84.82	
	Total Removal	41.26	43.58	45.57	40.75	37.17	48.43	45.93	41.66	
TSS	Initial, mg/L	827.5	848.5	856.5	870.5	878.4	878.5	894	920	
	Final, mg/L	850	858	872	880	880	896	900		
	Feed (TOC) mg/L	279.6	282.1	282.7	287.8	268.1	287.1	282.1	282.1	
	Time	Cycle 30	Cycle 31	Cycle 32	Cycle 33	Cycle 34	Cycle 35	Cycle 36	Cycle 37	
	0	129.6	140	153	156.7	137	158.3	148.5	148.5	
	24	105.3	100.1	109.6	122	92	114	105	105	
	48	88.6	84.14	79.8	101.2	85.7	100.7	85	75.1	
	72	80	81.3	79	78.1	74.7	89.8	72.7	65	
	96	74.7	79.21		75.2		72.4			
	120	64.5	64.15	63	64.6	65.1	65.16	62	62.7	
	Total Removal	50.23	54.18	58.82	58.77	52.48	58.84	58.25	57.78	

LAS measured as MBAS

Time	Cycle 1	Cycle 3	Cycle 5	Cycle 7	Cycle 10	Cycle 11	Cycle 12	Cycle 13	Cycle 14
0	314.67	265.8	260.1	262.51	255.89	268.4	260.5	221	206.88
24								130	138.88
48								67.2	138.99
72								40.3	64.68
96								22.8	38.99
120	77.98	95.62	100.7	57.89	51.16	52.44	48.2	19.26	33.48
Total Removal								91.29	83.82

Time	Cycle 15	Cycle 16	Cycle 17	Cycle 18	Cycle 19	Cycle 20	Cycle 21	Cycle 22
0	211.46	194.9	222.01	184.47	155	264.77	247.61	226.24
24	71.55	122.02	78.89	61.09	65.95	85.05	181.23	115.17
48	28.9	51.83	36.67	55.23	46.82	47.815	77.93	72.16
72	33.03	35.77	30.74	40.69	41.05	33.43	46.26	57.85
96	23.85				38.88	24.27	35.54	
120	21.56	32.11	27.04	31.23	33.14	18.35	21.55	28.77
Total Removal	89.80	83.52	87.82	83.07	78.62	93.07	91.30	87.28

Time	Cycle 30	Cycle 31	Cycle 32	Cycle 33	Cycle 34	Cycle 35	Cycle 36	Cycle 37
0	175.07	208.75	236.14	260.12	202.2	244	232.45	208.75
24	96.38	95.28	86.52	64.4	89.8	67.99	60	95.28
48	42.31	35.78	34.55	44.87	43.16	48.32	40.05	35.78
72	37.94	32.58	29.23	39.44	29.45	32.12	28.44	32.58
96	30.21	29.89		30.02		28.5		29.89
120	27.98	27.74	27.13	26.44	24.4	25.16	24.96	27.74
Total Removal	84.02	86.71	88.51	89.84	87.93	89.69	89.26	86.71

Overall Removal per cycle (TOC-MBAS)

Cycle	TOC , mg/L			MBAS , mg/L		
	Influent	Effluent	% Removed	Influent	Effluent	% Removed
1	284.6	125.1	56.04	436.50	77.98	82.14
3	284.6	105.42	62.96	436.50	95.62	78.09
5	287.4	95.85	66.65	440.80	100.7	77.16
7	286.3	96.83	66.18	439.11	57.89	86.82
10	300	84.91	71.70	460.12	51.16	88.88
11	300	83.4	72.20	460.12	52.44	88.60
12	292.3	80.9	72.32	448.31	48.22	89.24
13	285.2	81.16	71.54	437.42	19.26	95.60
14	285.1	87.4	69.34	437.27	33.48	92.34
15	285.1	89.7	68.54	437.27	21.56	95.07
16	286.5	87.5	69.46	439.42	32.11	92.69
17	282.1	78	72.35	432.67	27.04	93.75
18	280.4	78.5	72.00	430.06	31.23	92.74
19	272.9	77.6	71.56	418.56	33.14	92.08
20	286.4	81.12	71.68	439.26	18.35	95.82
21	285.4	82.19	71.20	437.73	21.55	95.08
22	282.1	84.82	69.93	432.67	28.77	93.35
30	279.6	64.5	76.93	428.83	27.98	93.48
31	282.1	64.15	77.26	432.67	27.74	93.59
32	282.7	63	77.71	433.59	27.13	93.74
33	268.1	64.6	75.90	411.20	26.44	93.57
34	287.1	65.1	77.32	440.34	24.44	94.45
35	282.1	65.16	76.90	432.67	25.16	94.18
36	282.1	62	78.02	432.67	24.96	94.23
37	282.1	62.7	77.77	432.67	27.74	93.59

APPENDIX 4: ICEAS DATA

TSS , TOC within cycles, and influent TOC

TSS	Initial	662.5	647.5	637	625	635						
	Final	650	642	630	645	650	660	665	675	685	700	1550
	Feed	285.1	286.5	282.1	280.4	272.9	286.4	285.4	282.1	279.6	282.1	282.7
Time	Cycle 1	Cycle 2	Cycle 3	Cycle 4	Cycle 5	Cycle 6	Cycle 7	Cycle 8	Cycle 9	Cycle 10	Cycle 11	
	0	79.16	118.54	73.98	90.92	87.41	72.13	64.59	64.54	78.92	80.78	75.81
	24	95.24	95.2	83.38	90.11	81.71	69.92	69.76	68.21	74.26	79.62	76.45
	48	105.96	89.4	82.52	88.07	96.1	68.57	63.83	65.46	75.04	79.27	72.5
	72	113.9	86.4	84.14	83.4	93.05	66.43	65.08	68.59	76.79	75.48	75.94
	96	105.58	81.4	90.92	77.31	74.94	66.71	67.59	72.81	75.88	73.83	74.49
	120	107.8	78.31	86.25	82.55	71.78	66.33	66.54	71.73	74.16	77.19	73.81

TSS	Initial											
	Final	1585	1612	1630	1643	1680	1683	1720	1745	1760	1780	1775
	Feed	566.99	268.1	287.1	282.1	282.1	571.1	282.1	287.8	394.9	268.1	287.1
Time	Cycle 12	Cycle 13	Cycle 14	Cycle 15	Cycle 16	Cycle 17	Cycle 18	Cycle 19	Cycle 20	Cycle 21	Cycle 22	
	0	112.4	75.19	72.51	69.63	60.45	92.64	76.06	67.12	66.06	65.14	62.88
	24	95.2	74.46	77.33	69.23	62.03	72.72	76.22	65.81	66.75	62.25	60.41
	48	74.35	73.10	78.54	70.48	60.57	78.13	71.95	60.2	63.85	60.33	61.02
	72	73.56	70.42	75.55	73.29	63.97	79.58	69.35	62.45	63.53	61.58	60.45
	96	72.08	78.86	79.95	66.36	62.44	77.27	66.56	63.01	65.19	60.7	60.75
	120	74.9	77.83	69.33	66.82	60.68	75.46	62.44	60.3	68.62	60.7	61.03

LAS measured as MBAS

Time	Cycle 1	Cycle 2	Cycle 3	Cycle 4	Cycle 5	Cycle 6	Cycle 7	Cycle 8	Cycle 9	Cycle 10	Cycle 11
0	31.2	88.53	22.68	44.95	32.92	22.06	19.76	19.74	24.14	24.71	23.19
24	69.3	66.05	34.4	39.91	24.99	21.39	21.34	20.87	22.72	24.36	23.39
48	92.2	44	32.11	38.1	29.4	20.98	19.53	20.03	22.96	24.25	20.64
72	105.9	40.4	35.8	27.06	28.47	20.32	19.91	20.98	23.49	23.09	23.47
96	83.9	31.65	44.95	12.66	22.93	20.41	20.68	22.67	23.21	22.59	22.06
120	91.5	20.2	44.95	30.03	21.96	20.29	20.36	21.94	20.67	23.62	19.53

Time	Cycle 12	Cycle 13	Cycle 14	Cycle 15	Cycle 16	Cycle 17	Cycle 18	Cycle 19	Cycle 20	Cycle 21	Cycle 22
0	96.89	23.41	19.98	19.6	60.45	55.26	26.89	21.84	19.52	26.62	22.44
24	81.44	23.1	23.16	19.5	62.03	30.5	26.41	20.45	19.06	19.06	19.62
48	30.4	22.85	25.22	19.81	60.57	25.36	20.45	21.16	18.42	18.64	19.5
72	23.04	21.9	23.47	20.45	63.97	25.6	19.55	18.44	21.85	18.7	19.5
96	20.78	25.61	22.01	19.04	62.44	21.85	19.04	19.7	22.45	19.21	19.5
120	21.55	24.8	19.85	18.55	60.68	20.66	18.87	18.6	21.45	19.05	19.5

Overall Removal per cycle (TOC-MBAS)

Cycle	TOC , mg/L			MBAS , mg/L		
	Influent	Effluent	% Removed	Influent	Effluent	% Removed
1	285.1	107.8	62.19	437.27	91.5	79.07
2	286.5	78.31	72.67	439.42	20.2	95.40
3	282.1	86.25	69.43	432.67	44.95	89.61
4	280.4	82.55	70.56	430.06	30.03	93.02
5	272.9	71.78	73.70	418.56	21.96	94.75
6	286.4	66.33	76.84	439.26	20.29	95.38
7	285.4	66.54	76.69	437.73	20.36	95.35
8	282.1	71.73	74.57	432.67	21.94	94.93
9	279.6	74.16	73.48	428.83	20.67	95.18
10	282.1	77.19	72.64	432.67	23.62	94.54
11	282.7	73.81	73.89	433.59	19.63	95.47
12	566.99	74.9	86.79	869.62	21.55	97.52
13	268.1	77.83	70.97	411.20	24.8	93.97
14	287.1	69.33	75.85	440.34	19.85	95.49
15	282.1	66.82	76.31	432.67	18.55	95.71
16	282.1	60.68	78.49	432.67	18.81	95.65
17	571.1	75.46	86.79	875.92	20.66	97.64
18	282.1	62.44	77.87	432.67	18.87	95.64
19	287.8	60.3	79.05	441.41	18.6	95.79
20	394.9	68.62	82.62	605.67	21.45	96.46
21	268.1	60.7	77.36	411.20	19.05	95.37
22	287.1	61.03	78.74	440.34	19.5	95.57

APPENDIX 5: SBBR DATA

TOC within cycles, and influent TOC

Feed														272.9	286.4	285.4	282.1
Time	Cycle 1	Cycle 2	Cycle 3	Cycle 4	Cycle 5	Cycle 6	Cycle 7	Cycle 8	Cycle 9	Cycle 10	Cycle 11	Cycle 12	Cycle 13	Cycle 14	Cycle 15	Cycle 16	Cycle 17
0	241.17	246.6	187.6	146	144.9	122.2	133.1	162.5	173.1	185.4	174.3	180.24	164.3	174.46	172.5	169.8	171.9
0.5								124.7									
1								105.8									
1.5								99.1									
2								95.7									
2.5								88.9									
5								85.9				132.37					
24	152.5	120.92	100.48	74.06	71.56	71.25	68.82	83.51	105.5	116.2	111.6	120.25	117.5	127.09	119.8	115.1	117.2
36																	
48	146.77	117.08	102.37	73.02	72.8				96.38	97.9	92.5	123.9					
60																	
72	147.31								106.6	96.17	90.2						
96																	
120																	
Feed	287.1	282.1	282.1	283	283.2	280.4	282.5	284.2	281.5	281.5	281.5	285.6	285.6	284.1	284.1	283.4	282.1
Time	Cycle 23	Cycle 24	Cycle 25	Cycle 26	Cycle 27	Cycle 28	Cycle 29	Cycle 30	Cycle 31	Cycle 32	Cycle 33	Cycle 34	Cycle 35	Cycle 36	Cycle 37	Cycle 38	Cycle 39
0	173.6	172.3	172	172	173.6	170.2	171.5	173.8	140	142	141.4	142.1	139.5	135.5	137.1	140	137.2
0.5										120.70	108.88	112.26	115.79	113.14	117.91	114.80	113.19
1																	
1.5																	
2										103.66	101.67	100.89	103.23	97.42	93.23	96.60	96.04
2.5																	
5										78.10	77.77	78.16	76.73	74.53	75.41	77.00	75.46
24	121	119	119	116	117	115.8	114.1	115.4	116	95.47	86	81.6	80.6	79	77.8	62.3	71.17
36																	
48									109.68	96.8	87.25	84.31	84.31	88.57	80.17	78.38	72.09
60																	
72																	
96																	
120																	
Feed	282.1	282.1	285.9	285.9	284.4	280	282.1	282.1	284	284	284	283.4	282	283.1	282.1	282.1	285
Time	Cycle 45	Cycle 46	Cycle 47	Cycle 48	Cycle 49	Cycle 50	Cycle 51	Cycle 52	Cycle 53	Cycle 54	Cycle 55	Cycle 56	Cycle 57	Cycle 58	Cycle 59	Cycle 60	Cycle 61
0	162.83	134.2	140.27	117.9	124.37	125	128.22	132.6	133.91	130.55	131.06	130.8	130.7	165.75	152.11	133.58	135.15
0.5	131.57	111.39	117.83	100.80	99.37	98.75	105.14	108.73						137.57	112.08	110.87	109.47
1																	
1.5																	
2	118.87	93.94	105.20	86.07	89.55	90.00	88.47	94.15	92.40	93.87	96.98	95.48	93.45	107.33	103.93	94.84	92.58
2.5																	
5	89.56	73.81	77.15	64.85	68.40	68.75	71.32	74.26	73.65	65.28	70.77	66.71	77.11	92.82	80.6	72.13	75.68
24	91.92	85.48	82.45	77.48	71.77	67.24	67.26	72.57	69.13	67.03	63.25	63.88	60.3	97.13	73.04	70.85	68.56
36														88.72	77.41		
48	73.2	86.05	78.64	74.04	69.01	66.42	65.21	62.99	64.07	62.79	61.23	60.96	58.06		75.24	68.74	62.93
60														84.74			
72								59.89				64.53		85.66			65.17
96																	
120														86.09			

LAS as MBAS

Feed														418.56	439.26	437.73	432.67
Time	Cycle 1	Cycle 2	Cycle 3	Cycle 4	Cycle 5	Cycle 6	Cycle 7	Cycle 8	Cycle 9	Cycle 10	Cycle 11	Cycle 12	Cycle 13	Cycle 14	Cycle 15	Cycle 16	Cycle 17
0	266.97	180.58	174.77	156.42	178.89	133.03	192.2	233.48	270.63	272.9	256.14	247.1	233.5	240	241	240	245
0.5																	
1																	
1.5																	
2																	
2.5																	
5																	
24						50.45	49.90	50.45	100.00			178.00	159.00	140.00	135.00	137.00	143.00
36																	
48		20.18	13.3	35.77	45.41												
60																	
72	37.16									66.15	61.27						
96																	
120																	
Feed	440.34	432.67	432.67	434.05	434.36	430.06	433.28	435.89	431.75	431.75	431.75	438.04	438.04	435.74	435.74	434.66	432.67
Time	Cycle 23	Cycle 24	Cycle 25	Cycle 26	Cycle 27	Cycle 28	Cycle 29	Cycle 30	Cycle 31	Cycle 32	Cycle 33	Cycle 34	Cycle 35	Cycle 36	Cycle 37	Cycle 38	Cycle 39
0	260	262	249	251	266	255	253	261	253	206	179.35	208.25	210.1	219.72	210.55	211.92	208.71
0.5																	
1																	
1.5																	
2																	
2.5																	
5																	
24	144.00	136.00	141	144	147	145	141	144	140	70.2	55.96	57.34	56.88	44.49	53.21	48.16	38.07
36																	
48									33.03	45.8	39.44	38.53	34.4	35.78	27.06	10.56	9.17
60																	
72																	
96																	
120																	
Feed	432.7	432.7	438.5	438.5	436.2	436.2	432.7	432.7	432.7	432.7	434.7	434.7	432.5	943.3	432.7	432.7	437.1
Time	Cycle 45	Cycle 46	Cycle 47	Cycle 48	Cycle 49	Cycle 50	Cycle 51	Cycle 52	Cycle 53	Cycle 54	Cycle 55	Cycle 56	Cycle 57	Cycle 58	Cycle 59	Cycle 60	Cycle 61
0	198.5	192.4	198.6	85.79	127.4	237.86	192.7	202.6	208.6	222.14	201.83	206.85	194.23	206	203.5	205.48	
0.5	159.05													155.78			
1																	
1.5																	
2	101.43													98.42			
2.5																	
5	88.74						26.62							92.74			
24	65	30.16	35.06	22.31	33.95	23.52	12.82	26.48	21.78	23.39	22.84	25.65	23.58	18.47	25.2	26.4	25.87
36														11.92			
48	17.23	15.01	16.16	12.5	12.04	11.41	8.94	11.47	6.54	7.72	7.05	6.54	8.04	7.63	8.22	6.01	7.15
60																	
72								3.86						5.37			5.28
96																	
120																	

Overall Removal cycles 32 to 66 (TOC-MBAS)

Cycle	TOC , mg/L			MBAS , mg/L		
	Influent	Effluent	% Removed	Influent	Effluent	% Removed
32	281.5	96.8	65.61	431.75	45.8	89.39
33	281.5	87.5	68.92	431.75	39.44	90.87
34	285.6	84.31	70.48	438.04	38.53	91.20
35	285.6	84.31	70.48	438.04	34.4	92.15
36	284.1	88.57	68.82	435.74	35.78	91.79
37	284.1	80.17	71.78	435.74	27.06	93.79
38	283.4	78.38	72.34	434.66	10.56	97.57
39	282.1	72.09	74.45	432.67	9.17	97.88
40	282.1	85.79	69.59	432.67	12.71	97.06
41	283.6	71.35	74.84	434.97	11.74	97.30
42	283.5	76.93	72.86	434.82	16.89	96.12
43	283.4	74.24	73.80	434.66	11.13	97.44
44	283.4	75.6	73.32	434.66	13.41	96.91
45	282.1	73.2	74.05	432.67	17.23	96.02
46	282.1	86.05	69.50	432.67	15.01	96.53
47	285.9	78.64	72.49	438.50	16.16	96.31
48	285.9	74.04	74.10	438.50	12.5	97.15
49	284.4	69.01	75.73	436.20	12.04	97.24
50	280	66.42	76.28	429.45	11.41	97.34
51	282.1	65.21	76.88	432.67	12.82	97.04
52	282.1	59.89	78.77	432.67	3.86	99.11
53	284	64.07	77.44	435.58	6.54	98.50
54	284	62.79	77.89	435.58	7.72	98.23
55	284	62.23	78.09	435.58	7.05	98.38
56	283.4	64.53	77.23	434.66	4.02	99.08
57	282	58.06	79.41	432.52	8.04	98.14
58	283.1	86.09	69.59	434.20	5.37	98.76
59	282.1	75.24	73.33	432.67	8.22	98.10
60	282.1	68.74	75.63	432.67	6.01	98.61
61	285	65.17	77.13	437.12	5.28	98.79
62	285	58.86	79.35	437.12	7.79	98.22
63	288	56.45	80.40	441.72	5.36	98.79
64	288	57.7	79.97	441.72	4.15	99.06
65	288	56.41	80.41	441.72	6.88	98.44
66	285	55.62	80.48	437.12	8.16	98.13

APPENDIX 6: COD BALANCE - K1

Influent	1175	260.9	405.61						
	mg/L	mg/L	mg/L	mg/L-hr	COD from				
Time, h	COD	TOC	MBAS	OUR	MBAS		OUR	Aveg OUR	
0	706	148.5	208.75	3.45	580.33		3.456	5.175	31.05
24	485	105	95.28	12.516	264.88		6.894	8.672	104.064
48	303	65	35.78	7.042	99.47		10.45	11.483	68.898
72	290	62.8	32.58	4.863	90.57		12.516	11.7845	70.707
96	284	62.8	29.89	3.32	83.09		11.053	9.1315	109.578
120	267	62.7	27.74	2.01	77.12		7.21	7.126	42.756
Overall removed	77.28	75.97	93.16						

Time, h	mg/L-h	mg/L-h	mg/L	mg Oxygen
0	3.456	5.175	31.05	77.625
6	6.894	8.672	104.064	260.16
18	10.45	11.483	68.898	172.245
24	12.516	11.7845	70.707	176.7675
30	11.053	9.1315	109.578	273.945
42	7.21	7.126	42.756	106.89
48	7.042	7.026	42.156	105.39
54	7.01	6.235	74.82	187.05
66	5.46	5.2995	31.797	79.4925
72	5.139	5.001	30.006	75.015
78	4.863	4.2305	50.766	126.915
90	3.598	3.46	20.76	51.9
96	3.322	3.091	18.546	46.365
102	2.86	2.528	30.336	75.84
114	2.196	1.893	11.358	28.395
120	1.59			

K1

Start of Cycle	
Residual	
COD	313 mg/L
Volume	1.25 L
COD mass	391.25 mg
Input	
COD	1175 mg/L
Volume	1.25 L
COD mass	1468.75 mg
Biomass	
TSS	894 mg/L
COD equ	1.42 mg COD/mg biomass
Volume	2.5 L
COD mass	3173.7 mg

End of Cycle	
Residual	
COD	267 mg/L
Volume	2.5 L
COD mass	667.5 mg
Oxygen consumed	
COD	
Volume	2.5 L
COD mass	1844 mg
Biomass	
TSS	900 mg/L
COD equ	1.42 mg COD/mg biomass
Volume	2.5 L
COD mass	3195 mg

Total COD,mg 5033.45

Total COD,mg 5706.5

COD BALANCE - K2

Influent	1175	260.9	405.61									
	mg/L COD	mg/L TOC	mg/L MBAS	mg/L-hr	COD from MBAS	Time, h	OUR mg/L-h	Volume L	Oxygen mg			
0	216	60.45	60.45		168.05	0	5.16	9.0735	54.441	1.25	1,281.5	69,766.14
24	217	62.03	62.03		172.44	6	12.967	15.3485	184.182	1.31	1.38	253.43
48	270	70.57	60.57		168.38	18	17.71	13.615	81.69	1.44	1.47	120.13
72	267	63.97	63.97		177.84	24	9.52	9.241	55.446	1.50	1.53	85.03
96	214	55.68	62.44		173.58	30	8.962	8.8855	106.626	1.57	1.63	173.59
120	216	60.99	60.68		168.69	42	8.809	8.775	52.65	1.69	1.72	90.69
						48	8.741	8.7015	52.209	1.75	1.79	93.22
Overall removed	81.62	76.62	85.04			54	8.662	8.21	98.52	1.82	1.88	185.22
						66	7.758	7.599	45.594	1.94	1.97	90.03
						72	7.44	7.336	44.016	2.01	2.04	89.68
						78	7.232	6.776	81.312	2.07	2.13	173.36
						90	6.32	6.187	37.122	2.20	2.23	82.65
						96	6.054	5.811	34.866	2.26	2.29	79.83
						102	5.568	5.4845	65.814	2.32	2.38	156.90
						114	5.401	4.9755	29.853	2.45	2.48	73.99
						120	4.55	4.55		2.51	2.51	0.00

K2

Start of Cycle	
Residual	
COD	216 mg/L
Volume	1.25 L
COD mass	270 mg
Input	
COD	1175 mg/L
Volume	1.25 L
COD mass	1468.75
Biomass	
TSS	3360 mg/L
COD equ	1.42 mg COD/mg biomass
Volume	1.25 L
COD mass	5964 mg

End of Cycle	
Residual	
COD	216 mg/L
Volume	2.5 L
COD mass	540 mg
Oxygen consumed	
COD	
Volume	2.5 L
COD mass	1817.50 mg
Biomass	
TSS	1683 mg/L
COD equ	1.42 mg COD/mg biomass
Volume	2.5 L
COD mass	5974.65 mg

Total COD,mg 7702.75

Total COD,mg 8332.15

APPENDIX 7: FOAM HEIGHT SBR

Foam height - 5-day cycle (Cycle 15,16 -21,22- 36)

Time , hours	Foam height , cm						K1		
	Cycle 15	Cycle 16	Cycle 25	Cycle 26	Cycle 34	Average	OUR	TOC	LAS
0.00	0	0	0	0	0	0		150.74	193.36
1.00	0.5	0	0.5	0.5	0	0.3			
1.50	1.5	1.1	1.3	1.5	1.3	1.34			
2.00	3	2.5	2.2	2.5	2.5	2.54			
2.33	5.5	5	4.8	5	5	5.06			
3.00	6	5.3	5.5	5	5.5	5.46			
3.50							3.456		
4.00	7	6.5	5.5	5.2	6	6.04			
5.00	8.5	7.5	8	8.2	8	8.04			
6.50	9.7	8.8	9.7	9.3	9.4	9.38			
8.00	11	9.4	10.5	9.4	9.4	9.94			
9.50							6.894		
12.00	12	9.6	10.3	9	9.4	10.06			
21.50							10.45		
24.50	10	8.7	9.6	8.3	8.5	9.02		114.78	109.99
27.50							12.516		
33.50							11.053		
36.00	6	5	7	5	4.6	5.52			
45.50							7.21		
48.50	4.2	3.3	2	3	2.5	3		92.744	54.17
51.50							7.042		
57.50									
69.50							7.01		
72.50	3.1	1.9	2	1.5	2.3	2.16		86.64	40.27
75.50							5.46		
81.50							5.139		
93.50							4.863		
96.50	1.2	0.8	0.8	0.6	1	0.88		86.76	29.70
99.50							3.598		
105.50							3.322		
117.50							2.86		
120.50	0.3	0.8	0.4	0.7	0.8	0.6	2.354	81.24	25.79

APPENDIX 8: BATCH SORPTION DATA - SBR AND ICEAS

		K1								
		Co mg/L	Ce mg/L	Co - Ce (mg/L)	X (mg)	Biomass		dry mass (m, mg)	(x/m)	Ce/(x/m)
						mass	moisture,%			
LAS	50 mg/L	34.87	28.65	6.22	0.7464	0.5157	95.32	24.13	0.03	926.39
		34.87	26.73	8.14	0.9768	1.2314	95.32	57.63	0.02	1577.02
		34.87	25.96	8.91	1.0692	1.3879	95.32	64.95	0.02	1577.07
	500 mg/L	358.1	393.21	-35.11	-4.2132	0.4778	95.32	22.36	-0.19	-2086.91
		358.1	331.64	26.46	3.1752	1.1456	95.32	53.61	0.06	5599.83
		358.1	318.53	39.57	4.7484	1.4484	95.32	67.79	0.07	4547.13
	1000 mg/L	717	620.02	96.98	11.6376	0.6518	95.32	30.50	0.38	1625.18
		717	589.85	127.15	15.258	1.313	95.32	61.45	0.25	2375.50
		717	549.4	167.6	20.112	1.2168	95.32	56.95	0.35	1555.60

		K2								
		Co (mg/L)	Ce (mg/L)	Co - Ce (mg/L)	X (mg)	Mass .g		dry mass (m, mg)	(x/m)	Ce/(x/m)
						mass	moisture,%			
LAS	50 mg/L	34.87	47.66	-12.79	-1.5348	1.1721	94.54	64.00	-0.02	-1987.28
		34.87	55.62	-20.75	-2.49	1.448	94.54	79.06	-0.03	-1766.01
		34.87	52.36	-17.49	-2.0988	1.6591	94.54	90.59	-0.02	-2259.92
	500 mg/L	358.1	415.02	-56.92	-6.8304	1.448	94.54	79.06	-0.09	-4803.79
		358.1	405.35	-47.25	-5.67	1.4558	94.54	79.49	-0.07	-5682.53
		358.1	408.2	-50.1	-6.012	2.8646	94.54	156.41	-0.04	-10619.66
	1000 mg/L	717	738.91	-21.91	-2.6292	1.2181	94.54	66.51	-0.04	-18691.47
		717	740.74	-23.74	-2.8488	1.7185	94.54	93.83	-0.03	-24397.54
		717	752.14	-35.14	-4.2168	1.8542	94.54	101.24	-0.04	-18057.80

VITA

Luis Alberto Espinoza Rodezno was born in Honduras on August 19, 1972. Luis received his Bachelor of Science in Chemical Engineering from Universidad Nacional Autónoma de Honduras, in 1995. He worked for Corporación Cressida as process engineer in the area of palm oil extraction and bleaching from 1996 to 1998, and then was hired by Unilever de Honduras as Quality Assurance manager for their chemical complex located in Comayagua. In 2001, Luis entered the graduate school of Louisiana State University to obtain a Master of Science in Civil Engineering under a Fulbright scholarship. He worked under the guidance of Dr. William M. Moe, testing operating strategies for bioreactors treating model industrial wastewater containing high concentrations of linear alkylbenzene sulfonate (LAS).