Effect of Feed Rate and Solid Retention Time (SRT) on Effluent Quality and Sludge Characteristics in Activated Sludge Systems using Sequencing Batch Reactors

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EFFECT OF FEED RATE AND SOLID RETENTION TIME (SRT) ON EFFLUENT QUALITY AND SLUDGE CHARACTERISTICS IN ACTIVATED SLUDGE SYSTEMS USING SEQUENCING BATCH REACTORS

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ABSTRACT

A critical element to the successful operation of activated sludge systems is efficient solid liquid separation achieved by bioflocculation. Bioflocculation refers to the process of microbial aggregation to form activated sludge flocs, dependent on the interaction of exocellular polymeric substances (EPS) to form the matrix that holds microbes, other organics and inorganic particles in a flocculent mass. Numerous factors affect bioflocculation; two key parameters are the Solid Retention Time (SRT) and the substrate loading rate. The latter is related to the two basic designs in activated sludge bioreactor configurations: the Plug Flow Reactor (PFR) and the Completely Stirred Tank Reactor (CSTR). PFR systems have a high substrate loading rate, whereas CSTRs have a low substrate loading rate. Research has shown that the PFR configurations produce better sludge quality, in terms of settleability and dewaterability, and subsequently better effluent quality than CSTR systems.

In this experiment, the effect of SRT and substrate loading rate on activated sludge was investigated using bench scale SBRs. PFR and CSTR configurations were simulated by adjusting the fill period to be shorter or longer respectively. A series of SBRs were operated, each with an operating volume of 6L, to obtain data for PFR (**fast feed**) versus CSTR (**slow feed**) configurations at 10 Day, 5 Day and 2 Day SRTs. Effluent quality was monitored by measuring effluent TSS, VSS, total and soluble COD and soluble biopolymers. Sludge quality was monitored for the aerobic phase by measuring total and suspended solids, total and suspended volatile solids, Sludge Volume Index (SVI), Capillary Suction Time (CST) and Zeta Potential. Anaerobic digestibility was measured for the sludge produced in these systems by measuring gas production, similar to estimating biogenic methane potential (BMP) and determining short term odor productions, specifically Total Volatile Organic Sulfur Compounds (TVOSCs).

As expected the change in feeding pattern and SRTs affected the effluent and sludge quality during the aerobic operation phase. Effluent quality was found to be better for the fast feed system at all SRTs, with all monitored parameters being of similar or significantly lower concentration than for the slow feed system. In terms of sludge quality, the fast feed system was found to retain more of its biomass in solution, indicating better flocculation and settleability in this system. COD was given a lower rank as an effluent quality indicator, since the 5 Day and 2 Day SRT datasets did not correlate well with other datasets, specifically effluent TSS and biopolymers. The data was included because it is believed that the trends were accurate

representations of fast versus slow feed system behavior. The trends were comparable to those of effluent TSS and solution biopolymer datasets.

In terms of anaerobic digestion potential, the fast feed sludge exhibited greater volumetric gas production per gram of solid at the 5 and 2 Day SRTs. Gas production was similar for both systems at the 10 Day SRT. Total and Volatile Solid reduction were however found to be higher for the slow feed sludge than for the fast feed. This may indicate higher gas and potential odor production per gram of solid degraded for the fast feed sludge. This theory is supported by the odor analyses, which revealed that the fast feed sludge had a higher TVOSC production at each SRT. This was related to the higher protein content of the sludge, indicated by the effluent biopolymers being much higher in protein content than carbohydrates. Shearing, which is part of the solids handling process at most plants, releases these proteins and makes them bioavailable, allowing them to be oxidized to produce TVOSCs and hence higher odors.

In conclusion it was found that the fast feed effluent and sludge quality appeared to be overall better at each SRT simulated; the higher TVOSC content may indicate a problem with solids handling, but research has shown that these can be overcome with the addition of iron. Additionally, both systems, the fast and slow feed systems operated better at longer SRTs, with the fast feed system performing better in all cases. The difference was not completely significant in all cases and this is attributed to being a by-product of operating at the optimal M:D salt ratio.

This project has strength in terms of its potential for large scale applications. SRT is the considered the most important design parameter and one of the more complicated parameters to manipulate due to its widespread effect on reactor behavior, specifically sludge and effluent quality. Additionally, the fast feed versus slow feed concept is one that has been gaining significant interest, since bioreactor configuration impacts the effluent and sludge quality. Feed configurations have been investigated more frequently within the past decade. The novel approach taken by this project is that it combines these two parameters, both of which are important to large scale plants, both industrial and municipal.

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CHAPTER 1. BACKGROUND

1.1 WASTEWATER TREATMENT AND THE ACTIVATED SLUDGE SYSTEM

1.1.1 The Activated Sludge Process.

Since its inception in 1914 (*Ardern et al. 1914*) the activated sludge system has become one of the most widely used biological wastewater treatment processes (*Sanin et al.2006*). It is an aerobic suspended growth system in which microorganisms are grown for the purpose of removing soluble organic matter (*Grady et al.1999*). The suspended microbes are flocculent in nature and are collectively referred to as biomass. This biomass is mixed into wastewater streams in large basins called biological reactors or bioreactors, in which aerobic conditions are maintained by mechanical aerators, such as diffused or surface aerators. With proper adjustments, the activated sludge technique can also be used to achieve nitrogen and phosphorus removal.

The performance of the activated sludge process is affected by several factors; these include temperature, pH, sludge return rates, dissolved oxygen levels, food to microorganism ratio (that is, organic matter to activated sludge ratio), aeration rates and wastewater toxicity. Consequently there are many different configurations of the activated sludge system, classified according to the nature of these factors. All configurations share a few basic features. The biomass is introduced into the aeration basin and mixed into the influent wastewater stream to form a flocculent slurry called **mixed liquor**. This medium is kept well aerated; dissolved oxygen levels are usually maintained at or above 2mg/L to limit filamentous bulking sludge characteristic of low DO concentrations. Aerobic bacteria metabolize the organic matter to form biological flocs, which are then removed via quiescent gravity settling in secondary sedimentation chambers to produce effluent that is low in suspended solids. This settled material is removed and a portion is returned to the beginning of the bioreactor basins as a concentrated slurry to maintain a viable biomass concentration; this fraction is referred to as return activated sludge (RAS). The remainder is disposed of and called waste activated sludge (WAS); the fraction of WAS is manipulated to maintain the desired SRT (*Grady et al. 1999*).

The central control point in the activated sludge process is the bioreactor. There are two ideal configurations that exist for designing these, based on the manner of introducing the waste stream. The simplest configuration is the continuously stirred tank reactor (CSTR), which comprises a completely mixed reactor with a constant liquid volume, where mixing is sufficient to maintain a constant concentration throughout the reactor. The concentration in the reactor is equal to the effluent concentration. The other configuration is the plug flow reactor (PFR) in which the influent material moves through the system in the same order that they enter without intermixing. This latter configuration can also be referred to as pulse feeding (*Grady et.al. 1999*).

1.1.2 Activated Sludge Structure.

A critical element to successful wastewater treatment by the activated sludge process is the achievement of proper solid-liquid separation. Mixed liquor, the key component of the activated sludge process, is a heterogeneous mixture of organic and inorganic particles, particularly microorganisms and colloidal material held together by a matrix of polymers called extracellular polymeric substances (EPS), and cations. The composition of the specific fractions depends on the origin of the sample (*Bruus et al. 1992; Murthy et al. 1998; Sanin et al.2006; Nguyen et al. 2008*)

Activated sludge structure has been described in terms of macrostructure and microstructure; research has suggested that the structure is comprised of bacteria and micro-colonies, which can be considered the microstructure, and flocs, or macrostructure (*Nguyen et al. 2008*). The first level of the microstructure is comprised of bacterial cells held in a polymeric matrix to form micro-colonies, which is considered the second level of microstructure. The macrostructure is comprised of these micro-colonies enmeshed in a matrix of polymeric substances and cations to form activated sludge flocs, or bioflocs. Researchers postulate that these polymeric substances are either the products of microbial metabolism and cell lysis or originate in the influent wastewater stream (*Urbain et al. 1993; Grady et al. 1999*). EPS is comprised of proteins, polysaccharides, humic compounds, nucleic acids and lipids (*Higgins et al. 2002*). Researchers state that the relative proportion of proteins to polysaccharides influence bioflocculation and subsequently the dewaterability; higher protein to carbohydrate ratios improve dewaterability, which improves sludge digestibility (*Sanin et al. 2006*)

1.2 Solid-Liquid Separation: The Role of Bioflocculation.

Solid-liquid separation is a physical process whereby solid particles, which are usually in a suspended or colloidal state after every stage of treatment, are removed. The removal of this suspended material, primarily composed of biomass from the biochemical operations, is done by gravity sedimentation. The efficiency of the activated sludge treatment process is correlated to a good solid-liquid separation, which is strongly determined by the biomass settling properties (*Govoreanu et al., 2003*). Efficient solid-liquid separation results from the aggregation of microbes and solids into activated sludge flocs, or bioflocs. This flocculation, called bioflocculation, is determined by the interaction between EPS and cations in the activated sludge macrostructure. These microbial adhesion mechanisms are critical to producing a good biofloc; an ideal biofloc is defined as one that is strong and compacts well so that it settles properly producing a dense sludge for recycle to the bioreactor and a high quality effluent. Bioflocculation impacts not only the settling properties of sludge, which are determined in the aeration basin, but also the dewaterability. The final stage of biosolid-liquid separation involves quiescent settling in secondary clarifiers; this step of the process is considered the limiting factor in producing high quality effluent. Poor bioflocculation in the aeration basin can be seen by poor

settling in the clarifiers, turbid effluent and adverse effects on dewatering (*Grady et al., 1999*; *Sanin et al. 2006; Nguyen et al.2008*).

There are several factors that relate to failure of solid liquid separation. One of the most commonly recognized is the proliferation of filamentous bacteria that grow in long strands that become intermeshed with the floc particles and affect sedimentation. Although some number of filaments is necessary to give strength to the floc, acting as a backbone that holds the floc together, proliferation can yield bulking which prevents proper settling. Factors which impact proper bioflocculation are discussed in the next section.

1.3 FACTORS AFFECTING BIOFLOCCULATION

Bioflocculation, the process of microbial aggregation to form activated sludge flocs, is dependent on the interaction of exocellular polymeric substances (EPS) to form the matrix that holds microbes, other organics and inorganic particles in a flocculent matrix that is activated sludge. EPS can be found attached to microbial cell surfaces or free in solution. The EPS network in which microbes are embedded is described as highly hydrated and is 'sticky' so that other material, such as particulates, humics and some dissolved substances can become enmeshed into the activated sludge flocs. Research has shown that the mass of EPS in the activated sludge flocs can be up to 80% of the mass of activated sludge (*Sanin et al. 2006; Higgins et al. 2002*). Ideal bioflocculation has EPS interactions which produce highly hydrated flocs with good settling and dewatering properties; however; several operational and physicochemical parameters affect this process.

1.3.1 The Role of Cations: Research has shown that the activated sludge flocs have an overall net negative charge and this has been attributed to the functional groups of the EPS. Researchers suggested that divalent cations played a critical role in forming the exocellular matrix, by complexation and ionic bonding between bacteria and the EPS surfaces (*Busch et al.1968; Bruus et al. 1992*). Consequently the role of cations in bioflocculation has been widely studied within recent times, with reference to both municipal and industrial waste streams. Higgins and Sobeck (2002) investigated three theories to explain the role of cations in bioflocculation. The results were summarized as follow:

- **The Double Layer Compression Theory:** Also called the Derjaguin, Landau, Verwey, and Overbeek (DLVO) Theory after the people who proposed it, this theory suggests cations improve flocculation by compression through charge neutralization of the double layer that surrounds particles. This compression reduces repulsive forces between particles, allowing increased contact. This theory was disproved by the work of Higgins and Sobeck (2002) due to noted conflictions with the results of monovalent versus divalent cation addition and their impacts on settling and dewatering properties of activated sludge. Specific reference was made to the impact of Na^+ in the deterioration of sludge settling and dewatering properties. These observations were supported by the work of Bruus et al (1992) as well as Higgins and

Novak (1997). Therefore the DLVO theory could not be used to explain this phenomenon since it suggests that addition of monovalent ions would improve bioflocculation.

- **The Alginate Theory:** First proposed by Bruus et al (1992), this theory suggests that alginate gels, produced from bacterial polysaccharides in the presence of Ca^{2+} drive bioflocculation. These authors showed displacement of Ca^{2+} from the EPS matrix by Na^{+} and Mg^{2+} ions, which resulted in deterioration in settling and dewatering properties. They therefore concluded that the EPS matrix has a greater affinity for Ca^{2+}, supporting the role of Ca^{2+} specific alginate in bioflocculation. However research conducted by Higgins and Sobeck (2002) showed similar improvements in addition of calcium or magnesium to the feed on bench scale reactors. The only exception reported was that Ca^{2+} showed greater increase in cake solids than Mg^{2+}. An explanation for this was provided through the work of Forster et al (1980) which showed that Ca^{2+} reduced the bound water content of flocs, whereas as Mg^{2+} did not. Additionally, Murthy et al (1998) showed Mg^{2+} was a suitable alternative to Ca^{2+} in field application to improve settleability and dewaterability at an industrial waste treatment plant. This disputes the specificity of Ca^{2+} for alginate gel production and bioflocculation proposed by this theory.

- **Divalent Cation Bridging Theory (DCBT):** This theory suggests that non-specific binding of divalent cations bridge the negatively charged functional groups within and between EPS stabilizing the biopolymer matrix and consequently the activated sludge floc. This theory was supported by the work of Higgins and Novak (1997) where they postulated that the ratio of monovalent to divalent cations, called the Monovalent to Divalent Cation Ratio (M:D) ratio, impacts sludge settleability and dewaterability. They defined this ratio as being the sum of monovalent ions (Na^{+}, NH_4^{+} and K^{+}) divided by the sum of divalent cations (Ca^{2+} and Mg^{2+}) expressed on as equivalents, and found that if this ratio exceeded 2, then there was a deterioration in floc properties (*Higgins and Novak. 1997*). Higgins and Sobeck (2002) showed that addition of Ca^{2+} and Mg^{2+} caused similar effects on settleability and dewaterability of activated sludge flocs. They used the DCBT to explain the results of Na^{+} trials, where Na^{+} displaced divalent ions from the EPS matrix, resulting in a lack of bridging and deterioration of the activated sludge flocs. Consequently it was concluded that the M:D ratio is an appropriate indicator of floc properties based on cationic concentrations.

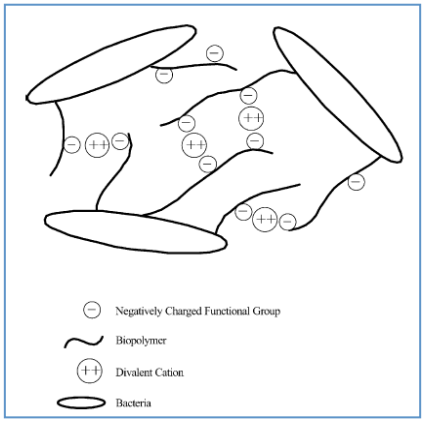


Figure 1: Depiction of the Divalent Cation Bridging Theory (Higgins and Sobeck.2002)

Use of the DCBT to explain stabilization or de-stabilization of the EPS matrix has been widely accepted (*Higgins and Sobeck 2002; Murthy et al. 1998; Novak et al. 1998*), elucidating the

importance of the M:D ratio as a sludge property indicator. The effects of monovalent and divalent cations are explained in greater detail below.

1.3.1.1 Monovalent Cations: Poor floc formation in activated sludge systems has been largely attributed to high concentrations of Na^+. High Na^+ concentrations results in a deterioration of sludge quality, observed by measuring settling properties such as floc density, capillary suction time (CST), sludge volume index (SVI). A corresponding deterioration in effluent quality is seen in the form of higher effluent solids (TSS) and COD, as well as increased effluent proteins and polysaccharides in the effluent (*Murthy and Novak, 2001*). This increase in the TSS is considered to be a sign of weak floc and it is attributed to a decrease in the bonding strength of exopolymer functional groups. Research suggests that at values lower than 10 meq/L sodium did not greatly impact the settling properties of activated sludge, but at concentrations higher than 10 meq/L there may be problems related to poor settling (*Higgins and Novak, 1997(c)*). Other authors suggest that problems related to settling and dewatering occur in activated sludge systems when the M:D ratio exceeded 1 and deteriorated significantly when the ratio exceeded 2. It was suggested that the poor settling and dewatering can be improved by raising the concentration of divalent cations, specifically Ca^{2+} and Mg^{2+} (*Higgins and Novak, 1997(a))*.

This is an important consideration particularly in industrial waste treatment, where Na^+ is present in the influent resulting from use of sodium based chemicals in manufacturing, and NaOH is often used to neutralize the characteristic low pHs. Drawing reference to the DCBT, Higgins and other researchers (2004) suggested improvement of bioflocculation in such situations by either **1)** direct addition of divalent cations, specifically Ca^{2+} and Mg^{2+}, or **2)** use of alternatives to Na^+ based chemicals, such as $Ca(OH)_2$ or $Mg(OH)_2$ for pH neutralization. The use of either application is based on the source of Na^+ ions. Field trials of both applications were successful, by bringing down the M:D ratio.

Other monovalent cations of interest in activated sludge include potassium (K^+) and ammonium (NH_4^+). The former, usually present in low concentrations, is a key component in cellular enzymes used for protein synthesis and therefore its effects on sludge properties cannot be explained simply by the M:D ratio. Studies showed that increasing concentrations of potassium, though causing poor dewaterability, produce higher strength flocs with good settleability, unlike with Na^+ ions (*Novak et al. 1998; Murthy et al. 1998; Park et al. 2006*). The ammonium ion was found to have adverse effects on sludge dewaterability, specifically CST and cake solids, accompanied by expulsion of soluble proteins in the effluent (*Murthy et al. 1998*). Ammonia is a principle constituent in influent waste streams and is usually converted to nitrate through nitrification by maintaining long enough SRTs to achieve this.

1.3.1.2 Divalent cations*:* The importance of divalent cations to the settleability and dewaterability of activated sludges can be elucidated from the preceding information. In summary, researchers concur that the divalent ions, specifically Ca^{2+} and Mg^{2+} ions, are critical

to good bioflocculation, and addition of these ions to activated sludge will continually improve floc formation. Research has also shown that some activated sludge plants require equimolar concentrations of Ca^{2+} and Mg^{2+} for efficient settling and dewatering. The concentrations of these required for good bioflocculation ranged from 0.72-2.0 meq/L each (*Higgins and Novak. 1997(c)*).

1.3.1.3 Multivalent Ions: The role of multivalents, specifically trivalent ions such as Fe^{3+} and Al^{3+} have been much less investigated than monovalent and divalent cations, though they are often found occurring in high concentrations in activated sludge. Compounds of these are used as coagulating and phosphorous removal agents in wastewater treatment, and researchers suggested that due to the higher charge valence, they may contribute greater floc stability (*Park et al. 2006*). A study conducted by Murthy and Novak (1998) on the cationic impacts on activated sludge effluent quality showed that iron-hydroxy compounds were important to the bioflocculation, by maintaining floc structure and retaining proteins within the EPS matrix, thereby reducing effluent COD. They postulated that better flocculation in municipal plants compared to industrial plants with similar M:D ratios was due to the higher iron content in the former. They suggested that addition of iron in conjunction with divalent cations could improve effluent quality in industrial waste treatment. Further investigation by Park et al (2006) found that for industrial waste streams with high Na^{+} concentrations, the adverse impacts on dewatering and effluent quality were offset by addition of Al^{3+} and Fe^{3+}. Specific findings showed an inverse relationship between effluent biopolymer and the sum of Al^{3+} and Fe^{3+}. Additionally they found that the ratio of Al/Na expressed as equivalents could be used as an indicator of sludge properties; increases in this ratio would yield improved sludge quality.

1.3.2 Kinetic Selection Theory and the role of filamentous organisms: Microbes involved in bioflocculation can be classified into two broad categories: floc formers and filamentous microbes. Both are required for proper flocculation. The latter provide a strong backbone for proliferation of flocculated bacteria, producing a large, dense, compact floc with good settleability. If the filamentous bacteria grow and extend beyond the biofloc, the particles compact poorly, increasing the sludge volume index (SVI) and negatively impacting the solid-liquid separation process. This produces a filamentous bulking sludge, which has been a continued nuisance to plant operators in activated sludge systems. An SVI of 150 mg/L is used as an indicator of the onset of bulking sludge (*Grady et al. 1999; Martins et al.2003*).

Settling problems in activated sludge are often attributed to an imbalance in the ratio of floc formers to filamentous microbes. Several factors affect the proliferation of filamentous microbes. These include low pH, low dissolved oxygen (DO) content, low Solid Retention Times (SRTs) and low substrate concentrations. Proliferation of filamentous microbes is controlled by imposing kinetic selection, in which conditions are provided to give floc formers the competitive advantage. One mechanism used to control proliferation of microbes is the Selector Activated Sludge (SAS) process, in which a selector is used upstream of the bioreactor to allow

proliferation of floc formers. This is achieved by imposing a high Process Loading Factor, which is explained further in the subsequent section. The impact of SRT is also important, since this has largely replaced the process loading factor as the principle design consideration in activated sludge treatment.

1.3.2.1 Substrate Loading Rate and the use of Sequencing Batch Reactors (SBRs): The Process Loading Factor (U), also called the Food to Microorganism (F/M) Ratio is defined as the mass of substrate applied to the reactor per unit time divided by the mass of microbes in the reactor. More specifically the F/M ratio is actually a measurement of the amount of incoming food measured as mass of influent Biochemical Oxygen Demand (BOD) divided by the mass of microorganisms in the system, measured as Mixed Liquor Volatile Suspended Solids (MLVSS). Prior to the use of SRT, the F/M ratio was the basic independent variable used in the design of CSTRs. The same concentration of substrate can be made available to biomass in a short period, characterized by a strong substrate gradient, or over a long period, characterized by a low substrate gradient. This gradient can be manipulated to minimize the proliferation of filamentous organisms through creation of a substrate concentration gradient at the bioreactor inlet that favors the growth of floc formers over filamentous organisms. According to the kinetic selection theory, explained previously, floc formers favor strong substrate gradients. The behavior of floc formers and filaments at different concentrations is explained by using the Monod equation, which is defined below:

$$\mu = \hat{\mu} \frac{S}{K_S + S} \qquad \textbf{(Eq. 1)}$$

Where

S = Substrate Concentration (mg/L)

μ = Specific growth rate coefficient

$\hat{\mu}$ = Maximum specific growth rate

K_S = Half saturation coefficient for subtrate

Some researchers believe that when the substrate concentration is low, based on the Monod equation, filamentous organisms have higher μ and K_s values than floc formers and therefore win the competition for substrate. This is true of CSTR systems, where the feed rates are low and continuous, producing a low influent and reactor concentration. The reverse is true if a substrate gradient is allowed in a way that the influent concentration is high, since floc-forming bacteria can now grow at the expense of the filamentous bacteria. A configuration of this type can be obtained in many different ways; for example, a reactor can be set to work like an ideal plug-flow reactor (PFR), or by adjusting the feeding pattern to obtain the higher substrate loading rate. This rate is usually much faster and shorter than CSTR, similar to pulse feed. These systems have been found to produce better settling sludge than the former.

Another theory of the competitive advantage of filaments at low substrate concentrations is that of diffusion limitation favors proliferation of filaments, due to their lower diffusional resistances

than in flocs. Researchers state that this hypothesis is sufficient does not require intrinsic kinetic difference between these two groups (*Martins et al. 2003; Dionisi et al. 2006*).

Laboratory simulation of these systems has successfully been achieved by using Sequencing Batch Reactors (SBRs), which are those reactors operated in a sequence of steps. Typical SBRs follow a cycle of Feed/Fill- React- Settle- Decant. SBRs are chosen in many laboratory models because they can be easily manipulated to reproduce many processes if the SRT and HRT (hydraulic retention time) are the same. The fill period of an SBR is an important feature since its length can be varied to allow the SBR to operate in a range between a PFR and a CSTR. Changing the fill period influences the Process Loading Factor and substrate gradient as described previously. A high process loading factor is achieved by a short fill period, whereby the biomass in the reactor receives a high initial amount of organic matter and nutrients; in this case the SBR will be analogous to a plug flow reactor. Conversely, for a longer fill period the process loading factor is small and the SBR will be analogous to a completely mixed reactor (*Grady et al., 1999*). Most of the reactions such as substrate utilization and biomass growth will take place during the filling period. Increasing the length of the feed creates a low substrate concentration, which can negatively affect the settling properties of the activated sludge. When the fill time is short, a high substrate gradient is present which promotes the substrate intake close to the maximum specific rate of bacteria, leading to good settleability. These conditions affect the ratio of floc-forming bacteria to filamentous organisms as described previously, and this is consistent with the idea that the feeding pattern had an influence on the microbial population dynamics and kinetics of activated sludge (*Martins et al., 2003*). Feed patterns, particularly as they are used in operation of SBRs, have been found to impact the settleability and dewaterability of sludges (*Martins et al 2003; Dionisi et al. 2006*).

1.3.2.2 The Effect of Solid Retention Time (SRT): Due to the importance of the particulates in the bioreactors, designers define SRT or Mean Cell Residence Time (MCRT) as being the mass particulates in the bioreactor divided by the mass discharged per unit time. This parameter is considered the most important design factor in determining the performance of the activated sludge system. SRT influences numerous factors in the activated sludge process, such as nitrification. Research has shown that a minimum SRT is required for bioflocculation; generally SRTs in excess of 2 days are used for achievement of efficient bioflocculation. SRTs have been found to affect floc macrostructure, by influencing the relative proportion of floc-forming bacteria and filamentous bacteria present in the floc. Studies show that low SRTs between 0.25 and 2 days produced dispersion of suspended growth biomass resulting in inadequate flocculation. Conversely, relatively high SRTs between 9 and 12 produce irregularly shaped pin point floc. The SRT is determined by the origin and nature of the influent biomass; activated sludge systems usually have long SRTs to stabilize entrapped biomass and particulates or to degrade xenobiotic or slowly biodegradable organics (*Grady et al., 1999*). The latter is generally the case in industrial waste treatment, which could be a possible reason for poorer flocculation and effluent quality associated with many of these.

Researchers using bench scale SBRs showed that increasing SRTs improved sludge dewaterability (*Sanin et al. 2006; Feng et al. 2009*) Sanin and other researchers found that as the SRT increased, the polymer protein to carbohydrate ratio of the EPS matrix increased, which is believed to improve flocculation and consequently dewaterability. Feng et al (2009) investigated the impact of SRTs on hydrolysis of WAS and found that as the SRT increased, the hydrolysis of the sludge increased until a point when hydrolysis became stable. Therefore one can conclude that higher SRTs improve sludge dewaterability until a point of stabilization, beyond which changes are minimal.

Considering the factors listed above that affect bioflocculation it is important to remember that these parameters seldom operate independently of each other. It is therefore important when doing bench scale simulations to consider several parameters, in order to be representative of field scale scenarios and ease the ability to scale up from lab to field trials.

CHAPTER 2. INTRODUCTION

From the literature reviewed it can be elucidated that bioflocculation is the principle factor influencing solid-liquid separation in wastewater treatment systems. The success of activated sludge systems is measured through the ability to develop a flocculent biomass that settles and compacts efficiently in the secondary clarifier. Failure to achieve this result in process failure, represented by poor quality effluent and sludge (Grady et al. 1999). It has also shown that a number of factors affect bioflocculation. The role of cations has been well documented and thoroughly investigated, as evidenced by the now widely accepted concept of the M:D ratio for achievement of proper flocculation (Higgins and Novak. 1997). Two other factors, which are also important in their influence but less investigated, are the effect of substrate loading rate and SRT.

The substrate loading rate is believed to directly impact the growth of filamentous and floc forming bacteria and therefore influences the nature of the sludge and effluent. Most bioflocculation problems can be related directly to the imbalance between these two microbial groups. Poor bioflocculation can result from dispersed growth, where bioflocculation is incomplete and biomass does not settle but is carried into the effluent producing a poor quality effluent. Inadequate filamentous bacteria result in pin point flocs which shear easily since there are insufficient filaments to form a strong backbone for the flocs. Excessive filaments lead to bulking sludge, which settles very slowly and compacts poorly, leading to excessive RAS flow rates.

It is believed that high substrate loading rates favor the growth of floc forming bacteria and low substrate loading rates favor the growth of filamentous bacteria. Manipulating the substrate loading rate is generally best achieved by manipulating the rate at which substrate is fed to the bioreactor. Manipulation of this feed rate is generally of two types: fast feed rates or pulse feed,

reminiscent of a PFR configuration and slow feed rates reminiscent of a CSTR configuration. The postulated relationship between the substrate loading factor and relative growth of floc formers and filaments leads to the belief that slow feed or CSTR bioreactor configurations would produce a poorer quality sludge and effluent than a PFR bioreactor configuration (*Grady et al. 1999; Martins et al. 2006; Verachtert et al. 1973*). In their research, Martins et al (2006) concluded that in highly dynamic systems such as wastewater treatment plants, only PFR configurations can achieve a strong substrate concentration gradient and properly control sludge settleability. Bench scale manipulation of these systems is often successfully achieved using SBR systems and adjusting the length of the fill period; shorter fill periods, or fast feed rates, produce higher substrate gradients, as with PFR configurations, and longer fill periods or slow feed rates produce lower substrate gradients as seen in CSTR systems (*Dionisi et al. 2006*).

SRT is the operational parameter that replaced the process loading factor as the key design parameter in activated sludge plants. In terms of its effect on bioflocculation it is believed that a minimum SRT is required in order to achieve efficient bioflocculation. This theory has been thought to suggest that a minimum SRT represents a balance between the rate of bacterial growth and the rate of production of EPS to achieve flocculation. At low SRTs the rate of growth exceeds the rate of EPS production and bioflocculation is incomplete resulting in system washout since the microbes are wasted from the system faster than they grow. At higher SRTs the quantity of EPS produced is increased and bioflocculation improves. The minimum SRT, defined as the value below which a specific group of microbes is unable to grow in suspended growth bioreactors, for activated sludge systems has been suggested to be 3 days. Plants have however successfully operated at SRTs as low as 1 day (*Grady et al. 1999*). This has been linked to the presence of active biomass and EPS in the influent which alters the relationship between SRT and biomass specific growth rates, since the biomass in the reactor no longer needs to grow as fast to maintain itself, allowing operation at lower SRTs (*Grady et al. 1999*). The SRT of choice generally depends on the objective of the plant. Nitrification SRTs are usually high due to the low specific growth rate of autotrophs, although nitrification has been found to occur at SRTs as low as 2 days. SRT is largely impacted by temperature, depending on the application. Some plants have chosen SRTs as low as 1 day to avoid nitrification and the oxygen demand associated with it. However, the SRT to be chosen in this case is site specific since nitrification is temperature dependent and the required design SRT has been found to vary from site to site (*Metcalf and Eddy. 2003; Barker and Dold. 1997; Fillos et al. 2000*). Substrate complexity additionally impacts the design SRT; municipal plants usually have lower SRTs than industrial plants, due to the greater complexity of the substrate in the latter (*Grady et al.1999*).

2.1 USE OF SEQUENCING BATCH REACTORS (SBRS) TO MONITOR THE COMBINED EFFECT OF SRT AND FEED RATES OF REACTOR PERFORMANCE

Having clearly defined the importance of SRT and substrate loading rates in achieving successful activated sludge operation, it is clear that further investigation of these two is required. The purpose of this project was to investigate the combined effect of SRTs and substrate loading rate on the quality of sludge and effluent obtained in activated sludge systems.

Investigations were conducted using bench scale SBR simulations and substrate loading rate was equated to feeding patterns or rates. The latter was manipulated by adjusting the length of the SBR fill cycle; a short fill period was used to achieve a high substrate loading rate, or PFR configuration, whereas a longer fill period was used to achieve a low substrate loading rate, or CSTR configuration. The PFR configuration was termed **fast feed** and the CSTR was termed **slow feed**. The SRTs chosen for this investigation ranged from 2 to 10 days, which was considered to be an operating range representative of most full scale wastewater treatment plants. The manipulation of SBRs to achieve desired SRTs and substrate gradients is described in the materials and method section.

2.1.2 Overall Goal and Objectives

The overall goal of this project was to determine if the effluent quality and sludge characteristics, specifically settling and dewatering properties, of bench-scale SBR reactors are different when subjected to different feeding patterns at given SRTs. This goal is broken into more specific goals and objectives below.

Effluent Quality Goal: To determine if the effluent quality for the fast feed configuration was better than that produced by the slow feed at all SRTs operated.

Effluent Quality Objectives: to use SBR simulations to:

- compare the effluent quality of fast feed systems versus slow feed systems for specific SRTs using SBR simulations by measuring and comparing specific effluent quality parameters at defined intervals during operation.

- compare the behavior of fast feed versus slow feed systems across a representative operating range of SRTs using SBR simulations.

Sludge Quality Goal: To determine if the sludge quality of the fast feed configuration was better than that produced by the slow feed at all SRTs operated

Sludge Quality Objectives: to use SBR simulations to:

- compare the sludge quality of fast feed versus slow feed systems for specific SRTs by measuring and comparing specific settleability and dewaterability parameters at defined intervals during operation.

- compare the settleability and dewaterability of fast versus slow feed systems across a representative operating range of SRTs.

2.2 MATERIALS AND METHODS

2.2.1 EXPERIMENTAL DESIGN

Laboratory scale sequencing batch reactors (SBRs), each with an operating volume of 6 L were used to study the effect of feeding pattern on activated sludge performance. The reactors were all seeded with 3L of Return Activated Sludge (RAS) from the Blacksburg/VPI Municipal Wastewater Treatment Plant. Seeding was done to ensure that MLSS concentration upon completion of the first feed cycle was ~2500 mg/L. For this study, three different SRTs were used, 10 Days, 5 Days and 2 Days. The SRTs were achieved by manipulating the solids wastage rate (F_w) such that it was equal to the operating volume of the reactor (V) divided by the desired SRT(θ_c).

$$F_w = V/\theta_c \qquad \textbf{(Eq.2)}$$

For each SRT two SBRs were operated each with a different feeding pattern, to generate a pulse feed or PFR and a continuously mixed or CSTR configuration. The PFR or fast feed reactor was operated with a fill period of 5 minutes. The CSTR or slow feed reactor was operated with a fill period of 3 hours. Both the fast and slow feed reactors were fed the same volume during their respective fill period. All SRTs used the same operating volume. Reactors were mixed using vigorous diffused aeration using air stones, in order to maintain a dissolved oxygen concentration above 2 mg/L. Solids wastage was conducted during the last 5 minutes of the aeration cycle; the flow rates were dictated by the SRT, as described previously. Each operation cycle culminated with a one hour non-aeration period, consisting of a 40 minute settle period followed by a 20 minute decant period. The common characteristics of the operating cycles by SRT are shown in Table 2.2.1(a) below.

Table 2.1: Operational Characteristics of SBRs used in this experiment

Operational Parameter	SRT (Days)		
	10	***5***	***2***
Operating Volume (L)	6	6	6
F_w(L/Day)	0.6	1.2	3
# of Cycles per day	2	2	3
Cycle Length (hours)	12	12	8
Aeration Cycle (hours)	11	11	7
Hydraulic Retention Time (HRT)/Days	1	1	0.5

2.2.1.1 *2Day SRT Bench Scale Design*

For the 2 Day SRT bench scale reactor, achieving the required SRT by solely adjusting the wastage rate would have resulted in an inadequate mass of solids to carry out the study objectives. To ensure that there were sufficient solids present at the end of operation to run batch digestion and odors analyses, the aeration cycle lengths and influent COD were manipulated to maintain a steady state MLSS of approximately 1800 mg/L. The cycle lengths and influent COD were determined using the following equation from *Grady et al. 1999*:

$$X_T \approx (\theta_C/HRT).\ Y_{obs}\ (S_0 - S) \qquad \textbf{(Eq. 3)}$$

Where $\mathbf{X_T}$ = Total Biomass Concentration (mg/L)

HRT =Hydraulic Retention Time

$\boldsymbol{\theta_C}$= Solid Retention Time (SRT/days)

$\mathbf{Y_{obs}}$ = Observed Yield (0.5 in this case)

($\mathbf{S_0}$-**S**)= COD (mg/L)

From the calculations it was determined that increasing the influent COD to 700mg/L and using an HRT of 0.5day or a flow rate (Q) of 12L/day should maintain a steady state MLSS of approximately 1800mg/L. Consequently the 2 Day SRT experimental set up was manipulated to have three 8 hour cycles per day. The aeration cycle lengths were decreased from 11 hours to 7 hours and F_w was increased to 3L/day. All other cycle lengths remained the same.

2.2.2 SYNTHETIC FEED COMPOSITION

The feed used in this experiment was a synthetic wastewater comprised of bactopeptone, which is a microbiological enzymatic digest of protein, acetate and cationic feeds as shown in the table below. The concentrations of the cations in the feed were taken from Cubas (2006) and adjusted for this experiment in order to maintain an M:D ratio as close as possible to 1:1. This ratio was found to be an ideal M:D operating ratio in previous studies, to ensure proper floc formation and settling (Higgins, 1997). For this reason the potassium ion (K+) concentration is higher than that used by Cubas (2006). The cation stock concentrations used are shown in the table 2.2.3.

Table 2.2 Cation Feed Concentration

Cation	Concentration (meq/L)	Compound used	Desired Influent Concentration (mg/L)
Ca^{2+}	1.5	$CaCl_2.2H_2O$	110
Mg^{2+}	1.5	$MgSO_4.7H_2O$	185
K^{+}	0.5	$KHSO_4$	228.48
Fe^{3+}	0.3	$FeCl_3$	17.5
Al^{3+}	0.18	$Al_2(SO_4)_3.18H_2O$	37

Adapted from Cubas.2006

The total COD of the feed was maintained at 350mg/L, except in the case of the 2Day SRT reactor as explained previously. The contributing COD reagents were bactopeptone and acetate, which were mixed together in a 3:1 ratio. The acetate source used was sodium acetate, except in the case of COD adjustment for the 2Day SRT reactors, where the acetate used was supplemented with calcium acetate. This was done to ensure the maintenance of a 1:1 M:D ratio. Bactopeptone was used on a mass equivalent, assuming that 1mg of Bactopeptone was equivalent to 1mg of COD. COD equivalents of sodium and calcium acetate were done using mass balance with chemical equivalents.

2.2.3 Sample Analyses

Effluent and sludge quality were measured using the following parameters, sampling frequencies and test procedures shown in table 2.2.2(a). Samples were collected throughout the operating phase and the datasets were used to define a steady state period as described in section 2.2.4. Effluent samples were collected during the decanting period, at the end of the operation cycle. Mixed Liquor samples were collected for both reactors during the aeration cycle, at the end of the slow feed fill period. The reactors were shut down when MLSS values declined to below 500 mg/L.

Table 2.3: Parameters measured and Sampling Frequencies

Parameter Measured		Sampling Frequency			Analysis Method
		10Day SRT	5Day SRT	2 Day SRT	
Sludge	*Sludge Volume Index (SVI)*	Every 3 Days	Every 2 Days	Every Day	2710D Standard Methods (1995)
	Capillary Suction Time (CST)				2710 G Standard Methods (1995)
	Total and Volatile Solids (TS & VS)				2540 G Standard Methods (1995)
	Total and Volatile Suspended Solids (TSS&VSS)				2540 D & E Standard Methods (1995)
	Zeta Potential	Single Measurement			Refer to sub-section 2.2.2.2(a)
	Soluble COD				Refer to sub-section 2.2.2.1(a)
	Odors				Refer to sub-section 2.2.2.1(d)
	Gas Production	Every Day			Batch Digestion –refer to sub-section 2.2.2.1 (b)
Effluent	*Total and Volatile Suspended Solids (TSS&VSS)*	Every 3 days	Every 2 days	Every Day	2540 D & E Standard Methods (1995)
	Total and Soluble COD				5220C Standard Methods (1995)
	Proteins				Hartree Modification of Lowry Method (1972)
	Polysaccharides				Dubois Method (Dubois et al 1965).

2.2.3.1 Additional Mixed Liquor Sample Analyses

2.2.3.1 (a) *Soluble COD Degradation*

Soluble COD was measured in the reactor by collecting samples at fixed intervals, commencing at the end of the fill period. Ten ml samples were taken every 10-15minutes for a period of three hours. The samples were collected using a syringe, and were then filtered through 0.45µm filter paper. The COD test was then conducted on these samples in accordance with Section 5220C from Standard Methods (1995). The soluble COD test was conducted once for each SRT after the reactor had reached steady state.

2.2.3.1 (b) *Batch Digestion*

At the end of the reactor run, aeration was shut off and the solids were concentrated by centrifugation. The thickened activated sludge was used to set up anaerobic batch digesters to measure biogas and odor production. The batch digesters were set in 250ml glass bottles and were seeded using 20% by volume of anaerobically digested sludge from the Pepper's Ferry Municipal Wastewater Treatment Plant. Batch digester initial total solid (TS) concentrations were maintained within a range of 1.5-2% solids and, upon initiation, the headspace was purged with nitrogen gas. Biogas production was measured daily over a 30day period, using a liquid column nanometer filled with a displacement solution comprised of an acid/salt solution as described in Standard Methods (1995). Gas production data was normalized by the solids concentration to provide a better comparison between fast and slow feed reactors for a given SRT.

2.2.3.1 (c) *Scanning Electron Micrographs (SEM)*

Mixed Liquor samples were observed using an FEI Quanta 600 scanning electron microscope, which was used in high-vacuum mode for this purpose. Samples were diluted in the following ratios: 1:10, 1:25 and 1:50. Dilutions were done using distilled water and samples were filtered through and fixed onto a 0.45 m nitrocellulose membrane filter. The filters were allowed to dry overnight in a desiccator. This was done on the 5 and 2 Day SRT reactors.

2.2.3.1 (d) *Odor Analysis*

Odor analysis was conducted using the headspace method developed by Glindemann et al. (2006). Dewatered sludge cake was incubated in a closed bottle and the headspace gas measured over time. Headspace gas was measured using an Agilent 6890 Gas Chromatograph/5973 Mass Spectrophotometer. The column was a DB-5 with a 0.25mm inner diameter and a 0.25µm film

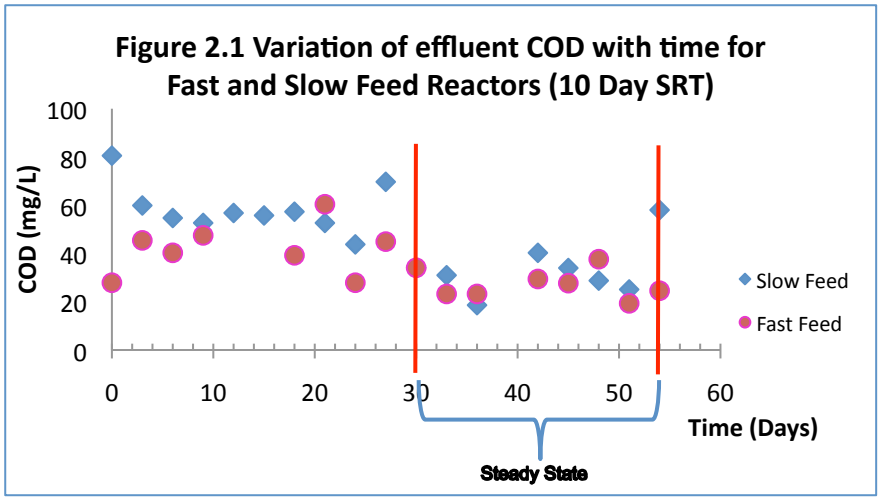
thickness. Biosolids were conditioned with 1% Clarifloc polymer, centrifuged and dewatered using a filter press. Cake solids were then placed in glass vials and sealed with septa. Odor analyses involved measurement of headspace gas concentrations. 25µl of gas was extracted from the vial and injected to the GC/MS through the sample port. Cryotrapping of the compounds was performed for two minutes, using liquid nitrogen. Once completed, the trap heater was engaged and the sample was heated to 260°C, until the end of the program. The peaks generated by the respective compounds were compared against standards to determine the concentrations. Compounds analyzed included Total Volatile Organic Sulfur Compounds (TVOSCs), Volatile Fatty Acids (VFAs) and *para*-cresol. Only the TVOSC data is presented in this paper; the long term odors were still being analyzed when this paper was written but will be presented in later research papers.

2.2.3.1(e) *Zeta Potential*

This was measured on the effluent but is included as a mixed liquor characteristic since it is used to analyze particle behavior. Zeta Potential was measured using the Zeta-Meter System 3.0. This system comprised a Zeta Meter 3.0+ unit, a microscope module and an electrophoresis cell. The microscope module and electrophoresis cell were both connected to the Zeta Meter 3.0+ unit. The undiluted effluent sample was poured into the electrophoresis cell as directed by the Zeta-Meter System 3.0 operating instruction manual. The appropriate voltage was chosen as 150V and particle movement was tracked using the Zeta Meter 3.0+ unit and the microscope module. The unit directly calculated zeta potential for the samples since they were aqueous.

2.2.4 STEADY STATE DETERMINATION

In order to determine when steady state was achieved in these systems, visual inspection of the data sets was used. It is a generally accepted rule for completely mixed systems that constant performance is observed after a period equal to the three times the SRT (3 SRTs), starting from the point of inoculation. This was observed to be the case for the 10 and 5 Day SRT reactor sets; the 2 Day SRT reactor appeared to achieve steady state faster. Cubas 2006 stated that "the system was considered to be at steady state when visual inspections of settling properties plotted as a function of time were stable and variability no greater than ≈ 25 % occurred". Figure 1 below is used to show how steady state was determined for each data set, using visual inspection of the plotted data. This latter method is represented in Figure 2.1 below.



In this case, steady state was considered to be between 30 and 54 days for effluent COD in the 10 Day SRT Reactor set. For the 5 Day and 2 Day SRT reactors, steady state was found to begin at 12 and 5 days respectively. Steady state datasets consisted of approximately 10 points for the 10 Day SRT systems, 12 points for the 5 Day SRT and 10 points for the 2 Day SRT systems.

2.2.5 Red Worms

The Fast Feed 10 Day SRT reactor developed a red worm infestation between days 12 and 15 of operation. The worms appeared as a bright red layer at the top of the sludge blanket during the settling phase. To get rid of them, aeration was shut off and the reactors were sealed to provide anaerobic conditions. The worms, being obligate aerobes, could not survive such conditions. After approximately 2-3 days of anaerobic conditions, the reactor was unsealed and aeration was restarted. After approximately 1day the reactor was back to normal working order and no red worms were observed through the rest of the run. There were no red worms in the slow feed 10 Day SRT reactor, nor in any of the 5 Day or 2 Day SRT reactors.

2.2.6 Statistical Analyses

Steady state means were analyzed as follows. To compare fast and slow feed reactors for a single SRT, t-tests were used. Comparison of the variability between fast and slow feed reactors, across the three SRTs was conducted using a Two Way ANOVA test, followed by a Tukey HSD test as a backup to the t-test to see which datasets exhibited significant differences. Statistical analyses were conducted using the R software package and a significant difference was considered when a p value less than 0.05 was obtained. Visual representation was achieved through use of boxplots, produced in R, as well as bar charts and line graphs produced in Excel. For the boxplots, the boxes represented the interquartile data range, and the whiskers represented the extremes of the data range.

CHAPTER 3.0 RESULTS AND DISCUSSION

All parameters measured were discussed in terms of comparison at specific SRTs and behavior across SRTs.

3.1 EFFLUENT QUALITY

3.1.1 Effluent Total and Volatile Suspended Solids

The following three boxplots show the effluent TSS for each of the SRTs used.

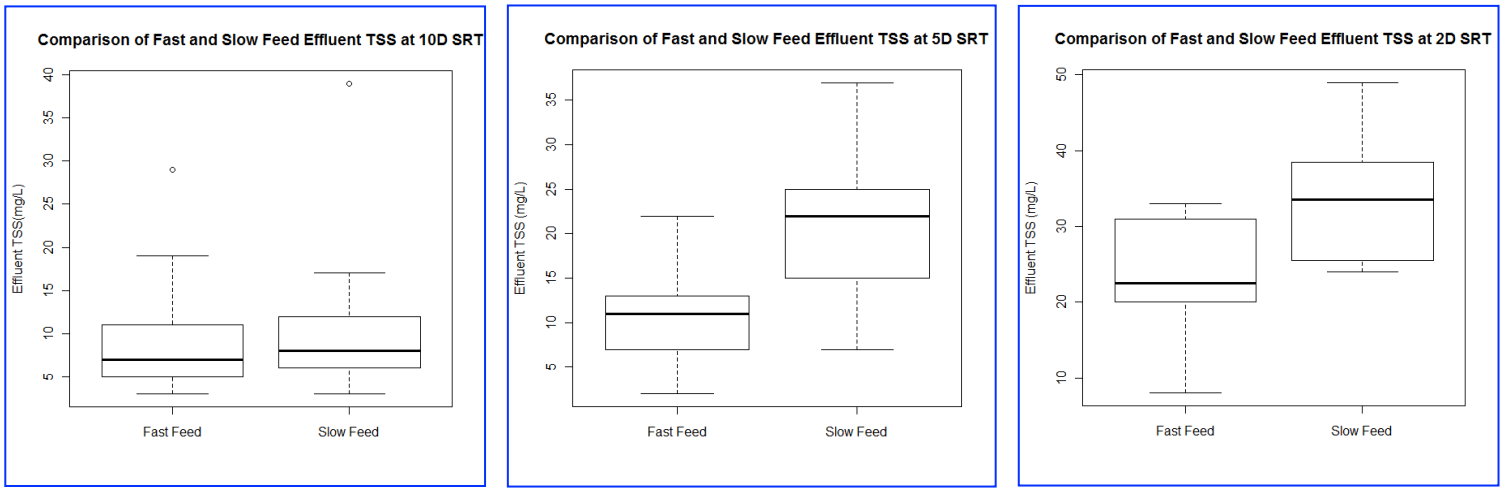
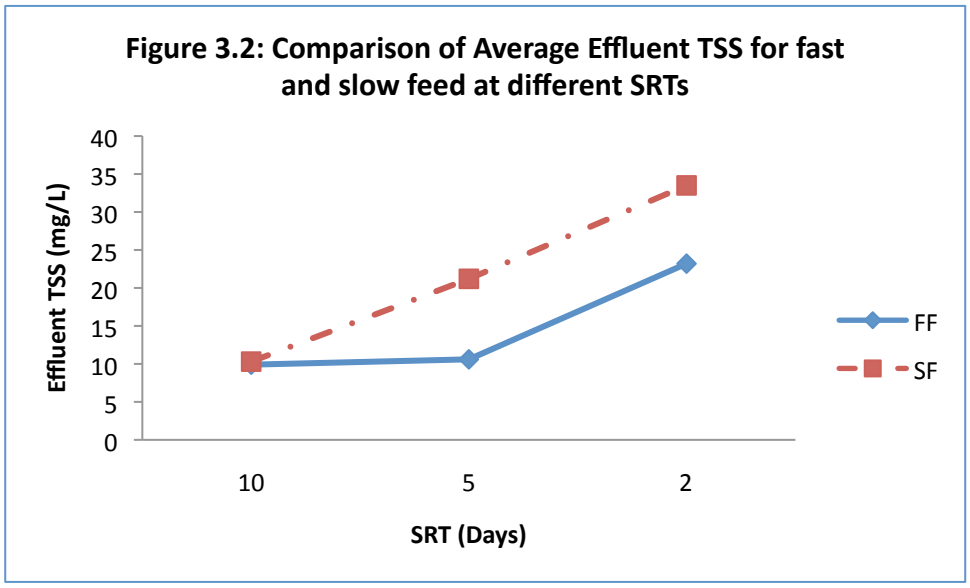


Figure 3.1: Boxplot comparison of fast versus slow feed effluent TSS at 10 Day, 5 Day and 2 Day SRTs respectively

Visual comparison showed no significant difference between fast and slow feed at the 10 Day SRT though the fast feed effluent TSS average appeared to be slightly lower than the slow feed. Significant differences were however observed at SRTs of 5 and 2 days. For the latter two cases the fast feed mean appears to be significantly lower. Individual t-tests were conducted for each SRT and it was found that for both 5 Day and 2 Day SRTs the fast feed effluent TSS was significantly lower than the slow feed. This implies that the fast feed bioreactor solids settled better than the slow feed solids during the settle cycle. Laboratory observations revealed the effluent for all slow feed configurations to be much more opaque than fast feed effluent. On several occasions it was not possible to filter the effluent samples for the slow feed reactor; this happened often with the slow feed effluent samples at 2 Day SRT.

Looking at the data across SRTs, it was seen that the effluent TSS became progressively higher as the SRT decreased for both the fast and slow feed configurations. This implied that the settleability of both configurations decreased with shorter SRTs, perhaps due to system washout, particularly at the 2 Day SRT. Despite this, it was also observed that the slow feed effluent TSS

remained higher than the fast feed at all SRTs operated. This is depicted in figure 3.2 below which is consistent with the description of effluent TSS behavior described above. As the SRT was decreased from 10 to 2 days, the difference between the fast and slow feed effluent TSS increased, implying that the variability of the slow feed effluent TSS was higher than the fast feed effluent TSS; in other words, the slow feed effluent TSS showed a higher rate of increase with decreasing SRT than the fast feed.



A two way ANOVA was conducted on the effluent TSS data and the comparison showed a significant difference in effluent TSS between fast and slow feed reactors and between SRTs. There was also evidence of some interaction taking place between SRTs and reactor configurations, implying that there was a combined effect of these two parameters on the effluent TSS concentrations. A Tukey Test was conducted and the results concurred with the evidence provided in the earlier discussion, showing no significant difference in effluent TSS at a 10 Day SRT, but found significant differences between fast and slow feed at 5 Day and 2 Day SRTs. Based on these calculations within and across SRTs, it was therefore concluded that the effluent TSS was lower for fast feed than slow feed at each given SRT.

Comparing effluent VSS, the results were similar to that found in comparing effluent TSS. Visual comparison provided in Figure 3.4 shows that there are significant difference between fast and slow feed mean effluent VSS at 5 Day and 2 Day SRTs, as well as between the 10 Day and 2 Day SRTs, but there is little difference between the mean values at the10 Day and 5 Day SRT. At the 10 Day SRT the fast feed effluent VSS appeared to be higher; a difference was most observable at the 5 Day SRT. There was also a noticeable difference at the 2 Day SRT. Individual t-tests conducted on each dataset confirmed these observations. Statistical analysis of

the 10 Day SRT revealed no significant difference between fast and slow feed. Statistical analyses of the 5 Day and 2 Day SRT datasets showed that slow feed effluent VSS was higher. This trend was similar to that exhibited by the effluent TSS for fast and slow feed at all SRTs.

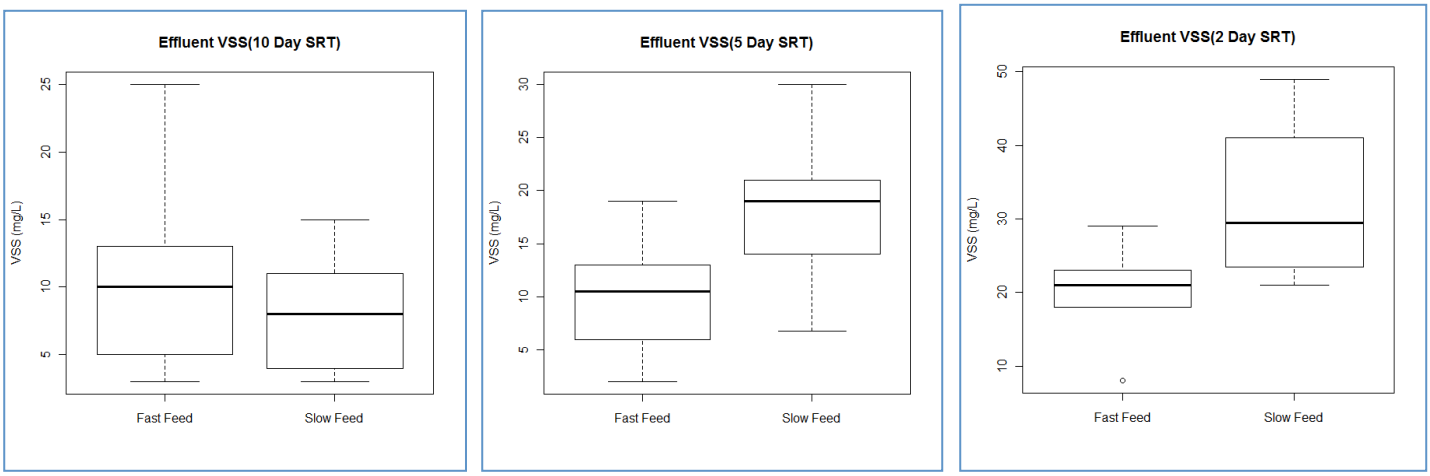


Figure 3.3: Boxplot comparison of fast versus slow feed effluent VSS at 10 Day, 5 Day and 2 Day SRTs respectively

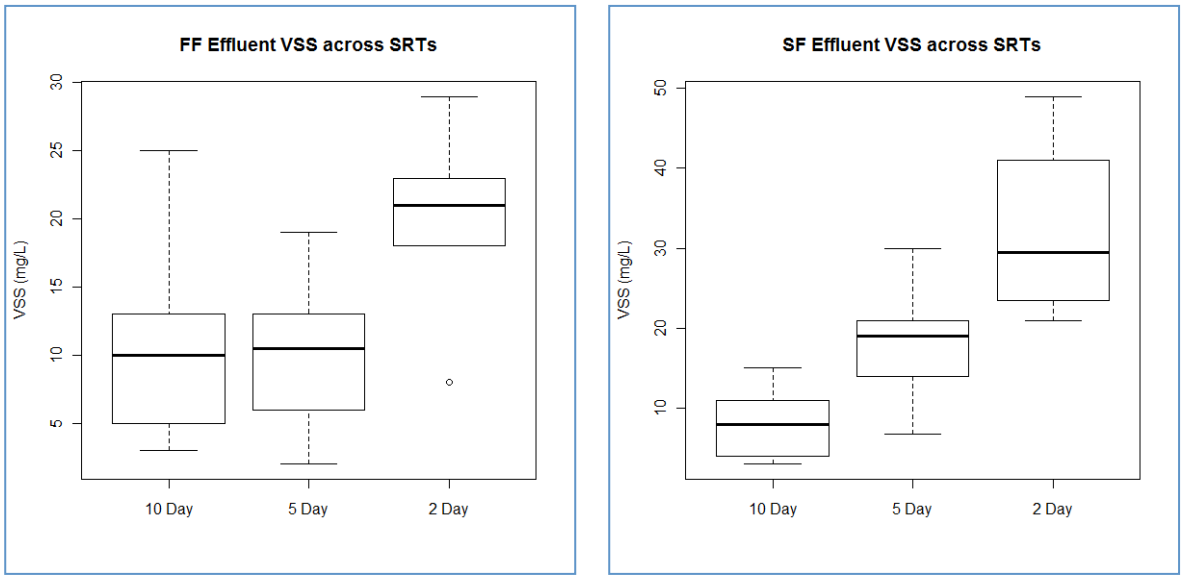


Figure 3.4: Boxplot comparison of fast feed and slow feed effluent VSS across SRTs

Visual comparison across SRTs, shown in Figures 3.4, revealed that effluent VSS was highest at the 2 Day SRT for both systems, and lowest at the 10 Day SRT for the slow feed system. The

fast feed 10 Day and 5 Day datasets were very similar. The trends observed here were almost identical to the behavior of the effluent TSS. A two way ANOVA was conducted on these datasets for comparison across SRTs and to show possible interactions between feed rates and SRTs. The results showed a significant difference in effluent VSS between SRTs and between fast and slow feed systems, with p-values less than α=0.05. There was also evidence of diagonal interaction between SRTs and reactor configurations. A Tukey Test was conducted for confirmation and the results showed significant differences between fast and slow feed systems at 2 Day and 5 Day SRTs. Based on all the analyses done here, it can be concluded that the effluent VSS was higher for the slow feed systems at lower SRTs and exhibited similar trends to the effluent TSS.

3.1.2 Effluent COD

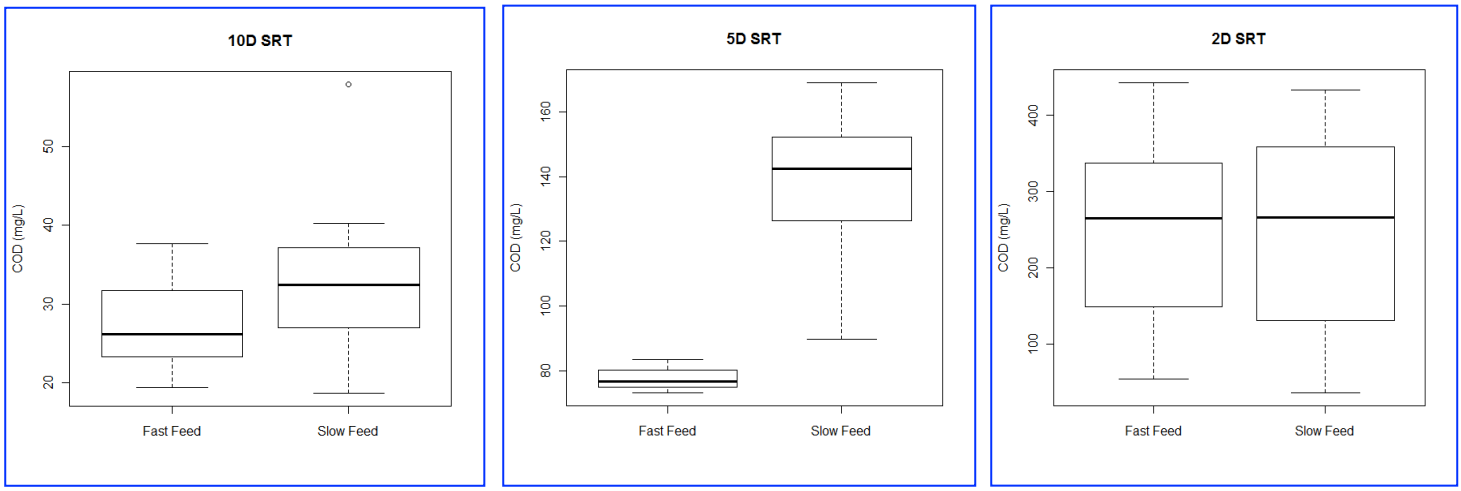
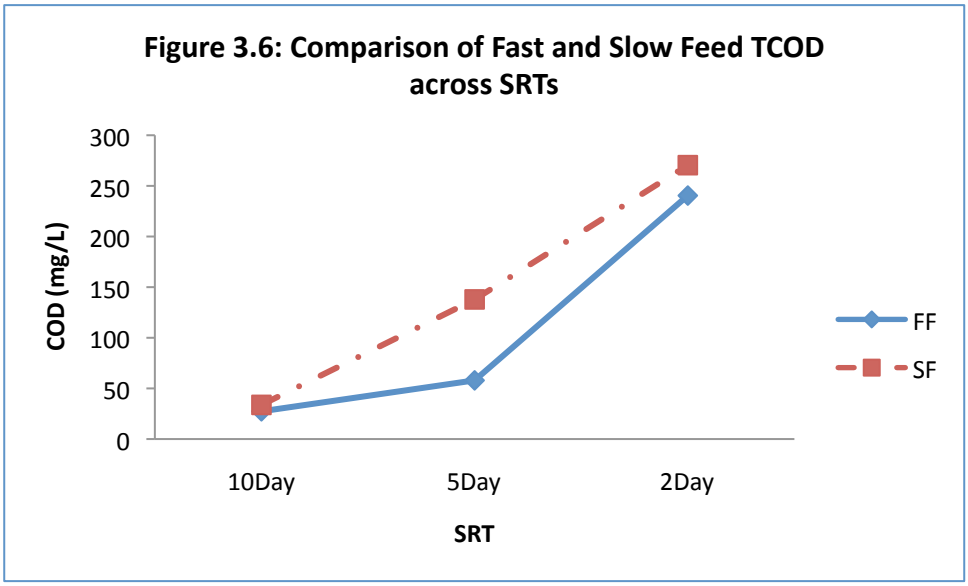


Figure 3.5: Boxplot comparison of fast versus slow feed effluent TCOD at 10 Day, 5 Day and 2 Day SRTs respectively

Visual comparison of total COD in Figure 3.5 showed that the effluent TCOD mean values showed no significant difference between fast and slow feed at the 2 Day SRT. At the 10 Day and 5 Day SRTs, however there was an observable difference in mean values, with the fast feed effluent TCOD being lower. The difference was largest at the 5 Day SRT. Individual t-tests run for each SRT revealed that there were no significant differences between fast and slow feed effluent TCOD at 10 Day and 2 Day SRTs; significant difference was found for the 5 Day SRT.

Comparison across SRTs showed that both fast and slow feed effluent COD increased with decreasing SRTs. It was also observed that the 2 Day effluent TCOD was significantly higher than at 10 or 5 Day SRTs for both fast and slow feed. One possible explanation was that a large

fraction of the biomass was being washed out into the effluent; this postulation is supported by observing that the trend in effluent TCOD across SRTs is similar to that observed for effluent TSS across SRTs. Biomass tends to be washed out at lower SRTs, since the rate of microbial growth exceeds the rate of production of EPS and therefore bioflocculation does not have sufficient time to occur. Figure 3.6 shows the increase in TCOD for fast and slow feed as the SRT decreased; it is observed that slow feed effluent TCOD was higher than for the fast feed systems at all SRTs operated.



A two way ANOVA was conducted to compare TCOD across SRTs, to observe if there were statistical differences between fast and slow feed systems, between SRTs and if there was any interaction between SRT and feed pattern. The results revealed significant differences in TCOD values between feed patterns and between SRTs, but no diagonal interaction was found between SRT and feed pattern. A follow up Tukey test was conducted and the results showed no significant differences between 10 and 2 Day SRT; there was significant difference between the 5 Day SRT datasets.

Comparing total and soluble CODs for each of the SRTs operated revealed that soluble COD represented more than 50% of the total COD for both the fast and slow feed systems. This implied that the total effluent COD was comprised largely of soluble microbial products, such as effluent biopolymers, with the remaining fraction being composed of larger suspended solids. The latter group is contributed as a result of high levels of deflocculation associated with low SRTs. The comparison between total and soluble effluent COD for fast and slow feed at each SRT is summarized in Figure 3.7 below.

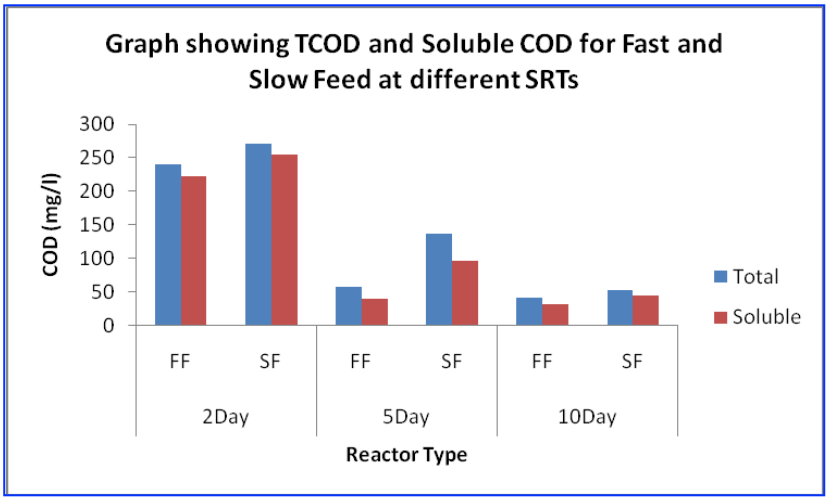


Figure 3.7: Graph comparing effluent TCOD and SCOD between fast and slow feed at different SRTs

Based on these analyses, it is concluded that effluent TCOD was observed to be higher and more variable at each SRT than for the fast feed system. Significant difference was only found between mean TCOD at the 5 Day SRT. Additionally, the effluent TCOD was comprised primarily of soluble COD (SCOD), implying the main constituents were soluble microbial products, with the non-soluble fraction being comprised of suspended solids produced by deflocculation.

3.1.3 Effluent Biopolymers

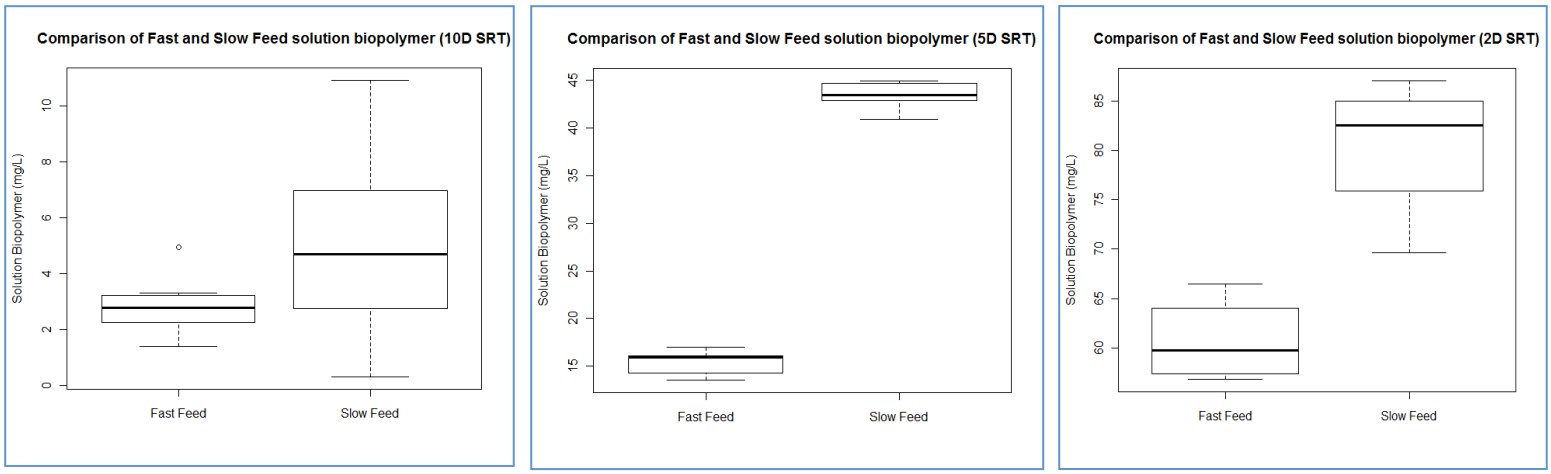


Figure 3.8: Boxplot comparison of fast versus slow feed effluent TCOD at 10 Day, 5 Day and 2 Day SRTs respectively

From Figure 3.8 it is seen that the mean solution biopolymer concentration is much lower for the fast feed system than the slow feed system for each SRT operated. The difference between fast and slow feed seems to be most significant at the 5 Day SRT. Individual t-tests conducted for

each SRT showed the fast feed solution biopolymer effluent concentrations to be significantly lower for the fast feed than the slow feed systems, at all SRTs operated.

Visual comparison of solution biopolymer concentrations for fast feed and slow feed systems across SRTs showed increasing concentrations for both systems as SRT decreased. This is observed in Figure 3.9. This trend was similar to that shown by effluent TSS as well as effluent TCOD and SCOD.

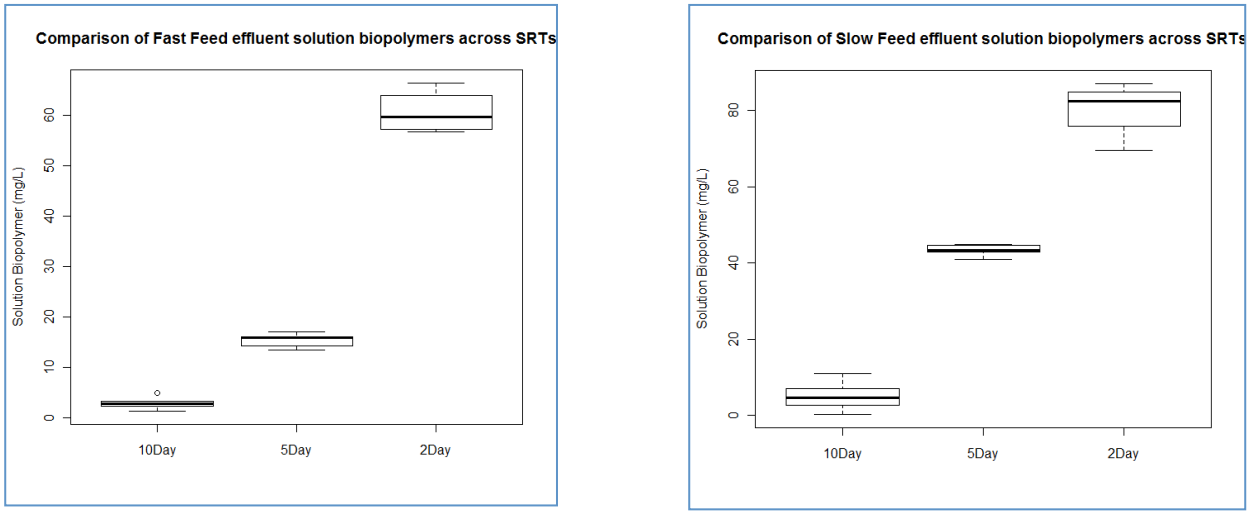


Figure 3.9: Boxplot comparison showing behavior of fast feed and slow feed effluent TCOD across SRTs

A two way ANOVA was conducted to see if there were significant differences between SRTs and between fast and slow feed at specific SRTs, as well as to see if there was some combined effect of SRT and feed rate on the concentration of solution biopolymers in the effluent. The results of the test showed that there were significant differences in datasets across SRTs and for fast and slow feed systems at specific SRTs. The results also provided evidence of a combined effect of SRT and feed rate on concentration of solution biopolymers in the effluent. A follow up Tukey test was conducted on the ANOVA output and revealed significant differences between fast and slow feed biopolymer concentrations at each SRT. This is concurrent with the results of statistical analyses conducted on specific SRT datasets. Based on these analyses, it can be concluded that there is a combined effect of feed rate and SRT on solution biopolymer concentrations that results in significantly lower concentrations for fast feed systems at each given SRT.

3.1.4 CONCLUSION FOR EFFLUENT QUALITY

Based on statistical analysis of the data for each of the effluent quality parameters measured, it can be seen that average fast feed values were lower in all cases, except for the 10 Day effluent VSS. This is in agreement with the work of Martins in 2003, which concluded that PFR systems can properly control sludge settleability versus the slow feed or CSTR systems have poorer settleability and this would result in solids being released into the effluent, as evidenced by the higher effluent TSS for the slow feed systems at each given SRT. High effluent TSS is a sign of weak flocs which results in the formation of less dense particles that are harder to settle and are washed out into the effluent (*Novak et al., 1998*). The effluent VSS exhibits a similar trend to the TSS since it is dependent on this parameter.

Since solids released are actually activated sludge biomass an increase in effluent solids is generally associated with an increase in effluent TCOD; this was observed for each SRT. The fraction of TCOD that was represented by SCOD was greater than 50%, suggesting that the effluent COD was largely contributed by the loss of soluble microbial products, particularly solution biopolymers. This is characteristic of incomplete flocculation or deflocculation associated with lower SRTs, resulting in the excursion of biomass and EPS into the effluent. This provides an explanation for the similarly large increase in solution biopolymers, observed for both fast and slow feed systems as the SRTs decreased. The higher concentration of effluent solution biopolymers coincides with high effluent TSS, which confirms the finding many studies which suggest that effluent biopolymers are a sign of poor flocculation (*Park et al., 2006*). Despite the literature stating that plants can operate at very low SRTs without biomass being washed out, it was also stated that this is usually achieved for systems that have influent biomass (*Grady et al. 1999*). In the bench scale systems operated in this experiment, the feed was a synthetic feed with no influent biomass; therefore it would be expected that rapid washout would occur when operating these systems at low SRTs. This theory was supported by the very high effluent TSS, COD and solution biopolymer concentrations observed for both systems at the 2 Day SRT. Research conducted by Dionisi in 2006 using similar systems found that despite eventual complete washout of both systems, the slow feed system always had poorer quality effluent than the fast feed system. This is supported by the effluent TSS, COD and solution biopolymers all being lower for the fast feed system than the slow feed system at the 2 Day SRT, as well as at the 10 Day and 5 Day SRTs.

The changes in these effluent properties could also be related to the differences in EPS composition of the activated sludge which depends on the microbial community. In their research, Sponza (2004) came to the conclusion that the quantity of EPS plays a very important role in flocculation, but it is not the sole consideration when relating EPS to settleability. The presence of a similar amount of soluble proteins and polysaccharides along with continuous deterioration of effluent quality as the system goes from fast to slow feed agree with studies that

state that settling and effluent quality parameters depend not only on the quantity but the composition of the activated sludge EPS. In some cases the composition of EPS is more important than the quantity of EPS in activated sludge. This change in EPS composition could be attributed to a change in the microbial community, which is expected due to difference in the feeding patterns. Research shows fast feed systems favor the proliferation of floc forming bacteria, while slow feed systems provide ideal conditions for the proliferation of filaments (*Martins et al. 2003; Dionisi et al. 2006*). The increase in solution biopolymers is a result of a decrease in retention of EPS in the floc which increase the effluent COD, suggesting that this increase is not due to a change in kinetics of microbial degradation (*Higgins et al. 2004(b)*)

One observation made was that the effluent COD data and biopolymer data did not correlate well. Though they exhibited similar qualitative trends both in terms of difference in reactor configuration datasets at specific SRTs and patterns across SRTs, the biopolymer concentration was several orders of magnitude lower than the effluent SCOD and TCOD data. This conflicts with the original thought that the biopolymers accounted for the majority of the effluent TCOD; in this case the solution biopolymers would have been very close in magnitude to the SCOD, but this was not observed. Additional attempts to correlate the data by linear regression yielded R^2 values less than 0.3, indicating that there was little to no correlation between the datasets. There are several inferences that can be made here. The first is that the low SRTs resulted in larger degrees of deflocculation and so the particulate matter that was released into the effluent was colloidal and therefore able to pass through the 0.45µm filters used to filter the effluent TCOD samples to obtain SCOD samples. In this case the fraction of biopolymers could not solely account for the effluent SCOD.

A final inference as to why the data did not correlate well was due to problems with sample storage, particularly with the biopolymer samples. The COD samples were preserved by adding concentrated sulfuric acid and refrigerating at 4°C in accordance with the Standard Methods. The biopolymer samples were required to be frozen. There were several instances where the freezer failed and sample integrity would have declined. Unfortunately due to time constraints, the reactors in many cases could not be re-run to obtain samples again. Samples were analyzed primarily to observe the difference in biopolymer concentrations between feed configurations. It is likely that the original concentrations were higher, and may have correlated better to the effluent COD. It can be inferred however that the patterns exhibited by the data are representative of the original effluent samples collected.

Comparing effluent TSS and COD values, it is observed that the magnitudes of COD obtained in the 5 Day and 2 Day SRT reactors are considerably higher than for the 10 Day SRT system, although the effluent TSS does not exhibit such a dramatic increase in magnitude. Based on this it can be inferred that there was some error within the COD datasets; the COD procedure is a very accurate method and any error made is greatly magnified by the multiplication factor of 8000 used in the calculation (*AWWA.*1995).the data is included in this report however to compare the behavior of fast versus slow feed systems at each SRT. Although it is possible that

the COD magnitudes may be erroneous, except for the 10 Day SRT system, the representation of the data trends of fast versus slow feed is considered valid.

It was interesting to note that the largest differences between fast and slow feed system effluent quality was observed at the 5 Day SRT but it is unclear why this occurred. All of the measured effluent quality parameters are directly linked to sludge settleability. In order to confirm the validity if these measurements and postulations for their trends, analysis of the sludge in the operating systems is required. This is addressed in the next section. At this point in the data analysis, the conclusion drawn was that when operating such systems, it is advisable to operate a PFR or fast feed configuration at lower SRTs in order to preserve effluent quality regardless of the SRT used.

3.2 Aerobic Phase Sludge Quality

The evidence provided by the effluent quality data analysis conducted suggests that the slow feed effluent quality is lower than fast feed at all of the SRTs operated. This implies that there was poorer settleability and consequently the sludge of the slow feed systems was of poorer quality. To confirm these findings several analyses were conducted on the biomass/solids in these operating systems. These findings were then compared to the effluent quality data to prove or disprove the current proposed theory for slow feed versus fast feed effluent quality.

3.2.1 Soluble COD Degradation

The soluble COD degradation was used as an indirect measure of substrate concentration or gradient in the operating systems. Figure 3.12 presents one of the soluble COD degradations observed for the fast and slow feed systems.

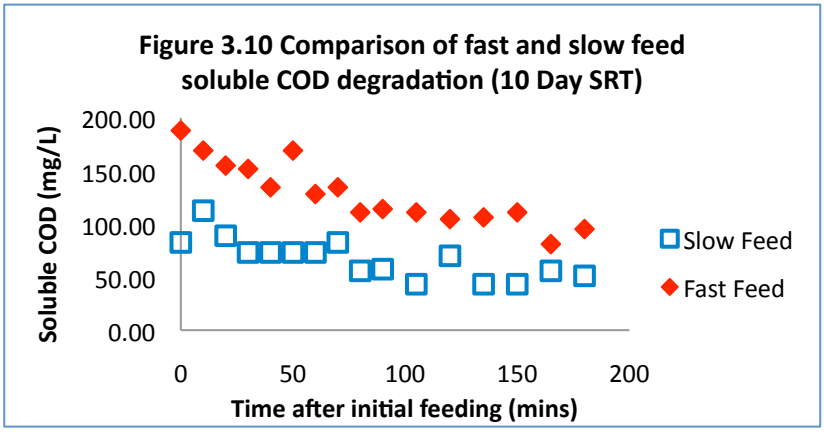
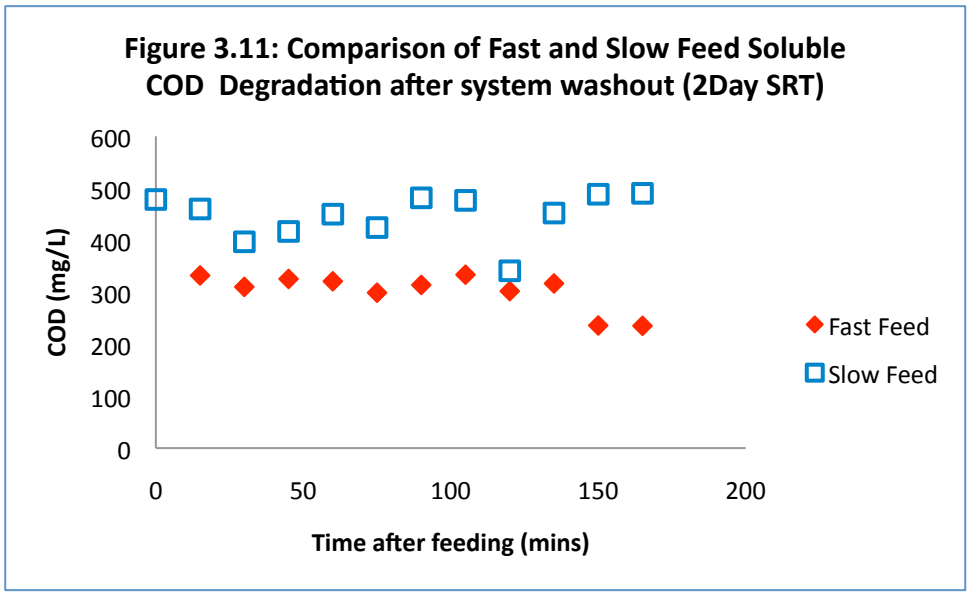


Figure 3.10 shows clearly the difference in soluble COD degradation exhibited by these two feed configurations. The initial soluble COD or substrate concentration in the fast feed system is high

and then declines rapidly until it reaches a steady concentration. This substrate loading pattern is similar to a PFR configuration and implies high substrate loading rate or substrate pressure. The slow feed system had a consistently low substrate concentration, characteristic of a CSTR system. The decline in COD concentration is much slower for this system and the COD is consistently at a low substrate pressure, characteristic of CSTR configurations. Therefore for the purposes of this experiment it can be concluded that the fast feed and slow feed systems behaved like a PFR and CSTR respectively.

It was interesting to note the change in soluble COD degradation as the systems achieved washout. At the 2 Day SRT, rapid washout occurred in these systems, as evidenced by the poor effluent quality. A soluble COD test was conducted late in the operation phase and showed COD uptake to be negligible in both systems. This was perhaps due to rapid system washout and biomass not being able to utilize the substrate fast enough. At this point the effluent quality was so poor that the reactors were discontinued. This data is shown in Figure 3.11 below.



3.2.2 Total and Volatile Solids

Reactor total and volatile solids were monitored to observe if there were any differences between the two systems at each SRT.

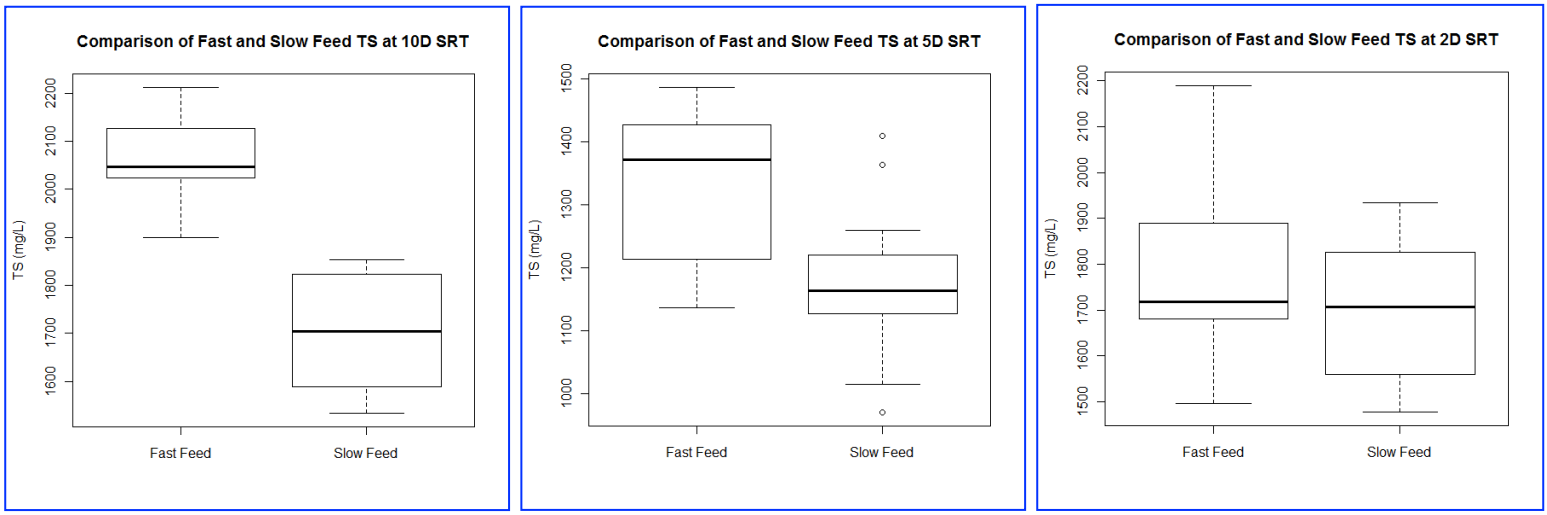


Figure 3.12: Boxplot comparison of fast versus slow feed TS at 10 Day, 5 Day and 2 Day SRTs respectively

Individual t-tests performed for each SRT showed that there were significant differences in total solids concentration between the fast and slow feed reactors at 10 Day and 5 Day SRTs. No significant difference was found at the 2 Day SRT.

A two way ANOVA was conducted to see if there were significant differences between SRTs and between fast and slow feed at specific SRTs, and to determine if there was some combined effect of SRT and feed rate on the bioreactor total solid concentration. The results of the test showed that there were significant differences in datasets across SRTs and for fast and slow feed systems at specific SRTs; there was also evidence of a combined effect of SRT and feed rate on bioreactor total solids. A follow up Tukey test was conducted on the ANOVA output and revealed significant differences between fast and slow feed total solids at the 10 and 5 Day SRTs but none was found at the 2 Day SRT. This is concurrent with the results of statistical analyses conducted on specific SRT datasets. Based on these analyses, it can be concluded that there is a combined effect of feed rate and SRT on total solids concentrations that results in significantly higher total solids concentrations for fast feed systems at each given SRT, with decreasing difference between the two systems at lower SRTs.

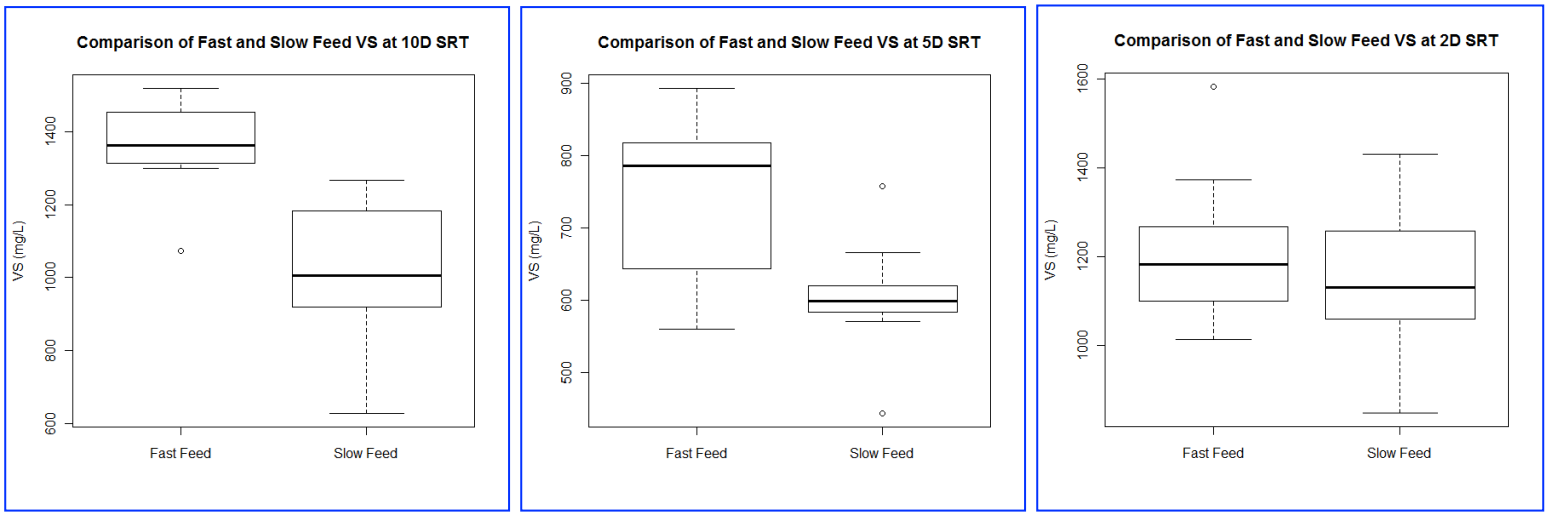


Figure 3.13: Boxplot comparison of fast versus slow feed VS at 10 Day, 5 Day and 2 Day SRTs respectively

Visual comparison of VS showed the fast feed VS to be higher at each SRT, with possible significant differences between fast and slow feed systems at the 10 Day and 5 Day SRT. Individual t-tests conducted for each SRT showed that there fast feed VS was significantly higher for the fast feed at the 10 Day and 5 Day SRT; no significant difference was found at the 2 Day SRT.

To compare behavior across SRTs a two way ANOVA was conducted. The results obtained showed significant differences between feed configurations, between SRTs and provided evidence of some combined interaction between feed configuration and SRT that influenced the VS trends in this experiment. A follow up Tukey test was conducted on the datasets and it was found that there was a significant difference between fast and slow feed VS for 10 Day and 5 Day SRTs; no significant difference was found for the 2 Day SRTs. This supports the findings of the individual t-tests; it was also observed that the VS trends are similar to the TS trends.

3.2.3 Mixed Liquor Suspended and Volatile Solids

Reactor total and volatile suspended solids were monitored to observe if there were any biomass differences between the two systems at each SRT. The mixed liquor suspended solids is perhaps the most important parameter of consideration in the successful operation of any wastewater treatment or activated sludge system. It represents the biomass, as well as other insoluble pollutants, which is responsible for the biodegradation that takes place within suspended growth systems. MLVSS is also a good indicator of system biomass (*Grady et al.1999*).

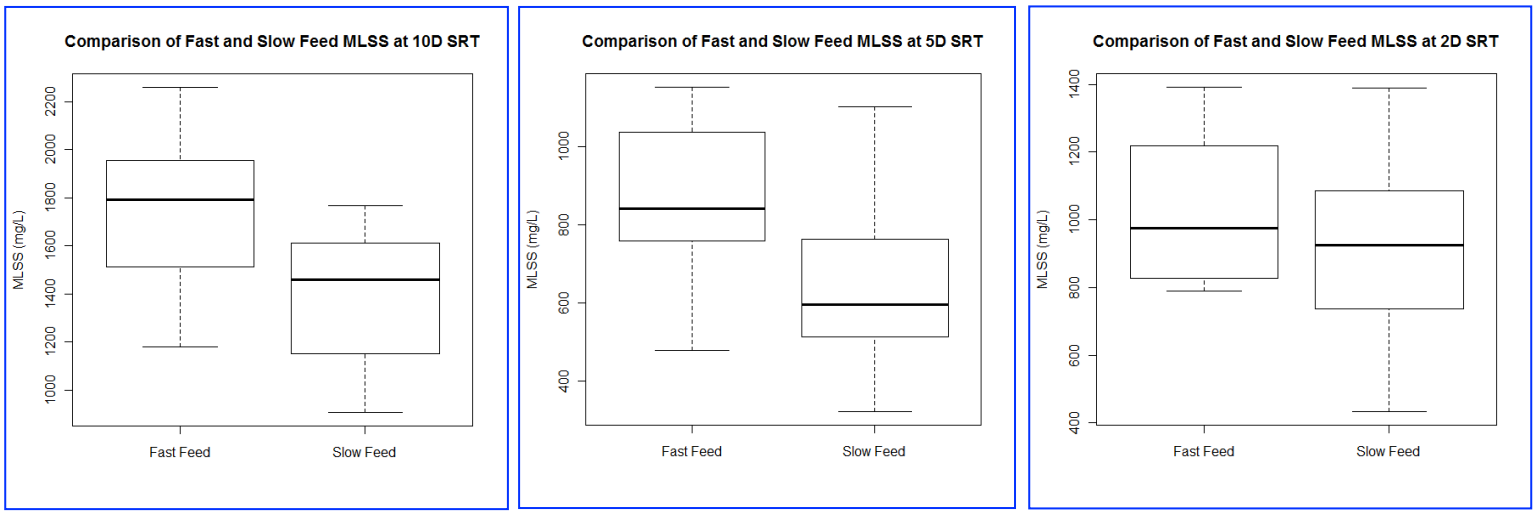


Figure 3.14: Boxplot comparison of fast versus slow feed MLSS at 10 Day, 5 Day and 2 Day SRTs respectively

Visual comparison of fast and slow feed systems at each SRT showed the fast feed system MLSS to be higher at each SRT. There appeared to be significant differences between fast and slow feed MLSS at 10 Day and 5 Day SRTs, but not at the 2 Day SRT. Individual t-tests conducted supported these observations and indicated that the fast feed MLSS was significantly higher than the slow feed MLSS at 10 and 5 Day SRTs; no significant difference was observed between these systems at the 2 Day SRT.

Comparing across SRTs, it appeared that the 10 Day SRT MLSS was the highest and the 5 Day SRT MLSS was lowest for both feed configurations. Observations of the scales in Figure 3.15 support the previous observation that the fast feed MLSS was higher at all SRTs. A two way ANOVA was conducted to determine if there were significant differences between feed configuration datasets, between SRTs datasets and if there was a possible combined interaction between SRT and feed configuration that impacted MLSS concentration. The results obtained indicated difference between feed configurations and between SRTs, but provided little or no evidence of a combined interaction of the two that impacted reactor MLSS.

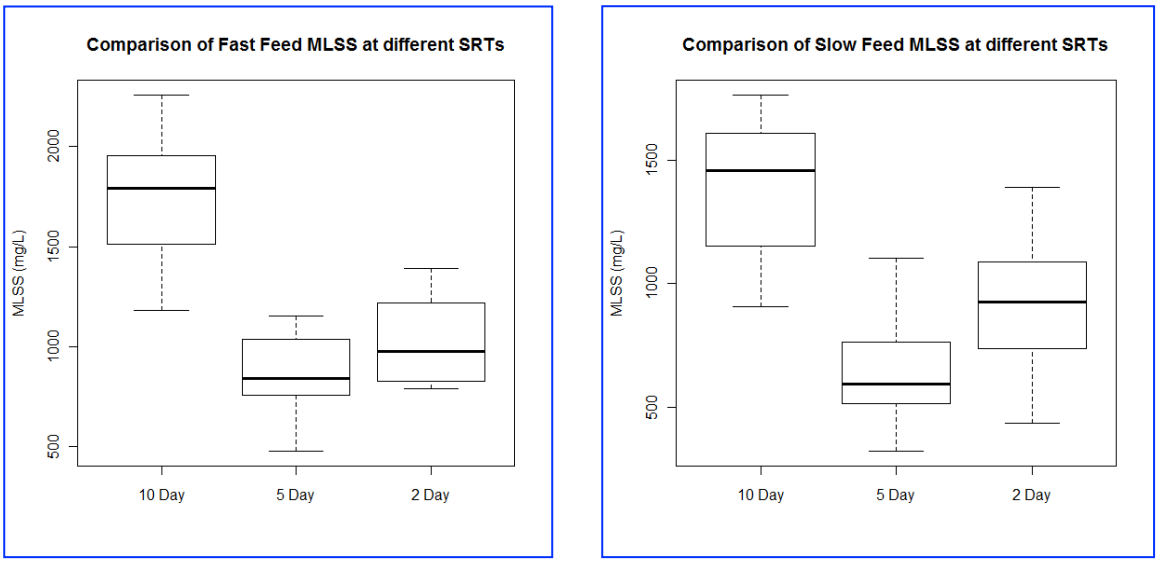


Figure 3.15: Boxplot comparison showing behavior of fast feed and slow feed MLSS across SRTs

Reactor MLVSS was also observed and visual comparisons are presented in Figure 3.16. Observations are similar to those made for reactor MLSS, with fast feed MLVSS being higher than slow feed at all SRTs. Possible significant difference were observed between fast and slow feed systems at 10 Day and 5 Day SRTs; the difference was less observable at the 2 Day SRT. Individual t-tests conducted confirmed these observations, showing that the fast feed was significantly higher than the slow feed at the 10 Day and 5 Day SRTs, with no significant difference being observed at the 2 Day SRTs.

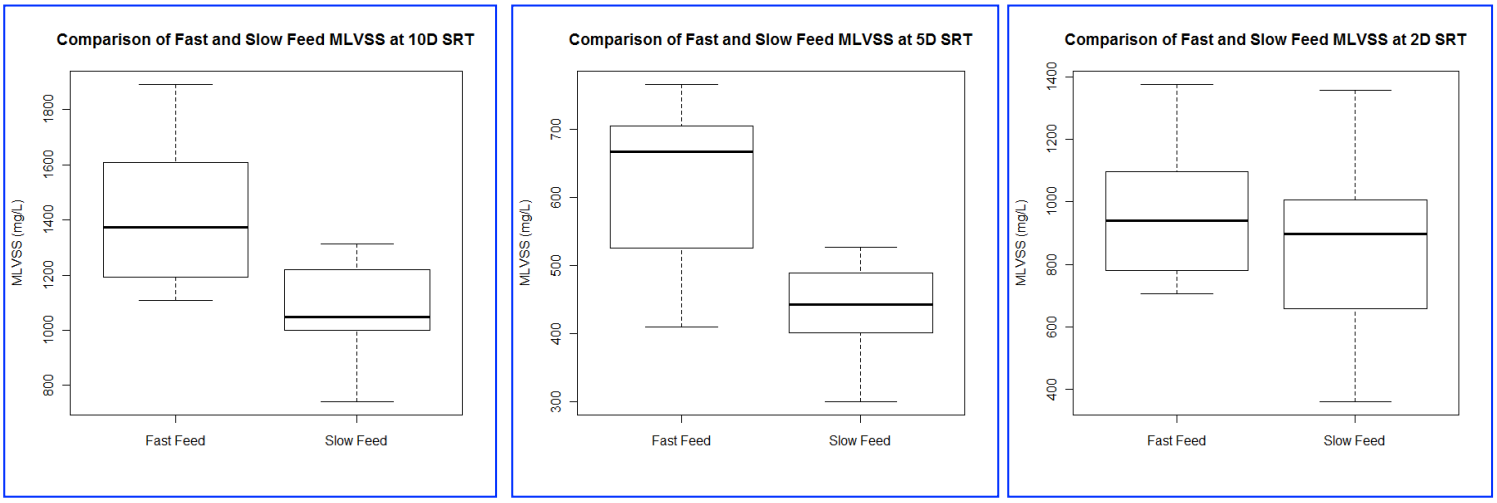


Figure 3.16: Boxplot comparison of fast versus slow feed MLSS at 10 Day, 5 Day and 2 Day SRTs respectively

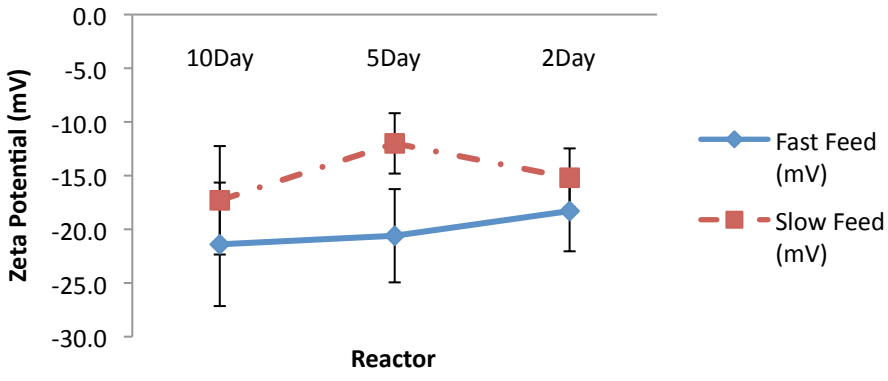
As with the previous datasets analyzed a two way ANOVA was conducted to determine overall significant difference between feed configurations, between SRTs and if there was combined interaction between the feed configuration and SRT that influence the MLVSS trends in this experiment. The results provided evidence to support all three of these postulations. A follow up Tukey Test was conducted and supported the results of the individual t-tests, showing significant differences between the 10 Day and 5 Day SRTs and no significant difference at the 2 Day SRT.

Observation and analyses in this section lead to the conclusion that the MLSS and MLVSS exhibit similar trends between feed configurations across the SRTs operated in this experiment. In each case the fast feed concentration is higher, with the difference seemingly most pronounced at the 5 Day SRT and no significant difference observed at the 2 Day SRT. It was also found that steady state values were highest at 10 Day SRT and lowest at 5 Day SRT.

3.2.4 Zeta Potential

Zeta potential is a measurement of the approximate electric potential of a particle's interfacial double layer. In the absence of charge, meaning surface charge is zero, then there is no electrostatic repulsion, and the particles tend to agglomerate. As the charge on the surface increases, be it positive or negative, then the electrostatic repulsion also increases and potential for agglomeration decreases. For identical particles, if the surface charge increases, repulsion between particles increases. A zeta potential measurement, the magnitude of the value indicates the extent of the surface charge. As the value approaches zero the likelihood that the particles can coagulate or agglomerate increases. The sign just indicates the type of charge, specifically positive versus negative surface charge. Theoretically, zeta potential should correspond to the surface potential of the fixed layer of ions attached to a particle, though in wastewater treatment operations it is used to measure the potential of the surface of the electrostatic cloud surrounding particles (*Metcalf and Eddy. 2003*). The zeta potential, though measured using the effluent samples for this experiment, can therefore be used as an indicator of sludge quality, since it is an indirect measure of particulate agglomeration and hence gives some idea of flocculation. The data obtained for these experiments are presented in Figure 3.17 below.

Figure 3.17: Comparison of Zeta Potential for Fast and Slow Feed across SRTs



As can be seen the surface charge of the particles for all reactors was negative, indicating that the particles all had a net negative surface charge. It is observed that the fast feed zeta potential has a higher magnitude than the slow feed at all SRTs, indicating greater particle agglomeration for the latter. The difference in zeta potentials between fast and slow feed is most significant at the 5 Day SRT. For the 10 Day and 2 Day SRTs the difference is smaller, but still indicates greater agglomeration in the slow feed systems due to their lower magnitudes.

Comparing fast feed data, the largest zeta potential magnitude was found at the 10 Day SRT and the lowest at the 2 Day SRT. This implied that the lowest amount of particle agglomeration occurred at the 10 Day SRT and the highest occurred at the 2 Day SRT. For the slow feed the highest zeta potential magnitude and hence lowest potential flocculation was found at the 10 Day SRT; the highest was found at the 5 Day SRT.

3.2.5 Sludge Volume Index (SVI)

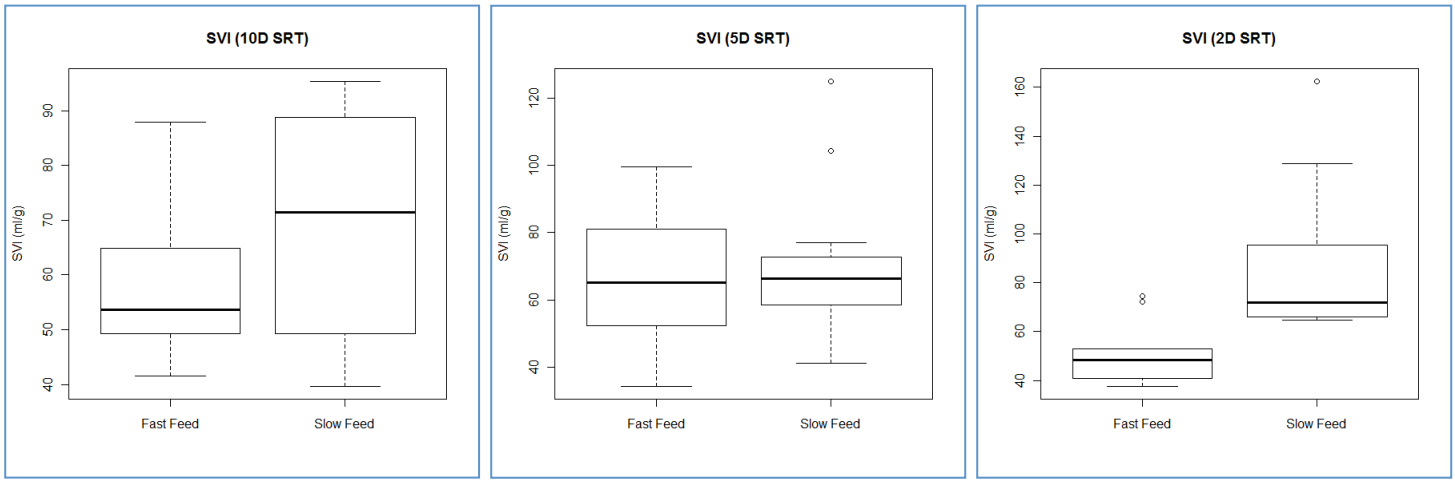


Figure 3.18: Boxplot comparison of fast versus slow feed SVI at 10 Day, 5 Day and 2 Day SRTs respectively

Sludge Volume Index (SVI) is commonly used as an indicator of sludge settleability. Visual comparison of SVI in Figure 3.18 revealed that the mean fast feed SVI appeared lower than the slow feed at all SRTs operated. The difference in means was least observable at the 5 Day SRT and most observable at the 2 Day SRT. Individual t-tests conducted revealed that the fast feed SVI was significantly lower at both the 10 Day and 2 Day SRT, but no significant difference was found between means at the 2 Day SRT.

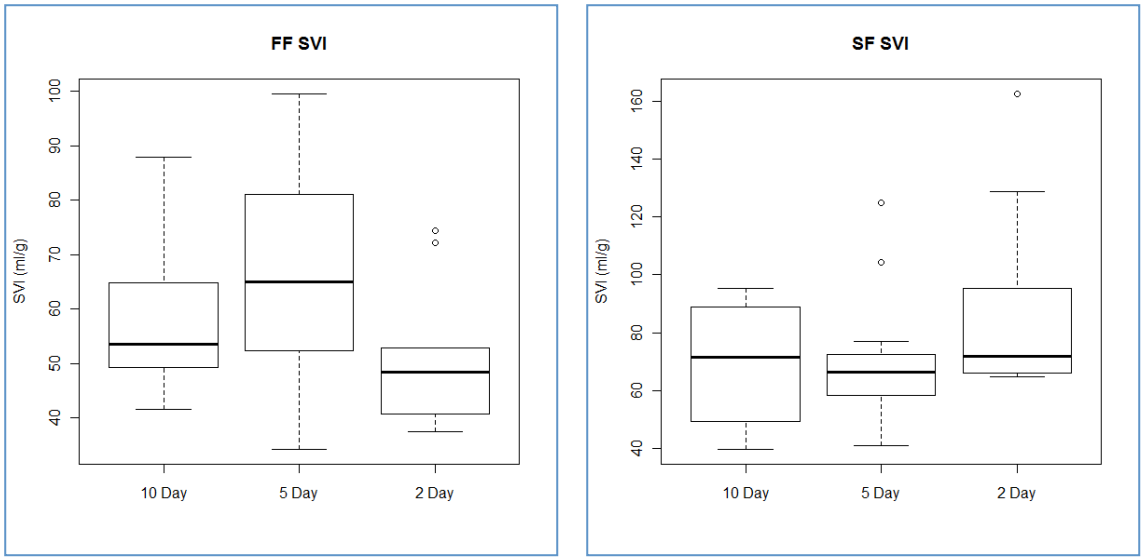


Figure 3.19: Comparison of Fast and Slow Feed SVI across SRTs

Comparison of fast and slow feed SVI behavior across SRTs is presented in Figure 3.19. The fast feed system has the highest mean SVI at the 5 Day SRT and the lowest at the 2 Day SRT. The slow feed mean SVIs are similar across SRTs. Analysis using a two way ANOVA showed evidence of significant differences between reactor configurations, but no significant differences between SRT datasets. There was also no evidence of combined interaction of SRT and reactor configuration on this parameter. A follow up Tukey HSD confirmed the findings of the t-tests, showing significant differences between the fast and slow feed systems at 10 and 2 Day SRTs but no significant differences at the 5 Day SRT,

3.2.6 Capillary Suction Time (CST)

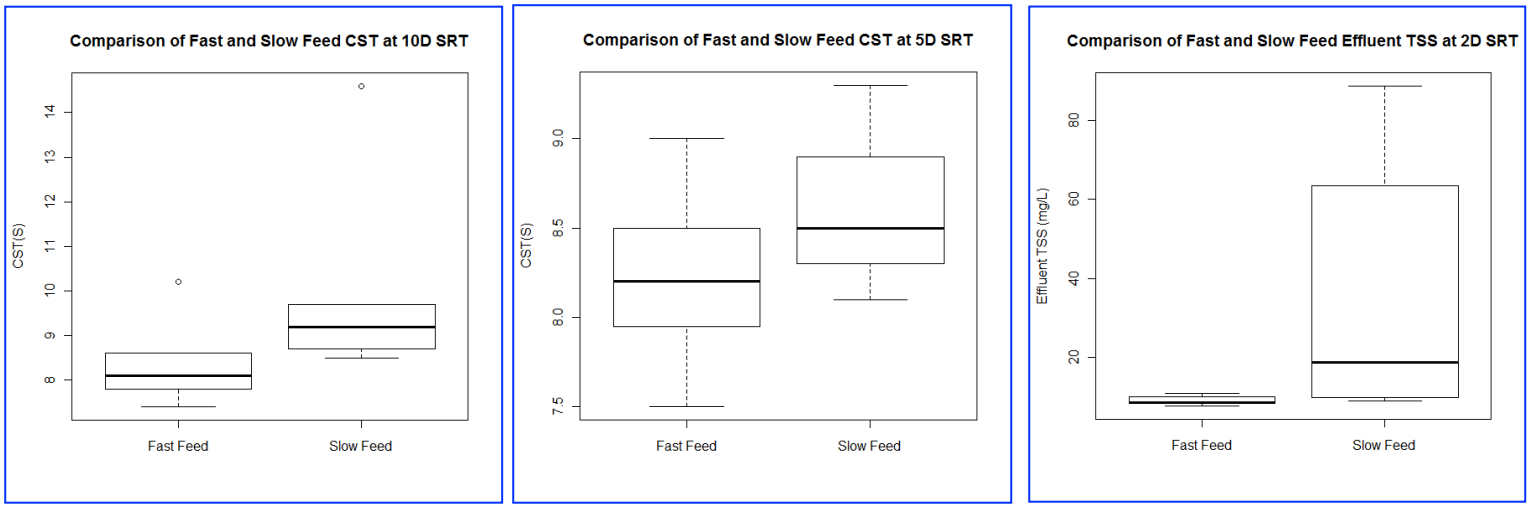
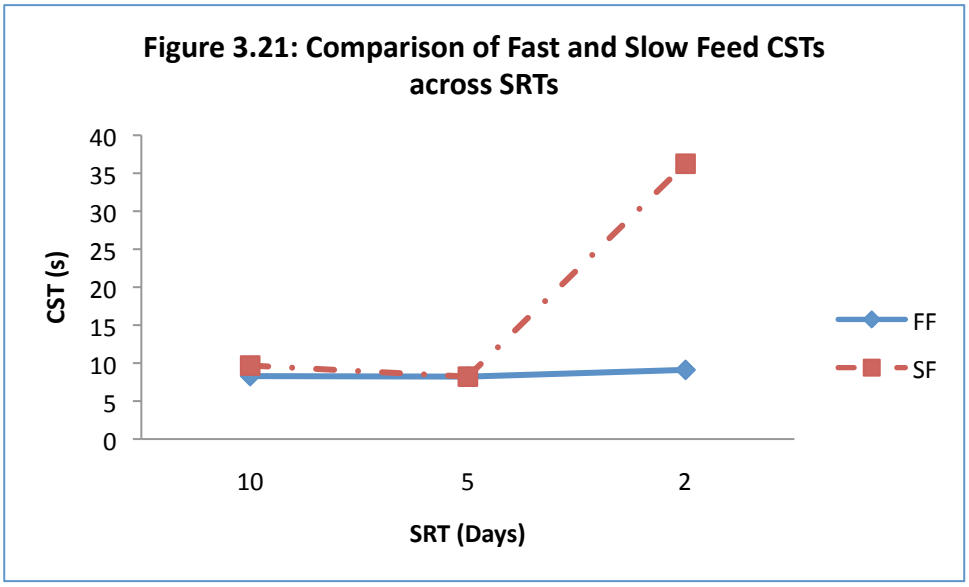


Figure 3.20: Boxplot comparison of fast versus slow feed MLSS at 10 Day, 5 Day and 2 Day SRTs respectively

Capillary Suction Time (CST) is used as an indicator of sludge dewaterability, where lower CSTs indicate better dewaterability and with an average CST of ~10 s being considered good. Visual comparison of Figure 3.20 shows that the fast feed CST is lower in all cases and it appears to be a significant difference in each case. Individual t tests were conducted for the 10 Day and 5 Day SRTs; a Wilcox test was used for the 2 Day SRT due to the skewed, non-normal slow feed dataset. The results of each test showed the fast feed CST to be significantly lower at each SRT. Observation of Figure 3.21 below shows the behavior of the CST data across SRTs; it is seen that the fast feed CST increases slowly yet consistently as SRT increases; the slow feed shows a decline from 10 Day to the 5 Day SRT, but then showed a dramatic increase at the 2 Day SRT.



3.2.7 Conclusion for Aerobic Phase Sludge Quality

Comparing the operation of fast and slow feed at each SRT indicates that changes in reactor feed configurations and changes in SRTs result in significant impacts on sludge quality. Comparing solid concentrations, the differences in reactor total and suspended solids indicated by TS and MLSS measurements showed slow feed concentrations were lower in all cases; this is observed in Figures 3.14 and 3.17 respectively. The differences observed between TS and MLSS value indicated that there was a high fraction of soluble microbial products in the biomass, specifically EPS. The MLSS was found to be highest at the 10 Day SRT and lowest at the 5 Day SRT; the higher biomass concentration at the 2 Day SRT could be attributed to the higher influent substrate concentration or soluble COD, which was implemented to counteract the effect of the low SRT on the system MLSS. The purpose of using the higher COD was to fix biomass in the system and reducing the rate at which washout occurred, since such a low SRT system is very

dynamic and would lead to rapid system washout if it had been operated with the exact parameter values as the 10 Day and 5 Day SRT systems. The reactor MLSS data can be related to the effluent quality parameters as follows.

In terms of effluent TSS, it is seen that this parameter concentration is higher for the slow feed than the fast feed, indicating greater loss in reactor suspended solids into the effluent. This corresponded to the lower MLSS seen in the slow feed systems, and is indicative of poor settleability and higher deflocculation in these systems. These occurrences are characteristic of CSTR configurations (*Grady et al. 1999*). High effluent TSS is related to poor flocculation in these systems, resulting in small particles that are difficult to settle; these particles usually stay in suspension and are washed out of the reactors through the wastage process which leads to a reduction in the biomass of the system (*Novak et al.1998*). This is precisely what was observed with the slow feed systems, with much more rapid increases in effluent TSS across SRTs.

A point to note here is that there is a higher MLSS in the 2 Day SRT system, possible due to the increase in influent COD concentration, which could be used as an explanation for the higher effluent TSS. It can be argued that it would not be good practice to compare datasets across SRTs since the 2 Day SRT system in this experiment had two parameter changes, specifically influent COD and number of cycles per day. Two points are raised to counter this argument. First, the SVI values indicated the sludge samples settled to levels comparable to those at the other SRTs and second the comparative behavior of the fast feed system at this SRT is similar to those observed at the other SRTs. Therefore, it is believed that these comparisons are valid for the purposes of this project.

Comparing the other effluent quality parameters revealed similar trends to the effluent TSS and corresponding inverse relationships with reactor MLSS. Loss of biomass indicates loss of activated sludge components into the effluent. This includes organic matter such as bacteria, matrix EPS and inorganic compounds (*Bruus et al. 1992; Murthy et al. 1998; Sanin.2006; Nguyen et al. 2008*). This leads to an increase in effluent biopolymers and in effluent TCOD and SCOD; these trends were observed for the slow feed systems at each SRT. This was additionally observed for both fast and slow feed across SRTs, accompanied by an increase in effluent TSS and a decline in reactor MLSS, indicating a decrease in system biomass.

The effluent quality and reactor MLSS data were compared to the zeta potential data presented in Figure 3.20. The zeta potential is a measure of surface charge and of electrostatic repulsion. It is therefore an indirect measure of potential particle agglomeration; the higher the magnitude, the greater is the particle agglomeration. It is seen that the fast feed had higher zeta potential values in all cases. This indicates that the particle agglomeration was higher in the slow feed systems than the fast feed systems. This data is contradictory to the data shown for sludge quality and effluent TSS, which indicates that the fast feed systems had less effluent TSS and hence biomass loss, implying better flocculation and greater settleability in these systems. In this case one possible interpretation is that in the fast feed effluent, those particles which were there exhibited

very high electrostatic surface charge and hence greater repulsion, so that they were not incorporated into the floc structure. Conversely it is noted that for the fast feed system, the zeta potential was highest for the 5 Day SRT, which corresponded to the much lower MLSS observed at this SRT. The lowest zeta potential was found at the 5 Day SRT, which corresponded to the lowest biomass; yet the lowest zeta potential indicates greater potential flocculation. One inference is that perhaps there is a proliferation of filamentous organisms in the slow feed systems. Particles would stick together because of the low zeta potential but would not settle properly due to excessive filaments; therefore there would be bulk loss of reactor biomass. This is supported by the effluent TSS found for the 5 Day SRT slow feed.

Behavior of the reactor MLSS can be related to the SVI data presented in figure 3.21. Since SVI is used as a settleability indicator, it could be expected that systems with better settleability have lower SVIs and higher effluent TSS. A good SVI is generally considered less than 80ml/g; greater than 150 ml/g is considered poor (*Grady et al.1999*). In their work Martins et al 2003 concluded that the SVI shows a strong increase with the increase in SBR fill time. Dionisi (2006) found that the biggest difference in operating fast and slow feed systems was in settling properties; slow feed systems were found to have higher SVIs. Additionally in her work in 2004 Sponza found that highly negative charged surfaces corresponded to lower SVI levels and good settling properties. Observation of the zeta potential data in Figure 3.20 shows that the fast feed zeta potential is highly negative; therefore one would expect the fast feed system to have a lower SVI than the slow feed system. This was observed for each of the systems operated, but the difference was not found to be significant at the 5 Day SRT.

CST was used as an indicator of dewaterability, with lower CSTs indicating greater potential for dewatering. Sanin (2006) found that higher MCRT or SRTs improved sludge dewaterability. They stated that the lower SRTs produced deflocculation and smaller particles sizes that clogged filters and increased Specific Resistance to Filtration (SRF). They also stated that the improved dewaterability at lower SRTs coincided with an increase in the total EPS, specifically the protein fraction. Their research also indicated that the dominant EPS fraction was protein, whereas the carbohydrate was found to decrease with increasing SRTs. CST measurements indicated in all cases that the fast feed sludge had a greater potential for dewaterability than did the slow feed sludge. This was inferred from the lower CSTs observed for the fast feed system at all SRTs operated. Low quality sludge would retain water, creating problems for solids handling at wastewater treatment plants due to the need to manipulate much larger volumes of sludge. CST measures are therefore good indicators of sludge's potential for digestion and of its conditioning requirements. From the data observed here it could be inferred that the fast feed sludge would digest better, with greater dewaterability, ease of sludge handling and greater digestibility, specifically reduction of TS and VS.

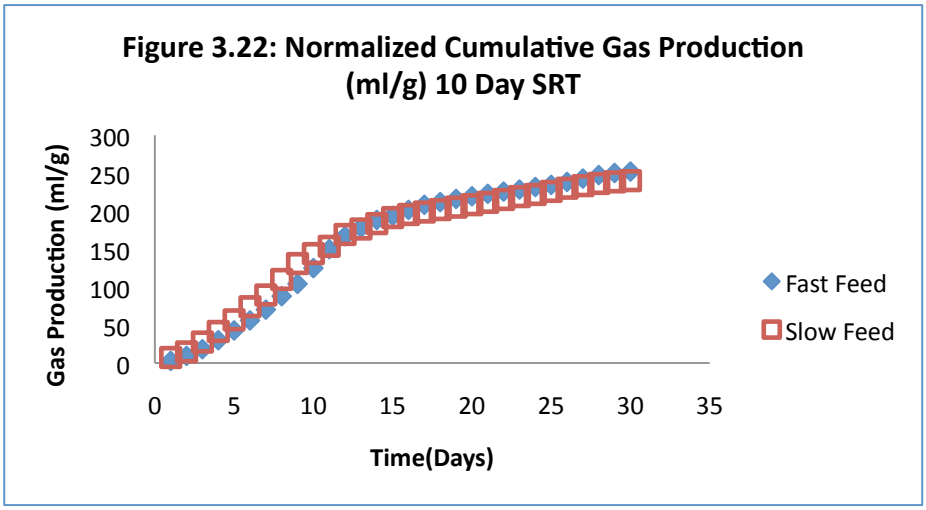
Furthermore, in considering effluent VS and VSS, the data shows these to be higher for the fast feed systems than the slow feed systems. One possible explanation for this is that the reactor TS

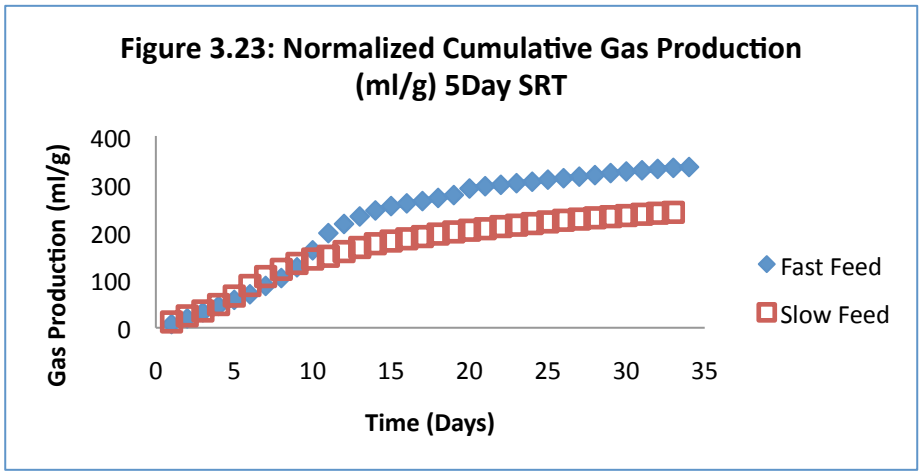
and MLSS are both higher for the fast feed system than the slow feed system. Comparison of the fast feed VS and MLVSS as a fraction of TS and MLSS respectively revealed them to be slightly higher for the fast feed system at all SRTs. Why this occurs is uncertain; one possibility is that it indicates higher odor content in the fast feed sludge. This can only be determined from conducting some form of digestion.

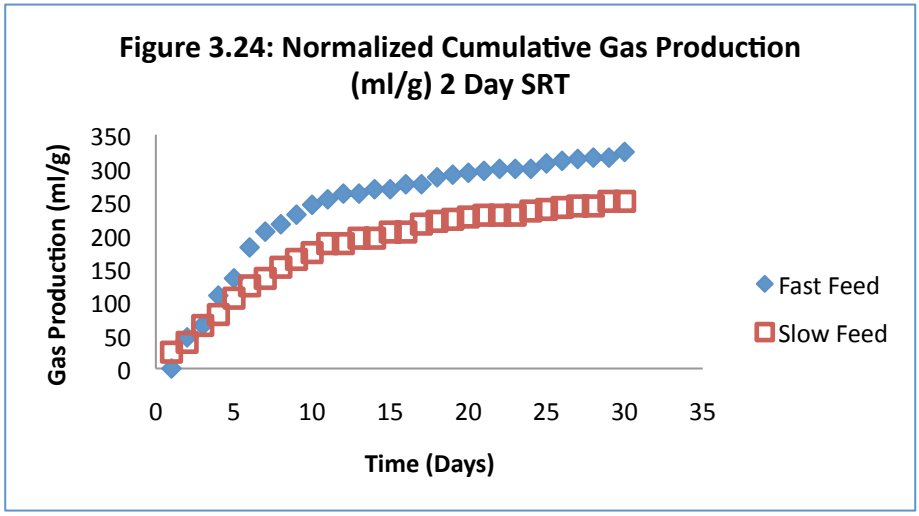
Based on all the data presented for sludge and effluent quality produced during the aerobic operation phase it appears that the fast feed system operates at a higher efficiency in terms of higher quality effluent and sludge, particularly in terms of sludge settleability and dewaterability. The concentration of the measured effluent quality parameters was used as an indicator of flocculation and sludge settleability; SVI and CST were used as indicators of settleability and dewaterability, both found to be better for the fast feed system.

3.3 Sludge Quality: Anaerobic Operation

3.3.1 (a) Batch Digestion and Gas Production







The fast and slow feed reactor sludge qualities were put to one further qualitative comparison: anaerobic digestion. After completion of the aerobic phase of operation, the fast and slow feed reactor solids were concentrated via centrifugation and digested using anaerobic batch digestion. Gas production was monitored over a 30 day period and this data is presented for the sludges obtained from the 10 Day, 5 Day and 2 Day SRT aerobic reactors in Figures 3.22, 3.23 and 3.24 above respectively. This method is similar to a biochemical methanogenic potential test, which is used to measure the biodegradability of wastewater and RAS. The volume of gas produced, presumed to be a combination of methane and carbon dioxide is considered the determinant of the success of mineralization, specifically TS and VS reduction in an anaerobic process.

Comparison of these three graphs reveals that the volumetric gas production was higher for the fast feed sludge obtained from 5 Day and 2 Day SRT aerobic reactors; the sludge obtained from the 10 Day SRT reactors had similar rates of production for both the fast and slow feed systems. The highest volumetric production was found at the 5 Day SRT, specifically for the fast feed system. Therefore from these graphs it can be inferred that the 10 Day fast and slow feed systems had similar TS reduction (TSR) and VS reductions (VSR), while for the 5 and 2 Day SRTs, TSR and VSR were higher for the fast feed system.

Interestingly, the TSR and VSR for the fast feed system was lower than for the slow feed system at all 3 SRTs. Fast feed TSR was approximately 31%, 30% and 32% for the 10 Day, 5 Day and 2 Day SRTs; slow feed TSRs were 37% for all three SRTs. VSR for the fast feed reactors were 38%, 36% and 40% for the 10 Day, 5 Day and 2 Day SRTs; for the slow feed system they were 44.3%, 45% and 49.4% respectively. It is unclear why this occurred. One possible conclusion that could be drawn here is that the fast feed effluent solids produced a greater volume of gas per gram of solid degraded than the slow feed reactor. Based on this postulation, it could be inferred that the biogenic methane potential of the fast feed system is greater than the slow feed.

3.3.1 (b) Short Term Odor Production –Total Volatile Organic Sulfur Compounds (TVOSCs)

For the purposes of this project only short term odors, specifically TVOSCs, are presented since the long term odors were still being analyzed when this paper was written. At the end of the 30 day anaerobic batch digestion period, the sludges were conditioned and dewatered to produce cake solids. These cake solids were stored and measured for short term odor production, specifically organic sulfur compounds, referred to as Total Volatile Organic Sulfur Compounds (TVOSCs). These compounds are generally produced over the first 10 days after storage of the dewatered solids and then decrease. After approximately 20 days, the concentration of these compounds in the headspace for stored cake solids is low. These compounds are of interest since TVOSCs are the most odorous compounds associated with sludge handling in wastewater treatment. These compounds are produced by the reduction of sulfur compounds contained with sludge. The main source of these sulfur compounds is the EPS matrix, specifically the protein component.

Figure 3.25: Comparison of TVOSC for fast and slow feed batch digestion (10 Day SRT)

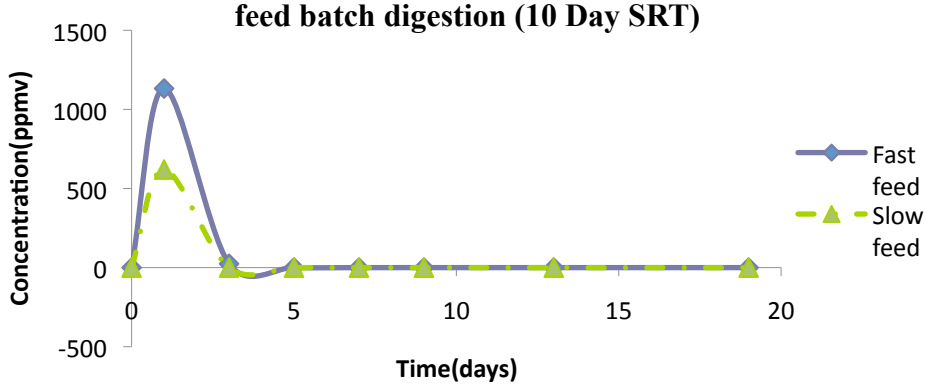
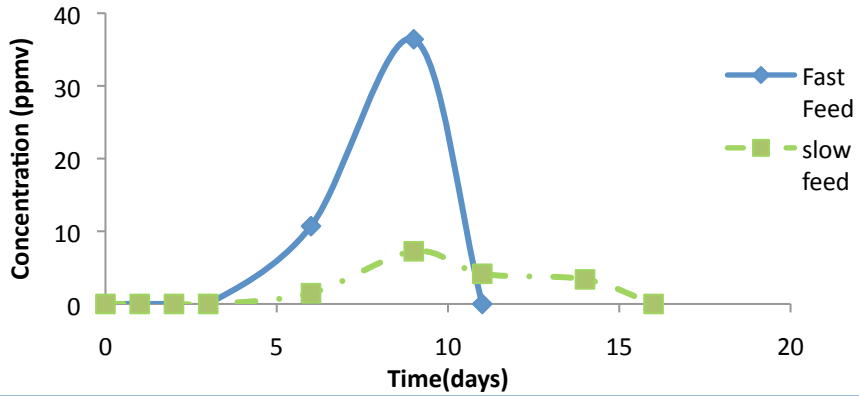
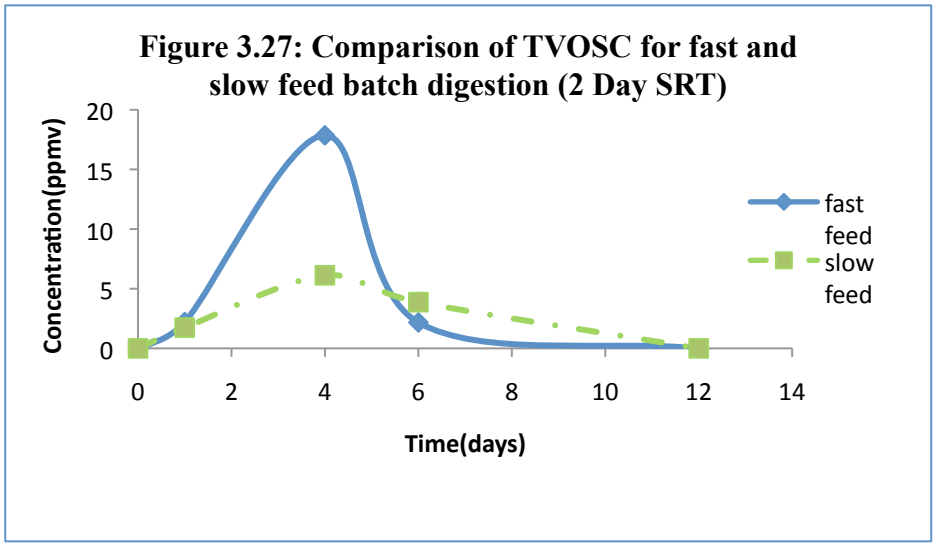


Figure 3.26: Comparison of TVOSC for fast and slow feed batch digestion (5 Day SRT)





Comparison of the data for these systems showed that the sludge from the fast feed aerobic reactors produced higher quantities of TVOSC than sludge from the slow feed systems for all of the aerobic SRTs used. It was also observed that the odors for sludge obtained from the 10 Day and 2 Day SRT aerobic systems peaked between 0-5 days, with the 10 Day SRT sample peaking at approximately 1 day and the 2 Day SRT samples peaking at about 4 days. The 5 Day SRT samples peaked at approximately 9 days.

Observation of the figures also revealed that the TVOSC production was several orders of magnitude higher for the 10 Day SRT than for the 5 Day or 2 Day SRTs. The 2 Day SRT TVOSC production was lowest. The peak and subsequent decline was also much sharper for the 10 Day SRT; for the 5 Day and 2 Day SRT samples this was more gradual, particularly for the slow feed samples.

3.3.2 Anaerobic Phase Sludge Quality Conclusion

Based on the observations for the fast and slow feed anaerobic batch digestion, it is seen that the fast feed has a higher gas production rate per gram of solid as well as a higher total and volatile solid reduction. This indicates higher quality digestion of sludges from fast feed digestion.

The Water Environment Research Foundation (WERF) has sponsored three phases of a long-term project entitled "Identifying and Controlling Odors in the Municipal Wastewater Environment." In Phase 2 of this research they concluded the following regarding sources and causes of biosolids cake odors:

1. Bio-available protein is the primary "food source" of odor-causing bacteria in anaerobically digested biosolids.

2. Some dewatering practices, such as high-solids centrifugation, tend to produce higher odors in biosolids cake than other dewatering practices. This is believed to be a result of exposure to shearing of polymer conditioned bioflocs, used for dewatering, which release proteins into solution and render them bioavailable.

3. Volatile organic sulfur compounds (VOSCs) are the primary constituents of odors from anaerobically digested biosolids.

4. Dewatered biosolids from thermophilic anaerobic digestion can have different odor characteristics and patterns of time release than biosolids cake from mesophilic anaerobic digestion.

5. Iron in sufficient concentrations in biosolids cakes appear to help bind bioavailable protein and reduce its odor production.

Odor produced from dewatered biosolids is a major cause of concern at many wastewater treatment plants and land application sites. Studies have shown that most of the odors are a result of sludge shearing by dewatering equipment. Phase 2 of the study also revealed that solids retention time (SRT) of digesters was associated with odor potential of digested product, whereby increased digester SRT reduced odor potential.

In terms of short term odor production, specifically TVOSCs, it is seen that the fast feed system is much higher for all SRTs operated. One source of organic sulfur in activated sludge systems is EPS, particularly proteins. Effluent biopolymers for these systems were found to consist of more than 50% protein, indicating that the activated sludge floc EPS network was primarily proteins (*Erdal et al. 2008*) In observing system effluent biopolymers, it was noted that the effluent biopolymers at each SRT were higher for the slow feed systems, indicating that these systems were losing more proteins from the biomass. Hence it could be expected that there would be more TVOSCs produced by the sludge from the fast feed systems due to the higher protein content retained within the sludge in these systems. The odor concentrations were very low for the 5 Day and 2 Day SRT systems, much lower than what is considered a normal range for anaerobically digested sludges, as seen in the 10 Day SRT systems. This may be due to the increased loss of biomass from these systems, seen by the declining MLSS and also by the much higher concentrations of effluent biopolymers. The most important trend for the purposes of this project is the behavior of the fast versus slow feed systems, and the trends here are considered accurate representations of these systems.

4.0 ADDITIONAL DISCUSSION

ESEM Images of Flocs for 5 and 2 Day SBRs.

In an effort to garner some understanding of the flocs that existed in these systems, electron micrographs were generated for the 5 and 2 Day SRTs. Unfortunately the 10 Day SRT reactor run had already been terminated when this was thought of. The method and equipment used is explained in the materials and method section. Several dilutions were used. Only a few images are included here for a brief discussion; the remainder is appended.

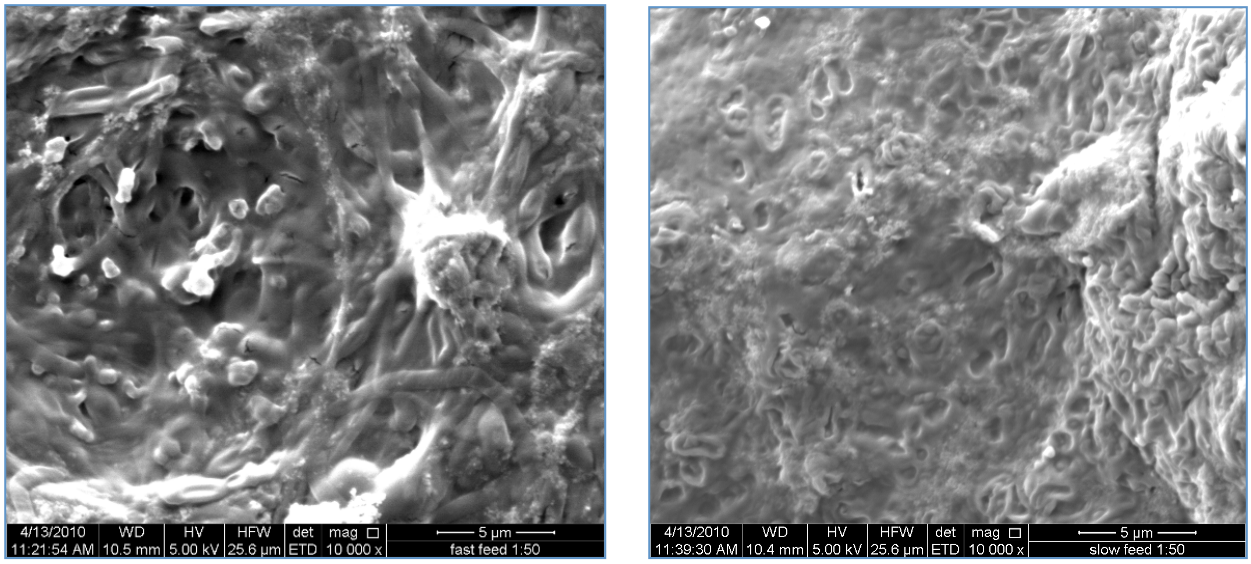


Figure 4.1: ESEM Image comparisons of 5 Day Fast and Slow feed MLSS samples respectively (X10000)

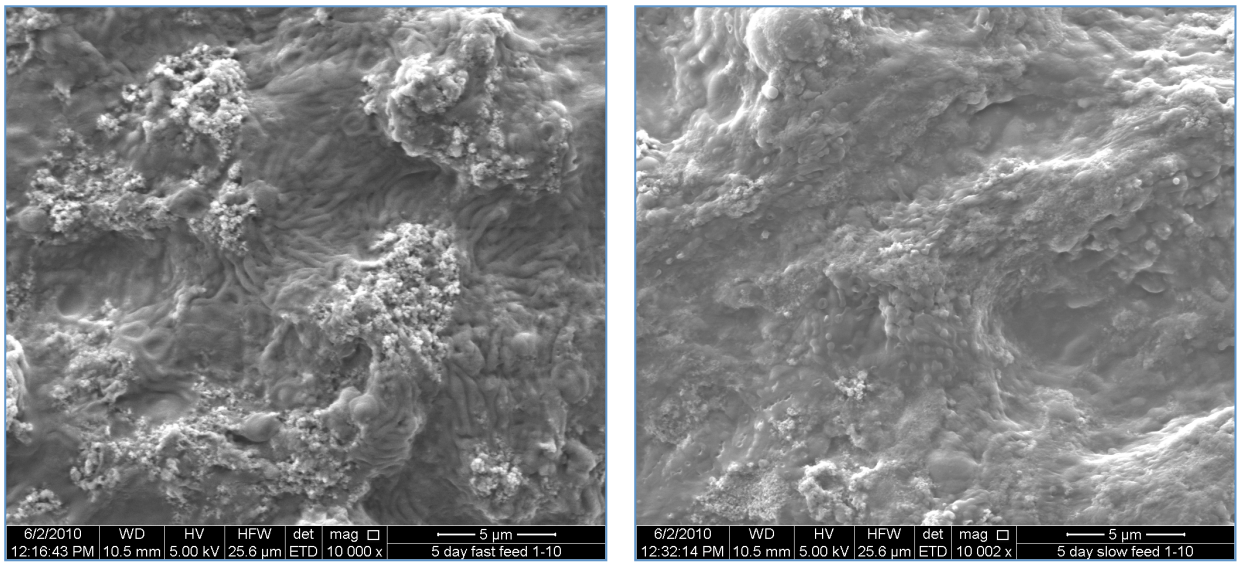


Figure 4.2: ESEM Image comparisons of 2 Day Fast and Slow feed MLSS samples respectively (X10000)

Visual inspection of the 5 Day SRT reactor samples revealed that the fast feed MLSS appeared to be more structured, with clear floc structure that appeared to be a network of filaments with bacterial cells enmeshed within. This is in keeping with the definition of activated sludge structure given in the background. The slow feed MLSS image shows no clear structure, nor any signs of bacterial cells. The material looks globular and appears to be more debris than actual living biomass. The fast feed structure additionally appears to be more porous than the slow feed; this is consistent with the CST results which showed the fast feed MLSS samples to dewater better than the slow feed.

For the 2 Day SRT, the comparison is similar to that made for the 5 Day SRT samples. It is noted this time that, despite the fast feed having a more defined structure than the slow feed, the fast feed appears much less defined than at the 5 Day SRT. Bacterial cells were still present in the fast feed samples; none were observed in the slow feed sample. Both also appear less porous; the slow feed sample in particular appeared very much less porous than any of the other samples observed. This was again consistent with the findings of the 2 Day SRT slow feed CST, which was significantly higher than any other CSTs measured.

Limitations and possible sources of error

In this experiment there were several shortfalls in terms of data quality and collection. On several occasions there were pump failures, as well as problems with timers and programs failing. All of these are inherent in operating an automated system. The learning curve could be observed as the work moved from a more trial and error phase into a structured experiment. Additionally there were problems with sample storage, particularly with storage of biopolymer samples, which were required to be frozen. Freezer failure resulted in possible decline in integrity of the samples. Despite this, data trends were generally quite consistent. It could therefore be concluded that these problems may have decreased the length of the steady state period, but sufficient points were still collected to make valid comparisons.

Perhaps the biggest shortfall of this experiment was the lack of time to repeat the experiments to ensure data reproducibility. The 10 and 5 Day SRT systems were operated twice; however the 10 Day SRT repeat was operated for too short a period to draw any valid conclusions from it. The 5 Day SRT repeat had some problems during the aerobic operating phase, as a result of moving the experimental set up during its operating phase. Some of the data obtained for the latter was found reproducible.

One factor to be considered is the concept of desired versus actual SRT. For these systems, the desired SRT was achieved by manipulating the solids wastage rate from the reactor aerobic phase. Solids were also removed from the system through the effluent wasted after the settle phase of SBR operation. Therefore, the actual system SRT may have been lower than the desired SRT. This is related to the quantity of solids lost in the effluent; the lower the quantity, the closer

the actual SRT is to the desired SRT. Therefore for this project, it is expected that the actual SRT of 10 Day SRT systems was approximately 10 Days; for the 2 Day SRT system this may have deviated more from the desired SRT, due to the higher effluent TSS. The data was still considered to be representative of the desired SRTs since the effluent solids were low compared to the quantity of solids wasted by the set F_w.

Another problem was found in the operation of the first 10 Day SRT reactor set, where the system suffered an infestation of red worms. This is explained in some more detail below.

The Impact of Red Worms

Worms are the largest organisms observed during the microscopic investigation of activated sludge systems. Red worms, so called for their color, are a type of aquatic segmented worm, or oligochaete that move peristaltically and are a commonly occurring nuisance in activated sludge systems where there are zones of improper mixing. The worms have been found to feed on the sludge and can lead to substantial sludge reduction (*Wei et al.2009*). They have also been found to occur in plants with a higher sludge age; in this experiment the worms were found to occur in the fast feed 10 Day SRT system, but not in the slow feed system. Numerous methods have been suggested to get rid of these worms, including decreasing sludge age by increasing sludge wastage, more vigorous and efficient aeration. Suggestions have been made about the usefulness of these worms in reducing the quantity of sludge for solid handling, but this has to be further investigated. Currently these worms are considered a nuisance due to their occurrence in the bioreactors and inhibition of proper bioflocculation. For the purposes of this experiment the aeration in the fast feed reactor was shut off for approximately 2-3 days and the reactor was sealed to generate anaerobic conditions. Once this was achieved aeration was switched back on and the red worms did not recur. There was a decline in reactor MLSS, but not a significant decline; the reactors recovered rapidly and achieved steady state along with the unaffected slow feed reactor.

5.0 CONCLUSION

Observation of all the data provided indicates that SRT and feed rate impact sludge quality and consequently effluent quality. The effluent and sludge quality parameters monitored revealed that the fast feed configuration operated more efficiently at each SRT. Fast feed effluent TSS, VSS, total and soluble COD and effluent soluble biopolymers were all lower at each SRT for the fast feed system. This indicated a better quality effluent produced from the fast feed configuration at each SRT. The SCOD fraction accounted for more than 50% of the TCOD, indicating that the effluent COD was primarily contributed by soluble microbial products or effluent biopolymers, lost from the reactor biomass due to poor flocculation and settleability, particularly in the case of the slow feed system. The lack of exact correlation between the effluent biopolymers and SCOD could have been due to experimental error, or degradation of

sample integrity through problems with sample preservation. The COD was given a lower rank as an effluent quality indicator, due to possible error in the data values, observed when trying to correlate it to other effluent parameters. The 10 Day effluent COD was considered accurate since it was comparable to both the effluent TSS and soluble COD degradation for those systems.

Observation of soluble COD degradation trends in the bioreactors for each SRT showed the difference in substrate gradients for the fast and slow feed systems, with the former exhibiting a high substrate loading rate or PFR gradient model, and the latter exhibiting a low substrate pressure or CSTR gradient model. Sludge quality monitored during the aerobic phase showed the fast feed sludge retained higher reactor MLSS, which was supported by the lower effluent TSS. The slow feed system showed a rapid decline in the MLSS, which was expected when observing the higher effluent TSS. Effluent COD and biopolymer trends supported the patterns in reactor MLSS shown for both the fast and slow feed configurations. Zeta potential readings showed an opposite trend to what was observed with the sludge quality; no definite reason has been concluded for this, though several postulations have been made. SVI was not considered a good indicator of settleability; effluent TSS was used as an indicator to back up questionable SVI data; the anomalies noted in SVI data were attributed to incomplete flocculation leading to incomplete settling of biomass and false representation of SVI. CST data gave a better, more consistent measure, showing that the fast feed dewaterability was better than slow feed, particularly at the 2 Day SRT.

A proposed explanation for the decline in MLSS was the occurrence of endogenous decay. Endogenous decay involves the degradation of energy sources within the biomass to meet energy requirements and biomass maintenance. The rate of endogenous decay increases with sludge age or more specifically with increasing SRTs, since the biomass is retained within the system and has more time to use up its exogenous energy sources and begin endogenous decay to supplement this. In this project it is unlikely that endogenous metabolism significantly accounted for any biomass loss, since the SRTs operated were low compared to standard SRTs used in many large scale plants, and the reactors were fed regularly with an exogenous substrate. The rate of biomass removal could therefore be considered to be primarily due to loss in solids wastage and effluent TSS.

It was observed overall for the aerobic phase that the performance of the fast feed system was superior to the slow feed system at all SRTs operated. It should be noted though that effluent and sludge quality declined for both the fast and slow feed systems as SRTs declined. This was expected with the lower SRTs providing unfavorable conditions for proliferation of floc forming bacteria and hence decreases the efficiency of bioflocculation.

In terms of anaerobic digestibility the sludge, it was shown that the fast feed system seemed to digest better than the slow feed sludge in terms of gas production at the 5 Day and 2 Day SRTs. 10 Day SRT gas production was similar. The TSR and VSR were lower, however for the fast feed system; the reason for this was unclear. The fast feed TVOSCs however were much higher

than the slow feed TVOSCs in all cases, indicating that the former system would produce sludge with much higher short term odors than the latter. The highest odor content was produced by the fast feed system at the 10 Day SRT. This was attributed to higher protein content in the fast feed sludge than the slow feed sludge. This odor can be taken care of by addition of iron; research has been successfully conducted in the use of Ferrate (VI) for this purpose (*Erdal et al.2008; Chun et al.2009*).

A point to bear in mind here is that this experiment was operated at what is considered the ideal M:D ratio, defined as 1:1 (*Higgins and Novak.1997*). Therefore the lack of statistical significant differences noted for several datasets may have been influenced by this parameter. It is expected that these results will change if the M:D ratio is manipulated. Postulations are in favor of the fast feed system still being the superior configuration in terms of effluent and sludge quality; however this requires bench scale testing before any definite conclusions can be drawn.

6.0 Engineering Significance and Future Recommendations

The implications of this study are widespread, since SRT is the key design parameters in all activated sludge systems. Manipulation of SRT has very dramatic impacts on the operation of activated sludge systems, including the required basin size and number or basins, the magnitude of aeration and it depends on the desired outcome of the plant. For example, construction is currently underway in Mexico City for a very large municipal wastewater treatment plant that is set to operate at a 1 day SRT. The reason for such a low SRT is that there is a desire to have no nitrification and retain ammonia in the effluent; this is because farmers downstream of the plant abstract water and use it both to irrigate and fertilize their crops. In Mexico City, the weather is warm year round and provides conditions for the promotion of nitrification. Therefore the plant's objective is to maintain a low SRT to prevent nitrification. The additional benefit of such a system is the cost saved in reduced aeration requirement, since nitrification is a very oxygen intensive process. Considering the results of this experiment it is advisable that this system uses a PFR or fast feed configuration for its operation.

Another engineering application of this project is in the use of PFR versus CSTR configurations to treat industrial wastewaters which have a large xenobiotic fraction. Based on SRT requirements for such compounds, the current default treatment involves very long SRTs in large basins with a CSTR configuration. The previous school of thought was the longer contact time would allow greater acclimation of microbes and yield better degradation and hence effluent quality. Despite the logic behind this, it was seldom found to be the case. Based on the results of this study, it can be inferred that a PFR configuration would achieve better effluent quality at the required higher SRT and reduce costs, due to decreased required basin size.

There are numerous potential applications for the results of this experiment and there is much more that can be done with the systems at the bench scale before launching into experimenting with full scale systems. The SRT- feed configuration interaction can be looked at across a wider

scale, and even incorporate variation in feed types. For example, the three SRTs operated in this paper could be operated and a third parameter could be incorporated such as variations in the M:D ratio, or more specifically the concentration of Na^+ ions, which is an ion of critical interest in industrial systems. This manipulation is similar to the work Francisco Cubas did in his research in 2006 (*Cubas. 2006*). Another way in which the feed could be manipulated is by varying feed complexity; complex substrates generally require longer retention times for decay. Additionally there is room for investigation of microbial community shifts in these systems. Other manipulations of the system can be done to determine the behavior of PFRs and CSTRs under varying conditions. Since these two are the basic designs underlying activated sludge systems, the importance of continued research in this area cannot be de-emphasized.

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APPENDIX A.

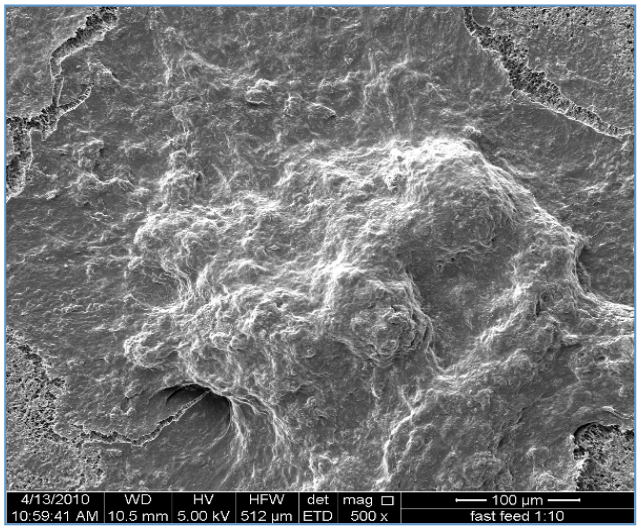
5 DAY SEM IMAGE COMPARISONS

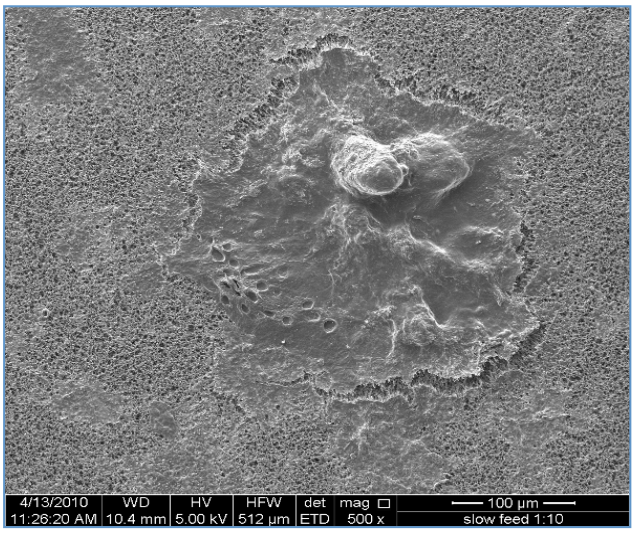
1:10 Dilution

FAST FEED

SLOW FEED

500x





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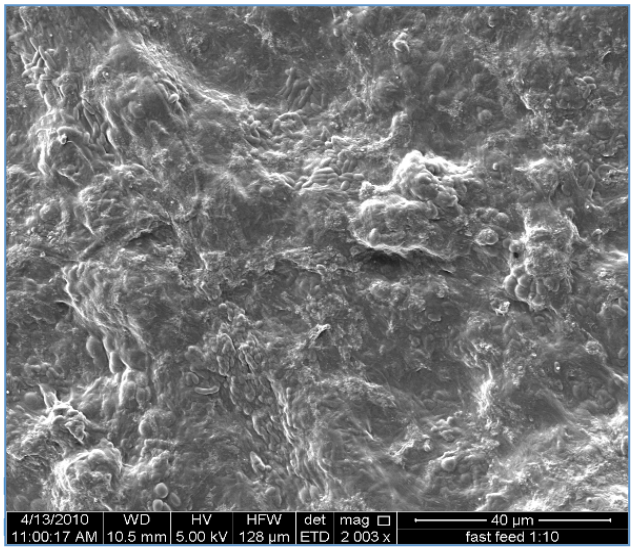
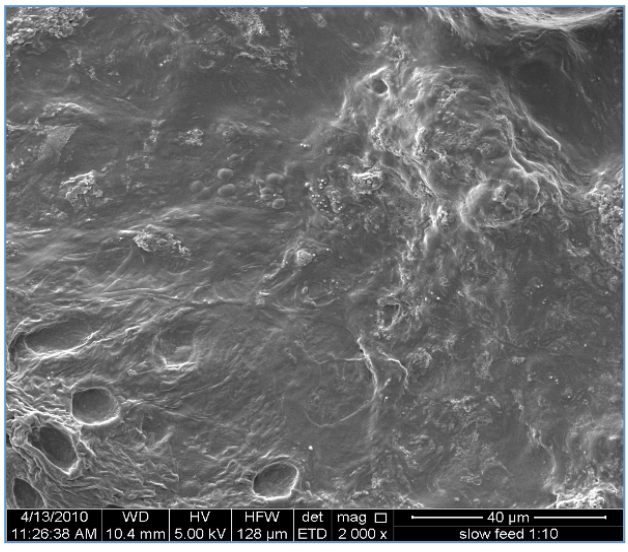
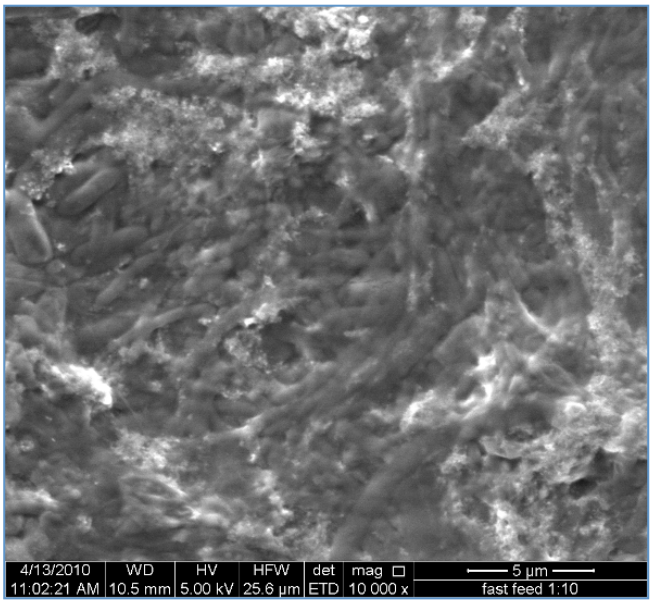
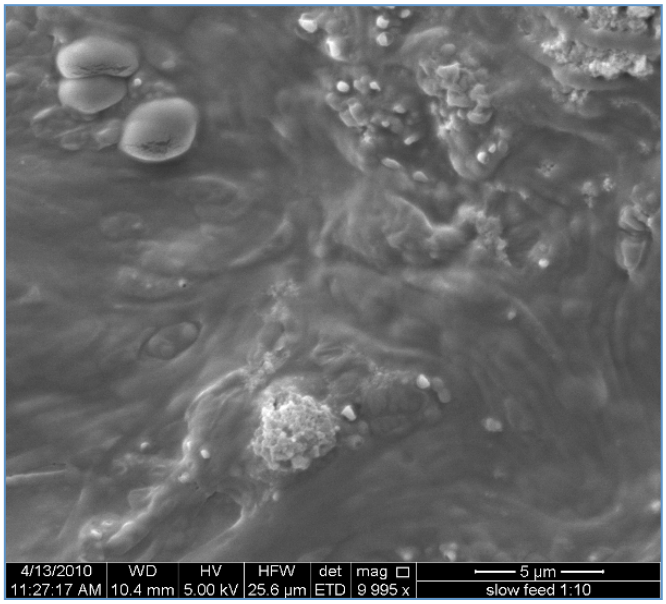




10000x



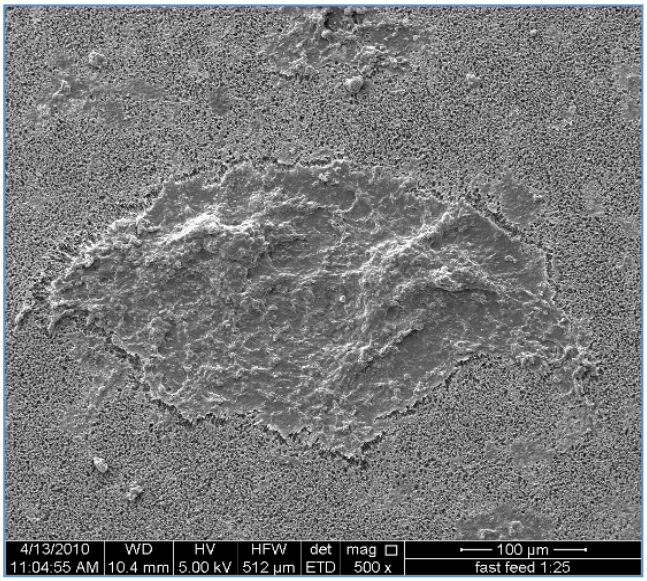


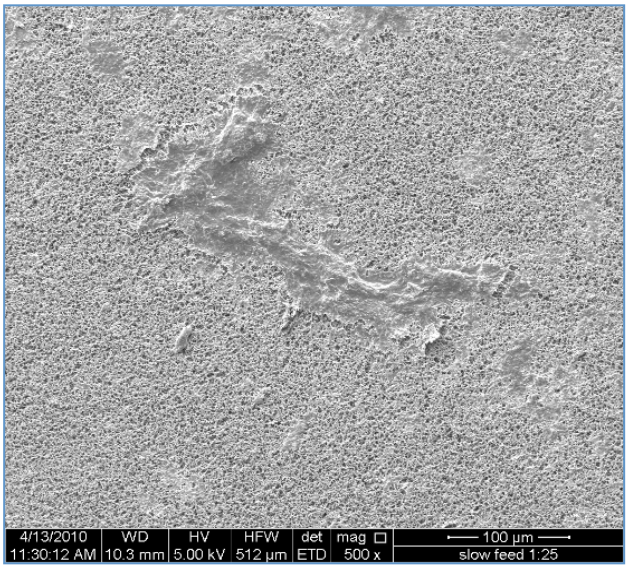
1:25 Dilution

FAST FEED

SLOW FEED

500x





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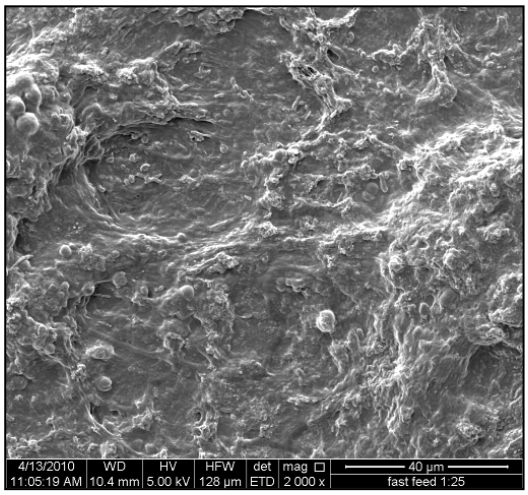

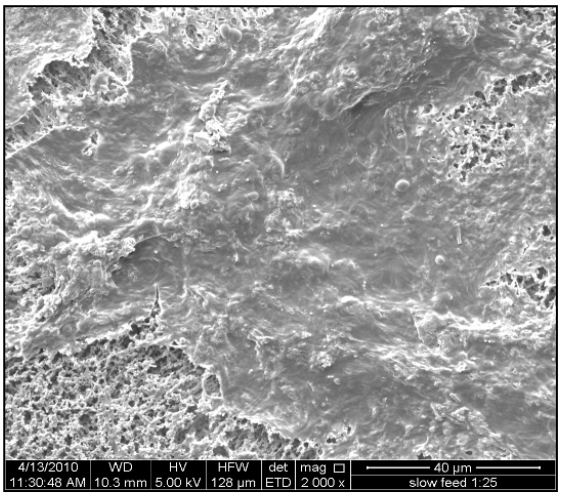

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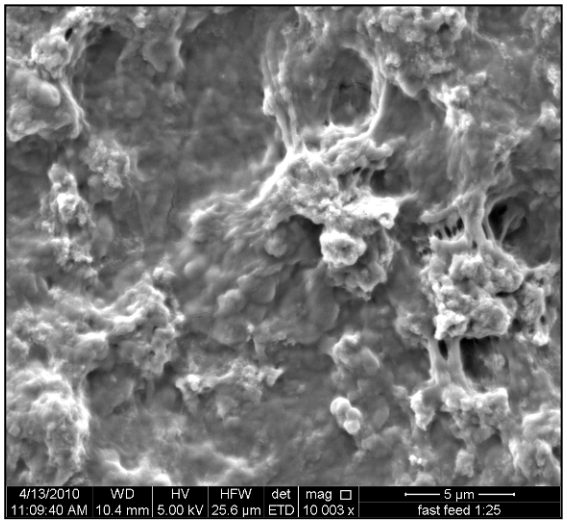
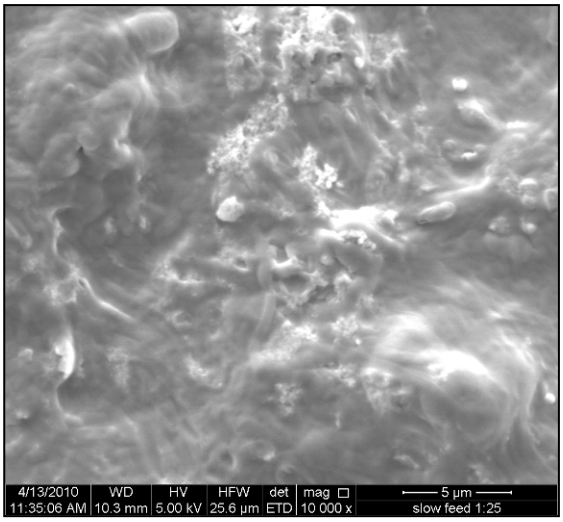
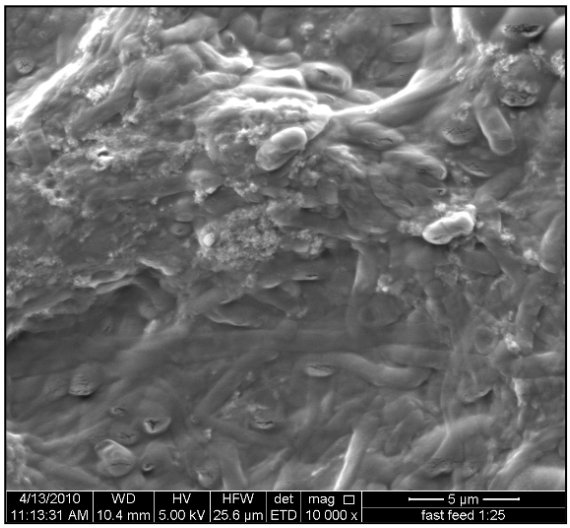




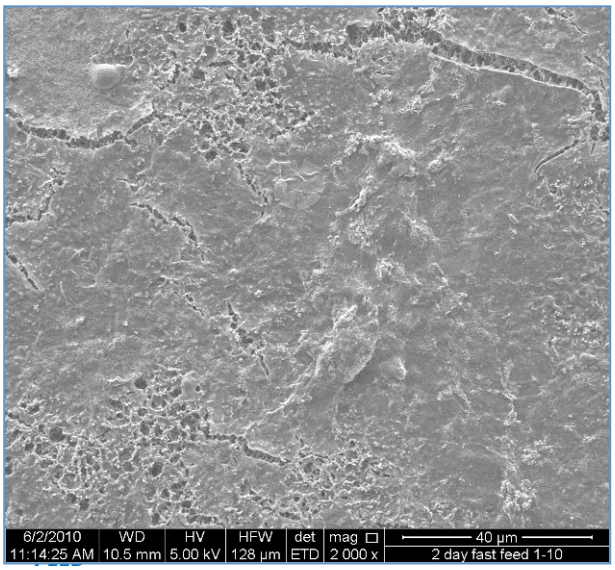


2DAY SEM COMPARISON

1:10 Dilution

FAST FEED

2000x



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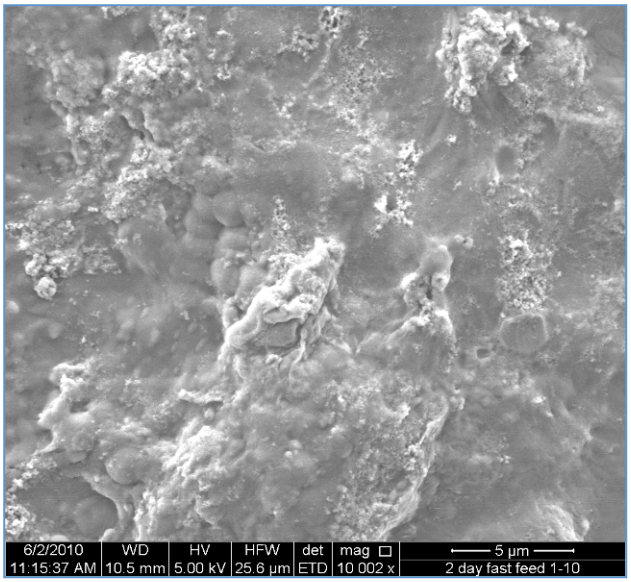
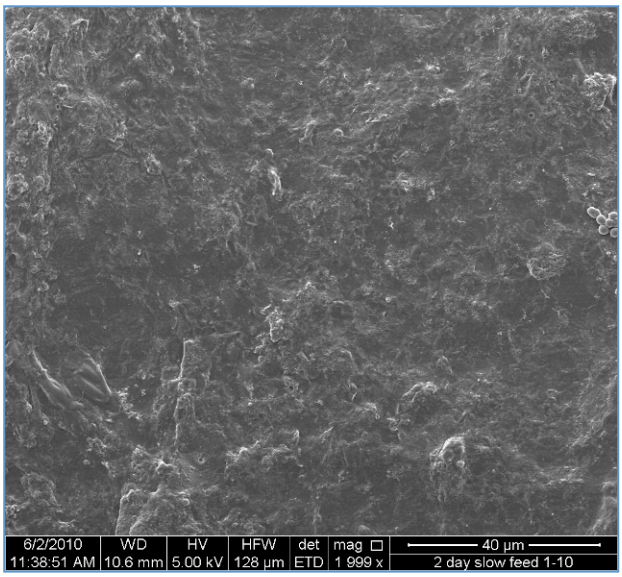


SLOW FEED

2000x



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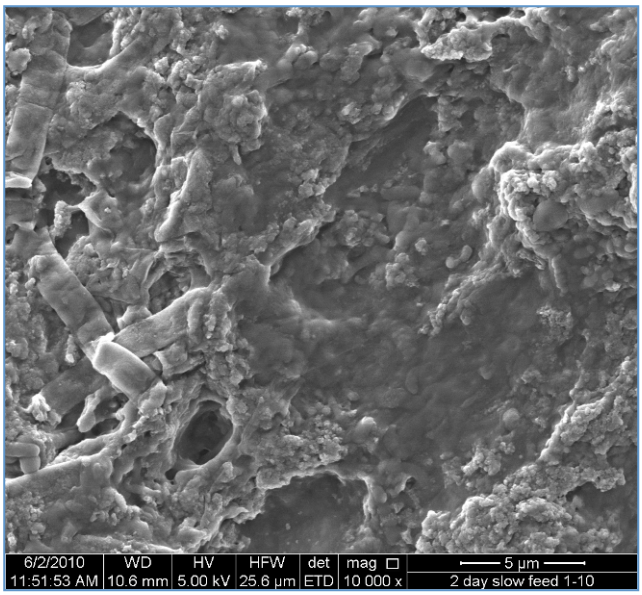
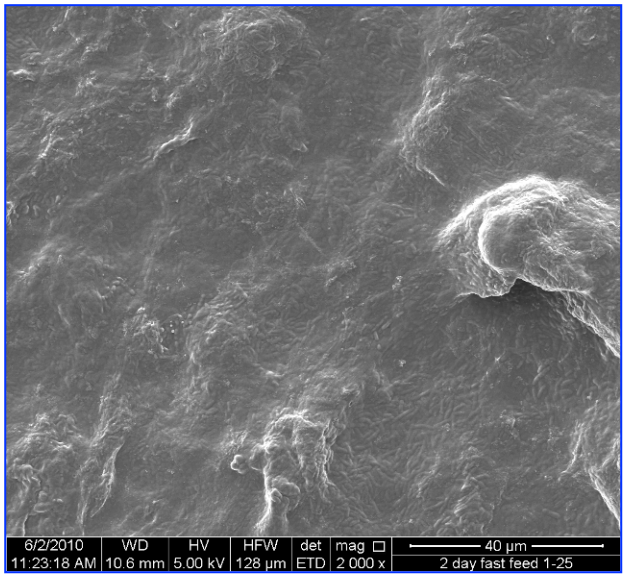
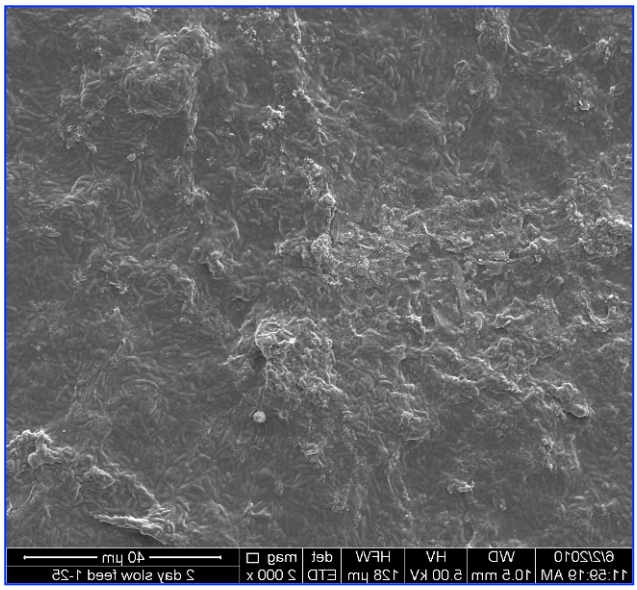


FAST FEED

SLOW FEED

2000x





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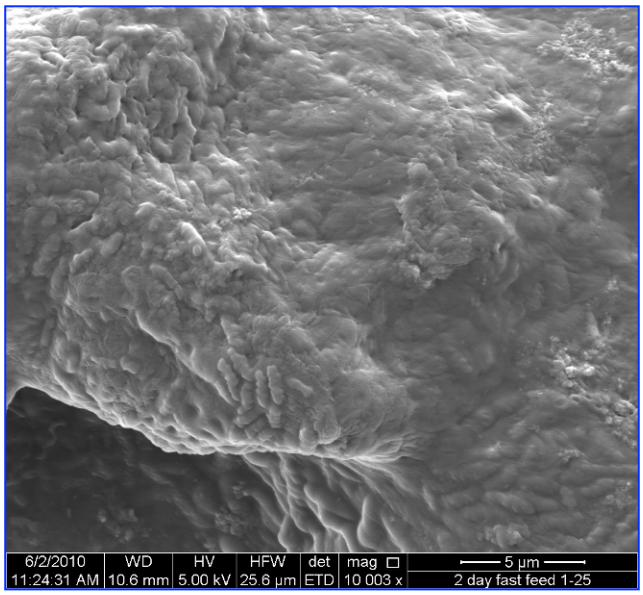
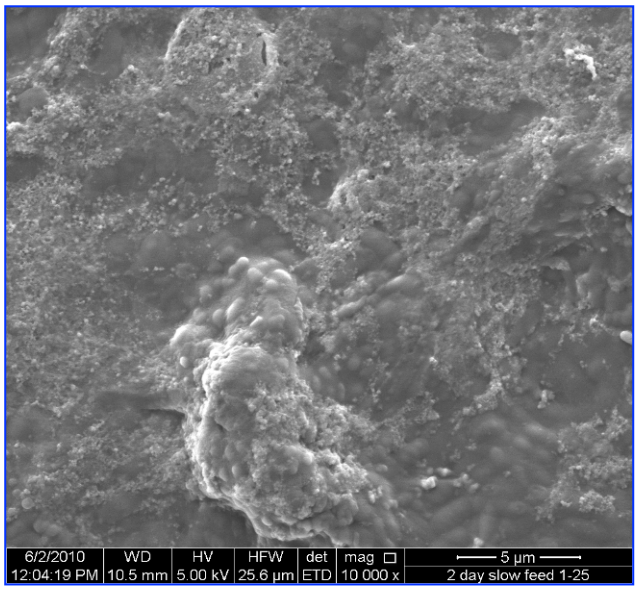
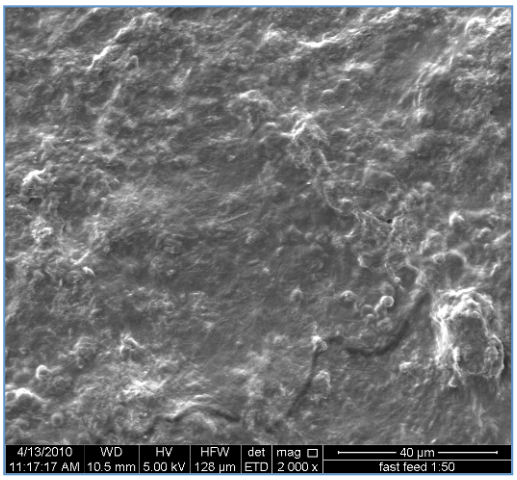


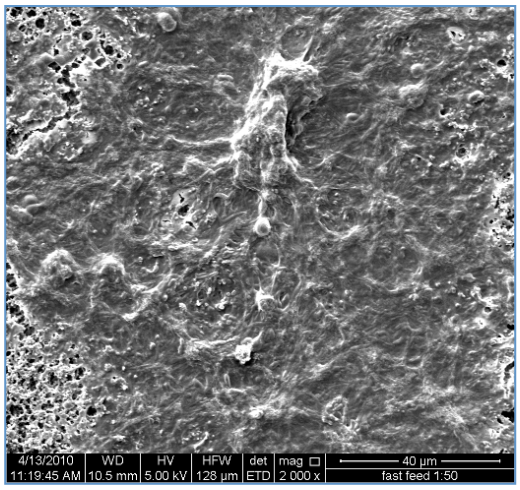


1:50 Dilution

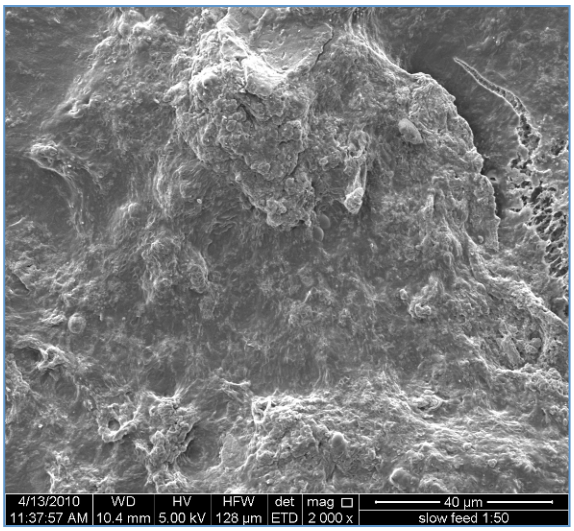
Fast Feed

2000x





Slow Feed



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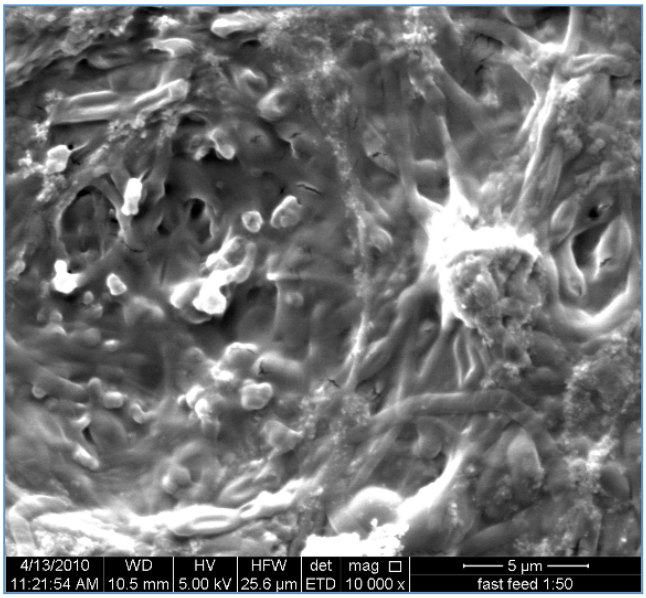
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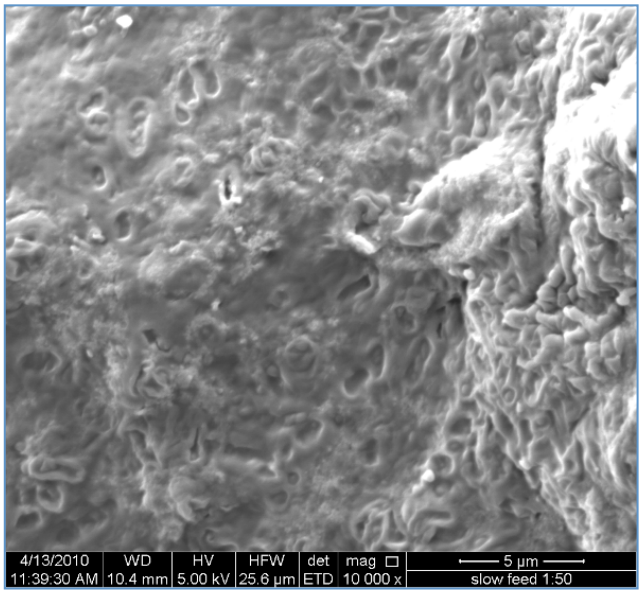
5 µm
fast feed 1:50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5 µm
slow feed 1:50