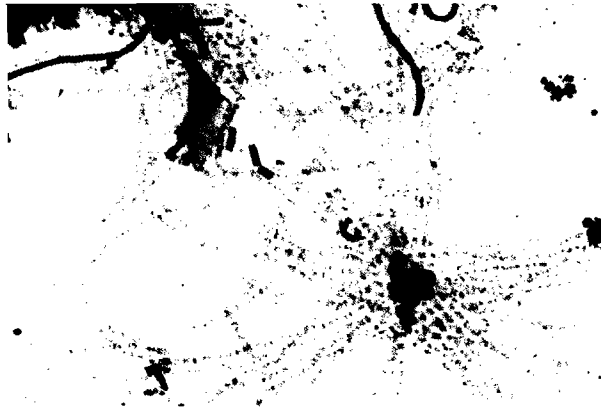


**Microscopic Evaluation of Activated Sludge
from Eleven Wastewater Treatment Plants in
Cape Town, South Africa**

Pamela Welz
Student number: 20161794

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Supervisor: Dr HA Esterhuysen

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ABSTRACT

From June to November 2007, a microscopic analysis was conducted on the activated sludge from eleven selected wastewater treatment plants (WWTP's) belonging to the City of Cape Town. The primary objective was the identification of the dominant and secondary filamentous organisms. Other important criteria included were the floc character, diversity, filament index (FI) and identification of the protozoan and metazoan communities. The operational data determined from routine analyses of the sludge, influent and effluent were used to assess the relationship of the filamentous population to wastewater characteristics and to compare this with previous findings. FI values of ≥ 3 and dissolved sludge volume indices (DSVI's) of ≥ 150 were chosen as representing the possibility of bulking conditions being present. The five most prevalent dominant filaments were Type 0092, Type 1851, actinomycetes, *Microthrix parvicella* and Type 021N, being present in 74%, 31%, 22%, 17% and 14% of samples respectively. Type 0092 did not appear to be associated with bulking in any of the WWTP's, although it was often incidentally present as a co-dominant species when bulking conditions existed. All three WWTP's with the Modified Ludzack-Ettinger configuration harboured Type 1851 as the major dominant species, irrespective of whether the plants treated domestic or industrial effluent. Conditions suggestive of bulking were present in two of these WWTP's. Contrary to expectations, Type 1851 was often found as a dominant species where domestic waste was the primary influent. Type 021N and actinomycetes were strongly implicated when bulking occurred. The overgrowth of these filaments appeared to be related to factors such as nutrient deficiency (Type 021N) or the presence of large amounts of low molecular weight substances in the influent. *Microthrix parvicella* did not cause major bulking problems. There was a strong association between low levels of nitrates/nitrites in the clarifier supernatant and good phosphorous removal, irrespective of the configuration of the WWTP. The converse was also true.

Keywords: Microscopic analysis, Activated sludge, Filamentous organisms, Operational data, Bulking

OPSOMMING

Gedurende Junie tot November 2007, is mikroskopiese analises op die geaktiveerde slyk van elf geselekteerde afvalwaterbehandelingsisteme van die Stad van Kaapstad uitgevoer. Die primêre doel was die identifisering van die dominante en sekondêre filamentagtige organismes. Ander belangrike kriteria het ingesluit die vlokarakter, diversiteit, filamentindeks (FI) en die identifikasie van die protosoë en metasoë populasies. Die bedryfs data van roetine analises van die slyk, invloei en uitvloei was gebruik in die assessering van die verwantskap tussen die filamentpopulasie tot afvalwatereienskappe en om te vergelyk met vorige bevindings. FI-waardes van ≥ 3 en opgeloste slyk volume indekse van ≥ 150 was gekies om die moontlikheid van uitduende slyk toestande te verteenwoordig. Die vyf mees oorwegend dominante filamente was Tipe 0092, Tipe 1851, *Actinomyces*, *Microthrix parvicella* en Tipe 021N, teenwoordig as 74%, 31%, 22%, 17% en 14% in die monsters, respektiewelik. Tipe 0092 was nie geassosieer met uitduende slyk in enige van die afvalwaterbehandelingsisteme nie, alhoewel dit dikwels as mede-dominante spesie, indien toestande vir uitduende slyk voorkom, teenwoordig was. In al die afvalwaterbehandelingsisteme met die Modified Ludzack-Ettinger konfigurasie was Tipe 1851 die hoof dominante spesie, ongeag of die sisteem huishoudelike of industriële uitvloeisel behandel. Toestande wat aanduidend is vir uitduende slyk was teenwoordig in twee van hierdie afvalwaterbehandelingsisteme. In teenstelling met die verwagtings, was Tipe 1851 dikwels as die dominante spesie in sisteme wat huishoudelike afval as primêre invloei het, gevind. Die teenwoordigheid van Tipe 021N en *Actinomyces* word sterk geassosieer met uitduende slyk. Die oorgroei van hierdie filamente is blykbaar verwant aan faktore soos voedingstoftekorte (Tipe 021N) of die teenwoordigheid van groot hoeveelhede lae molekulêre gewig bestanddele in die invloei. *Microthrix parvicella* veroorsaak nie groot uitduende slyk probleme nie. Daar is 'n sterk assosiasie tussen lae vlakke van nitrate/nitriete in die supernatant van die besinkingsdam en goeie fosfaatverwydering, ongeag die konfigurasie van die geaktiveerde slykbehandlingsisteem. Die teenoorgestelde is ook waar.

Sleutelwoorde: Mikroskopiese analises, Geaktiveerde slyk, Filamentagtige organismes, Bedryfs data, Uitduende

ABBREVIATIONS

AA: Anaerobic/aerobic
AMF: Aerobic mass fraction
ATP: Adenosine triphosphate
AOO: Ammonium oxidizing organism
AS: Activated sludge
BNR: Biological nitrogen removal
BPR: Biological phosphorous removal
BOD: Biological oxygen demand
BOD_L: Ultimate biological oxygen demand
BOD₅: Five day biological oxygen demand
COD: Chemical oxygen demand
COD/N: Chemical oxygen demand to nitrogen ratio
COD/P: Chemical oxygen demand to phosphorous ratio
DO: Dissolved oxygen
DPOA: Denitrifying phosphorous accumulating organism
DSVI: Dissolved sludge volume index
EFF: Effluent
EPBR: Enhanced biological phosphate removal
FI: Filament index
F/M: Food/microorganism ratio
GAO: Glycogen accumulating organism
HMW: High molecular weight
IAND: Intermittently aerated nitrification-denitrification
IN: Influent
LMW: Low molecular weight
MCRT: Mean cell residence time
MLE: Modified Ludzack-Ettinger
MLR: Mixed liquor recycle
MLSS: Mixed liquor suspended solids
MLVSS: Mixed liquor volatile suspended solids
MUCT: Modified UCT
NDBEPR: Nitrification-denitrification biological excess phosphorous removal
NOO: Nitrite oxidizing organism
OUR: Oxygen utilization rate
P: Phosphorous
PAO: Phosphorous accumulating organism
PHA: Polyhydroxyalkanoate
PHB: Polyhydroxybutyrate
PST: Primary settling tank
RAS: Return activated sludge
RASSS: Return activated sludge suspended solids
RBCOD: Readily biodegradable COD
SBCOD: Slowly biodegradable COD
SR: Sludge recycle
SRT: Solids retention time
SS: Suspended solids
SUR: Substrate utilization rate
SVI: Sludge volume index
TKN: Total Kjeldahl nitrogen

μ_{\max} : Maximum specific growth rate
UCT: University of Cape Town
VFA: Volatile fatty acid
VFA/N: Volatile fatty acid to nitrogen
VSS: Volatile suspended solids
WAS: Waste activated sludge
WF: Waste flow
WWTP: Wastewater treatment plant
2RND: 2-reactor nitrification-denitrification

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CHAPTER 1

INTRODUCTION

1.1 REASONS FOR STUDY

The primary practical objective of the study was to conduct a microscopic analysis over a period of six months on the activated sludge from eleven selected wastewater treatment plants (WWTP's) belonging to the City of Cape Town. This took place from June to November 2007.

Literature research has revealed only two publications on filamentous organism surveys conducted in South Africa: Blackbeard *et al.* (1988) and Lacko *et al.* (1999). In the first study, 33 WWTP's from over South Africa were included. At this time, biological nutrient removal was in its infancy and knowledge on the control of bulking was limited. This resulted in the situation where filamentous bulking sludges were found in approximately three quarters of the study WWTP's. It stands to reason that with a more modern approach, the microbial composition may have changed. In the survey by Lacko *et al.* (1999) the frequency, dominance and seasonal variation of filamentous organisms from six WWTP's from the KwaZulu-Natal were determined. In both of these surveys, the filamentous population was identified primarily by morphology ascertained by light microscopy.

The proposed study was more extensive than either of the above and some of the objectives were:

- To microscopically determine the current composition of the filamentous population in the activated sludge in the Cape Town geographical area. (Routine microscopic analysis is not performed on activated sludge samples from WWTP's in this city).
- To perform a more extensive microscopic analysis, not only limited to the frequency and dominance of the filamentous organism population.
- To compare the microbial composition so determined to that obtained from other geographical locations, both local and global, where available.
- To assess the relationship of the filamentous population to wastewater characteristics and to compare these with expected findings, chiefly those reported by the leading authors in this field, namely Eikelboom (2000) and Jenkins *et al.* (2004).
- To assess any changes from winter to summer in both the prokaryotic and eukaryotic microbiota and to compare these with expected findings in a similar manner.
- To obtain operational data from the WWTP's included in the study.
- To expand the analysis of the microscopic results to encompass the use of the operational data as a marker of plant efficiency, especially in terms of bulking and nutrient removal.
- To extrapolate mechanisms obtained using bench-scale experimentation to full-scale operation on a number of WWTP's and to assess the functional validity thereof. The bulking hypothesis of Casey *et al.* (1999) was taken as a particular point of interest as there is to date no literature proving or refuting this hypothesis as a common cause of bulking in full-scale wastewater treatment plants.

1.2 DATA SOURCES

The investigation consisted of three parts that were dealt with individually and in an integrated manner, namely the plant configuration and operating parameters, the microscopic analysis and the operational data.

1.2.1 PLANT CONFIGURATION AND OPERATING PARAMETERS

This information was supplied by the individual WWTP managers and the Scientific Services department of the City of Cape Town and included: flow rates, design dissolved oxygen (DO) levels and reactor volumes. Loading rates, important in filament analysis and measured in the amount of chemical oxygen (mgCOD) to mixed liquor suspended solids ratio (mgMLVSS) per day (mgCOD/mgMLVSS.day), were calculated using this information, together with the operational data.

1.2.2 MICROSCOPIC ANALYSIS

This was performed exclusively by the masters' student, using the methods of Eikelboom (2000) and supplemented by those of Jenkins *et al.* (2004). The analysis was performed on grab samples of mixed liquor from the weir overflow site exiting the bioreactor. The results included:

- Floc characterization
- Analysis of bacterial diversity
- The presence and quantitation of monocolonies, spirochaetes and spirils
- The determination of filament index
- Filament identification and quantitation
- Identification and quantitation of protozoa and metazoa

Although a variety of molecular methods have been developed or are in the process of being developed for microbial sludge analysis, there is still a place for classical direct microscopy in assessing the functional efficiency of WWTP's. A holistic approach incorporating all the available methodology is ideal, but not always practically feasible. This fact is elucidated upon in chapter 2.

1.2.3 OPERATIONAL DATA

These results were included retrospectively. This was to prevent any influence that the knowledge of plant parameters may have had on the microscopic results - if results were known prior to the microscopic analysis, it may have inadvertently influenced the outcome. The influent values used were either from results obtained from the supernatant of the primary settling tanks (PST's), or in the absence of PST's, the raw influent into the bioreactor. Mixed liquor samples were as per the microscopic analysis and effluent samples were those obtained from the clarifier supernatant. Routine weekly testing is performed on samples from the WWTP's belonging to the City of Cape Town. The data below were included in the study. Additional data was included at a later date if deemed necessary to explain pertinent features of the analysis.

- Influent and effluent chemical oxygen demand (mgCOD/L)
- Influent, effluent and mixed liquor suspended solids (105°Cmg/L)
- Influent total Kjeldahl nitrogen and ammonia (Nmg/L)
- Effluent ammonia and nitrates/nitrites (Nmg/L)

- Influent total phosphates, influent and effluent ortho-phosphates (Pmg/L)
- Influent and effluent chloride (Cl⁻mg/L)
- Influent and effluent alkalinity (CaCO₃mg/L)
- Influent and effluent conductivity (mS/m)
- Mixed liquor volatile suspended solids (mg/L)
- Mixed liquor settleable solids (30min ml/L)
- Mixed liquor sludge volume index and dissolved sludge volume index (mL/g)
- Sludge age (days)

1.3 DATA ANALYSIS

As mentioned previously, the analysis was divided into two sections, namely the individual plant analyses and the meta-analysis.

1.3.1 INDIVIDUAL PLANT ANALYSES

Each WWTP was assessed as a single entity, with some inter-plant comparisons being made. The content included analysis of the efficiency of the WWTP, especially in terms of bulking and nutrient removal. The presence of bulking conditions was assessed using the dissolved sludge volume index (DSVI) and the filament index (FI) as indicators. Nutrient removal was assessed by analysis of the available operational data. The reason/s for any loss of efficiency was/were speculated upon according to literature findings. This was tied in with an analysis of the microbial community, according to prevailing conditions. The similarities and differences to the associations reported in the literature were elucidated upon.

1.3.2 META-ANALYSIS

This included combined data from all eleven WWTP's and was structured according to:

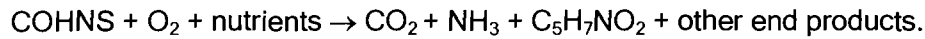
- Population dynamics of the filamentous organism population
 - Prevalence and comparison with similar South African studies
 - Changes from winter to summer over a six month period
 - Correlation with DSVI and comparison to findings of Eikelboom (2000)
 - Discussion of the most prevalent filament types regarding the documented organism characteristics, and comparison with study findings
- Population dynamics of the protozoa and metazoa
 - Changes from winter to summer over a six month period
 - Discussion of the documented organism characteristics and comparison with the findings of the study

CHAPTER 2

BACKGROUND LITERATURE

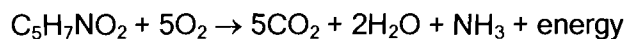
2.1 BACTERIAL METABOLISM AND WASTEWATER TREATMENT

In a functional activated sludge system under aerobic conditions the synthesis of biomass ($C_5H_7NO_2$) from organic matter (COHNS) takes place according to the equation:



(Tchobanglous and Burton, 1991)

Decomposition of the biomass takes place by a process of endogenous respiration. This is enhanced when nutrients have been expended and is related to the equation:



(Tchobanglous and Burton, 1991)

At the beginning of aeration, the nutrient concentration, or food (F) is at its highest, allowing aerobic bacteria to grow quickly, increasing the biomass (M). Thus, during aeration the food/microorganism (F/M) ratio decreases logarithmically until if and when the supply of nutrients is expended. At low F/M ratios, the cells of many strains of bacteria stick together forming flocs, which can be separated from the water by settling in a clarifier (Eikelboom, 2000).

For optimal settling results in the clarifier, microbial growth in the mixed liquor should be in the endogenous phase and excess sludge should be wasted to maintain the correct F/M ratio, desired MLSS and sludge age (Veissman and Hammer, 1998).

For process stability, the aim is to create conditions that allow for culture of the desired bacterial strains. These strains should consume the largest part of the influent nutrients. Due to ongoing competition for available nutrients, the population is constantly changing due to differences in a variety of factors. The operational parameters and physical conditions that have the largest effect are: sludge load, influent composition, DO and temperature (Eikelboom, 2000).

The configuration of the plant has an effect on the nutrient concentration and loading rate of the activated sludge (Eikelboom, 2000)

2.2 NUTRIENT REMOVAL AND BULKING

2.2.1 Nitrification/denitrification

The bulk of nitrogen removal takes place by assimilation and nitrification/denitrification. In the former, ammonia is incorporated into cell mass that may be released when the cell lyses and dies. The nitrifiers are sensitive to a variety of inhibitors, temperature, pH and oxygen concentration. By nitrification, the oxygen demand of ammonia is reduced and a large amount of alkalinity is consumed (Metcalf and Eddy, 1991).

The nitrifiers are chemolithotrophs that obtain their energy from the oxidation of either ammonia or nitrite (or both) that serve as electron donors for the chemiosmotic generation of adenosine triphosphate (ATP). They do not need preformed organic substances, but use inorganic CO_2 as the source of carbon for the synthesis of organic cellular compounds (Atlas, 1997).

In contrast, the heterotrophs are chemoorganotrophs that use organic molecules as their source of energy, electrons and carbon. A relatively low amount of energy is produced by nitrification, and about 35 moles of ammonia or 100 moles of nitrite are needed to fix one mole of carbon dioxide. This leads to a situation where the nitrification process is very high, but the growth of the bacteria very slow, especially in comparison to the heterotrophic population (Atlas, 1997).

Low oxygen conditions give many heterotrophic bacteria a competitive advantage over nitrifiers. Nitrifiers conserve less electrons in biomass and have a maximum yield coefficient that is about one quarter that of heterotrophs. The maximum specific growth rate is so small that large mean cell residence times (MCRT's), especially at low temperatures, in conjunction with adequate dissolved oxygen (DO), stable pH, and low inhibitor levels are needed to prevent washout (Rittman and McCarty, 2001).

Metcalf and Eddy (1991) have published a ratio of BOD₅/TKN of 1-3 to be expected in separate stage nitrification. They have determined that this will yield an estimated fraction of nitrifiers in the biomass of 0.21 (at 1), to 0.083 (at 3). Furthermore, they state that a ratio of approximately >5 would be expected in combined carbon oxidation and nitrification processes.

Denitrification is employed when complete nitrogen removal from wastewater is required. Most wastewater contains reduced nitrogen that must be oxidized to nitrate or nitrite to allow for denitrification - this is performed chiefly by facultatively aerobic heterotrophs. In oxygen-limited conditions, these organisms use nitrate and/or nitrite as a final electron acceptor during anaerobic respiration, thereby reducing nitrate or nitrite. Some denitrifiers are autotrophs that denitrify using hydrogen or reduced sulphur instead of organic carbon as electron donors. In contrast to nitrification, denitrification produces alkalinity (Rittman and McCarty, 2001).

| TABLE 2.1 THE REACTIONS AND THE CATALYZING ENZYMES FOR DISSIMILATORY NITRATE REDUCTION | |
|--------------------------------------------------------------------------------------------------------|--------------------------------|
| Equation | Enzyme |
| $\text{NO}_3^- + 2\text{e}^- + 2\text{H}^+ \rightarrow \text{NO}_2^- + \text{H}_2\text{O}$ | <i>nitrite reductase</i> |
| $\text{NO}_2^- + \text{e}^- + 2\text{H}^+ \rightarrow \text{NO} + \text{H}_2\text{O}$ | <i>nitrate reductase</i> |
| $2\text{NO} + 2\text{e}^- + 2\text{H}^+ \rightarrow \text{N}_2\text{O} + \text{H}_2\text{O}$ | <i>nitric oxide reductase</i> |
| $\text{N}_2\text{O} + 2\text{e}^- + 2\text{H}^+ \rightarrow \text{N}_2(\text{g}) + \text{H}_2\text{O}$ | <i>nitrous oxide reductase</i> |

(Rittman and McCarty, 2001)

According to Rittman and McCarty (2001), oxygen controls denitrification in two ways: by repression of the reductase genes and by inhibiting the activity of the enzymes themselves. The latter takes place at a much higher level of DO than the former, so that denitrification can still take place in the presence of oxygen, especially within flocs where the concentration is lower than in the bulk liquid. Nitrate reductase is repressed less than the other reductases at high DO, so the intermediates, which are greenhouse gases, may accumulate. This can also happen when the concentration of electron donor that drives the half-reactions is low.

Komorowska-Kaufman *et al.* (2006), employed batch testing that mimicked a Bardenpho configuration to demonstrate the effects of various factors on denitrification. These included the effect of additional carbon source, the effect of the volatile fatty acid to nitrate (VFA/N) ratio and the effect of the primary nitrate concentration in the anoxic zone. The authors found that with additional carbon, intensive denitrification took place in the first hour of anoxic conditions being established. If the VFA concentration was low and there was no additional carbon added, the intensive period was only thirty minutes and the overall process took double the time. An increase in the initial nitrate concentration caused enhanced denitrification efficiency, but not enough to prevent a lengthening of time required for denitrification. They also showed that intensive denitrification was optimal at a ratio of acetate to initial nitrate concentration of 1.67 mg CH₃COOH/mg N. From a lower value, as the ratio increased towards the optimum, the intensive denitrification time shortened and the intensive denitrification rate and efficiency increased. They determined that an increase over this optimum caused a decrease in denitrification rate.

2.2.2 Phosphorous removal

Orthophosphate, polyphosphate and organically bound phosphate occur in wastewater. After primary sedimentation, most is in the soluble form. Conventional secondary activated sludge (AS) processes reduce phosphorous to around 6 mg/L, but for protected watersheds a maximum value of around 1mg/L should be found. This is because it is the primary limiting nutrient in most natural water systems and the presence of this element is thus strongly related to eutrophication. Phosphorus may be removed by precipitation before, during or after biological treatment, or solely by biological means. If the removal of nitrogenous waste is also a requisite, then the longer solids retention times (SRT's) necessary for nitrification will lead to lower phosphorus removal rates (Rittman and McCarty, 2001).

A recent review article by Oehman *et al.* (2007) exhaustively details all elements of the enhanced biological phosphorous removal (EBPR) process in order to endeavour to explain why disturbances and prolonged periods of poor removal have been seen in full-scale plants under seemingly favourable conditions. In the absence of external forces such as high rainfall causing over-aeration in the anaerobic zone, excessive nitrate loading to the anaerobic zone or nutrient limitation, the chief reason for failure of phosphorus removal seems to be an increased ratio of glycogen accumulating organisms (GAO's) to phosphorous accumulating organisms (PAO's). There is plausible evidence that *Accumulibacter* spp. is a common PAO as this organism has been identified by molecular techniques in many efficient EBPR pilot-scale and full-scale plants (Oehman *et al.* 2007). The review article points out that these findings have been substantiated by metabolic studies, such as those conducted by McMahon *et al.* (2002), where they detected the gene sequence for polyphosphate kinase as well as the gene products of this enzyme in *Accumulibacter* spp. *Competibacter* spp. has likewise been found to belong to the GAO organism group and studies often use these two organisms as indicator species. Both GAO's and PAO's compete for substrate, mainly VFA's formed by fermentation of influent slowly biodegradeable chemical oxygen demand (SBCOD) and particulate organics. In a domestic WWTP setting, VFA's consist chiefly of acetate and to a lesser extent propionate. Concentrations of the latter can be increased by pre-fermentation of the influent. Amino acids and sugars as substrate may be relevant in an industrial setting. Oehman *et al.* (2007) also cite strong evidence that when conditions favour the growth of PAO's over GAO's, phosphorous removal is enhanced and *vice versa*.

Cech and Hartman (1993) found that acetate supported the growth of both organism types, but when used as the sole substrate, the PAO's grew preferentially and for the most part phosphorous removal was good. There were short periods when performance deteriorated, which the researchers ascribed to predation of the PAO's by a flagellate bloom. When glucose was used as a carbon source in addition to acetate, GAO's dominated the population.

Liu *et al.* (1997) determined that the ratio of phosphorous to carbon was an important factor influencing dominance of either PAO's or GAO's. Ratios of less than 2:100 favoured the growth of GAO's, while higher concentrations of 10:100 or more favoured the growth of PAO's. According to the authors, a chemical oxygen demand to phosphorous concentration ratio (COD:P) of 10-20mgCOD/mgP should favour the PAO's. Their theory is: PAO's take up acetate at a faster rate than GAO's anaerobically (substantiated experimentally). They proposed that because the uptake mechanism is energy dependent and relies on the stores of polyphosphate that have accumulated under aerobic conditions that if the supply of phosphorous is below the threshold level, the GAO's with their glycogen energy source can out-compete the PAO's for substrate. Furthermore, providing there is sufficient acetate, GAO's will still be present when phosphorous levels are high and the ratio of PAO's to GAO's is dependent on the concentration of substrate and phosphorous.

In their article, Oehman *et al.* (2007) also elucidated on the organisms dubbed as denitrifying phosphate accumulating organisms (DPAO's). These PAO's are capable of using nitrate/nitrite as terminal electron acceptors and can take up phosphate and denitrify simultaneously under anoxic conditions. They point out the not only a low COD:P ratio, but sufficient VFA's need to be present for EBPR. Propionate has proven to be a better substrate than acetate for EBPR ostensibly because *Accumulibacter* spp. seem to be able to switch between propionate and acetate while *Competibacter* spp. much prefer acetate. However, other GAO's show a preference for propionate. Oehman *et al.* (2007) postulate that GAO's prefer only one substrate, be it propionate or acetate and that by manipulating pre-fermentation conditions to enhance substrate switches it would lead to a preferential growth of PAO's. The authors refer to studies incorporating both substrates when alternation of substrate eliminated GAO's almost entirely.

The physical parameters of pH and temperature are also factors to be taken into consideration. A pH over about 7.25 leads to a better substrate uptake by PAO's in comparison to GAO's with EBPR. The converse also holds true (Oehman *et al.* 2007). This is because the energy consumption by the former for the uptake of acetate is higher at increased pH, leading to increased anaerobic polyphosphate degradation and release. Lower temperatures favour the growth of PAO's and higher temperatures that of GAO's. EBPR is still satisfactory at 20°C, but is retarded at 30°C. Saito *et al.* (2004) found that accumulation of nitrite in the anoxic zone inhibits the growth rate of PAO's.

In terms of the process conditions, the presence of nitrate in the anaerobic zone allows the ordinary heterotrophic population to compete for VFA's as substrate. Pre-denitrification demands high mixed liquor recycle from the aerobic to the anoxic zone to provide polyhydroxyalkanoate (PHA) rich PAO sludge with nitrates for denitrifying phosphorous removal and for denitrification of ammonia from the aerobic zone. The mixed liquor recycle from the anoxic to the anaerobic zone should be at the end of the anoxic zone and contain little nitrates and nitrites (Oehman *et al.*, 2007).

2.3 BULKING

Some bacteria, fungi or algae do not become detached from one another on cell division and grow into filaments. Certain bacteria form filaments under almost all conditions and there are around 30 species that are observed in activated sludge, with 10 being frequently found. Filamentous organisms are normally present in activated sludge. When conditions are created that allow them to out-compete the other bacteria to such an extent that they grow en masse, problems may be encountered: bulking sludge, deterioration of settling and dewatering properties and sometimes scum formation. The number of filaments present is not the only parameter determining sludge settling and the SVI or DSVI does not always correlate directly with the filament index (Eikelboom 2000).

Jenkins *et al.* (2004) advocates the following approach to correct solids separation problems:

- > Perform a microscopic examination.
- > Determine the probable cause of the problem using the information from the microscopic examination, coupled with knowledge of plant operating conditions and wastewater characteristics.
- > Make appropriate operating changes. These may be minor or major. Problems needing minor changes include septic wastewater and nutrient deficiency. Major changes would include design or operating changes such as installing additional aeration capacity or changing the aeration basin configuration.

If clarifier capacity is sufficient, even sludges with high DSVI's may settle sufficiently. The following equation gives a basic understanding of this:

$G_a = X (Q+Q_r)/A$, where

- G_a is the allowable applied suspended solids (SS) loading rate
- X is the mixed liquor suspended solids (MLSS) concentration
- Q is the influent flow rate
- Q_r is the return activated sludge (RAS) flow rate
- A is the clarifier surface area
- Q_r/A is the underflow RAS rate

This equation is simplified and does not take into account factors such as the settling characteristics of the AS itself. More complicated calculation techniques exist, that relate G_a , RASSS, Q_r/A and a range of sludge volume indices (SVI's) in a diagrammatic form. These Clarifier Operating Diagrams are used to empirically determine loading rates by correlation of settling tests with points on the diagrams. Ideally, empirical values should be compared with actual performance as a method of calibration (Jenkins *et al.*, 2004).

Under growth limiting conditions, the growth rate of floc-forming organisms generally decreases more than that of filamentous organisms. Only at very high nutrient (loading) levels, can the faster growing floc-formers out-compete the filaments. Some filamentous organisms also have competitive nutrient requirements. If certain elements or compounds are present in large enough quantities, these filaments have an advantage. For example, *Thiothrix* spp. and Type 021N, amongst others, can utilize reduced sulphur for growth. Most filamentous organisms are aerobic, so the creation of anoxic or anaerobic conditions will generally allow the growth of anaerobic or facultative floc-formers, but not filaments (Jenkins *et al.*, 2000). Eikelboom (2000) postulates that this is

because most filaments can absorb free higher fatty acids from the water phase under anaerobic and anoxic conditions, whereas most floc-formers can only absorb and process them aerobically.

Prolific growth of so-called “low F/M filaments” cannot be controlled by the selector effect in intermittently aerated systems used for biological nitrogen removal (BNR) such as Carrousel® and Orbal plants. This is also the case for multi-reactor anaerobic-anoxic-aerobic nitrification-denitrification enhanced biological phosphorous removal (NDBEPR) plants such as the modified University of Cape Town (MUCT) or Bardenpho configurations. True “low F/M filaments” are classified by Jenkins *et al.* (2004) as being selected when loading rates are in the range of 0.05-0.2 kg BOD⁵/kgMLSS.day.

They are as follows:

- > Type 0675, Type 1851, Type 0803
- > Type 0041 (may also be selected if there is pre-mixing of influent with RAS).
- > Type 0092, Type 0581, Type 0914 (These may also be selected in conditions of high concentrations of low molecular weight (LMW) organic acids. In addition, the presence of H₂S favours the growth of Type 0914).
- > *Microthrix parvicella* (there are many other conditions that favour the growth of this problem species such as abundant fatty acids, low temperatures (<15°C), reduced nitrogenous and sulphurous waste).

Lakay *et al.* (1999) conducted a number of laboratory-scale experiments in order to determine factors contributing to the growth of “low F/M filaments”. The authors mention the classification system of Jenkins *et al.* (2004) and refer to “low F/M filaments” in this context. In reality, many of the filaments identified at increased DSVI values were not classical “low F/M” filaments according to Jenkins *et al.* (2004). *Haliscomenobacter hydrosis* (*H. hydrosis*) in particular was referred to repeatedly as being “low F/M”. (Perhaps using terminology such as “possible bulking conditions” or “filamentous overgrowth” in the article would have been more accurate).

The substrates employed in the experiments by Lakay *et al.* (1999) were:

- > Municipal sewage
- > Municipal sewage subjected to ultrafiltration to give both readily biodegradable chemical oxygen demand (RBCOD) and slowly biodegradable chemical oxygen demand (SBCOD)
- > Defined substrate. The reason for the defined substrate was to eliminate variation in composition that may have affected low F/M filament growth.

Nitrate dosing was employed in all experiments in order to maintain the anoxic conditions and compare nitrate utilization in some instances. Filament identification for dominant, secondary and tertiary organisms was performed approximately every thirty days, or more randomly in some cases, according to requirements.

Evidence from the feeding regimes and filament identification led Lakay *et al.* (1999) to suggest that bulking can occur with either municipal or defined substrate (RBCOD and SBCOD). However, two of the three dominant filaments, namely *H. hydrosis* and Type 021N are not classified by Jenkins *et al.* (2004) as “low F/M”.

Lakay *et al.* (1999) found an increase in DSVI associated with anoxic conditions and concluded that RBCOD supplied under anoxic conditions resulted in “low F/M” proliferation. A similar experiment by the same authors employing defined substrate

yielded substantially higher DSVI values associated with *H. hydrosis* and type 1851. *H. hydrosis* was also the dominant species during protracted anoxic conditions, where it was associated with low DSVI values in this occasion. DSVI values increased during periods of intermittent aeration compared to either aerobic or anoxic conditions. The results of Lakay *et al.* (1999) with the unfiltered sewage and alternating continual and intermittent aeration concurred with those of Ketley *et al.* (1991) who found that intermittent aeration caused an increased DSVI associated with *Microthrix parvicella* (*M. parvicella*) dominance. The highest DSVI values were obtained with a 30-40% aerobic mass fraction for both intermittently aerated nitrification-denitrification (IAND) and two modified UCT (MUCT) systems. With the MUCT systems, type 0092 was the dominant species for the duration of the experiment.

Warburton *et al.* (1991) showed that there was little effect on DSVI, filament proliferation or filament species when the sludge age was decreased from 20 to 10 days. When sludge age was further decreased, filament population remained similar, but the organisms were less proliferative. However, such sludge ages are not appropriate in nutrient removal plants.

Casey *et al.* (1994a) demonstrated that lower DSVI values were obtained for 2 reactor nitrification-denitrification systems (2RND) operated at the same aerobic mass fraction as an IAND system. Further to this, the author compared differences between both systems and sought to establish clarity on the facets discussed in the following paragraphs:

- > The frequency of exposure of IAND and 2RND systems to alternating anoxic-aerobic cycles. For IAND systems this is usually high (>30/day), while that of 2RND systems is low (<5/day) due to the low aerobic-anoxic and sludge recycle ratios. Lakay *et al.* (1999) refer to studies by Ketley *et al.* (1991) and Hulsman *et al.* (1992) who determined that the frequency did not affect "low F/M" proliferation under the influent composition and operating conditions they used.
- > RBCOD concentration differences between the two systems. The rationale for these experiments by Lakay *et al.* (1999) was that both RBCOD and SBCOD are fed to an IAND system in proportion to the respective aerobic/anoxic mass fractions. In 2RND systems, the RBCOD will be utilized either aerobically (Wuhrman plant configuration) or anaerobically (Modified Ludzack-Ettinger plant configuration). In a typical domestic WWTP, operating at an aerobic mass fraction of 30%, all the RBCOD is utilized in the first reactor. The second reactor thus only has SBCOD as substrate. A 2RND system was operated in Modified Ludzack-Ettinger (MLE) configuration with a steady-state DSVI of approximately 130 ml/g. Influent sewage (approximately 20% RBCOD) was then diverted into both the aerobic and anoxic reactors to simulate an IAND system. There was no significant change in DSVI, demonstrating that the system configuration and thus environmental conditions does not affect "low F/M" growth.
- > The DO concentration in the aerobic period or reactor. In IAND systems, there is a high DO concentration at the start of aeration and then a decreasing gradient until oxygen is expended by biological action. With a 2RND system, oxygen is either present at a high concentration in the aerobic reactor, or entirely absent in the anoxic reactor. Results of the experiments by Lakay *et al.* (1999) provided evidence that "low F/M" proliferation in IAND configurations is not due to decreasing DO gradient.

- Nitrate concentrations in the anoxic period of IAND and 2RND systems. The findings of these experiments by Lakay *et al.* (1999) were novel and form the basis for an important model on the causes of filamentous bulking, so the discussion is more protracted and detailed:

Nitrate concentration in an IAND system is high at the end of the aerobic period due to nitrification. The nitrate concentration then decreases during the anoxic period due to denitrification and may be completely expended if the anoxic period is long enough. However, there is little variation in nitrate concentration with short anoxic periods. In an IAND system, there is a large gradual change in redox potential chiefly due to the varying concentrations of the electron acceptors nitrate, nitrite and oxygen. Oxygen levels are high at the beginning of the aeration cycle, and this element is used as the preferential electron acceptor, until depletion. At this stage, nitrates and then nitrites assume this role, until they too are depleted and anaerobic conditions ensue. In contrast, the anoxic zone of a 2RND system will have a constant (usually low to zero) nitrate concentration depending on the nitrate load from the recycle and the denitrification potential of the system. A different but constant redox potential is established in the aerobic and anoxic zones. This means that biota being transferred from one reactor to another will be subjected to large and sudden changes in redox potential (Lakay *et al.*, 1999).

Lakay *et al.* (1999) measured the redox potential for over a month in a 2RND system (MLE) and found an average value of -81mV in the anaerobic zone and +48mV in the aerobic zone. When the same system was changed to an IAND configuration, the DSVI values increased substantially. In one complete 8-hour cycle, the redox potential increased during the aeration cycle to a maximum of +40mV and declined to a minimum of -80mV during the anoxic cycle. These are almost identical to the figures obtained from the 2RND system. The authors deduced experimentally that the presence of nitrites and nitrates present throughout the cycle created a propensity for overgrowth of "low F/M" filaments.

Musvoto *et al.* (1999) conducted further experimentation in order to ascertain the relative implication of nitrate and/or nitrite as the causative agent of bulking in nutrient removal plants. At this point, it was recognized that what they had termed "low F/M bulking" was perhaps a misleading label, because their research had shown that the most probable cause of filamentous bulking in nutrient removal WWTP's was alternating anoxic/aerobic conditions. They thus termed this group of organisms "AA filaments". MUCT systems were dosed separately with excess nitrate and nitrite. Dissolved sludge volume index (DSVI) values were determined and nitrate and nitrite levels were measured. It was found that the rate of DSVI increase for nitrate dosing (with subsequent high nitrate and low nitrite levels in the second anoxic reactor) was 1.0 ml/g/day. The rate of DSVI increase for nitrite dosing (with high nitrite and low nitrate levels in the second anoxic tank) was 1.5ml/g/day. Filaments identified during nitrate dosing included dominant filaments Type 0092 and Type 0914 and secondary filaments Type 0092, Type 0041, *M. parvicella* and *Beggiatoa* spp. During nitrite dosing, filaments were identified only once with Type 0092 being dominant and type 021N and Type 0675 secondary. All of the dominant filaments fall into the "low F/M" category of Jenkins *et al.* (2004).

Musvoto *et al.* (1999) concluded that in MUCT nitrogen and phosphorous removal systems, filament proliferation takes place when the sludge is subjected to sequential anoxic-aerobic conditions. In addition, nitrate and or nitrite levels of >2.0 and >1.0mgN/L respectively must be present in the anoxic zone immediately preceding the aerobic zone.

Furthermore, Musvoto *et al.* (1999) made the assumption that denitrification is mediated predominantly by facultative heterotrophs (filaments and floc formers). In this case, they pointed out that alternating anoxic and aerobic conditions would force these organisms to switch between the nitrates/nitrites and oxygen as the terminal electron acceptor and hypothesized that bulking occurs due to the efficiency of this switching mechanism by some filamentous organisms.

Casey *et al.* (1999c) proposed a biochemical model to explain AA bulking using those assumptions. Initially, a literature review of the biochemistry of heterotrophic respiratory metabolism was conducted Casey *et al.* (1999a). This was followed firstly by the formulation of a conceptual biochemical model of the behaviour of heterotrophic facultative aerobic organisms when subjected to aerated and unaerated conditions and secondly by a series of experiments to test this model (Casey *et al.*, 1999b). His theory is that, assuming both filaments and floc-formers are facultative aerobes, under completely aerobic or anoxic conditions, floc-forming heterotrophs out-compete filaments for substrate because they have higher substrate utilization rates (SUR's). However, when configurations are such that there are alternating aerobic and anoxic conditions, RBCOD levels are low or absent and the SUR of floc-formers is inhibited under aerobic conditions. The mechanisms proposed by Casey *et al.* (1999c) for the proliferation of AA filaments are summarized simplistically in the ensuing paragraph:

Cytochrome d is integral to aerobic respiration. It is inhibited by nitric oxide. When there is insufficient RBCOD, the intermediate nitric oxide is formed intracellularly in floc-formers, resulting in the inhibition of cytochrome d . When cytochrome d is inhibited, aerobic respiration is inhibited and electrons are directed to the reductase enzymes. Oxygen changes the permeability of the membrane so that nitrate cannot diffuse across the membrane to reach nitrate reductase. The pathway for aerobic denitrification of nitrite to nitric oxide (and further) is thus followed. When the supply of electrons is restricted, there is a preferential electron flow to nitrite reductase over nitric oxide reductase, leading to accumulation of nitric oxide. This obviously occurs when the levels of nitrite are high enough (as is the case with alternating aerobic anoxic conditions) for two reasons - firstly, the denitrifying bacteria are inhibited under aerobic conditions, so in subsequent anoxic periods, there is an accumulation of nitrite. Secondly, the inhibition of NOO's (and to a lesser extent other nitrifiers) under oxygen limitation also leads to nitrite accumulation. If there are sufficient electrons (as is the case with RBCOD) however, the supply of electrons from NADH is not restricted. The intracellular concentration of nitric oxide is kept low enough by its reductase to prevent inhibition of aerobic respiration. In contrast, the AA filaments only produce the enzyme for the reduction of nitrate to nitrite (nitrate reductase). The reductases responsible for the conversion of nitrite to nitric oxide and further are not manufactured. Therefore, under anoxic conditions the cytochrome d inhibitor, nitric oxide does not accumulate. Under ensuing aerobic conditions, aerobic respiration is not inhibited. This gives the AA filaments a competitive advantage.

The results of a variety of batch tests on AS were reported by Casey *et al.* (1999c) and these substantiated the theoretical model. For example it was determined that when nitrite was added to a system following an anoxic phase the inhibition of the OUR during the aerobic phase increased with increased nitrite concentration (at the start of the aerobic phase). There was no inhibition of aerobic respiration when nitrite was absent. When RBCOD in the form of sewage was added, inhibition was relieved.

Casey *et al.* (1999c) also determined that sludge with a high DSVI was assumed to have a high proportion of filamentous organisms and that with a low DSVI, a high proportion of

floc-formers. In 8/10 tests, sludge from an anoxic system with a high DSVI accumulated nitrite, but did not accumulate dinitrogen when subjected to nitrogen reduction tests. The converse was true of the sludge with low DSVI. Casey *et al.* (1999c) took this as supportive of the hypothesis that the enzyme system of AA filaments does not allow for the conversion of nitrite to dinitrogen - denitrification of nitrate to nitrite only occurs.

In light of the above, Casey *et al.* (1999c) proposed some control measures to prevent bulking in nutrient removal plants, including:

- > Aerobic mass fraction: avoid ranges between 30-40%
- > Reduce nitrite levels: reduce to near zero prior to sludge entering the aerobic zone. This can be achieved by reducing the a-recycle or increasing the unaerated mass fraction to ensure complete denitrification in the anoxic zone.
- > Reduce nitric oxide: reduce intracellular nitric oxide significantly if the above is impractical due to inflexible plant configuration or inherent wastewater characteristics. This can be achieved by the inclusion of a small aerobic or anoxic reactor upstream of the main aerobic reactor to which RBCOD (sewage wastewater) is fed.

Note:

There are many causes of bulking and means of correcting this phenomenon. An in-depth analysis of every possibility is beyond the scope of this study. Only factors playing a role in many sections of chapters 3-6 have been discussed. Other more specific facts are elucidated upon in chapters 3-6 if pertinent to a particular WWTP or microorganism. Similarly, although scum formation is an important cause of poor plant performance, it was not a major focus of the study, so is not discussed in detail.

2.4 MICROORGANISM POPULATION IN MIXED LIQUOR

Microscopic analysis enables the evaluation of conditions over a longer period of time than chemical data (Jenkins *et al.* 2004). Within the last decade, there has been a move to identify the microbial population from WWTP's by molecular methods. There are advantages and disadvantages to each approach, and although microscopic identification of filaments is out of vogue in academic research, it still has many positive attributes that should not be disregarded. The main reasons for the ineffectiveness of the traditional method is that it does not allow distinction of different organisms that are morphologically the same, nor does it allow for identification of the same organisms exhibiting variable morphology (Hug *et al.*, 2005).

The most popular molecular methods employed are Fluorescent *in situ* hybridization (FISH) and Denaturing Gradient Gel Electrophoresis (DGGE). 16S rRNA DGGE analysis has excellent sensitivity and allows the identification of minority constituents. Muyzer *et al.* (1992) found the method able to identify organisms representing only 1% of the population. Drawbacks are that the method is time-consuming and requires establishment of extensive clone libraries. In addition, the clone frequencies in the clone libraries do not reflect the *in situ* quantities of the organisms thus identified (Eschenhagen *et al.*, 2003). Sekiguchi *et al.* (2001) discovered that when DGGE methodology, several phylogenetic species formed part of a single band, and that in order to differentiate these organisms, it was necessary to repeat the procedure in triplicate.

FISH allows for the definitive identification of a range of microorganisms and is particularly useful in detecting non-filamentous organisms such as the species that form the PAO and GAO population (Eschenhagen *et al.*, 2003). Using FISH, it has been discovered that some Eikelboom morphotypes such as *M. parvicella* appear to represent single species (Liu *et al.*, 2001b). However, this is not always the case. The microscopic identification of *Nostoicoida limicola* (*N. limicola*) has proven to be problematic. Liu *et al.* (2001b) designed oligonucleotide probes for this group of filaments and discovered that *N. limicola* II can consist of a number of phylogenetically unrelated filaments and that *N. limicola* I and *N. limicola* II are probably the same organism that changes morphologically according to culture conditions. However, the technique they used required numerous probes and mismatches (a common problem) did occur.

FISH can be used successfully to quantitate species or groups in the sludge. Hug *et al.*, (2005) have devised a rapid system based on mathematical principles to quantitate organisms using FISH. However, this method is limited to those filaments that are not part of the flocs. Eschenhagen *et al.* (2003) performed a comparative study employing FISH and 16S rDNA analysis using DGGE and T-RFLP. They found the latter were far superior to FISH for the detection of community diversity. Due to the nature of the methodology employed by FISH, the identification can only be as reliable as the oligonucleotide probes employed. Mismatches are bound to occur, especially while the technology is still relatively new. As new probes are designed, this phenomenon should decrease (Muyzer *et al.* 1992).

Apart from the difficulties with the traditional microscopic identification of *N. limicola*, there are reported inconsistencies in the identification of Eikelboom Type 021N. Kanagawa *et al.* (2000) performed a variety of morphological, biochemical and molecular tests to definitively identify 15 strains of this morphological type isolated from bulking sludges. The molecular methods employed were the sequencing of 16S rDNA (employing PCR and automated sequencing) and FISH to allow differentiation of the so-called 021N-*Thiothrix* cluster. They used the results to differentiate the 021N morphotypes into three distinct groups (I, II and III). The FISH technique required the use of three probes and the authors discovered mismatches with type II.

Molecular methods, especially FISH have also been used successfully to differentiate between bacteria that are loosely attached to flocs with a propensity to become free-swimming and those bacteria that tend to form an integral part of the floc (Morgan-Sagastume *et al.*, 2007). The population was classified broadly, for example as *Beta*-, *Alpha*- and *Deltaproteobacteria* with 10 different group specific probes. The results obtained by Wilen *et al.* (2007) employing shear testing and FISH identification concurred that the population prone to shear is different from that which is strongly associated with floc formation.

A drawback of conventional microscopic filament identification is that the filaments inside the flocs are not visible. Liao *et al.* (2003) has used this as argument to promote the use of FISH. This was a drawback also reported by Gulez *et al.* (2008), who employed pilot-scale reactors and used conventional microscopy, FISH and DGGE in an effort to compare the methods for filament identification in activated sludge. Only three filaments were identified in the study - *Sphaerotilus natans* (*S. natans*), *Thiothrix* spp. and *N. limicola*. Even with this limited number of species, they discovered anomalies with each method and advocated multiple approaches to filament identification. They pointed out the problems inherent in each methodology: FISH has been biased by poor permeability of the cell walls of certain species. Indeed, in their study, *N. limicola* was not detected

from all samples using this method, possibly due to impermeability despite treatment with lysozyme and mutanolysin (supposed to prevent this occurrence). This strain was also not detected by DGGE, ostensibly due to the cell wall composition affecting DNA extraction. Besides these problems with FISH, Gulez *et al.* (2008) cite literature findings of bias due to low ribosome content, lack of specificity of probes, a higher order structure of either the target or the probe and fluorescence either causing background interference or being bleached (FISH). DGGE may give artefactual bands, single bands may represent more than one organism or there may be more than one band per organism. DGGE is a time-consuming procedure relying on nucleic acid extraction, PCR amplification, visualization of PCR products on a gel, excision of these products and subsequent sequencing Gulez *et al.* (2008). The advantages and disadvantages of the three methods are summed up in chapter 6.

Microscopic evaluation also allows a rapid assessment of floc quality and the composition of the protozoan and metazoan communities (Jenkins *et al.*, 2004). Microscopic analysis can be used effectively to qualitatively assess the sludge and to aid in troubleshooting WWTP's that are not functioning to expectation. The characteristics of the flocs formed in the mixed liquor play a large role in determining the settling characteristics of the sludge in the clarifier. Robust, compact flocs settle best. The more diverse the range of microorganisms encountered in the sludge, the more robust and flexible the degradation process. Low diversity usually only occurs when there is a lack of nutrients, such as would be encountered in a high-loaded industrial treatment plant (Eikelboom, 2000).

Floc characteristics from plants with similar process conditions closely resemble one another and have similar protozoa and metazoa. Eikelboom (2000) has established that 90% of flocs in Dutch WWTP's fall into three groups based on loading rates and type of aeration employed. Surface aeration tends to break up flocs, so they will be smaller than those from plants employing diffuse aeration. Flocs from industrial treatment plants often differ considerably from plant to plant and are often small and fragmented, especially in plants where chemical waste is being treated and surface aeration is used. The flocs may be irregular because of the growth of masses of filamentous organisms. With industrial plants, he advocates that sludge should be examined over a period of time and the individual floc characteristics established. Further samples can then be compared to this norm (Eikelboom, 2000).

Protozoa and metazoa may utilize bacteria and particulate organic matter as a source of energy, thereby performing a polishing function of the treatment process. The protozoan/metazoan population is indicative of the process conditions in the WWTP, with some species being used as indicator organisms. The contribution of bacteria to the overall treatment process is far greater than that of these organisms (Eikelboom, 2000).

Salvado *et al.* (1995) determined that ciliates are best used as indicator organisms for seven different physico-chemical parameters when the population density in a sample is high. The parameters used in the study were: effluent and settled sewage BOD₅, retention time in aeration tank, organic loading rate, mixed liquor volatile suspended solids (MLVSS), mean cell retention time (MCRT) and dissolved oxygen (DO) in the aeration tank. They concluded that an increase in the abundance of ciliate species reflects the optimal range conditions for growth of that species. This allows for the assessment of the quality of effluent.

On start-up of a new plant, there is a distinct species succession with time. By recycling all sludge in the initial phases (large solids retention time), the resident population can be established. At first the nutrient supply is abundant and the bacteria that are present are not capable of fully processing this. Large numbers of flagellates and amoebae develop. Later, free-swimming ciliates appear and the numbers of flagellates and amoebae decline. With the increase in biomass, the loading levels decrease and so does the growth rate of the free-living ciliates due to nutrient deficiency. As the solids retention time (SRT) is decreased, many species are washed out. This is when the crawling and sessile ciliates appear, as they are not vulnerable in this way. Metazoa are established when the plant has been in operation for sufficient time. This gives an indication of the different conditions needed for the growth of the protozoa and metazoa. For example, at moderate loading levels many ciliates, testate amoebae and some metazoa are usually present. If the DO levels are too low, there is a decline in the removal of COD and this favours the growth of flagellates and amoebae. The presence of toxins results in the disappearance of the protozoan and metazoan population, followed by an explosion in the numbers of protozoa after a few days. This is because of the large number of free-living bacteria available for consumption after the absence of their predators (Eikelboom, 2000).

The higher bacterial prey densities found in high loaded plants favours the growth of flagellates, amoebae and small, free-swimming ciliates - these three groups all exhibit inefficient chase and capture feeding mechanisms. Attached ciliates, rotifers and other invertebrates are selected at lower loading levels - they are attached to the flocs and feed by more efficient mechanisms. A balance between free-swimming and attached ciliates and rotifers is likely to be encountered in sludge when AS performance is good. Generally speaking, an overabundance of flagellates, amoebae or free-swimming ciliates indicates high F/M. Conversely, an overabundance of attached ciliates, rotifers and other life forms indicates low load. Low DO, high temperatures, unfavourable pH (<6.6 or > 8.5), as well as toxicity can lead to floc stress with resultant release of numerous bacteria as a food source. Blooms of flagellates and/or small free-swimming ciliates ensue Jenkins *et al.* (2004).

2.5 SCUM FORMATION

Eikelboom (2000), has observed that filamentous scum reaches a peak in winter months in The Netherlands and that there is a direct correlation with filament index of the scum. By comparing the population of filaments found in the mixed liquor and scum of problem WWTP's, he deduced that *M. parvicella*, actinomycetes and *N. limicola* (to a lesser extent), float selectively. These organisms have Gram positive and thus hydrophobic cell walls. Together with their morphology of flexible, bowed filaments, their hydrophobicity confers a predilection for forming networks over the hydrophobic surfaces of gas bubbles. Both *M. parvicella* and the actinomycetes compete for the hydrophobic fat fraction of the wastewater, with higher temperature and sludge load favouring the latter. Most of the other Gram-positive filaments found in low loaded plants are straight and have sheaths, so the hydrophobic cell wall is not in contact with the mixed liquor directly. When scum formation is not excessive, control measures such as skimming, removal and destruction of scum or spraying effluent on the scum can be employed. However, if scum formation is excessive, controlling the growth of the responsible filamentous organisms is necessary (Eikelboom, 2000).

Wastewater with high concentrations of oil or grease may lead to a high population of actinomycetes, while there is anecdotal evidence that wastewaters such as those found

from paper and pulp processing with no oil or grease rarely harbour these organisms (Jenkins *et al.*, 2004).

Under anoxic and anaerobic conditions, *M. parvicella* can take up and store long chain fatty acids, possibly because of the presence of surface-associated lipases. It is thought that when no oxygen is present, hydrolysis rates of particulate organics by the sludge population is decreased. Under these circumstances, the floc-formers are unable to metabolize lipid substrates, leading to a competitive advantage for *M. parvicella* (Jenkins *et al.*, 2004).

*Additional literature is incorporated into the body of the text, where discussion on a particular feature of the study takes place. There is no duplication of this content in this chapter (chapter 2). This was due to the wide-ranging content of the study and to obviate the need for constant cross-referencing between the body of the text and chapter 2.

CHAPTER 3 METHODS

3.1 SAMPLE COLLECTION

This task is ordinarily performed on a weekly basis by the operational staff at each of the WWTP's of the City of Cape Town. Grab samples are taken in plastic bottles from the weir overflow site exiting the bioreactor. The site of collection is clearly marked at each WWTP. Samples are transported to the Scientific Services Department of the City of Cape Town and routinely analyzed on the day of collection. During the study period, the microscopic analysis was conducted on the same day as sample collection in most circumstances. On the rare occasion when this was not possible, the delay was never greater than 24 hours post collection.

3.2 OPERATIONAL DATA

The staff employed by the Scientific Services Department of the City of Cape Town conducted all of the tests listed in this section.

- COD
- VSS
- Settleable solids
- SVI
- DSVI
- Conductivity

According to the methods described by Clesceri *et al.* (2005)

Seven of the parameters were conducted using an automated system. A Lachat flow injection analysis (FIA) analyzer was employed to quantitate the amounts of ions present according to QuickChem methods. The parameters are given below together with the relevant QuickChem method number:

- | | |
|---------------------|---------------|
| • TKN | 10-107-06-2-D |
| • Ammonia | 10-107-06-1-A |
| • Nitrates/nitrites | 10-107-04-1-A |
| • Total Phosphates | 10-115-01-1-E |
| • Ortho-phosphates | 10-115-01-1-A |
| • Chloride | 10-117-07-1-A |
| • Alkalinity | 10-303-31-1-A |

3.3 MICROSCOPIC ANALYSIS

The microscopic analysis was performed on samples without prior knowledge of the parameters given in section 3.2 so as to not inadvertently skew any results. The methodology given by Eikelboom, (2000) in his manual and accompanying CD was used as the basis for the analysis. Wherever possible, comparisons were made with micrographs from the manual and accompanying CD of Eikelboom (2000), supplemented with the text and micrographs from the manual of Jenkins *et al.* (2004). All parameters were recorded on a separate chart for each mixed liquor sample.

3.3.1 Wet mounts

Three wet mounts were prepared from each mixed liquor sample. The edges of the coverslips were sealed with Vaseline to prevent desiccation during examination. The slides were examined using 10X, 20X and 40X phase contrast objective lenses to give final magnifications of 100X, 200X and 400X respectively. If there was a need to observe further detail, a 100X lens was employed using oil immersion to give a final magnification of 1000X. The wet mounts were used to determine:

3.3.1 (a) Morphological characteristics of the flocs

- Size (diameter): small <25µm, medium 25-250µm, large >250µm
- Shape: round or irregular
- Structure: compact or open
- Strength: firm or weak

3.3.1 (b) Filament index

The filament index is a semi-quantitative assessment of the number of filaments between the flocs. Sample micrographs of sludges with different indices are located on the CD accompanying the manual by Eikelboom, (2000) as comparative measures. These were used extensively in order to maintain consistency of results throughout the study. Filament index was classed on a scale of 0-5, incorporating half measures where appropriate (for example 1.5).

3.3.1 (c) Filament characteristics

Certain characteristics pertaining to the filaments were noted, including motility, the presence or absence of sheaths, cell shape, the presence or absence of septa, filament shape, filament diameter and attached growth. At times, some of these characteristics were better visualized on the stained preparations. This was especially apparent when different filaments shared similar features. Thus, the unstained and stained slides were used in conjunction with one another to determine the characteristics of the filaments necessary for reliable identification.

3.3.1 (d) Diversity

The diversity of the bacterial population was noted as being low (1), moderate (2) or high (3).

3.3.1 (e) Monocolonies

The presence of monocolonies was quantified as absent (0) or present in either scanty (1), moderate (2) or numerous (3) amounts.

3.3.1 (f) Free-living cells, spirochaetes and spirils

The presence of free-living cells, spirochaetes and spirils was quantified as absent (0) or present in either scanty (1), moderate (2) or numerous (3) amounts.

Initially a number of wet mounts were prepared simultaneously, sealed and then examined. Midway into the study, this was changed as it was noted that numbers of free-living cells and spirochaetes appeared to be greater in slides that had been left standing for longer periods. At this point, it was decided to prepare the wet mounts and examine immediately for free-living cells and spirochaetes. After 10 minutes, the mounts were re-examined. This was referred to as "comparative timed analysis" and the results are divided into those obtained before and after the introduction of this "comparative timed analysis".

3.3.1 (g) Identification of protozoa and metazoa

These organisms were identified exclusively from the text of Eikelboom's (2000) manual and accompanying CD. Due to the lack of any prior experience, no attempt to speciate within any of the genera was made. When there was any uncertainty, the microbes were merely classed as unidentified protozoa, unidentified free-living ciliates or unidentified crawling ciliates. There were no unidentified microbes in any of the other categories. Numbers were enumerated semi-quantitatively as scanty, moderate or numerous.

3.3.2 Sulphur storage test

This was performed on all mixed liquor samples according to the method described in the manual of Eikelboom, (2000).

3.3.3 Gram stain

One slide was prepared from each of the undiluted mixed liquor samples and stained according to the method reference number BSOP TP 39i1 described in the manual of the Standard Operating Procedures of the Health Protection Agency (HPA) of the United Kingdom. The initial stain employed was 0.5% crystal violet. The decolouriser used was acetone and the counterstain was diluted carbol fuschin. The choice of Gram stain was due to the personal preference and experience of the microscopist.

(source: <http://www.evaluations-standards.org.uk>.)

The slides were examined at 1000X magnification under oil-immersion. The staining characteristics as well as the apparent morphological characteristics of the filaments were noted. The latter were compared to the results observed in the wet mounts, after staining with the Neisser technique and after the sulphur storage test, to give a holistic picture of each filament type.

3.3.4 Neisser stain

One slide was prepared from each of the undiluted mixed liquor samples and stained according to the method described in the manual of Eikelboom, (2000).

The slides were examined at 1000X magnification under an oil-immersion lens. The staining characteristics as well as the apparent morphological characteristics of the filaments were noted. The latter were compared to the results observed in the wet mounts, after staining with the Gram technique and after the sulphur storage test, to give a holistic picture of each filament type.

3.3.5 Calculations

3.3.5 (a) Sludge age

This value is a measure of the age of the microbial population inherent in the sludge.

$$\frac{\text{Reactor volume (m}^3\text{)} \times \text{mixed liquor volatile suspended solids (MLVSS) (g/L)}}{\text{Sludge wasted (kg/day)}}$$

3.3.5 (b) F/M ratio

This value is a measure of the amount of nutrients supplied to the biomass within the reactor.

$$\frac{\text{Influent flow rate into bioreactor (ML/day)} \times \text{Chemical oxygen demand (COD) (mgCOD/L)}}{\text{Bioreactor volume (ML)} \times \text{MLVSS (mg/L)}}$$

4.1 ATHLONE WWTP

4.1.1 PLANT CONFIGURATION AND OPERATING PARAMETERS

Athlone WWTP is a nitrification-denitrification biological excess phosphorous removal (NDBEPR) plant with a design capacity to treat approximately 120ML effluent per day. The plant houses six identical bioreactors named from A to F, which operate in a University of Cape Town (UCT) configuration with diffuse aeration. The raw influent consists of approximately 70% industrial effluent and 30% domestic effluent. Influent to the primary settling tank (PST) consists of raw influent as well as the liquid component from the digesters and thickeners. The plant operates at a dissolved oxygen (DO) concentration of 2.5 to 3mg/L, anaerobic mass fraction of 15%, anoxic mass fraction of 29% and aerobic mass fraction of 56%.

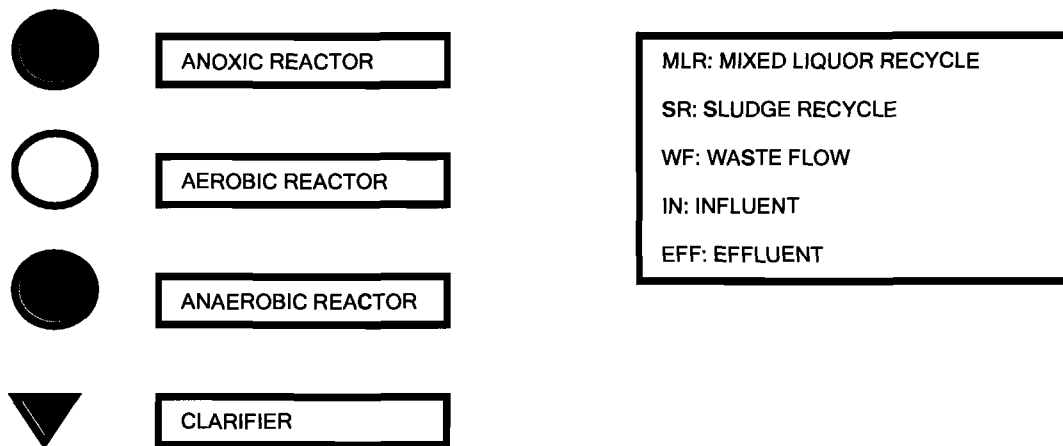


Figure 4.1.1 Key for WWTP configurations

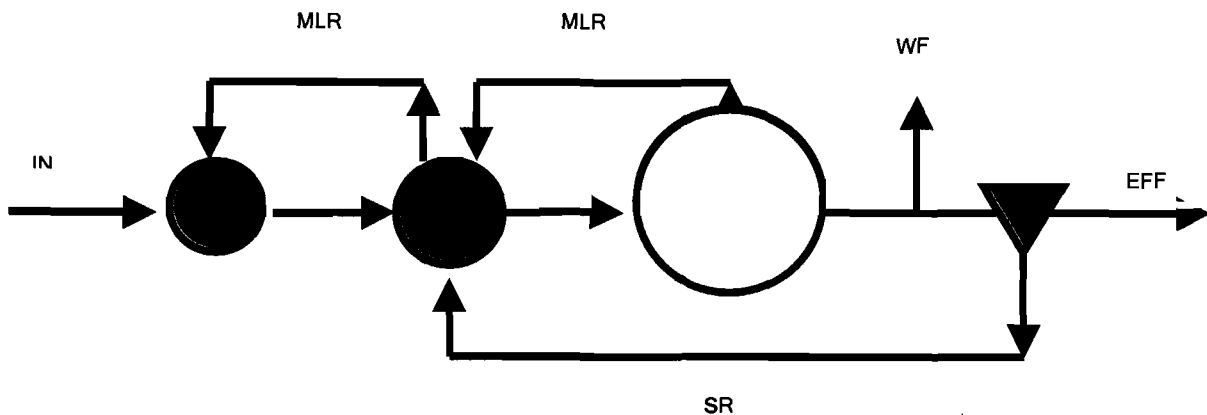


Figure 4.1.2 UCT process configuration (Metcalf and Eddy, 1991)

4.1.2 RESULTS

4.1.2 (a) Operational data

The influent (I) that flows into the reactors A-F is supernatant from the PST as well as the liquid component from the digesters and thickeners.

The mixed liquor (ML) from reactor A flows into the appropriate clarifier.

The effluent (E) is the clarifier supernatant.

*Note: breaks in the graph lines occur when no data was available

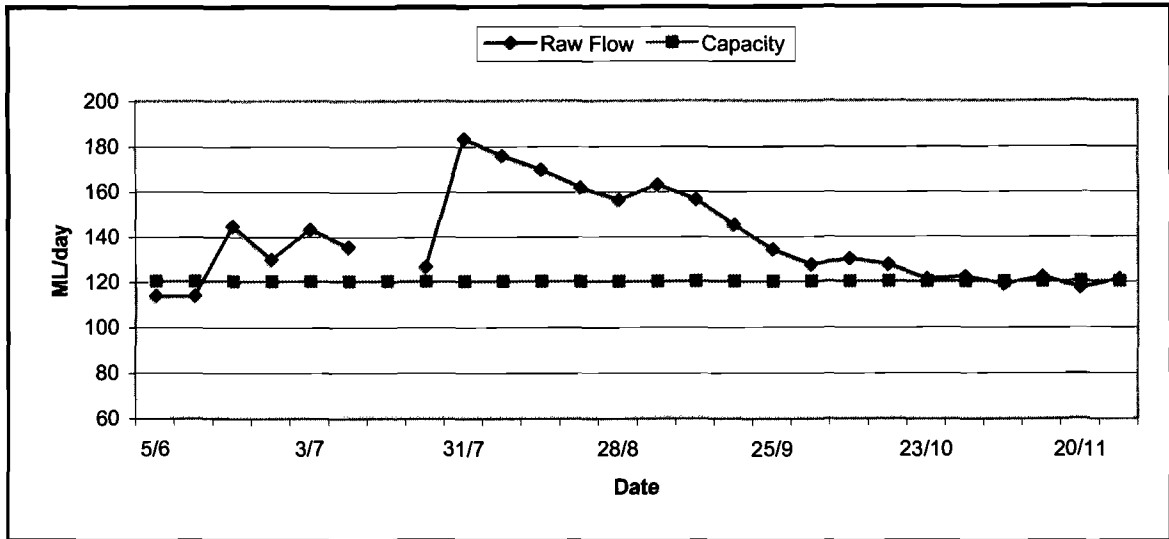
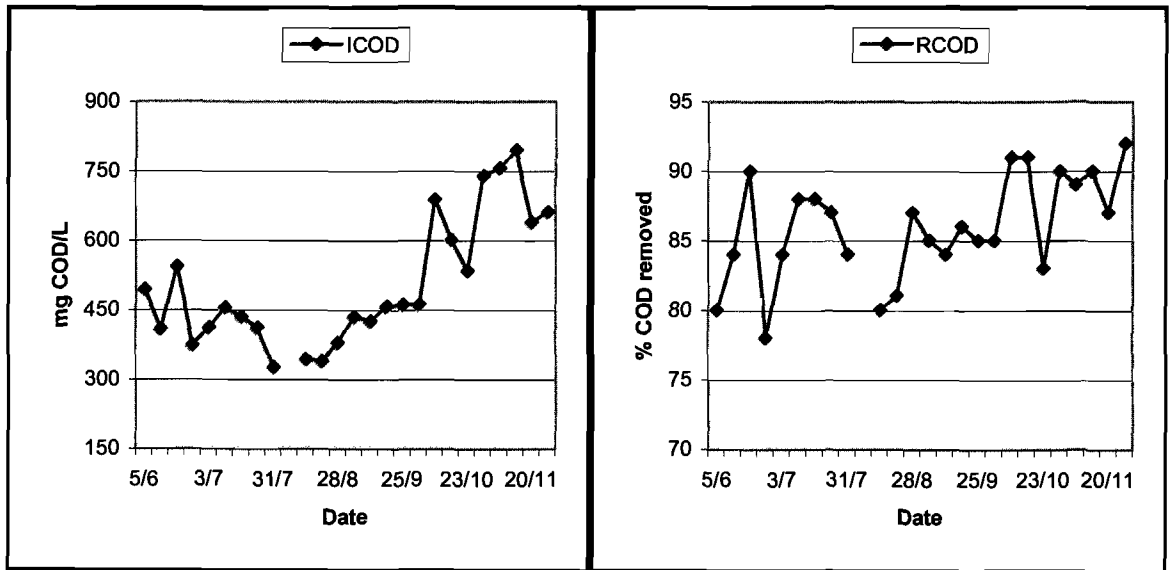


Figure 4.1.3 Graph depicting the raw flow into Athlone WWTP and the design capacity of the WWTP (2007).



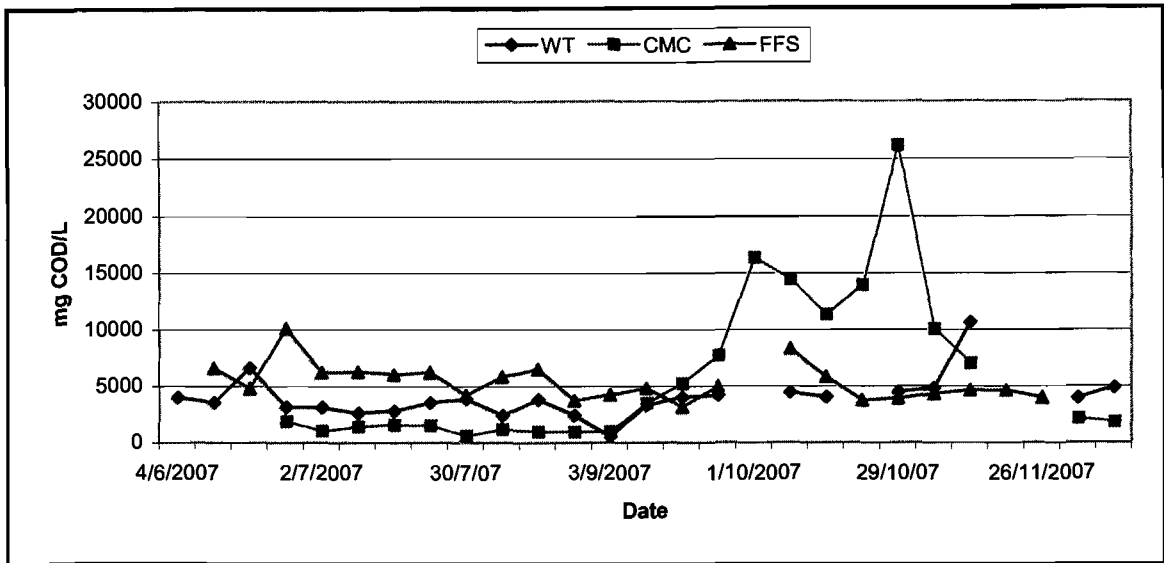


Figure 4.1.6 Graph depicting the chemical oxygen demand (COD) of the leachate from Wastetech (WT), Cape Metropolitan Council (CMC) and Vissershoek (FFS) processed at the Athlone WWTP during the study period

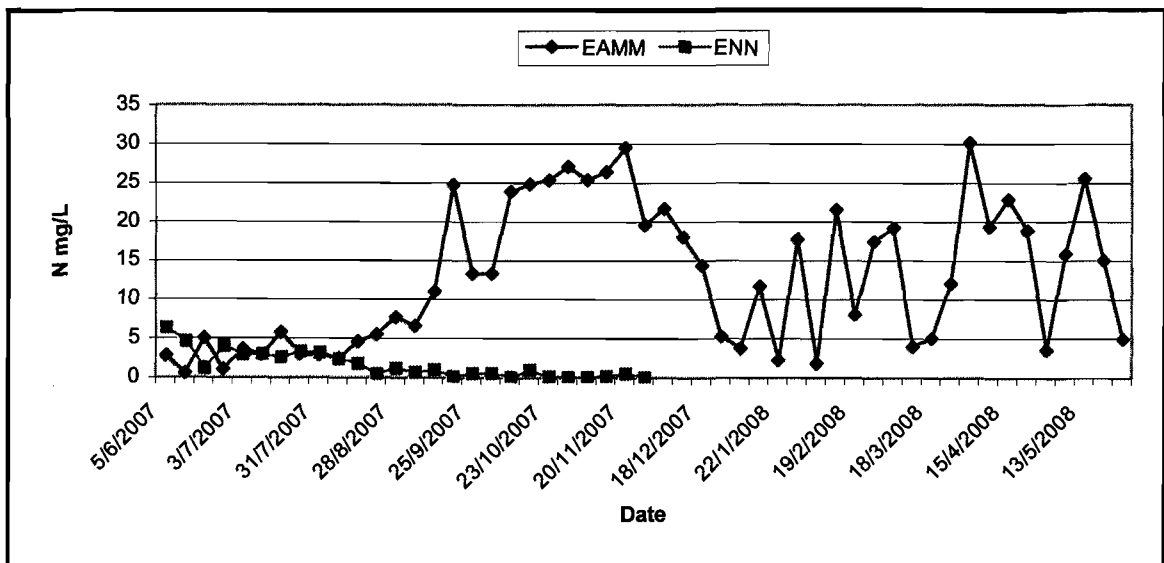


Figure 4.1.7 Graph depicting the effluent ammonia levels (EAMM) from June 2007 to June 2008 and effluent nitrate and nitrite levels (ENN) from June to November 2007 at Athlone WWTP

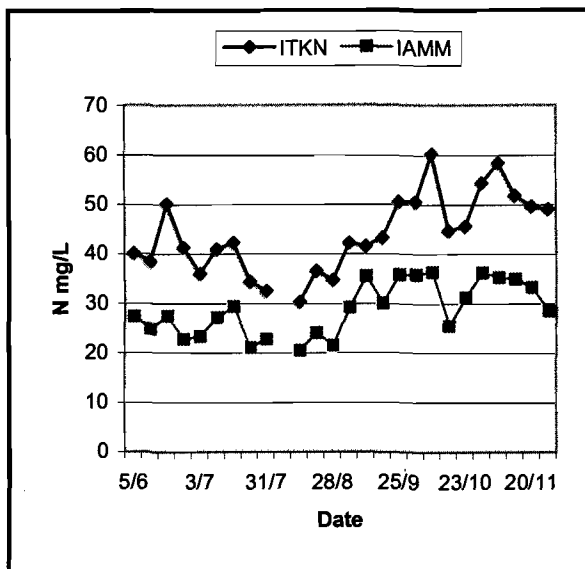


Figure 4.1.8 Graph depicting the influent levels of Total Kjeldahl nitrogen (ITKN) and ammonia (IAMM) at Athlone WWTP in 2007

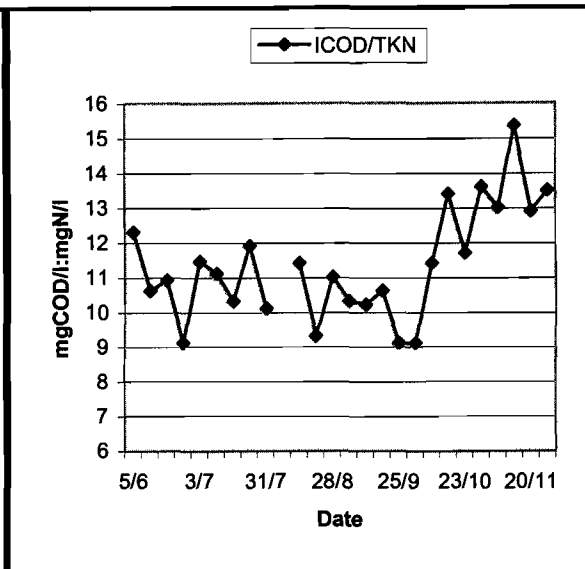


Figure 4.1.9 Graph depicting the ratio of the influent COD (ICOD) to Total Kjeldahl nitrogen (TKN) at Athlone WWTP in 2007

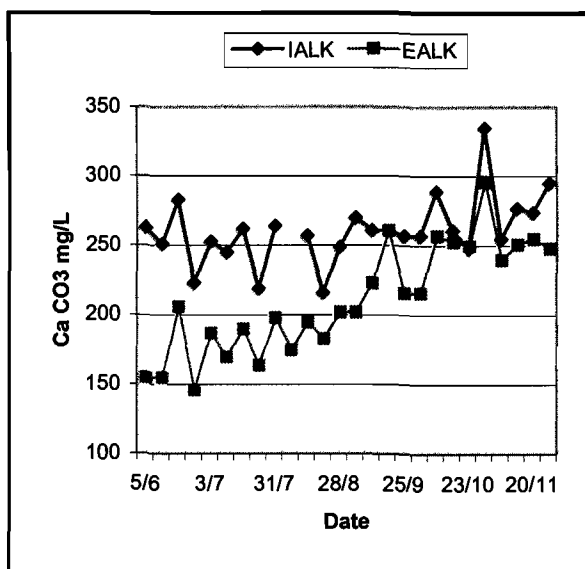


Figure 4.1.10 Graph depicting the influent alkalinity levels (IALK) and the effluent alkalinity levels (EALK) at Athlone WWTP in 2007

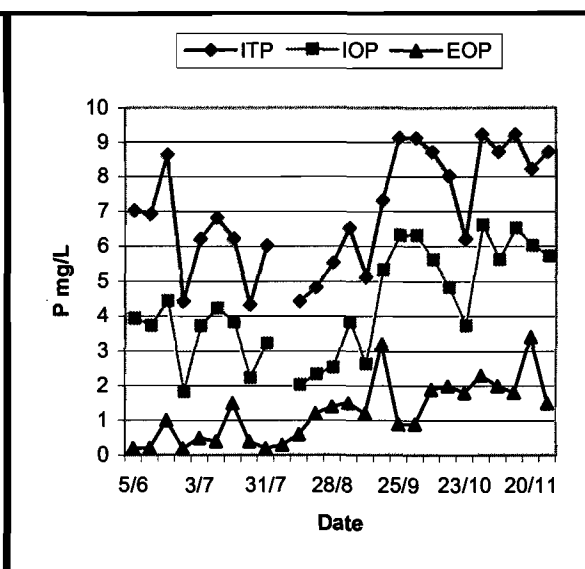


Figure 4.1.11 Graph depicting the influent total phosphate (ITP), influent sigma-phosphate (IOP) and effluent sigma-phosphate levels at Athlone WWTP in 2007

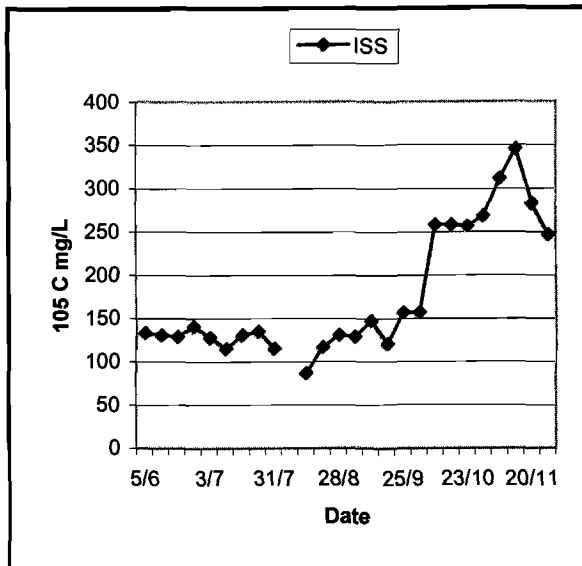


Figure 4.1.12 Graph depicting the influent levels of suspended solids (ISS) at Athlone WWTP in 2007

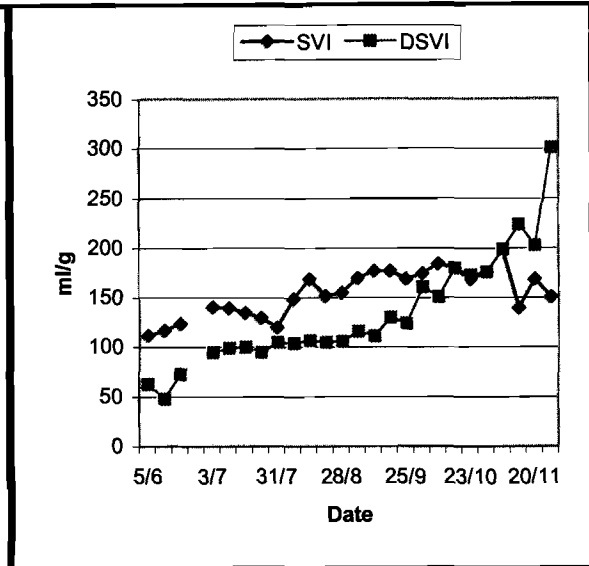


Figure 4.1.13 Graph depicting the rising sludge volume index (SVI) and dissolved sludge volume index (DSVI) in bioreactor A of Athlone WWTP in 2007

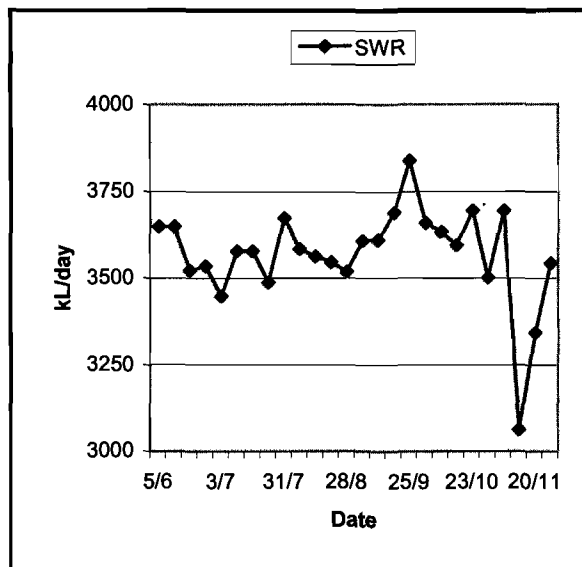


Figure 4.1.14 Graph depicting the sludge wasting rate (SWR) from the bioreactors at Athlone WWTP in 2007

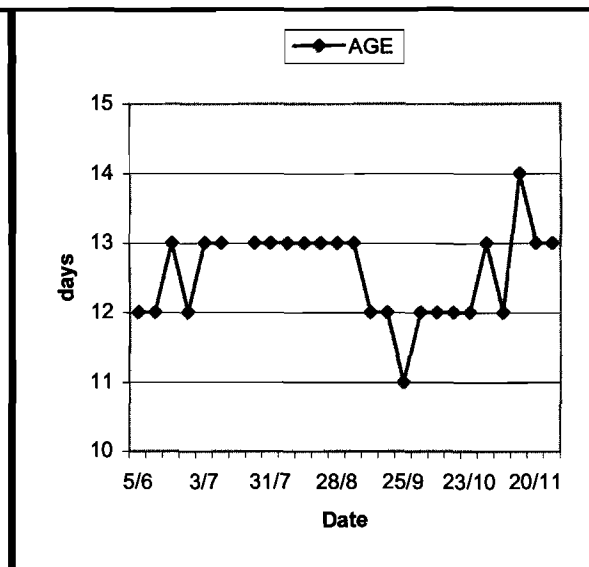


Figure 4.1.15 Graph depicting the calculated sludge age of bioreactor A at Athlone WWTP in 2007

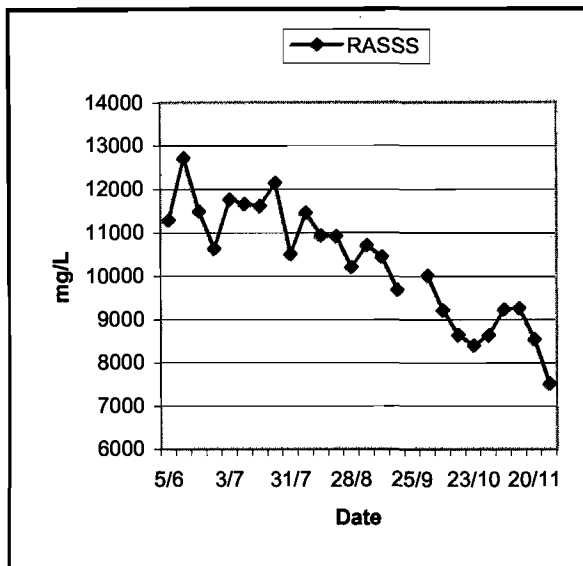


Figure 4.1.16 Graph depicting the decreasing amount of suspended solids (SS) in the return activated sludge (RAS) at Athlone WWTP in 2007

| Time after start of test (min) | Ave OUR (mg O ₂ /hr) |
|--------------------------------|---------------------------------|
| 0:15 - 0:30 | 57 |
| 0:30 - 1:00 | 39 |
| 1:00 - 1:30 | 27 |
| 1:30 - 2:00 | 22 |
| 2:00 - 2:30 | 19 |
| 2:30 - 3:00 | 16 |
| 3:00 - 3:30 | 15 |

The OUR was measured as the amount of oxygen consumed per liter of composite sample in one hour (mg O₂/hr). The composite sample consisted of equal parts of mixed liquor and raw influent.

| Parameter | Units | 25/10/2007 | 26/10/2007 | 27/10/2007 | 28/10/2007 |
|-----------|--------|------------|------------|------------|------------|
| Aluminium | ugAl/l | 2090 | 1910 | 1760 | 811 |
| Arsenic | ugAs/l | 2.0 | 2.0 | <1.0 | 3.2 |
| Cadmium | ugCd/l | 1.0 | <1.0 | <1.0 | <1.0 |
| Chromium | ugCr/l | 104 | 40 | 24 | 16 |
| Copper | ugCu/l | 144 | 78 | 71 | 48 |
| Iron | ugFe/l | 1800 | 1370 | 1400 | 1060 |
| Manganese | ugMn/l | 77 | 74 | 72 | 53 |
| Nickel | unNi/l | 48 | 22 | 14 | 11 |
| Lead | ugPb/l | 20 | 7.6 | 8.6 | 11 |
| Zinc | ugZn/l | 405 | 249 | 221 | 210 |

| TABLE 4.1.3 PHENOL INDEX ON RAW INFLUENT AND LEACHATE | |
|--------------------------------------------------------------|----------------------------|
| Sample | Phenol Index (mg/l) |
| Raw Influent | |
| 25/10/2007 | 0.33 |
| 26/10/2007 | 0.28 |
| 27/10/2007 | 0.32 |
| Vissershoek leachate | |
| 22/10/2007 | 30.97 |
| 29/10/2007 | 27.40 |
| Cape Metropolitan Council leachate | |
| 22/10/2007 | 12.92 |
| 29/10/2007 | 8.13 |
| Wastetech leachate | |
| 29/10/2007 | 23.59 |

| TABLE 4.1.4 FOOD TO MICROORGANISM (F/M) RATIO IN REACTOR A OF ATHLONE WWTP IN 2007 | |
|-----------------------------------------------------------------------------------------------|----------------------|
| June | 0.22 mgCOD/mgVSS.day |
| July | 0.22 mgCOD/mgVSS.day |
| August | 0.26 mgCOD/mgVSS.day |
| September | 0.32 mgCOD/mgVSS.day |
| October | 0.26 mgCOD/mgVSS.day |
| November | 0.41 mgCOD/mgVSS.day |

4.1.2 (b) Microscopic sludge analysis

This was performed monthly on the following dates: 26/6/07, 23/7/07, 28/8/07, 24/9/07, 22/10/07 and 26/11/07.

● Floc structure

In June, July and August, the flocs were round, compact, firm and medium in size, conforming to Eikelboom (2000) floc type 2. In September, the flocs were more open with bridging and filaments protruding from the flocs and between flocs. Many gonidia and rosettes were apparent. In October and November, there was an abundance of filamentous organisms both bridging the flocs and free between the flocs. Many gonidia and rosettes were once again present. Filamentous growth was so abundant that it was not possible to discern the floc shape and structure accurately, although the flocs appeared as small and round and joined by filamentous growth.

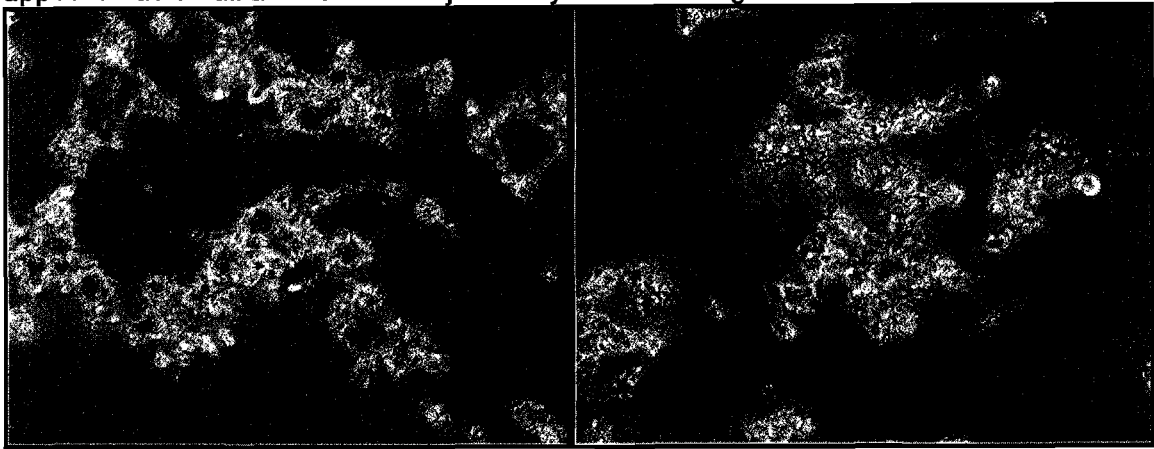


Figure 4.1.17: Micrographs of wet mounts of mixed liquor from Athlone reactor A showing the deterioration in floc character from June 2007 (left) to November 2007 (right). The flocs are smaller in November with more inter-floc bridging

● Diversity

In October, there was low diversity in both the prokaryotic and eukaryotic organisms present, with overwhelming numbers of *Aspidisca* spp. present and very scanty numbers of other protozoa/metazoa.

● Filament index

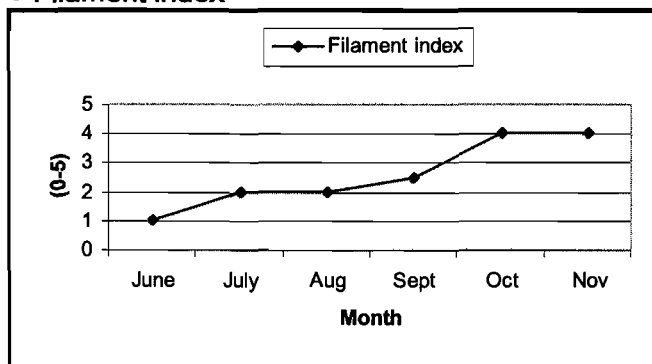


Figure 4.1.18 Graph showing the increasing filament index from June to November 2007

| TABLE 4.1.5 DOMINANT AND SECONDARY FILAMENTOUS ORGANISMS IDENTIFIED IN THE MIXED LIQUOR OF ATHLONE WWTP REACTOR A IN 2007 | | | | | | | | |
|---------------------------------------------------------------------------------------------------------------------------|------|------|-----|------|-----|-----|-------------------------|-------------------------|
| AS DOMINANT FILAMENTS | | | | | | | | |
| | June | July | Aug | Sept | Oct | Nov | % Prevalence (dominant) | % Prevalence (overall) |
| Type 0092 | X | X | X | X | | | 66 | 100 |
| Type 021N | | | | X | X | X | 50 | 83 |
| Type 0041 | X | | | | | | 17 | 66 |
| Actinomycetes | X | | | | | | 17 | 50 |
| <i>N. limicola</i> III | | | | | | X | 17 | 50 |
| AS SECONDARY FILAMENTS | | | | | | | | |
| Type 0092 | | | | | X | X | } | Also dominant filaments |
| Type 021N | | X | X | | | | | |
| Type 0041 | | X | X | X | | | | |
| Actinomycetes | | X | | | | X | | |
| <i>N. limicola</i> III | | | | X | X | | | |
| <i>M. parvicella</i> | | X | X | X | X | X | 0 | 83 |
| Type 1851 | | | X | X | X | | 0 | 50 |
| <i>Thiothrix</i> spp. | | X | X | | | | 0 | 33 |
| <i>Flexibacter</i> spp. | X | | | | | | 0 | 17 |
| <i>H. hydrosis</i> | | | X | | | | 0 | 17 |
| Type 0914 | | | X | | | | 0 | 17 |

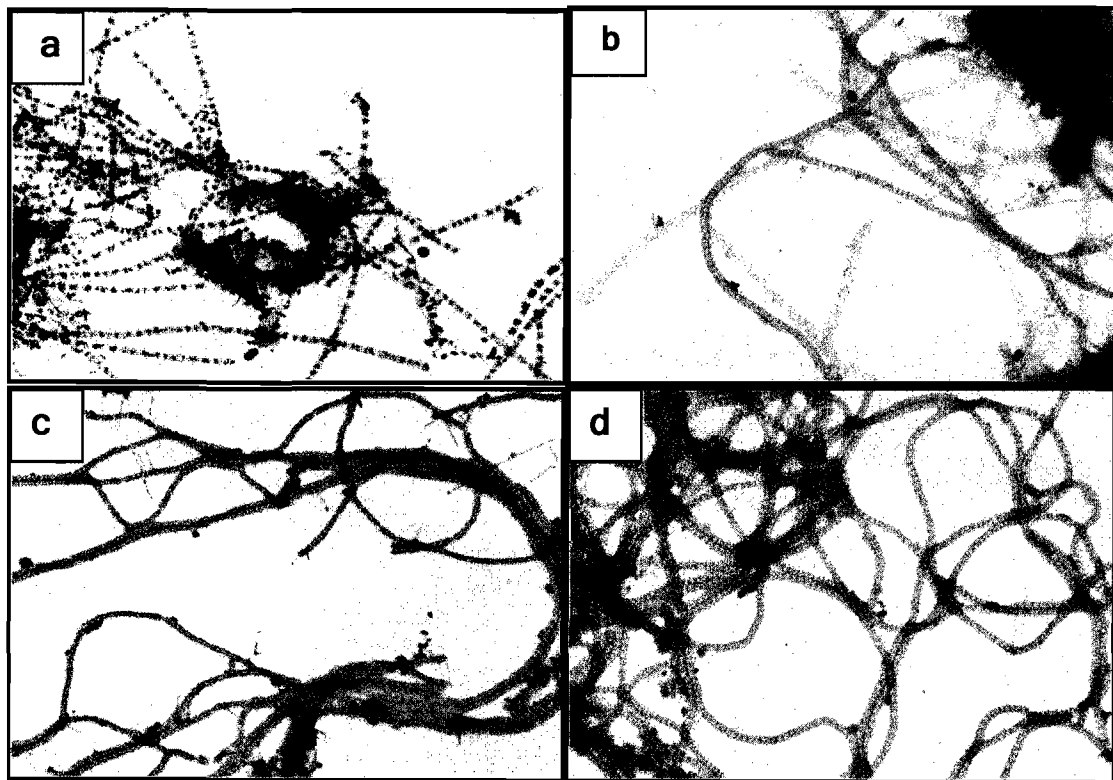


Figure 4.1.19 Micrographs of various filamentous prokaryotic microorganisms detected in mixed liquor from Athlone WWTP in November 2007

(a) Gram stain of type 021N

(b) Neisser stain of *N. limicola* III and Type 021N

(c) Gram stain of *N. limicola* III

(d) Neisser stain of *N. limicola* III

TABLE 4.1.6 PROTOZOA/METAZOA IDENTIFIED IN THE MIXED LIQUOR OF ATHLONE WWTP REACTOR A IN 2007

| | | SCANTY | | | | | |
|----------------------|-----------------------------|----------|------|-----|------|-----|-----|
| | | June | July | Aug | Sept | Oct | Nov |
| Sessile ciliates | <i>Carchesium</i> spp. | | X | | | | |
| | <i>Epistylis</i> spp. | | | X | | | X |
| | Suctorean | X | | | | | |
| | <i>Vorticella</i> spp. | | | X | X | X | |
| Crawling ciliates | <i>Aspidisca</i> spp. | X | X | X | | | X |
| | <i>Chilodonella</i> spp. | | X | | | | |
| | <i>Trachelophyllum</i> spp. | | | X | X | X | |
| Free-living ciliates | <i>Colpidium</i> spp. | | | | | | X |
| | <i>Euplotes</i> spp. | | X | | | X | |
| | <i>Litonotus</i> spp. | | X | X | | | |
| | unidentified | | | X | | | |
| Flagellates | <i>Bodo</i> spp. | X | | X | X | X | |
| | <i>Peranema</i> spp. | X | X | X | | X | |
| | <i>Monsiga</i> spp. | | | | | X | |
| | <i>Trepomonas</i> spp. | X | | | | | |
| Amoebae | | | | | | | |
| Heliozoa | | | | | | | |
| Rotifers | | | | | | | |
| Nematodes | | | | | | | |
| | | MODERATE | | | | | |
| Sessile ciliates | <i>Vorticella</i> spp. | X | | | | | |
| Crawling ciliates | <i>Aspidisca</i> spp. | | | | X | | |
| Flagellates | <i>Peranema</i> spp. | | | | X | | |
| | | NUMEROUS | | | | | |
| Crawling ciliates | <i>Aspidisca</i> spp. | | | | | X | |

4.1.3 DISCUSSION

Figures 4.1.3 to 4.1.6 and 4.1.12 show that Athlone WWTP was overloaded for almost the entire winter of 2007 and that although there was an increase in both chemical oxygen demand (COD) and suspended solids (SS) in September, October and November, the COD removal remained relatively efficient throughout the trial period. The increased COD corresponded to a high COD in the leachate that originated from the Cape Metropolitan Council source, but this may be circumstantial. Table 4.1.4 gives the figures for the food to microorganism ratio (F/M), which also show an increase in September and November. The return activated sludge suspended solids (RASS) levels decreased (figure 4.1.16).

Regarding nutrient removal, the removal of phosphorous was satisfactory (figure 4.1.11), but the plant experienced problems with the removal of nitrogenous waste. Towards the end of September, the performance of the plant deteriorated considerably. This was chiefly reflected by the substantial increase in effluent ammonia levels, despite only a small increase in influent ammonia and TKN. These results are reflected in figures 4.1.7 and 4.1.8. The effluent nitrate and nitrite levels decreased to zero when nitrification was poor.

At the same time, the dissolved sludge volume index (DSVI) and filament index (FI) values increased to those predictive of bulking conditions as depicted by figures 4.1.13 and 4.1.18. In addition, there was a considerable change in the protozoan and metazoan community. In September, there was a sharp decrease in the diversity and quantity of these organisms, such as would be encountered with a toxic event. In October, there was still a lack of diversity, but there were overwhelming numbers of *Aspidisca* spp. with numerically few other protozoa and metazoa. By November, *Aspidisca* spp. and *Peranema* spp. were both moderately abundant. There was a slight recovery in the numbers of other protozoa and metazoa. Although only semi-quantitative results are given in the results section, special notes were made on the results sheet regarding the protozoan and metazoan communities at Athlone WWTP. This was due to the striking difference in the diversity and abundance in comparison with the other study samples.

Sludge was imported from another WWTP in December, and nitrification performance improved. Unfortunately, it was not possible to obtain worthwhile information on this retrospectively. The nitrification results as given by effluent ammonia levels for the study period plus a further six months have been given in figure 4.1.7. Here it can be seen that after the initial improvement, the situation once again deteriorated. The pattern at this stage was somewhat different with nitrification being more erratic, but never entirely satisfactory. Over the problem period in 2007, the aerators experienced some periods with mechanical failure, but the exact dates could not be established.

There are a number of likely explanations for the sudden loss of nitrification - insufficient DO in the aerobic zone or an influx of toxic substances being the most obvious according to the results of the OUR testing and the loss of diversity. These are expanded on and discussed in the following pages as far as possible with the data available. Causes such as the increased influent COD to Total Kjeldahl nitrogen (ICOD/TKN) ratio (shown in graph 4.1.9), increased F/M ratio (table 4.1.4), septic conditions, decreased DO, sludge age (figure 4.1.15) and possible nitrifier washout are also taken into account as contributing factors. It was unfortunately not possible to obtain the operational DO results for the study period, which would have made the ensuing discussion less speculative.

The reasons for postulating that toxicity and not lack of DO was the determining factor are:

- Species of the genera *Nitrosomonas* and *Nitrobacter* are the main organisms responsible for the conversion of ammonia to nitrite and nitrite to nitrate respectively. The nitrifiers are sensitive to a variety of inhibitors, temperature, pH and oxygen concentration. Re-establishment of an effective nitrifier population may take weeks after an adverse event due to their slow specific growth rate. During nitrification, a large amount of alkalinity is consumed (Metcalf and Eddy 1991). Figure 4.1.10 shows that despite a relatively stable influent alkalinity, there is a rise in effluent levels during the problem period, coinciding with the loss of nitrification.
- The nitrifying population is more sensitive to toxins than the heterotrophs and the first step in the nitrification sequence shows the most sensitivity (Gerhaey *et al.*, 1997). Many decades back, Tomlinson *et al.* (1966) tested a wide range of substances for toxicity to nitrifiers. By 1977, this was still considered to be the most complete analysis of nitrification toxicity (Hockenbury, 1977). A literature search in 2008 did not reveal anything more complete. The chemicals analysed by Tomlinson *et al.* (1966) included those likely to be produced by industry and thus found in WWTP's treating a component of industrial effluent. For instance, a wide range of sulphur compounds that were used in the vulcanization of rubber were tested and found to be toxic. The concentration of phenol found to reduce ammonium oxidation by 75% was determined at 5.6 mg/L. By referring to table 4.1.3, it can be seen that the leachate phenol concentrations were substantially higher than this value before dilution. Heavy metals have also been implicated in nitrifier toxicity in wastewater, but the concentrations of cadmium, chromium, lead and zinc shown in table 4.1.2 that were found in the raw influent were not sufficient to cause loss of nitrification as determined by Madoni *et al.* (1999). Aluminium and lead levels appear high, but no literature could be found to assess the impact of these metals on the nitrifier population. Hockenbury *et al.* (1977) determined toxicity levels of a number of organic compounds by nitrifier toxicity testing. These researchers also found that ammonia exerted substrate inhibition on its own oxidation by *Nitrosomonas* spp. and that by increasing the levels of ammonia this increased the toxic effects of some organic compounds such as aniline. It is not conceivable to test for levels of every possible toxin, and the results given in table 4.1.2 and 4.1.3 and figure 4.1.6 are all that were available retrospectively. It is possible that the treatment of leachate at Athlone WWTP affected the nitrifying population, but this practice was abandoned at the end of 2007 and only resumed in June 2008. During this period, nitrification only improved slightly, albeit the pattern was more erratic than previously. The weekly leachate samples do differ from batch to batch, but limited information is routinely provided on the content of these. Athlone also has a high industrial waste component (70%), so the possibility of toxic substances from this source is also valid. Also, the influent to the PST's includes the liquid component from the digesters and thickeners that increases the likelihood of septic conditions being present (with resultant chemicals such as sulphides).
- Nitrification improved when sludge was imported and deteriorated again later even although at this stage the mechanical problems (and thus presumably any problems with DO levels) had been rectified. The results of the OUR tests conducted on the sludge from Athlone WWTP (shown in table 4.1.1) suggest that there were insufficient nitrifiers present. Oxygen utilization is a function of both the heterotrophic and the autotrophic nitrifying population, so this test is not

specific. The results of the OUR tests were interpreted by the laboratory (Scientific Services Wastewater laboratory) in a comment accompanying the test results:

“After an initial high, there was a rapid rate of decline of the OUR in a short period of time. This would seem to indicate that the biodegradable COD does get consumed, but there are insufficient nitrifiers present to convert most of the ammonia to nitrites and nitrates and hence much less oxygen is required after the biodegradable COD is depleted”.

To explain this more fully - the degradation of readily biodegradable COD (RBCOD) by the heterotrophic population is rapid in the presence of sufficient oxygen (Casey *et al.*, 1999b). This source of nutrition on its own is expended quickly resulting in an accompanying rapid drop in the OUR. If the nitrifier population was intact at this point, in the presence of ammonia, the OUR rate would be maintained at a moderate level due to the process of nitrification and the ammonia would be converted to nitrates and nitrites (Metcalf and Eddy, 1991). Thus, the interpretation that there were insufficient nitrifiers is plausible, in that the OUR rate declined precipitously and nitrification itself was grossly retarded for a NDBEPR WWTP. There are a number of specific tests designed to detect the loss of nitrification such as those described by Gerhaey *et al.* (1997). The test developed by these authors is based on the principle that the nitrification potential of known cultures of nitrifying organisms will be inhibited when subjected to sludge containing substances toxic to this population (measured against a control population). The results of this type of test would have been more conclusive in determining the cause of poor nitrification at Athlone WWTP, but was too specialized for the laboratory in question. Furthermore, the test has to be performed when the toxins are present in the sludge, which will not be the case if nitrifier loss is due to a single toxic event, unless monitoring is continuous. Such testing as a routine may be considered as an option in WWTP's that are known to experience problems with nitrification (either ongoing or intermittent).

- COD removal remained satisfactory throughout. The ICOD/TKN ratio shown in figure 4.1.9 increased at the beginning of the period when nitrification initially deteriorated. This, together with the concurrent increase in F/M may have exacerbated the condition. According to Metcalf *et al.* (1991), a BOD₅/TKN ratio of 1-3 would be expected in separate stage nitrification. The lower the ratio, the greater the fraction of nitrifiers. Most work on ratios of carbon to nitrogen has been performed at low temperatures, when nitrification is considerably reduced, or using BOD, not COD values. It is not possible to extrapolate BOD from COD values from a WWTP with such a high component of industrial effluent. In addition, the values given in figure 4.1.9 used influent COD values. With the UCT configuration, much COD removal takes place in the anoxic zone preceding the aerobic zone, so figures would realistically fall somewhere between the influent and effluent values. It is therefore difficult to assess whether the increase in COD played a role in the nitrification problem.
- The microorganism population is a good reflection of the prevailing conditions (Jenkins *et al.*, 2004). There was a loss of diversity amongst both the prokaryotic and eukaryotic organisms. Spirochaetes, spirils and filaments associated with low DO (Eikelboom, 2000), were not prevalent. Ciliates outnumbered flagellates, also pointing to lack of oxygen not being excessive (Eikelboom, 2000). Some *Aspidisca* species have been shown to be tolerant of high metal concentrations. (Abraham *et al.* 1998). *Nostocoida limicola* III appeared in September and was dominant by December. This organism is known to be strictly aerobic (Eikelboom, 2000). The lack of diversity can also be caused by lack of nutrients (Jenkins *et*

al., 2004). Besides COD, nitrites/nitrates and phosphates, no routine monitoring of other micronutrients or macronutrients is performed on the sludge from Athlone WWTP. However, due to the loss of nitrification, there were little nitrates and nitrites present in the latter months of the study. Type 021N, that was dominant in September, October and November is amongst other factors, associated with nitrogen deficiency (Jenkins *et al.*, 2004).

- The sludge wasting rate and sludge age shown in figures 4.1.14 and 4.1.15 respectively did not change substantially during the course of events, although there was a downward trend in the RASSS (figure 4.1.16). The latter may be due to the steady increase in the filamentous population that was associated with a high DSVI and thus poor settling. A substantial increase in the filaments would still allow COD removal to take place, and there may well have been a decrease in the ordinary heterotrophic population. It is possible that some washout of the nitrifying population contributed to the loss of nitrification, as the mean cell residence time (MCRT) needed for the growth of these organisms would increase under adverse conditions. Rittman and McCarty, (2001) calls for a solids retention time (SRT) of >15 days, with even longer times being implemented under conditions where toxins are present, or DO and/or temperature is low. However, the presence of rotifers and nematodes points to a long sludge age (Eikelboom, 2000).

In light of the above, the ongoing problems with nitrification were most likely caused initially by lack of DO and poisoning of the nitrifying population coupled with increased ICOD/TKN and F/M. After “recovery” and the cessation of leachate addition, there were again problems in 2008. However, at this stage periods of good nitrification punctuated the poor periods. If a decrease in the nitrifying population was to blame in 2008, the effluent ammonia levels would not show such repeated rapid recovery. Figure 4.1.7 shows that in 2008, nitrification performance varied constantly. This is in contrast to 2007, where performance was consistently poor for a protracted period. No mixed liquor samples were examined during 2008 (out of study period) and the cause of poor nitrification in 2008 may very well have been entirely different from that experienced in the latter stages of 2007.

In summary, on a practical level: it is not possible to continuously monitor influent for all possible toxins. Further microscopic analysis, in conjunction with detailed DO, ICOD/TKN and F/M ratios and ideally nitrifier toxin testing would help to pinpoint the problem. During the study period, plant-operating parameters in terms of the historical data and physical functioning of the WWTP's were unknown. This was purposeful, so as not to inadvertently skew microscopic results to fit known parameters (blind). Unfortunately, this led to the situation where the microscopic analysis could not reach its full potential as a troubleshooting aid. If microscopic analysis was part of the routine testing protocol, this situation could very well have been rectified. For example, when the change in the protozoan and metazoan population was noted, levels of possible toxins in the influent could have been determined in an endeavour to source the toxic waste and prevent future loss of nitrification.

Table 4.1.4 shows the results regarding the identification of the filamentous population: there was a distinct change in the dominant population after the onset of decreased nitrification. The true low F/M species of Type 0041 and Type 0092 were present in June, together with actinomycetes. Type 0092 continued to co-dominate until September, after which it became secondary. Type 021N became co-dominant in September and continued to dominate for the remainder of the study period, together with *N. limicola* III in November.

Type 021N is not restricted to low F/M environments and is associated with the presence of easily biodegradable compounds and/or sulphur compounds such as would occur under septic conditions (Eikelboom, 2000). Of the four organism types associated with the presence of hydrogen sulphide and/or the presence of easily biodegradable compounds and/or nutrient deficient conditions (Jenkins *et al.*, 2004), three were found as either dominant or secondary species in the Athlone plant, both before and after the “problem” period: *Thiothrix* spp. in July and August, Type 021N from July to November and Type 0914 in August. For the first three months of the study, the most likely reasons for the growth of these filaments are: the inherent composition of the industrial raw influent and/or septic influent either raw from the sewers or from the returned influent from the digesters and thickeners.

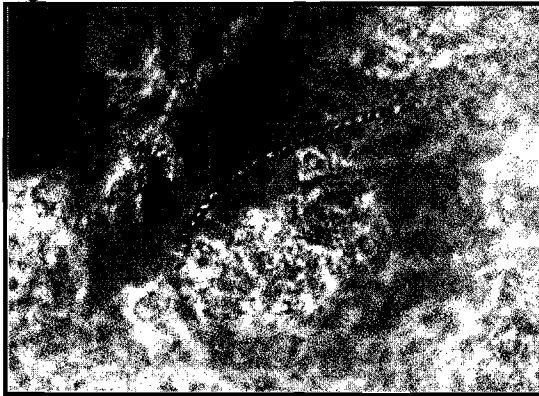


Figure 4.1.20 Wet mount of *Thiothrix* spp. from mixed liquor from bioreactor A at Athlone WWTP in July 2007 after application of the sulphur storage test

Nutrient deficiency would be unlikely to be associated with the growth of all of these species before the loss of nitrification potential led to a lack of nitrates and nitrites. Type 021N and *N. limicola* were the major dominant species at the stage when the DSVI was increasing. Neither of these organisms are associated with low DO (Jenkins *et al.*, 2004). Furthermore, the filaments that are associated with low DO by this author, namely *S. natans*, Type 1701 and *H. hydrosis*, were absent (except for one occasion when *H. hydrosis* was present as a secondary species). According to Jenkins *et al.* (2004), under nutrient deficient conditions, Type 021N forms gonidia and rosettes. These were seen abundantly in the mixed liquor samples from Athlone reactor A during the “problem” period. This lends credence to the hypothesis that due to impaired nitrification, there was a lack of nutrition in the form of nitrates and nitrites. This would be exacerbated with an increased influent COD. Both Jenkins *et al.* (2004) and Eikelboom (2000) have associated *N. limicola* with nutrient deficiency.

In terms of bacterial composition, it seems likely that the conditions encountered over the first three months of the project allowed the growth of a variety of mainly low F/M filaments prevalent in most of the Cape Town plants. At this stage competition between these filaments and the heterotrophic population did not allow filamentous overgrowth and bulking conditions were absent. Due to lack of nitrification over the latter three months, a nutrient deficiency (nitrates and nitrites) ensued. This allowed the filaments suited to these conditions to out-compete the heterotrophic population and bulking conditions arose. This is entirely different to the bulking hypothesis postulated by Casey *et al.* (1999), as in this case the bulking was probably caused by the absence and not the presence of nitrates and nitrites.

4.2 BELLVILLE WWTP

4.2.1 PLANT CONFIGURATION AND OPERATING PARAMETERS

The three industrial bioreactors (named North, Center and South), at Bellville have the capacity to treat approximately 60ML effluent per day. The configuration is the Modified Ludzack-Ettinger (MLE) and the method of aeration is diffuse. The influent consists of raw, unsettled influent of industrial origin that is supplemented by the primary sludge from the PST of the nearby domestic WWTP. The plant operates at a DO of 1-2 mg/L, an anoxic mass fraction of 20% and an aerobic mass fraction of 80%.

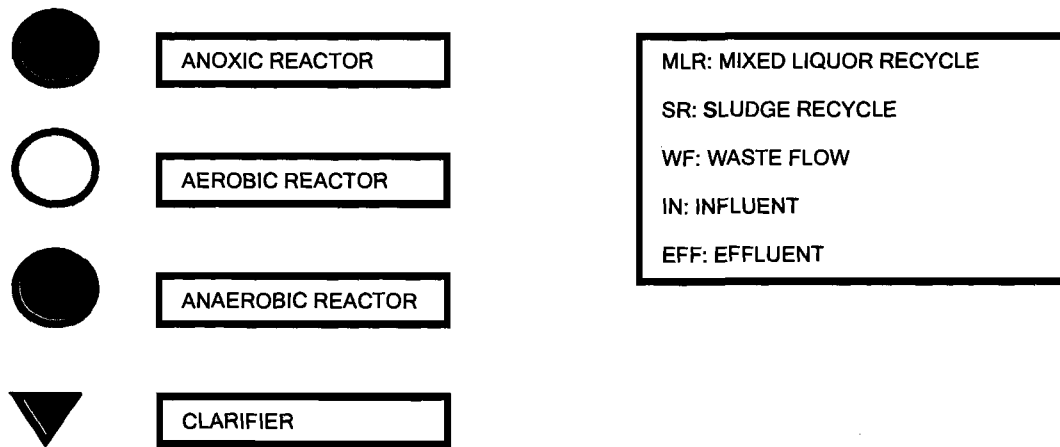


Figure 4.2.1 Key for WWTP configurations

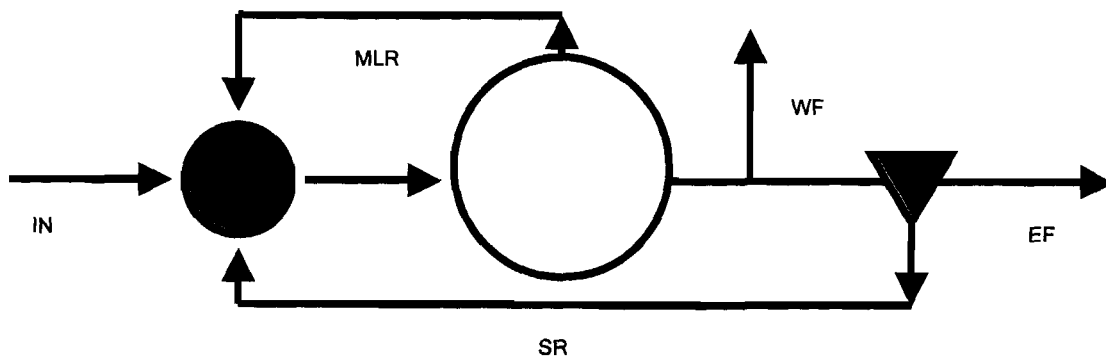


Figure 4.2.2 MLE configuration
(Metcalf and Eddy, 1991)

4.2.2 RESULTS

4.2.2 (a) Operational data

The influent (I) flows into 3 reactors. The North reactor was used for the study.

The mixed liquor (ML) from the North reactor flows into the clarifier.

The effluent (E) is the supernatant from the appropriate clarifier.

*Note: breaks in the graph lines occur when no data was available

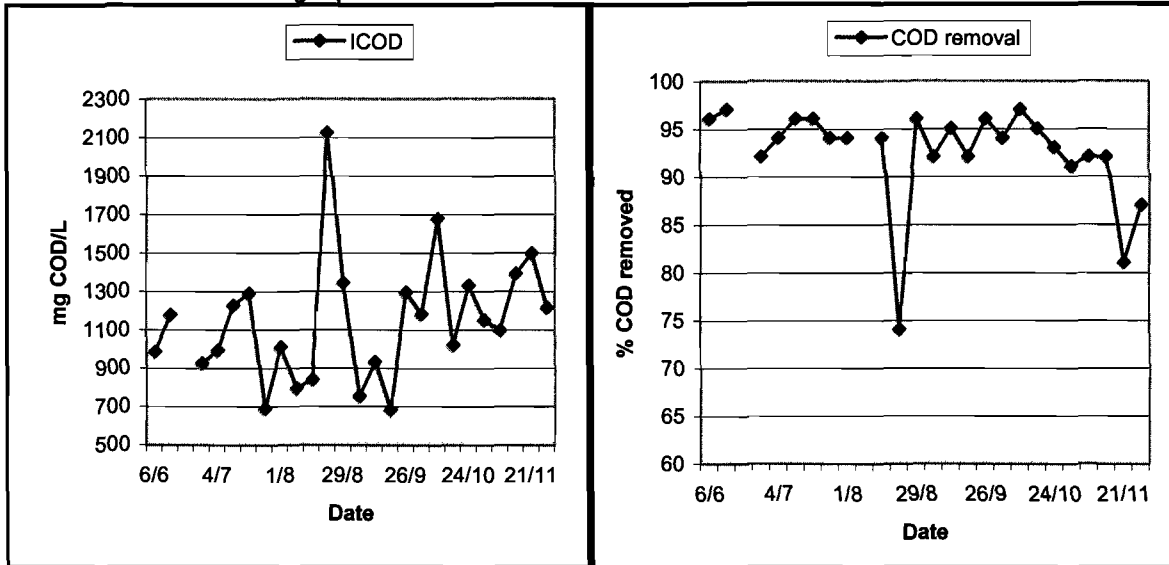


Figure 4.2.3 Graph depicting the influent COD (ICOD) at Bellville WWTP in 2007

Figure 4.2.4 Graph depicting the %COD removal at Bellville WWTP in 2007

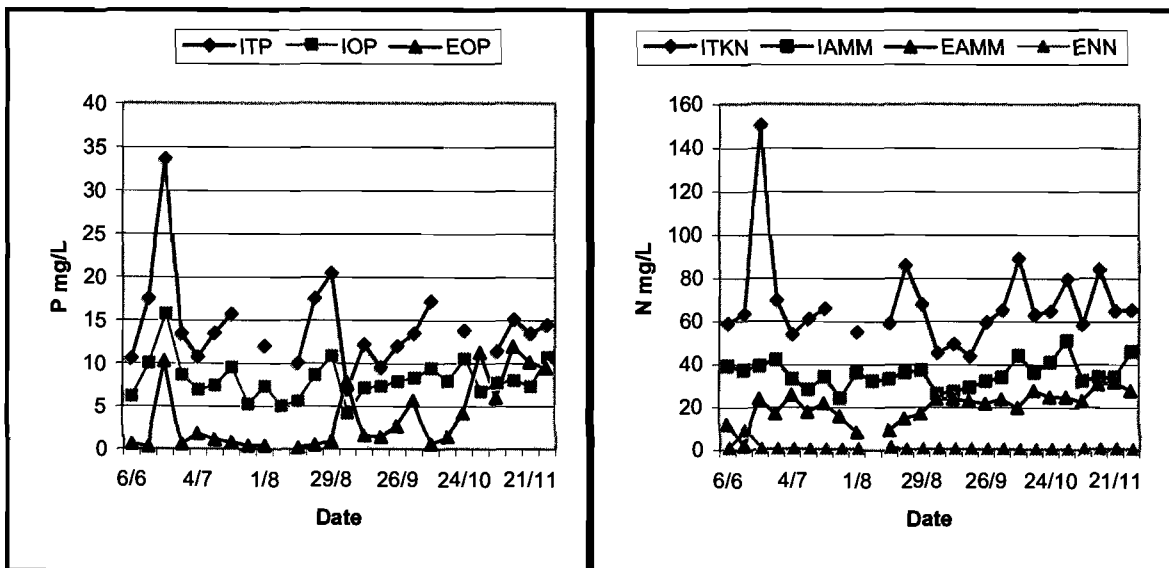


Figure 4.2.5 Graph depicting the influent total phosphate (ITP), influent σ -phosphate (IOP) and effluent σ -phosphate (EOP) levels at Bellville WWTP in 2007

Figure 4.2.6 Graph depicting the influent Total Kjeldahl nitrogen (ITKN) influent ammonia (IAMM), effluent ammonia (EAMM) and effluent nitrates and nitrites (ENN) at Bellville WWTP in 2007

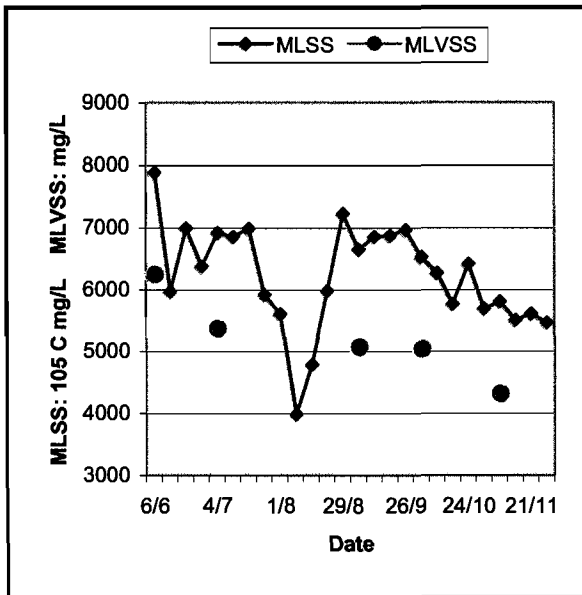


Figure 4.2.7 Graph depicting the influent mixed liquor suspended solids (MLSS) and mixed liquor volatile suspended solids (MLVSS) from the North reactor at Bellville WWTP in 2007

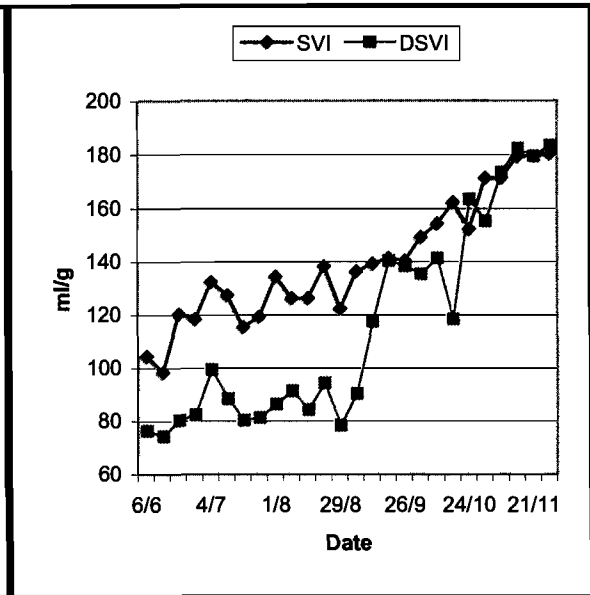


Figure 4.2.8 Graph depicting the sludge volume index (SVI) and dissolved sludge volume index (DSVI) from the mixed liquor from the North reactor at Bellville WWTP in 2007

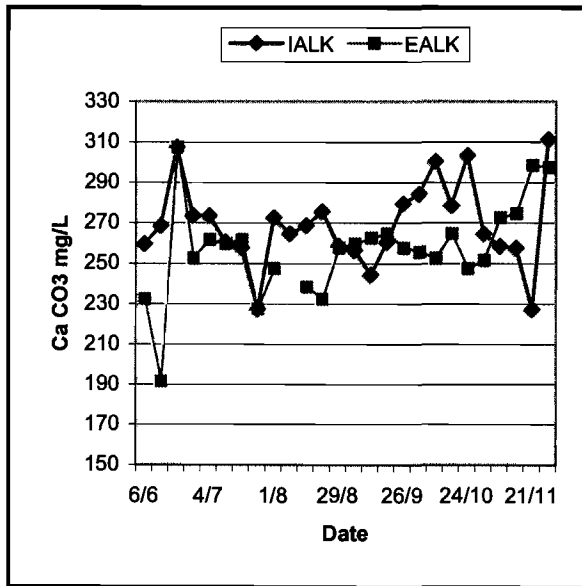


Figure 4.2.9 Graph depicting the influent alkalinity (IALK) and effluent alkalinity (EALK) at Bellville WWTP in 2007

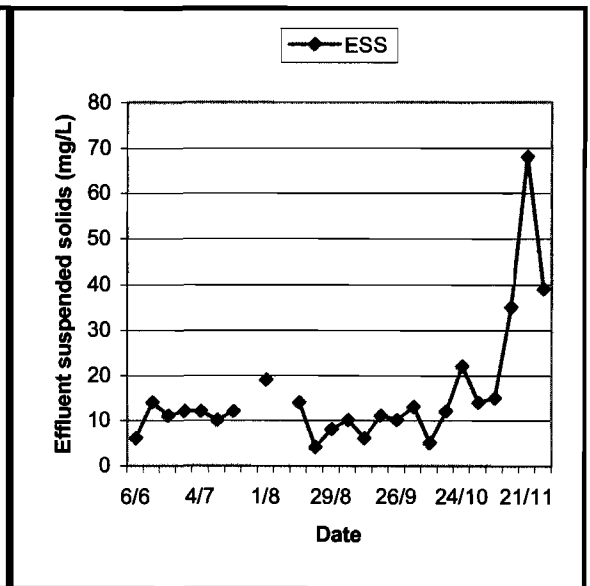


Figure 4.2.10 Graph depicting the effluent suspended solids (ESS) at Bellville WWTP in 2007

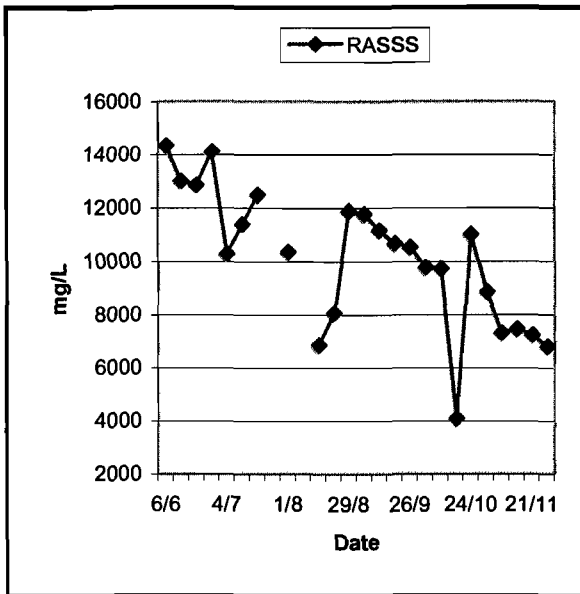


Figure 4.2.11 Graph depicting the influent return activated sludge suspended solids (RASSS) at Bellville WWTP in 2007

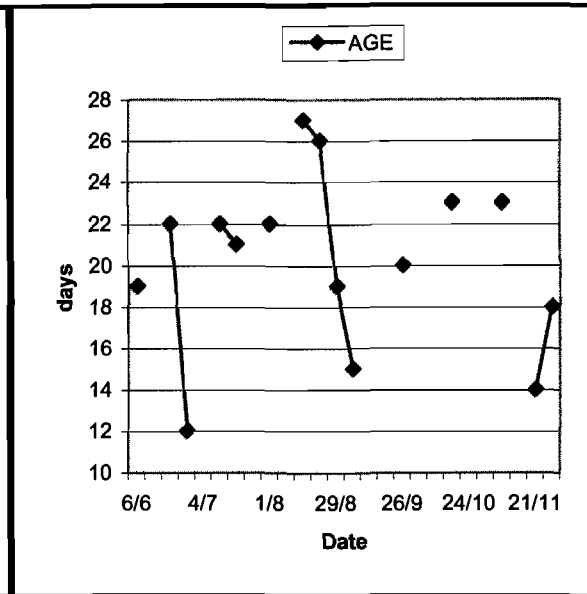


Figure 4.2.12 Graph depicting the calculated sludge age of mixed liquor (age) of the North reactor at Bellville WWTP in 2007

| TABLE 4.2.1 FOOD TO MICROORGANISM (F/M) RATIO IN NORTH REACTOR OF BELLVILLE WWTP IN 2007 | |
|------------------------------------------------------------------------------------------|----------------------|
| June | 0.46 mgCOD/mgVSS.day |
| July | 0.57 mgCOD/mgVSS.day |
| August | 0.59 mgCOD/mgVSS.day |
| September | 0.65 mgCOD/mgVSS.day |
| October | 0.70 mgCOD/mgVSS.day |

4.2.2 (b) Microscopic sludge analysis

This was performed monthly on the following dates: 28/6/07, 26/7/07, 23/8/07, 29/9/07, 25/10/07 and 22/11/07.

● Floc structure

In June and July, the flocs were irregular in shape, open in structure and firm in strength. In August and September, the flocs were irregular in shape, compact in structure and firm in strength. In October, the flocs were irregular in shape, open in structure and firm in strength. In November, the flocs were irregular in shape, open in structure and weak in strength.

The average flocs were large in size over the duration, until November, when the floc structure was divided between pin flocs or large open flocs (seemingly many small flocs joined by filaments).

There was bridging between filaments in July and August and from September there were also increasing numbers of tangled masses of filaments between flocs. There was an overall deterioration of the floc character over the period of the study.

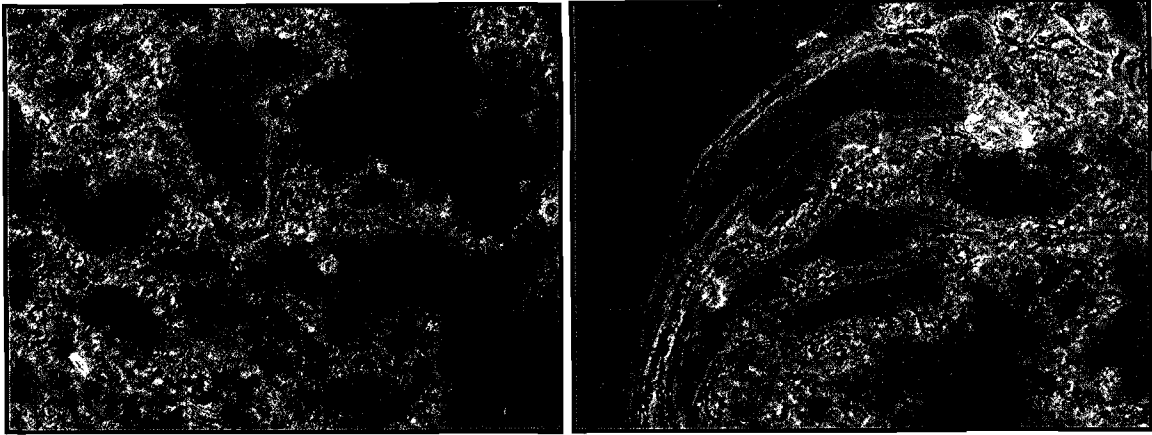


Figure 4.2.13 Micrographs of wet mounts of mixed liquor from the north reactor at Bellville WWTP taken in June 2007 (left) showing the large, open floc structure and September 2007 (right), showing the tangled masses of filaments

- Diversity

There was a medium amount of diversity over the duration. Scanty monocolonies were present in all samples.

- Filament index

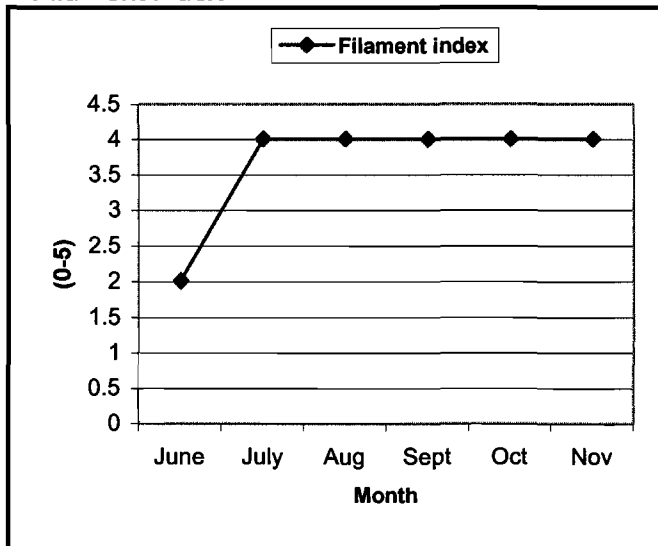


Figure 4.2.14 Graph depicting the filament index of mixed liquor from the north reactor at Bellville WWTP in 2007

TABLE 4.2.2 DOMINANT AND SECONDARY FILAMENTOUS ORGANISMS IDENTIFIED IN THE MIXED LIQUOR OF ATHLONE WWTP REACTOR A IN 2007

| AS DOMINANT FILAMENTS | | | | | | | | |
|------------------------|------|------|-----|------|-----|-----|-------------------------|------------------------|
| | June | July | Aug | Sept | Oct | Nov | % Prevalence (dominant) | % Prevalence (overall) |
| Type 1851 | X | X | X | X | X | X | 100 | 100 |
| Type 021N | X | X | X | | | X | 67 | 100 |
| AS SECONDARY FILAMENTS | | | | | | | | |
| Type 021N | | | | X | X | | } | Also dominant |
| Actinomycetes | X | X | X | X | X | | 0 | 83 |
| Type 0092 | | X | X | X | X | | 0 | 67 |
| Type 0041 | | | X | X | X | X | 0 | 67 |
| <i>Beggiatoa</i> spp. | | | | | | X | 0 | 17 |

TABLE 4.2.3 PROTOZOA/METAZOA IDENTIFIED IN THE MIXED LIQUOR OF ATHLONE WWTP REACTOR A IN 2007

| SCANTY | | | | | | | | | |
|----------------------|-----------------------------|------|------|-----|------|-----|-----|---|---|
| | | June | July | Aug | Sept | Oct | Nov | | |
| Sessile ciliates | <i>Carchesium</i> spp. | X | | X | | | | | |
| | <i>Epistylis</i> spp. | | X | | | | | | |
| | <i>Opercularia</i> spp. | | | X | | | | | |
| | Suctorean | | | X | | | | | |
| | <i>Vorticella</i> spp. | | X | X | X | | | | |
| Crawling ciliates | <i>Aspidisca</i> spp. | | X | | X | | | | |
| | <i>Trachelophyllum</i> spp. | X | X | | | | | | |
| | Unidentified | | X | | | | | | |
| Free-living ciliates | <i>Blespharisma</i> spp. | X | X | X | X | | | | |
| | <i>Euplotes</i> spp. | | X | | | | | | |
| | <i>Litonotus</i> spp. | | | X | | | | | |
| | <i>Spirostomum</i> spp. | X | X | X | | | | | |
| | Unidentified | | | X | X | | | | |
| Flagellates | <i>Bodo</i> spp. | | X | X | | | | | |
| | <i>Peranema</i> spp. | X | X | X | | | | | |
| | <i>Pleuromonas</i> spp. | | | | | | | | X |
| | <i>Trepomonas</i> spp. | | | | | | X | | |
| Amoebae | | X | X | X | X | X | | | |
| Rotifers | | X | X | X | X | X | | | |
| Unidentified "worms" | | | | | X | X | | | |
| MODERATE | | | | | | | | | |
| Free-living ciliates | <i>Spirostomum</i> spp. | | | | X | | | | |
| Crawling ciliates | <i>Trachelophyllum</i> spp. | | | X | | | | | |
| Flagellates | <i>Bodo</i> spp. | | | | X | X | | X | |
| | <i>Trepomonas</i> spp. | | | | | | | | X |
| Unidentified "worms" | | X | X | X | | | | | |



Figure 4.2.15 Wet mount of mixed liquor showing unidentified worm-like organism. It is possible to compare the size to the surrounding spirochaetes

4.2.3 DISCUSSION

By examining figures 4.2.3 and 4.2.4, it can be ascertained that although influent COD was comparatively high (albeit erratic), removal was satisfactory until October. At this stage there was a marked deterioration in COD removal that coincided with increased levels of effluent suspended solids (figure 4.2.10).

Phosphate removal is not part of the MLE design process as there is no anaerobic zone, but surprisingly this function was good in most samples until September. From October there was also deterioration of this process, as reflected in figure 4.2.5. In an exhaustive literature review, Oehman *et al.* (2007) implicated the COD/P ratio, the presence of adequate volatile fatty acids (VFA's) and the lack of nitrate in the anaerobic zone as being the most important parameters for efficient phosphorous removal. Such conditions allow the phosphorous accumulating organisms (PAO's) to successfully out-compete the glycogen accumulating organisms (GAO's). (The latter is discussed in more detail in chapter 2). It can be assumed that the level of VFA's would be adequate in a plant treating mixed industrial effluent. The COD/P values at Bellville ranged from 66.0-121.8mgCOD/mgP with a mean of 89.9mgCOD/mgP. According to Liu *et al.* (1997), a ratio of 10-20mgCOD/mgP should favour the growth of PAO's, while levels greater than 50mgCOD/mgP should enhance the growth of GAO's. This fact would lead to decreased, not enhanced phosphorous removal.

The explanation for the unexpected results is probably due to the fact that nitrification, and thus nitrification products were almost entirely lacking at Bellville WWTP. This would render the anoxic reactor a de facto anaerobic reactor with the result that the plant became capable of functioning as an enhanced biological phosphorous removal (EBPR) WWTP. Another mechanism for phosphorous removal could very well have been precipitation with ionic components (such as iron or aluminium), or absorption with components that were inherent in the industrial waste.

By examining figure 4.2.6, it can be seen that influent ammonia levels were not high and removal of this chemical was consistently poor. Nitrates and nitrites in the clarifier effluent were absent for almost the entire six months of the study. In the only sample where nitrates/nitrites were present, ammonia levels were zero. Alkalinity, reflected in figure 4.2.9, was high. From these observations, it can clearly be deduced that

nitrification was poor, and that the denitrification potential of the plant was sufficient to remove any nitrification products. It thus stands to reason that in the MLE system, with an anoxic zone preceding the aerobic zone, no nitrates/nitrites were present as the sludge passed into the aerobic zone. Therefore, the bulking conditions encountered could not be linked to the conditions required in the AA bulking hypothesis of Casey *et al.* (1999).

The two most likely causes of the poor nitrification achieved at Bellville are the high F/M (Eikelboom, 2000) or the presence of substances toxic to the nitrifier population (Gerhaey *et al.*, 1997), or a combination of these factors. Adequate overall sludge age, shown in graph 4.2.12 that is backed up by the presence of slow-growing metazoa such as rotifers, make the possibility of nitrifier washout unlikely. The population of protozoa and metazoa point to a plant with a high load and/or low DO (Eikelboom, 2000) - over the study period, there was a gradual increase in the flagellate to ciliate ratio that indicates these conditions. In fact, there were no ciliates present in the samples from either October or November.

A healthy ciliate population is indicative of sufficient oxygen and/or a low floc load (Eikelboom, 2000). The fact that the food to microorganisms (F/M) ratio (given in table 4.2.1) exhibits an increasing trend points to the loading rates being the "culprit". The increased effluent COD substantiates the likelihood that COD overloading lead to poor plant performance.

With an increase in F/M, the oxygen requirements are elevated. Unless the DO was increased to cater for the higher loading, lack of oxygen for both COD removal and nitrification would play a role in poor plant performance.

The presence of moderate numbers of *Bodo* spp. in September, October and November and *Trepomonas* spp. in November is further evidence - both of these flagellates are associated with high sludge load and/or lack of oxygen according to Eikelboom (2000).

The fact that none of the filaments exclusively associated with low DO by Jenkins *et al.* (2004), namely *Sphaerotilus natans*, Type 1701 and *Haliscomenobacter hydrosis* were found in any of the samples tends to implicate F/M rather than D/O as being more plausible. It is impossible to accurately speculate on whether toxins such as heavy metals were responsible for a diminished nitrifier population, as no additional results are available (as in the case of Athlone WWTP). However, if there were large quantities of heavy metals in the influent, this could explain both the lack of nitrification (due to toxicity) and the good phosphorus removal (due to precipitation).

Figure 4.2.7 depicts the gradual decrease in MLVSS, figure 4.2.8 the increase in DSVI and SVI and figure 4.2.14 the increase in FI. Musvoto *et al.* (1999) demonstrated a link between decreasing VSS and increasing DSVI that is borne out by these results. The high filament index pointed to the existence of filamentous bulking conditions from July, but although there was a steady increase in the DSVI, the value did not exceed 150mg/L until October. The deteriorating sludge quality in terms of FI, DSVI, floc character and the increase in numbers of tangled filament masses, was accompanied by a gradual decrease in RASSS. The link between poor sludge quality and decreasing RASSS was a phenomenon also noted at the Athlone WWTP.

The dominant filaments were Type 1851, (for the duration of the study period), and Type 021N (in all months bar September and October). In chapter 5, the association of both of these organism types with bulking conditions in the study WWTP's was established. According to Eikelboom (2000), Type 1851 is supposedly associated with industrial

WWTP's such as Bellville. It is classed as a low F/M filament by Jenkins *et al.* (2004) and Eikelboom (2000). However, Eikelboom (2000) does concede that there is limited knowledge about this organism. In this case the F/M values (given in table 4.2.1) were not low, and showed an increasing trend from June to October.

Dominant growth of Type 1851 must therefore be linked to other factors besides low F/M conditions in this instance. Both type 1851 and type 021N are associated with low molecular weight (LMW) compounds (Eikelboom, 2000), and the latter is not considered low F/M. It can be speculated that the numerical abundance of these two filament types was associated with increased availability of LMW compounds that is in turn reflected by the increasing F/M. In addition, a lack of nutrients (nitrates/nitrites) may be implicated for reasons outlined in 4.1.3 (Athlone WWTP discussion). The continual presence of actinomycetes, albeit as secondary filaments, supports the fact that there was a large LMW component (Eikelboom, 2000).

Type 0092, so prevalent in the study as a dominant species was found as a secondary species in four of the six samples. Although this filament and Type 0041 are classed as low F/M by both Jenkins *et al.* (2004) and Eikelboom (2000), Noutsopolous *et al.* (2006) determined the preferred range for growth of Type 0092 to be between 0.19 and 0.45 mgCOD/mgVSS.day. This organism was found at much higher F/M ratios in this study. By November, when the F/M ratio was highest, Type 0092 was no longer apparent, but Type 0041 was. These findings question the validity of classifying both of these filaments as low F/M.

In summary, then, the evidence points to an overloaded industrial WWTP, with an increasing F/M ratio that lead to inadequate nitrification and increasingly poor COD removal. The presence of toxic substances may or may not have played a role in poor nitrification. The high F/M also had a negative effect on floc formation, the high level of LMW compounds provided a competitive advantage for certain filaments over their heterotrophic rivals, leading to the presence of bulking conditions. This may have been exacerbated by nutrient (nitrate) deficiency. In addition, although phosphorus removal is not an inherent feature of WWTP's with the MLE configuration, this function did indeed take place. This was probably due to the fact that the anoxic zone became an effective anaerobic zone, allowing for functional EBPR.

4.3 BORCHERDS QUARRY WWTP

4.3.1 PLANT CONFIGURATION AND OPERATING PARAMETERS

Borcherd's Quarry WWTP has a design capacity of 32ML/day with an influent of approximately 40% industrial and 60% domestic effluent. The plant processes the night soil from the entire black bucket system of the mega city. Landfill leachate is processed on weekends only, so the influent is highly variable from day to day. The influent that flows into the two bioreactors (named B and C) in equal volumes is the supernatant from the PST. The configuration is the same as that of the Cape Flats WWTP, namely the 5-stage Bardenpho (Phoredox). The DO concentration in the primary aerobic reactor is 2.3mg/L and the method of aeration is diffuse. The plant experiences intermittent problems with scum formation.

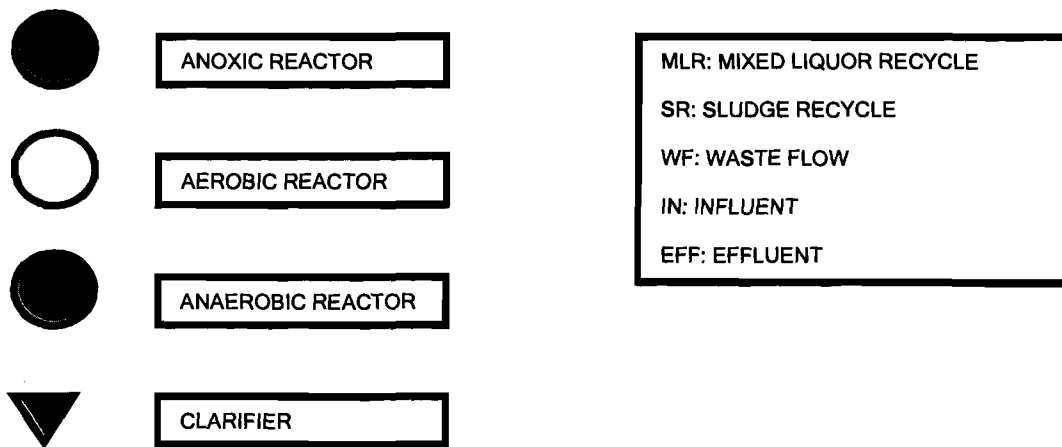


Figure 4.3.1 Key for WWTP configurations

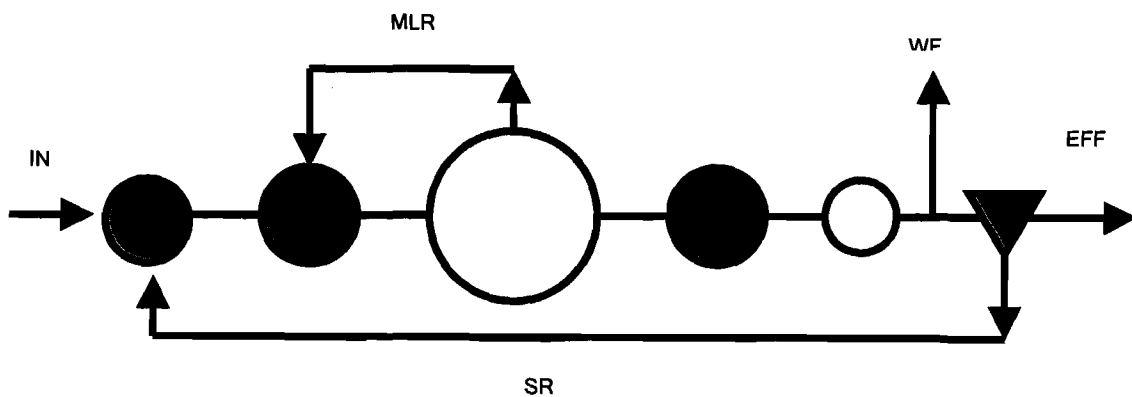


Figure 4.3.2 5-stage Bardenpho (Phoredox) configuration (Metcalf and Eddy, 1991)

4.3.2 RESULTS

4.3.2.1 (a) Operational data

The influent (I) into is supernatant from the primary settling tank (PST) that flows into bioreactor B and bioreactor C.

The mixed liquor (ML) from reactor B and reactor C flows into clarifier B and clarifier C respectively.

The effluent (E) is the supernatant from the clarifier B and C.

*Note: breaks in the graph lines occur when no data was available

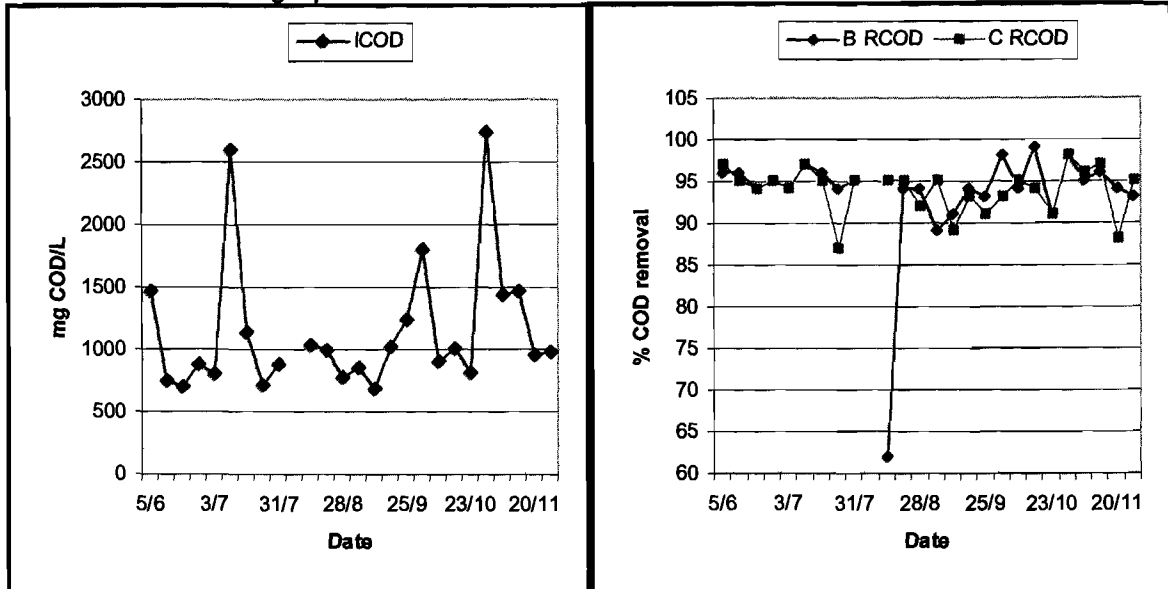


Figure 4.3.3 Influent COD (ICOD) into bioreactors of Borchers quarry WWTP in 2007

Figure 4.3.4 % removal of COD by bioreactor B (B RCOD) C (C RCOD) at Borchers quarry WWTP in 2007

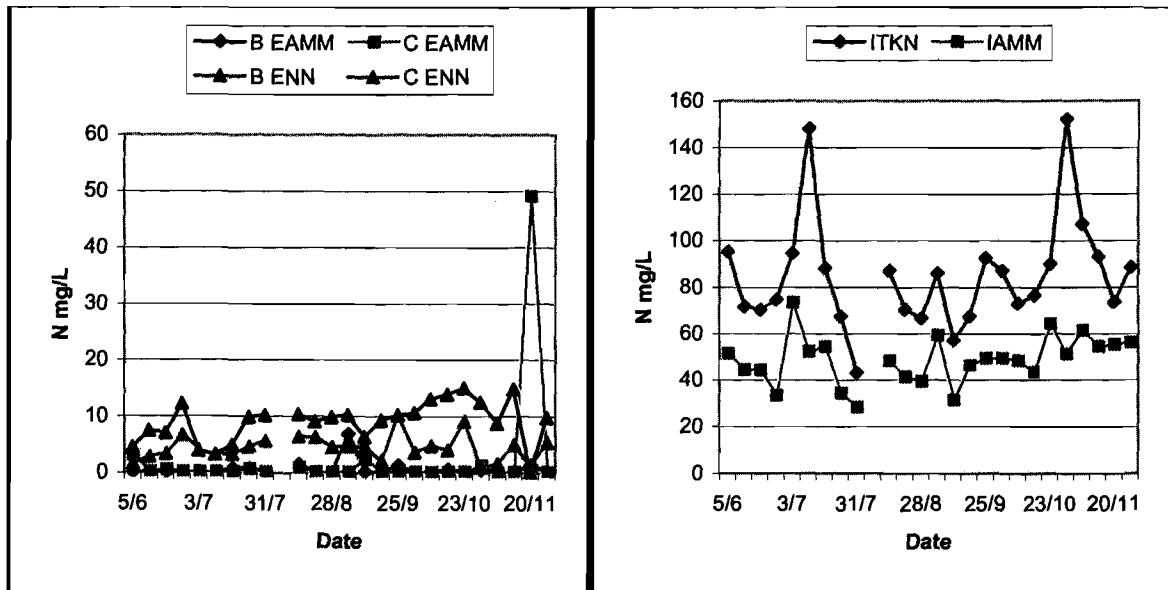


Figure 4.3.5 Effluent ammonia from clarifier B (B EAMM) and C (C EAMM) and effluent nitrates/nitrites from clarifier B (B ENN) and C (C ENN) at Borchers quarry WWTP in 2007

Figure 4.3.6 Influent Total Kjeldahl ammonia (ITKN) and influent ammonia (IAMM) into bioreactors of Borchers quarry WWTP in 2007

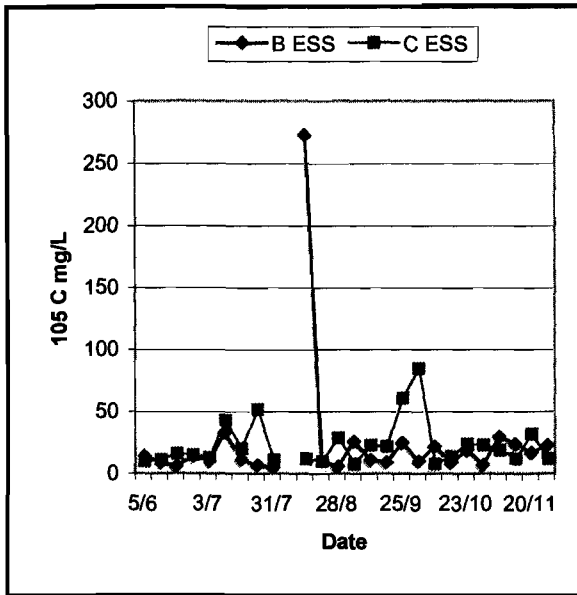


Figure 4.3.7 Effluent suspended solids from clarifier B (B ESS) and C (C ESS) at Borchers quarry WWTP in 2007

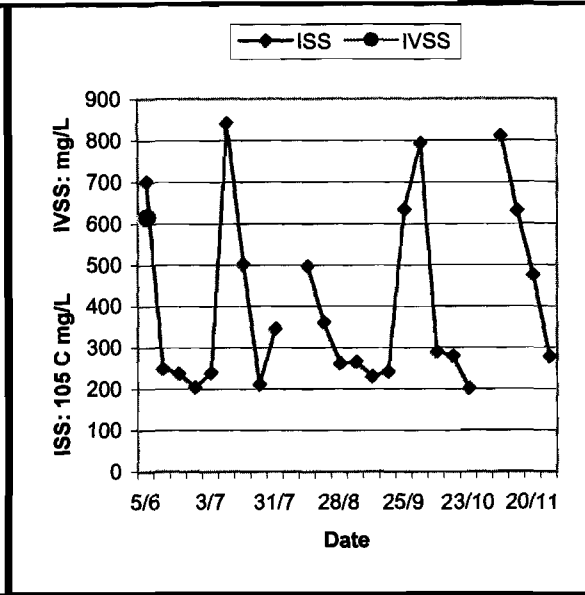


Figure 4.3.8 Influent suspended solids (ISS) and influent volatile suspended solids (IVSS) from the primary settling tank at Borchers quarry WWTP in 2007

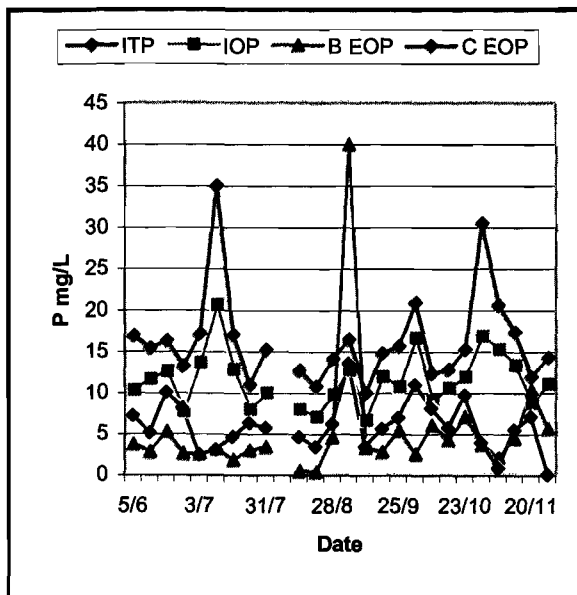


Figure 4.3.9 Influent total phosphates (ITP), influent σ -phosphates (IOP), and effluent σ -phosphates from clarifier B (B EOP) and C (C EOP) at Borchers quarry WWTP in 2007

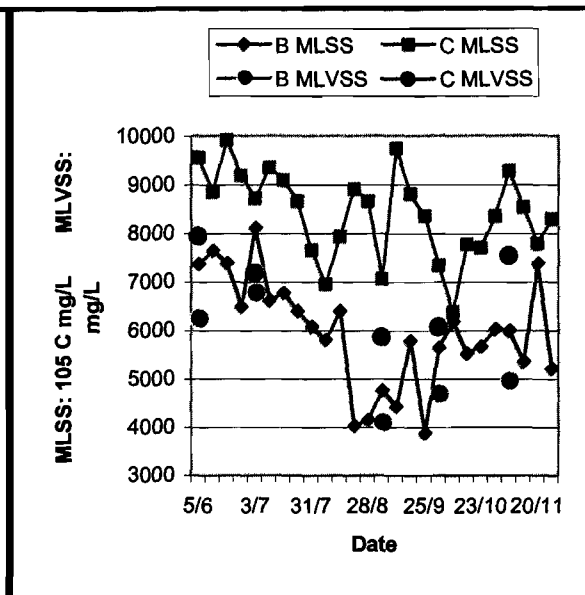


Figure 4.3.10 Mixed liquor suspended solids from reactor B (B MLSS) and C (C MLSS) and mixed liquor volatile suspended solids from reactor B (B MLVSS) and reactor C (C MLVSS) at Borchers quarry WWTP in 2007

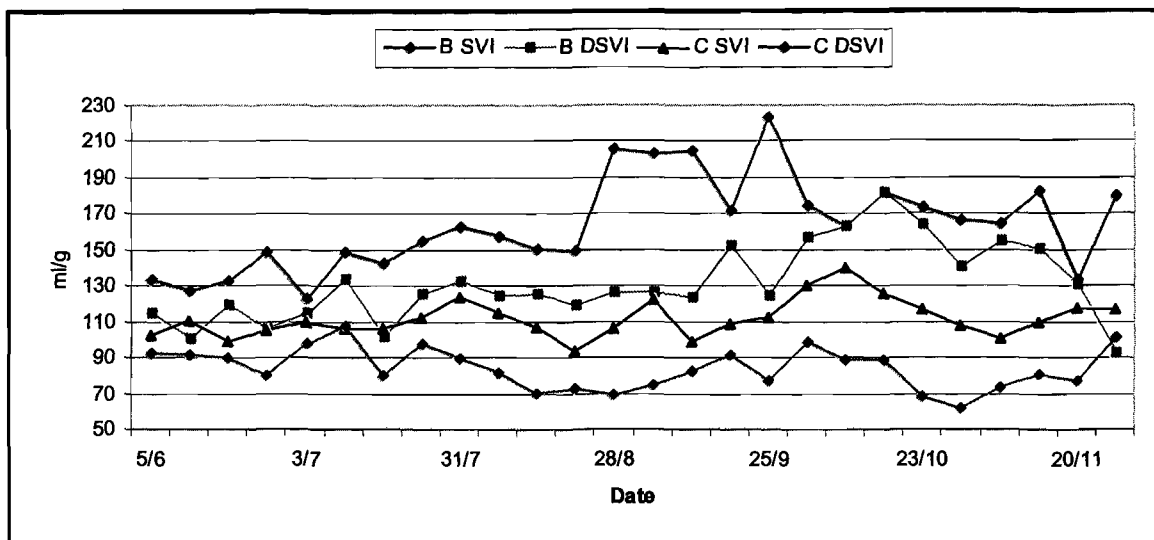


Figure 4.3.11 Sludge volume index from reactor B (B SVI) and reactor C (C SVI) and dissolved sludge volume index from reactor B (B DSVI) and reactor C (C DSVI) at Borchers quarry WWTP in 2007

| | REACTOR B | REACTOR C |
|-----------|----------------------|----------------------|
| June | 0.26 mgCOD/mgVSS.day | 0.20 mgCOD/mgVSS.day |
| July | 0.13 mgCOD/mgVSS.day | 0.12 mgCOD/mgVSS.day |
| September | 0.23 mgCOD/mgVSS.day | 0.15 mgCOD/mgVSS.day |
| October | 0.42 mgCOD/mgVSS.day | 0.31 mgCOD/mgVSS.day |
| November | 0.31 mgCOD/mgVSS.day | 0.20 mgCOD/mgVSS.day |

4.3.2 (b) Microscopic sludge analysis

This was performed monthly on samples from reactor B and reactor C on the following dates: 20/6/07, 18/7/07, 29/8/07, 19/9/07, 24/10/07 and 21/11/07. In addition, filament identification was performed on scum from reactor B on 3/10/07.

• Floc Structure

Reactor B

The flocs were round in shape and firm in strength for the duration of the study, except for September, when flocs were both round and irregular in shape. With regard to the structure of the flocs, they were compact in June and October and open in July, August, September and November. The flocs were moderate in size from June to August. In September, the flocs were dispersed between being small, medium and large in size, while in October and November the flocs were predominantly small. In September, there was much bridging between flocs and in October and November there were numerous rosettes present with filaments protruding from the flocs and free in the sludge (see figure 4.3.11).

Reactor C

The flocs were round in shape, open in structure and firm in strength for the duration of the study, with the exception of October and November, when they were both round and irregular in shape.

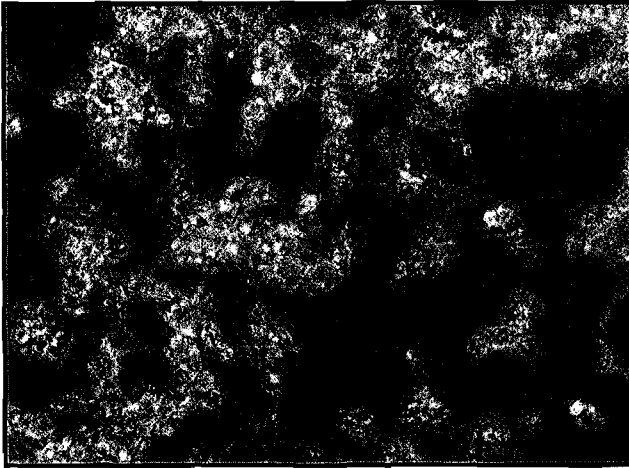


Figure 4.3.12 Wet mount of mixed liquor from reactor B at Borchers quarry WWTP in October 2007, showing numerous Type 021N rosettes and filaments protruding from pin flocs

● Diversity

Reactor B and reactor C

There was a moderate amount of microbial diversity for the duration of the study in both reactors. There were scanty monocolonies in all samples, except those from reactor B in October and reactor C in August, when there was a moderate number of monocolonies.

● Filament index

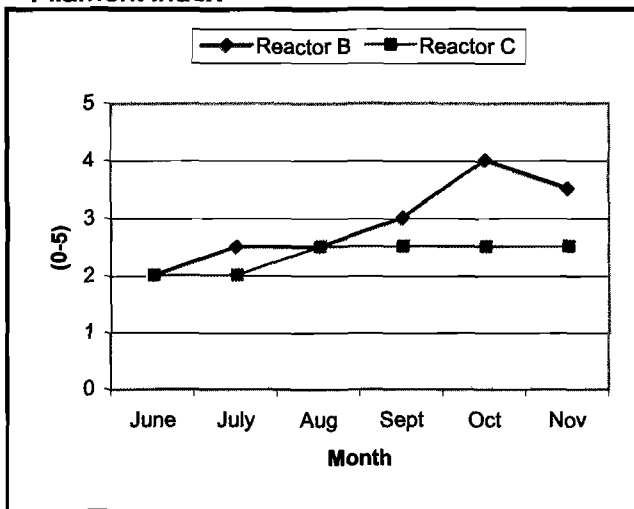


Figure 4.3.13 Filament index from mixed liquor of reactor B and reactor C at Borchers quarry WWTP in 2007

TABLE 4.3.2 DOMINANT AND SECONDARY FILAMENTOUS ORGANISMS IDENTIFIED FROM THE MIXED LIQUOR OF REACTOR B AND REACTOR C AT BORCHERDS QUARRY WWTP IN 2007

| AS DOMINANT FILAMENTS | | | | | | | | |
|------------------------|---------|---------|---------|---------|---------|---------|---------------------------------|--------------------------------|
| | June | July | Aug | Sept | Oct | Nov | % Prevalence B and C (dominant) | % Prevalence B and C (overall) |
| Type 0092 | X (B,C) | X (B,C) | X (B,C) | X (B,C) | X (B,C) | X (B,C) | 100 | 100 |
| Type 021N | | | X (B) | X (B) | X (B) | X (B,C) | 42 | 92 |
| Actinomycetes | | | X (C) | | | | 8 | 17 |
| AS SECONDARY FILAMENTS | | | | | | | | |
| Type 021N | X (C) | X (B,C) | X (C) | X (C) | X (C) | | } | Also dominant filaments |
| Actinomycetes | | X (C) | | | | | | |
| <i>M. Parvicella</i> | | X (B,C) | X (B,C) | X (B,C) | X (B) | | 0 | 58 |
| Type 0041 | | | X (C) | | X (B,C) | X (B) | 0 | 33 |
| <i>N. limicola III</i> | | | | | X (B,C) | X (C) | 0 | 25 |
| Type 0803 | X (B) | | | | | | 0 | 8 |

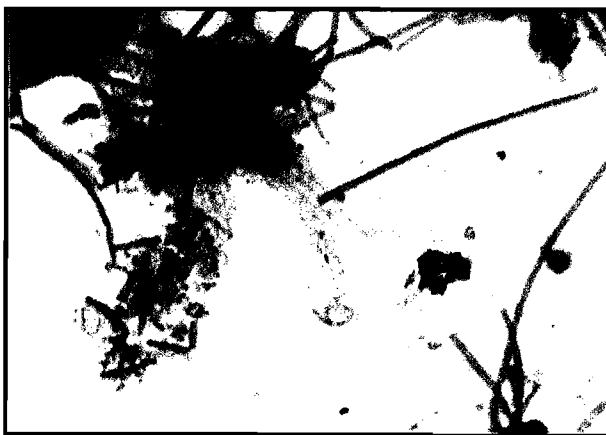


Figure 4.3.14 Neisser stain of Type 021N (Neisser negative) and Type 0092 (Neisser positive - blue) from reactor B Of Borchers quarry WWTP in November 2007

TABLE 4.3.3 PROTOZOA/METAZOA IDENTIFIED IN THE MIXED LIQUOR FROM REACTOR B AND REACTOR C AT BORCHERD'S QUARRY WWTP IN 2007

SCANTY

| | | June | July | Aug | Sept | Oct | Nov |
|----------------------|-----------------------------|------------|------------|------------|------------|------------|------------|
| Sessile ciliates | <i>Carchesium</i> spp. | X (C) | X (B,C) | X (B) | X (B) | | X (B) |
| | <i>Epistylis</i> spp. | X (B) | | X (B,C) | X (B) | X (B,C) | X (C) |
| | <i>Vaginicola</i> spp. | | | | | X (C) | |
| | <i>Vorticella</i> spp. | X (B,C) | X (B,C) | X (B,C) | X (C) | | X (C) |
| Crawling ciliates | <i>Aspidisca</i> spp. | X (B,C) | X (B,C) | X (B,C) | X (B) | X (B,C) | X (B,C) |
| | <i>Chilodonella</i> spp. | | X (C) | X (C) | | | |
| | <i>Paramecium</i> spp. | | | | | X (B) | |
| | <i>Trachelophyllum</i> spp. | | X (C) | X (B,C) | X (B,C) | X (C) | X (B) |
| | <i>Blepharisma</i> spp. | X (C) | X (C) | X (C) | X (C) | X (C) | |
| Free-living ciliates | <i>Colpidium</i> spp. | X (B,C) | X (B,C) | X (B,C) | X (B,C) | X (B,C) | X (B,C) |
| | <i>Euplotes</i> spp. | | X (B) | | | | |
| | <i>Litonotus</i> spp. | | | X (B,C) | | X (B) | |
| | <i>Colpidium</i> spp. | | | X (B) | | X (B) | X (B) |
| | <i>Spirostomum</i> spp. | X (C) | X (C) | X (C) | X (C) | X (C) | |
| | unidentified | X (C) | | | | | |
| Flagellates | <i>Bodo</i> spp. | X (B) | X (B,C) | X (C) | X (C) | | X (B) |
| | <i>Peranema</i> spp. | X (B,C) | X (B,C) | X (B,C) | X (B,C) | X (B,C) | X (B,C) |
| | Unidentified | X (B,C) | X (B) | X (B,C) | X (B,C) | X (B,C) | X (C) |
| Amoebae | X (B,C) | X (B) | X (B,C) | X (C) | X (B) | X (B) | |
| Testate amoebae | | | X (C) | | | | |
| Heliozoa | | X (C) | | | | | X (B) |
| Rotifers | | | X (B,C) | X (B,C) | X (B,C) | X (B) | |
| Tardigrades | | | X (C) | | | | |
| Oligochaeta | | X (C) | | | X (C) | | |

TABLE 4.3.3 (CONTINUED) PROTOZOA/METAZOA IDENTIFIED FROM THE MIXED LIQUOR OF REACTOR B AND REACTOR C AT BORCHERDS QUARRY WWTP IN 2007

| MODERATE | | | | | | | |
|-------------------|------------------------|--|--|--|--|----------|----------|
| Sessile ciliates | <i>Epistylis</i> spp. | | | | | X (C) | |
| Crawling ciliates | <i>Aspidisca</i> spp. | | | | | X (C) | |
| Oligochaeta | | | | | | | X (C) |
| Rotifera | | | | | | | X (B) |
| NUMEROUS | | | | | | | |
| Flagellates | <i>Trepomonas</i> spp. | | | | | | X (C) |

4.3.3 DISCUSSION

Borcherds Quarry experienced a comparatively high influent COD and SS. Apart from one anomalous result in each case, removal of both COD and SS was efficient (refer to figures 4.3.3, 4.3.4, 4.3.7 and 4.3.8).

Regarding the removal of nitrogen reflected in figures 4.3.5 and 4.3.6, nitrification efficiency was excellent, despite the sometimes relatively short calculated sludge age and high influent TKN. There was also one unexplained anomalous result in the case of effluent ammonia, where it seems that almost no nitrification took place. In this instance, there was complete denitrification, perhaps because there was insufficient nitrification product to denitrify. As with Cape Flats (also 5-stage Bardenpho), complete denitrification did not take place. Residual nitrate/nitrite levels in the clarifier supernatant were higher from reactor C than reactor B. The influent COD/TKN ratio ranged from 8.4 to 20.3 mgCOD/mgTKN with a mean of 13.1 mgCOD/mgTKN. These values were substantially higher than the values found at Cape Flats WWTP (mean of 7.8mgCOD/mgTKN). Even the lowest value of 8.4 mgCOD/mgN that was obtained at Borcherds quarry equaled that reported by Kujawa *et al.* (1996) as necessary for complete denitrification. There was no correlation between the influent COD/TKN ratio and effluent nitrates/nitrites as was the case with Cape Flats WWTP (see section 4.4.3). At Borcherds Quarry, it is highly unlikely that lack of electron donor was the reason for incomplete denitrification. The most probable reasons for the presence of residual nitrates/nitrites would thus be plant operating conditions, for example, insufficient mixed liquor recycle from the aerobic to the first anoxic zone or insufficient retention time in the anoxic zone.

Regarding the removal of phosphorous, shown in figure 4.3.9, results were inconsistent, but poor for this type of configuration. The COD/P ratio ranged between 46.5-91.7mgCOD/mgP with a mean of 69.4mgCOD/mgP. According to Liu *et al.* (1997), a ratio of 10-20mgCOD/mgP should favour the growth of PAO's, while levels greater than 50mgCOD/mgP should enhance the growth of GAO's. Theoretically, then according to this ratio, the growth of GAO's should have been favoured with a resultant negative effect on phosphorous removal. In addition, the likely presence of nitrates in the anaerobic zone probably also played a role and is substantiated by the fact that reactor C, with the highest levels of nitrates/nitrites in the clarifier effluent, also showed higher effluent phosphorous levels (poor removal). Both reactors receive the same influent, but

there is a link between denitrification efficiency that seemingly translates into phosphorous removal efficiency. In addition to the fact that the presence of nitrates in the anaerobic zone allows the ordinary heterotrophs to compete for substrate, there is another biological link - some PAO's are capable of denitrification in the anoxic zone. The polyhydroxybutyrate (PHB) formed when phosphate is released serves as a carbon source under anoxic conditions. Nitrate serves as the electron acceptor and phosphorous is taken up simultaneously. In a situation where the growth of heterotrophs and GAO's is favoured over the growth of PAO's, this will also then negatively affect denitrification (Kuba *et al.*, 1996).

An interesting result was that reactor C, which exhibited the worst performance in terms of nutrient removal, gave consistently better results with both of the bulking indicators DSVI and FI, as depicted by graphs 4.3.11 and 4.3.13 respectively. None of the mixed liquor samples taken from reactor C gave values indicating the presence of bulking conditions for either of these parameters. In fact, the DSVI values never rose much over 100ml/g for the duration of the study. In contrast, both parameters for the mixed liquor samples from reactor B indicated bulking conditions from September to November.

The presence of higher levels of nitrates/nitrites in reactor B and bulking conditions being present only in reactor C would seem to refute the AA bulking hypothesis of Casey *et al.* (1999) as being pertinent in this case. Importantly, in the Cape Flats WWTP with similar levels of nitrates/nitrites in the effluent, bulking was not apparent. There are no results for the concentrations of these nitrogenous substances in the reactors themselves, so this is merely an assumption.

Type 021N was never dominant at Cape Flats and was only dominant for one month in reactor C at Borchers Quarry. However, this filament was co-dominant for four of the six months of the study period in reactor B. This may give a clue as to the reason for bulking conditions being present only in reactor B. If Type 021N was indeed the filament responsible for bulking conditions, the AA bulking hypothesis of Casey *et al.* (1999) is again implausible as according to Eikelboom (2000), this filament is aerobic, and not facultative in nature.

The reported calculated sludge age was low at times, but nitrification that requires long MCRT's was good throughout. There was an absence of metazoa towards the end of the study. These organisms grow slowly (Eikelboom, 2000) and are also reliant upon long retention times. There was a healthy and diverse ciliate population in both reactors throughout, reflecting good oxygenation and probable lack of toxic substances (Jenkins *et al.*, 2004).

From September, the MLVSS was lower in reactor B. Once again, a link is established between increasing DSVI and decreasing MLVSS (Casey *et al.*, 1999b). In this case a clear comparison can be seen between both reactors. It is not clear which comes first - increasing DSVI or decreasing MLVSS (see figure 4.3.10).

The microscopic quality of the sludge mirrored the DSVI results obtained from reactor B. There was a steady deterioration and by November the flocs were predominantly pin forms and numerous filaments were seen both protruding from the flocs and between the flocs. Many rosettes were seen. This is shown in figure 4.3.12.

By examining table 4.3.2, it can be ascertained that the dominant filaments consisted of Type 0092 in all samples from both reactors and Type 021N which co-dominated in reactor B from August to the end of the study. Type 021N was constantly present as a

secondary species in reactor C, and also became co-dominant by November. *Microthrix parvicella* was prevalent in 58% of samples albeit as a secondary species. The only noticeable difference in the filament composition between the two reactors was the abundant numbers of Type 021N found in the mixed liquor samples from reactor B. It is not clear whether the overgrowth of this filament caused a decrease in the MLVSS with a resultant increase in F/M of reactor B (in comparison to reactor C), or whether an increased F/M resulted in the overgrowth of Type 021N, which in turn lead to a decreased MLVSS. The F/M values are given in table 4.3.1. Type 021N also became dominant in the Athlone WWTP at a time when the plant exhibited bulking tendencies. *M. parvicella* is also associated with LMW compounds, as is *N. limicola* III (Eikelboom, 2000).

Unfortunately, samples from Borchers Quarry WWTP are only taken on a particular day of the week, so influent composition only mirrors the circumstances on those days, complicating analysis of results.

The growth of Type 021N is enhanced by the presence of easily biodegradable compounds and/or sulphides. Such substances could be found from either the night soil (septic sewage) or the industrial effluent. Under nutrient removal conditions, the easily biodegradable fraction is usually expended in the anaerobic and anoxic zones, so the aerobic Type 021N does not have access to nutrients in the oxic zone (Eikelboom, 2000). It is thus a possibility that with an increase in F/M due to decreased VSS or vice versa, there was more substrate still available for this organism under aerobic conditions. The decrease in VSS presupposes that there was a decrease in the ordinary heterotrophic population. Thus, there would also have been less competition for substrate. Jenkins *et al.* (2004) associates the presence of rosettes and gonidia of Type 021N with nutrient deficiency, but this is unlikely as neither total denitrification nor phosphorous removal took place, and sufficient amounts of these should by inference have been available.

In summary, both reactors exhibited sub-optimal performance in terms of expected nutrient removal. Poor phosphorous removal was probably due to the high COD/P ratio coupled to the presence of nitrate in the anaerobic zone. The reactor with superior performance in terms of nutrient removal developed poor settling that was associated with decreased VSS, increased F/M, overgrowth of filament Type 021N and poor floc formation. The latter was most likely due to an increased availability of easily biodegradable compounds and possibly sulphides in the aerobic zone.

4.4 CAPE FLATS WWTP

4.4.1 PLANT CONFIGURATON AND OPERATING PARAMETERS

Cape Flats is a NDBEPR WWTP with a design capacity to treat approximately 200ML effluent per day. The plant has six identical bioreactors that operate in a 5-stage Bardenpho configuration with diffuse aeration. The raw influent consists of approximately 25% industrial effluent and 75% domestic effluent. The configuration is the same as that at Borchers quarry WWTP and is depicted by figure 4.3.1 in the previous section. The plant operates at a DO concentration of 2.5 mg/L in the first aerobic zone and 1 to 1.5 mg/L in the second aerobic zone and an aerobic mass fraction of 35%. The plant was often volumetrically overloaded in winter 2007 (see figure 4.4.1)

4.4.2 RESULTS

4.4.2 (a) Operational data

The influent (I) into the bioreactor G and bioreactor H is supernatant from the PST. The mixed liquor (ML) from both reactor G and reactor H flows into the clarifier. The effluent (E) is the supernatant from the clarifier.

*Note: breaks in the graph lines occur when no data was available

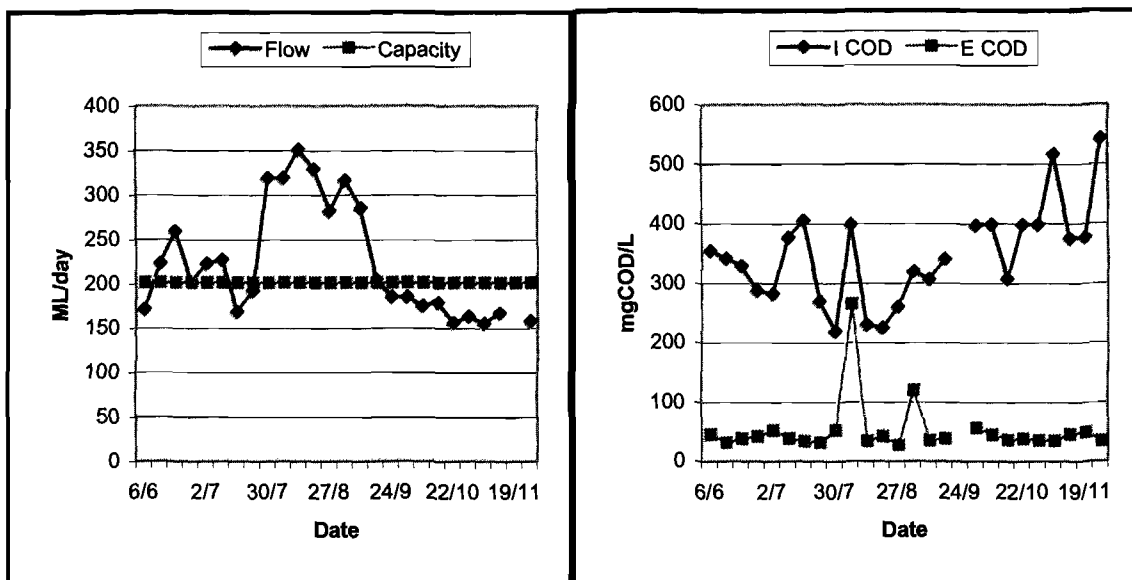


Figure 4.4.1 Graph depicting the overloading of Cape Flats WWTP in the winter of 2007

Figure 4.4.2 Graph depicting the influent (ICOD) and effluent COD (ECOD) levels at Cape Flats WWTP in 2007

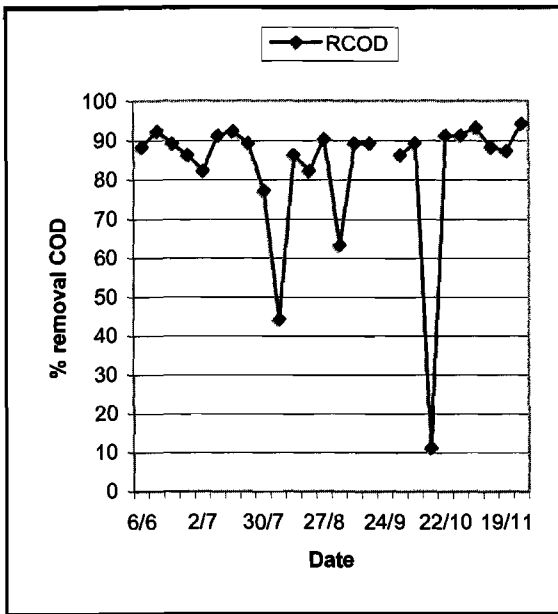


Figure 4.4.3 Graph depicting the removal efficiency of COD (RCOD) at Cape Flats WWTP in 2007.

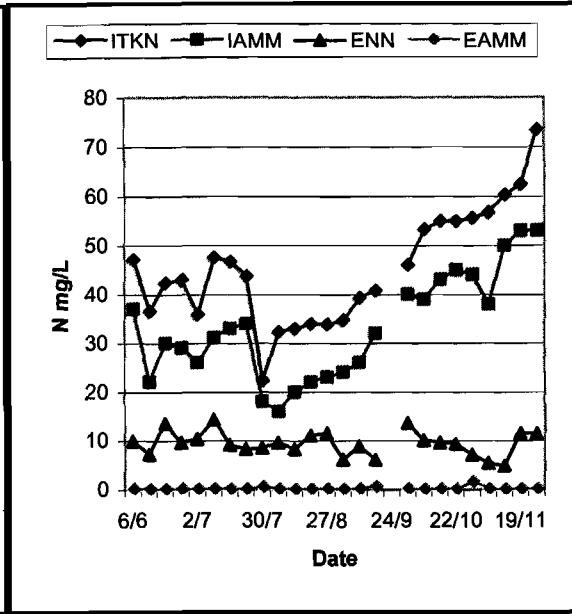


Figure 4.4.4 Graph depicting the influent TKN (ITKN) and ammonia (IAMM) as well as the effluent ammonia (EAMM) and nitrates/nitrites (ENN) at Cape Flats WWTP in 2007

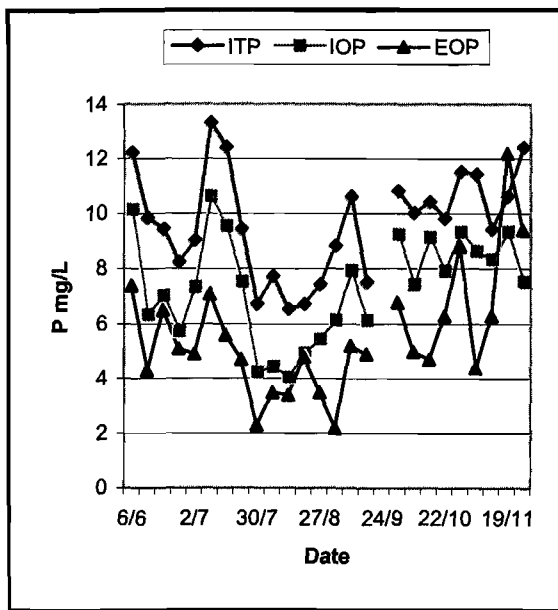


Figure 4.4.5 Graph depicting the levels of influent total phosphates (ITP) and influent σ -phosphates (IOP) as well as the effluent levels of σ -phosphates at Cape Flats WWTP in 2007

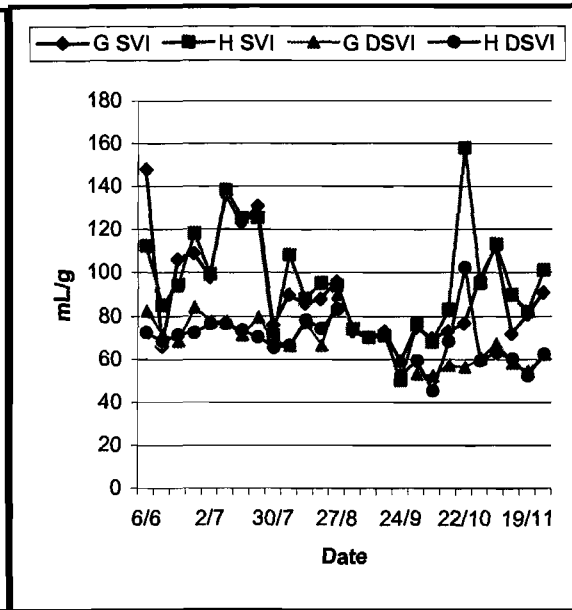


Figure 4.4.6 Graph depicting the values for the SVI (G SVI) and (H SVI) and DSVI (G DSVI and H CSVI) of the mixed liquor in reactor G and H respectively at the Cape Flats WWTP in 2007

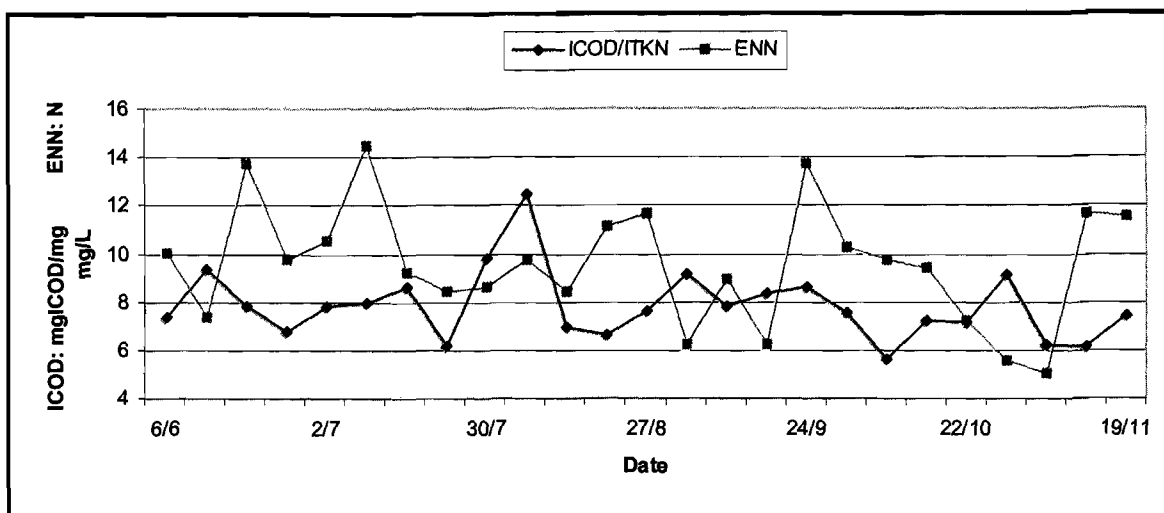


Figure 4.4.7 Relationship between the influent COD and TKN ratio (ICOD/TKN) and the effluent levels in nitrates/nitrites at the Cape Flats WWTP in 2007

| TABLE 4.4.1 FOOD TO MICROORGANISM (F/M) RATIO IN REACTOR G AND REACTOR H OF CAPE FLATS WWTP IN 2007 | | |
|-----------------------------------------------------------------------------------------------------|---------------------|---------------------|
| | REACTOR G | REACTOR H |
| June | 0.11mgCOD/mgVSS.day | 0.11mgCOD/mgVSS.day |
| July | 0.12mgCOD/mgVSS.day | 0.12mgCOD/mgVSS.day |
| August | 0.31mgCOD/mgVSS.day | 0.28mgCOD/mgVSS.day |
| October | 0.16mgCOD/mgVSS.day | 0.15mgCOD/mgVSS.day |
| November | 0.15mgCOD/mgVSS.day | 0.13mgCOD/mgVSS.day |

4.4.2 (b) Microscopic sludge analysis

This was performed monthly on the following dates: 18/6/07, 16/7/07, 20/8/07, 17/9/07, 22/10/07 and 19/11/07.

● Floc structure

Reactor G and reactor H

For both reactors, on each analysis, the structure was round, compact and firm and the average floc was medium in size (see figure 4.4.8).

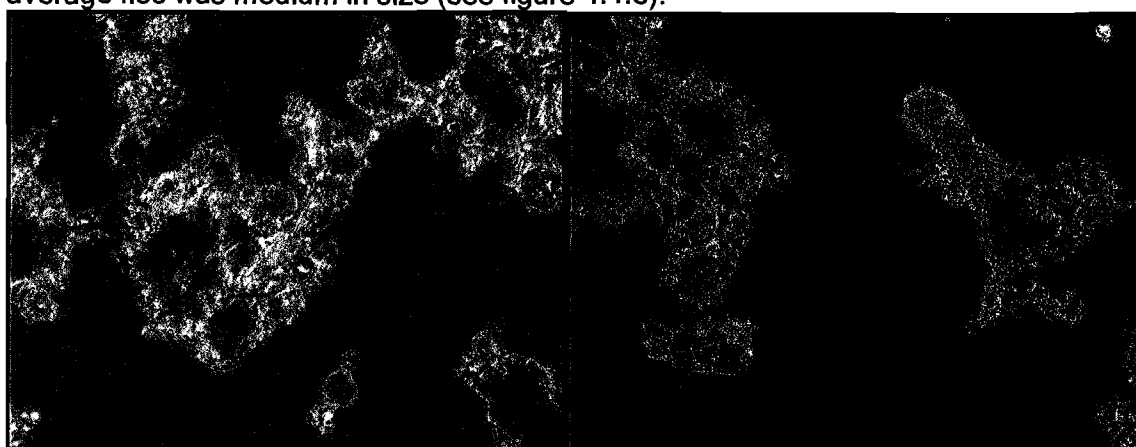


Figure 4.4.8 Wet mounts of mixed liquor from reactor H on the left and reactor G on the right from the Cape Flats WWTP in June 2007, showing the type of structure found in both reactors throughout the study

• Filament index

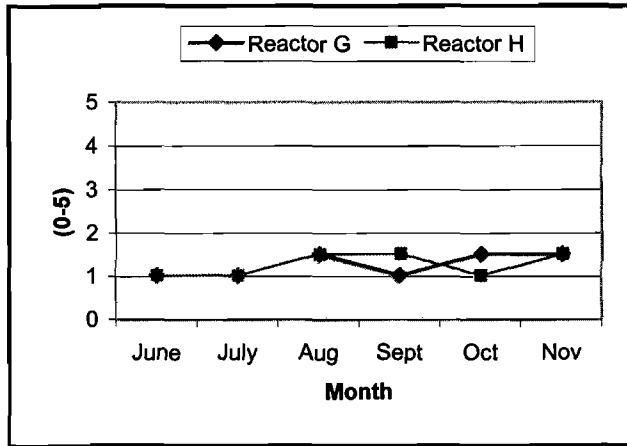


Figure 4.4.9 Graph depicting the low filament index values in both reactor G and H in 2007

| TABLE 4.4.2 DOMINANT AND SECONDARY FILAMENTOUS ORGANISMS IDENTIFIED IN THE MIXED LIQUOR OF REACTOR G (G) AND REACTOR H (H) AT BORCHERD'S QUARRY WWTP IN 2007 | | | | | | | | |
|--------------------------------------------------------------------------------------------------------------------------------------------------------------|---------|---------|---------|---------|---------|---------|---------------------------------|--------------------------------|
| AS DOMINANT FILAMENTS | | | | | | | | |
| | June | July | Aug | Sept | Oct | Nov | % Prevalence B and C (dominant) | % Prevalence B and C (overall) |
| Type 0092 | X (G,H) | X (G,H) | X (G,H) | X (G,H) | X (G,H) | X (G,H) | 100 | 100 |
| <i>H. hydrosis</i> | | | | | X (G) | X (G) | 17 | 50 |
| <i>N. limicola</i> | | | | | X (G) | X (G) | 17 | 33 |
| AS SECONDARY FILAMENTS | | | | | | | | |
| <i>H. hydrosis</i> | | | X (G,H) | | X (H) | X (H) | } | Also dominant filaments |
| <i>N. limicola</i> III | | | | | X (H) | X (H) | | |
| <i>M. parvicella</i> | | | X (G,H) | X (G,H) | X (G,H) | X (G,H) | 0 | 67 |
| Type 0041 | | | X (H) | X (G,H) | X (G,H) | X (G,H) | 0 | 58 |
| Type 1701 | | X (G,H) | X (G,H) | X (G,H) | | | 0 | 50 |
| Type 1851 | | X (G) | X (G,H) | X (H) | X (H) | X (H) | 0 | 50 |
| Type 021N | | | X (G) | X (G,H) | X (G,H) | X (H) | 0 | 50 |
| Type 0581 | X (G,H) | X (G,H) | X (G) | | | | 0 | 42 |
| <i>Flexibacter</i> spp. | | | | X (G,H) | | X (G,H) | 0 | 33 |

TABLE 4.4.3 PROTOZOA/METAZOA IDENTIFIED IN THE MIXED LIQUOR FROM REACTOR G AND REACTOR H AT CAPE FLATS WWTP IN 2007

| | | SCANTY | | | | | |
|-----------------------------|-----------------------------|------------|------------|------------|------------|------------|------------|
| | | June | July | Aug | Sept | Oct | Nov |
| Sessile ciliates | <i>Carchesium</i> spp. | X (G,H) | X (G) | | | | |
| | <i>Epistylis</i> spp. | | X (G,H) | X (G) | X (G) | X (G,H) | X (G) |
| | <i>Opercularia</i> spp. | | | X (G) | | | |
| | <i>Vaginicola</i> spp. | | X (G) | X (G,H) | X (G,H) | X (G,H) | |
| | <i>Vorticella</i> spp. | | X (G,H) | X (G,H) | X (G,H) | X (G,H) | X (G) |
| Crawling ciliates | <i>Aspidisca</i> spp. | X (H) | X (G) | X (G,H) | X (G,H) | X (G,H) | X (G,H) |
| | <i>Chilodonella</i> spp. | | X (G,H) | X (G) | | X (H) | |
| | <i>Trachelophyllum</i> spp. | | | X (G,H) | X (G,H) | X (H) | X (G,H) |
| | Unidentified | | | X (H) | | | |
| | <i>Blepharisma</i> spp. | | X (H) | | X (G) | X (H) | |
| Free-living ciliates | <i>Euplotes</i> spp. | | | | X (H) | | |
| | <i>Litonotus</i> spp. | X (H) | X (G,H) | X (G,H) | X (H) | | X (G,H) |
| | <i>Paramecium</i> spp. | | | | X (G) | | |
| | <i>Spirostomum</i> spp. | | | X(G,H) | X (G) | X (G) | X (G,H) |
| | Unidentified | | | X (G) | | | |
| Flagellates | <i>Bodo</i> spp. | X (H) | X (G,H) | X (G) | X (G) | X (H) | X (G,H) |
| | <i>Peranema</i> spp. | | X (G,H) | X (G,H) | | X (G,H) | X (H) |
| | <i>Monsiga</i> spp. | | | | | | X (G) |
| Amoebae | X (G,H) | X (G,H) | X (G,H) | X (G,H) | X (H) | X (H) | |
| Testate amoebae | | | | X (H) | | | |
| Heliozoa | | | | X (H) | X (H) | X (G) | |
| Rotifers | X (G,H) | X (G,H) | X (G,H) | X (G,H) | X (G,H) | X (G,H) | |
| Nematodes | | | | | X (H) | | |
| Tardigrades | | | X (G) | X (H) | X (G,H) | X (G) | |

| TABLE 4.4.3 (CONTINUED) PROTOZOA/METAZOA IDENTIFIED IN THE MIXED LIQUOR FROM REACTOR G AND REACTOR H AT CAPE FLATS WWTP IN 2007 | | | | | | |
|---------------------------------------------------------------------------------------------------------------------------------|-----------------------|----------|--|----------|----------|----------|
| Oligochaeta | | X (G) | | | | X (G) |
| MODERATE | | | | | | |
| Sessile ciliates | <i>Epistylis</i> spp. | | | X (H) | X (H) | X (H) |

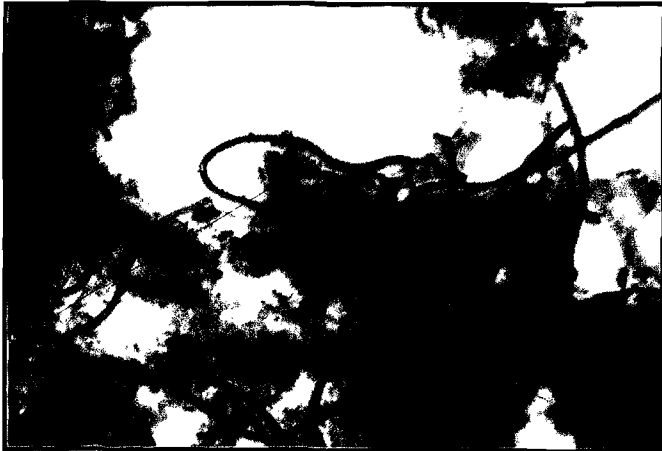


Figure 4.4.10 Neisser stain of Type 0092 from mixed liquor from reactor G at Cape Flats WWTP in June 2007.

4.4.3 DISCUSSION

Both reactor G and H of the Cape Flats WWTP showed good results for COD and ammonia removal for the entire study period, as can be seen from figures 4.4.2 and 4.4.3.

Figure 4.4.4 shows the presence of residual nitrate/nitrite, presumably due to incomplete denitrification in the anoxic zones. (The level of nitrate/nitrite in the clarifier supernatant was continuously around the 10mgN/L mark). Unfortunately, routine testing understandably does not include more detailed results on the nitrogenous compounds in various stages of the bioreactor. A plant of this configuration with two anoxic zones should achieve complete denitrification. The presence of readily available electron donor in the form of COD in the anoxic zone is necessary for this function (Tchobangous and Burton, 1991).

An article by Komorowska-Kaufman *et al.* (2006), describes evidence of the effects of wastewater characteristics and biological process conditions on nitrogen removal effectiveness in Bardenpho mode reactors. They found that denitrification was enhanced by the addition of extra carbon source in the form of acetate until the VFA/N ratio was equal to 1.67 mg acetate/mg N, after which no extra benefit was derived.

The authors refer to a study by Kujawa *et al.* (1996), who determined that a COD/N ratio of 8.4mgCOD/mgN was required for complete denitrification of wastewater. The ratio in

the influent at Cape Flats over the six-month study period ranged between 6.1 and 13.7, with a mean of 7.8mgCOD/mgTKN in the influent. Obviously requirements will differ from plant to plant because of substrate differences and the amount of utilization or absorption of COD taking place in the anaerobic zone preceding the first anoxic zone. In the second anoxic reactor in the Bardenpho system, denitrification takes place by the slower mechanism of endogenous respiration, and is significantly retarded by even low levels of DO. The effect of DO on nitrification is not significantly affected at high COD levels (Jeill *et al.*, 1999).

To ascertain whether lack of electron donor was the reason for incomplete denitrification, graph 4.4.7 was plotted comparing the influent COD/TKN ratio to effluent levels of nitrates and nitrites. There appears to be a link in that on most occasions, an increased trend on the influent COD/TKN ratio is followed by a decreased trend in the effluent nitrate/nitrite levels. It merits further work to ascertain whether this is indeed the case, as not only is the level of nitrates/nitrites in the effluent affected, but also possibly the phosphorous levels (see below). The other most obvious causes are:

- insufficient mixed liquor recycle from the aerobic to the first anoxic zone
- insufficient retention time in the anoxic zones
- carry over of DO in the mixed liquor recycle and/or from the aerobic to the second anoxic zone
- insufficient retention time in the anoxic zone

Phosphorous removal, graphically depicted in graph 4.4.5, was less than ideal for a system designed for this purpose. The presence of nitrates/nitrites in the clarifier effluent indicates that these nitrogenous compounds were probably also present in the RAS that enters the anaerobic zone.

Assuming this was the case, the anaerobic zone would become a de facto anoxic zone. This can be compared to exactly the opposite scenario experienced at the Bellville plant (MLE) that possibly resulted in unexpectedly good phosphorous removal. There is also a link between denitrification and phosphorous removal that was alluded to in the previous section (4.3, Borchers Quarry).

Carlsson *et al.* (1995), found that the concentration of VFA's was the most important parameter associated with the removal of phosphorous and that the relationship between the VFA/P ratio and removal of this nutrient was linear. The most important source of VFA's is the influent and this is supplemented by the formation of these fatty acids in the anaerobic zone by fermentation and hydrolysis. An important parameter to be considered is the retention time in the sewer - the longer the retention time, the higher the concentration of VFA's.

Under anoxic conditions, the ordinary heterotrophic population can compete with PAO's for VFA substrate. If sufficient VFA substrate and anaerobic conditions are present, biological phosphorous removal is chiefly dependent on conditions that result in PAO's having a competitive advantage over GAO's. This is described extensively in chapter 2. Both organism types compete for VFA's in the influent although substrate preferences in terms of acetate and propionate do differ. A ratio of P/COD of 10-20mgCOD/mgP favours the growth of PAO's. A ratio of 50mgCOD/mgP enhances the growth of GAO's (Liu *et al.*, 1997). The ratio encountered at Cape Flats ranged from 28.5-45.3mgCOD/mgP with a mean value of 35.8mgCOD/mgP. The assumption can thus be made that conditions did not favour either PAO's or GAO's. Thus the first steps to ascertain why phosphorous removal is inefficient would be to determine the levels of nitrates, nitrites and VFA's in the anaerobic zone.

The F/M ratio (shown in table 4.4.1), measured in mgCOD/mgMLVSS.day, was low in June, July, October and November and was slightly higher in August in both reactors.

From table 4.4.2, it can be seen that Type 0092, classed as low F/M by Jenkins *et al.* (2004), was dominant throughout the study in both reactors, which correlates well with the F/M results obtained. Co-dominant *N. limicola* III and *H. hydrosis* were only found in 2 of the 12 samples, but their presence in terms of F/M was not unexpected as they can both occur over a wide range of F/M values.

The assumption that denitrification was incomplete can be coupled to the hypothesis on AA bulking by Casey *et al.* (1999c) and lead to the possibility that conditions conducive to the overgrowth of AA filaments were present (the presence of sufficient levels of nitrates/nitrites in the mixed liquor entering the aerobic zones). However the DSVI values depicted in graph 4.4.6 and FI values depicted in graph 4.4.9 did not reflect the presence of bulking conditions at any time over the study period.

The floc character was as expected from a low loaded WWTP treating predominantly domestic effluent and with diffuse aeration (Eikelboom, 2000). The even spread of protozoa and metazoa in the sludge samples reflected the prevailing conditions of aerobic (ciliates) and anaerobic/anoxic (flagellates, amoeba). The presence of bacteriovorous free-living ciliates reflected the finding of free-living cells in many samples. The presence of rotifers, nematodes, tardigrades and oligochaete worms similarly reflected the generally long sludge ages (Eikelboom, 2000).

In summary, the reactors used in the study did not exhibit bulking tendencies in terms of elevated DSVI and/or FI from June to September 2007. This was despite the probability that there were nitrates/nitrites present in the anoxic zone preceding the aerobic zone. Nutrient removal was poor, most likely due to insufficient VFA's in the anaerobic zone, and/or too much nitrate in the anaerobic zone and/or lack of electron donor in the first anoxic zone.

4.5 KRAAIFONTEIN WWTP

4.5.1 PLANT CONFIGURATION AND OPERATING PARAMETERS

Kraaifontein is a relatively small NDEBPR WWTP for the treatment of domestic wastewater. The configuration of the plant is classic UCT (see figure 4.1.2) and surface aeration is employed. There is only one bioreactor and the aerobic, anoxic and anaerobic mass fractions are 59%, 30% and 11% respectively and the average DO concentration is 2.8mg/L. Influent is first settled in PST's before entering the bioreactor.

4.5.2 RESULTS

4.5.2 (a) Operational data

The influent (I) is the supernatant from the PST's combined.

The mixed liquor (ML) results are from the bioreactor.

The effluent (E) is the averaged results from the supernatant of three clarifiers.

*Note: breaks in the graph lines occur when no data was available

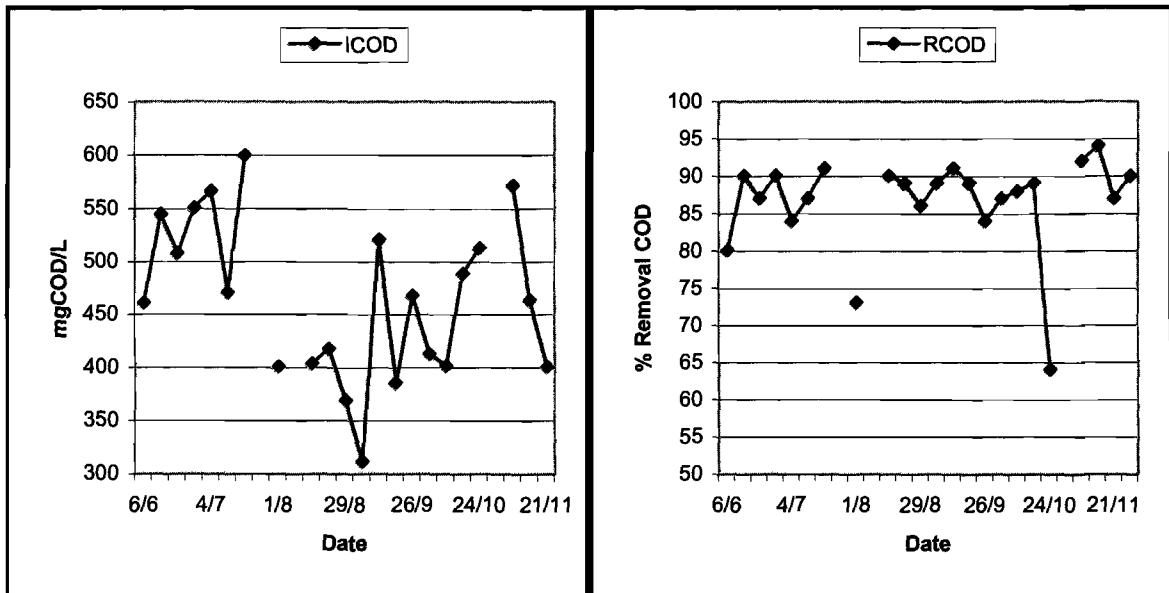


Figure 4.5.1 Graph depicting the influent COD levels at Kraaifontein WWTP in 2007

Figure 4.5.2 Graph depicting the COD removal efficiency (RCOD) at Kraaifontein WWTP in 2007

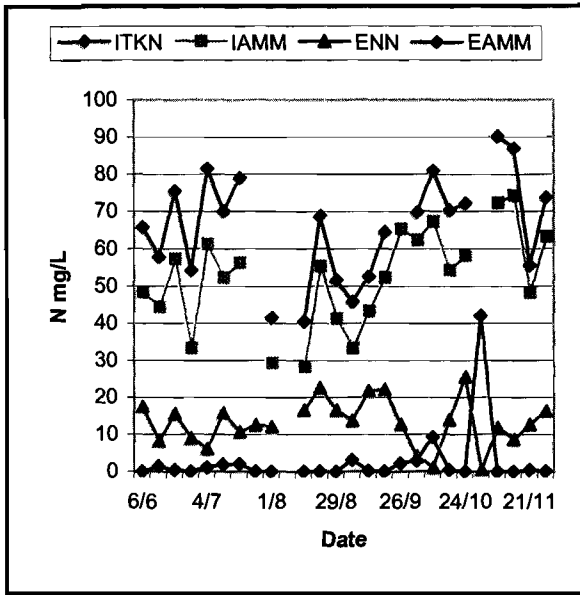


Figure 4.5.3 Graph depicting the influent TKN (ITKN), influent ammonia (IAMM), effluent nitrates/nitrites (ENN) and effluent ammonia (EAMM) levels at Kraaifontein WWTP in 2007

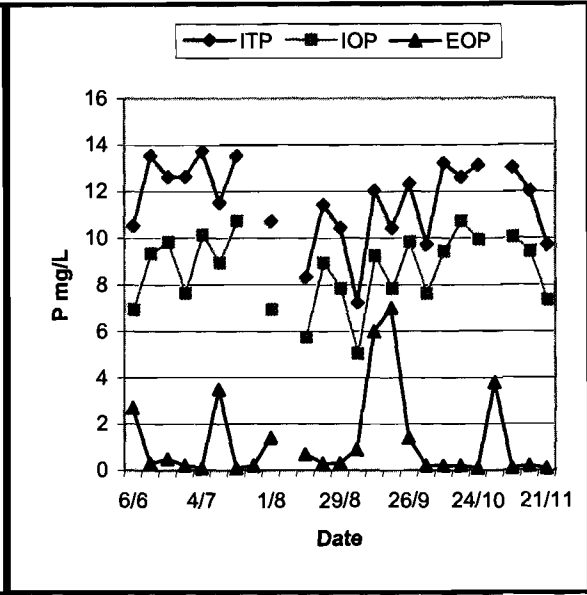


Figure 4.5.4 Graph depicting the influent total phosphate (ITP), influent σ -phosphate and effluent σ -phosphate levels at Kraaifontein WWTP in 2007

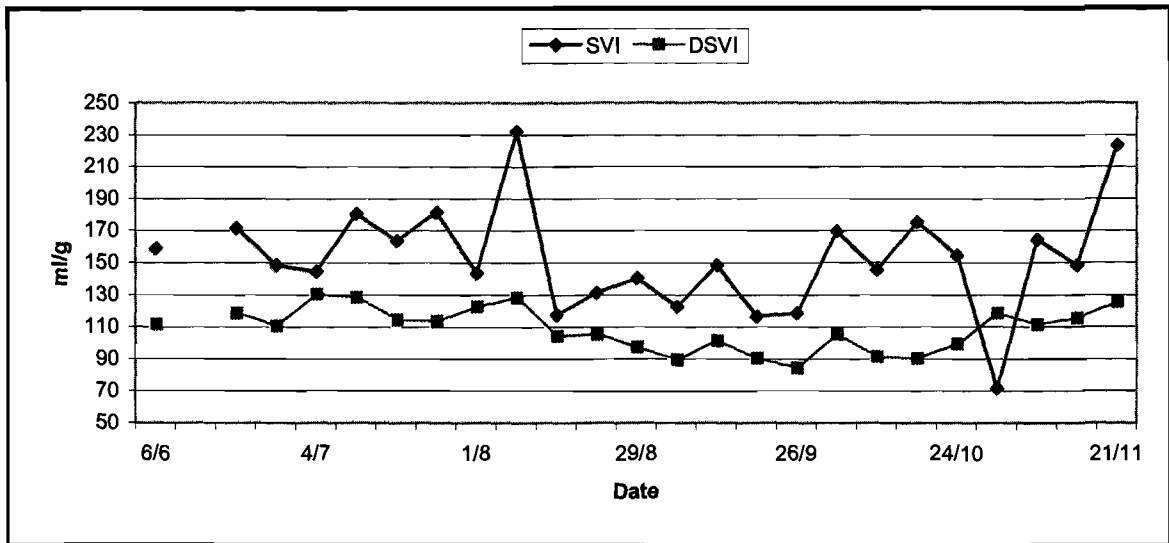


Figure 4.5.5 Graph depicting the SVI and DSVI values obtained at Kraaifontein WWTP in 2007

| | |
|-----------|----------------------|
| June | 0.08 mgCOD/mgVSS.day |
| July | 0.31 mgCOD/mgVSS.day |
| September | 0.19 mgCOD/mgVSS.day |
| October | 0.16 mgCOD/mgVSS.day |
| November | 0.14 mgCOD/mgVSS.day |

4.5.2(b) Microscopic sludge analysis

This was performed monthly on the following dates: 28/6/07, 26/7/07, 23/8/07, 20/9/07, 25/10/07 and 22/11/07.

- **Floc structure**

The flocs were open in structure and firm in strength for the duration of the study. The shape of the flocs differed from round, in June, August and November, irregular in July and both irregular and round in September and November. The size of the flocs was medium in June, small (pin flocs) and medium in July, medium and large in August, September and October and medium again in November.

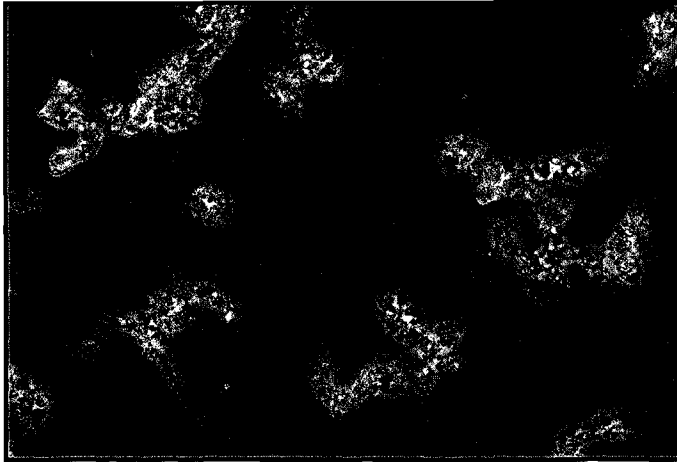


Figure 4.5.6 Micrograph of wet mount of mixed liquor showing irregular floc structure at Kraaifontein WWTP in September 2007

- **Diversity**

Diversity was moderate to low for the duration of the study, and there were a moderate number of monocolonies, staining principally as GAO bacteria (see section 4.5.3 and figure 4.5.9).

- **Filament index**

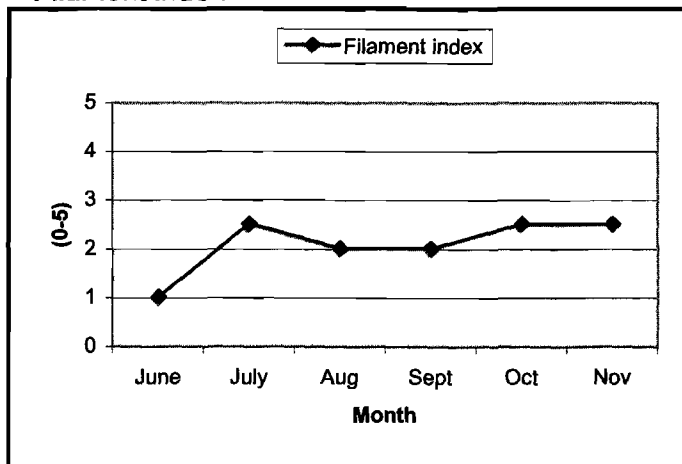


Figure 4.5.7 Graph showing the FI values obtained from the mixed liquor at Kraaifontein WWTP in 2007

TABLE 4.5.2 DOMINANT AND SECONDARY FILAMENTOUS ORGANISMS IDENTIFIED FROM THE MIXED LIQUOR AT KRAAIFONTEIN WWTP IN 2007

| AS DOMINANT FILAMENTS | | | | | | | | |
|------------------------|------|------|-----|------|-----|-----|-------------------------|-------------------------|
| | June | July | Aug | Sept | Oct | Nov | % Prevalence (dominant) | % Prevalence (overall) |
| Type 0092 | X | X | X | X | X | X | 100 | 100 |
| <i>M. parvicella</i> | | | | X | X | X | 50 | 100 |
| AS SECONDARY FILAMENTS | | | | | | | | |
| <i>M. parvicella</i> | X | X | X | | | | } | Also dominant filaments |
| Type 1851 | X | X | X | X | X | X | 0 | 100 |
| Type 0041 | | | X | X | X | X | 0 | 67 |
| Type 021N | | X | | | | | 0 | 17 |
| Type 1863 | | | | | | X | 0 | 17 |

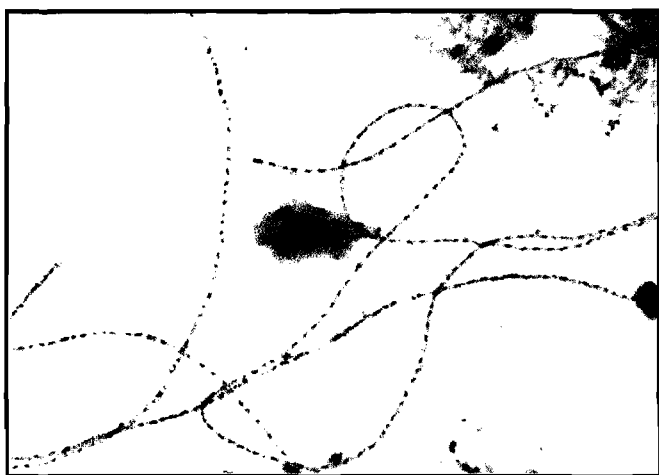


Figure 4.5.8 Neisser stain of *Microthrix parvicella* from mixed liquor sample taken at Kraaifontein WWTP in October 2007.

TABLE 4.5.3 PROTOZOA/METAZOA IDENTIFIED IN THE MIXED LIQUOR FROM KRAAIFONTEIN WWTP IN 2007

| SCANTY | | | | | | | |
|----------------------|-----------------------------|------|------|-----|------|-----|-----|
| | | June | July | Aug | Sept | Oct | Nov |
| Sessile ciliates | <i>Carchesium</i> spp. | X | | X | X | X | |
| | <i>Epistylis</i> spp. | | | X | X | X | X |
| | <i>Vorticella</i> spp. | X | | X | | X | X |
| Crawling ciliates | <i>Aspidisca</i> spp. | X | | X | X | X | X |
| | <i>Chilodonella</i> spp. | | | X | | X | X |
| | <i>Trachelophyllum</i> spp. | | | X | X | | |
| Free-living ciliates | <i>Colpidium</i> spp. | X | | | | | |
| | Unidentified | | | X | X | | |
| Flagellates | <i>Bodo</i> spp. | | | | X | X | X |
| | <i>Peranema</i> spp. | | X | | X | X | |
| | Unidentified | | | X | X | | |
| Amoebae | | | | X | | X | X |
| Testate amoebae | <i>Arcella</i> spp. | | | | | | |
| Heliozoa | | | | X | | | |

| TABLE 4.5.3 (CONTINUED) PROTOZOA/METAZOA IDENTIFIED IN THE MIXED LIQUOR FROM KRAAIFONTEIN WWTP IN 2007 | | | | | | | |
|--------------------------------------------------------------------------------------------------------|-----------------------------|------|------|-----|------|-----|-----|
| | | June | July | Aug | Sept | Oct | Nov |
| Rotifers | | | X | | | | |
| Nematodes | | | | | X | X | X |
| MODERATE | | | | | | | |
| Sessile ciliates | <i>Epistylis</i> spp. | | X | | | | |
| Crawling ciliates | <i>Aspidisca</i> spp. | | X | | | | |
| | <i>Trachelophyllum</i> spp. | | X | | | | |
| Free-living ciliates | <i>Euplotes</i> spp. | | | X | | | |
| Flagellates | <i>Peranema</i> spp. | | | | | | X |
| Amoebae | | | | | X | | |

4.5.3 DISCUSSION

By studying figures 4.5.1, 4.5.2 and 4.5.3, it can be seen that the removal of ammonia and COD at Kraaifontein WWTP was relatively efficient over the study period, except for at the end of October, when there were some high aberrant results for COD and suspended solids in the clarifier effluent. The problem may have been linked to the clarifier and not the bioreactor.

There was residual nitrate/nitrite in the clarifier effluent for most of the trial period. On the odd occasion when nitrates/nitrites were absent, there was a link to an increase in ammonia levels, which is demonstrated by figure 4.5.3. This infers that nitrification of ammonia is usually complete, but the amount of nitrates/nitrites thus formed exceed denitrification potential of this system, resulting in the presence of residual nitrate/nitrites. However, when nitrification is incomplete, the conversion of ammonia into nitrates/nitrites is not achieved to the same degree and the biological denitrification system can fully cope with the lower levels of nitrate/nitrite.

The possible reasons for lack of denitrification have already been elucidated upon in the previous sections of this chapter (4.2.3 and 4.3.3). Regarding the lack of electron donor, the influent COD/TKN ratio ranged from 5.0-10.2mgCOD/mgTKN, with a mean of 7.2mgCOD/mgTKN. The mean was thus below the value of 8.4mgCOD/mgTKN reported by Kujawa *et al.* (1996) as being necessary for complete denitrification. There was no obvious correlation between this ratio and effluent nitrate/nitrite values, but this does not necessarily rule out the lack of electron acceptor as being the major cause of complete denitrification. The nitrate/nitrite levels in the clarifier effluent only serve as a guideline under these circumstances and do not necessarily reflect the true daily reactor conditions.

The erratic nature of phosphorus removal efficiency is reflected in figure 4.5.4. There were periods of almost complete or complete removal interspersed with poor removal, especially around the end of August. Ekama *et al.* (1984) found that with a TKN/COD ratio of <0.08, no external energy source was necessary for complete denitrification to occur. In a UCT system, a TKN/COD ratio of >0.14 usually results in a nitrate being carried over from the primary anoxic reactor to the anaerobic reactor with a resultant negative effect on phosphorous removal. In fact, at this level, Ekama *et al.* (1984) found it unlikely that BPR would be achieved with any consistency because of insufficient denitrification. The TKN/COD range at Kraaifontein from June to November 2007 was 0.098-0.201mgN/mgCOD, with a mean value of 0.14mgN/mgCOD.

To prevent carry over of nitrates, the practice is to increase the mixed liquor recycle from the aerobic zone and effectively increase the anoxic retention time. The presence of varying amounts of nitrate in the mixed liquor recycle to the anaerobic zone may explain the erratic nature of phosphorous removal, but the fact that a moderate number of GAO's were seen in the mixed liquor presupposes that conditions were created that allowed these organisms to compete with the PAO's.

The COD/P ratio ranged between 27.6-48.6mgCOD/mgP with a mean of 39.8mgCOD/mgP. At this level, the growth of neither the GAO's nor the PAO's should be enhanced over the other (Liu *et al.*, 1997).

However, there were moderate numbers of monoclonies in all samples. These stained principally as GAO bacteria (see figure 4.5.9 below) and the phenomenon is discussed in more detail later in this chapter under section 4.9.

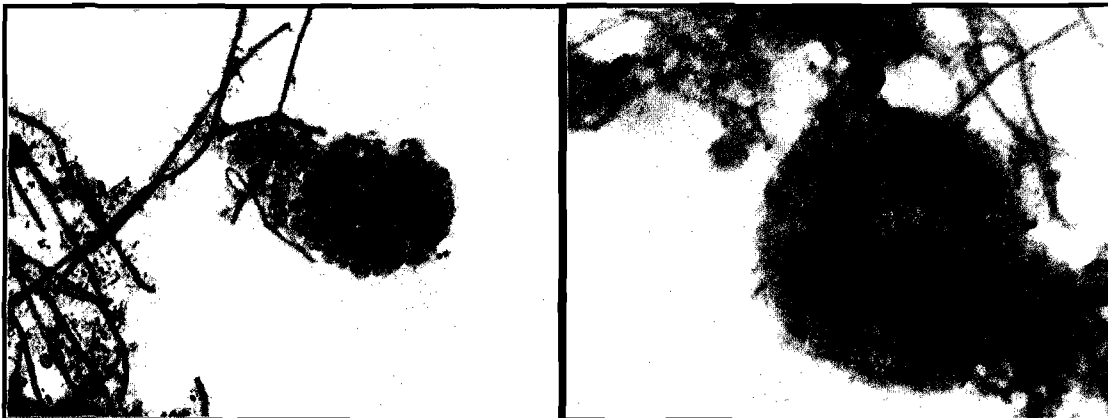


Figure 4.5.9 Gram stain of GAO's from mixed liquor from Kraaifontein WWTP in August 2007 (left) and Neisser stain of GAO's from mixed liquor from Kraaifontein WWTP in August 2007 (right).

The floc structure of the mixed liquor was open, which is a characteristic of surface aeration (Eikelboom, 2000). Apart from the flocs occasionally being irregular in shape, and the presence of pin flocs June, the overall floc character was fair.

Both dominant filaments, namely Type 0092 and *M. parvicella* are classified by Jenkins *et al.* (2004) as "low F/M". This correlates well with the calculated values given in table 4.5.1. The value in July was higher than the other months of the study, and in this month, there was a concomitant increase in the number of sessile and crawling ciliates. This can be seen by examining figure 4.5.3. *Epistylis* spp., *Aspidisca* spp. and *Trachelophyllum* spp. were all found in moderate amounts in this month. However, on the whole, the even spread of ciliates and the presence of amoeba, rotifers and nematodes reflect the varied DO levels, lack of high levels of toxic substances and long sludge age (Eikelboom, 2000).

It can be speculated that the presence of residual amounts of nitrate/nitrite would be conducive to AA bulking using the metabolic hypothesis of Casey *et al.* (1999). In fact, the indicator FI and the DSVI values shown in figure 4.5.7 and 4.5.5 respectively were never a cause for concern.

4.6 MACASSAR WWTP

4.6.1 PLANT CONFIGURATION AND OPERATING PARAMETERS

Macassar WWTP has a design capacity of 34ML/day. The plant is equipped with two Carousel® systems operating in UCT mode (figures 4.1.1 and 4.1.2). The influent is primarily of domestic origin. DO concentrations in the reactors should range from 0.2-0.9mg/L. The plant experiences intermittent problems with scum formation.

4.6.2 RESULTS

4.6.2 (a) Operational data

The influent (I) is the raw wastewater.

The mixed liquor (ML) results are from the two bioreactors differentiated as reactor 1 and reactor 2. The mixed liquor from these reactors flows into separate the clarifiers, also named 1 and 2.

The effluent (E) is the supernatant from clarifiers.

*Note: breaks in the graph lines occur when no data was available

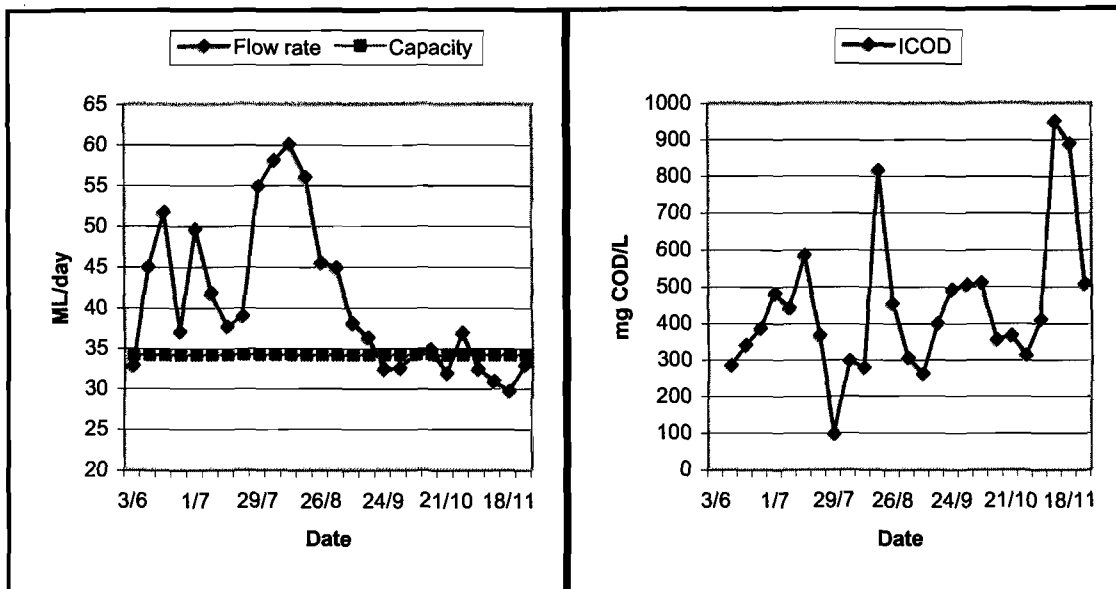


Figure 4.6.1 Graph depicting the design capacity and the actual flow rate into the Macassar WWTP, showing the theoretical gross volumetric overloading in 2007

Figure 4.6.2 Graph depicting the influent COD (ICOD) levels at Macassar WWTP in 2007

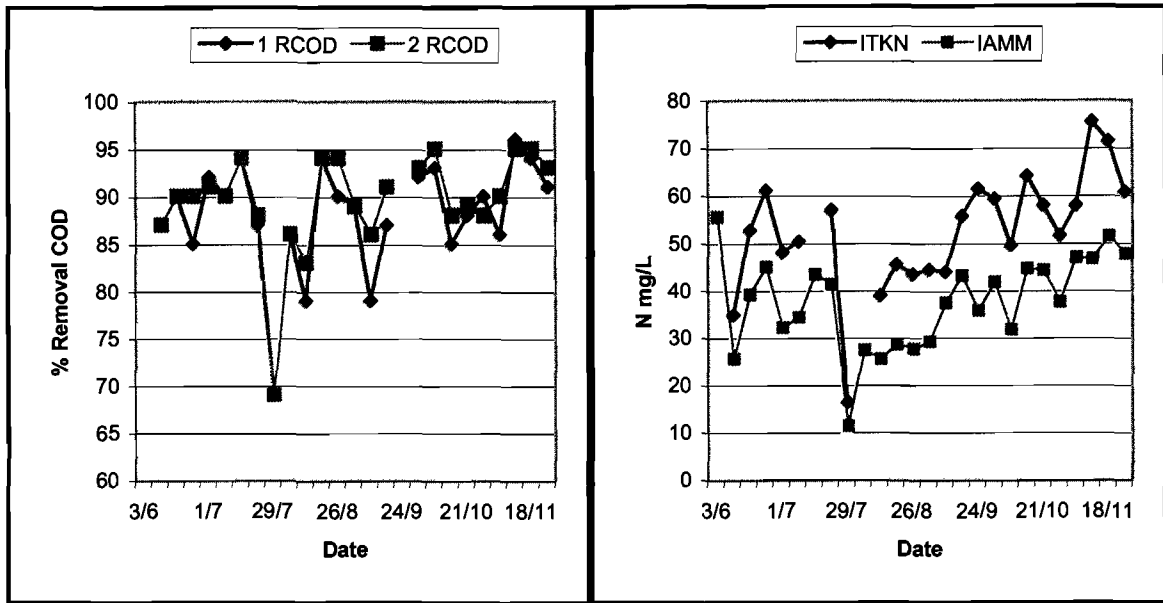


Figure 4.6.3 Graph depicting the COD removal efficiency of reactor 1 (1 RCOD) and reactor 2 (2 RCOD) at Macassar WWTP in 2007

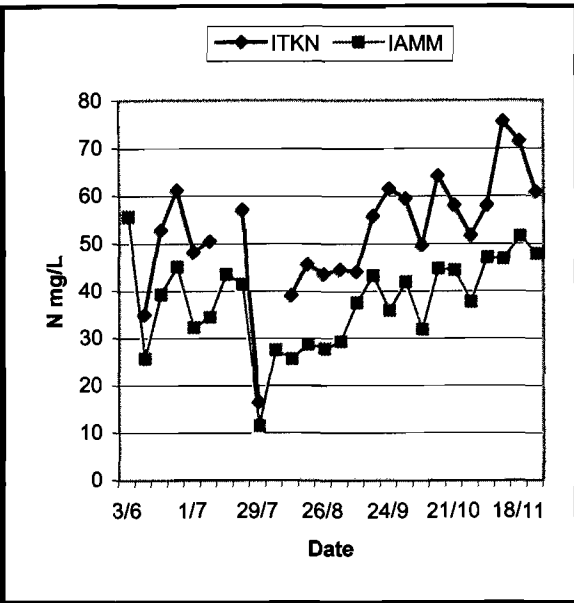


Figure 4.6.4 Graph depicting the influent TKN (ITKN) and influent ammonia (IAMM) levels at Macassar WWTP in 2007

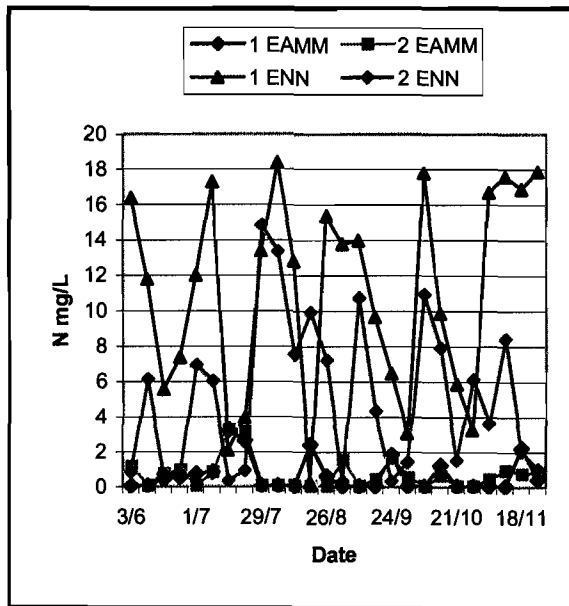


Figure 4.6.5 Graph depicting the effluent ammonia levels from clarifier 1 (1 EAMM) and clarifier 2 (2 EAMM) and the effluent nitrates/nitrites from clarifier 1 (1 ENN) and clarifier 2 (2 ENN) at Macassar WWTP in 2007

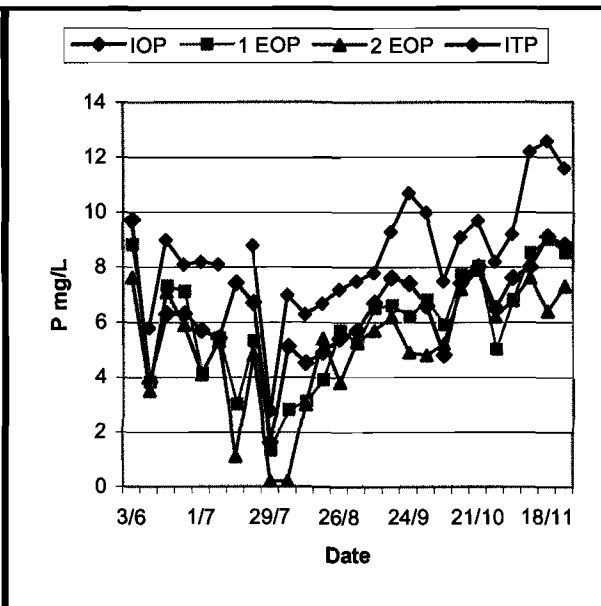


Figure 4.6.6 Graph depicting the influent σ -phosphates (IOP) and total phosphates (ITP) and the effluent σ -phosphates from clarifier 1 (1 EOP) and clarifier 2 (2 EOP) at Macassar WWTP in 2007

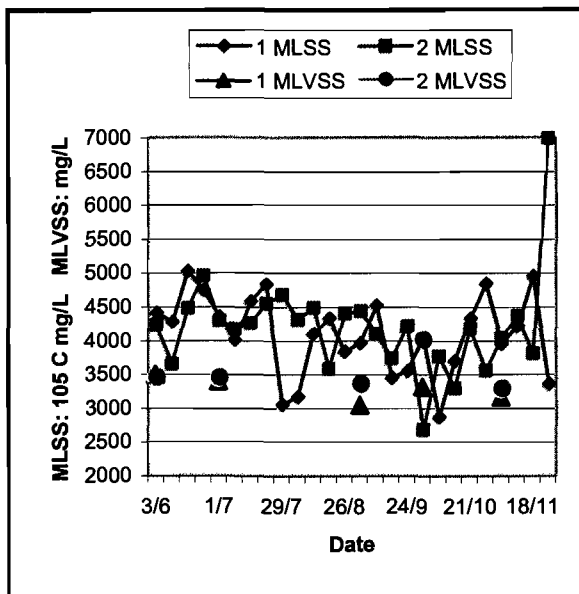


Figure 4.6.7 Mixed liquor suspended solids from reactor 1 (1 MLSS) and reactor 2 (2 MLSS) and mixed liquor volatile suspended solids from reactor 1 (1 MLVSS) and reactor 2 (2 MLVSS) at Macassar WWTP in 2007

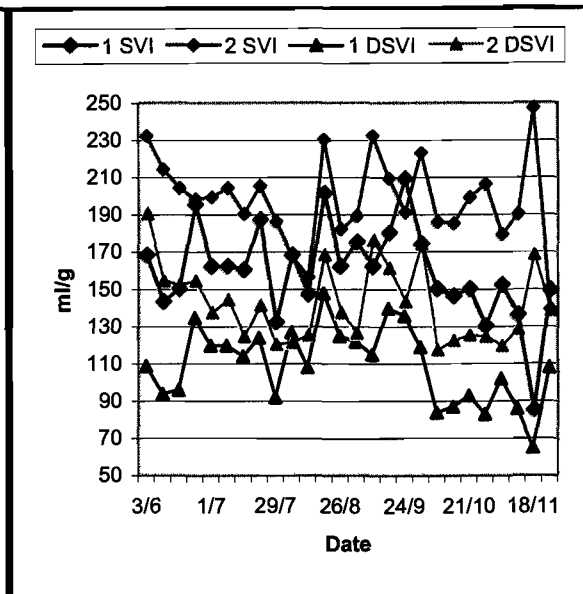


Figure 4.6.8 Sludge volume index from reactor 1 (1 SVI) and reactor 2 (2 SVI) and dissolved sludge volume index from reactor 1 (1 DSVI) and reactor 2 (2 DSVI) at Macassar WWTP in 2007

| TABLE 4.6.1 FOOD TO MICROORGANISM (F/M) RATIO IN REACTOR 1 AND REACTOR 2 OF MACASSAR WWTP IN 2007 | | |
|---------------------------------------------------------------------------------------------------|----------------------|----------------------|
| | REACTOR 1 | REACTOR 2 |
| July | 0.14 mgCOD/mgVSS.day | 0.14 mgCOD/mgVSS.day |
| September | 0.09 mgCOD/mgVSS.day | 0.08 mgCOD/mgVSS.day |
| October | 0.10 mgCOD/mgVSS.day | 0.08 mgCOD/mgVSS.day |
| November | 0.09 mgCOD/mgVSS.day | 0.08 mgCOD/mgVSS.day |

4.6.2 (b) Microscopic sludge analysis

This was performed monthly on the following dates: 26/6/07, 23/7/07, 28/8/07, 24/9/07, 22/10/07 and 26/11/07.

- Floc structure

Reactor 1

In June, the flocs were irregular in shape, open in structure and firm in strength. In July, October and November, the flocs were round in shape, open in structure and firm in strength. In August and September the flocs were round in shape, compact in structure and firm in strength. The flocs were medium-sized for the duration of the study.

Reactor 2

In June, July, September, October and November the flocs were round in shape, open in structure and firm in strength. In August, the flocs were round in shape, compact in structure and firm in strength. The flocs were medium-sized for the duration of the study.

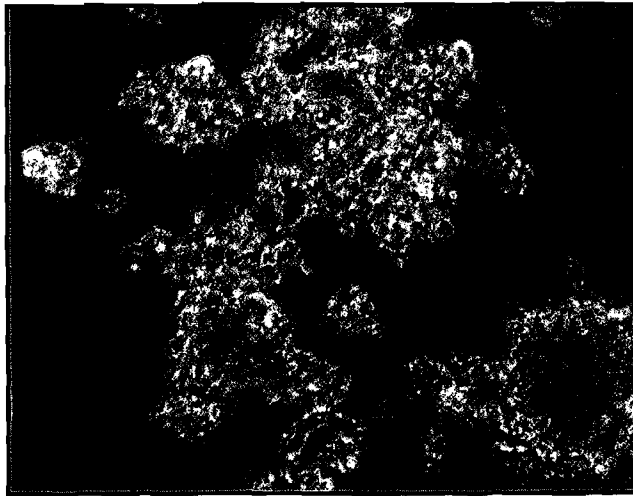


Figure 4.6.9 Wet mount of mixed liquor from reactor 2 at Macassar WWTP in July showing round, open and firm floc structure

● Diversity

Microscopic examination revealed moderate diversity in all samples from both reactors. Scanty monocolonies were present in all samples except those from reactor 1 in June, August, and October and reactor 2 in June, October and November, when no monocolonies were observed.

● Filament index

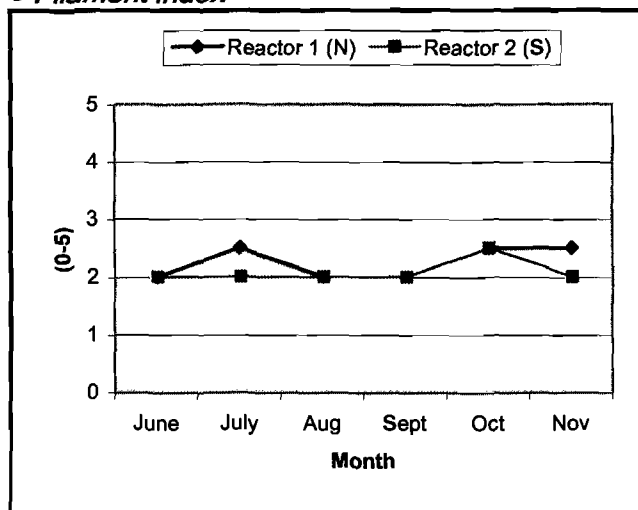


Figure 4.6.10 Graph depicting the filament index of reactor 1 and reactor 2 at Macassar WWTP in 2007. In August and September the FI for both reactors was the same.

TABLE 4.6.2 DOMINANT AND SECONDARY FILAMENTOUS ORGANISMS IDENTIFIED FROM THE MIXED LIQUOR OF REACTOR 1 AND REACTOR 2 AT MACASSAR WWTP IN 2007

| AS DOMINANT FILAMENTS | | | | | | | | |
|------------------------|----------|------------|------------|------------|------------|------------|---------------------------------------|--------------------------------------|
| | June | July | Aug | Sept | Oct | Nov | % Prevalence 1 and 2 (dominant) | % Prevalence 1 and 2 (overall) |
| <i>M. parvicella</i> | | X (1,2) | X (1,2) | X (1,2) | X (1) | X (1,2) | 75 | 83 |
| Actinomycetes | | X (2) | | X (1,2) | X (1,2) | X (2) | 50 | 92 |
| Type 0092 | X (2) | X (2) | | X (2) | | X (1) | 33 | 100 |
| Type 1851 | | | | | X (1) | X (1,2) | 25 | 67 |
| <i>H. hydrosis</i> | | X (2) | | X (2) | | | 17 | 75 |
| Type 0041 | | X (2) | | | | | 8 | 83 |
| AS SECONDARY FILAMENTS | | | | | | | | |
| <i>M. parvicella</i> | | | | | X (2) | | } Also dominant filaments | |
| Actinomycetes | X (1) | X (1) | X (1,2) | | | X (1) | | |
| Type 0092 | X (1) | X (1) | X (1,2) | X (1) | X (1,2) | X (2) | | |
| Type 1851 | | | X (1,2) | X (1,2) | X (2) | | | |
| <i>H. hydrosis</i> | | | X (1,2) | X (1) | X (1,2) | X (1,2) | | |
| Type 0041 | X (2) | | X (1,2) | X (1,2) | X (1,2) | X (1,2) | | |
| Type 021N | | X (2) | X (1,2) | X (1) | X (1) | X (1,2) | | 0 |
| Type 0581 | | | | | | X (1) | 0 | 8 |
| Type 0914 | X (1) | | | | | | 0 | 8 |
| Type 1701 | | | X (1) | | | | 0 | 8 |

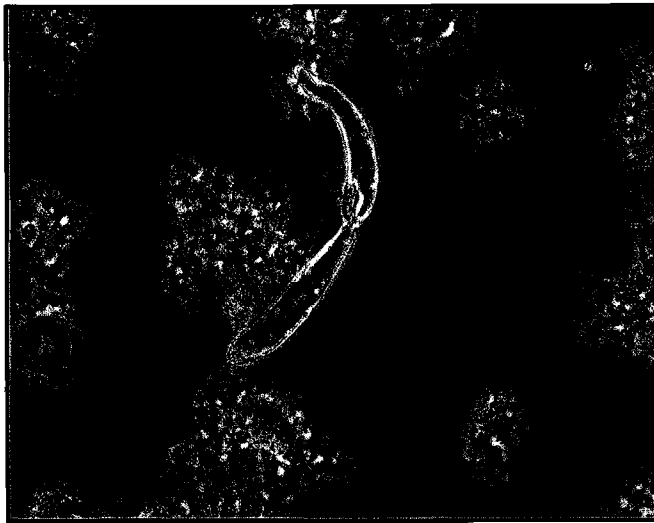


Figure 4.6.11 Wet mount of mixed liquor from reactor 1 at Macassar WWTP in November 2007 showing *Spirostomum* spp., apparently mating.

| TABLE 4.6.3 PROTOZOA/METAZOA IDENTIFIED IN THE MIXED LIQUOR FROM REACTOR 1 AND REACTOR 2 AT MACASSAR WWTP IN 2007 | | SCANTY | | | | | |
|-------------------------------------------------------------------------------------------------------------------|-----------------------------|------------|----------|------------|------------|------------|------------|
| | | June | July | Aug | Sept | Oct | Nov |
| Sessile ciliates | <i>Epistylis</i> spp. | | X (2) | X (1,2) | | | X (1) |
| | <i>Opercularia</i> spp. | X (1) | X (1) | | | | |
| | <i>Vaginicola</i> spp. | | | | | X (1) | |
| | <i>Vorticella</i> spp. | X (1) | | X (1,2) | X (1,2) | X (1,2) | X (1,2) |
| Crawling ciliates | <i>Aspidisca</i> spp. | X (1) | | X (1,2) | X (1) | X (1) | X (2) |
| | <i>Chilodonella</i> spp. | | X (2) | X (1,2) | | | |
| | <i>Trachelophyllum</i> spp. | X (1) | X (1) | X (2) | | | |
| Free-living ciliates | <i>Blepharisma</i> spp. | | | | | X (1) | |
| | <i>Colpidium</i> spp. | X (1) | | | X (1,2) | X (1,2) | X (1,2) |
| | <i>Euplotes</i> spp. | | | X (2) | | | |
| | <i>Litonotus</i> spp. | | X (2) | X (2) | | X (1) | X (2) |
| | <i>Paramecium</i> spp. | | | | | X (1,2) | |
| | <i>Spirostomum</i> spp. | X (1,2) | | | | X (1) | X (2) |

TABLE 4.6.3 (CONTINUED) PROTOZOA/METAZOA IDENTIFIED IN THE MIXED LIQUOR FROM REACTOR 1 AND REACTOR 2 AT MACASSAR WWTP IN 2007

| | | June | July | Aug | Sept | Oct | Nov |
|----------------------|-------------------------|------------|------------|------------|----------|------------|------------|
| | Unidentified | X (2) | X (2) | X (2) | | | |
| Flagellates | <i>Bodo</i> spp. | | | X (1,2) | | X (1,2) | X (2) |
| | <i>Peranema</i> spp. | | | X (1) | | | |
| | <i>Trepomonas</i> spp. | | | X (1) | | | |
| Amoebae | | X (1,2) | X (1,2) | X (1) | X (1) | X (1) | X (1,2) |
| Rotifera | | | X (1,2) | X (1,2) | | X (1) | X (1,2) |
| Nematodes | | | | X (1,2) | X (1) | X (1) | X (1,2) |
| MODERATE | | | | | | | |
| Attached ciliates | <i>Vaginicola</i> spp. | | | | | X (2) | |
| | <i>Vorticella</i> spp. | | X (1,2) | | | | |
| Crawling ciliates | <i>Aspidisca</i> spp. | | X (1) | | | | X (1) |
| Free-living ciliates | <i>Euplotes</i> spp. | | X (1) | | | | |
| | <i>Spirostomum</i> spp. | | | | | | X (1) |
| Flagellates | <i>Bodo</i> spp. | | | | | X (1) | |
| Amoebae | | | | X (2) | | | |
| Rotifera | | X (1,2) | | | X (1) | | |
| NUMEROUS | | | | | | | |
| Crawling ciliates | <i>Aspidisca</i> spp. | | | | X (2) | | |
| Flagellates | <i>Pleuromonas</i> spp. | | | | X (1) | | |

4.6.3 DISCUSSION

Macassar WWTP was severely overloaded in terms of design capacity, which is clearly depicted in figure 4.6.1. However, due to the low influent COD, the removal of this substrate was efficient (figures 4.6.2 and 4.6.3). The influent COD was consistently lower than that of most other WWTP's included in the study, perhaps due to the lack of industrial component.

Nitrification was satisfactory but denitrification was often poor, with erratic results that can be seen by perusing figures 4.6.4 and 4.6.5. The levels of residual nitrates/nitrites were extremely erratic (0-15mg/L). There was no obvious correlation between influent COD/TKN ratios and effluent nitrate/nitrite levels. This fact does not preclude the possibility of electron donor deficiency in the anoxic zone being the reason for poor denitrification, as from the values in table 4.6.1, it can be seen that the F/M ratio (mgCOD/mgMLVSS.day) was constantly low in both reactors:

High levels of nitrates/nitrites were often, but not exclusively found when total nitrification (zero ammonia) had taken place. Again, the inference in these circumstances is that when nitrification is incomplete, for whatever reason, the biological denitrification system can fully cope with the lower levels of nitrate/nitrite produced. The influent COD/TKN ratio ranged from 5.5-17.9mgCOD/mgTKN with a mean value of 8.3mgCOD/mgTKN. The latter is close to the value of 8.4mgCOD/mgTKN reported by Liu *et al.* (1996) as being necessary for complete denitrification. In some instances neither complete nitrification nor complete denitrification was achieved. Although nitrification was similar in both reactors, denitrification was usually markedly better in reactor 2.

The phosphorous removal efficiency of the WWTP was extremely poor, despite the relatively low levels in the influent. This can be seen in figure 4.6.6. Possible reasons have been outlined in previous sections of chapter 4 and are applicable here, especially lack of VFA's or increased COD/P ratio in the influent and/or the presence of nitrate in the anaerobic zone. The fact that reactor 2, with consistently less nitrates/nitrites measured in the effluent also showed consistently better phosphorous removal leads to the assumption that the presence of nitrates in the anaerobic zone played at least a partial role. The COD/P ratio ranged from 33.2-77.6mgCOD/mgP with a mean of 50.9mgCOD/mgP. According to Liu *et al.* (1997), this figure suggests that the GAO's would have a competitive advantage over the PAO's, adversely affecting phosphorous removal.

The DSVI values reflected in figure 4.6.8 were consistently higher in reactor 2 than in reactor 1 and were above 150mg/L on a number of occasions. The values for reactor 1 ranged between 100 and 150mg/L. However, the FI, shown in figure 4.6.10 was similar for both reactors throughout. As with Borchers Quarry (section 4.3), the reactor that performed better in terms of nutrient removal, seemed to exhibit a propensity, albeit small in this case, for bulking. However, at Macassar there was no significant difference in F/M between the reactors. In this case the reactor with the lower DSVI values (reactor 1) also had the lower VSS concentrations in the mixed liquor (figure 4.6.7), except for one month, when levels were equal in the two reactors.

A point of interest is that filament composition in the two reactors differed slightly from month to month, even although the influent was the same. In addition, although the FI was considered normal for both reactors, the DSVI was "raised" in reactor 2. Perhaps the link between FI and MLVSS would be better than that between DSVI and MLVSS, because the FI is a more quantitative measurement of filament abundance, whereas DSVI reflects settling, and can thus be influenced by a host of other factors.

It can once again be speculated that the presence of residual amounts of nitrate/nitrite would be conducive to AA bulking using the metabolic hypothesis of Casey *et al.* (1999). Once again, the reactor with the higher levels of nitrates/nitrites in the effluent, performed better in terms of DSVI, making this unlikely.

By referring to table 4.6.2 it can be seen that *M. parvicella* and actinomycetes were the most prevalent dominant filaments, and the problem with scum formation is thus not surprising. It can be speculated that the cause was the presence of large amounts of dietary fats in the influent, with other process conditions playing a contributory role. Four of the six dominant filaments encountered fall into the "low F/M" category according to Jenkins *et al.* (2004). These are: *M. parvicella*, actinomycetes, Type 0041, and Type 1851. The other two, namely *H. hydrosis* and actinomycetes can be found at a wide range of loading levels (Jenkins *et al.*, 2004). *M. parvicella* did not show any variation from winter to summer, and was still a dominant filament in November.

Notable was the presence of Type 1851 in high numbers and the fact that this organism, according to Eikelboom (2004), is commonly found, but almost never dominates in domestic WWTP's, and is more likely to prevail in large numbers in industrial plants. In such WWTP's there is an abundance of LMW compounds. It would be interesting to analyse the composition of the influent to Macassar, and other domestic WWTP's in Cape Town with Type 1851 as a dominant filament, to ascertain whether influent composition is the reason for it's abundance or whether there are other factors at play. For example, the plant manager was worried that incomplete mixing and the creation of septic conditions was taking place in some areas within the Carousel system. If this is true, these septic conditions could be a contributory factor.

M. parvicella, actinomycetes and Type 0041 were dominant in random scum samples from both reactors. The implication of the former two filaments with scum formation is well documented and there is further discussion on this topic in Chapter 5 under sections 5.1.2.4 and 5.1.2.6. The presence of Type 0041 is not so clear-cut. The organism has a hydrophobic Gram-positive cell wall, but it usually has a sheath. However, if the sheath became damaged, the organism would presumably also have a propensity to float. Also, the filaments were so intertwined that it is possible *M. parvicella* and actinomycetes provided the means of flotation for the entire mass.

The protozoan and metazoan composition given in table 4.6.3, was such that it reflected the presence of varying concentrations of dissolved oxygen, low F/M ratio and a long sludge age. The presence of high numbers of *Pleuromonas* spp., such as encountered in reactor 2 usually indicates a lack of oxygen or a high sludge load (Eikelboom, 2000). The latter can be ruled out, so lack of oxygen is the more likely cause. However, nitrification and COD removal were not dramatically affected during this period. Perhaps there was a toxic event that led to the demise of most of the ciliate population and to the abundance of flagellates thereafter. However, without further data and online DO readings, it is difficult to definitively speculate whether the bloom was due to such an occurrence, insufficient oxygen or another reason entirely.

In summary, Macassar WWTP performed poorly in terms of the removal of nitrates/nitrites and phosphorous. Lack of phosphorous removal was likely due to the high COD/P ratio and the presence of nitrates in the anaerobic zone. The filaments encountered were indicative of low F/M conditions, supported by actual values obtained. The presence of *M. parvicella* and actinomycetes points to high levels of dietary fat in the influent and explains the problems with intermittent scum formation. The presence of bulking conditions was marginal for one of the reactors.

4.7 MITCHELLS PLAIN WWTP

4.7.1 PLANT CONFIGURATION AND OPERATING PARAMETERS

Mitchells plain WWTP is a NDBEPR plant with a UCT configuration (figure 4.1.1 and 4.1.2). The design capacity is 37 ML/day and there are eight bioreactors named alphabetically from A to H, that treat wastewater primarily of domestic origin. Thirty five percent of the flow is channeled into 4 of the reactors (C, D, E and F) and the other 65% into the larger reactors (G and H). One reactor from each of these groups, namely reactor C and reactor G were chosen for the study. The DO in the aerobic zone is maintained in the range of 2.0 to 2.5mg/L by means of diffuse aeration. The plant does experience intermittent problems with scum formation, particularly in the anoxic zone. The scum tends to break up before the weir overflow site and there is no carry over. Scum samples from the plant were not analyzed.

4.7.2 RESULTS

4.7.2 (a) Operational data

The influent (I) is the supernatant from the alphabetically named PST's supplying reactor G (PST C-F) and reactor H (PST GH).

The mixed liquor (ML) results are from reactor C and reactor G, the former being fed from PST (C-F) and the latter from PST (GH). The mixed liquor from the reactors flows into the clarifiers also alphabetically named. Clarifier CD is supplied by reactor C and clarifier GH is supplied by reactor G.

The effluent (E) is the supernatant from clarifiers CD and GH.

*Note: breaks in the graph lines occur when no data was available

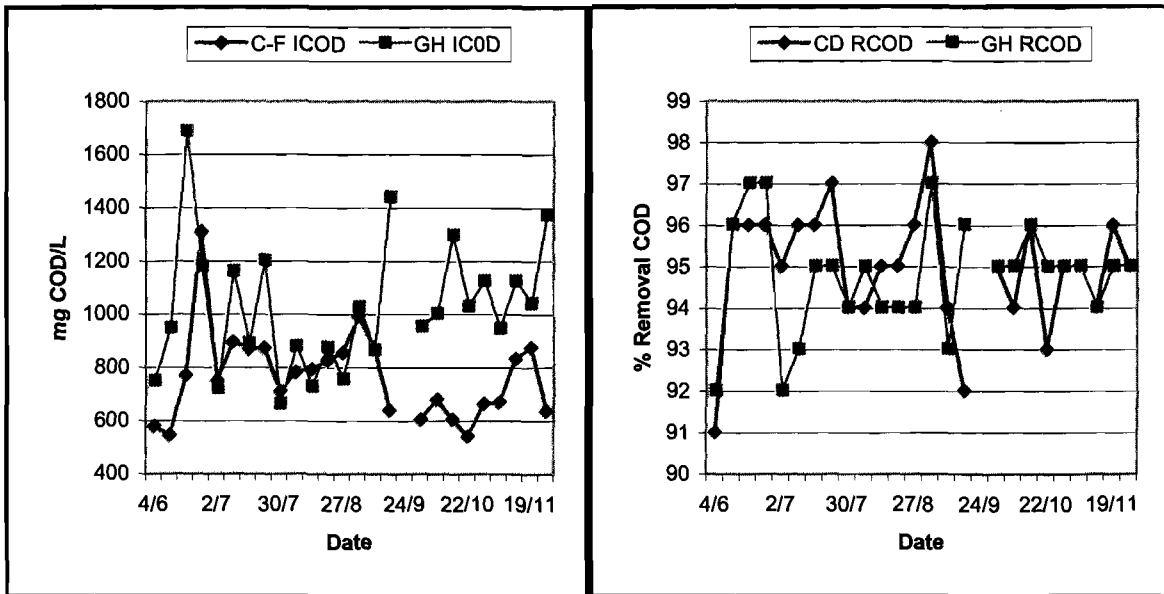


Figure 4.7.1 Influent COD levels from PST C-F (C-F ICOD) and PST GH (GH ICOD) at Mitchells plain WWTP in 2007

Figure 4.7.2 Removal efficiency of COD by bioreactors CD (CD RCOD) and GH (GH RCOD) at Mitchells plain WWTP in 2007

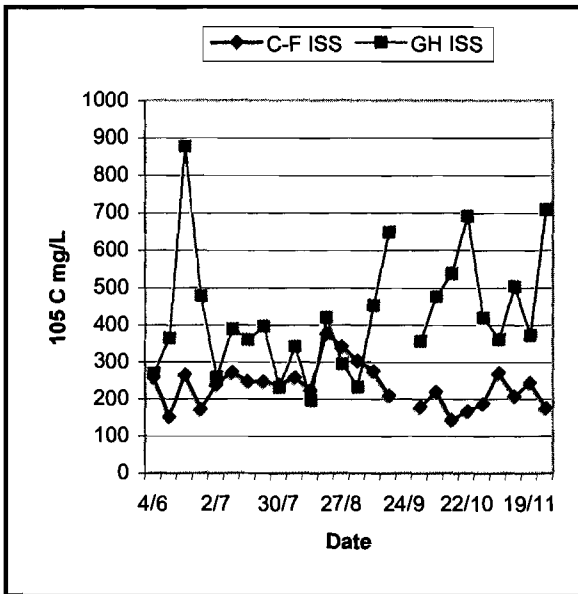


Figure 4.7.3 Influent suspended solids from PST C-F (C-F ISS) and PST GH (GH ISS) at Mitchells plain WWTP in 2007

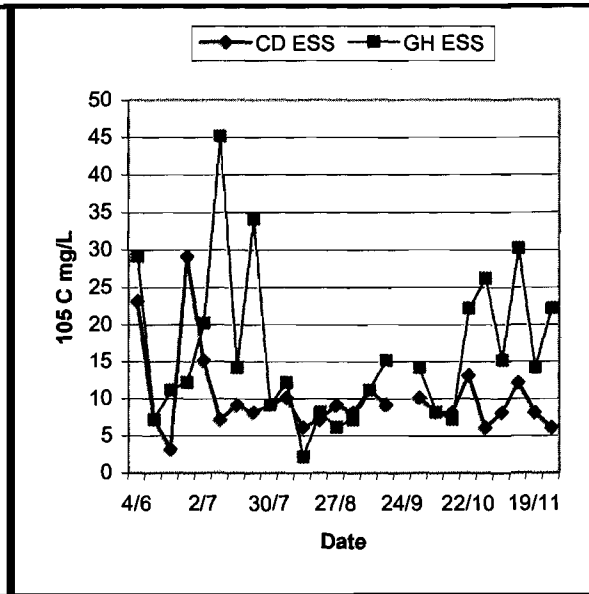


Figure 4.7.4 Effluent suspended solids from clarifier CD (CD ESS) and clarifier GH (GH ESS) at Mitchells plain WWTP in 2007

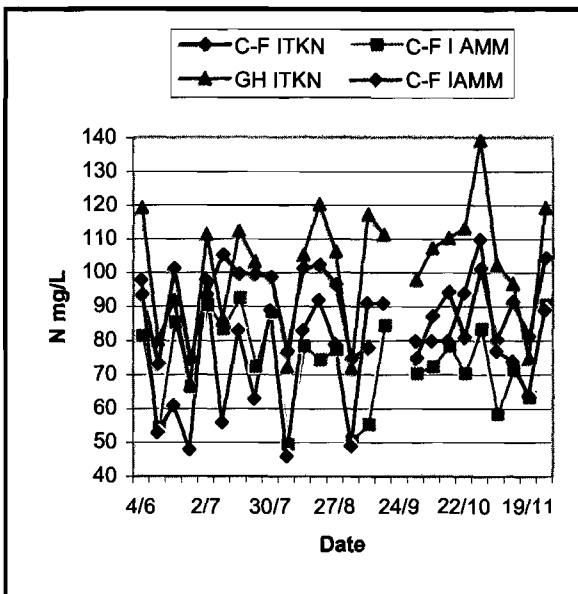


Figure 4.7.5 Influent TKN from PST C-F (C-F ITKN) and PST GH (GH ITKN) and influent ammonia from PST C-F (C-F IAMM) and PST GH (GH IAMM) at Mitchells plain WWTP in 2007

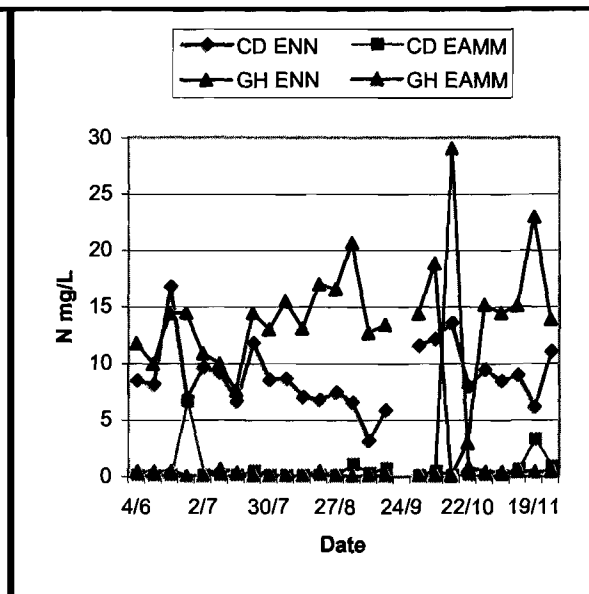


Figure 4.7.6 Effluent nitrates/nitrites from clarifier CD (CD ENN) and clarifier GH (GH ENN) and effluent ammonia from clarifier CD (CD EAMM) and clarifier GH (GH EAMM) at Mitchells plain WWTP in 2007

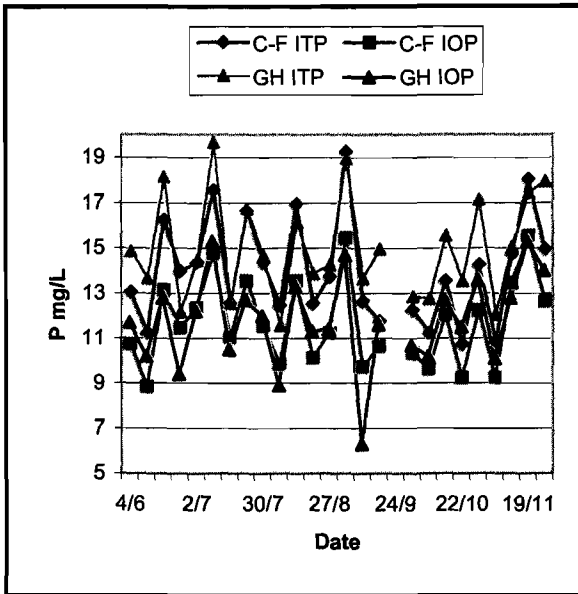


Figure 4.7.7 Influent total phosphates from PST C-F (C-F ITP) and PST GH (GH ITP) and influent σ -phosphates from PST C-F (C-F IOP) and PST GH (GH IOP) at Mitchells plain WWTP in 2007

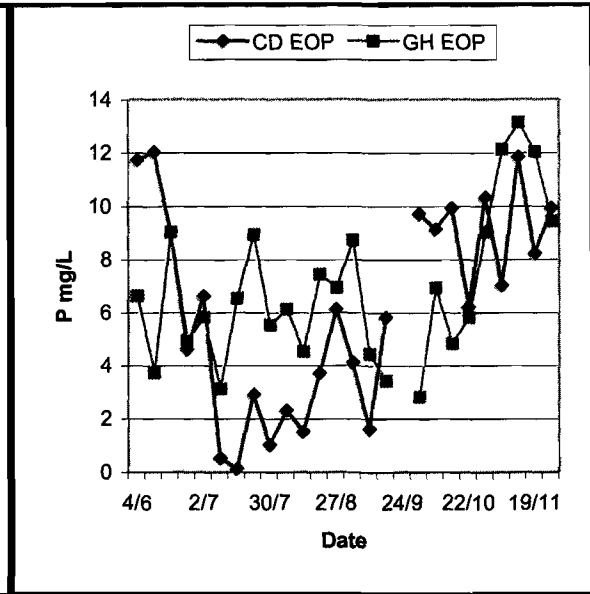


Figure 4.7.8 Effluent σ -phosphates from clarifier CD (CD EOP) and clarifier GH (GH EOP) at Mitchells plain WWTP in 2007

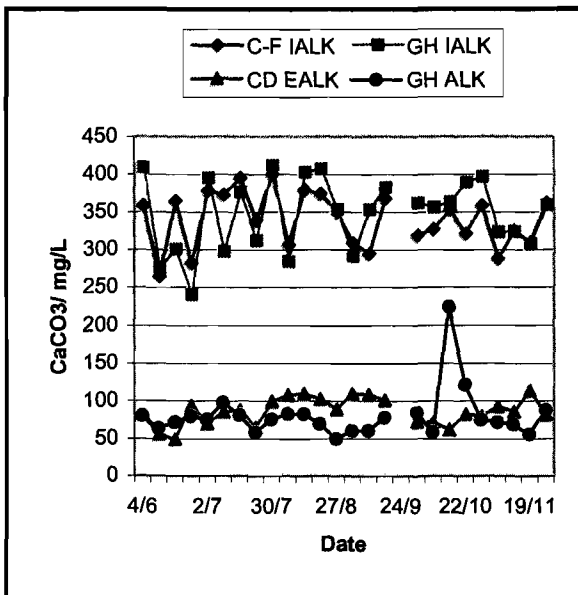


Figure 4.7.9 Influent alkalinity from PST C-F (C-F IALK) and PST GH (GH IALK) and effluent alkalinity from clarifier CD (CD EALK) and clarifier GH (GH ALK) at Mitchells plain WWTP in 2007

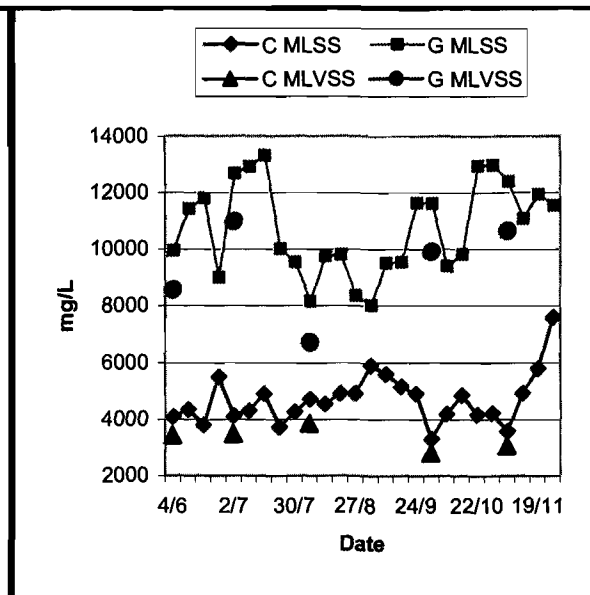


Figure 4.7.10 Values for mixed liquor suspended solids in reactor C (C MLSS) and reactor G (G MLSS) and mixed liquor volatile suspended solids in reactor C (C MLVSS) and reactor G (G MLVSS) at Mitchells plain WWTP in 2007

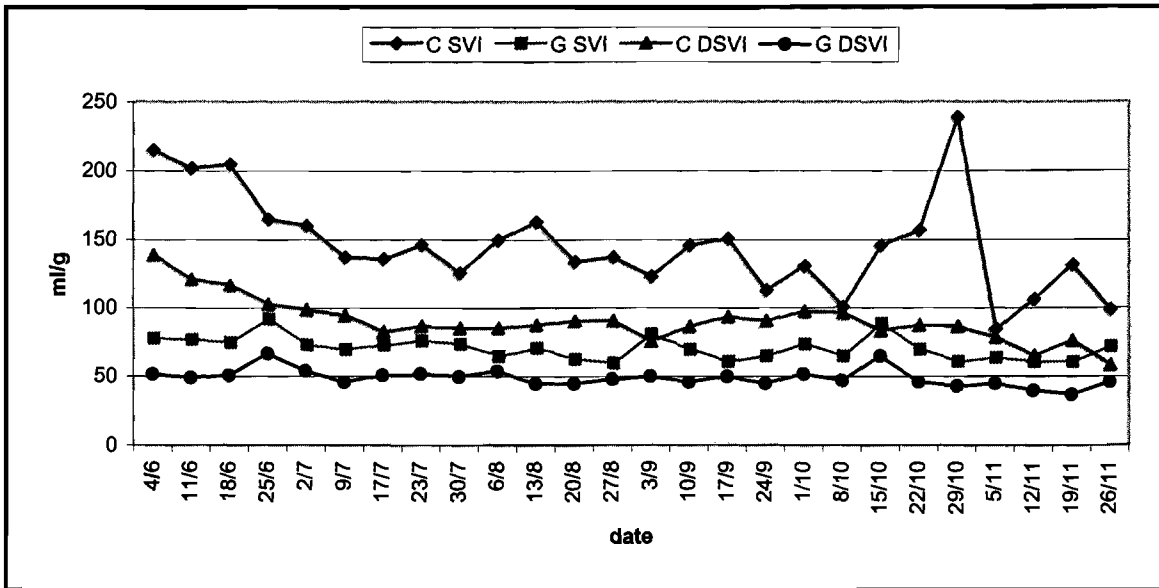


Figure 4.7.11 Values of sludge volume index for reactor C (C SVI) and reactor G (G SVI) and dissolved sludge volume index for reactor C (C DSVI) and reactor G (G DSVI) at Mitchells plain WWTP in 2007

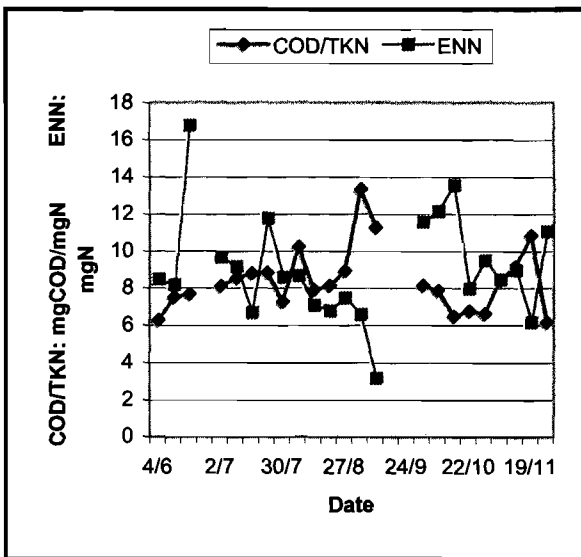


Figure 4.7.12 Ratio of influent COD/TKN plotted against the effluent levels of nitrates/nitrites from reactor C at Mitchells plain WWTP in 2007

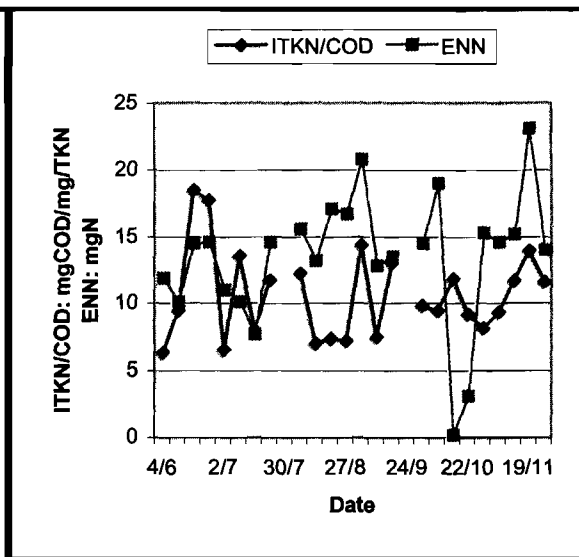


Figure 4.7.13 Ratio of influent COD/TKN plotted against the effluent levels of nitrates/nitrites from reactor G at Mitchells plain WWTP in 2007

| TABLE 4.7.1 FOOD TO MICROORGANISM (F/M) RATIO IN REACTOR C AND REACTOR G OF MITCHELLS PLAIN WWTP IN 2007 | | |
|----------------------------------------------------------------------------------------------------------|----------------------|----------------------|
| | REACTOR C | REACTOR G |
| June | 0.19 mgCOD/mgVSS.day | 0.18 mgCOD/mgVSS.day |
| July | 0.17 mgCOD/mgVSS.day | 0.10 mgCOD/mgVSS.day |
| August | 0.23 mgCOD/mgVSS.day | 0.27 mgCOD/mgVSS.day |
| October | 0.21 mgCOD/mgVSS.day | 0.17 mgCOD/mgVSS.day |
| November | 0.19 mgCOD/mgVSS.day | 0.14 mgCOD/mgVSS.day |

4.7.2 (b) Microscopic sludge analysis

This was performed monthly on the following dates: 18/6/07, 16/7/07, 20/8/07, 24/9/07, 22/10/07 and 26/11/07.

- **Floc structure**

Reactor C

The flocs were round in shape, compact in structure and firm in strength for the duration of the study except for the structure in October, which was open. The floc size was predominantly large (>250µm) in August, but medium-sized for the balance.

Reactor G

The flocs were round in shape, compact in structure and firm in strength for the duration of the study (see figure 4.7.14). The floc size was medium in June and July, but predominantly large from August to November.

- **Diversity**

There was a moderate amount of diversity with scanty monocolonies in all samples, except the sample from reactor C in October, which had a moderate number of monocolonies, probably G-bacteria.

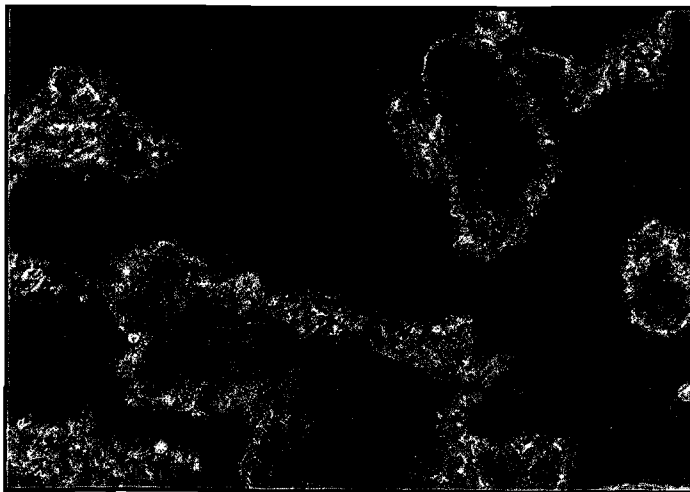


Figure 4.7.14 Micrograph of Wet mount showing the floc structure encountered at Mitchells plain reactor G in August 2007, namely large, compact and firm

• Filament index

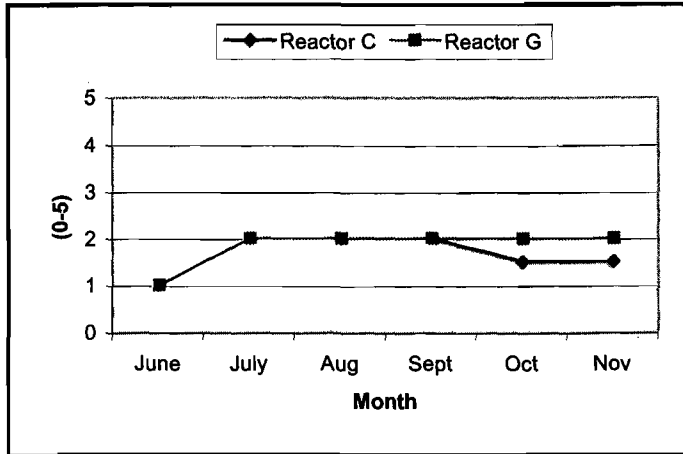


Figure 4.7.15 Filament index of reactor C and reactor G at Mitchells plain WWTP in 2007. The results were the same in June, July and August

TABLE 4.7.2 DOMINANT AND SECONDARY FILAMENTOUS ORGANISMS IDENTIFIED FROM THE MIXED LIQUOR OF REACTOR C AND REACTOR G AT MITCHELLS PLAIN WWTP IN 2007

| AS DOMINANT FILAMENTS | | | | | | | | |
|-------------------------|------------|------------|------------|------------|------------|------------|---------------------------------|--------------------------------|
| | June | July | Aug | Sept | Oct | Nov | % Prevalence C and G (dominant) | % Prevalence C and G (overall) |
| Type 0092 | X (C,G) | X (C,G) | X (C,G) | X (C,G) | X (C) | X (C,G) | 92 | 100 |
| Type 1851 | | | | X (G) | X (G) | X (G) | 25 | 92 |
| <i>M. parvicella</i> | | | X (C) | | | | 8 | 25 |
| AS SECONDARY FILAMENTS | | | | | | | | |
| Type 0092 | | | | | X (G) | | } Also dominant filaments | |
| Type 1851 | X (G) | X (C,G) | X (C,G) | X (C) | X (C) | X (C) | | |
| <i>M. parvicella</i> | | | | X (C) | X (C) | | | |
| Type 0041 | X (C) | X (C,G) | X (C,G) | X (C,G) | X (C,G) | X (C,G) | 0 | 92 |
| Type 1701 | | X (G) | X (G) | | | | 0 | 17 |
| <i>Flexibacter</i> spp. | | | | | | X (C,G) | 0 | 17 |
| Type 021N | | | | | X (G) | X (G) | 0 | 17 |
| Type 0803 | | X (C) | | | | | 0 | 8 |
| <i>Thiothrix</i> spp. | | | X (G) | | | | 0 | 8 |

TABLE 4.7.3 PROTOZOA/METAZOA IDENTIFIED IN THE MIXED LIQUOR FROM REACTOR C AND REACTOR G AT MITCHELLS PLAIN WWTP IN 2007

| | | SCANTY | | | | | |
|----------------------|-----------------------------|------------|------------|------------|------------|------------|------------|
| | | June | July | Aug | Sept | Oct | Nov |
| Sessile ciliates | <i>Carchesium</i> spp. | | X (C,G) | | X (C,G) | X (G) | X (G) |
| | <i>Epistylis</i> spp. | X (C) | | X (C,G) | X (C) | X (C,G) | X (C,G) |
| | Suctorean | | | X (G) | | | |
| | <i>Vaginicola</i> spp. | | | | | X (C,G) | |
| | <i>Vorticella</i> spp. | X (G) | X (C,G) | X (C) | X (C,G) | X (C,G) | X (C,G) |
| Crawling ciliates | <i>Aspidisca</i> spp. | X (C,G) | | X (C) | X (C,G) | | X (C,G) |
| | <i>Chilodonella</i> spp. | | X (C) | X (G) | X (C,G) | | X (C,G) |
| | <i>Trachelophyllum</i> spp. | | X (G) | | X (G) | X (G) | X (G) |
| Free-living ciliates | <i>Blepharisma</i> spp. | | X (G) | X (G) | X (G) | | |
| | <i>Colpidium</i> spp. | | | X (C,G) | X (C) | X (C) | |
| | <i>Euplotes</i> spp. | | X (G) | X (C,G) | | | |
| | <i>Litonotus</i> spp. | | X (G) | X (C) | X (C,G) | | |
| | <i>Spirostomum</i> spp. | | | | | | X (C) |
| | Unidentified | | | X (C) | X (G) | | |
| Flagellates | <i>Bodo</i> spp. | | X (C) | X (G) | X (C,G) | X (G) | X (C,G) |
| | <i>Peranema</i> spp. | | X (C) | | X (C) | | X (C,G) |
| | Unidentified | | X (C) | | | X (G) | |
| Amoebae | | X (C) | X (C) | X (C,G) | X (G) | X (C,G) | |
| Testate amoebae | | | | X (C,G) | | | |
| Heliozoa | | | | X (C) | | X (C) | |
| Oligochaeta | X (C) | X (G) | | X (C,G) | | X (G) | |
| Rotifera | X (G) | X (C,G) | X (C,G) | X (C,G) | X (C,G) | X (C,G) | |
| Tardigrades | | | | X (G) | X (C) | | |

TABLE 4.7.3 (CONTINUED) PROTOZOA/METAZOA IDENTIFIED IN THE MIXED LIQUOR FROM REACTOR C AND REACTOR G AT MITCHELLS PLAIN WWTP IN 2007

| | | MODERATE | | | | | |
|-------------------|------------------------|----------|----------|----------|------|------------|----------|
| | | June | July | Aug | Sept | Oct | Nov |
| Attached ciliates | <i>Vorticella</i> spp. | | | X (G) | | | |
| Crawling ciliates | <i>Aspidisca</i> spp. | | X (G) | X (C) | | X (C,G) | |
| Tardigrades | | | | | | | X (C) |

4.7.3 DISCUSSION

The influent into each of the two reactors was not from the same PST source. Therefore influent levels of certain substrates differed considerably during the trial period, notably the influent COD, SS, and TKN. Levels of these were usually higher from the PST supplying reactor G. Influent ammonia levels also differed, but neither PST was prone to supplying consistently higher levels than its counterpart.

Both reactor C and G performed well in terms of the removal of COD (figure 4.7.2), SS (figure 4.7.3 and 4.7.4) and ammonia (figure 4.7.7 and 4.7.8). There was a large drop in alkalinity, depicted in figure 4.7.9, from PST effluent to clarifier effluent, due to the high degree of nitrification taking place.

Although nitrification was efficient in both reactors for most of the study period, residual nitrates and nitrites were constantly present, shown in figure 4.7.8. Denitrification took place to a larger extent in reactor C, with satisfactory levels of nitrates/nitrites in the clarifier effluent being achieved at times. Complete denitrification is not expected in a WWTP with the UCT configuration as there is no second anoxic zone to enable denitrification of all nitrification products formed in the aerobic zone. Although there should be a high mixed liquor re-circulation from the aerobic to the anoxic zone, there will still be some nitrification products that exit the aerobic zone. However, values obtained were high, especially from reactor G. As described for other WWTPs in previous sections of this chapter, on the occasions that ammonia levels increased, there was an accompanying decrease in nitrates/nitrites.

Operating conditions leading to poor denitrification were discussed in section 4.4.3. Regarding the lack of electron donor as having an adverse effect on denitrification: the influent COD/TKN ratio for reactor C ranged between 6.2-13.3mgCOD/mgTKN (mean 8.3mgCOD/mgTKN). The mean for reactor G was 9.9mgCOD/mgTKN. Thus, the mean from reactor C was much closer to the value of 8.4mgCOD/mgTKN reported by Kujawa *et al.* (1996), as being necessary for complete denitrification. If this figure is correct, and if lack of electron donor did play a role in poor denitrification in reactor C, it would explain the more erratic nature of denitrification from this reactor - there would have been periods where electron donor was insufficient interspersed with periods of sufficiency. This is substantiated by the fact that when the COD/TKN ratio was plotted against effluent nitrates/nitrites, there was a correlation between increased influent COD/TKN and more efficient denitrification in reactor C. This is especially apparent around the beginning of September 2007. There was no such correlation in reactor G. These relationships can be seen in figures 4.7.12 and 4.7.13. This suggests that denitrification

in reactor C, with a smaller COD/TKN ratio, was adversely affected by lack of electron donor to some extent. The mechanism for poor denitrification in the case of reactor G, with a higher COD/TKN ratio, overshadowed any effect that a lack of electron donor may have had (if any).

The efficiency of phosphorous removal shown in graphs 4.7.7 and 4.7.8, was erratic in reactor C, but was constantly poor in reactor G. In all cases where more than one reactor per WWTP was included, there was a comparative correlation between denitrification efficiency and phosphorous removal. Reactors C and G at Mitchells plain are not identical and receive influent from different PST's. Even so, reactor C, with lower levels of nitrates and nitrites in the clarifier effluent generally performed better regarding phosphorous removal than reactor G. Values for reactor G were constantly poor, while those from reactor C were more erratic.

Once again, it can be inferred that the presence of nitrates in the anaerobic zone interfered with the biological process of phosphorous removal. The influent COD/P ratio in reactor C was also lower than that of reactor G: 42.6-69.9mgCOD/mgP (mean 53.6mgCOD/mgP) and 66.9-83.5 (mean 66.9mgCOD/mgP). These levels should enhance the growth of GAO's over PAO's according to Liu *et al.* (1997), but would be more pronounced for reactor G.

The other consistent comparison that was a feature of the study was that the reactor that performed better in terms of nitrate/nitrite removal showed consistently higher DSVI values. These are reflected in figure 4.7.11. This phenomenon also occurred at Mitchells plain WWTP and is elucidated upon in chapter 6. However, in this case neither the DSVI values nor the FI values were ever high enough to indicate the presence of bulking conditions at Mitchells Plain WWTP.

There was a large difference in the MLSS and MLVSS values of reactor C and G, seen in figure 4.7.10, but the operation of the reactors was such that loading rates, given in table 4.7.1 remained similar. The incomplete denitrification could theoretically lead to AA bulking according to the hypothesis of Casey *et al.* (1999). However, the chosen bulking indicators of FI and DSVI were never raised, and the DSVI was in fact lower for the reactor with the most residual nitrates/nitrites. In addition, as already noted, the reactor with the lower values of nitrates/nitrites in the clarifier effluent exhibited the higher DSVI values.

Referring to table 4.7.2, it can be seen that Type 0092 was the major dominant filament encountered in both reactors. *M parvicella* was present as a dominant species on one occasion in reactor C. In reactor G, Type 1851 was co-dominant from September, possibly being linked to the higher settling values from this reactor. Once again, this filament was unexpectedly found as a dominant species in a domestic wastewater removal plant (Eikelboom, 2000). All three dominant filaments are classed as low F/M by Jenkins *et al.* (2004). This correlates well with the calculated loading rates. The association of the ubiquitous Type 0092 with non-bulking conditions was established in the meta-analysis, and conditions leading to the competitive growth of this organism should be encouraged.

In summary, both reactors from the Mitchells Plain WWTP gave sub-optimal results in terms of nutrient removal (phosphorous and nitrates/nitrites). The reactor that showed the worst nutrient removal record exhibited the lowest DSVI. The dominant filaments were all classed as low F/M and bulking conditions were not encountered at any stage.

4.8 PAROW WWTP

4.8.1 PLANT CONFIGURATION AND OPERATING PARAMETERS

Parow WWTP is a small plant treating designed to treat 1ML effluent/day. There is no primary settling and the configuration of the bioreactor is MLE with surface aeration (refer to figure 4.2.1 and 4.2.2). The theoretical operational DO is 2.3 mg/L. The plant is generally overloaded in terms of capacity and is due to be upgraded in the near future. There are often operational problems with wasting leading to MLSS values above the ideal range.

4.8.2 RESULTS

4.8.2 (a) Operational data

The influent (I) is raw influent. There are no PST's.

The mixed liquor (ML) results are from the bioreactor.

The effluent (E) is the supernatant from the clarifier.

*Note: breaks in the graph lines occur when no data was available

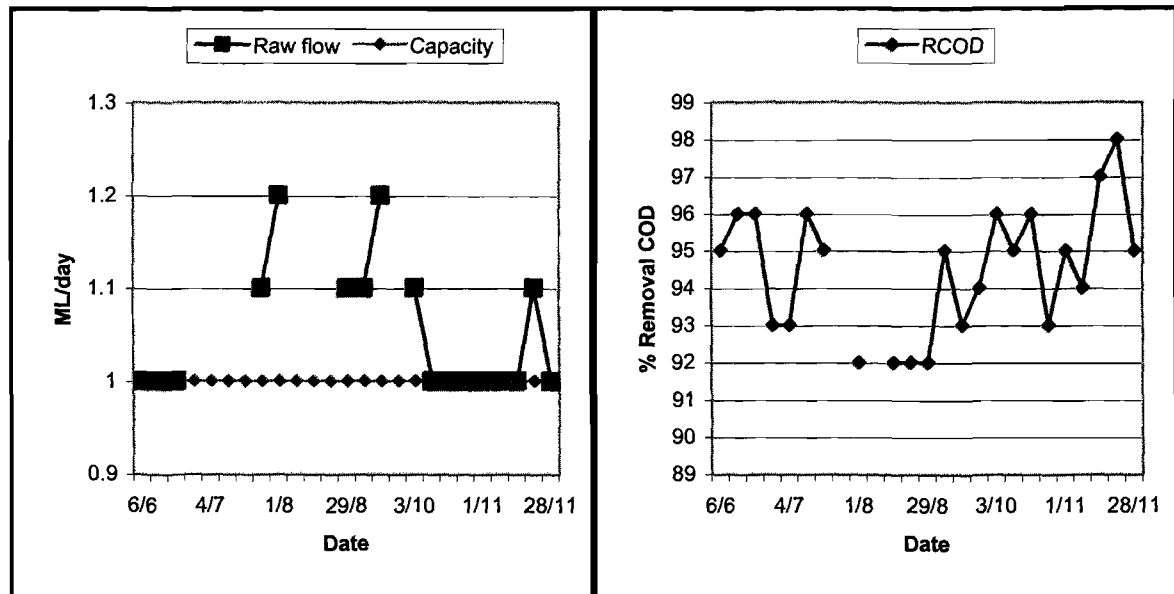


Figure 4.8.1 Graph of inflow of raw wastewater into Parow WWTP plotted against the design capacity of the plant in 2007

Figure 4.8.2 Graph depicting the removal of COD (RCOD) from Parow WWTP in 2007

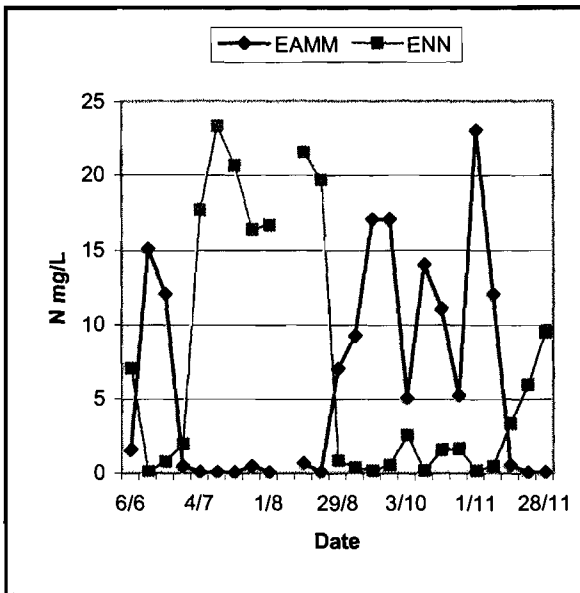


Figure 4.8.3 Graph depicting the effluent ammonia (EAMM) and effluent nitrates/nitrites (ENN) from Parow WWTP in 2007

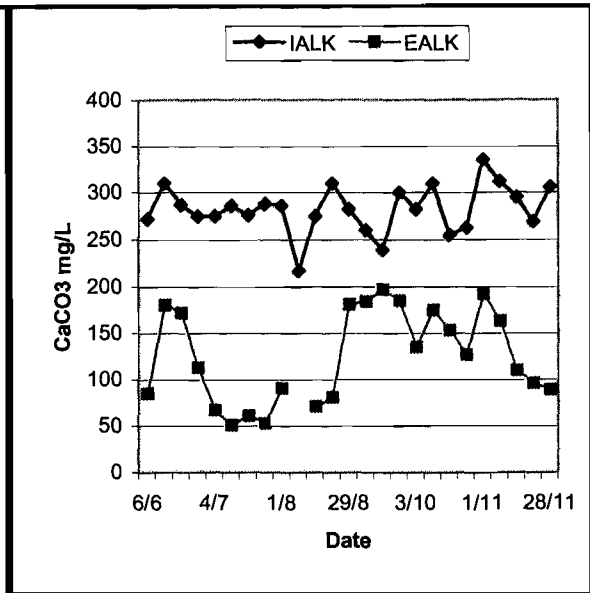


Figure 4.8.4 Graph depicting the influent alkalinity (IALK) and effluent alkalinity (EALK) values at Parow WWTP in 2007

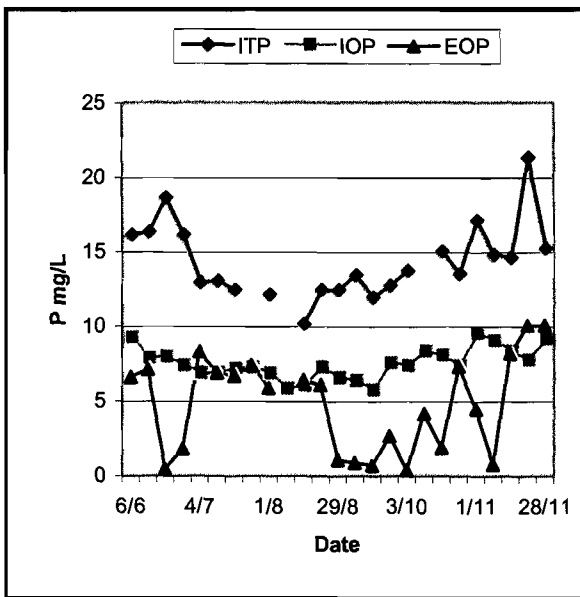


Figure 4.8.5 Graph showing the influent total phosphates (ITP) and influent orthophosphates (IOP) as well as the effluent orthophosphates (EOP) at Parow WWTP in 2007

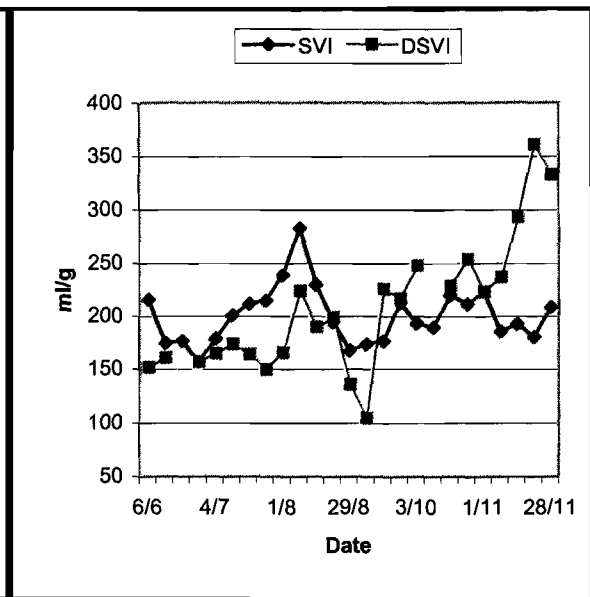


Figure 4.8.6 Graph showing the SVI and DSVI values from the mixed liquor at Parow WWTP during 2007

| TABLE 4.8.1 FOOD TO MICROORGANISM (F/M) RATIO IN REACTOR OF PAROW WWTP IN 2007 | |
|--------------------------------------------------------------------------------|----------------------|
| June | 0.19 mgCOD/mgVSS.day |
| July | 0.15 mgCOD/mgVSS.day |
| September | 016 mgCOD/mgVSS.day |
| October | 0.23 mgCOD/mgVSS.day |
| November | 0.20 mgCOD/mgVSS.day |

4.8.2.1 Microscopic sludge analysis

This was performed monthly on the following dates: 28/6/07, 26/7/07, 23/8/07, 24/9/07, 25/10/07 and 22/11/07.

● Floc structure

The flocs were round in shape, open in structure and firm in strength from June to September. In October and November, the shape changed from round to irregular. The flocs were medium-sized from June to August, became large in September and remained large in October. In these latter months, the flocs exhibited extensive bridging, the large flocs appearing to be smaller flocs joined together by bridging. In November, this phenomenon was so extensive, that it was impossible to accurately determine floc size. In this month the flocs appeared as large open mats. In September and October, the large number of nematodes that were voraciously feeding on the flocs disturbed the floc structure, leaving holes in the flocs (see figure 4.8.7).



Figure 4.8.7 Wet mounts of mixed liquor samples taken from Parow WWTP in September 2007. Micrograph A shows the structure of the flocs that has been disturbed by voracious feeding of the large nematode population. Micrograph B shows an example of two of these nematodes.

● Diversity

There was a moderate amount of diversity with scanty monocolonies in all samples, except for November, when there were no monocolonies seen.

● Filament index

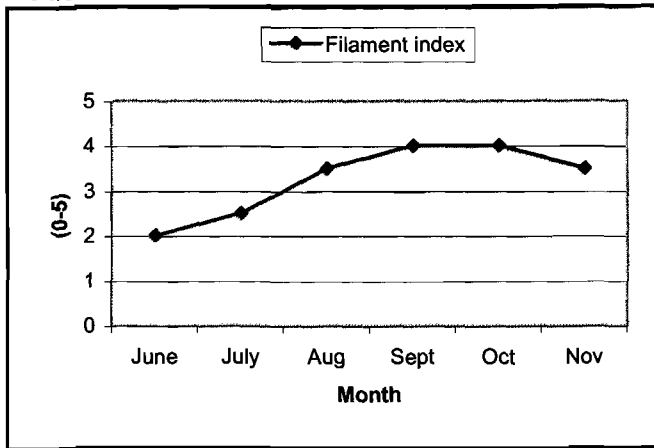


Figure 4.8.8 Graph depicting the rising filament index from the activated sludge at Parow WWTP from June to October 2007

| TABLE 4.8.2 DOMINANT AND SECONDARY FILAMENTOUS ORGANISMS IDENTIFIED IN THE MIXED LIQUOR OF PAROW WWTP IN 2007 | | | | | | | | |
|---------------------------------------------------------------------------------------------------------------|------|------|-----|------|-----|-----|---------------------------|------------------------|
| AS DOMINANT FILAMENTS | | | | | | | | |
| | June | July | Aug | Sept | Oct | Nov | % Prevalence (dominant) | % Prevalence (overall) |
| Actinomycetes | X | X | X | | X | X | 83 | 100 |
| Type 1851 | X | X | X | X | X | | 83 | 100 |
| Type 0092 | | X | X | | | X | 50 | 100 |
| Type 0041 | X | X | | | | | 33 | 50 |
| Actinomycetes | | | | X | | | } Also dominant filaments | |
| Type 1851 | | | | | | X | | |
| Type 0092 | X | | | X | X | | | |
| Type 0041 | | | X | | | | | |
| Type 021N | | | X | X | X | X | 0 | 67 |
| <i>M. parvicella</i> | | | X | X | X | | 0 | 50 |
| <i>H. hydrosis</i> | | | | | | | 0 | 17 |

| TABLE 4.8.3 PROTOZOA/METAZOA IDENTIFIED IN THE MIXED LIQUOR OF PAROW WWTP IN 2007 | | | | | | | |
|-----------------------------------------------------------------------------------|--------------------------|------|------|-----|------|-----|-----|
| SCANTY | | | | | | | |
| | | June | July | Aug | Sept | Oct | Nov |
| Sessile ciliates | <i>Epistylis</i> spp. | X | | X | X | X | |
| | <i>Opercularia</i> spp. | X | X | | | | |
| Crawling ciliates | <i>Aspidisca</i> spp. | X | | | | X | X |
| | <i>Chilodonella</i> spp. | X | | | | | |
| | Unidentified | X | | | | | |
| Free-living ciliates | <i>Euplotes</i> spp. | | | | X | | |
| Flagellates | <i>Bodo</i> spp. | | X | X | | | |
| | <i>Peranema</i> spp. | | | | | | |
| | <i>Pleuromonas</i> spp. | | | | | | |
| | <i>Trepomonas</i> spp. | | | | | | |
| Rotifers | | X | | X | X | X | X |
| Nematodes | | | | | X | X | |
| MODERATE | | | | | | | |
| Sessile ciliates | <i>Epistylis</i> spp. | | | | | X | |
| Crawling ciliates | <i>Aspidisca</i> spp. | | | X | | | |
| Nematodes | | | | | X | X | |
| NUMEROUS | | | | | | | |
| Crawling ciliates | <i>Aspidisca</i> spp. | | | | X | | |
| Flagellates | <i>Monsiga</i> spp. | X | | | | | |

4.8.3 DISCUSSION

Removal of COD was good throughout (figures 4.8.1 and 4.8.2). From graph 4.8.8, it can be seen that nitrification and denitrification was extremely unstable. At times effluent ammonia levels were high, and effluent nitrate/nitrites were low to absent. At other times the converse was true. The other WWTP's discussed in this chapter generally exhibited either poor nitrification or poor denitrification, but none gave such aberrant results with both. As expected, enhanced consumption of alkalinity mirrored periods of good nitrification and vice versa, graphically depicted in figures 4.8.3 and 4.8.4.

Phosphorous removal too was extremely unstable (figure 4.8.5). There was a very strong link between poor denitrification and poor phosphorous removal. This is evidence that the presence of nitrates/nitrites in the anoxic zone was responsible for deterioration in phosphorous removal. Conversely, when there were no residual nitrates/nitrites in the effluent, and by association, none in the anoxic zone, phosphorous removal was enhanced. The latter also occurred in Bellville WWTP, which has the same configuration. It thus lends credence to the speculation that lack of nitrates/nitrites due to poor nitrification, did indeed convert the anoxic zone into a de facto anaerobic zone with resulting EBPR. This occurred intermittently at Parow WWTP and on a continual basis at Bellville WWTP.

The DSVI values reflected in figure 4.8.6 indicated the presence of bulking conditions for most of the study period, with especially high values during the latter stages of the study. The FI values depicted in figure 4.8.8 also indicated the presence of bulking conditions from August to November.

Table 4.8.2 reflects the dominant and secondary filaments identified. No noticeable change in species dominance occurred with an increase in DSVI. In fact, Type 0092, not strongly associated with bulking conditions by the meta-analysis, was one of the dominant filaments at this stage. There was an increase in loading rates, given in table 4.8.1 at this stage that may have played a role in filament abundance.

All of the dominant species except the actinomycetes are classed as low F/M by Jenkins *et al.* (2004). Actinomycetes are able to grow over a range of loading rates. Type 1851 was also found as a dominant in other WWTP's, notably at Bellville, another MLE configured plant, where there was an increased DSVI and FI but also a high F/M. It is unlikely that Type 0092 had any effect on the advent of bulking conditions, as this filament tends to hide in the flocs. The increased DSVI and FI may have been due to a relative change in the ratio of heterotrophs to filaments induced by predation of nematodes that allowed the existing filamentous population to flourish.

The diversity of the protozoa and metazoa was low. This was probably due to the continual change in environmental conditions that must have been present to create such erratic nutrient removal results. Only the species able to adapt to changing conditions would prevail. There were interesting blooms of particular species at times:

- The flagellate *Monsiga* spp. an indicator organism for high sludge load and or shortage of oxygen (Eikelboom, 2000) was present in large numbers in June. Concurrent with this was a poor rate of nitrification. As the calculated F/M values were low, loss of nitrification was probably associated with low DO.
- Large numbers of nematodes were feeding on the flocs. These worms were present from July, but increased in number until October. The physical disturbance of the flocs as well as consumption of heterotrophs possibly played a role in the increased DSVI. The nematodes were not capable of feeding on the filamentous population, giving the latter a competitive advantage.

In summary, Parow WWTP functioned in a particularly unstable manner in terms of nutrient removal and was prone to bulking. There was excellent evidence of a link between the absence of nitrates/nitrites in the anoxic zone and phosphorous removal efficiency that can be extrapolated to other MLE WWTP's such as Bellville. Changes in nitrification/denitrification patterns were most likely associated with changes in DO. Unfortunately, the DO results could not be obtained to substantiate this fact unequivocally.

4.9 POTSDAM WWTP

4.9.1 PLANT CONFIGURATION AND OPERATING PARAMETERS

Potsdam WWTP uses both activated sludge bioreactors and fixed medium (stone) trickling filter systems to treat the influent. This influent consists of primarily domestic waste with an industrial component. The trickling filter system is ageing and function is poor. The plant is presently being upgraded to increase both capacity and efficiency. Approximately half of the influent is treated in the two identical bioreactors after undergoing primary settling. The plant operates in the UCT configuration with surface aeration. Potsdam WWTP experiences intermittent scum problems, mainly in winter and during periods of overloading.

4.9.2 RESULTS

4.9.2 (a) Operational data

Influent (I) is the supernatant from the PST.

The mixed liquor (ML) results are from combined samples from both bioreactors.

The effluent (E) is the supernatant from the appropriate clarifier.

*Note: breaks in the graph lines occur when no data was available

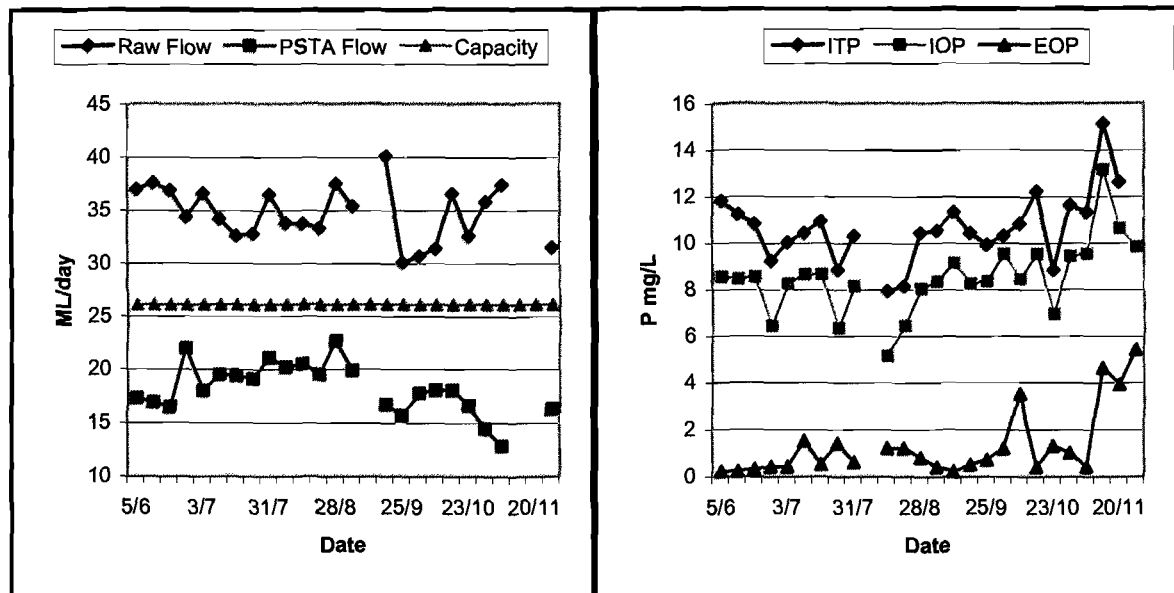


Figure 4.9.1 Graphs showing the raw flow into Potsdam WWTP in 2007 as well as the design capacity. In addition, the flow into the PST feeding the bioreactors is given (PSTA).

Figure 4.9.2 Graph depicting the levels of influent total phosphates (ITP) and σ -phosphates as well as the effluent levels of σ -phosphates at Potsdam WWTP in 2007

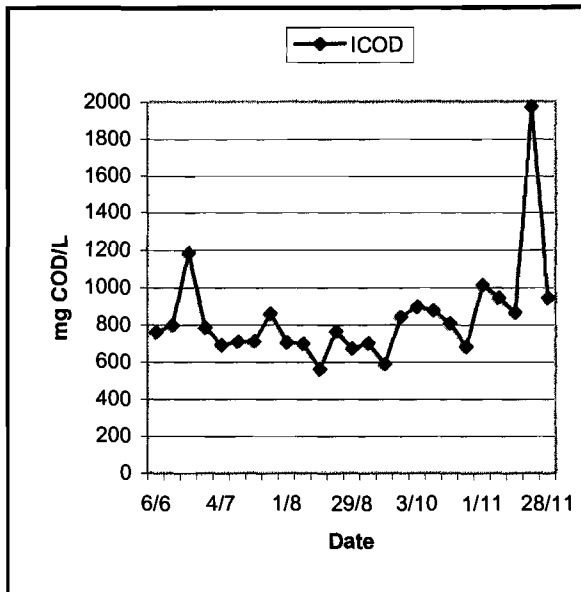


Figure 4.9.3 Influent COD (ICOD) from the PST at Potsdam WWTP in 2007

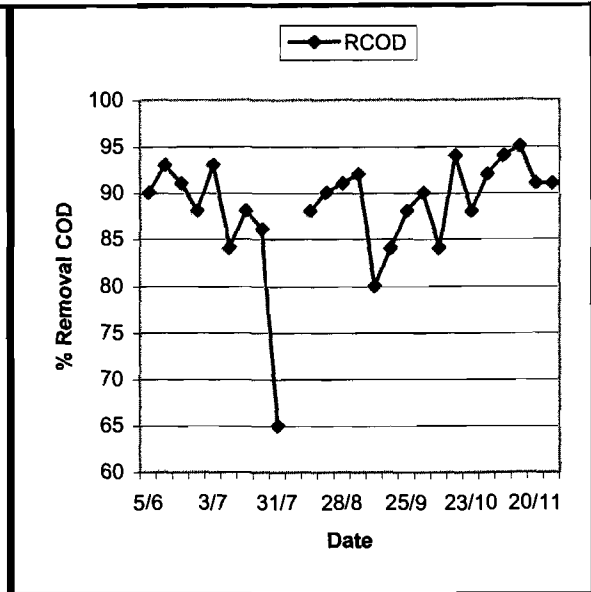


Figure 4.9.4 COD removal efficiency (RCOD) by the bioreactor at Potsdam WWTP in 2007

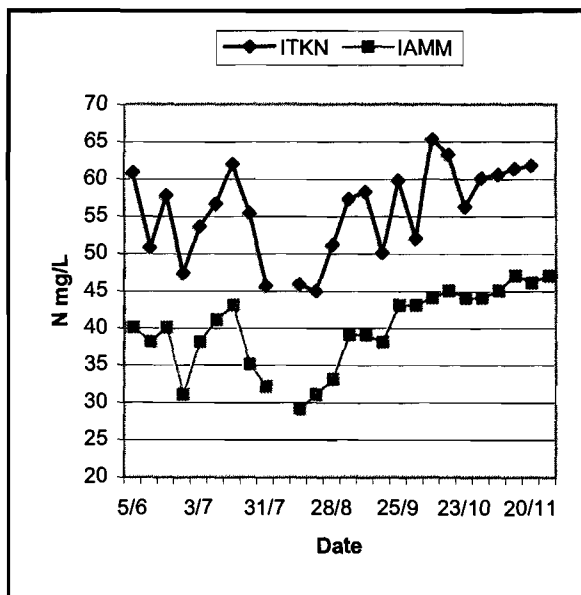


Figure 4.9.5 Graph depicting the influent ammonia (IAMM) and influent TKN (ITKN) at Potsdam WWTP in 2007

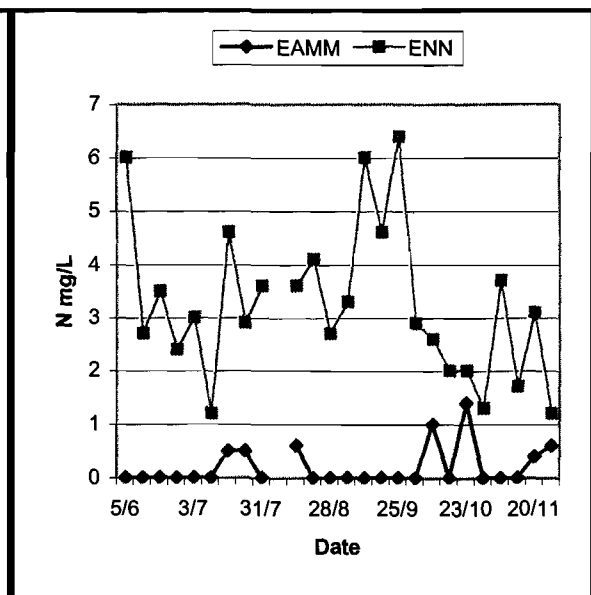


Figure 4.9.6 Graph depicting the effluent ammonia (EAMM) and nitrates/nitrites (ENN) at Potsdam WWTP in 2007

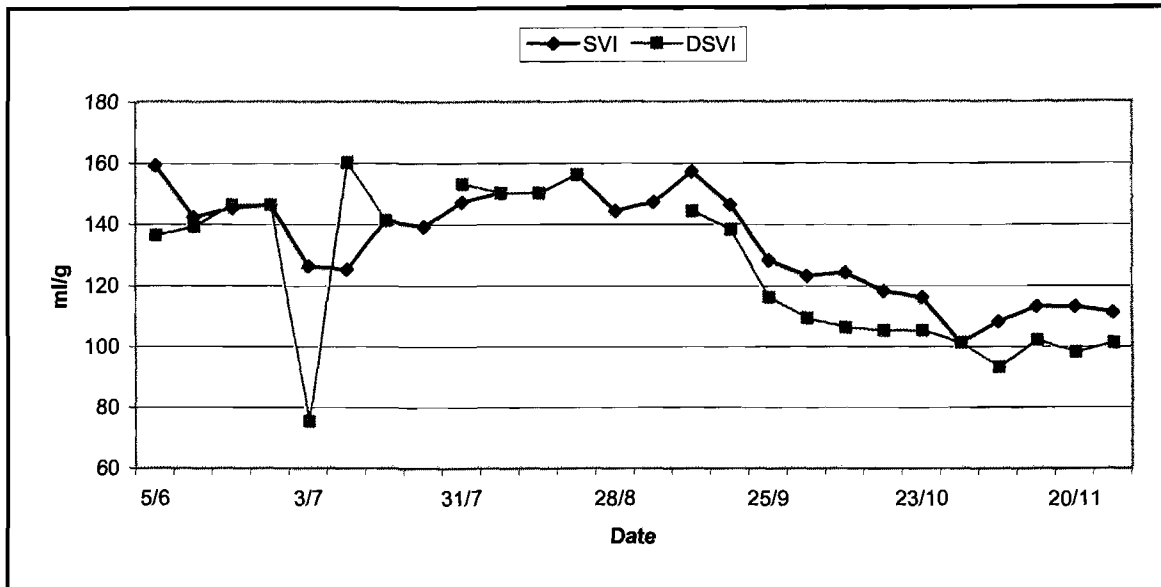


Figure 4.9.7 Graph depicting the SVI and DSVI values experienced from the mixed liquor at Potsdam WWTP in 2007

| Month | F/M Ratio (mgCOD/mgVSS.day) |
|-----------|-----------------------------|
| June | 0.12 |
| July | 0.09 |
| September | 0.13 |
| October | 0.08 |
| November | 0.06 |

4.9.2 (b) Microscopic sludge analysis

This was performed monthly on the following dates: 20/6/07, 18/7/07, 29/8/07, 19/9/07, 24/10/07 and 21/11/07.

● Floc structure

The flocs were round in shape, open in structure and firm in strength in all samples. The flocs were medium in size in June and September. In June, there was bridging between the flocs. In July and August, there were numerous pin flocs as well as large open flocs with extensive bridging. In October and November, the flocs were large and open with extensive bridging (see figure 4.9.8A).

● Diversity

Diversity was low in June, July, August and November, medium-low in September and medium in October. Notable was the abundance of monoclonies from June to October (see figure 4.9.8B). However, by November the numbers were low. On staining, the majority appeared to be GAO's (see section 4.9.3 for further details).

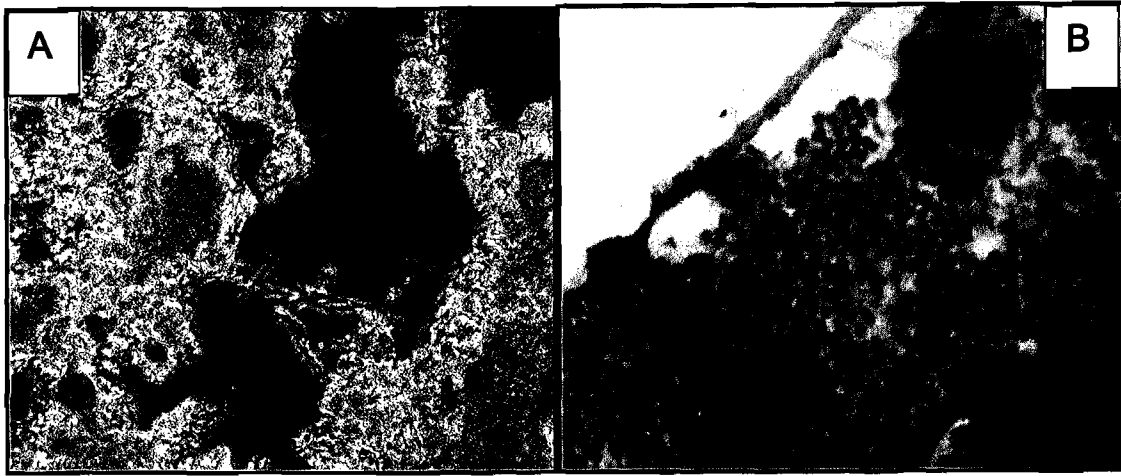


Figure 4.9.8 Wet mount (A) of mixed liquor sample taken from Potsdam WWTP in July 2007 showing floc structure with numerous filaments and bridging between the flocs and Neisser stain (B) of mixed liquor taken from Potsdam WWTP in August 2007 showing monocolonies staining as GAO's.

• Filament index

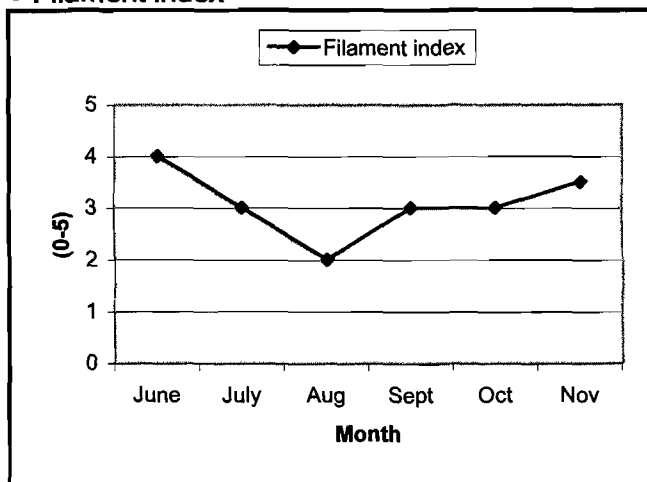


Figure 4.9.9 Graph representing the FI values obtained at Potsdam WWTP in 2007

TABLE 4.9.2 DOMINANT AND SECONDARY FILAMENTOUS ORGANISMS IDENTIFIED FROM THE MIXED LIQUOR AT POTSDAM WWTP IN 2007

| AS DOMINANT FILAMENTS | | | | | | | | |
|------------------------|------|------|-----|------|-----|-----|-------------------------|-------------------------|
| | June | July | Aug | Sept | Oct | Nov | % Prevalence (dominant) | % Prevalence (overall) |
| Type 0092 | X | X | X | X | X | X | 100 | 100 |
| <i>M. parvicella</i> | | X | | | | | 16 | 100 |
| AS SECONDARY FILAMENTS | | | | | | | | |
| <i>M. parvicella</i> | X | | X | X | X | X | } | Also dominant filaments |
| Type 1851 | | | X | X | X | X | 0 | 67 |
| Actinomycetes | | X | X | | X | X | 0 | 67 |
| Type 021N | | | X | | X | X | 0 | 50 |
| Type 0041 | | | X | | X | X | 0 | 50 |
| <i>N. limicola</i> III | | | | | X | | 0 | 16 |

TABLE 4.9.3 PROTOZOA/METAZOA IDENTIFIED IN THE MIXED LIQUOR FROM KRAAIFONTEIN WWTP IN 2007

| SCANTY | | | | | | | |
|----------------------|-----------------------------|------|------|-----|------|-----|-----|
| | | June | July | Aug | Sept | Oct | Nov |
| Sessile ciliates | <i>Carchesium</i> spp. | | | | | X | |
| | <i>Epistylis</i> spp. | | | | X | X | |
| | <i>Vorticella</i> spp. | | X | X | X | X | |
| Crawling ciliates | <i>Aspidisca</i> spp. | X | | X | X | X | |
| | <i>Chilodonella</i> spp. | | | | | X | X |
| | <i>Trachelophyllum</i> spp. | X | X | X | | | X |
| Free-living ciliates | <i>Colpidium</i> spp. | | X | | X | | X |
| | <i>Litonotus</i> spp. | X | | X | X | | |
| | Unidentified | | | X | X | X | |
| Flagellates | <i>Bodo</i> spp. | | | | | X | |
| | <i>Peranema</i> spp. | | | X | | X | X |
| | Unidentified | | | | | | X |
| Amoebae | | X | | X | | | |
| Rotifera | | X | X | X | X | X | X |
| MODERATE | | | | | | | |
| Crawling ciliates | <i>Aspidisca</i> spp. | | X | | | | |
| Flagellates | <i>Peranema</i> spp. | | | | X | | |
| Rotifera | | | | | X | | |

4.3 DISCUSSION

Potsdam WWTP was overloaded in terms of volume for the entire study period as shown in figure 4.9.1. Despite this, the removal of COD was generally efficient, albeit erratic during a period in July (unexplained). The influent COD and COD removal values are reflected in figures 4.9.3 and 4.9.4.

Nitrification was efficient for most of the study period, but residual nitrates/nitrites were present in the clarifier effluent (figures 4.9.5 and 4.9.6). The values were never very high and not unexpected for a WWTP with a UCT configuration.

Most of the study WWTP's that exhibited residual nitrates/nitrites in the clarifier effluent showed poor phosphorous removal. This was not the case for Potsdam WWTP, and removal of this nutrient was generally excellent, except for a period in November, when both influent and effluent values increased slightly (refer to figure 4.9.3). In the earlier months, moderate numbers of monoclonies were observed in the mixed liquor samples. These numbers were far greater than those from any of the other WWTP's (barring some samples from Kraaifontein). In November, there was both a decrease in monoclonies and phosphorous removal. The majority of the monoclonies stained as GAO's (or G-bacteria). The cell wall, but not the contents of G-bacteria stain with the Neisser stain, but genres may be either Gram positive or Gram negative. The microbial composition in terms the presence of large numbers of GAO organisms as well as the filamentous and protozoan/metazoan population was remarkably similar to that found at Kraaifontein WWTP. By comparing the relevant tables from each of these WWTPs (table 4.5.2 with 4.9.2 and 4.5.3 with 4.9.3), this can be clearly seen. Both WWTPs have the same configuration (UCT) and mode of aeration (surface). Both experienced similar low loading rates and influent COD values (figures 4.5.1 and 4.9.3 and tables 4.5.1 and 4.9.1).

The COD/P ratio at Potsdam WWTP ranged from 51.6-65.9mgCOD/mgP with a mean value of 57.1mgCOD/mgP. These values, according to Liu *et al.* (1997), should favour the growth of GAO's over PAO's. However, WWTP's with lower COD/P values than those obtained at Potsdam were far less efficient for the removal of phosphorous. The amount of VFA's and nitrates in the anaerobic zone were unfortunately unknown. In terms of nutrient removal, the performance at Potsdam WWTP was also similar to that experienced at Kraaifontein WWTP.

Conditions in terms of the COD/P ratio encountered at Potsdam WWTP would appear to support the growth of GAO's in preference to PAO's, according to Liu *et al.* (1997). In addition, many GAO's were seen in the mixed liquor samples. Despite these findings, phosphorous removal was efficient. This seemingly presents an anomaly. To enhance the understanding of the dynamics of phosphorous removal at the Cape Town WWTP's there is a need to perform comparative measurements of VFA's, nitrates and oxygen in the anaerobic zones of the bioreactors.

In chapter 2, the hypothesis of Liu *et al.* (1997), outlining the metabolic mechanism and operating conditions responsible for competition between the GAO's and PAO's is described in detail. However, it seems that not all representatives of the phylogenetically diverse group of GAO's are capable of anaerobic synthesis of intracellular polysaccharides. Seviour *et al.* (2000) examined pure cultures of *Amaricoccus* spp. and ascertained that although some stains can synthesize intracellular polysaccharides in large amounts under aerobic conditions using either glucose or acetate as substrate, they were not capable of doing so under anaerobic conditions. Thus, with this organism,

there is no direct evidence to support the theory of Liu *et al.* (1997). In the case of Potsdam, with large numbers of GAO's and high COD/P ratios concurrent with efficient phosphorous removal, the supposition is that the resident G-bacteria (of unknown phylogeny) did not compete against the PAO's under anaerobic conditions. Without further details on the composition of nutrients in the various reactor zones, it is not possible to speculate further on the relative abundance of these organisms. All of the above assumes that the Potsdam organisms were indeed GAO's and not PAO's. Identification by one of the molecular techniques would be necessary to definitively identify these bacteria. It must be pointed out that organisms staining as classic PAO's were also observed in the mixed liquor, but were considerably outnumbered by the GAO's.

The DSVI values shown in graph 4.9.7 were only higher than 150ml/g in July and August. This was the indicator value chosen to represent the presence of bulking conditions. The FI however, was less than the similarly chosen indicator value only in August. Thus, both the appearance of the flocs and the filament index point to the presence of possible bulking, but this was not reflected in the DSVI values. The dominating filament throughout the study was Type 0092. In the meta-analysis, this organism was not highly associated with bulking in terms of DSVI and FI. This filament is known for growing inside the flocs and not affecting settling (Eikelboom, 2000). The secondary filaments that grew outside and between the flocs, such as *M. parvicella*, Type 1851 and actinomycetes, were probably responsible for the high FI values. Both Type 1851 and actinomycetes were linked to bulking in the meta-analysis, so other factors, such as the large number of monoclonies in the mixed liquor may have played a role in the enhanced settling.

The dominant filament, Type 0092 is classed as low F/M by Jenkins *et al.* (2004), which substantiates this finding. The constant presence of *M. parvicella* and actinomycetes may possibly be related to dietary fats in the influent. It is also highly likely that these organisms are responsible for the intermittent problems with scum formation experienced by Potsdam WWTP. There was no variation in the prevalence of *Micorhrix parvicella* from winter to summer. This phenomenon was not unique to Potsdam.

Apart from the similarity of the protozoan and metazoan community to that at Kraaifontein WWTP, there was nothing notable in the range of these organisms encountered at Potsdam WWTP. The presence of a number of ciliates, flagellates, amoeba and rotifera throughout the study period adequately reflect the alternating anaerobic and aerobic conditions, lack of toxic substances and adequate sludge age. Neither the presence of *Peranema* spp. or rotifera in moderate numbers in September is a cause for concern at this point as no literature was found linking either of these organisms with specific process conditions.

In summary, the bioreactors at Potsdam WWTP performed well in terms of nutrient removal. The removal of phosphorous was especially efficient in comparison to most of the other WWTP's with residual nitrates/nitrites in the clarifier effluent. The presence of a high COD/P ratio and large numbers of GAO's should, in theory, result in decreased phosphorous removal due to the competitive advantage of GAO's over PAO's. The results obtained were thus anomalous, and further identification of the monoclonies, as well as comparative values for nitrates and VFA's in the anaerobic zones would be necessary to endeavour to explain this phenomenon.

4.10 WESFLEUR WWTP

4.10.1 PLANT CONFIGURATION AND OPERATING PARAMETERS

Wesfleur WWTP treats effluent from a variety of industrial sources in the Atlantis area. The plant also houses a reactor that treats domestic waste. Only the industrial reactor was included in the study, in order to enable comparisons between the filaments found in this type of WWTP and those treating either mixed industrial and domestic effluent or those treating effluent entirely of a domestic nature. The configuration of the reactor is the Modified Ludzack-Ettinger with an operational DO of 2.8mg/L created by diffuse aeration (refers to figures 4.2.1 and 4.2.2). The influent is not subjected to primary settling and is thus of a raw nature.

4.10.2 RESULTS

4.10.2 (a) Operational data

The influent (I) is raw industrial influent. There are no PST's.

The mixed liquor (ML) results are from the single industrial bioreactor.

The effluent (E) is the supernatant from the clarifier.

*Note: breaks in the graph lines occur when no data was available

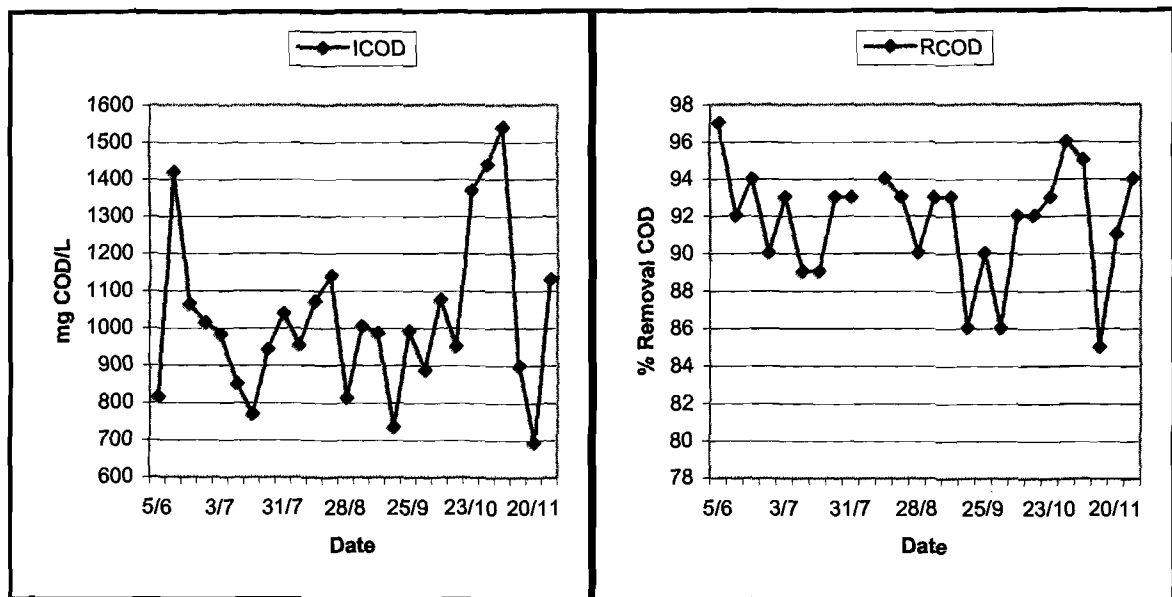


Figure 4.10.1 Graph depicting the influent COD values at Wesfleur WWTP in 2007

Figure 4.10.2 Graph depicting the COD removal efficiency (RCOD) at Wesfleur WWTP in 2007

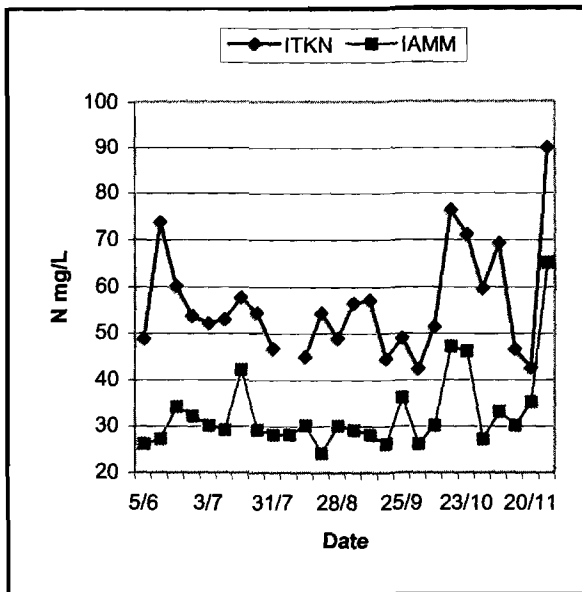


Figure 4.10.3 Graph depicting the influent levels of ammonia (IAMM) and TKN (ITKN) at Wesfleur WWTP in 2007

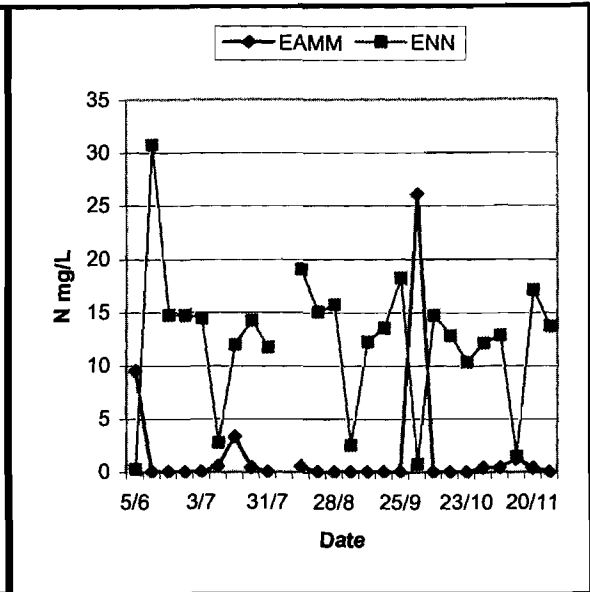


Figure 4.10.4 Graph depicting the effluent levels of ammonia (EAMM) and nitrates/nitrites (ENN) at Wesfleur WWTP in 2007

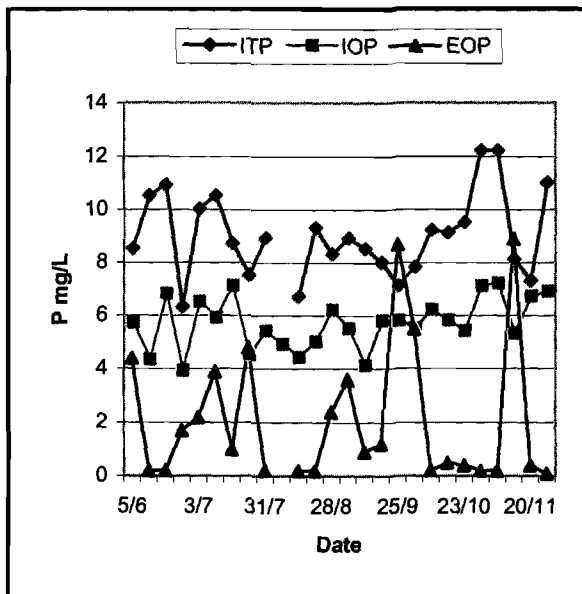


Figure 4.10.5 Graph depicting the influent total phosphates (ITP) and influent orthophosphates (IOP) as well as the effluent orthophosphates at Wesfleur WWTP in 2007

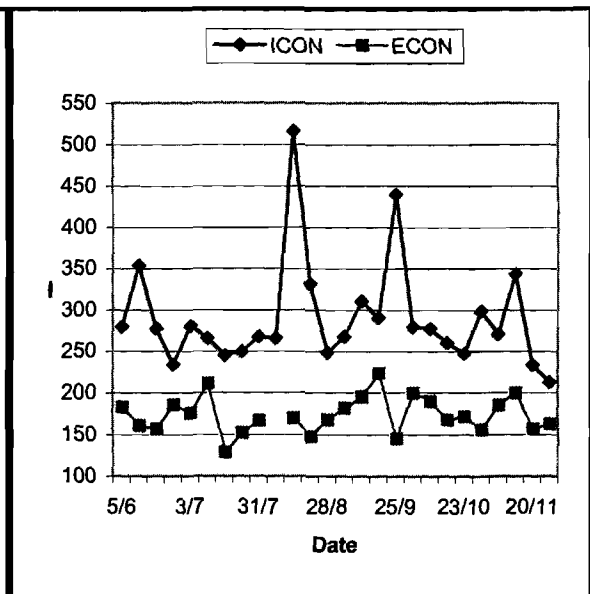


Figure 4.10.6 Graph depicting the influent conductivity values (ICON) and effluent conductivity values at Wesfleur WWTP in 2007

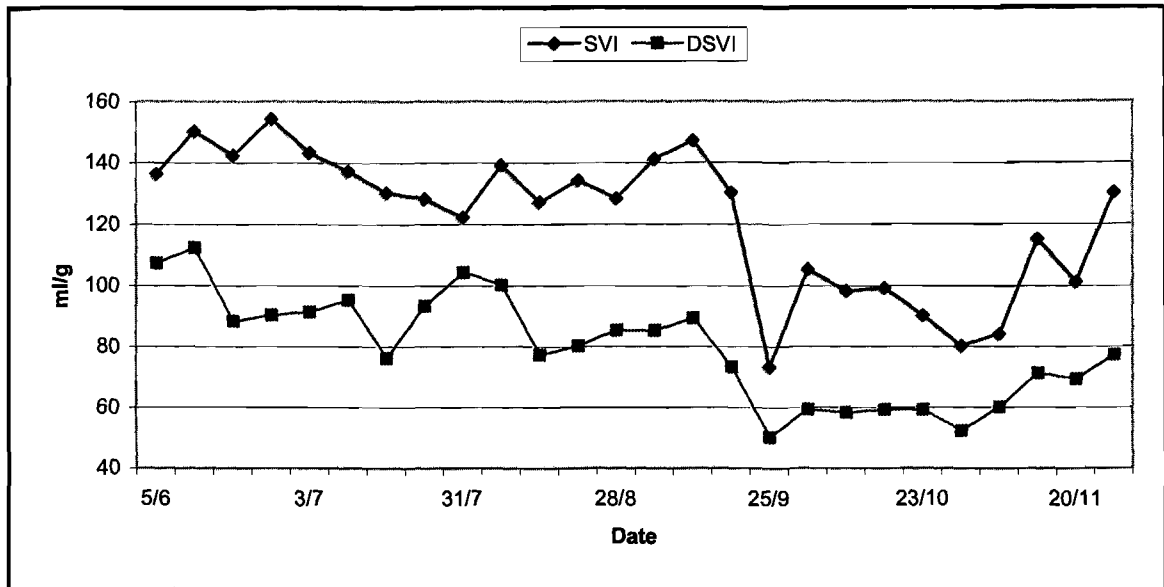


Figure 4.10.7 Graph showing the values for the sludge volume index (SVI) and the dissolved sludge volume index for the mixed liquor at Wesfleur WWTP in 2007

| TABLE 4.10.1 FOOD TO MICROORGANISM (F/M) RATIO IN INDUSTRIAL BIOREACTOR OF WESFLEUR IN 2007 | |
|---------------------------------------------------------------------------------------------|----------------------|
| June | 0.09 mgCOD/mgVSS.day |
| July | 0.11 mgCOD/mgVSS.day |
| September | 0.11 mgCOD/mgVSS.day |
| October | 0.10 mgCOD/mgVSS.day |
| November | 0.19 mgCOD/mgVSS.day |

4.10.2 (b) Microscopic sludge analysis

This was performed monthly on the following dates: 20/6/07, 18/7/07, 29/8/07, 19/9/07, 24/10/07 and 21/11/07.

- **Floc structure**

The shape of the flocs was round from June to October, but in November there were also irregularly shaped flocs present. The flocs were compact in structure in all samples. The flocs were medium-sized in June, July and November, whilst in August, September and October they were small and large, the large flocs appeared as small flocs bound by bridging.

- **Diversity**

There was a moderate amount of diversity in all samples.

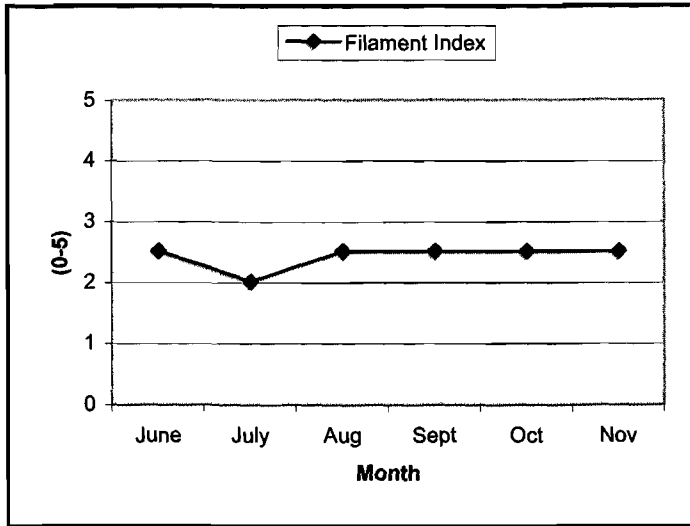


Figure 4.10.8 Graph showing the Filament index values obtained from the mixed liquor at Wesfleur WWTP in 2007

| TABLE 4.10.2 DOMINANT AND SECONDARY FILAMENTOUS ORGANISMS IDENTIFIED FROM THE MIXED LIQUOR AT WESFLEUR WWTP IN 2007 | | | | | | | | |
|---------------------------------------------------------------------------------------------------------------------|------|------|-----|------|-----|-----|---------------------------|------------------------|
| AS DOMINANT FILAMENTS | | | | | | | | |
| | June | July | Aug | Sept | Oct | Nov | % Prevalence (dominant) | % Prevalence (overall) |
| Type 1851 | X | X | X | X | X | X | 100 | 100 |
| Type 0092 | | | | X | X | X | 50 | 100 |
| Type 0041 | | | X | X | | X | 50 | 83 |
| AS SECONDARY FILAMENTS | | | | | | | | |
| Type 0092 | X | X | X | | | | } Also dominant filaments | |
| Type 0041 | | X | | | X | | | |
| <i>N. limicola</i> III | | | | | X | X | 0 | 33 |
| <i>H. Hydrosis</i> | | | | | X | X | 0 | |

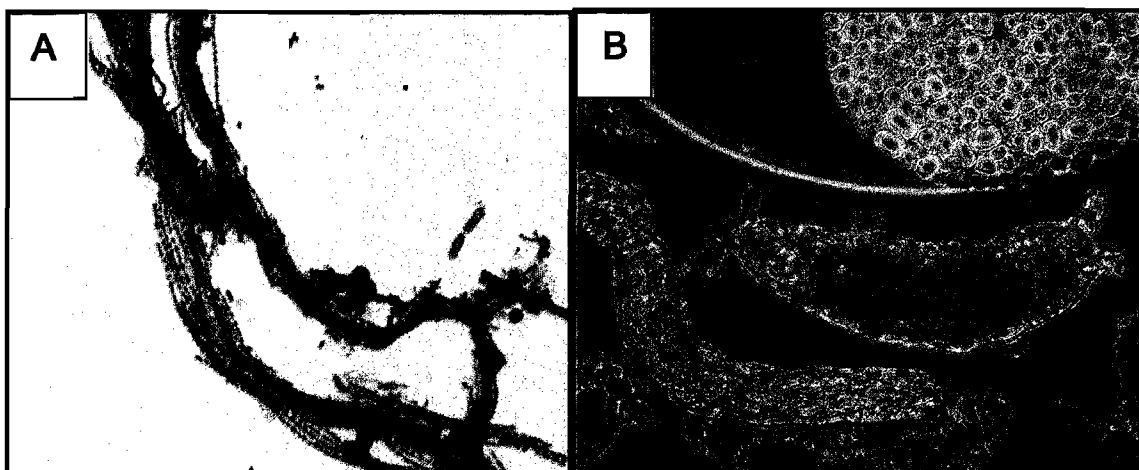


Figure 4.10.9 A, Gram stain of Type 1851 from mixed liquor sample showing “clean” filaments associated with industrial wastewater from Wesfleur WWTP and B, wet mount of mixed liquor from Wesfleur WWTP taken in October 2007 showing a Tardigrade (above) and an Oligochaete worm (below).

| TABLE 4.10.3 PROTOZOA/METAZOA IDENTIFIED IN THE MIXED LIQUOR FROM WESFLUER WWTP IN 2007 | | | | | | | |
|-----------------------------------------------------------------------------------------|-----------------------------|------|------|-----|------|-----|-----|
| SCANTY | | | | | | | |
| | | June | July | Aug | Sept | Oct | Nov |
| Sessile ciliates | <i>Carchesium</i> spp. | | | | | X | |
| | <i>Vorticella</i> spp. | X | | | | X | X |
| | <i>Vaginicola</i> spp. | | | X | | | |
| Crawling ciliates | <i>Aspidisca</i> spp. | X | X | | X | X | X |
| | <i>Trachelophyllum</i> spp. | X | | | X | | |
| Free-living ciliates | <i>Colpidium</i> spp. | | | | | | X |
| | <i>Euplotes</i> spp. | | X | | | | |
| | <i>Litonotus</i> spp. | | | X | | | |
| | <i>Spirostomum</i> spp. | | | X | X | X | X |
| Flagellates | <i>Bodo</i> spp. | | | | X | X | X |
| Amoebae | | | X | | X | | X |
| Heliozoa | | X | X | | | | |
| Rotifers | | | | X | X | X | X |
| Oligochaeta | | X | X | X | X | X | |
| Tardigrades | | | X | | | X | |
| MODERATE | | | | | | | |
| Crawling ciliates | <i>Aspidisca</i> spp. | | | | | | |
| | <i>Chilodonella</i> spp. | | | | | | |
| Free-living ciliates | <i>Spirostomum</i> spp. | | | | | | |

4.10.3 DISCUSSION

The Wesfleur industrial bioreactor received effluent with a high COD content, but removal was good as can be seen by the removal efficiency reflected in figures 4.10.1 and 4.10.2.

Nitrification was satisfactory for most of the study period as shown by the influent and effluent levels of ammonia in figures 4.10.3 and 4.10.4. The latter graph also shows the erratic results of effluent nitrates/nitrites. The levels of these nitrogenous compounds decreased with an increase in ammonia, but also randomly at other times. Complete denitrification is not expected with the MLE configuration, but nitrate/nitrite levels were often high. Lack of electron donor can almost certainly be ruled out as a cause - the COD/TKN ratio was extremely high with a range between 12.6-24.1mgCOD/mgTKN and a mean of 18.5mgCOD/mgTKN. Not surprisingly, there was no correlation between this result and the residual levels of nitrates/nitrites. Operational conditions probably led to poor denitrification.

When nitrification was poor, especially during the peak level at the end of September, the cause of low nitrates/nitrites was most likely due to aeration problems. Lack of DO would lead to the dual effects of reduced nitrification (and thus less substrate for denitrification) as well as possible continuation of denitrification within the aerobic reactor. The latter could occur in severe cases, such as with mechanical breakdown of aerators.

The presence of high residual nitrates/nitrites at times when nitrification was adequate was probably due to either insufficient mixed liquor recycle between the aerobic and

anoxic zone (so that nitrates/nitrites formed in the aerobic zone could not be denitrified and were present in the clarifier effluent), or too high DO in the anoxic zone.

No biological phosphorus removal is expected with the MLE configuration, but removal did occur - this was erratic but substantial at times as seen by figure 4.10.5. By examining figure 4.10.6, it can be seen that the conductivity of the influent was high. The industrial nature of this influent would result in the presence of a range of chemicals. It can be speculated that phosphorus removal took place was by chemical co-precipitation. However, in the two other MLE configured WWTP's, namely Bellville WWTP and Parow WWTP, there was also substantial phosphorous removal that coincided with increased levels of effluent ammonia and decreased nitrates/nitrites. Lack of nitrates and nitrites (and presumably also oxygen) in the anoxic zone probably turned this section of the bioreactor into a de facto anaerobic zone with resultant EBPR. The high COD/P ratio would make conditions unsuitable for a large PAO population to flourish. (The COD/P ratio was 91.6-125.3mgCOD/mgP with a mean of 112.4mgCOD/mgP - double that of Parow WWTP).

The parameters for the presence of bulking conditions used in the study, namely FI and DSVI, shown in graph 4.10.8 and 4.10.7 respectively were both within the defined limits for the duration of the study period.

The calculated monthly F/M ratios shown in table 4.10.1 were low. This supports the fact that the three dominant filaments, namely Type 1851, Type 0041 and Type 0092 are classed as low F/M by Jenkins *et al.* (2004). The association of Type 1851, with industrial waste by Eikelboom, (2000) holds true in this instance (but is not the case with some of the domestic WWTP's included in the study). The filaments found at Wesfleur were often without a sheath and attached growth, a phenomenon also noted by Eikelboom, (2000) as being more typical for industrial waste.

The mixed liquor supported a healthy population of protozoa and metazoa adapted to the prevailing conditions. This, coupled with the presence of healthy nitrifier population, points to the fact that toxicity levels were never a problem. All these populations are extremely sensitive to a range of toxins (discussed in section 4.1.3)

In summary, the operation and microbial population and dynamics of this industrial WWTP were satisfactorily stable for the entire six months. The high COD levels were considerably reduced throughout the study and there were no obvious signs of poisoning at any stage. The filament population reflected the low F/M conditions that prevailed.

4.11 WILDEVOELVLEI WWTP

4.11.1 PLANT CONFIGURATION AND OPERATING PARAMETERS

Wildevoevlei WWTP is designed to treat approximately 14.5ML of domestic effluent per day. The plant houses two different bioreactors: reactor A is a carousel-type system that operates in a 3-stage Phoredox mode from the anaerobic zone. This reactor operates with a DO of 2.0ppm and receives 54% of the daily influent while reactor B receives 46%. Reactor B is configured and operated in the UCT (see figure 4.1.1) mode with an operational DO of 2.2 ppm. Both reactors have an aerobic mass fraction of 50%, an anaerobic mass fraction of 19% and an anoxic mass fraction of 31%.

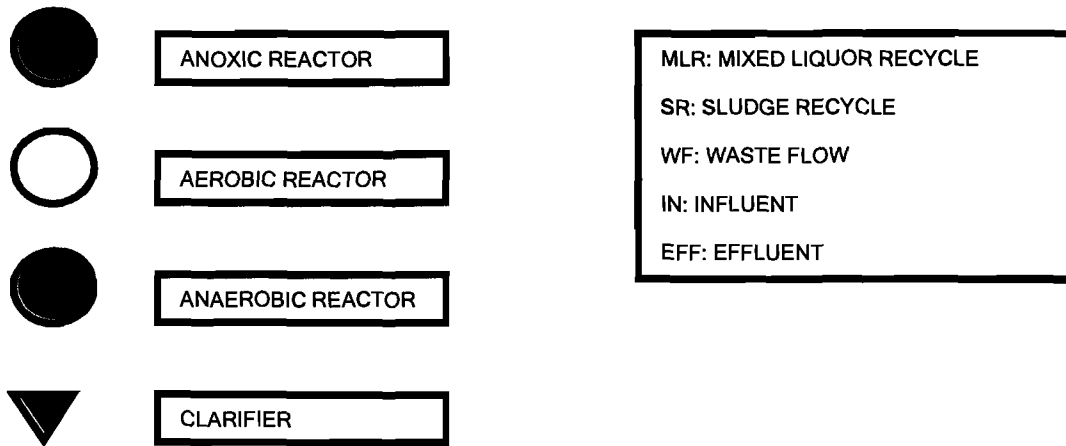


Figure 4.11.1 Key for WWTP configurations

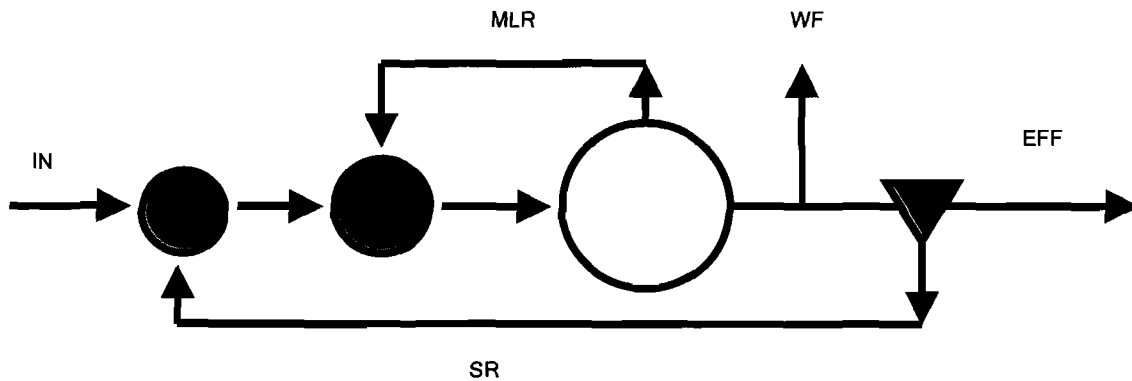


Figure 4.11.2 3-stage Phoredox configuration
(Metcalf and Eddy, 1991)

4.11.2 RESULTS

4.11.2 (a) Operational data

The raw influent (I) is distributed to bioreactor A and reactor B. There is no PST.

The mixed liquor (ML) from bioreactor A and B flows into the clarifiers A and B respectively.

The effluent (E) is the supernatant from the plant clarifiers A and B.

*Note: breaks in the graph lines occur when no data was available

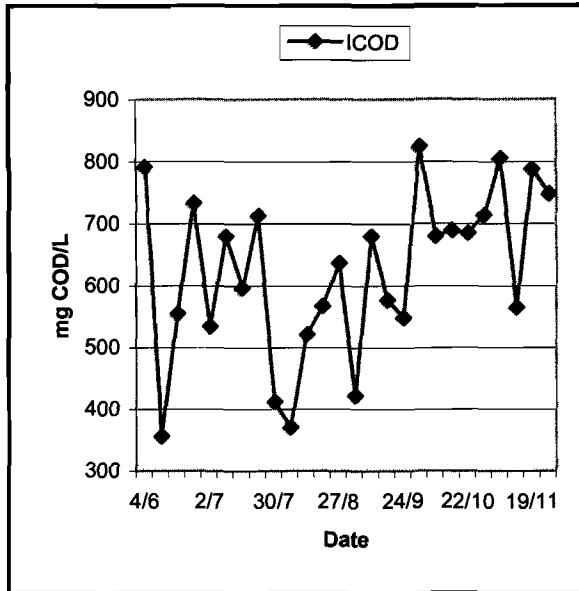


Figure 4.11.3 Graph showing the influent COD (ICOD) at Wildevoelveli WWTP in 2007

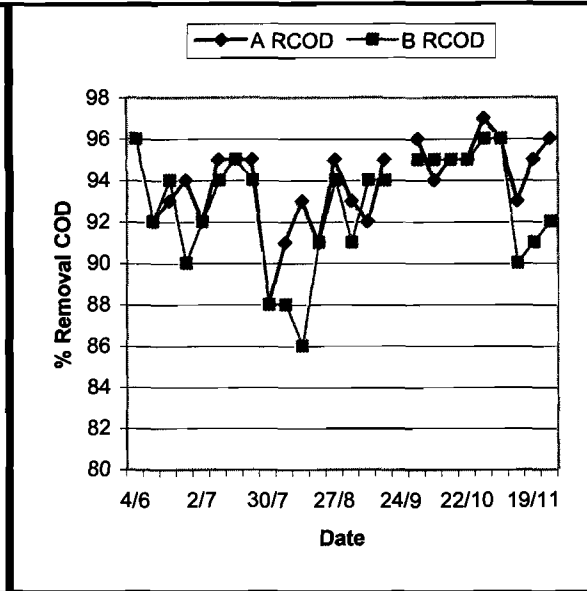


Figure 4.11.4 Graph showing COD removal efficiency from clarifier A (A RCOD) and clarifier B (B RCOD) at Wildevoelveli WWTP in 2007

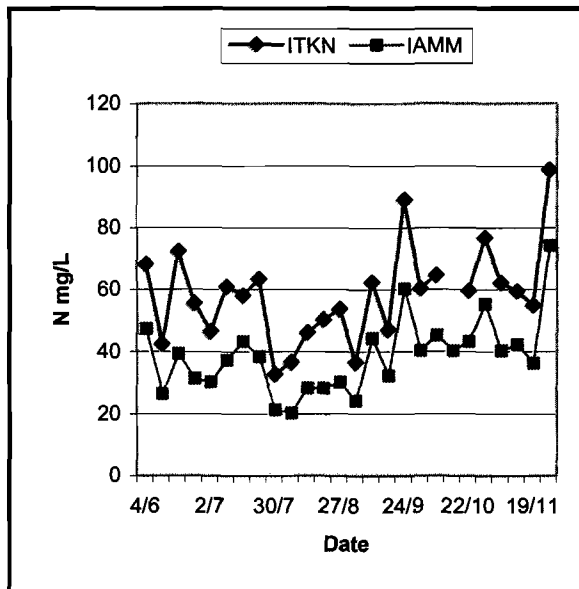


Figure 4.11.5 Graph showing the influent TKN (ITKN) and influent ammonia (IAMM) levels at Wildevoelveli WWTP in 2007

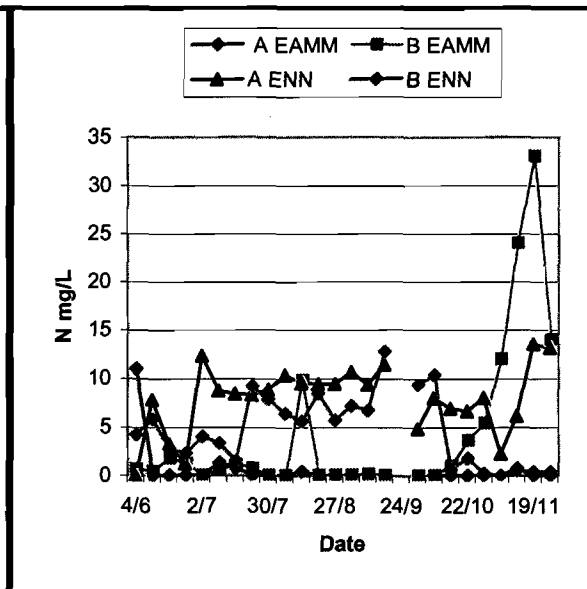


Figure 4.11.6 Graph showing the effluent ammonia levels from clarifier A (A EAMM) and B (B EAMM) and the effluent nitrates/nitrites from clarifier A (A ENN) and B (B ENN) at Wildevoelveli WWTP

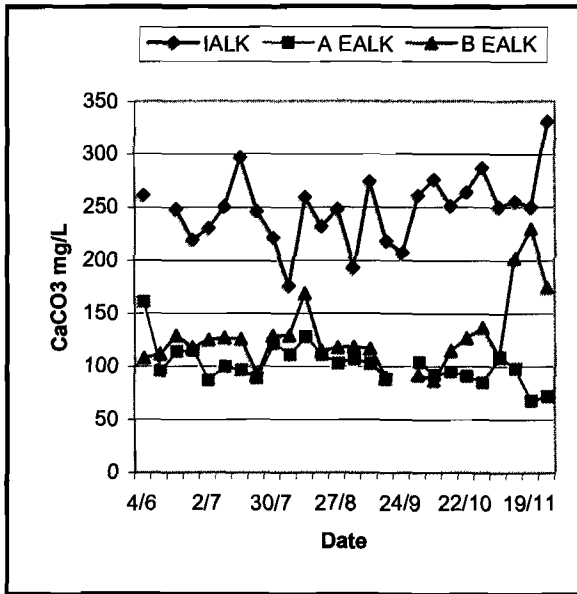


Figure 4.11.7 Graph depicting the influent alkalinity (IALK) and the effluent alkalinity from clarifier A (A EALK) and B (B EALK) at Wildevoelvie WWTP in 2007

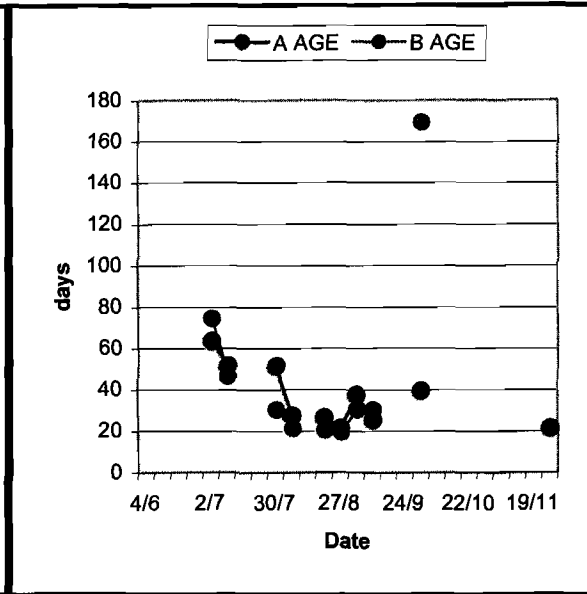


Figure 4.11.8 Graph showing the sludge age of the mixed liquor in reactor A (A AGE) and reactor G (B AGE) at Wildevoelvie WWTP in 2007

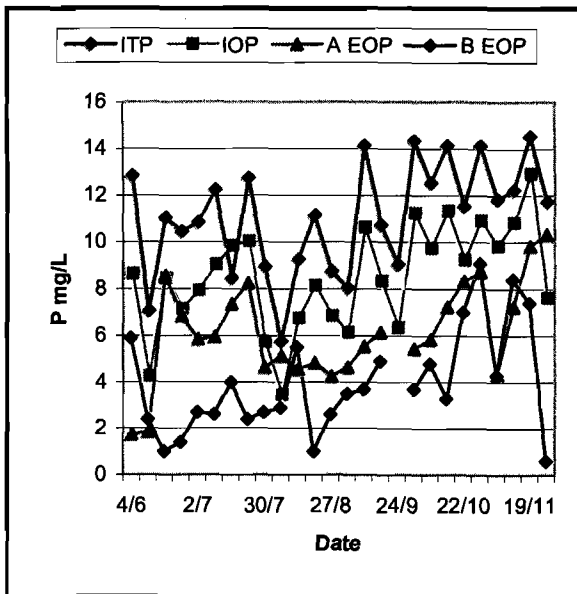


Figure 4.11.9 Graph showing the influent total phosphates (ITP) and influent α -phosphates (IOP) as well as the effluent α -phosphates from clarifier A (A EOP) and clarifier B (B EOP) at Wildevoelvie WWTP in 2007

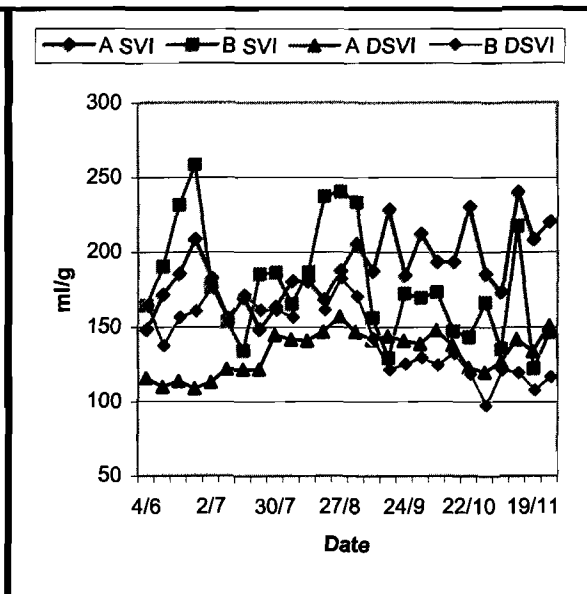


Figure 4.11.10 Graph showing the SVI from the mixed liquor of reactor A (A SVI) and B (B SVI) as well as the DSVI values from the mixed liquor of reactor A (A DSVI) and reactor B (B DSVI) at Wildevoelvie WWTP in 2007

| TABLE 4.11.1 FOOD TO MICROORGANISM (F/M) RATIO IN REACTOR A AND REACTOR B OF WILDEVOELVLEI WWTP IN 2007 | | |
|---------------------------------------------------------------------------------------------------------|----------------------|----------------------|
| | REACTOR A | REACTOR B |
| June | 0.10 mgCOD/mgVSS.day | 0.10 mgCOD/mgVSS.day |
| July | 0.18 mgCOD/mgVSS.day | 0.13 mgCOD/mgVSS.day |
| August | 0.12 mgCOD/mgVSS.day | 0.09 mgCOD/mgVSS.day |
| September | 0.12 mgCOD/mgVSS.day | 0.13 mgCOD/mgVSS.day |
| October | 0.32 mgCOD/mgVSS.day | 0.14 mgCOD/mgVSS.day |
| November | 0.32 mgCOD/mgVSS.day | 0.09 mgCOD/mgVSS.day |

4.11.2 (b) Microscopic sludge analysis

This was performed monthly on samples from reactor A and reactor B on the following dates: 26/6/07, 25/7/07, 21/8/07, 19/9/07, 24/10/07 and 27/11/07.

● Floc Structure

Reactor A

The flocs were round in shape, open in structure, firm in strength and medium in size from June to August. Although other parameters remained the same for the last three months, the shape of the flocs was both irregular and round. There was bridging between flocs from July to November and in October there were tangled masses of filaments.

Reactor B

In June, July, October and November the flocs were round in shape, open in structure firm in strength and medium in size. In August, the shape was irregular and in September, both irregular and round. Many filaments were in tangled masses from June to October and there was bridging in all samples.

● Diversity

Reactor A and reactor B

There was moderate diversity and scanty monocolonies were present in all samples.

● Filament index

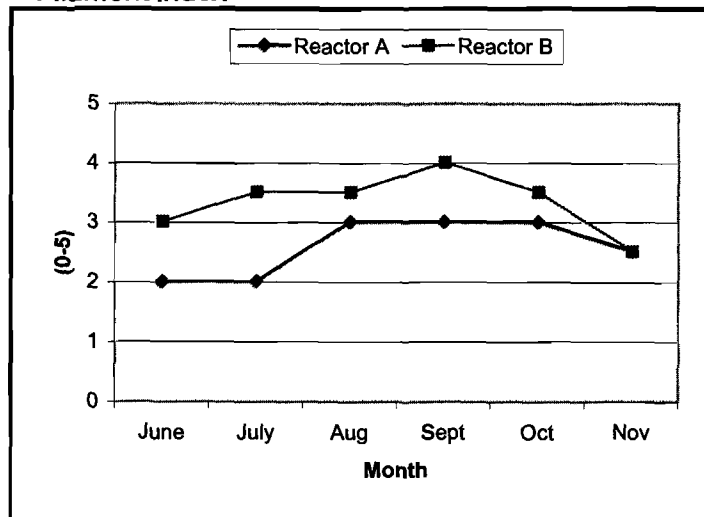


Figure 4.11.11 Graphs depicting the FI values obtained from the mixed liquor of reactor A and reactor B at Wildevoelvrei WWTP in 2007

TABLE 4.11.2 DOMINANT AND SECONDARY FILAMENTOUS ORGANISMS IDENTIFIED FROM THE MIXED LIQUOR OF REACTOR A AND REACTOR B AT WILDEVOELVLEI WWTP IN 2007

| AS DOMINANT FILAMENTOUS | | | | | | | | |
|--------------------------|------------|------------|------------|------------|------------|------------|---------------------------------|--------------------------------|
| | June | July | Aug | Sept | Oct | Nov | % Prevalence A and B (dominant) | % Prevalence B and C (overall) |
| Type 0092 | X (A,B) | X (A,B) | X (A,B) | X (A,B) | X (B) | X (B) | 83 | 100 |
| Actinomycetes | X (B) | X (A,B) | X (A) | X (A,B) | X (B) | | 58 | 100 |
| Type 1851 | | X (A) | | X (A,B) | X (A,B) | X (A,B) | 58 | 83 |
| <i>M. parvicella</i> | | | | X (B) | | X (B) | 17 | 67 |
| Type 021N | X (A) | | | | | | 8 | 92 |
| AS SECONDARY FILAMENTOUS | | | | | | | | |
| Type 0092 | | | | | X (A) | X (A) | } Also dominant filaments | |
| Actinomycetes | X (A) | | X (B) | | X (A) | X (A,B) | | |
| Type 1851 | | X (B) | X (A,B) | | | | | |
| <i>M. parvicella</i> | | X (A,B) | X (B) | X (A) | X (B) | X (A) | | |
| Type 021N | X (B) | X (A,B) | X (B) | X (A,B) | X (A,B) | X (A,B) | | |
| Type 0041 | X (B) | X (A) | X (A,B) | X (B) | X (A,B) | X (A,B) | 0 | 75 |
| <i>H. hydrosis</i> | | | | X (B) | X (A) | | 0 | 17 |
| Type 1701 | X (A) | | | | | | 0 | 8 |
| <i>Flexibacter</i> spp. | | X (B) | | | | | 0 | 8 |

TABLE 4.11.3 PROTOZOA/METAZOA IDENTIFIED IN THE MIXED LIQUOR FROM REACTOR B AND REACTOR C AT BORCHERDS QUARRY WWTP IN 2007

| SCANTY | | | | | | | |
|------------------|-------------------------|------------|------------|------------|----------|------------|------------|
| | | June | July | Aug | Sept | Oct | Nov |
| Sessile ciliates | <i>Carchesium</i> spp. | | | X (B) | | | |
| | <i>Epistylis</i> spp. | X (A,B) | | X (B) | X (B) | X (B) | X (B) |
| | <i>Opercularia</i> spp. | | X (B) | X (B) | | | |
| | <i>Vaginicola</i> spp. | | | X (A) | | X (A,B) | X (A) |
| | <i>Vorticella</i> spp. | X (B) | X (A,B) | X (A,B) | | X (B) | X (A,B) |

TABLE 4.11.3 (CONTINUED) PROTOZOA/METAZOA IDENTIFIED FROM THE MIXED LIQUOR OF REACTOR A AND REACTOR B AT WILDEVOELVLEI WWTP IN 2007

| | | June | July | Aug | Sept | Oct | Nov |
|----------------------|-----------------------------|------------|------------|------------|------------|------------|------------|
| Crawling ciliates | <i>Aspidisca</i> spp. | X (B) | X (A,B) | X (B) | X (A,B) | X (A,B) | X (A,B) |
| | <i>Trachelophyllum</i> spp. | | | X (A) | X (A) | X (A) | X (A) |
| Free-living ciliates | <i>Blepharisma</i> spp. | | | | X (A) | X (A) | |
| | <i>Euplotes</i> spp. | | X (B) | | | | X (A) |
| | <i>Litonotus</i> spp. | | X (A) | | | | |
| | <i>Colpidium</i> spp. | | | X (A) | | | |
| | <i>Spirostomum</i> spp. | | | X (A) | X (A) | X (A) | X (A) |
| | Unidentified | X (B) | | | | | X (A) |
| | <i>Bodo</i> spp. | X (A,B) | X (A) | X (A) | X (A,B) | X (B) | |
| Flagellates | <i>Hexamitus</i> spp. | | | | | | X (A) |
| | <i>Peranema</i> spp. | | X (A) | X (A) | X (B) | X (B) | X (A) |
| | <i>Pleuromonas</i> spp. | | | | | X (A) | |
| | <i>Trepomonas</i> spp. | | | | | X (B) | X (A) |
| | Unidentified | | X (B) | | X (A) | | |
| Amoebae | | X (A) | | X (A,B) | X (B) | X (B) | |
| Testate amoebae | | | | X | | | |
| Nematodes | | X (A) | | | | X (B) | |
| Rotifers | X (A,B) | X (A,B) | X (A,B) | X (A,B) | X (A,B) | | |
| Tardigrades | | | X (A) | X (B) | X (A,B) | X (A) | |
| Oligochaeta | | | | X (A) | | | |

| TABLE 4.11.3 (CONTINUED) PROTOZOA/METAZOA IDENTIFIED FROM THE MIXED LIQUOR OF REACTOR A AND REACTOR B AT WILDEVOELVLEI WWTP IN 2007 | | | | | | | |
|-------------------------------------------------------------------------------------------------------------------------------------|------------------------|----------|----------|----------|----------|-----|----------|
| MODERATE | | | | | | | |
| | | June | July | Aug | Sept | Oct | Nov |
| Attached ciliates | <i>Vaginicola</i> spp. | | | | X (A) | | |
| Crawling ciliates | <i>Aspidisca</i> spp. | X (A) | | X (A) | | | |
| Free-living ciliates | <i>Paramecium</i> spp. | | | | | | X (A) |
| Flagellates | <i>Bodo</i> spp. | | X (B) | X (B) | | | |
| | <i>Peranema</i> spp. | | | | | | X (B) |

3.11.3 DISCUSSION

Removal of COD at Wildevoelveli WWTP was good (figure 4.11.3 and 4.11.4), with results from reactor A being more consistent. Figure 4.11.6 and graph 4.11.7 show that nitrification and denitrification efficiency also differed between the two reactors. Reactor A produced mostly excellent results for nitrification, while the results from reactor B were inconsistent, being particularly poor from October. Loss of nitrification in this reactor was accompanied by increased effluent alkalinity as reflected in figure 4.11.7. Toxic poisoning of the nitrifier population can be ruled out, as reactor A was unaffected. Overloading is also ruled out, as the F/M (table 4.11.1) remained low throughout in reactor B (in fact, reactor A showed an increased F/M in October and November). Unfortunately, no MLVSS results were available for the calculation of F/M ratios in August and September, but results were extrapolated using the percentage of MLVSS to MLSS in other months. This figure was very consistent with a range of between 82 and 85%, with a mean of 83% for reactor A, and a range of 81 to 83%, with a mean of 82% for reactor B. The sludge age, depicted in graph 4.11.8 was also long enough to support the growth of sufficient numbers of nitrifiers. The most likely cause then of poor nitrification in reactor B would be insufficient DO.

The levels of residual nitrates/nitrites were also consistent in reactor A, (mostly between 5-10mgN/L) and erratic in reactor B. There were no residual nitrates/nitrites in the clarifier effluent from reactor B when there was loss of nitrification. This type of result was common in the WWTP's included in the study. The influent COD/TKN ratio ranged from 6.1-14.4mgCOD/mgTKN with a mean of 10.9mgCOD/mgTKN. The mean is thus higher than the value of 8.4mgCOD/TKN reported by Kujawa *et al.* (1996) as being necessary for complete denitrification, but the range fell on either side of this value. However, there was no correlation between the levels of effluent nitrates/nitrites and the COD/TKN ratio. In addition, complete denitrification is not expected in either of these types of reactors with only one anoxic zone. The most probable reasons for the intermittent presence of high levels of residual nitrates/nitrites would thus be plant operating conditions for example, insufficient mixed liquor recycle from the aerobic to the anoxic zone or insufficient retention time in the anoxic zone.

Phosphorous removal, shown in figure 4.11.9 from reactor A was almost consistently poor, while that from reactor B was generally better, but not entirely adequate. The reactor with the higher levels of nitrates/nitrites in the clarifier effluent was the worst performer in terms of phosphorous removal. Notwithstanding the fact that the recycle for the two reactors is different, it can be speculated once again that the phosphorous removal function was retarded, at least to some degree by the presence of nitrates in the anaerobic reactor. The influent COD/P ratio was in the range of 46.2 to 70.8mgCOD/mgP with a mean of 56.6mgCOD/mgP. At this level, according to Liu *et al.* (1996), the growth of GAO's over PAO's would be favoured.

The DSVI values shown in figure 4.11.10 were only slightly above 150mg/L mark during periods of August and November in reactor A. In reactor B, the values were above this level from June until August and subsequently decreased. The filament index was raised to a level suggestive of bulking for periods in both reactors - from August to October in reactor A and from June to October in reactor B. The FI, given in graph 4.11.11 was always higher in reactor B. There was often substantial bridging between the flocs and the filaments formed tangled masses. In this WWTP sludge settling was better than the amount of filamentous growth and the floc characteristics would suggest.

Regarding the filament species given in table 4.11.2, the presence of Type 0092 as a dominant filament does not affect settling as it tends to form the backbone of flocs and grows within the flocs. However, it would also not be included in the filament index evaluations for the same reason. Type 1851 was found as a dominant species in both reactors in more than half of the samples. The association of this filament as a dominant species not only with industrial effluent (as expected), but also with domestic effluent was a feature of the study. The presence of Type 1851 correlated with the presence of bulking conditions according to the meta-analysis, but only to a moderate degree. The correlation of actinomycetes with bulking conditions in the meta-analysis was significant. This organism was also present as a dominant species in more than half of the samples from Wildevoelviei WWTP. The presence of large number of filaments associated with bulking conditions with only moderately increased DSVI values presents an anomaly. It can be speculated that there were other unknown factors present in the wastewater that enhanced settling even under these circumstances.

Although the F/M values were similar in June and July, there was a marked difference in October and November. The presence of *M. parvicella* and actinomycetes suggests high levels of dietary fats in the influent. Type 0092 and Type 1851 are considered as low F/M by Jenkins *et al.* (2004). Type 1851 was present as a dominant species in both reactors in October and November. Type 0092 was present in both reactors as a dominant species until September, after which it only present as a dominant species in reactor B. This establishes an excellent correlation between type 0092 and low F/M conditions. Filament composition was similar for both reactors, reflecting that in this case, influent composition was more important than plant configuration for species succession.

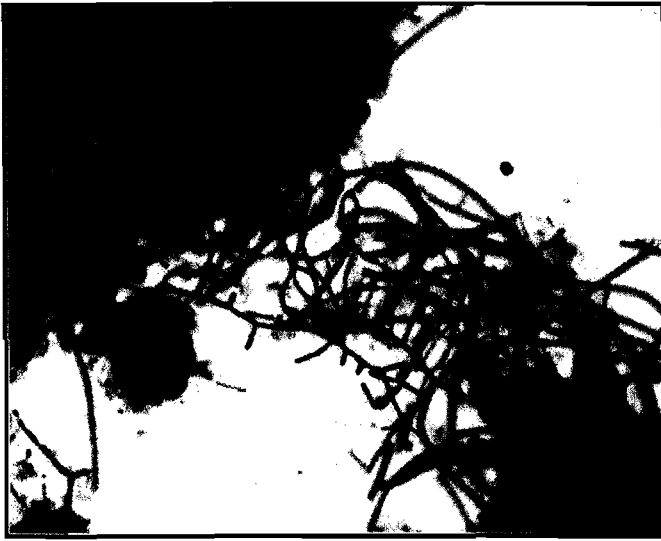


Figure 4.11.12 Gram stain of Gram negative (pink) Type 0092 and Gram positive Blue) branching actinomycetes from mixed liquor sample of Wildevoelwei WWTP taken from reactor A in November 2007.

Regarding the protozoa and metazoa - there were substantial differences between ciliates and flagellates. The identification results are given in table 4.11.3. Reactor B harboured a greater variety of sessile ciliates and reactor A a greater variety of free-living ciliates. In reactor B in July and August, the flagellates outnumbered the ciliate population. This usually indicates a high sludge load and/or a lack of DO. The most abundant flagellate was *Bodo* spp., which is an indicator organism for the same conditions (Eikelboom, 2000).

The calculated F/M for reactor B was low throughout the study period, so lack of DO must be considered. However, nitrification and COD removal remained satisfactory during this period and only deteriorated at a later date. One speculative explanation is that there was a chronic lack of DO that was not initially apparent by analysis of the chemical results but was reflected in the protozoan and metazoan population. Furthermore, there may have been just sufficient DO to achieve satisfactory COD removal and nitrification until the influent COD increased from the end of September. At this stage, the amount of available DO became insufficient to achieve both COD removal and nitrification and the nitrifier population was adversely affected, leading to high effluent ammonia.

In summary, reactor A functioned in a more stable manner than reactor B in terms of nutrient removal. There was a period of high effluent ammonia in reactor B, probably due to lack of DO resulting in a retarded nitrifier population. Phosphorous removal was adversely affected, ostensibly by a high COD/P ratio and the presence of nitrates in the anaerobic zone. The microscopic sludge quality relating to floc character and filament index suggested bulking conditions to a greater extent than the DSVI values would imply. The filament population was similar in both reactors but the protozoan population in reactor B pointed to low DO in this reactor.

CHAPTER 5
META-ANALYSIS OF COMBINED RESULTS
FROM WASTEWATER TREATMENT PLANTS

5.1 POPULATION DYNAMICS OF FILAMENTOUS ORGANISMS FROM JUNE TO NOVEMBER 2007

5.1.1 Results

5.1.1 (a) Prevalence and comparison with similar South African studies

- Sample distribution

Number of samples: 96

Number of reactors: 16

Number of WWTP's: 11

16 Filament species found as dominant or secondary species.

- Prevalence

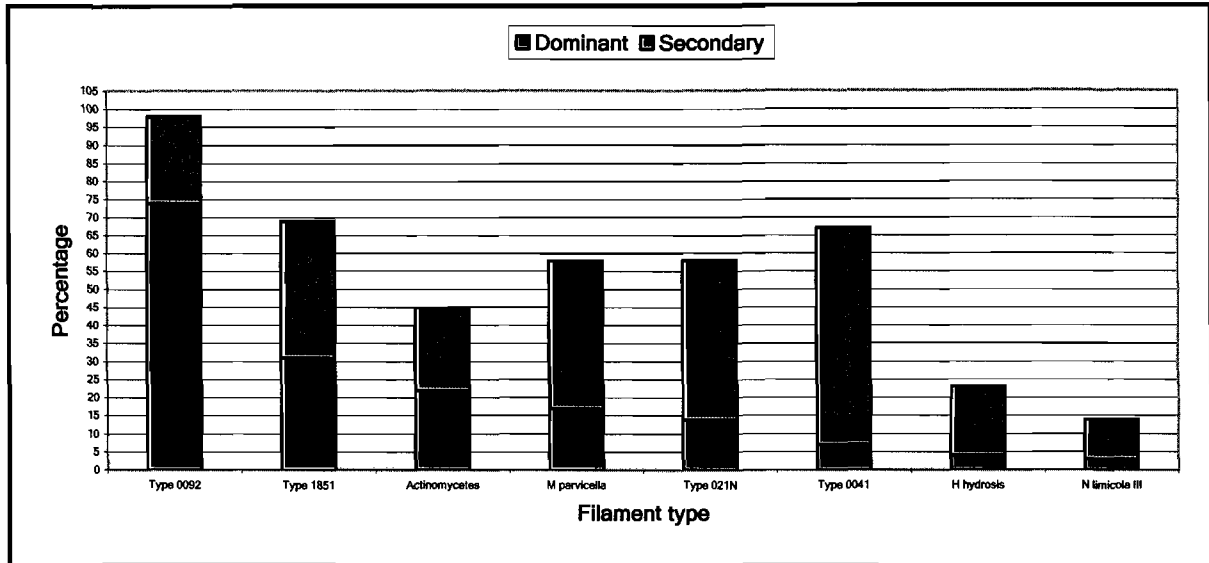


Figure 5.1.1 This bar chart depicts the percentage prevalence of dominant filamentous microorganisms in the 96 samples examined during the course of the study. The prevalence of these organisms as secondary species is also represented.

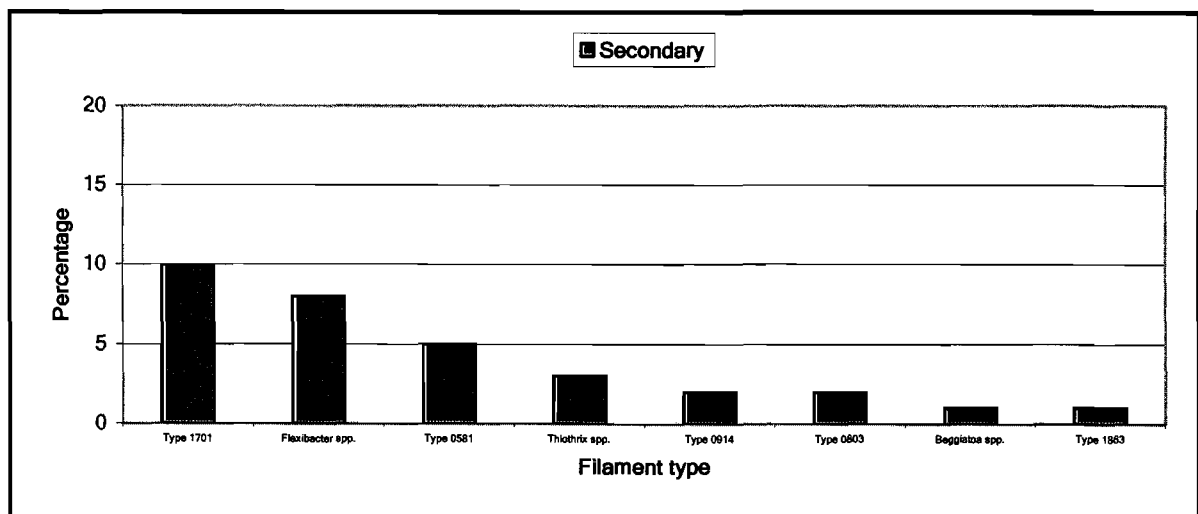


Figure 5.1.2 Percentage prevalence of filamentous microorganisms that were identified only as secondary species during the study period

● Comparison with other filamentous surveys conducted in South Africa

Two surveys for filamentous organisms in South Africa were found in the literature, one from KwaZulu-Natal (KZN), conducted in 1999 (Lacko *et al.*, 1999) and the other from nutrient removal plants distributed over the country (SA), conducted in 1986 (Blackbeard *et al.*, 1988). In the KZN study, mixed liquor samples were taken from 5 nutrient removal plants in the KwaZulu-Natal (KZN) area every fortnight from May-October in the late 1990's. Filaments were identified and data was analyzed according to frequency of occurrence (prevalence) and abundance (Lacko *et al.*, 1999). A decade earlier, Blackbeard *et al.* (1988), examined the mixed liquor of 33 nutrient removal plants from over South Africa. At the time, nutrient removal was in its infancy, and the scope of the study represented three quarters of these plants operating in the country at the time. In addition, the presence of bulking sludge was extremely common compared to the situation today. The reason is that adjustments to operating parameters to deter bulking in nutrient removal plants have been instituted over the years - evidence of the causes of the bulking phenomenon have been uncovered using data from pilot scale laboratory observations and empirical data from functioning WWTP's. Unfortunately, the geographical distribution of the plants is not given, so comparisons in terms of filament abundance and location cannot be made.

| FILAMENT | KZN (OP) 1999 | SA (OP) 1986 | SA (D) 1986 | CT (OP) 2007 | CT (D) 2007 |
|-----------------------------------------|------------------|-----------------|----------------|-----------------|----------------|
| Type 0092 | 0 | 94 | 82 | 98 | 74 |
| Type 1851 | 100 | 58 | 21 | 69 | 31 |
| <i>M. parvicella</i> | 100 | 76 | 33 | 58 | 17 |
| Type 0041 | 67 | 85 | 39 | 67 | 7 |
| Type 021N | 50 | 0 | 0 | 58 | 14 |
| Actinomycetes/ <i>Nocardia</i> spp * | 67 | 24 | 15 | 45 | 22 |
| <i>H. hydrosis</i> | 33 | 21 | 12 | 23 | 4 |
| <i>N. limicola</i> III/III ** | 83 | 21 | 6 | 14 | 3 |
| Type 1701 | 50 | 0 | 0 | 10 | 0 |
| <i>Flexibacter</i> spp. | 0 | 0 | 0 | 8 | 0 |
| Type 0581 | 0 | 0 | 0 | 5 | 0 |
| <i>Thiothrix</i> spp.*** | 50 | 6 | 3 | 3 | 0 |
| Type 0914 | 50 | 70 | 33 | 2 | 0 |
| Type 0803 | 0 | 27 | 17 | 2 | 0 |
| <i>Beggiatoa</i> spp. | 33 | 0 | 0 | 1 | 0 |
| Type 1863 | 0 | 9 | 6 | 1 | 0 |
| Type 1702 | 0 | 3 | 0 | 0 | 0 |
| Type 0675 **** | 67 | 73 | 45 | Type 0041 | Type 0041 |
| Type 0961 | 0 | 3 | 0 | 0 | 0 |
| <i>S. natans</i> | 33 | 0 | 0 | 0 | 0 |

*Eikelboom (2000) prefers the term Actinomycetes, Jenkins *et al.* (2004) use the term *Nocardia* spp. for this group of organisms with similar morphological characteristics.

**Eikelboom (2000) no longer classifies *N. limicola* II as a separate entity. Both *N. limicola* II and *N. limicola* III are now classed as *N. limicola* III that includes all strains with similar morphological characteristics. Blackbeard *et al.* (1986) do not separate *N. limicola* I, II or III, merely naming *N. limicola* per se.

***Lacko *et al.* (1999) separate *Thiothrix* I (17% occurrence and dominance) and *Thiothrix* II (33% occurrence and dominance). As they were found in different plants, the percentages have been added in the above table.

****Eikelboom (2000) now classifies Type 0675 as part of Type 0041.

Table 5.1.1 shows that the results for certain filaments differed markedly from those in the Cape Town 2007 study and some of the more notable discrepancies are discussed more fully in section 5.1.2. that deals with filamentous microorganisms that were found in more than 2% of the study. However, there were four filaments that were found in significant numbers in one or both of the previous studies (Lacko *et al.*, 1999 and Blackbeard *et al.*, 1988) and were scanty or absent in this study:

- Type 0914 was prevalent in 50% of the samples examined by Lacko *et al.* (1999) and 33% of samples examined by Blackbeard *et al.* (1988). This organism occurs in low loaded plants with H₂S in the influent (Eikelboom, 2000).
- Type 0803 was prevalent only in the study conducted by Blackbeard *et al.* (1988). It is a filament commonly found in industrial wastewater (Eikelboom, 2000).
- *Beggiatoa* spp. was prevalent in the study by Lacko *et al.* (1999). This filament is an indicator species for the presence of many reduced sulphides and/or low DO conditions (Eikelboom, 2000). The 3 organisms, namely Type 0914, Type 0803 and *Beggiatoa* spp. have similar requirements. Conditions in the Cape Town WWTPs that were examined were probably not conducive to the growth of these filaments.
- *Sphaerotilus natans* was found in 33% of the samples examined by Lacko *et al.* (1999), but only from one of the WWTPs included in their study. According to Eikelboom (2000), this previously problematic filament is rare in modern nutrient removal plants. The filament enjoys moderate loading conditions. It is associated with nutrient deficiency (including oxygen) and complete mixing in the aerobic zone (Eikelboom, 2000). In addition, the filament enjoys the presence of high amounts of LMW substances. Lacko *et al.* (1999) postulate that the presence of large amounts of complex carbohydrates in the wastewater from a nearby brewery may have led to the proliferation of this organism.

This highlights that different populations occur in WWTPs in different geographical locations and that results from one area cannot necessarily be extrapolated to another. For example, the mining and industry in Gauteng will necessitate a larger industrial component in the WWTPs in comparison to the Western Cape.

5.1.1 (b) Correlation with DSVI and FI

Eikelboom, (2000) deduced the effect of different filament types on SVI by data analysis, as outlined in table 5.1.2.

| SIGNIFICANT EFFECT | MODERATE EFFECT | MINIMAL EFFECT |
|---------------------------------------------------------------------------------------------|----------------------------------------------------------------------------------------------|---------------------------------|
| <i>M. parvicella</i> <i>S. natans</i> <i>Thiothrix</i> spp. Type 021N Type 1851 | Actinomycetes <i>H. hydrosis</i> Type 0041 Type 0092 Type 0803/0914 Type 1701 | <i>N. limicola</i> Type 1863 |

- The effect may be enhanced for Type 0092 and Type 0041 in industrial WWTP's.
- Eikelboom (2000) uses the wording "large, medium and small" not significant, moderate and minimal for the effect on filament type.

For this study DSVI was chosen over the SVI as it is deemed to be a better indicator of bulking conditions (Jenkins *et al.*, 2004). Deterioration of settling is often seen with an FI of 3 or above. Although the FI is a measure of the number of filaments present, it does not take into account filaments that are within the flocs (Eikelboom, 2000). Correlation between SVI/DSVI and FI is not always consistent because settling is also affected by other factors besides the number of filaments (Eikelboom, 2000). A figure of ≥ 150 ml/g for DSVI and FI of ≥ 3 was deemed to be indicative of bulking conditions being present. Eight out of 16 reactors (50%) in 7 of the 11 WWTP's (64%) had DSVI's that were ≥ 150 ml/g for at least one month during the study period. Seven of the 16 reactors (44%) in 6 of the 11 WWTP's (54%) had FI's that were ≥ 3 for at least one month during the same period. Although DSVI's were performed on a weekly basis, the FI's were only performed monthly. If the DSVI was ≥ 150 ml/g at any time during a given month, it was included. In order that the two parameters could be compared, each of the six months constituted one "sample" for each WWTP, giving a total of 96 samples for both the FI and the DSVI.

5.1.1 (bi) DSVI results

The results when the DSVI was ≥ 150 ml/g are given below. This was the case for 24% of the samples.

| | June | July | Aug | Sept | Oct | Nov |
|--------------------|------|------|-----|------|-----|-----|
| Athlone A | | | | | X | X |
| Bellville N | | | | | X | X |
| Borcherds quarry B | | | | X | X | X |
| Macassar 2 | X | | X | X | | X |
| Parow | X | X | X | X | X | X |
| Potsdam | | X | X | | | |
| Wildevoëlvei A | | | X | | | |
| Wildevoëlvei B | X | X | X | | | |

*The figures in red denote the samples where the FI results were not ≥ 3 (30%)

| Filament type | % Prevalence DSVI ≥ 150 ml/g | % Overall Dominance | Monthly distribution | | | | | |
|------------------------|-----------------------------------|---------------------|----------------------|------|-----|------|-----|-----|
| | | | June | July | Aug | Sept | Oct | Nov |
| Type 0092 | 61 | 74 | 2 | 3 | 4 | 2 | 1 | 2 |
| Actinomycete | 43 | 22 | 2 | 2 | 2 | 1 | 1 | 2 |
| Type 1851 | 35 | 31 | 1 | 1 | 1 | 1 | 2 | 1 |
| Type 021N | 26 | 14 | 1 | | | | 2 | 3 |
| <i>M. parvicella</i> | 13 | 17 | | 1 | 1 | 1 | | |
| Type 0041 | 9 | 7 | 1 | 1 | | | | |
| <i>H. Hydrosis</i> | 4 | 4 | | | | 1 | | |
| <i>N. limicola</i> III | 4 | 3 | | | | | | 1 |

*For overall dominance refer to figure 5.1.1

5.1.1 (bii) FI results

The results when the FI ≥ 3 are given below. This was the case for 28% of the samples.

| | June | July | Aug | Sept | Oct | Nov |
|--------------------|------|------|-----|------|-----|-----|
| Athlone A | | X | X | X | X | X |
| Bellville N | | | | | X | X |
| Borcherds quarry B | | | | X | X | X |
| Parow | | | X | X | X | X |
| Postdam | X | X | | X | X | X |
| Wildevoëlvei A | | | X | X | X | |
| Wildevoëlvei B | X | X | X | X | X | |

*The figures in red denote the samples where the DSVI results were not ≥ 150 ml/g (41%)

| Filament type | % Prevalence FI ≥ 3 | % Overall Dominance | Monthly distribution | | | | | |
|------------------------|-----------------------------|------------------------|----------------------|------|-----|------|-----|-----|
| | | | June | July | Aug | Sept | Oct | Nov |
| Type 0092 | 63 | 74 | 2 | 2 | 3 | 4 | 3 | 3 |
| Type 1851 | 44 | 31 | 1 | | 2 | 4 | 4 | 1 |
| Actinomycete | 33 | 22 | 1 | 1 | 2 | 2 | 2 | 1 |
| Type 021N | 30 | 14 | | 1 | 1 | 1 | 2 | 3 |
| <i>M. parvicella</i> | 7 | 17 | | 1 | | 1 | | |
| <i>N. limicola</i> III | 4 | 3 | | | | | | 1 |

*For overall dominance refer to figure 5.1.1

5.1.1.2 (bii) Combined FI and DSVI results

The results of the dominant filaments encountered when the DSVI values were ≥ 150 ml/g and the FI values were ≥ 3 are given in the table below (5.1.7). This occurred in 17% of samples.

| Filament type | % Prevalence DSVI ≥ 150 ml/g and FI ≥ 3 | % Overall Dominance | Monthly distribution | | | | | |
|------------------------|---------------------------------------------------------|------------------------|----------------------|------|-----|------|-----|-----|
| | | | June | July | Aug | Sept | Oct | Nov |
| Type 0092 | 63 | 74 | 1 | 2 | 3 | 1 | 1 | 2 |
| Actinomycete | 38 | 22 | 1 | 1 | 2 | | 1 | 1 |
| Type 021N | 38 | 14 | | | | 1 | 2 | 3 |
| Type 1851 | 31 | 31 | | | 1 | 1 | 2 | 1 |
| <i>M. parvicella</i> | 6 | 17 | | 1 | | | | |
| <i>N. limicola</i> III | 6 | 3 | | | | | | 1 |

The results of the dominant filaments encountered when the DSVI values were ≥ 150 ml/g or the FI values were ≥ 3 are given in the table below (5.1.8). This occurred in 35% of samples.

| Filament type | % Prevalence DSVI ≥ 150 ml/g or FI ≥ 3 | % Overall Dominance | Monthly distribution | | | | | |
|------------------------|-----------------------------------------------------------|------------------------|----------------------|------|-----|------|-----|-----|
| | | | June | July | Aug | Sept | Oct | Nov |
| Type 0092 | 62 | 74 | 3 | 3 | 4 | 5 | 3 | 3 |
| Actinomycete | 38 | 22 | 2 | 2 | 2 | 3 | 2 | 2 |
| Type 1851 | 44 | 31 | 1 | 2 | 2 | 4 | 4 | 2 |
| Type 021N | 24 | 14 | | 1 | 1 | 1 | 2 | 3 |
| <i>M. parvicella</i> | 15 | 17 | 1 | | 1 | 2 | | 1 |
| Type 0041 | 6 | 7 | 1 | 1 | | | | |
| <i>H. Hydrosis</i> | 3 | 4 | | | | 1 | | |
| <i>N. limicola</i> III | 3 | 3 | | | | | | 1 |

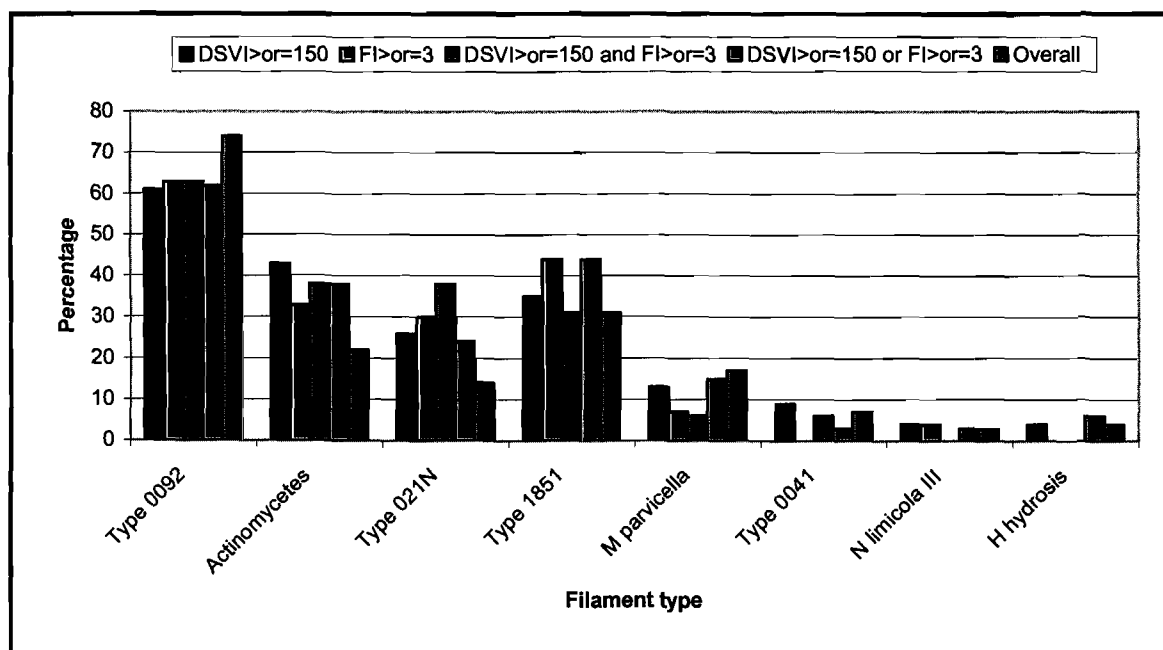


Figure 5.1.3 Bar chart showing the relationship between elevated FI and/or DSVI and filament type

The association depicted by figure 5.1.3 was made in an attempt to ascertain whether certain filament types were more likely to be involved in the advent of bulking conditions in the Cape Town WWTPs. For example, the overall percentage of samples containing Type 0092 as a dominant species was less than those samples with an elevated DSVI and/or FI by more than 10% in each case, so the association of Type 0092 with bulking tendencies is low. In contrast, using the same logic, the association of Type 021N with bulking conditions is high.

5.1.2 Discussion

Twelve of the sixteen filament types found are discussed, in order of prevalence. Four filaments that were prevalent in 2% or less of samples and never dominant have been omitted. These are Type 0914, Type 0803, *Beggiatoa* spp. and Type 1863 and are mentioned in section 5.1.1 and chapter 4 if significant. Filaments are discussed in light of the overall findings in this chapter, and together with plant configuration and operating parameters under the individual plant analyses in chapter 4.

The discussion will take place for each filament under three headings, namely:

- Documented organism characteristics
- Occurrence in selected WWTP's in Cape Town from June-November 2007
- Analysis of findings

5.1.2 (a) Type 0092

• Documented organism characteristics

Very little is known about the physiology of this organism. The bacterium grows within the flocs, so it does not have a profound effect on the DSVI (Eikelboom, 2000). Logically, this would then also hold true for filament index. Type 0092 is associated with low sludge loading levels and has been termed as a "low F/M" filament. In his manual, Jenkins *et al.* (2004) gives growth parameters of MCRT's of between 8 and 50 days and a F/M of between 0.05 and <0.2 kgBOD₅/kgMLSS/day, the latter being similar to the loading rates documented by Eikelboom, (2000).

Evidence points to that fact that it has the ability to utilize slowly biodegradeable chemical oxygen demand (SBCOD) for growth and metabolism. Particulates and slowly metabolizable substrates are not readily degraded in the anaerobic or anoxic zones. In the aerobic zones of biological nutrient removal (BNR) WWTPs, they are hydrolyzed to produce low concentrations of soluble organics. It is thought that this source of nutrient supply favours the growth of Type 0092 (amongst other filaments).

Eikelboom, (2000) suggests that *M. parvicella* and Type 0092 have similar substrate requirements, but different temperature requirements - at very high MCRT's, the latter will predominate at temperatures over 15°C and the latter at lower temperatures. He also reports an association between raw influent and the growth of this organism.

In a study by Noutsopoulos *et al.* (2006) it was found that although the reduction in sludge age below six days at <18°C suppressed the growth of *M. parvicella*, it did not affect the growth of Type 0092. The organic loading range used in these experiments was 0.19-0.45 kgCOD/kgVSS.day, thus conflicting with the categorization of this filament as strictly low F/M.

• Occurrence in selected WWTP's in Cape Town from June-November 2007

Percentages given are from monthly analyses and are given in sequential chronological order from June to November 2007. Type 0092 was the most frequently encountered as well as the most frequently dominant species.

Type 0092 was ubiquitous, being found in between 94% (June and October) and 100% (July, August, September, November) of samples. From June to November 2007, it was dominant or co-dominant in 75%, 81%, 75%, 81%, 56% and 75% of samples. However,

as can be seen from figure 5.1.3, in samples that were deemed to be prone to bulking by virtue of elevated DSVI and/or FI, this filament was only found in 61-63% of the samples.

- **Analysis of findings**

Figure 5.1.3 shows that Type 0092 was in terms of frequency overwhelmingly the most common filament both in the elevated DSVI and/or FI (bulking) and overall categories. However, this was not strongly related to elevated DSVI or FI, as the figures derived for these conditions were consistently more than 10% less for those of overall dominance of this filament. Thus, in this study it can be said that the effect of Type 0092 on DSVI is minimal and not in agreement with Eikelboom (2000). (Refer to table 5.1.2). However, as previously stated and generally accepted, DSVI is a more reliable indicator of bulking than SVI. In addition, no conditions are given for the derived data found in table 5.1.2.

Furthermore, it can be deduced that there is no obvious change in the prevalence of this organism from winter to summer in Cape Town. All of the WWTP's included in the study were nutrient removal plants with high mean cells residence times (MCRT's) and low loading levels. This supports the theory that this filament enjoys conditions of low F/M and can utilize SBCOD for growth and metabolism. Given the Mediterranean climatic conditions in Cape Town, the temperature of the activated sludge is unlikely to decrease to the point where growth of Type 0092 is inhibited. This is borne out by the fact that growth was prodigious throughout the duration of the study from mid-winter to summer. It is not known what the situation would be when conditions had been warmer for extended periods as the study did not continue into midsummer. Thus, the seasonal variation with temperature variation seen in other parts of the world can neither be categorically proven nor refuted, although evidence does suggest that Cape Town WWTP's will not experience the same patterns as those found in colder climates.

This filament was found in the study by Blackbeard *et al.* (1988) as the most common organism, but was not found at all in the study conducted in KwaZulu-Natal. Some possibilities for this finding are outlined in the paragraphs below.

The influent characteristics and/or climatic conditions experienced in KwaZulu-Natal are sufficiently different to those experienced in Cape Town to allow for the ubiquitous growth only in the latter area. This is unlikely to occur for the entire year, especially as Type 0092 is deemed to prefer warmer conditions.

There are discrepancies with the differentiation of *N. limicola* and Type 0092. This is the idea forwarded by Lacko *et al.* (1999) in their paper on the topic, and is discussed further under the section on *N. limicola* in section 5.2.1.8.

5.1.2 (b) Type 1851

- **Documented organism characteristics**

This filament is also present in low loaded plants. Eikelboom, (2000) gives a preferential sludge load of <0.15 kgBOD₅/kgMLSS/day. According to Jenkins *et al.* (2004), growth of Type 1851 may be suppressed by the use of a selector. Anoxic or anaerobic conditions and compartmentalization of the selector enhance this phenomenon. Intermittent feeding has also been shown to suppress the growth of this filament. According to Eikelboom (2000), Type 1851 is commonly found, but almost never dominates in domestic WWTP's and he also states that it can be present in large numbers in industrial plants especially from the agro industry, where there is an abundance of low molecular weight (LMW) compounds.



Figure 5.1.4 Gram stain of type 1851 from Bellville WWTP.

- Occurrence in selected WWTP's in Cape Town from June-November 2007

This filament was the second most common and the second most dominant filament encountered. From June to November 2007, it was found in 31%, 62%, 88%, 81%, 81% and 69% of samples and was dominant in 19%, 25%, 19%, 38%, 44% and 44% respectively.

- Analysis of findings

There was an upward trend in the occurrence of this organism from June to August. There is no obvious reason for this occurrence.

Type 1851 was found to be dominant in a number of domestic WWTP's in Cape Town. This is discussed under the pertinent individual plant analyses in chapter 4. It was also a common isolate in the study by Lacko *et al.* (1999) and Blackbeard *et al.* (1988) and was dominant in about one third of the samples that were positive for the presence of Type 1851.

From figure 5.1.3 it can be deduced that this organism is slightly more likely to be found in samples that are associated with bulking conditions in terms of elevated DSVI and/or FI as the prevalence in these samples is higher than the overall dominance in most categories. However, the effect can be classed as moderate, which conflicts with that of Eikelboom, (2000) selected in table 5.1.2, namely a large effect on SVI. It must be reiterated that DSVI is a better indicator of bulking conditions than SVI and that conditions for the data in table B are unknown.

5.1.2 (c) Type 0041

- Documented organism characteristics

Like Type 0092, 0041 can grow on SBCOD substrate and can thus be problematic in nutrient removal plants. Pre-mixing of influent and RAS before aeration allows this organism to grow on the particulate fraction. It is classed by both Jenkins *et al.* (2004) and Eikelboom (2000) as preferring conditions of low F/M.

- Occurrence in selected WWTP's in Cape Town from June-November 2007

Type 0041 was the third most commonly encountered and the sixth most dominant filament. From June to November 2007, this organism was found in 31%, 43%, 88%,

75%, 88% and 75% of samples, and was dominant in 13%, 13%, 6%, 6%, 0% and 6% respectively.

- Analysis of findings

This filament was commonly encountered in all three South African studies (see table 5.1.1). There appeared to be a spring maximum in the prevalence of this organism, but levels of dominance did not vary considerably. Values of overall dominance are slightly greater than those for elevated DSVI and/or FI in the Cape Town plants, agreeing with Eikelboom's (2000) categorization of this organism as having a moderate effect on SVI. However, these results must be taken with caution, as numbers were low.

5.1.2 (d) *Microthrix parvicella*

- Documented organism characteristics

M. parvicella is found commonly in domestic low F/M WWTP's and is the most common cause of bulking sludge and scum formation in nutrient removal plants in many countries. In colder climates, there is a winter growth maximum, as this filament prefers temperatures of <15°C for optimal growth. Domestic wastewater, and other wastewaters containing higher fatty acids e.g. oleic acid provide selective nutrition. Nielsen *et al.* (2002) have postulated that surface lipases allow *M. parvicella* to take up and store long chain fatty acids under anoxic as well as anaerobic conditions. Under these conditions, the organism thus has an advantage because of the slow hydrolysis rates of particulate organics by the rest of the biota.

Growth can also be supported by simple carbon substrates such as glucose, acetate and ethanol. Long retention times in sewers, PST's or anaerobic zones allow for hydrolysis of fats into fatty acids, therefore creating conditions where this organism can proliferate. Conditions of low DO in the aerobic reactor or a large anoxic mass fraction also contribute to overgrowth of this problem species. The presence of reduced sulphur and nitrogen, as may be found when recycling water from dewatering units, is another factor that has been implicated (Jenkins *et al.* 2004).

In 2006, Noutsopoulos *et al.* conducted a study to determine the effect of solids retention time on the growth of *M. parvicella*. They used a series of bench-scale continuous-flow experiments and growth kinetic studies. The inoculum consisted of mixed liquor from plants experiencing bulking and foaming problems supplemented with oleic acid in the form of Tween 80. The latter was employed to enhance the proliferation of *M. parvicella*. The experiments were only conducted in the temperature range of 14°C to 18°C. The authors deduced that the maximum sludge age to avoid foaming caused by *M. parvicella* at these temperatures was 6 days. The filaments were shorter and did not stain uniformly with the Gram method under these conditions. The loss of hydrophobicity alleviated the foaming problems, but significant growth suppression was only achieved at sludge ages of less than 5.7 days.

The kinetic studies were conducted according to the experimental protocol of Ekama *et al.* (1986). They found that the biomass with a high proportion of *M. parvicella* gave low but stable μ_{\max} values. With a decreasing *M. parvicella* to floc-former ratio, there was a concurrent increase in μ_{\max} . Thus the results of kinetic studies of biomass with a large *M. parvicella* count were deemed to be attributable to this filament as opposed to the floc-formers that were chiefly responsible for results at low *M. parvicella* counts. The low μ_{\max} values for *M. parvicella* of 0.67 1/d and 0.53 1/d for aerobic and anoxic conditions respectively were determined. The maintenance co-efficient for *M. parvicella* was 30% lower than that of the floc-formers, reflecting the fact that this filament needs significantly less energy to perform its basic biochemical processes. This gives the former an

advantage under the starvation conditions encountered in nutrient removal systems. In practice, there is a major problem with decreasing MCRT to control bulking and foaming and that is the adverse affect on nitrification.

- **Occurrence in selected WWTP's in Cape Town from June-November 2007**

Together with Type 021N, this classically problem filament was the fourth most abundant organism encountered. It was also the fourth most dominant organism identified in the samples. From June to November 2007, it was present in 13%, 56%, 75%, 81%, 69% and 56% of samples and was dominant in 13%, 19%, 19%, 25%, 13% and 25% respectively.

- **Analysis of findings**

Apart from the low frequency in June there was no obvious change in the occurrence of this organism from winter to summer in Cape Town. Given the Mediterranean climatic conditions in Cape Town, the temperature of the activated sludge is unlikely to decrease to the point where growth of *M. parvicella* is enhanced. This is assuming that other criteria for the growth of this organism are met and is borne out by the fact that *M. parvicella* was present throughout the duration of the study from mid-winter to summer. It is not known whether there would be less growth during extended warmer periods, as the study did not continue into midsummer. Thus, the seasonal variation with temperature variation seen in other parts of the world can neither be categorically proven nor refuted, although evidence does suggest that Cape Town WWTP's will not experience the same patterns as those found in colder climates.

In the KZN study (Lacko *et al.*, 1999), the organism was found in 100% of mixed liquor samples, but was never dominant. In the foam samples, it was found in 3 of the five WWTP's and exhibited a preference for the winter months in two of these. The climate in that region is sub-tropical and the temperatures are warmer throughout the year than those experienced in the Western Cape. In KZN, this filament was never dominant, probably because temperatures were never low enough to allow abundant growth, and in summer it is possible that temperatures were too high to allow for any growth at all in foam samples. It can be postulated that the Cape Town temperatures are not cold enough to allow for prolific growth in winter, neither are they warm enough to prevent growth in early summer.

The findings in the 1986 survey by Blackbeard *et al.* (1988) mirror those of the latest Cape Town survey in terms of the percentage of dominance from "positive" samples.

The expected bulking problems associated with this organism were not apparent. From figure 5.3, it can be seen that the prevalence in samples with elevated DSVI's and FI's, was less than the overall prevalence. Thus, in the Cape Town WWTP's, association can be classed as minimal, and not as significant (Table 5.1.2). If, as postulated, prevailing temperatures prevented mass overgrowth, it would explain this phenomenon.

5.1.2 (e) Type 021N

- **Documented organism characteristics**

This strict aerobe can grow over a broad range of loading levels. According to Eikelboom (2000) it is more likely to grow in massive amounts if the load is >0.1 kgBOD/kgMLSS.day. Growth is promoted if the influent contains easily biodegradable compounds (such as fatty acids) that can arise in the sewer or in industrial effluent. Type 021N can utilize hydrogen sulphide as a source of energy, so together with the predilection for biodegradable compounds abundant growth under septic conditions can occur.

Other factors that are deemed to favour its growth are nutrient (nitrogen, phosphorous, oxygen) shortages (Eikelboom, 2000). Substrate is removed from biological nutrient removal plants with initial anaerobic and anoxic zones, and because this filament is aerobic, it is thought to only cause bulking problems in industrial treatment plants and domestic plants without nutrient removal. Configurations with complete mixing in the aeration tank and continuous feeding can also promote the growth of Type 021N (Jenkins *et al.* 2004). According to Jenkins, (2004), they form gonidia and rosettes under nutrient deficient conditions.

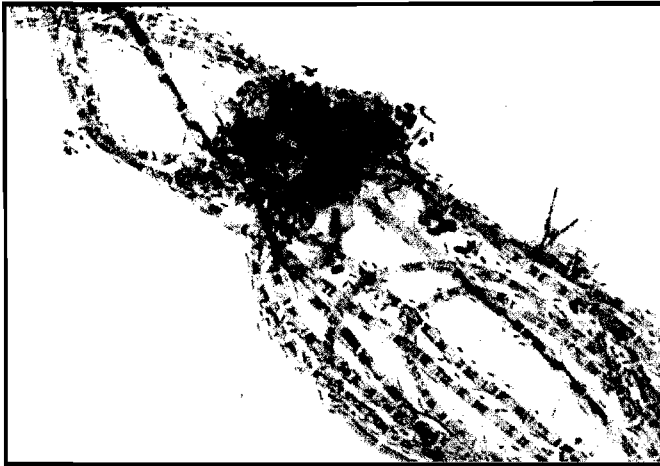


Figure 5.1.5 Gram stain of Type 021N from Parow WWTP, showing rosette formation.

- Occurrence in selected WWTP's in Cape Town from June-November 2007

Together with *M. parvicella*, this filament was the fourth most abundant organism encountered. It was the fifth most dominant filament. From June to November 2007, it was found in 25%, 50%, 63%, 69%, 75% and 69% of samples and was dominant in 13%, 16%, 13%, 13%, 13% and 25% respectively.

- Analysis of findings

This filament had a moderate prevalence, supporting the fact that it has specific growth requirements. However, if conditions were such to allow for the presence of Type 021N, it can be seen from figure 5.1.3, that it was significantly more likely to cause bulking as determined by elevated DSVI and/or FI. From table 5.1.2, it can be seen that this concurs with the findings of Eikelboom, (2000). Blackbeard *et al.* (1988) did not identify this organism in any samples, but numbers in the KwaZulu-Natal study were also moderately high. Perhaps this can be attributed to changes in wastewater composition or other parameters since the 1980's, but it is difficult to make any assumptions with the available details.

5.1.2 (f) Actinomycetes

- Documented organism characteristics

Jenkins *et al.* (2004) refers to this group of filaments as nocardioform organisms and includes many genera. Whichever term is used (actinomycetes or nocardioforms), for this dissertation, the filaments included are all Gram positive branching bacilli, albeit of mixed genera and growth requirements.

The combined results of Richard *et al.* (1982) and Strom and Jenkins (1984), cited by Jenkins *et al.* (2004), show that this group of organisms were the most common filaments in 270 bulking and foaming plants in the US.

They are not considered low F/M filaments, but can grow over a broad range of MCRT's including long sludge ages associated with low F/M conditions. According to Eikelboom (2000), however, the actinomycetes prefer higher sludge loads to *M. parvicella*. Both of these organisms can grow on the fat fraction of the wastewater. Like *M. parvicella*, the actinomycetes have a Gram positive cell wall that is not covered by a sheath or attached growth. They are thus hydrophobic and lipophilic so they tend to float and become attached to the fat fraction of the wastewater (Eikelboom, 2000). There is thus a strong association between the presence of actinomycetes and scum formation. Due to the fact that they float preferentially, the numbers are often greater than those in the mixed liquor. Conditions that are known to favour the growth of the actinomycetes are the presence of fats or other hydrophobic compounds, the presence of surface-active materials, recycling of floating material and temperatures > 15°C (Jenkins *et al.*, 2004).

- Occurrence in selected WWTP's in Cape Town from June-November 2007

This group of organisms was the sixth most common filament, but the third most dominant. In almost half the samples where it was encountered, it occurred as a dominant species group. From June to November 2007, it was found in 37%, 56%, 50%, 37%, 44% and 44% of samples and was dominant in 19%, 25%, 19%, 25%, 25% and 13% respectively.

- Analysis of findings

There was no obvious change in the occurrence of this organism from winter to summer in Cape Town. This ties in well with the results obtained for Type 0092, which has similar temperature preferences.

When present, there was a significant likelihood of this filament being dominant. From figure 5.1.3, it can also be assumed that the actinomycetes are closely associated with bulking conditions in terms of elevated DSVI and/or FI. In this category, dominance was 10-20% higher than the overall dominance (depending on the FI/DSVI category). Once again, there is conflict with Table 5.1.2, which denotes a moderate affect of this organism on SVI. It must be re-iterated that DSVI is a better indicator of bulking conditions than SVI and that conditions for the data in table 5.1.2 are unknown.

5.1.2 (g) *Halscomenobacter hydrosis*

- Documented organism characteristics

According to Jenkins *et al.* (2004), *H. hydrosis* is associated with low DO and/or nutrient (phosphorous and/or nitrogen) deficiency. The preferential sludge loads he cites are between >0.05 and <0.8 kgBOD/kgMLSS.day, while Eikelboom (2000) gives a value of >0.2kgBOD.kgMLSS.day. He also associates this filament with low DO and a phosphate deficiency and he links the growth of *H. hydrosis* to the presence of LMW compounds and/or high nitrogen concentration in the influent, and/or complete mixing in the aeration tank.

- Occurrence in selected WWTP's in Cape Town from June-November 2007

H. hydrosis was the seventh most prevalent and the seventh most dominant species. It was not present in any of the samples in June, but from July to November 2007, it occurred in 6%, 38%, 25%, 38% and 31% of samples and was dominant in 6%, 0%, 6%, 6%, and 6% respectively.

- **Analysis of findings**

The organism was not detected in any samples in June and only 6% of samples in July. Thereafter, *H. hydrosis* was present in similar numbers for the rest of the study period.

Referring to figure 5.1.3, it appears that the filament can be associated with elevated DSVI, but not elevated FI. However, when dominant, *H. hydrosis* was not the only dominant species and numbers are too small to make any assumptions. It is interesting to note that studies by Lakay *et al.* (1999) at UCT also found an association between high DSVI and *H. hydrosis*, except during protracted anoxic conditions.

5.1.2 (h) *Nostocoida limicola* III

- **Documented organism characteristics**

Eikelboom (2000) includes the organism species previously named *N. limicola* II into this group of species. He cites the F/M range favourable for growth as being a load of 0.1-0.3 kgBOD/kgMLSS.day, whereas Jenkins *et al.* (2004) gives the range for *N. limicola* II as >0.05-0.6 kgBOD/kgMLSS/day. Other conditions Eikelboom (2000) cites are: pre-settlement of influent, low water temperature, complete mixing in the aeration tank and the presence of easily metabolizable compounds (such as found in industrial waste). He also alludes to the possibility of nutrient shortages being a factor.

Jenkins *et al.* (2004) concurs that easily degraded compounds (specifically LMW organic acids) and nutrient deficiency play a role.

- **Occurrence in selected WWTP's in Cape Town from June-November 2007**

N. limicola III was the eighth most prevalent and dominant filament. It was not found in June, July or August, but from September, October and November was identified in 6%, 44% and 13% of samples and was dominant in 0%, 6% and 13% respectively.

- **Analysis of findings**

N. limicola was found to a much larger extent in the KwaZulu-Natal study, supporting the hypothesis by Lacko *et al.* (1999) that there may be errors with identification between Type 0092 and this filament.

According to the literature source, the identification methods used by this group of researchers was based on the 1984 Manual by Jenkins *et al.* and differentiated the two filaments mainly on cell shape (the assumption is thus made that the majority of the filaments were Gram negative), and by the presence of PHB granules. In the 2007 study in Cape Town, the filaments were differentiated according to Eikelboom (2000) and supported by Jenkins (2004). Both filaments can be Gram negative and blue-grey after Neisser staining, although *N. limicola* III is most often Gram positive. It is difficult to assess unstained preparations microscopically as both of these filaments tend to grow within the flocs.

Notable differences were:

- Gram stain: the majority of the Cape Town *N. limicola* III were Gram positive.
- Septa: Individual cells could be clearly discerned for *N. limicola* III after either a Gram or Neisser stain had been performed, whereas no septa could be seen, even at 1000X magnification for Type 0092.
- Filament shape: Filaments of *N. limicola* III were more coiled by comparison with Type 0092.
- Comparisons with micrographs: Eikelboom provides a number of micrographs in a CD that accompanies his 2000 Manual that allows more accurate identification.

No PHB granules were noted in either of these filaments. It would be interesting to perform cross-identification to determine whether identification methodology was indeed the reason for the discrepant results. Newer methodologies such as fluorescent *in-situ* hybridization (FISH) should minimize such occurrences.

Without further details, there is no plausible argument for the large increase in the presence of this filament in late spring and the phenomenon can merely be noted at this stage. Any findings of dominance on an individual plant basis are discussed in chapter 4.

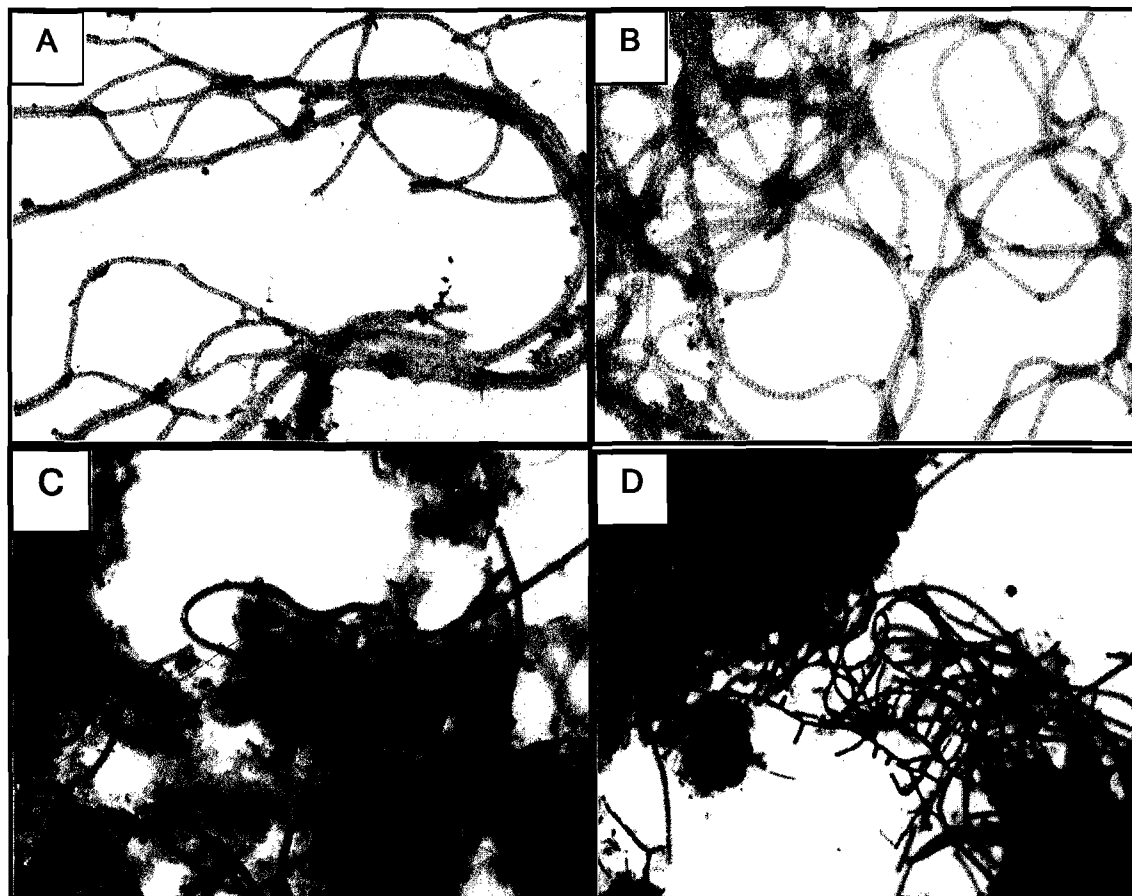


Figure 5.1.6

A Micrograph of a Gram stain of a Gram positive (blue) strain of *N. limicola* III typically found in the study

C Micrographs of a Neisser stain (blue-positive) of Type 0092 found in the study

B Micrographs of a Neisser stain of a Neisser positive (grey-blue) strain of *N. limicola* III typically found in the study

D Micrograph of a Gram stain of Gram negative (pink) Type 0092 from the study (The Gram positive organisms are actinomycetes)

5.1.2 (i) Type 1701

• Documented organism characteristics

The sludge load that Jenkins *et al.* (2004) cites for optimal growth of this filament is >0.1 to <0.8 kgBOD/kgMLSS/day, which is very similar to the value of >0.2 kgBOD.kgMLSS.day, given by Eikelboom (2000). Both authors agree that low DO also encourages growth of Type 1701. In addition, Eikelboom (2000) ties high levels of carbohydrates, complete mixing and temperature of $>15^{\circ}\text{C}$ as contributory factors to the

prevalence of this filament. He also states that Type 1701 “almost never” grows in domestic nutrient removal plants, but can cause high SVI in plants treating effluent from the agro industry.

- Occurrence in selected WWTP’s in Cape Town from June-November 2007

This was the 8th most prevalent filament, but was never present as a dominant species. It did not occur in October and November, but was present from June to September in 6%, 19%, 25% and 13% of samples respectively. It occurred in 50% of samples from the Kwazulu-Natal study (refer to table 5.1.1).

- Analysis of findings

The filament was only found as a secondary species and numbers were too small to make any overall assumptions.

5.1.2 (j) *Flexibacter* spp.

- Documented organism characteristics

There is little known about the occurrence of this filament except that it has no effect on settling (Eikelboom, 2000).

- Occurrence in selected WWTP’s in Cape Town from June-November 2007

This group comprised the 9th most prevalent filaments, but they were never dominant. They were absent in August and October, but were present in June, July, September and November in 6%, 6%, 13% and 25% of samples respectively.

- Analysis of findings

Numbers were too limited and knowledge of this organism too scanty to perform any analysis on its presence.

5.1.2 (k) Type 0581

- Documented organism characteristics

Jenkins *et al.* (2004) links the growth of this organism to elevated LMW compounds and classes it as low F/M. Little is known about this filament.

- Occurrence in selected WWTP’s in Cape Town from June-November 2007

Type 0581 was the 10th most common filament, but was never dominant. It was not present from September to November, but in June, July and August was found in 13%, 13% and 6% of samples respectively.

- Analysis of findings

There appears to be a winter maximum in the growth of this filament, but numbers were too low to make any definite assumptions. This filament was not identified in the other studies quoted in table 5.1.1.

5.1.2 (l) *Thiothrix* spp.

- Documented organism characteristics

Both Eikelboom, (2000) and Jenkins *et al.* (2004) agree that this filament has a broad range of growth requirements in terms of sludge load and is associated with elevated LMW carbon compounds and/or reduced sulphur and/or nutrient deficiency (nitrogen/phosphorous). In addition, Eikelboom (2000) links the growth of this organism to lack of oxygen and complete mixing in the aeration tank. He also found that *Thiothrix* spp. are only found in nutrient removal plants where there are high levels of sulphide, such as encountered with stale sewage.

- **Occurrence in selected WWTP's in Cape Town from June-November 2007**

This filament was only found in 2 WWTP's and was never present as a dominant species. It was found in 6% of the samples from July and 13% of the samples from August.

- **Analysis of findings**

It is not possible to make any overall assumptions about this filament, as it is only encountered under very specific conditions in nutrient removal plants. This is discussed under the pertinent plants in the individual analyses in chapter 4. In the KwaZulu Natal study, *Thiothrix* spp. occurred in 50% of samples, the authors postulating that the increase in industrialization in the region was responsible for this high figure.

5.2 POPULATION DYNAMICS OF PROTOZOA/METAZOA FROM JUNE TO NOVEMBER 2007

5.2.1 Results

Number of samples: 96

Number of reactors: 16

Number of WWTP's: 11

The prevalence was calculated using the number of samples. The abundance (scanty, moderate or numerous) was not taken into account in this calculation. The object of this chapter is to elucidate on the extent of these life forms in the Cape Town WWTP's over the study period. If the abundance of a particular organism is deemed to be significant, it is discussed under the analysis of individual plant in which it is found in chapter 4. Figures are given in the same manner as for filamentous organisms - in descending order of prevalence with the number of samples in which the organism was found and the percentage of samples in which that organism was found in parenthesis.

5.2.1 (a) Protozoa

- Ciliates

- Sessile ciliates

Prevalence from all samples (n=96)

Vorticella spp. (64/66%), *Epistylis* spp. (48/50%), *Carchesium* spp. (29/30%), *Vaginicola* spp. (18/19%), *Opercularia* spp. (8/8%), Sucroreans (3/3%). At the top of the following page is the monthly distribution graph (figure 5.2.1) for the three genera of sessile ciliates present in $\geq 20\%$ of samples.

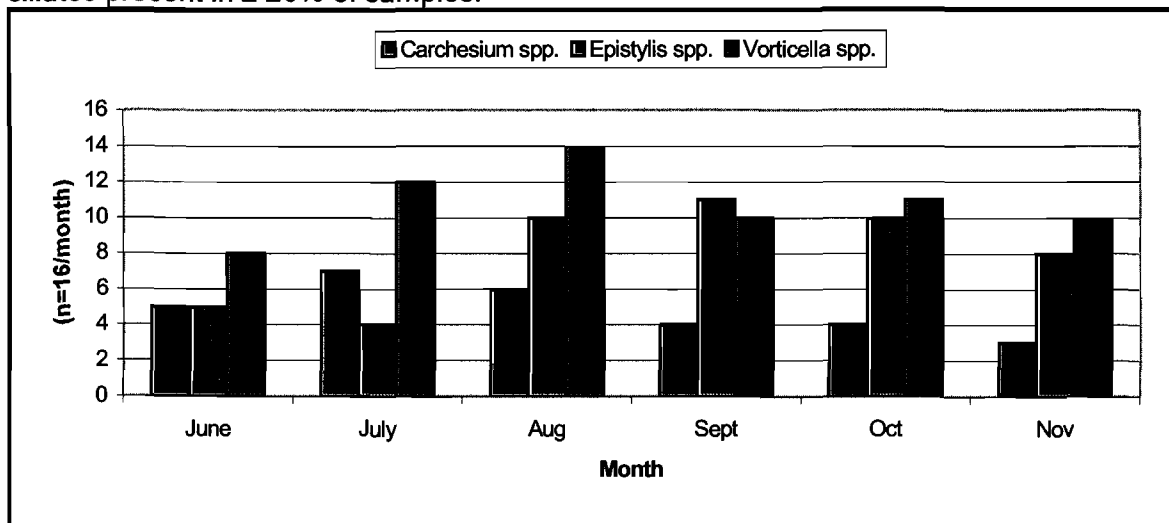


Figure 5.1.7 Bar chart showing the monthly distribution of sessile ciliates from the mixed liquor of the study WWTPs

- Crawling ciliates

Prevalence from all samples (n=96)

Aspidisca spp. (84/88%), *Trachelophyllum* spp. (39/41%), *Chilodonella* spp. (23/24%), unidentified crawling ciliates (3/3%).

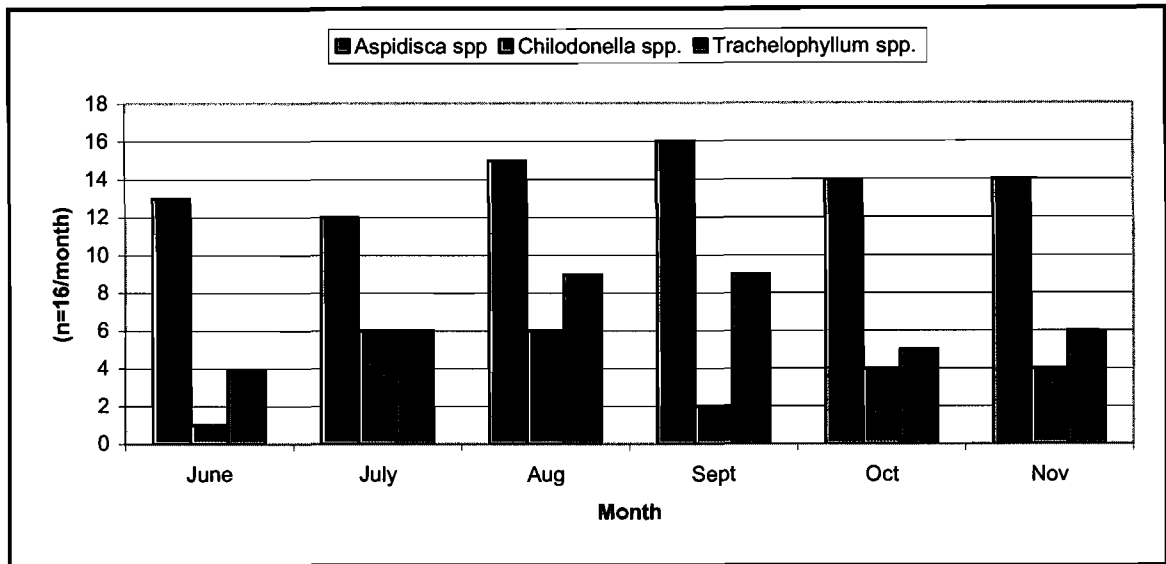


Figure 5.1.9 Bar chart showing the monthly distribution of crawling ciliates present in >20% of mixed liquor samples examined during the study

● Free-living ciliates

Prevalence from all samples (n=96):

Spirostomum spp. (33/35%), *Litonotus* spp. (26/27%), *Colpidium* spp. (21/22%), Unidentified free-living ciliates (16/17%), *Euplotes* spp. (15/16%), *Blepharisma* spp. (15/16%), *Paramecium* spp. (5/5%).

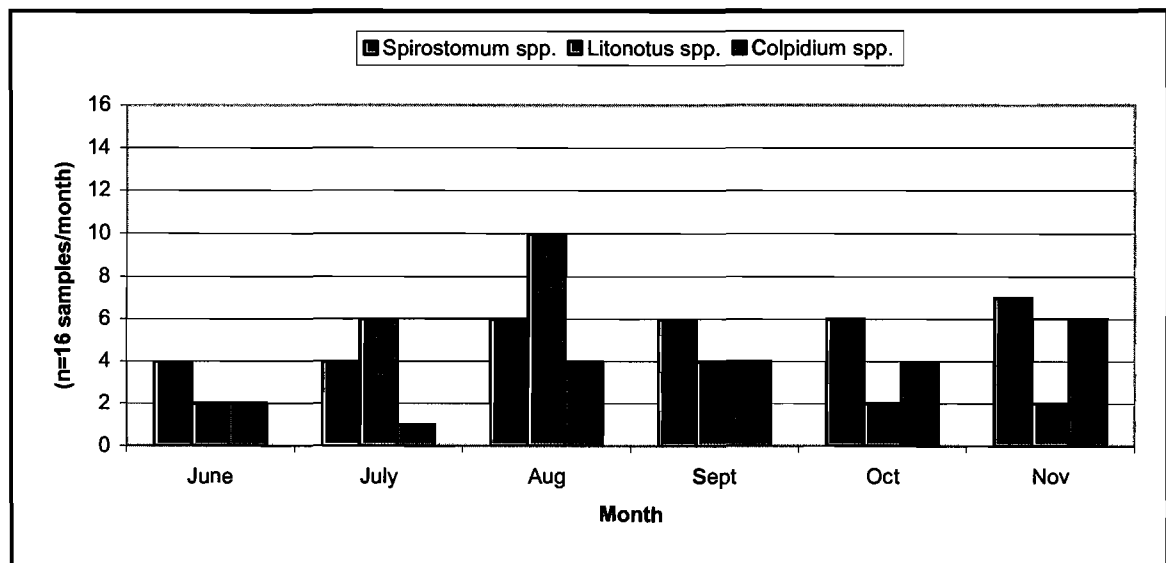


Figure 5.1.10 Bar chart showing the monthly distribution of free-living ciliates present in >20% of mixed liquor samples examined during the study

● **Flagellates**

Prevalence from all samples (n=96):

Bodo spp. (53/55%), *Peranema* spp. (47/49%), unidentified flagellates (10/10%), *Trepomonas* spp. (7/7%), *Monsiga* spp. (3/3%), *Pleuromonas* spp. (3/3%), *Hexamitus* spp. (1/1%).

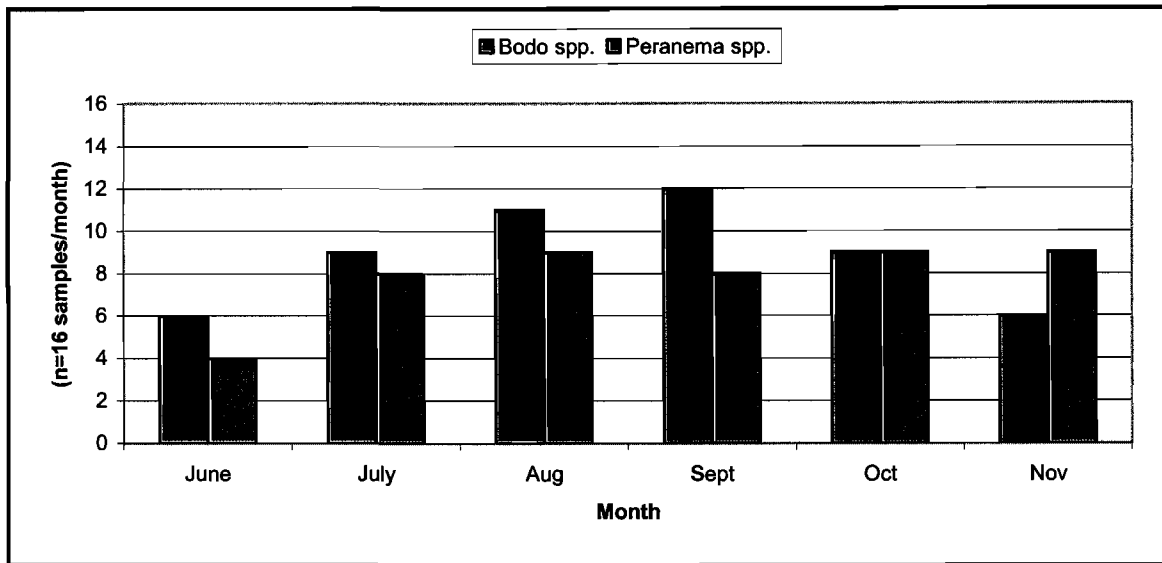


Figure 5.1.11 Bar chart showing the monthly distribution of flagellates present in >20% of mixed liquor samples examined during the study

● **Amoebae, testate amoebae and heliozoa**

Prevalence from all samples (n=96)

Amoebae (55/57%), heliozoa (11/11%) and testate amoebae (5/5%).

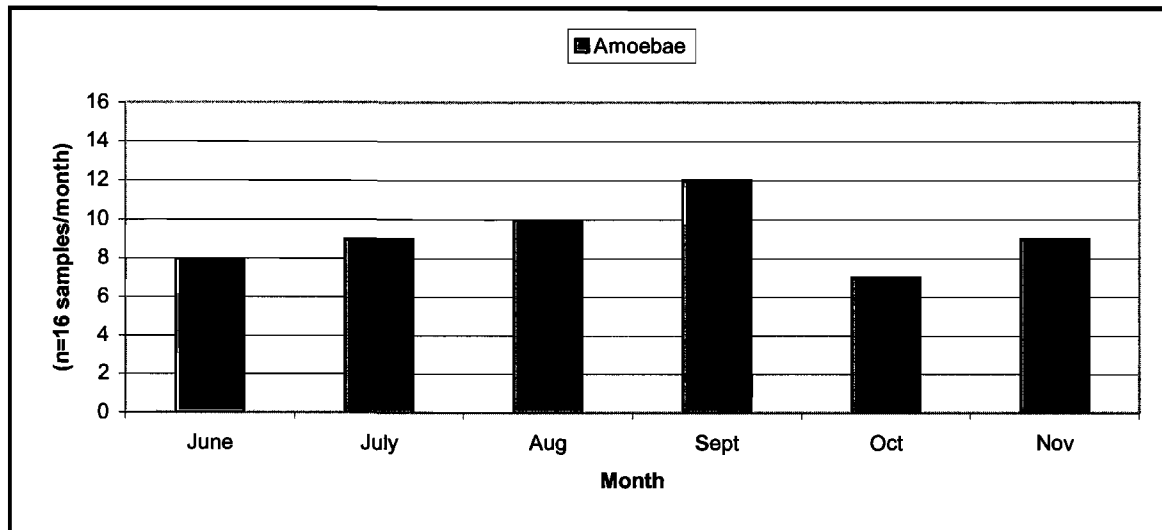


Figure 5.1.12 Bar chart showing the monthly distribution of amoebae (present in >20% of mixed liquor samples examined during the study)

5.2.1 (b) Metazoa

- Rotifers, Nematodes, Tardigrades and Oligochaete worms

Rotifers (83/86%), Tardigrades (18/19%), Nematodes (16/17%), Oligochaete worms (16/17%).

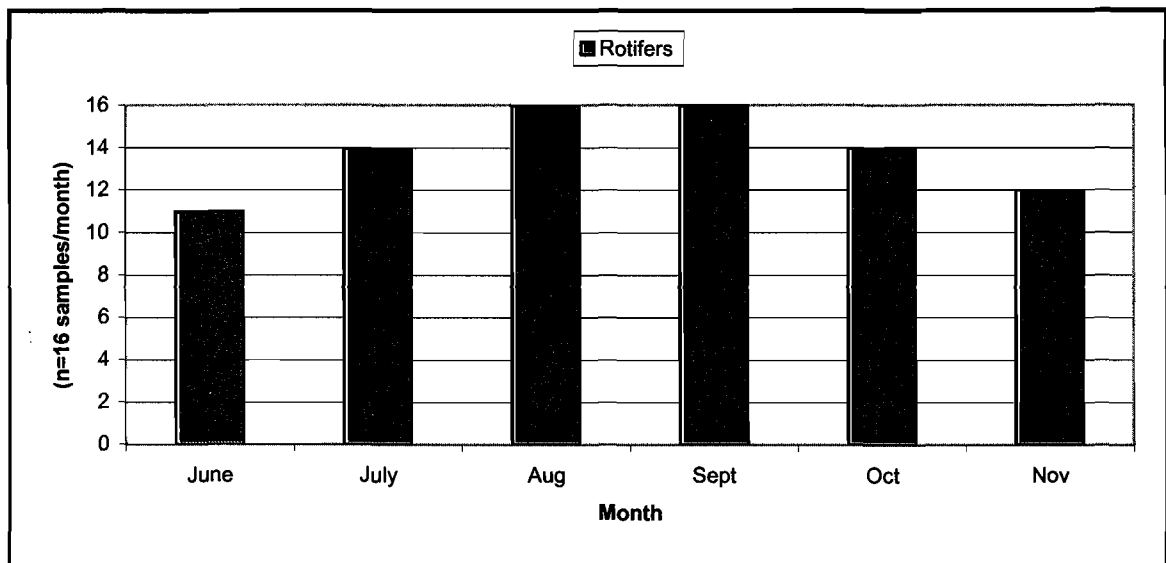


Figure 5.1.13 Bar chart showing the monthly distribution of rotifera (present in >20% of mixed liquor samples examined during the study)

5.2.2 Discussion

Only the ciliates and flagellates that were present in >20% of samples are discussed further in this chapter. If any of the others are relevant in the context of the individual WWTP in which they were found, they will be elucidated upon in the relevant section in chapter 4. The organisms were only identified to genus level (ciliates and flagellates), so any changes from winter to summer does not take into account that there may have been species succession within genera.

5.2.2 (a) Protozoa

- Ciliates

- Documented organism characteristics

Unless otherwise stated, the facts contained in this section are taken from Eikelboom, (2000).

- Sessile ciliates

- *Carchesium* spp.

This genus predaes mainly on bacteria. It is a common ciliate and is associated with sludge loads of <0.2kgBOD/kgMLSS.day. In a pilot plant study to assess the use of ciliates as bio-indicators, Lee *et al.* (2002) found significant statistical correlation between the presence of *Carchesium* spp. and low DO, evaluating it to be a good bio-indicator of these conditions.

- *Epistylis* spp.

This commonly found organism has similar food and loading (0.1 to 0.2kgBOD/kgMLSS.day) requirements to *Carchesium* spp.

- ***Vorticella* spp.**
As with the other stalked ciliates, *Vorticella* spp. is ubiquitous and predated on bacteria. It can tolerate higher loading rates (<0.4kgBOD/kgMLSS.day) than the other two sessile ciliates discussed above. Lee *et al.* (2002), found *Vorticella campanula*, to be an indicator of good quality effluent and good settling by association with low BOD and SVI respectively. Other species within this genus did not show significant correlation with low BOD, but were not deemed to be indicators of high BOD as determined in previous studies by Madoni *et al.* (1994) and Salvado *et al.* (1995). The findings of Lee *et al.* (2002) are backed up by those of Zhou *et al.* (2006), who found high numbers of *Vorticella convallaria* during periods of good plant performance and a strong negative correlation coefficient with BOD (the study was conducted over the period of a year at a sewage treatment plant in China).
- ***Opercularia* spp.**
Although this stalked ciliate was only found in 8% of samples, it is mentioned in this text because of the correlation of *Opercularia microdiscus* with high wasting rates as determined by Lee *et al.* (2002), and its resistance to the presence of heavy metals as determined by Madoni *et al.* (1996). Zhou *et al.* (2006) found a significant positive correlation of *Opercularia coarctata* with BOD and SS that suggested its use as an indicator of low quality effluent.
- **Crawling ciliates**
 - ***Aspidisca* spp.**
This ciliate preys on bacteria. It is frequently found providing the sludge load is <0.2kgBOD/kgMLSS.day. As with *Chilononella* spp. (below), Lee *et al.* (2002) found a significant association of *Aspidisca* spp. with low F/M and of *Aspidisca cicada* with old sludge age.
 - ***Chilodonella* spp.**
Chilodonella spp. are also bacteriovores and are common in plants with the same sludge load as for *Aspidisca* spp. In a study toxicological conducted by Puigagut *et al.* (2005), *Chilodonella uncinata* (together with *Acineria uncinata*), were the most tolerant to raised levels of ammonia, where they became the dominant species. Lee *et al.* (2002) suggested that *Chilodonella* spp. could be used as bio-indicators of high organic loading and poor effluent quality as determined by factor analysis with high SVI and MLSS. There was a similar relationship between the abundance of this organism and long SRT, so they also deduced this genus to be indicative of old sludge. In addition, they found that there was a relationship between these (and other) crawling ciliates and low F/M and postulated a possible metabolic advantage in competition for substrate.
 - ***Trachelophyllum* spp.**
These prey mainly on other ciliates as are thus deemed to be carnivorous. It occurs commonly at sludge loads of <0.4kgBOD/kgMLSS.day.
- **Free-living ciliates**
 - ***Colpidium* spp.**
This ciliate is a bacteriovore with a propensity for sludge loads of 0.1 to 0.4 kgBOD/kgMLSS.day. It is not frequently encountered.
 - ***Litonotus* spp.**
Litonotus spp. is a carnivore and is encountered in plants with sludge loads of <0.4kgBOD/kgMLSS.day. Lee *et al.* (2002) determined that statistically *Litonotus lamella* and other carnivorous protozoa were associated with poorly settling sludge and that this relationship was opposite to that of the stalked *Vorticella* spp. Zhou *et al.* (2006), found a correlation between poor settling conditions and

Litonotus obtusus. (This was the only species in the genus that was frequently present).

➤ ***Spirostomum* spp.**

These are the largest ciliates found in mixed liquor and occur as common protozoans in WWTP's with sludge loads of <0.2 kgBOD/kgMLSS.day.

● Occurrence in selected WWTP's in Cape Town from June-November 2007

● Sessile ciliates

By examining figure 5.1.7, it can be seen that *Vorticella* spp. was the most frequently found sessile ciliate with a maximum in August. *Epistylis* spp. was the second most frequent, being found in similar numbers to *Carchesium* spp. in winter, but was much more common in spring (maximum) and summer, where it competed numerically with *Vorticella* spp.

● Crawling ciliates

The results depicted by figure 5.1.8 show that *Aspidisca* spp. were the most commonly encountered crawling ciliates. Indeed, they were the most frequently found protozoan. There was little fluctuation in numbers over the study period, but *Trachellophyllum* spp., the second most common crawling ciliate, did show a distinct maximum frequency in August and September.

● Free-living ciliates

From figure 5.1.9, it can be seen that although *Spirostomum* spp. was the most common ciliate in this group overall, *Litonotus* spp. was found more frequently in July and August, after which numbers declined from September to November. Conversely, the numbers of *Colpidium* spp. increased toward the end of the study period, when they were greater than those of *Litonotus* spp. and similar to those of *Spirostomum* spp.

● Analysis of findings

The common sessile and free-living ciliates exhibited a succession. In addition, so did *Trachellophyllum* spp. and to a lesser extent *Chilodonella* spp. Of the crawling ciliates. *Aspidisca* spp. did not exhibit much fluctuation in numbers over the duration of the study. The latter is the most commonly encountered protozoan in most WWTP's, so the findings in terms of frequency for the Cape Town plants, is expected. The common occurrence of all the ciliates, barring *Colpidium* spp. concurs with Eikelboom's descriptions. The latter was a common isolate in the Cape Town WWTP's. The fact that *Vorticella* spp. was a common isolate suggests many reactors had low BOD and/or good settling according to literature findings.

● Flagellates

● Documented organism characteristics

➤ ***Bodo* spp.**

The presence of this organism is indicative of either a high sludge load and/or a lack of oxygen.

➤ ***Peranema* spp.**

No specific process conditions are linked to this organism and as such it plays no indicator role.

● Occurrence in selected WWTP's in Cape Town from June-November 2007

Looking at figure 5.1.11, it can be seen that *Bodo* and *Peranema* spp. increased in numbers from June to August, where after the number of *Bodo* spp. declined, so that they were less than those of *Peranema* spp. by November. The frequency of the latter remained at similar figures from August to November.

- **Analysis of findings**

The commonly encountered flagellates exhibited succession.

- **Amoebae, testate amoebae and heliozoa**

- **Documented organism characteristics**

According to Eikelboom, (2000), amoebae are seldom observed in low loaded plants and are usually associated with loads of 0.1 to 0.4 kgBOD/kgMLSS.day. and/or oxygen shortages.

- **Occurrence in selected WWTP's in Cape Town from June-November 2007**

Amoebae were found in a large number of samples and increased in number towards spring, after which they declined.

- **Analysis of findings**

Low sludge loads as found in nutrient removal plants should be not normally associated with the presence of amoebae, so the fact that this was a commonly observed organism may be explained by the low oxygen conditions encountered in the anaerobic and/or anoxic zones.

5.2.2 (b) Metazoa

- **Rotifers, Nematodes, Tardigrades and Oligochaete worms**

The metazoa are usually seen at loading levels of <0.15kgBOD/kgMLSS.day. Although they are larger than the bacteria and protozoa, they generally only play a minor role in the treatment of AS.

- **Documented organism characteristics**

Due to the size of their feeding apparatus, rotifers and nematodes cannot consume large particles. The major portion of their diet is thus restricted to free-living bacteria and small floc particles. Rotifers usually only reach large numbers of plants that treat waste from the agro-industry. Tardigrades graze on the surface of the flocs. Worms such as the oligochaeta can consume flocs and floc particles. At times they may bloom to large numbers in low loaded plants with pre-settled influent. In such circumstances there may be a concomitant decrease in sludge production.

- **Occurrence in selected WWTP's in Cape Town from June-November 2007 and analysis of findings**

There was little unexpected about the occurrence of the metazoa in the Cape Town WWTP's. The rotifers were the most abundant and exhibited a spring maximum. The exceptions are discussed further under the individual plants in chapter 4.

5.2.3 Additional observations

Due to the lack of technical expertise, the protozoa and metazoa were not speciated in this study. Most literature findings on the use of protozoa as bio-indicators point to different species being associated with certain operational parameters. Assumptions to genus level can often be made, but in many cases, there is not enough evidence to state that these findings would hold true for every species within a genus. For the analysis of protozoa in sludge, maximum benefit would be derived from linking the abundance of individual species with operating conditions for individual plants and then using these as bio-indicators in later samples. The problem with this is that it is extremely time-consuming and requires experienced personnel, which cannot be justified for routine monitoring of plant operation for the amount of benefit derived from such investigations. Thus, this type of work is likely to remain academic until such time as more data becomes available from research work to use a number of protozoa to make categorical

links between a genus and a variety of operating parameters. In this text and in the individual plant analyses in chapter 4, postulates are made according to previous research findings.

5.3 POPULATION DYNAMICS OF SPIROCHAETES AND FREE-LIVING CELLS FROM JUNE TO NOVEMBER 2007

5.3.1 Results

A total of 61 samples had been processed with random timing before comparative timed analysis was instituted (see methods, chapter 2). A further 35 samples were examined at 0 minutes and again at 10 min.

• Spirochaetes

After the introduction of comparative timed analysis, 80% of samples showed an increase in the number of spirochaetes detected between 0 and 10 minutes. Of the remaining 20%, all but 1 sample had no spirochaetes at both 0 and 10 minutes.

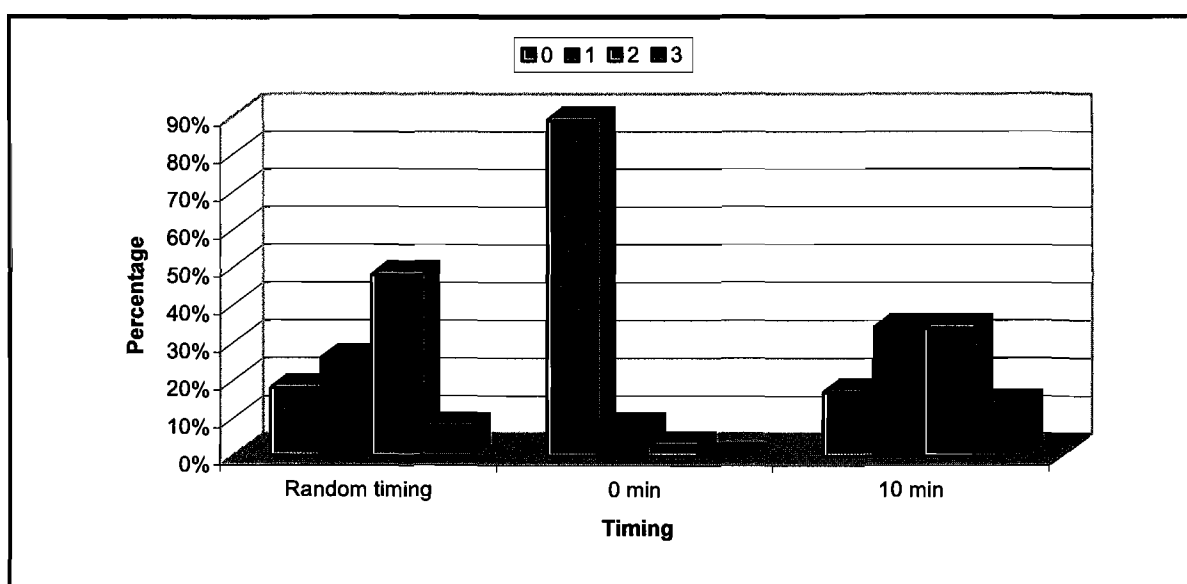


Figure 5.1.14 Abundance of spirochaetes in mixed liquor samples during random timing and after the introduction of comparative timed analysis

• Free-living cells

After the introduction of comparative timed analysis, 69% of samples showed an increase in the number of free-living cells detected between 0 and 10 minutes. Of the remaining 31%, all but 2 samples had no free-living cells at both 0 and 10 minutes.

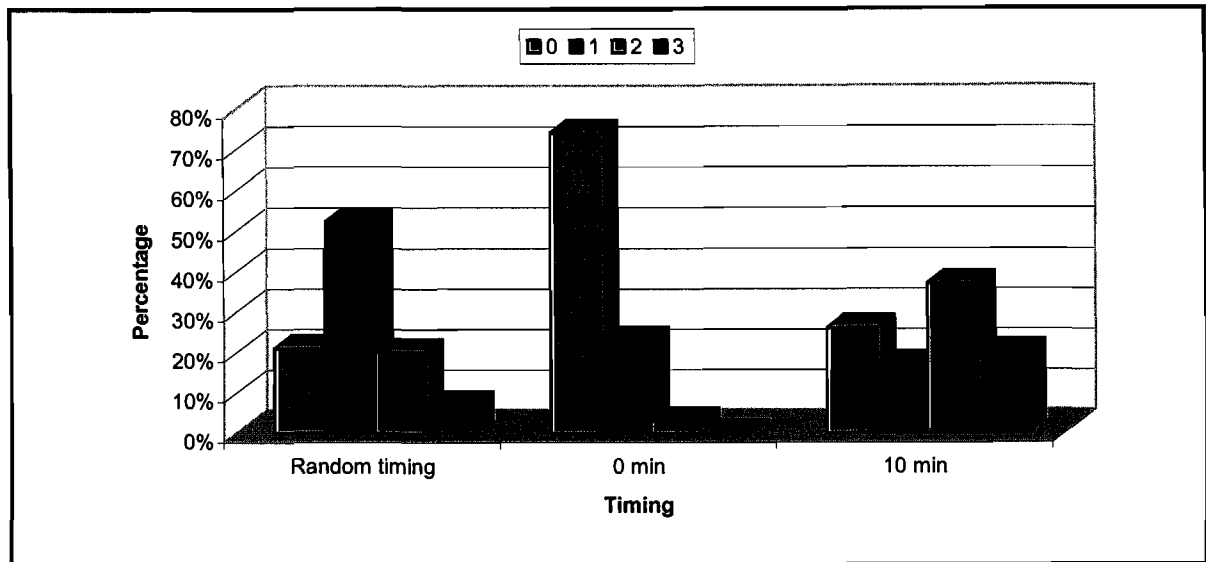


Figure 5.1.15 Abundance of free-living cells in mixed liquor samples during random timing and after the introduction of comparative timed analysis

5.3.2 Discussion

• Documented organism characteristics

Jenkins *et al.* (2004), associates the finding of spirochaetes with high organic acid and low DO concentrations, as may be found in septic wastewater. Eikelboom, (2000) equates their presence with anoxic conditions.

• Occurrence in selected WWTP's in Cape Town from June-November 2007

The data outlined in this section is mirrored in figures 5.1.14 and 5.1.15. In the samples where there were no free-living cells or spirochaetes, the figures were similar when the samples were viewed randomly or after 10 minutes: 21% and 26% of samples with free-living cells and 18% and 17% of samples with spirochaetes observed at random timing and 10 minutes respectively.

Although it may be assumed that some of the random samples were viewed immediately and thus should have higher numbers with no organisms, this was not the case. This can be attributed to the fact that the microscopic analysis is a lengthy process, and organisms that emerged from the flocs during the examination were included. Indeed, the common occurrence of this phenomenon led to the introduction of comparative timed analysis to prove this observed effect.

The presence of both of these organisms was ubiquitous in the Cape Town WWTP's tested, being found in close to approximately 80% of samples with both random timing and after 10 minutes of making a wet mount. However, figures taken from when the wet mount was examined immediately for spirochaetes and free-living cells they were found in only 11% and 26% of samples respectively. With regard to the spirochaetes, not only was there a 71% increase in the observation (absent to present) of this bacterium between 0 and 10 minutes, in all save 1 sample, there was an increase in abundance in the samples that were already positive. With the free-living cells, there was a 48% increase in the observation and only 2 samples did not show an increase in abundance.

Most of the samples with free-living cells did not have a large diversity of these organisms, with most seeming to have only one morphological type. Very few were immobile and a large number exhibited a tumbling type of motility reminiscent of that exhibited by *Listeria monocytogenes*.

● **Analysis of findings**

From the results, the following deductions can be made:

- Even in fresh samples, there was a high prevalence of spirochaetes and free-living cells in mixed liquor from the Cape Town WWTP's included in the study.
- These organisms live chiefly within the flocs and emerge under the cover slip of a wet mount (wet mounts were sealed with Vaseline).
- The conditions that cause this phenomenon are unknown. It can be speculated that lack of oxygen is responsible. However, spirochaetes are associated with anoxic conditions, the fact that they reside within flocs supports this fact.
- Although Eikelboom, (2000) notes for spirochaetes that "their number can increase during microscopic investigation" because spirochaetes are often "hidden in the flocs" and that "spirochaetes disappear if the sludge sample is stored for a few days in a refrigerator", he gives no guidance as to whether these emerging bacteria should be included in the analysis or not. For instance, if the microscopic examination is brief and the slide is prepared immediately before examination, they may easily be missed, or at the very least underestimated.

5.3.3 Additional observations

Work to ascertain more about the nature of these organisms in the Cape Town WWTP's could include sampling from different stages in the reactor cycles. These samples could be observed microscopically for the comparative abundance of spirochaetes and free-living cells. Perhaps these organisms grow in the free liquid phase during anaerobic and anoxic cycles and enter the flocs when the DO levels increase during the aerobic cycle. The spirochaetes and most of the free-living cells were highly motile, which would allow them to move in and out of flocs according to prevailing conditions.

No additional supporting literature could be found on this topic.

CHAPTER 6 SUMMATION

6.1 WWTP CONFIGURATION, NUTRIENT REMOVAL, BULKING AND FILAMENTOUS GROWTH

The tables below briefly summarize the nutrient removal results (table 6.1) and the bulking and notable filamentous characteristics (table 6.2) of the study WWTP's.

| TABLE 6.1 COMPARISON OF NUTRIENT REMOVAL EFFICIENCY IN STUDY WWTP'S | | | |
|---------------------------------------------------------------------|---------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| WWTP | CONFIG. | NUTRIENT REMOVAL | MOST PROBABLE REASON/S FOR GOOD OR POOR REMOVAL |
| Bellville | MLE | Ammonia - poor Nitrates/nitrites - good Phosphorous - good | Overloading Decreased supply of substrate due to poor nitrification Favourable influent composition. Lack of nitrates/nitrites creating anaerobic conditions in anoxic zone |
| Parow | MLE | Ammonia - erratic Nitrates/nitrites - erratic Phosphorous - erratic | Low DO Plant operation Intermittent nitrates/nitrites in anoxic zone creating alternating anaerobic and anoxic conditions. |
| Wesfleur | MLE | Ammonia - erratic Nitrates/nitrites - erratic Phosphorous - erratic | Low DO Plant operation Intermittent nitrates/nitrites in anoxic zone creating alternating anaerobic and anoxic conditions. |
| Athlone | UCT | Ammonia - poor Nitrates/nitrites - good Phosphorous - good | Demise of nitrifier population due to toxins Favourable plant operation. Reduced supply of substrate when nitrification poor Favourable influent composition. Lack of nitrates/nitrites in anaerobic zone |
| Kraaifontein | UCT | Ammonia - good Nitrates/nitrites - moderately poor Phosphorous - erratic | Favourable plant operation. Sufficient DO. Healthy nitrifier population Unfavourable plant operation. Possible lack of electron donor Intermittent nitrates/nitrites in anaerobic zone |
| Mitchells Plain | UCT | Reactor C: Ammonia - good Nitrates/nitrites - moderate, erratic Phosphorous - moderate Reactor G: Ammonia - good Nitrates/nitrites - poor Phosphorous - poor | Favourable plant operation. Sufficient DO. Healthy nitrifier population Intermittent lack of electron donor. Plant operation. Intermittent presence of nitrates/nitrites in anaerobic zone. Competition for substrate by GAO's. Same as for reactor C Unfavourable Plant operation Nitrates/nitrites in anaerobic zone. Competition for substrate by GAO's |
| Potsdam | UCT | Ammonia - good Nitrates/nitrites - good Phosphorous - good | Favourable plant operation. Sufficient DO. Healthy nitrifier population Favourable plant operation. Sufficient electron donor Favourable influent composition. Lack of nitrates/nitrites in anaerobic zone |

| TABLE 6.1 CONTINUED | | | |
|---------------------|-------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| WWTP | CONFIG. | NUTRIENT REMOVAL | MOST PROBABLE REASON/S FOR GOOD OR POOR REMOVAL |
| Macassar | UCT (carousel) | Reactor 1/reactor 2: Ammonia - good Nitrates/nitrites - erratic (poor to good) Phosphorous - erratic (moderate to poor) | Favourable plant operation. Sufficient DO. Healthy nitrifier population Insufficient electron donor. Nitrates/nitrites in anaerobic zone. |
| Borcherds quarry | 5-stage Bardenpho | Reactor B/reactor C: Ammonia - good Nitrates/nitrites - poor Phosphorous - poor | Favourable plant operation. Sufficient DO. Healthy nitrifier population Unfavourable plant operation Nitrates/nitrites in anaerobic zone. Competition for substrate by GAO's. |
| Cape Flats | 5-stage Bardenpho | Reactor G/reactor H: Ammonia - good Nitrates/nitrites - poor Phosphorous - poor | Favourable plant operation. Sufficient DO. Healthy nitrifier population Lack of electron donor Nitrates/nitrites in anaerobic zone. Possible lack of VFA's in anaerobic zone. |
| Wildevoevlei | 3-stage Phoredox | Reactor A: Ammonia - erratic Nitrates/nitrites - poor Phosphorous - poor Reactor B: Ammonia - good Nitrates/nitrites - erratic Phosphorous - moderately poor | Changes in DO Unfavourable plant operation Nitrates/nitrites in anaerobic zone. Competition for substrate by GAO's Favourable plant operation. Sufficient DO. Healthy nitrifier population Unfavourable plant operation Nitrates/nitrites in the anaerobic zone. Competition for substrate by GAO's |

In order to fully understand the nature of the unexpectedly poor phosphorous removal in some of the Cape Town WWTP's a study incorporating all of the NDBEPR WWTP's in the area would be prudent. Testing should include separate quantitation of nitrates in the various sections of the bioreactors as well as influent levels of acetic and propionic acid. The above measures are uncomplicated and inexpensive. The additional use of molecular methods to assess the microbial population in terms of GAO's and PAO's would add useful information. Such research would be beneficial and applied. Results would allow the implementation of substrate switches by changing operating conditions to improve nutrient removal.

In addition, the use of microscopy to aid in troubleshooting when confronted with loss of WWTP performance, such as occurred at Athlone WWTP is recommended. Regular assessment, especially regarding the protozoan and metazoan succession at the time would have been valuable. The use of FISH or DGGE to assess the nitrifier population at the time would have been definitive.

| TABLE 6.2 COMPARISON OF BULKING CONDITIONS AND FILAMENTOUS MICROORGANISMS IN STUDY WWTP'S | | | | |
|-------------------------------------------------------------------------------------------|---------------------------------|---------------------------------------------------------------|------------------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| WWTP | BULKING CONDITIONS | MAJOR DOMINANT SPECIES | CASEY'S HYPOTHESIS SUPPORTED | FURTHER COMMENTS |
| Bellville | Yes | Type 1851 (Type 021N)* | No | Filamentous population probably associated with increased LMW compounds in influent. Frequency of type 021N less than that of Type 1851 |
| Parow | Yes | Actinomycetes Type 1851 | No | Type 1851 dominant in domestic WWTP |
| Wesfleur | No | Type 1851 | No | "Clean" Type 1851 filaments |
| Athlone | Yes | Type 0092 (Type 021N)* | No | Type 0092 decreasing and Type 021N (with gonidia and rosettes) increasing in dominance with bulking conditions. Bulking may be associated with nitrogen deficiency. |
| Kraaifontein | No | Type 0092 | No | Large similarities with Potsdam WWTP |
| Mitchells Plain | C - no G - no | Type 0092 Type 0092 | No | Reactor with better nutrient removal had higher DSVI values. Type 1851 also dominant (domestic WWTP). |
| Potsdam | Yes | Type 0092 | No | Secondary organisms responsible for high FI. Enhanced settling, possibly by large number of monocolonies |
| Macassar | 1 - no 2 - yes (marginal) | <i>M. parvicella</i> <i>M. parvicella</i> Actinomycetes | No | Reactor with better nutrient removal had higher DSVI values. Type 1851 also dominant (domestic WWTP). |
| Borcherds quarry | B - yes C - no | Type 0092 (Type 021N)* Type 0092 | No | Reactor with better nutrient removal had higher DSVI values. Type 0092 decreasing and Type 021N (with gonidia and rosettes) increasing in dominance with bulking conditions in reactor B. |

| TABLE 6.2 (CONTINUED) | | | | |
|-----------------------|----------------------------------|-------------------------------------|------------------------------|------------------------------------------------------------------|
| WWTP | BULKING CONDITIONS | MAJOR DOMINANT SPECIES | CASEY'S HYPOTHESIS SUPPORTED | FURTHER COMMENTS |
| Cape Flats | G - no H - no | Type 0092 Type 0092 | No | |
| Wildevoevllei | A - yes (marginal) B - yes | Type 0092 Type 1851 Type 0092 | No | Type 1851 found as a major dominant filament in a domestic WWTP. |

*The major dominant filaments in table 6.2 are those with the highest frequency as dominant filaments. Although some of the dominant filaments did not have the highest frequency over the study period, their occurrence was equally high or higher during times of bulking. These filaments have been included in brackets.

By examining table 6.1 in conjunction with table 6.2, some strong associations can be determined. In terms of the WWTP configurations, the following was noted:

MLE configuration

All three plants with this configuration harboured Type 1851 as the major dominant species, irrespective of whether the plants treated domestic or industrial effluent. Type 021N at Bellville and actinomycetes at Parow were also present as major dominant species and in these two WWTP's, bulking conditions existed during the study period. At Wesfleur, Type 1851 was the sole major dominant species, and there was no indication of bulking.

Phosphorous removal was unexpectedly good at Bellville, for a WWTP of the MLE configuration. There was a strong relationship between the presence or absence of nitrates/nitrites in the effluent (and thus presumably the anaerobic zone) and phosphorous removal in the three plants.

UCT configuration

Four of the five WWTP's with this configuration harboured Type 0092 as the major dominant species. The structure and operating parameters of Macassar is considerably different to the other four and *M. parvicella* was the major dominant species in both reactors from this plant, along with actinomycetes in reactor 2. In this reactor, bulking conditions existed during the study period. Of the other four WWTP's, bulking conditions only existed at Athlone and Potsdam. At Athlone, the bulking was related to Type 021N rather than Type 0092, and at Potsdam, the secondary organisms may have been responsible for the high FI. The microbial populations (filaments, GAOs and protozoa/metazoa) at Kraaifontein WWTP and Potsdam WWTP showed remarkable similarities.

Ammonia removal was good in all of these plants, barring Athlone. The levels of nitrates/nitrites in the clarifier supernatant were absent to low only at Potsdam and Athlone WWTP's, and these were the only plants that showed good phosphorous removal. As with the MLE plants, there was a strong association between the presence or absence of nitrates/nitrites in the effluent and phosphorous removal. This must lead to the conclusion that the major cause of poor phosphorous removal in the Cape Town WWTP's is the presence of nitrates in the anaerobic zone.

5-stage Bardenpho configuration

There were only two WWTP's with this configuration. Due to the fact that the configuration includes two anoxic and aerobic zones, the removal of nitrates/nitrites should be complete or nearly complete. This was not the case for either WWTP. Once again, the presence of nitrates/nitrites in the effluent and poor phosphorous removal occurred concurrently.

3-stage Phoredox configuration

There was only one WWTP with this configuration. Type 0092 was the major dominant species, along with Type 1851 in reactor A. In the latter, bulking conditions did exist during the study period, but were borderline.

Thus, there were many similarities between the WWTPs in each configuration type, seemingly more strongly associated with configuration types than with influent composition. A recommendation that a larger survey including more WWTP's in each group to further establish or refute this link would be plausible. The perceived benefit of this is that corrective procedures applied to one WWTP with similar configuration and microbial dynamics would have application for other WWTPs in the same "category".

6.2 IDENTIFICATION OF MICROORGANISMS

The study results would have been more complete if molecular methodology was employed concurrently to assess the prokaryotic population. However, the City of Cape Town does not (or have the intention of introducing) the identification of microorganisms as a routine measure in the WWTP's in the area. As such, the study has provided valuable information that otherwise would not have been available.

In addition, there are advantages and disadvantages to all the commonly used methodologies (FISH and DGGE and conventional microscopic morphological identification, that have been discussed in chapter 1). A summarized comparison of the advantages and disadvantages of FISH and conventional microscopy is presented in table 6.3. (In reality DGGE only has an academic function due to the protracted nature of the test, the expertise required and the need for expensive and specialized equipment).

| TABLE 6.3 SUMMARY OF ADVANTAGES AND DISADVANTAGES OF CONVENTIONAL MICROSCOPIC INVESTIGATION, DGGE AND FISH FOR THE MICROBIOLOGICAL CHARACTERIZATION OF ACTIVATED SLUDGE | |
|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------|---------------------------------------------------------------------------------------------------------------|
| CONVENTIONAL MICROSCOPY | |
| Advantages | Disadvantages |
| It allows semi-quantitative analysis of the microbial population | It does not allow for accurate identification of non-filamentous bacteria, for example GAOs, PAOs, nitrifiers |
| It is simple to use once trained | It does not allow for identification of different species within the same morphotype |
| It requires minimal equipment | It does not always allow for identification of different morphotypes of the same species |
| It allows for assessment of floc character | *Problems may be experienced in identifying Gram negative filaments within the flocs |
| It allows assessment of filaments, protozoa and metazoa | |

| TABLE 6.3 (CONTINUED) SUMMARY OF ADVANTAGES AND DISADVANTAGES OF CONVENTIONAL MICROSCOPIC INVESTIGATION, DGGE AND FISH FOR THE MICROBIOLOGICAL CHARACTERIZATION OF ACTIVATED SLUDGE | |
|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-------------------------------------------------------------------------------------------|
| CONVENTIONAL MICROSCOPY | |
| Advantages | Disadvantages |
| It is inexpensive | |
| There is well-established data linking various microorganisms identified in this manner to operational conditions | |
| For the above reasons, it is widely used in Routine wastewater laboratories. Thus academic results obtained by this method are still of practical value | |
| FISH | |
| Advantages | Disadvantages |
| Theoretically allows for identification of the entire microbial population, for example filaments, PAOs, GAOs, nitrifiers | A number of probes need to be used and are not yet standardized |
| Allows tiered identification from group to species level | Mismatches occur |
| Methods have been described for quantitation of organisms | Permeability problems may occur |
| High level of specificity with correct probes | Autofluorescence or quenching of fluorescence may occur |
| | Does not allow for floc characterization and identification of protozoa and metazoa (yet) |

*Regarding the identification of Gram negative filaments within flocs, during this study no problems were experienced and this may have been due to the different staining techniques employed.

6.3 OTHER IMPORTANT STUDY FINDINGS

M. parvicella did not cause major bulking problems in the Cape Town WWTP's. Little work was performed on scum formation. At Macassar, both *M. parvicella* and actinomycetes were found in the scum samples. However, these two filaments were also present as major dominant species at that WWTP. It is possible that these organisms are implicated in the intermittent scum problems experienced at WWTP's such as Potsdam, where this phenomenon mainly occurs during the winter months. Having said that, there was no variation in the occurrence of either *M. parvicella* or Type 0092 during the course of the study.

Regarding the spirochaetes and free-living cells, very little could be deduced as these organisms move out of the flocs due to their motility, even in fresh samples. Unless standard methods are developed, it is difficult to interpret inter-laboratory results.

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ANNEXURE 1.1 RAW DATA FROM ATHLONE WWTP

INFLUENT

| Date: | Flow | SS | SETS | COD | TKN | AMM | TP | OP | CL | PH | CON | ALK |
|-------|-------|-----|------|-----|------|------|-----|-----|-----|-----|-----|-----|
| 5/6 | 113.6 | 132 | <0.1 | 492 | 39.9 | 27.2 | 7.0 | 3.9 | 200 | 7.1 | 129 | 262 |
| 12/6 | 113.6 | 130 | 0.5 | 407 | 38.3 | 24.7 | 6.9 | 3.7 | 196 | 7.5 | 128 | 250 |
| 19/6 | 144.3 | 128 | 1.0 | 542 | 49.9 | 27.3 | 8.6 | 4.4 | 209 | 7.5 | 135 | 282 |
| 26/6 | 129.7 | 140 | 2.0 | 372 | 41.1 | 22.6 | 4.4 | 1.8 | 170 | 7.4 | 112 | 222 |
| 3/7 | 143.0 | 127 | <0.1 | 410 | 35.8 | 23.2 | 6.2 | 3.7 | 196 | 7.7 | 126 | 252 |
| 10/7 | 135.1 | 114 | <0.1 | 453 | 40.8 | 27.0 | 6.8 | 4.2 | 235 | 7.4 | 145 | 244 |
| 17/7 | ND | 130 | ND | 434 | 42.1 | 29.2 | 6.2 | 3.8 | 231 | 7.3 | 142 | 261 |
| 24/7 | 126.6 | 134 | ND | 410 | 34.2 | 20.9 | 4.3 | 2.2 | 211 | 7.3 | 124 | 218 |
| 31/7 | 183.2 | 114 | 2.0 | 325 | 32.3 | 22.6 | 6.0 | 3.2 | 199 | 7.8 | 131 | 263 |
| 7/8 | 175.6 | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| 14/8 | 169.6 | 86 | 1.0 | 342 | 30.1 | 20.3 | 4.4 | 2.0 | 155 | 7.5 | 108 | 256 |
| 21/8 | 161.5 | 116 | 1.0 | 338 | 36.4 | 23.9 | 4.8 | 2.3 | 185 | 7.7 | 122 | 215 |
| 28/8 | 156.1 | 130 | 1.0 | 378 | 34.5 | 21.3 | 5.5 | 2.5 | 158 | 7.7 | 118 | 248 |
| 4/9 | 162.8 | 128 | <0.1 | 434 | 42.0 | 29.0 | 6.5 | 3.8 | 198 | 7.6 | 133 | 269 |
| 10/9 | 156.3 | 146 | <0.1 | 424 | 41.5 | 35.3 | 5.1 | 2.6 | 181 | 7.7 | 125 | 260 |
| 18/9 | 145.1 | 120 | <0.1 | 456 | 43.1 | 29.9 | 7.3 | 5.3 | 215 | 7.7 | 137 | 260 |
| 25/9 | 134.2 | 156 | 0.1 | 460 | 50.4 | 35.6 | 9.1 | 6.3 | 188 | 7.7 | 130 | 256 |
| 2/10 | 127.4 | 156 | 0.1 | 460 | 50.4 | 35.6 | 9.1 | 6.3 | 188 | 7.7 | 130 | 256 |
| 9/10 | 130.1 | 258 | 1.0 | 688 | 60.1 | 36.2 | 8.7 | 5.6 | 200 | 7.5 | 134 | 288 |
| 16/10 | 127.5 | 258 | 8.0 | 600 | 44.6 | 25.3 | 8.0 | 4.8 | 208 | 7.5 | 131 | 260 |
| 23/10 | 121.1 | 256 | 4.0 | 534 | 45.5 | 31.1 | 6.2 | 3.7 | 205 | 7.1 | 126 | 247 |
| 30/10 | 122.1 | 268 | 5.0 | 739 | 54.2 | 36.1 | 9.2 | 6.6 | 236 | 7.2 | 146 | 334 |
| 6/11 | 118.9 | 311 | 8.0 | 756 | 58.3 | 35.1 | 8.7 | 5.6 | 181 | 7.4 | 126 | 254 |
| 13/11 | 122.2 | 345 | 10.0 | 794 | 51.7 | 34.8 | 9.2 | 6.5 | 194 | 7.2 | 127 | 276 |
| 20/11 | 117.2 | 282 | 4.0 | 638 | 49.5 | 33.1 | 8.2 | 6.0 | 188 | 7.1 | 126 | 273 |
| 27/11 | 121.0 | 246 | 6.0 | 660 | 48.9 | 28.3 | 8.7 | 5.7 | 188 | 7.3 | 125 | 294 |

MIXED LIQUOR REACTOR A

SLUDGE (ALL REACTORS)

| Date | SS | SETS | VSS | SVI | DSVI | PH | SWR | AGE | RASS |
|-------|------|------|------|-----|------|-----|------|-----|-------|
| 5/6 | 6490 | 720 | 5185 | 111 | 62 | 6.7 | 3648 | 12 | 11280 |
| 12/6 | 5575 | 650 | | 116 | 47 | 6.4 | 3648 | 12 | 12695 |
| 19/6 | 6100 | 750 | | 123 | 72 | 6.8 | 3520 | 13 | 11480 |
| 26/6 | 3610 | 900 | | 249 | 155 | 6.6 | 3533 | 12 | 10620 |
| 3/7 | 6800 | 950 | 5490 | 140 | 94 | 6.8 | 3445 | 13 | 11750 |
| 10/7 | 6530 | 910 | | 139 | 98 | 6.8 | 3576 | 13 | 11650 |
| 17/7 | 7240 | 970 | | 134 | 99 | 6.6 | 3576 | ND | 11600 |
| 24/7 | 7620 | 980 | | 129 | 94 | 6.6 | 3485 | 13 | 12130 |
| 31/7 | 4630 | 550 | | 119 | 104 | 6.8 | 3672 | 13 | 10490 |
| 7/8 | 5810 | 860 | 4740 | 148 | 103 | 6.7 | 3582 | 13 | 11450 |
| 14/8 | 5650 | 950 | | 168 | 106 | 6.6 | 3561 | 13 | 10920 |
| 21/8 | 5770 | 870 | | 151 | 104 | 6.8 | 3544 | 13 | 10910 |
| 28/8 | 4550 | 700 | | 154 | 105 | 6.9 | 3516 | 13 | 10190 |
| 4/9 | 5550 | 940 | 4570 | 169 | 115 | 7.0 | 3606 | 13 | 10700 |
| 10/9 | 5440 | 960 | | 176 | 110 | 7.0 | 3610 | 12 | 10430 |
| 18/9 | 5580 | 980 | | 176 | 129 | 7.0 | 3688 | 12 | 9670 |
| 25/9 | 5530 | 930 | | 168 | 123 | 7.0 | 3837 | 11 | ND |
| 2/10 | 5750 | 1000 | 4680 | 174 | 160 | 6.9 | 3658 | 12 | 10000 |
| 9/10 | 5330 | 980 | | 184 | 150 | 7.1 | 3633 | 12 | 9200 |
| 16/10 | 5590 | 1000 | | 179 | 179 | 7.0 | 3594 | 12 | 8630 |
| 23/10 | 5820 | 980 | | 168 | 172 | 7.1 | 3694 | 12 | 8380 |
| 30/10 | 5720 | 1000 | | 175 | 175 | 6.9 | 3500 | 13 | 8620 |
| 6/11 | 5040 | 1000 | 4350 | 198 | 198 | 6.8 | 3694 | 12 | 9220 |
| 13/11 | 7190 | 1000 | | 139 | 223 | 6.9 | 3061 | 14 | 9250 |
| 20/11 | 5950 | 1000 | | 168 | 202 | 6.9 | 3340 | 13 | 8520 |
| 27/11 | 6660 | 1000 | | 150 | 300 | 6.9 | 3541 | 13 | 7500 |

| EFFLUENT | | | | | | | | | | | |
|----------|----|-----|-----|------|------|------|-----|-----|-----|-----|-----|
| Date | SS | DS | COD | CODF | AMM | NN | OP | CL | PH | CON | ALK |
| 5/6 | 26 | ND | 94 | ND | 2.7 | 6.2 | 0.2 | 182 | 7.2 | 104 | 154 |
| 12/6 | 13 | 586 | 65 | ND | 0.5 | 4.5 | 0.2 | 159 | 7.4 | 97 | 154 |
| 19/6 | 2 | ND | 52 | ND | 4.9 | 1.1 | 1.0 | 193 | 7.1 | 110 | 205 |
| 26/6 | 17 | ND | 83 | 51 | 1.0 | 3.9 | 0.2 | 155 | 7.1 | 93 | 145 |
| 3/7 | 15 | ND | 66 | ND | 3.5 | 2.8 | 0.5 | 184 | 7.4 | 111 | 186 |
| 10/7 | 7 | 598 | 53 | 49 | 2.8 | 2.9 | 0.4 | 200 | 7.1 | 116 | 169 |
| 17/7 | 5 | ND | 51 | 51 | 5.7 | 2.5 | 1.5 | 231 | 7.0 | 118 | 189 |
| 24/7 | 14 | ND | 55 | ND | 2.9 | 3.2 | 0.4 | 201 | 7.0 | 111 | 163 |
| 31/7 | 4 | ND | 51 | ND | 2.8 | 3.1 | 0.2 | 169 | 7.1 | 108 | 197 |
| 7/8 | 5 | ND | 48 | ND | 2.4 | 2.3 | 0.3 | 163 | 7.0 | 97 | 174 |
| 14/8 | 8 | ND | 69 | ND | 4.5 | 1.7 | 0.6 | 158 | 7.0 | 94 | 194 |
| 21/8 | 6 | ND | 65 | ND | 5.4 | 0.4 | 1.2 | 165 | 7.2 | 106 | 182 |
| 28/8 | 10 | ND | 54 | ND | 7.6 | 1.1 | 1.4 | 163 | 7.2 | 110 | 201 |
| 4/9 | 7 | ND | 66 | ND | 6.5 | 0.6 | 1.5 | 166 | 7.3 | 105 | 201 |
| 10/9 | 9 | 619 | 69 | ND | 10.9 | 0.9 | 1.2 | 172 | 7.4 | 113 | 222 |
| 18/9 | 8 | ND | 65 | ND | 24.7 | 0.1 | 3.2 | 184 | 7.4 | 127 | 260 |
| 25/9 | 16 | ND | 71 | ND | 13.2 | 0.5 | 0.9 | 179 | 7.3 | 111 | 215 |
| 2/10 | 16 | ND | 71 | ND | 13.2 | 0.5 | 0.9 | 179 | 7.3 | 111 | 215 |
| 9/10 | 13 | ND | 65 | ND | 23.8 | <0.1 | 1.9 | 181 | 7.4 | 122 | 256 |
| 16/10 | 17 | ND | 55 | ND | 24.8 | 0.9 | 2.0 | 181 | 7.9 | 121 | 252 |
| 23/10 | 24 | ND | 91 | ND | 25.3 | 0.1 | 1.8 | 190 | 7.4 | 125 | 249 |
| 30/10 | 13 | ND | 73 | ND | 27.0 | <0.1 | 2.3 | 188 | 7.4 | 125 | 295 |
| 6/11 | 14 | ND | 81 | ND | 25.3 | <0.1 | 2.0 | 170 | 7.3 | 117 | 239 |
| 13/11 | 20 | 595 | 82 | ND | 26.3 | 0.1 | 1.8 | 185 | 7.4 | 120 | 250 |
| 20/11 | 17 | ND | 80 | ND | 29.5 | 0.5 | 3.4 | 183 | 7.3 | 121 | 254 |
| 27/11 | 12 | ND | 55 | ND | 19.5 | <0.1 | 1.5 | 182 | 7.5 | 115 | 247 |

- AGE:** Sludge age (days)
ALK: Alkalinity (CaCO₃ mg/L)
AMM: Ammonia (N mg/L)
CL: Chloride (Cl⁻ mg/L)
COD: Chemical oxygen demand (mg COD/L)
CODF: Chemical oxygen demand (filtered)
CON: Conductivity (mS/m)
DS: Dissolved solids (mg/L)
DSVI: Dissolved sludge volume index (ml/g)
Flow: Raw flow (ML/day) for entire plant
ND: Not determined
NN: Nitrate and nitrite (N mg/L)
OP: ortho-phosphate (P mg/L)
RASF: RAS flow rate (s) (kL/day)
RASSS: RAS suspended solids (mg/L)
SETS: Settleable solids (30 min ml/L)
SS: Suspended solids (105°C mg/L)
SVI: Sludge volume index (ml/g)
SWR: Sludge wasting rate (kL/day)
TKN: Total Kjeldahl nitrogen (N mg/L)
TP: Total phosphorus (P mg/L)
VSS: Volatile suspended solids (mg/L)

Figures in red are anomalies and not transcription errors from the laboratory printouts. These results do not fit the general trend and have been left out of the graphs in the results and discussion section.

| ANNEXURE 1.2 RAW DATA FROM BELLVILLE WWTP | | | | | | | | | | | | | |
|----------------------------------------------------|------|------|-----|-------|------|------|-----|------|------|-----|-----|-----|-----|
| RAW INDUSTRIAL WASTEWATER INTO REACTORS (INFLUENT) | | | | | | | | | | | | | |
| Date: | Flow | SS | VSS | SETS | COD | TKN | AMM | TP | OP | PH | CL | ALK | CON |
| 6/6 | 50.7 | 504 | 434 | 18.0 | 978 | 58.5 | 39 | 10.4 | 6.0 | 7.1 | 99 | 259 | 97 |
| 13/6 | ND | 835 | ND | 37.0 | 1172 | 62.9 | 37 | 17.4 | 9.9 | 7.2 | 97 | 268 | 105 |
| 20/6 | 55.0 | 1650 | ND | 120.0 | 6010 | 150 | 39 | 33.5 | 15.6 | 6.9 | 115 | 307 | 111 |
| 27/6 | 54.6 | 634 | ND | 30.0 | 921 | 69.4 | 42 | 13.3 | 8.5 | 7.3 | 101 | 273 | 99 |
| 4/7 | ND | 592 | ND | 17.0 | 987 | 53.5 | 33 | 10.6 | 6.8 | 7.1 | 122 | 273 | 112 |
| 11/7 | 54.5 | 560 | ND | 30.0 | 1223 | 60.8 | 28 | 13.4 | 7.3 | 6.9 | 135 | 260 | 115 |
| 18/7 | 48.5 | 535 | ND | 22.0 | 1285 | 65.6 | 34 | 15.6 | 9.4 | 6.5 | 110 | 257 | 108 |
| 25/7 | 58.5 | 297 | ND | 10.0 | 683 | ND | 24 | ND | 5.1 | 7.2 | 112 | 227 | 96 |
| 1/8 | 78.9 | 568 | ND | 25.0 | 1003 | 54.7 | 36 | 11.9 | 7.2 | 7.1 | 124 | 272 | 104 |
| 8/8 | ND | 498 | ND | 20.0 | 788 | ND | 32 | ND | 4.9 | 7.4 | 108 | 264 | 95 |
| 15/8 | 63.6 | 488 | ND | 27.0 | 834 | 58.9 | 33 | 9.9 | 5.5 | 7.3 | 100 | 268 | 100 |
| 22/8 | 58.0 | 907 | ND | 50.0 | 2119 | 85.6 | 36 | 17.4 | 8.5 | 7.2 | 103 | 275 | 102 |
| 29/8 | 61.6 | 828 | ND | 26.0 | 1339 | 67.5 | 37 | 20.3 | 10.7 | 7.2 | 124 | 258 | 111 |
| 5/9 | 70.0 | 276 | 219 | 11.0 | 748 | 44.9 | 26 | 6.8 | 4.0 | 7.4 | 112 | 256 | 100 |
| 12/9 | ND | 535 | ND | 26.0 | 927 | 49.3 | 27 | 12.1 | 7.0 | 7.4 | 150 | 244 | 108 |
| 19/9 | ND | 158 | ND | <0.1 | 679 | 43.4 | 29 | 9.4 | 7.2 | 7.3 | 116 | 260 | 105 |
| 26/9 | 48.2 | 840 | ND | 38.0 | 1290 | 59.3 | 32 | 11.9 | 7.8 | 7.2 | 115 | 279 | 109 |
| 3/10 | ND | 844 | ND | 27.0 | 1178 | 64.9 | 34 | 13.4 | 8.2 | 7.1 | 110 | 284 | 104 |
| 10/10 | ND | 1260 | ND | 58.0 | 1672 | 88.7 | 44 | 17.1 | 9.3 | 6.8 | 101 | 300 | 107 |
| 17/10 | 47.7 | 498 | ND | 16.0 | 1016 | 62.8 | 36 | ND | 7.8 | 7.1 | 112 | 278 | 109 |
| 24/10 | ND | 792 | ND | 3.0 | 1326 | 64.5 | 41 | 13.7 | 10.4 | 7.0 | 114 | 303 | 108 |
| 1/11 | 49.6 | 624 | ND | 28.0 | 1145 | 79.3 | 51 | ND | 6.6 | 7.3 | 98 | 264 | 101 |
| 8/11 | 48.4 | 670 | 495 | 25.0 | 1092 | 58.1 | 32 | 11.3 | 7.6 | 7.3 | 102 | 258 | 97 |
| 14/11 | ND | 987 | ND | 54.0 | 1387 | 83.7 | 34 | 15.0 | 7.9 | 7.4 | 97 | 257 | 99 |
| 21/11 | 52.9 | 890 | ND | 42.0 | 1492 | 64.7 | 34 | 13.4 | 7.2 | 7.2 | 92 | 227 | 88 |
| 28/11 | 51.5 | 560 | ND | 28.0 | 1211 | 65.1 | 46 | 14.4 | 10.6 | 7.2 | 102 | 311 | 104 |

| MIXED LIQUOR REACTOR N | | | | | | | | | | |
|------------------------|------|------|------|-----|------|-----|------|------|-----|-------|
| Date | SS | SETS | VSS | SVI | DSVI | PH | SWR | RASF | AGE | RASSS |
| 6/6 | 7870 | 820 | 6225 | 104 | 76 | 6.9 | 508 | ND | 19 | 14300 |
| 13/6 | 5950 | 580 | ND | 98 | 74 | 7.2 | ND | ND | ND | 12970 |
| 20/6 | 6980 | 840 | ND | 120 | 80 | 6.7 | 432 | ND | 22 | 12810 |
| 27/6 | 6360 | 750 | ND | 118 | 82 | 6.7 | 659 | ND | 12 | 14070 |
| 4/7 | 6900 | 910 | 5350 | 132 | 99 | 6.4 | ND | ND | ND | 10240 |
| 11/7 | 6840 | 870 | ND | 127 | 88 | 6.6 | 467 | ND | 22 | 11350 |
| 18/7 | 6970 | 800 | ND | 115 | 80 | 6.7 | 467 | ND | 21 | 12440 |
| 25/7 | 5900 | 700 | ND | 119 | 81 | 6.6 | 433 | ND | ND | ND |
| 1/8 | 5590 | 750 | ND | 134 | 86 | 6.6 | 400 | ND | 22 | 10320 |
| 8/8 | 3970 | 500 | ND | 126 | 91 | 6.6 | ND | ND | ND | ND |
| 15/8 | 4770 | 600 | ND | 126 | 84 | 6.6 | 428 | 1284 | 27 | 6820 |
| 22/8 | 5960 | 820 | ND | 138 | 94 | 6.8 | 500 | 1500 | 26 | 8030 |
| 29/8 | 7210 | 880 | ND | 122 | 78 | 6.7 | 561 | ND | 19 | 11850 |
| 5/9 | 6630 | 900 | 5055 | 136 | 90 | 6.9 | 667 | ND | 15 | 11740 |
| 12/9 | 6840 | 950 | ND | 139 | 117 | 6.7 | ND | ND | ND | 11140 |
| 19/9 | 6860 | 970 | ND | 141 | 140 | 7.0 | ND | ND | ND | 10640 |
| 26/9 | 6950 | 970 | ND | 140 | 138 | 6.9 | 567 | ND | 20 | 10500 |
| 3/10 | 6510 | 970 | 5020 | 149 | 135 | 6.9 | ND | ND | ND | 9740 |
| 10/10 | 6250 | 960 | ND | 154 | 141 | 6.8 | ND | ND | ND | 9690 |
| 17/10 | 5750 | 930 | ND | 162 | 118 | 7.0 | 1067 | ND | 23 | 4060 |
| 24/10 | 6400 | 970 | ND | 152 | 163 | 6.9 | ND | ND | ND | 10980 |
| 1/11 | 5680 | 970 | ND | 171 | 155 | 7.1 | ND | ND | ND | 8820 |
| 8/11 | 5790 | 990 | 4300 | 171 | 173 | 7.1 | 600 | ND | 23 | 7270 |
| 14/11 | 5490 | 980 | ND | 179 | 182 | 6.9 | ND | ND | ND | 7430 |
| 21/11 | 5595 | 1000 | ND | 179 | 179 | 6.9 | 833 | ND | 14 | 7205 |
| 28/11 | 5450 | 980 | ND | 180 | 183 | 6.9 | 700 | ND | 18 | 6760 |

| CLARIFIER N (EFFLUENT) | | | | | | | | | |
|------------------------|----|-----|-----|------|------|-----|-----|-----|-----|
| Date: | SS | COD | AMM | NN | OP | PH | CL | ALK | CON |
| 6/6 | 6 | 37 | 12 | 0.4 | 0.6 | 7.1 | 102 | 232 | 89 |
| 13/6 | 14 | 40 | 1.9 | 8.8 | 0.3 | 7.7 | 108 | 191 | 86 |
| 20/6 | 11 | 73 | 24 | <0.1 | 10.3 | 7.4 | 115 | 307 | 111 |
| 27/6 | 12 | 76 | 17 | <0.1 | 0.6 | 7.4 | 106 | 252 | 93 |
| 4/7 | 12 | 63 | 26 | 0.2 | 1.8 | 7.4 | 119 | 261 | 103 |
| 11/7 | 10 | 51 | 18 | <0.1 | 1.1 | 7.6 | 120 | 259 | 105 |
| 18/7 | 12 | 48 | 22 | <0.1 | 0.8 | 7.4 | 110 | 261 | 100 |
| 25/7 | ND | ND | 16 | <0.1 | 0.4 | 7.6 | 112 | 227 | 96 |
| 1/8 | 19 | 59 | 8.5 | 0.2 | 0.4 | 7.4 | 117 | 247 | 98 |
| 8/8 | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| 15/8 | 14 | 48 | 9.4 | 1.0 | 0.2 | 7.3 | 98 | 238 | 90 |
| 22/8 | 4 | 55 | 15 | <0.1 | 0.5 | 7.6 | 93 | 232 | 86 |
| 29/8 | 8 | 48 | 17 | <0.1 | 0.9 | 7.4 | 118 | 257 | 99 |
| 5/9 | 10 | 62 | 24 | <0.1 | 7.9 | 7.7 | 111 | 259 | 102 |
| 12/9 | 6 | 44 | 24 | <0.1 | 1.6 | 7.8 | 106 | 262 | 98 |
| 19/9 | 11 | 55 | 23 | <0.1 | 1.4 | 7.8 | 100 | 264 | 98 |
| 26/9 | 10 | 55 | 22 | <0.1 | 2.7 | 7.7 | 104 | 257 | 98 |
| 3/10 | 13 | 75 | 24 | 0.1 | 5.7 | 7.5 | 102 | 255 | 95 |
| 10/10 | 5 | 44 | 20 | <0.1 | 0.6 | 7.7 | 104 | 252 | 93 |
| 17/10 | 12 | 55 | 28 | <0.1 | 1.4 | 7.8 | 101 | 264 | 98 |
| 24/10 | 22 | 99 | 25 | 0.2 | 4.2 | 7.5 | 100 | 247 | 93 |
| 1/11 | 14 | 105 | 25 | <0.1 | 11.3 | 7.9 | 111 | 251 | 97 |
| 8/11 | 15 | 90 | 23 | <0.1 | 6.0 | 7.6 | 112 | 272 | 99 |
| 14/11 | 35 | 114 | 31 | 0.1 | 12.0 | 7.6 | 109 | 274 | 102 |
| 21/11 | 68 | 279 | 32 | 0.4 | 10.2 | 7.5 | 103 | 298 | 97 |
| 28/11 | 39 | 163 | 28 | <0.1 | 9.5 | 7.6 | 101 | 297 | 95 |

- AGE: Sludge age (days)
 ALK: Alkalinity (CaCO₃ mg/L)
 AMM: Ammonia (N mg/L)
 CL: Chloride (Cl⁻ mg/L)
 COD: Chemical oxygen demand (mg COD/L)
 CON: Conductivity (mS/m)
 DSVI: Dissolved sludge volume index (ml/g)
 Flow: Raw Flow (ML/day) for entire industrial plant (into north, central and south reactors)
 ND: Not determined
 NN: Nitrate and nitrite (N mg/L)
 OP: ortho-phosphate (P mg/L)
 RASF: RAS flow rate (s) (kL/day)
 RASSS: RAS suspended solids (mg/L)
 SETS: Settleable solids (30 min ml/L)
 SS: Suspended solids (105°C mg/L)
 SVI: Sludge volume index (ml/g)
 SWR: Sludge wasting rate (kL/day)
 TKN: Total Kjeldahl nitrogen (N mg/L)
 TP: Total phosphorus (P mg/L)
 VSS: Volatile suspended solids (mg/L)

Figures in red are anomalies and not transcription errors from the laboratory printouts.

ANNEXURE 1.3 RAW DATA FROM BORCHERDS QUARRY WWTP

INFLUENT FROM PST

| Date: | Flow | SS | VSS | SETS | COD | TKN | AMM | TP | OP | PH | CL | ALK | CON |
|-------|------|-----|-----|-------|------|------|-----|------|------|-----|-----|-----|-----|
| 5/6 | 33 | 700 | 613 | 28.0 | 1464 | 95.0 | 51 | 16.8 | 10.2 | 7.0 | 113 | 346 | 117 |
| 12/6 | 33 | 247 | ND | 2.0 | 745 | 71.4 | 44 | 15.3 | 11.6 | 7.1 | 108 | 333 | 117 |
| 19/6 | 33 | 236 | ND | 4.0 | 690 | 70.1 | 44 | 16.2 | 12.5 | 7.4 | 116 | 311 | 106 |
| 26/6 | 33 | 202 | ND | 20.0 | 873 | 74.4 | 33 | 13.1 | 7.6 | 7.0 | 95 | 243 | 92 |
| 3/7 | 33 | 238 | ND | <1.0 | 790 | 94.3 | 73 | 17.0 | 13.5 | 7.8 | 124 | 370 | 128 |
| 10/7 | 33 | 840 | ND | 130.0 | 2584 | 148 | 52 | 34.9 | 20.5 | 7.1 | 121 | 321 | 124 |
| 17/7 | 32 | 500 | ND | 3.0 | 1133 | 88.0 | 54 | 16.9 | 12.7 | 6.6 | 117 | 336 | 126 |
| 24/7 | 33 | 209 | ND | 5.0 | 705 | 67.2 | 34 | 10.9 | 7.9 | 7.1 | 94 | 255 | 94 |
| 31/7 | 34 | 344 | ND | 12.0 | 869 | 42.9 | 28 | 15.1 | 9.9 | 7.4 | 110 | 290 | 112 |
| 7/8 | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| 14/8 | ND | 496 | ND | 15.0 | 1020 | 87.0 | 48 | 12.6 | 7.9 | 7.5 | 158 | 394 | 110 |
| 21/8 | ND | 360 | ND | 12.0 | 981 | 70.3 | 41 | 10.7 | 7.0 | 7.5 | 117 | 312 | 114 |
| 28/8 | 32 | 260 | ND | 2.0 | 775 | 66.4 | 39 | 14.0 | 9.7 | 7.6 | 110 | 317 | 115 |
| 4/9 | 33 | 264 | ND | 2.0 | 851 | 85.8 | 59 | 16.4 | 12.7 | 7.7 | 112 | 328 | 123 |
| 11/9 | 33 | 229 | ND | 2.0 | 674 | 56.9 | 31 | 9.9 | 6.6 | 7.5 | 108 | 286 | 105 |
| 18/9 | 32 | 240 | ND | 2.0 | 1015 | 67.3 | 46 | 14.7 | 11.9 | 7.6 | 113 | 310 | 113 |
| 25/9 | ND | 633 | ND | 23.0 | 1235 | 92.4 | 49 | 15.6 | 10.7 | 7.6 | 114 | 334 | 118 |
| 2/10 | 33 | 792 | ND | 30.0 | 1798 | 86.9 | 49 | 20.8 | 16.6 | 6.9 | 110 | 330 | 111 |
| 9/10 | 32 | 288 | ND | 3.0 | 892 | 72.9 | 48 | 12.3 | 9.0 | 7.5 | 107 | 304 | 110 |
| 16/10 | 32 | 278 | ND | 2.0 | 1006 | 76.3 | 43 | 12.7 | 10.5 | 7.1 | 117 | 321 | 114 |
| 23/10 | 32 | 200 | ND | <1.0 | 808 | 89.8 | 64 | 15.2 | 11.9 | 7.1 | 111 | 352 | 122 |
| 30/10 | 30 | ND | ND | 100.0 | 2731 | 152 | 51 | 30.4 | 16.8 | 6.7 | 105 | 429 | 122 |
| 6/11 | 32 | 810 | ND | 25.0 | 1437 | 107 | 61 | 20.5 | 15.2 | 7.3 | 105 | 335 | 116 |
| 13/11 | 32 | 630 | ND | 28.0 | 1467 | 92.9 | 54 | 17.3 | 13.3 | 6.9 | 114 | 324 | 119 |
| 20/11 | 32 | 474 | ND | 18.0 | 945 | 73.5 | 55 | 11.9 | 8.7 | 7.3 | 104 | 312 | 107 |
| 27/11 | 32 | 277 | ND | 1.0 | 970 | 88.5 | 56 | 14.2 | 11.0 | 7.0 | 118 | 391 | 122 |

MIXED LIQUOR REACTOR B

| Date | SS | SETS | VSS | SVI | DSVI | PH | SWR | AGE | RASS |
|-------|------|------|------|-----|------|-----|------|-----|-------|
| 5/6 | 7350 | 980 | 6225 | 133 | 114 | 6.7 | 1150 | 13 | 13090 |
| 12/6 | 7630 | 970 | ND | 127 | 100 | 6.9 | 1250 | 12 | 12850 |
| 19/6 | 7370 | 970 | ND | 132 | 119 | 6.9 | 990 | 15 | 10730 |
| 26/6 | 6470 | 960 | ND | 148 | 105 | 6.7 | 990 | 14 | 12860 |
| 3/7 | 8100 | 990 | 6750 | 122 | 114 | 7.1 | ND | ND | 12640 |
| 10/7 | 6600 | 980 | ND | 148 | 133 | 6.7 | ND | ND | 12820 |
| 17/7 | 6750 | 960 | ND | 142 | 101 | 6.4 | ND | ND | 9430 |
| 24/7 | 6380 | 980 | ND | 154 | 125 | 6.7 | 1190 | 12 | 12320 |
| 31/7 | 6060 | 980 | ND | 162 | 132 | 6.8 | 1090 | 13 | 11400 |
| 7/8 | 5790 | 810 | ND | 157 | 124 | 6.8 | ND | ND | ND |
| 14/8 | 6380 | 960 | ND | 150 | 125 | 6.6 | ND | ND | 12090 |
| 21/8 | 4020 | 600 | ND | 149 | 119 | 6.6 | ND | ND | 8110 |
| 28/8 | 4140 | 850 | ND | 205 | 126 | 7.0 | ND | ND | 9580 |
| 4/9 | 4750 | 960 | 4080 | 202 | 126 | 7.0 | 2100 | 7 | 16230 |
| 11/9 | 4410 | 900 | ND | 204 | 123 | 7.2 | 1340 | 11 | 11100 |
| 18/9 | 5780 | 990 | ND | 171 | 152 | 6.7 | 1270 | 11 | 11510 |
| 25/9 | 3860 | 860 | ND | 223 | 124 | 7.1 | ND | ND | 5420 |
| 2/10 | 5630 | 980 | 4680 | 174 | 156 | 6.6 | ND | ND | 11810 |
| 9/10 | 6160 | 1000 | ND | 162 | 162 | 6.8 | 2423 | 6 | 9990 |
| 16/10 | 5510 | 1000 | ND | 181 | 181 | 6.7 | 824 | 17 | 9600 |
| 23/10 | 5660 | 980 | ND | 173 | 163 | 7.0 | 1150 | 12 | 9500 |
| 30/10 | 6020 | 1000 | ND | 166 | 140 | 6.8 | 1080 | 14 | 12270 |
| 6/11 | 5980 | 980 | 4940 | 164 | 154 | 6.9 | 1050 | 13 | 4960 |
| 13/11 | 5340 | 970 | ND | 182 | 150 | 7.0 | 1180 | 12 | 9230 |
| 20/11 | 7365 | 980 | ND | 133 | 130 | 6.9 | 1190 | 12 | 9675 |
| 27/11 | 5200 | 930 | ND | 179 | 92 | 6.9 | 1160 | 12 | 11640 |

| MIXED LIQUOR REACTOR C | | | | | | | |
|------------------------|------|------|------|-----|------|-----|-------|
| Date | SS | SETS | VSS | SVI | DSVI | PH | RASS |
| 5/6 | 9540 | 980 | 7955 | 103 | 92 | 6.8 | 13200 |
| 12/6 | 8840 | 980 | ND | 111 | 91 | 6.9 | 11350 |
| 19/6 | 9900 | 980 | ND | 99 | 89 | 6.9 | 13920 |
| 26/6 | 9170 | 960 | ND | 105 | 79 | 6.7 | 17990 |
| 3/7 | 8700 | 960 | 7190 | 110 | 97 | 7.0 | 13840 |
| 10/7 | 9340 | 990 | ND | 106 | 107 | 6.7 | 13690 |
| 17/7 | 9080 | 960 | ND | 106 | 79 | 6.5 | 12220 |
| 24/7 | 8650 | 970 | ND | 112 | 97 | 6.8 | 13200 |
| 31/7 | 7640 | 950 | ND | 124 | 89 | 6.9 | 11780 |
| 7/8 | 6930 | 800 | ND | 115 | 81 | 6.9 | ND |
| 14/8 | 7920 | 850 | ND | 107 | 70 | 6.7 | 11830 |
| 21/8 | 8890 | 840 | ND | 94 | 72 | 6.7 | 13100 |
| 28/8 | 8640 | 920 | ND | 106 | 69 | 7.0 | 13010 |
| 4/9 | 7050 | 860 | 5880 | 122 | 74 | 7.0 | 11340 |
| 11/9 | 9730 | 960 | ND | 99 | 82 | 7.0 | 14400 |
| 18/9 | 8800 | 960 | ND | 109 | 91 | 6.7 | 12410 |
| 25/9 | 8340 | 930 | ND | 112 | 77 | 7.0 | 10410 |
| 2/10 | 7320 | 950 | 6080 | 130 | 98 | 6.5 | 12330 |
| 9/10 | 6370 | 890 | ND | 140 | 88 | 6.9 | 8930 |
| 16/10 | 7760 | 980 | ND | 126 | 88 | 6.7 | 10090 |
| 23/10 | 7690 | 900 | ND | 117 | 68 | 7.0 | 11730 |
| 30/10 | 8350 | 900 | ND | 108 | 62 | 6.8 | 12540 |
| 6/11 | 9270 | 950 | 7550 | 101 | 73 | 6.9 | 6510 |
| 13/11 | 8520 | 940 | ND | 110 | 80 | 7.0 | 12070 |
| 20/11 | 7775 | 920 | ND | 118 | 77 | 7.0 | 13845 |
| 27/11 | 8290 | 970 | ND | 117 | 101 | 6.9 | 12240 |

| CLARIFIER B (EFFLUENT) | | | | | | | | | |
|------------------------|-----|-----|------|------|------|-----|-----|-----|-----|
| Date: | SS | COD | AMM | NN | OP | PH | CL | Aik | CON |
| 5/6 | 13 | 55 | <0.4 | 1.5 | 3.6 | 7.1 | 118 | 166 | 78 |
| 12/6 | 8 | 33 | <0.4 | 2.7 | 2.7 | 7.4 | 103 | 163 | 76 |
| 19/6 | 5 | 41 | <0.4 | 3.3 | 5.2 | 7.2 | 114 | 173 | 80 |
| 26/6 | 13 | 47 | <0.4 | 6.6 | 2.5 | 6.9 | 100 | 122 | 71 |
| 3/7 | 9 | 51 | <0.4 | 4.0 | 2.5 | 7.4 | 122 | 169 | 84 |
| 10/7 | 32 | 77 | <0.4 | 3.3 | 3.1 | 7.1 | 118 | 160 | 84 |
| 17/7 | 10 | 48 | 0.7 | 3.2 | 1.7 | 7.0 | 123 | 165 | 83 |
| 24/7 | 6 | 44 | 0.6 | 4.5 | 2.9 | 7.0 | 111 | 156 | 82 |
| 31/7 | 4 | 41 | <0.4 | 5.5 | 3.3 | 7.3 | 112 | 193 | 91 |
| 7/8 | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| 14/8 | 272 | 392 | 1.4 | 6.3 | 0.4 | 7.1 | 122 | 168 | 80 |
| 21/8 | 9 | 55 | <0.4 | 6.2 | 0.3 | 7.5 | 112 | 163 | 89 |
| 28/8 | 5 | 49 | <0.4 | 4.5 | 4.5 | 7.5 | 106 | 174 | 86 |
| 4/9 | 25 | 95 | 6.8 | 4.6 | 39.9 | 7.1 | 112 | 211 | 99 |
| 11/9 | 10 | 58 | 0.1 | 4.5 | 3.2 | 7.3 | 116 | 167 | 85 |
| 18/9 | 8 | 62 | <0.4 | 2.0 | 2.7 | 7.5 | 124 | 175 | 88 |
| 25/9 | 24 | 83 | 1.2 | 10.1 | 5.4 | 7.2 | 131 | 149 | 91 |
| 2/10 | 9 | 44 | <0.4 | 3.5 | 2.5 | 7.1 | 111 | 147 | 76 |
| 9/10 | 21 | 55 | <0.5 | 4.7 | 6.0 | 7.4 | 113 | 149 | 80 |
| 16/10 | 8 | 37 | 0.5 | 3.9 | 4.2 | 7.5 | 116 | 163 | 82 |
| 23/10 | 18 | 73 | <0.4 | 9.0 | 7.1 | 7.4 | 106 | 138 | 80 |
| 30/10 | 6 | 46 | 0.4 | 0.9 | 3.6 | 7.3 | 102 | 182 | 76 |
| 6/11 | 29 | 66 | <0.4 | 1.6 | 2.0 | 7.4 | 102 | 174 | 77 |
| 13/11 | 23 | 60 | <0.4 | 4.9 | 4.4 | 7.4 | 112 | 157 | 77 |
| 20/11 | 16 | 58 | 1.3 | 1.1 | 10.3 | 7.5 | 110 | 163 | 79 |
| 27/11 | 22 | 66 | <0.4 | 5.3 | 5.6 | 7.4 | 121 | 174 | 83 |

| CLARIFIER C (EFFLUENT) | | | | | | | | | | |
|------------------------|----|-----|------|------|------|-----|-----|-----|-----|--|
| Date: | SS | COD | AMM | NN | OP | PH | CL | ALK | CON | |
| 5/6 | 9 | 47 | 2.9 | 4.5 | 7.3 | 7.0 | 114 | 169 | 81 | |
| 12/6 | 10 | 40 | <0.4 | 7.5 | 5.2 | 7.3 | 104 | 144 | 77 | |
| 19/6 | 15 | 45 | 0.4 | 7.1 | 10.1 | 7.1 | 112 | 162 | 82 | |
| 26/6 | 14 | 47 | <0.4 | 12.3 | 8.2 | 7.0 | 103 | 115 | 74 | |
| 3/7 | 12 | 47 | <0.4 | 4.0 | 2.5 | 7.4 | 122 | 169 | 84 | |
| 10/7 | 42 | 74 | <0.4 | 3.3 | 3.1 | 7.1 | 118 | 160 | 84 | |
| 17/7 | 19 | 58 | <0.4 | 5.0 | 4.7 | 7.1 | 118 | 152 | 87 | |
| 24/7 | 51 | 91 | 0.5 | 9.9 | 6.4 | 6.9 | 109 | 139 | 82 | |
| 31/7 | 10 | 48 | <0.4 | 10.2 | 5.8 | 7.2 | 110 | 175 | 92 | |
| 7/8 | ND | ND | ND | ND | ND | ND | ND | ND | ND | |
| 14/8 | 11 | 51 | 0.8 | 10.4 | 4.7 | 7.0 | 121 | 153 | 84 | |
| 21/8 | 9 | 51 | <0.4 | 9.1 | 3.5 | 7.4 | 116 | 155 | 89 | |
| 28/8 | 28 | 61 | <0.4 | 9.9 | 6.3 | 7.2 | 105 | 149 | 87 | |
| 4/9 | 7 | 44 | <0.4 | 10.3 | 13.6 | 7.3 | 111 | 144 | 89 | |
| 11/9 | 22 | 76 | 2.3 | 6.4 | 3.5 | 7.1 | 112 | 170 | 86 | |
| 18/9 | 21 | 76 | <0.4 | 9.3 | 5.7 | 7.3 | 122 | 151 | 90 | |
| 25/9 | 60 | 108 | <0.4 | 10.2 | 7.1 | 7.1 | 123 | 146 | 90 | |
| 2/10 | 84 | 129 | <0.4 | 10.6 | 11.1 | 6.8 | 107 | 127 | 80 | |
| 9/10 | 7 | 44 | <0.4 | 13.1 | 8.2 | 7.2 | 114 | 122 | 74 | |
| 16/10 | 13 | 58 | <0.4 | 13.9 | 5.8 | 7.2 | 118 | 128 | 84 | |
| 23/10 | 23 | 73 | <0.4 | 15.0 | 9.8 | 7.2 | 108 | 116 | 83 | |
| 30/10 | 22 | 62 | 1.1 | 12.5 | 4.0 | 7.1 | 101 | 141 | 79 | |
| 6/11 | 18 | 59 | <0.4 | 8.8 | 0.9 | 7.3 | 105 | 148 | 77 | |
| 13/11 | 11 | 45 | <0.4 | 14.9 | 5.6 | 7.3 | 111 | 121 | 80 | |
| 20/11 | 31 | 116 | 49 | 0.1 | 7.2 | 7.5 | 102 | 307 | 106 | |
| 27/11 | 11 | 48 | <0.4 | 9.8 | 0.1 | 7.5 | 122 | 163 | 83 | |

AGE: Sludge age (days)
 ALK: Alkalinity (CaCO₃ mg/L)
 AMM: Ammonia (N mg/L)
 CL: Chloride (Cl⁻ mg/L)
 COD: Chemical oxygen demand (mg COD/L)
 CON: Conductivity (mS/m)
 DSVI: Dissolved sludge volume index (ml/g)
 Flow: Raw flow for entire plant (ML/day)
 ND: Not determined
 NN: Nitrate and nitrite (N mg/L)
 OP: ortho-phosphate (P mg/L)
 RASSS: RAS suspended solids (mg/L)
 SETS: Settleable solids (30 min ml/L)
 SS: Suspended solids (105°C mg/L)
 SVI: Sludge volume index (ml/g)
 SWR: Sludge wasting rate (kL/day)
 TKN: Total Kjeldahl nitrogen (N mg/L)
 TP: Total phosphorus (P mg/L)
 VSS: Volatile suspended solids (mg/L)

Figures in red are anomalies and not transcription errors from the laboratory printouts.

| ANNEXURE 1.4 RAW DATA FROM CAPE FLATS WWTP | | | | | | | | | | | |
|--------------------------------------------|------|-----|-----|------|------|------|------|-----|-----|-----|-----|
| INFLUENT FROM PST EFGH | | | | | | | | | | | |
| Date: | Flow | SS | COD | TKN | AMM | TP | OP | PH | CL | ALK | CON |
| 6/6 | 170 | 142 | 353 | 47.0 | 37.0 | 12.2 | 10.1 | 7.0 | 130 | 239 | 100 |
| 11/6 | 222 | 196 | 340 | 36.5 | 22.0 | 9.8 | 6.3 | 6.9 | 98 | 194 | 86 |
| 18/6 | 258 | 138 | 328 | 42.3 | 30.2 | 9.4 | 7.0 | 7.1 | 112 | 231 | 92 |
| 25/6 | 201 | 128 | 286 | 42.9 | 28.8 | 8.2 | 5.7 | 7.1 | 97 | 212 | 89 |
| 2/7 | 222 | 108 | 281 | 35.8 | 25.6 | 9.0 | 7.3 | 7.3 | 114 | 221 | 94 |
| 9/7 | 226 | 152 | 375 | 47.4 | 30.9 | 13.3 | 10.6 | 7.2 | 104 | 223 | 97 |
| 17/7 | 167 | 164 | 404 | 46.6 | 32.7 | 12.4 | 9.5 | 7.2 | 98 | 218 | 99 |
| 23/7 | 191 | 100 | 268 | 43.6 | 33.9 | 9.4 | 7.5 | 7.0 | 117 | 229 | 102 |
| 30/7 | 318 | 98 | 217 | 22.2 | 17.6 | 6.7 | 4.2 | 7.2 | 93 | 207 | 88 |
| 6/8 | 318 | 233 | 397 | 32.2 | 15.7 | 7.7 | 4.4 | 6.8 | 77 | 159 | 65 |
| 13/8 | 350 | 88 | 228 | 32.9 | 19.5 | 6.5 | 4.0 | 7.2 | 82 | 197 | 88 |
| 20/8 | 328 | 102 | 223 | 33.9 | 21.8 | 6.7 | 4.9 | 7.5 | 109 | 206 | 92 |
| 27/8 | 281 | 114 | 259 | 33.8 | 22.9 | 7.4 | 5.4 | 7.5 | 117 | 215 | 98 |
| 3/9 | 315 | 124 | 318 | 34.7 | 24.1 | 8.8 | 6.1 | 7.4 | 95 | 220 | 92 |
| 10/9 | 284 | 136 | 305 | 39.2 | 26.4 | 10.6 | 7.9 | 7.4 | 93 | 220 | 90 |
| 17/9 | 204 | 134 | 339 | 40.7 | 32.4 | 7.5 | 6.1 | 7.4 | 100 | 236 | 96 |
| 24/9 | 184 | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| 1/10 | 184 | 148 | 395 | 45.9 | 40.2 | 10.8 | 9.2 | 7.4 | 109 | 257 | 113 |
| 8/10 | 174 | 144 | 398 | 53.3 | 38.5 | 10.0 | 7.4 | 7.4 | 99 | 250 | 93 |
| 15/10 | 178 | 128 | 306 | 54.9 | 42.8 | 10.4 | 9.1 | 7.4 | 106 | 260 | 98 |
| 22/10 | 155 | 164 | 397 | 54.9 | 44.8 | 9.8 | 7.9 | 7.3 | 99 | 255 | 94 |
| 29/10 | 162 | 136 | 397 | 55.5 | 43.8 | 11.5 | 9.3 | 7.3 | 115 | 255 | 101 |
| 5/11 | 154 | 260 | 516 | 56.6 | 38.2 | 11.4 | 8.6 | 7.4 | 96 | 242 | 95 |
| 12/11 | 166 | 106 | 374 | 60.2 | 49.7 | 9.4 | 8.3 | 7.5 | 119 | 278 | 106 |
| 19/11 | ND | 124 | 376 | 62.3 | 52.8 | 10.6 | 9.3 | 7.5 | 114 | 286 | 107 |
| 26/11 | 157 | 242 | 541 | 73.3 | 52.6 | 12.4 | 7.5 | 7.5 | 118 | 289 | 109 |

| MIXED LIQUOR REACTOR G | | | | | | | | | |
|------------------------|------|------|------|-----|------|-------|-----|------|-----|
| Date | SETS | SS | VSS | SVI | DSVI | RASSS | SWR | RASF | AGE |
| 6/6 | 800 | 5400 | 4305 | 148 | 82 | 7930 | 690 | ND | 26 |
| 11/6 | 260 | 2965 | ND | 66 | 71 | 8485 | 690 | ND | 22 |
| 18/6 | 440 | 4140 | ND | 106 | 68 | 6210 | 690 | ND | 19 |
| 25/6 | 500 | 4280 | ND | 109 | 84 | 8850 | 690 | ND | 20 |
| 2/7 | 510 | 5190 | 4180 | 98 | 77 | 8900 | 690 | ND | 40 |
| 9/7 | 780 | 5740 | ND | 136 | 77 | 9760 | 690 | ND | 47 |
| 17/7 | 830 | 6760 | ND | 123 | 71 | 9990 | ND | ND | ND |
| 23/7 | 860 | 6590 | ND | 131 | 79 | 11600 | ND | ND | ND |
| 30/7 | 270 | 3550 | ND | 76 | 68 | 10980 | 690 | 0 | 35 |
| 6/8 | 380 | 4220 | 3290 | 90 | 66 | 8640 | 690 | 0 | 7 |
| 13/8 | 270 | 3130 | ND | 86 | 77 | 7600 | 690 | ND | 62 |
| 20/8 | 320 | 3630 | ND | 88 | 66 | 6620 | 690 | ND | 27 |
| 27/8 | 300 | 3110 | ND | 96 | 90 | 6710 | 690 | ND | ND |
| 3/9 | 150 | 2050 | ND | 73 | ND | 4610 | 690 | ND | 16 |
| 10/9 | 230 | 3280 | ND | 70 | ND | 6300 | 690 | ND | 29 |
| 17/9 | 240 | 3270 | ND | 73 | ND | 5610 | 690 | ND | 35 |
| 24/9 | 250 | 4270 | ND | 59 | ND | ND | 690 | ND | ND |
| 1/10 | 340 | 4560 | 3750 | 75 | 53 | ND | 690 | ND | 34 |
| 8/10 | 320 | 4600 | ND | 70 | 52 | 6800 | 691 | ND | 25 |
| 15/10 | 360 | 4930 | ND | 73 | 57 | 7530 | 691 | ND | 27 |
| 22/10 | 500 | 6470 | ND | 77 | 56 | 8680 | 691 | ND | 37 |
| 29/10 | 780 | 8030 | ND | 97 | 60 | 11720 | 691 | ND | 22 |
| 5/11 | 670 | 5960 | 4730 | 112 | 67 | 9070 | 691 | ND | 32 |
| 12/11 | 300 | 4150 | ND | 72 | 58 | 9610 | 691 | ND | 46 |
| 19/11 | 480 | 5950 | ND | 81 | 54 | 7510 | ND | ND | ND |
| 26/11 | 530 | 5800 | ND | 91 | 62 | 7560 | 691 | ND | 31 |

| MIXED LIQUOR REACTOR H | | | | | | | | |
|------------------------|------|------|------|-----|------|-------|-----|-----|
| Date | SETS | SS | VSS | SVI | DSVI | RASSS | SWR | AGE |
| 6/6 | 620 | 5530 | 4425 | 112 | 72 | 7930 | 690 | 26 |
| 11/6 | 350 | 4135 | ND | 85 | 68 | 8485 | 690 | 22 |
| 18/6 | 370 | 3950 | ND | 94 | 71 | 6210 | 690 | 19 |
| 25/6 | 590 | 5010 | ND | 118 | 72 | 8850 | 690 | 20 |
| 2/7 | 520 | 5230 | 4230 | 99 | 76 | 8900 | 690 | 40 |
| 9/7 | 800 | 5810 | ND | 138 | 76 | 9760 | 690 | 47 |
| 17/7 | 820 | 6540 | ND | 125 | 73 | 9990 | ND | ND |
| 23/7 | 850 | 6820 | ND | 125 | 70 | 11600 | ND | ND |
| 30/7 | 260 | 3670 | ND | 71 | 65 | 10980 | 690 | 35 |
| 6/8 | 460 | 4260 | 3710 | 108 | 66 | 8640 | 690 | 7 |
| 13/8 | 270 | 3080 | ND | 88 | 78 | 7600 | 690 | 62 |
| 20/8 | 360 | 3770 | ND | 95 | 74 | 6620 | 690 | 27 |
| 27/8 | 320 | 3390 | ND | 94 | 83 | 6710 | 690 | ND |
| 3/9 | 150 | 2040 | ND | 74 | ND | 4610 | 690 | 16 |
| 10/9 | 230 | 3300 | ND | 70 | ND | 6300 | 690 | 29 |
| 17/9 | 230 | 3230 | ND | 71 | ND | 5610 | 690 | 35 |
| 24/9 | 270 | 5380 | ND | 50 | 52 | ND | 690 | ND |
| 1/10 | 360 | 4720 | 3820 | 76 | 59 | ND | 690 | 34 |
| 8/10 | 300 | 4400 | ND | 68 | 45 | 6800 | 691 | 25 |
| 15/10 | 390 | 4680 | ND | 83 | 68 | 7530 | 691 | 27 |
| 22/10 | 620 | 3930 | ND | 158 | 102 | 8680 | 691 | 37 |
| 29/10 | 770 | 8080 | ND | 95 | 59 | 11720 | 691 | 22 |
| 5/11 | 720 | 6390 | 5150 | 113 | 63 | 9070 | 691 | 32 |
| 12/11 | 480 | 5330 | ND | 90 | 60 | 9610 | 691 | 46 |
| 19/11 | 500 | 6120 | ND | 82 | 52 | 7510 | ND | ND |
| 26/11 | 590 | 5840 | ND | 101 | 62 | 7560 | 691 | 31 |

| CLARIFIER GH (EFFLUENT) | | | | | | | | | |
|-------------------------|-----|-----|------|------|------|-----|-----|-----|-----|
| Date: | SS | COD | AMM | NN | OP | PH | CL | ALK | CON |
| 6/6 | 9 | 44 | <0.4 | 10.0 | 7.4 | 6.9 | 115 | 82 | 74 |
| 11/6 | 4 | 30 | <0.4 | 7.3 | 4.3 | 6.6 | 106 | 98 | 75 |
| 18/6 | 6 | 37 | <0.4 | 13.7 | 6.5 | 7.0 | 109 | 78 | 74 |
| 25/6 | 8 | 41 | <0.4 | 9.7 | 5.1 | 6.7 | 102 | 89 | 71 |
| 2/7 | 10 | 51 | <0.4 | 10.5 | 4.9 | 6.9 | 108 | 86 | 75 |
| 9/7 | 6 | 37 | <0.4 | 14.4 | 7.1 | 7.0 | 105 | 81 | 76 |
| 17/7 | 5 | 33 | <0.4 | 9.2 | 5.6 | 6.9 | 110 | 90 | 75 |
| 23/7 | 5 | 30 | <0.4 | 8.4 | 4.7 | 6.9 | 111 | 94 | 76 |
| 30/7 | 17 | 51 | 0.4 | 8.6 | 2.3 | 7.0 | 87 | 113 | 75 |
| 6/8 | 173 | 263 | <0.4 | 9.7 | 3.5 | 6.6 | 105 | 110 | 75 |
| 13/8 | 4 | 33 | <0.4 | 8.4 | 3.4 | 7.0 | 117 | 114 | 71 |
| 20/8 | 5 | 41 | <0.4 | 11.1 | 4.8 | 7.4 | 103 | 99 | 78 |
| 27/8 | 3 | 26 | <0.4 | 11.6 | 3.5 | 7.2 | 89 | 88 | 71 |
| 3/9 | 58 | 118 | <0.4 | 6.2 | 2.2 | 7.3 | 90 | 125 | 73 |
| 10/9 | 4 | 34 | <0.4 | 8.9 | 5.2 | 7.3 | 96 | 92 | 73 |
| 17/9 | 4 | 37 | 0.5 | 6.2 | 4.9 | 7.4 | 97 | 112 | 73 |
| 24/9 | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| 1/10 | 18 | 55 | <0.4 | 13.7 | 6.8 | 7.2 | 105 | 83 | 81 |
| 8/10 | 10 | 44 | <0.4 | 10.2 | 5.0 | 7.3 | 103 | 90 | 72 |
| 15/10 | 8 | 35 | <0.4 | 9.7 | 4.7 | 7.4 | 102 | 89 | 73 |
| 22/10 | 8 | 37 | <0.4 | 9.4 | 6.3 | 7.2 | 104 | 98 | 73 |
| 29/10 | 9 | 34 | 1.5 | 7.2 | 8.8 | 7.1 | 111 | 108 | 77 |
| 5/11 | 7 | 34 | <0.4 | 5.5 | 4.4 | 7.3 | 95 | 104 | 68 |
| 12/11 | 11 | 45 | <0.4 | 5.0 | 6.3 | 7.3 | 111 | 116 | 75 |
| 19/11 | 9 | 48 | <0.4 | 11.6 | 12.2 | 7.2 | 108 | 99 | 74 |
| 26/11 | 8 | 33 | <0.4 | 11.5 | 9.4 | 7.0 | 107 | 95 | 73 |

| | |
|---------------|-------------------------------------------------------|
| AGE: | Sludge age (days) |
| ALK: | Alkalinity (CaCO ₃ mg/L) |
| AMM: | Ammonia (N mg/L) |
| CL: | Chloride (Cl ⁻ mg/L) |
| COD: | Chemical oxygen demand (mg COD/L) |
| CON: | Conductivity (mS/m) |
| DSVI: | Dissolved sludge volume index (ml/g) |
| Flow: | Flow (ML/day) for entire plant into PST ABCD and EFGH |
| ND: | Not determined |
| NN: | Nitrate and nitrite (N mg/L) |
| OP: | ortho-phosphate (P mg/L) |
| RASF: | RAS flow rate(s) (kL/day) |
| RASSS: | RAS suspended solids (mg/L) |
| SETS: | Settleable solids (30 min ml/L) |
| SS: | Suspended solids (105°C mg/L) |
| SVI: | Sludge volume index (ml/g) |
| SWR: | Sludge wasting rate (kL/day) |
| TKN: | Total Kjeldahl nitrogen (N mg/L) |
| TP: | Total phosphorus (P mg/L) |
| VSS: | Volatile suspended solids (mg/L) |

Figures in red are anomalies and not transcription errors from the laboratory printouts. It was not in the scope of this study to speculate on reasons for anomalies.

ANNEXURE 1.5 RAW DATE FROM KRAAIFONTEIN WWTP

PST (INFLUENT)

| Date: | FLOW | SS | COD | TKN | AMM | TP | OP | PH | CON | CL | ALK |
|-------|------|-----|-----|------|-----|------|------|-----|-----|-----|-----|
| 6/6 | 12.8 | 242 | 460 | 65.5 | 48 | 10.5 | 6.9 | 7.2 | 95 | 104 | 247 |
| 13/6 | 30.8 | 215 | 544 | 57.5 | 44 | 13.5 | 9.3 | 7.5 | 113 | 140 | 283 |
| 20/6 | 26.0 | 156 | 507 | 75.2 | 57 | 12.6 | 9.8 | 7.3 | 109 | 122 | 290 |
| 27/6 | 21.6 | 206 | 550 | 53.9 | 33 | 12.6 | 7.6 | 7.3 | 107 | 131 | 254 |
| 4/7 | 18.0 | 236 | 566 | 81.2 | 61 | 13.7 | 10.1 | 7.4 | 122 | 130 | 315 |
| 11/7 | 16.6 | 190 | 470 | 69.8 | 52 | 11.5 | 8.9 | 7.2 | 125 | 55 | 294 |
| 18/7 | 23.2 | 240 | 599 | 78.7 | 56 | 13.5 | 10.7 | 7.3 | 113 | 125 | 281 |
| 25/7 | 23.9 | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| 1/8 | 32.3 | 162 | 400 | 41.2 | 29 | 10.7 | 6.9 | 7.1 | 107 | 128 | 251 |
| 8/8 | 29.2 | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| 15/8 | 32.1 | 132 | 403 | 40.2 | 28 | 8.3 | 5.7 | 7.1 | 98 | 123 | 253 |
| 22/8 | 25.8 | 140 | 417 | 68.5 | 55 | 11.4 | 8.9 | 7.8 | 118 | 125 | 323 |
| 29/8 | 24.6 | 137 | 368 | 51.3 | 41 | 10.4 | 7.8 | 7.6 | 102 | 141 | 279 |
| 5/9 | 24.5 | 151 | 311 | 45.5 | 33 | 7.2 | 5.0 | 7.6 | 87 | 93 | 235 |
| 12/9 | 25.2 | 180 | 520 | 52.3 | 43 | 12.0 | 9.2 | 7.3 | 109 | 128 | 272 |
| 19/9 | 23.6 | 140 | 385 | 64.2 | 52 | 10.4 | 7.8 | 7.7 | 106 | 113 | 282 |
| 26/9 | 21.1 | 176 | 467 | ND | 65 | 12.3 | 9.8 | 7.7 | 112 | 107 | 308 |
| 3/10 | 21.2 | 124 | 412 | 69.5 | 62 | 9.7 | 7.6 | 7.7 | 100 | 92 | 288 |
| 10/10 | 20.6 | 148 | 401 | 80.7 | 67 | 13.2 | 9.4 | 7.8 | 115 | 125 | 309 |
| 17/10 | 19.3 | 148 | 488 | 70.1 | 54 | 12.6 | 10.7 | 7.6 | 100 | 100 | 270 |
| 24/10 | 19.6 | 240 | 512 | 71.9 | 58 | 13.1 | 9.9 | 7.6 | 109 | 112 | 295 |
| 1/11 | 20.1 | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| 8/11 | 19.6 | 178 | 571 | 89.9 | 72 | 13.0 | 10.0 | 7.6 | 117 | 130 | 309 |
| 14/11 | 19.8 | 152 | 463 | 86.7 | 74 | 12.0 | 9.4 | 7.9 | 111 | 115 | 311 |
| 21/11 | 19.2 | 151 | 400 | 55.3 | 48 | 9.7 | 7.3 | 7.5 | 81 | 80 | 226 |
| 28/11 | 20.2 | 177 | 400 | 73.5 | 63 | 14.5 | 10.7 | 7.7 | 105 | 111 | 303 |

MIXED LIQUOR

| Date | SETS | SS | VSS | SVI | DSVI | PH | RASSS | SWR |
|-------|------|------|------|-----|------|-----|-------|------|
| 6/6 | 970 | 6120 | 4715 | 158 | 111 | 6.8 | 12430 | ND |
| 13/6 | ND | ND | ND | ND | ND | ND | ND | ND |
| 20/6 | 980 | 5740 | ND | 171 | 118 | 6.9 | 9060 | ND |
| 27/6 | 970 | 6550 | ND | 148 | 110 | 6.8 | 9910 | ND |
| 4/7 | 970 | 6760 | 5050 | 144 | 130 | 6.5 | 10420 | ND |
| 11/7 | 960 | 5330 | ND | 180 | 128 | 6.7 | 10590 | 797 |
| 18/7 | 970 | 5950 | ND | 163 | 114 | 6.3 | 10600 | 752 |
| 25/7 | 960 | 5300 | ND | 181 | 113 | 6.8 | ND | 564 |
| 1/8 | 610 | 4260 | ND | 143 | 122 | 6.9 | 7630 | 890 |
| 8/8 | 800 | 3450 | ND | 232 | 128 | 7.1 | ND | 958 |
| 15/8 | 450 | 3830 | ND | 117 | 104 | 6.7 | 7990 | 1076 |
| 22/8 | 500 | 3820 | ND | 131 | 105 | 7.0 | 5570 | 866 |
| 29/8 | 520 | 3720 | ND | 140 | 97 | 7.1 | 4510 | ND |
| 5/9 | 440 | 3600 | 2765 | 122 | 89 | 7.0 | 7590 | 730 |
| 12/9 | 470 | 3180 | ND | 148 | 101 | 6.9 | 5570 | 900 |
| 19/9 | 310 | 2680 | ND | 116 | 90 | 7.0 | 2670 | 807 |
| 26/9 | 450 | 3830 | ND | 118 | 84 | 6.9 | 7000 | 616 |
| 3/10 | 770 | 4570 | 3590 | 169 | 105 | 6.9 | 9330 | 686 |
| 10/10 | 700 | 4830 | ND | 145 | 91 | 6.9 | 7160 | ND |
| 17/10 | 940 | 5360 | ND | 175 | 90 | 7.0 | 7710 | 558 |
| 24/10 | 750 | 4860 | ND | 154 | 99 | 6.8 | 10360 | ND |
| 1/11 | 50 | 700 | ND | 71 | 118 | 7.6 | 1040 | ND |
| 8/11 | 950 | 5780 | 4610 | 164 | 111 | 7.6 | 12850 | ND |
| 14/11 | 360 | 2440 | ND | 148 | 115 | 6.9 | 3250 | ND |
| 21/11 | 930 | 4175 | ND | 223 | 125 | 6.9 | 10865 | ND |
| 28/11 | 980 | 4600 | ND | 213 | 148 | 6.9 | 6140 | ND |

| AVERAGE FOR FOR CLARIFIER 1, 2 AND 3 (EFFLUENT) | | | | | | | | | | |
|-------------------------------------------------|-----------|------------|------|------|------------|-----|-----|-----|-----|--|
| Date: | SS | COD | AMM | NN | OP | PH | CON | CL | ALK | |
| 6/6 | 65 | 92 | <0.4 | 17.2 | 2.7 | 6.4 | 69 | 114 | 27 | |
| 13/6 | 24 | 52 | 1.4 | 7.9 | 0.3 | 7.2 | 88 | 146 | 125 | |
| 20/6 | 26 | 66 | 0.4 | 15.3 | 0.5 | 6.9 | 75 | 124 | 76 | |
| 27/6 | 9 | 55 | <0.4 | 8.7 | 0.2 | 7.2 | 83 | 132 | 106 | |
| 4/7 | 49 | 91 | 1.1 | 5.9 | 0.1 | 7.1 | 81 | 132 | 109 | |
| 11/7 | 34 | 62 | 1.9 | 15.5 | 3.5 | 6.9 | 89 | 138 | 83 | |
| 18/7 | 14 | 56 | 2.1 | 10.4 | 0.1 | 7.0 | 81 | 127 | 92 | |
| 25/7 | ND | ND | <0.4 | 12.3 | 0.2 | 7.4 | 84 | 134 | 102 | |
| 1/8 | a) 13/37 | c) 43/109 | <0.4 | 11.8 | 1.4 | 7.2 | 89 | 135 | 113 | |
| 8/8 | ND | ND | ND | ND | ND | ND | ND | ND | ND | |
| 15/8 | 7 | 35 | <0.4 | 16.3 | 0.7 | 7.2 | 81 | 118 | 110 | |
| 22/8 | 6 | 48 | <0.4 | 22.3 | 0.3 | 7.2 | 83 | 119 | 62 | |
| 29/8 | 10 | 50 | <0.4 | 16.3 | 0.3 | 7.2 | 88 | 137 | 87 | |
| 5/9 | 9 | 35 | 3.1 | 13.4 | 0.9 | 7.2 | 75 | 103 | 97 | |
| 12/9 | 10 | 46 | 0.2 | 21.5 | 6.0 | 6.9 | 77 | 111 | 43 | |
| 19/9 | 6 | 45 | <0.4 | 21.9 | 7.0 | 6.9 | 77 | 109 | 39 | |
| 26/9 | 31 | 73 | 2.2 | 12.5 | 1.4 | 7.2 | 72 | 103 | 85 | |
| 3/10 | 12 | 52 | 2.9 | 3.8 | 0.2 | 7.3 | 69 | 106 | 118 | |
| 10/10 | 20 | 49 | 9.3 | 0.9 | 0.2 | 7.5 | 78 | 117 | 153 | |
| 17/10 | 47 | 52 | 0.4 | 13.7 | 0.2 | 7.1 | 66 | 98 | 72 | |
| 24/10 | b) 79/150 | d) 112/187 | <0.4 | 25.3 | 0.1 | 6.5 | 69 | 104 | 16 | |
| 1/11 | 65 | 143 | 42 | 0.2 | e) 2.3/3.8 | 7.6 | 99 | 116 | 243 | |
| 8/11 | 12 | 47 | <0.4 | 11.5 | 0.1 | 7.2 | 71 | 112 | 78 | |
| 14/11 | 5 | 30 | <0.4 | 8.4 | 0.2 | 7.2 | 68 | 109 | 79 | |
| 21/11 | 18 | 53 | 0.4 | 12.4 | 0.1 | 7.2 | 66 | 104 | 77 | |
| 28/11 | 18 | 38 | <0.4 | 16.0 | 5.9 | 7.1 | 66 | 107 | 51 | |

Note: The results in most cases for all three clarifiers are very close. In the case where there are anomalous results, 2 results are given in red. The first figures are those excluding the anomalous result and the second includes the anomalous result. The latter are the figures used for the graphs in the "Results and Discussion".

The anomalous results are as follows:

SS: (a) 1st Aug - STE 1 - 187 (b) 24th Aug - STE 1 - 292
 COD: (c) 1st Aug - STE 1 - 240 (d) 24th Aug - STE 1 - 337
 OP: (e) 1st Nov - STE3 - 7.0

No results were given for STE 3 on the 14th Nov.

ALK: Alkalinity (CaCO₃ mg/L)
 AMM: Ammonia (N mg/L)
 CL: Chloride (Cl⁻ mg/L)
 COD: Chemical oxygen demand (mg COD/L)
 CON: Conductivity (mS/m)
 DSVI: Dissolved sludge volume index (ml/g)
 Flow: Flow (ML/day) raw flow for entire plant
 ND: Not determined
 NN: Nitrate and nitrite (N mg/L)
 OP: ortho-phosphate (P mg/L)
 RASSS: RAS suspended solids (mg/L)
 SETS: Settleable solids (30 min ml/L)
 SS: Suspended solids (105°C mg/L)
 SVI: Sludge volume index (ml/g)
 SWR: Sludge wasting rate (kL/day)
 TKN: Total Kjeldahl nitrogen (N mg/L)
 TP: Total phosphorus (P mg/L)
 VSS: Volatile suspended solids (mg/L)

Figures in red are anomalies and not transcription errors from the laboratory printouts.

ANNEXURE 1.6 RAW DATA FROM MACASSAR WWTP

RAW INFLUENT (NO PST)

| Date: | Flow | SS | VSS | SETS | COD | TKN | AMM | TP | OP | PH | CON | CL | ALK |
|-------|------|-----|-----|------|-----|------|------|------|-----|-----|-----|-----|-----|
| 3/6 | 32.7 | ERR | ND | 60 | ERR | ERR | 55.5 | ERR | 9.7 | 6.9 | 126 | 165 | 327 |
| 10/6 | 44.9 | 162 | ND | 4 | 282 | 34.8 | 25.5 | 5.8 | 3.8 | 7.0 | 106 | 146 | 249 |
| 17/6 | 51.6 | 84 | ND | <1 | 339 | 52.7 | 39.1 | 9.0 | 6.3 | 7.2 | 115 | 151 | 290 |
| 24/6 | 36.9 | 119 | ND | 0 | 384 | 61.0 | 44.9 | 8.1 | 6.3 | 7.4 | 117 | 154 | 271 |
| 1/7 | 49.5 | 202 | 168 | 4 | 479 | 48.0 | 32.1 | 8.2 | 5.7 | 7.5 | 123 | 163 | 292 |
| 8/7 | 41.7 | 208 | ND | 8 | 440 | 50.3 | 34.3 | 8.1 | 5.4 | 7.5 | 117 | 159 | 269 |
| 15/7 | 37.5 | 262 | ND | 7 | 583 | ND | 43.3 | ND | 7.4 | 7.2 | 129 | 185 | 282 |
| 22/7 | 38.8 | 98 | ND | <1 | 365 | 56.9 | 41.2 | 8.8 | 6.7 | 7.2 | 123 | 161 | 282 |
| 29/7 | 54.7 | 30 | ND | 0 | 95 | 16.4 | 11.3 | 2.8 | 1.6 | 7.5 | 102 | 146 | 215 |
| 5/8 | 57.9 | 118 | 106 | <1 | 295 | ND | 27.3 | 7.0 | 5.1 | 7.1 | 125 | 181 | 269 |
| 12/8 | 59.9 | 125 | ND | <1 | 276 | 38.9 | 25.4 | 6.3 | 4.5 | 7.3 | 122 | 175 | 271 |
| 19/8 | 55.9 | 178 | ND | 8 | 813 | 45.5 | 28.6 | 6.7 | 4.9 | 7.6 | 128 | 176 | 270 |
| 26/8 | 45.4 | 242 | ND | 5 | 452 | 43.4 | 37.6 | 7.2 | 5.4 | 7.4 | 108 | 133 | 255 |
| 2/9 | 44.8 | 88 | 74 | <1 | 303 | 44.4 | 29.1 | 7.5 | 5.7 | 7.7 | 121 | 165 | 257 |
| 9/9 | 38.0 | 54 | ND | <1 | 259 | 43.9 | 37.3 | 7.8 | 6.7 | 7.7 | 123 | 168 | 262 |
| 16/9 | 36.2 | 111 | ND | 1 | 398 | 55.7 | 43.0 | 9.3 | 7.6 | 7.7 | 130 | 173 | 288 |
| 24/9 | 32.3 | 268 | ND | 4 | 491 | 61.4 | 35.7 | 10.7 | 7.4 | 7.6 | 118 | 155 | 177 |
| 30/9 | 32.4 | 238 | 218 | 8 | 502 | 59.3 | 41.7 | 10.0 | 6.6 | 7.5 | 132 | 164 | 266 |
| 7/10 | 34.1 | 300 | ND | 6 | 508 | 49.5 | 31.7 | 7.5 | 4.8 | 7.5 | 111 | 154 | 247 |
| 14/10 | 34.7 | 112 | ND | <1 | 353 | 64.1 | 44.6 | 9.1 | 7.4 | 7.5 | 134 | 208 | 281 |
| 21/10 | 31.8 | 122 | ND | <1 | 365 | 57.9 | 44.2 | 9.7 | 8.0 | 7.5 | 124 | 184 | 267 |
| 28/10 | 36.8 | 120 | ND | <1 | 311 | 51.6 | 37.5 | 8.2 | 6.5 | 7.6 | 205 | 439 | 257 |
| 4/11 | 32.3 | 214 | 190 | 0 | 408 | 57.9 | 46.9 | 9.2 | 7.6 | 7.5 | 127 | 196 | 268 |
| 11/11 | 30.8 | 492 | ND | 19 | 947 | 75.5 | 46.6 | 12.2 | 8.0 | 7.5 | 140 | 223 | 270 |
| 18/11 | 29.6 | 480 | ND | 12 | 886 | 71.4 | 51.4 | 12.6 | 9.1 | 7.5 | 128 | 191 | 282 |
| 25/11 | 32.8 | 190 | ND | 4 | 506 | 60.7 | 47.6 | 11.6 | 8.8 | 7.5 | 130 | 184 | 281 |

MIXED LIQUOR REACTOR 1

| Date | SETS | SS | VSS | SVI | DSVI | PH | SWR | AGE | RASF | RASSS |
|-------|------|------|------|-----|------|-----|------|-----|------|-------|
| 3/6 | 740 | 4400 | 3505 | 168 | 109 | 6.9 | 909 | 36 | ND | 6310 |
| 10/6 | 610 | 4265 | ND | 143 | 94 | 6.8 | 893 | 37 | ND | 7595 |
| 17/6 | 750 | 5010 | ND | 150 | 96 | 7.0 | 955 | 39 | 32.0 | 6910 |
| 24/6 | 920 | 4730 | ND | 195 | 135 | 6.9 | 660 | 52 | 37.0 | 8330 |
| 1/7 | 700 | 4340 | 3400 | 162 | 120 | 7.0 | 826 | 43 | 35.0 | 7550 |
| 8/7 | 650 | 4000 | ND | 162 | 120 | 6.9 | 1002 | 34 | ND | 6510 |
| 15/7 | 730 | 4570 | ND | 160 | 114 | 6.8 | 1002 | 36 | ND | 7850 |
| 22/7 | 900 | 4820 | ND | 187 | 124 | 6.7 | 600 | 62 | ND | 7500 |
| 29/7 | 400 | 3040 | ND | 132 | 92 | 6.7 | 615 | 58 | ND | 8720 |
| 5/8 | 530 | 3160 | ND | 168 | 127 | 6.9 | 526 | 48 | ND | 7210 |
| 12/8 | 600 | 4090 | ND | 147 | 108 | 6.9 | 711 | 38 | 33.0 | 7440 |
| 19/8 | 870 | 4320 | ND | 201 | 148 | 7.1 | 739 | 47 | 33.0 | 7780 |
| 26/8 | 620 | 3830 | ND | 162 | 125 | 7.1 | 739 | 46 | 32.0 | 6590 |
| 2/9 | 690 | 3950 | 3050 | 175 | 122 | 7.2 | 681 | 54 | 30.0 | 7100 |
| 9/9 | 730 | 4510 | ND | 162 | 115 | 7.2 | 813 | 46 | 30.0 | 5140 |
| 16/9 | 620 | 3440 | ND | 180 | 140 | 7.1 | 1022 | 34 | 31.0 | 4710 |
| 24/9 | 740 | 3540 | ND | 209 | 136 | 7.1 | 1022 | ND | 31.0 | ND |
| 30/9 | 700 | 4030 | 3320 | 174 | 119 | 7.1 | 826 | 44 | 33.0 | 5150 |
| 7/10 | 430 | 2860 | ND | 150 | 84 | 5.9 | 835 | 42 | 28.0 | 5770 |
| 14/10 | 540 | 3690 | ND | 146 | 87 | 7.2 | 522 | 69 | 18.0 | 6370 |
| 21/10 | 650 | 4320 | ND | 150 | 93 | 7.1 | 114 | 268 | 18.0 | 6840 |
| 28/10 | 630 | 4830 | ND | 130 | 83 | 6.9 | ND | ND | 18.0 | 7830 |
| 4/11 | 600 | 3940 | 3170 | 152 | 102 | 7.1 | 914 | 37 | 18.0 | 7390 |
| 11/11 | 570 | 4200 | ND | 136 | 86 | 7.0 | 392 | 93 | 19.0 | 7750 |
| 18/11 | 420 | 4940 | ND | 85 | 65 | 7.2 | 715 | 50 | 18.0 | 6660 |
| 25/11 | 500 | 3350 | ND | 149 | 108 | 7.1 | 531 | 59 | 18.0 | 7600 |

| MIXED LIQUOR REACTOR 2 | | | | | | | | | | |
|------------------------|------|------|------|-----|------|-----|------|-----|------|-------|
| Date | SETS | SS | VSS | SVI | DSVI | PH | SWR | AGE | RASF | RASSS |
| 3/6 | 980 | 4220 | 3445 | 232 | 190 | 6.8 | 1020 | 34 | ND | 5350 |
| 10/6 | 780 | 3645 | ND | 214 | 154 | 6.9 | 990 | 32 | ND | 6625 |
| 17/6 | 910 | 4470 | ND | 204 | 152 | 6.5 | 962 | 38 | 28.0 | 6190 |
| 24/6 | 980 | 4950 | ND | 198 | 154 | 6.8 | 586 | 63 | 35.0 | 5410 |
| 1/7 | 870 | 4280 | 3440 | 199 | 137 | 6.9 | 1007 | 34 | 37.0 | 6730 |
| 8/7 | 850 | 4160 | ND | 204 | 144 | 6.8 | 1037 | 35 | ND | 6300 |
| 15/7 | 860 | 4250 | ND | 190 | 124 | 6.7 | 1037 | 34 | ND | 6920 |
| 22/7 | 930 | 4540 | ND | 205 | 141 | 6.7 | 604 | 61 | ND | 6860 |
| 29/7 | 870 | 4670 | ND | 186 | 120 | 6.6 | 595 | 61 | ND | 8550 |
| 5/8 | 720 | 4300 | ND | 167 | 121 | 6.8 | 646 | 47 | ND | 7490 |
| 12/8 | 700 | 4480 | ND | 156 | 125 | 6.9 | 818 | 44 | 31.0 | 6830 |
| 19/8 | 820 | 3570 | ND | 230 | 168 | 7.0 | 747 | 43 | 36.0 | 5880 |
| 26/8 | 800 | 4390 | ND | 182 | 137 | 7.0 | 747 | 49 | 36.0 | 6250 |
| 2/9 | 840 | 4430 | 3350 | 189 | 126 | 7.1 | 696 | 54 | 35.0 | 6340 |
| 9/9 | 950 | 4090 | ND | 232 | 176 | 7.2 | 777 | 48 | 33.0 | 6020 |
| 16/9 | 780 | 3730 | ND | 209 | 161 | 7.1 | 1043 | 35 | 34.0 | 5280 |
| 24/9 | 800 | 4200 | ND | 191 | 143 | 7.1 | 1043 | - | 34.0 | ND |
| 30/9 | 820 | 2670 | 4000 | 223 | 174 | 7.0 | 856 | 43 | 35.0 | 5160 |
| 7/10 | 700 | 3760 | ND | 186 | 117 | 7.1 | 861 | 42 | 31.0 | 6350 |
| 14/10 | 610 | 3290 | ND | 185 | 122 | 7.1 | 887 | 42 | 35.0 | 4840 |
| 21/10 | 830 | 4170 | ND | 199 | 125 | 7.1 | 618 | 59 | 32.0 | 5060 |
| 28/10 | 730 | 3550 | ND | 206 | 124 | 6.9 | 1079 | 34 | 35.0 | 13870 |
| 4/11 | 720 | 4030 | 3280 | 179 | 119 | 7.1 | 573 | 61 | 33.0 | 6080 |
| 11/11 | 830 | 4360 | ND | 190 | 128 | 7.0 | 492 | 75 | 32.0 | 6140 |
| 18/11 | 940 | 3800 | ND | 247 | 168 | 7.0 | 900 | 42 | 30.0 | 7180 |
| 25/11 | 970 | 6980 | ND | 139 | 138 | 7.0 | 696 | 52 | 30.0 | 7180 |

| CLARIFIER 1 (EFFLUENT) | | | | | | | | | |
|------------------------|----|-----|------|------|-----|-----|-----|-----|-----|
| Date: | SS | COD | AMM | NN | OP | PH | CON | CL | ALK |
| 3/6 | 23 | 76 | <0.4 | 16.4 | 8.8 | 7.2 | 92 | 144 | 69 |
| 10/6 | 13 | 38 | <0.4 | 11.8 | 3.8 | 7.3 | 85 | 129 | 102 |
| 17/6 | 2 | 41 | 0.4 | 5.6 | 7.3 | 7.8 | 90 | 143 | 131 |
| 24/6 | 10 | 58 | 0.5 | 7.4 | 7.1 | 7.5 | 90 | 149 | 102 |
| 1/7 | 6 | 37 | 0.7 | 12.0 | 4.1 | 7.7 | 89 | 146 | 108 |
| 8/7 | 11 | 44 | 0.8 | 17.3 | 5.4 | 7.4 | 94 | 149 | 84 |
| 15/7 | 10 | 33 | 3.2 | 2.1 | 3.0 | 7.5 | 100 | 174 | 145 |
| 22/7 | 2 | 47 | 2.6 | 4.0 | 5.3 | 7.5 | 97 | 161 | 139 |
| 29/7 | 3 | 30 | <0.4 | 13.4 | 1.3 | 7.7 | 92 | 136 | 137 |
| 5/8 | 15 | 40 | <0.4 | 18.4 | 2.8 | 7.4 | 108 | 177 | 124 |
| 12/8 | 21 | 58 | <0.4 | 12.8 | 3.1 | 7.7 | 101 | 161 | 136 |
| 19/8 | 6 | 51 | 2.4 | 0.3 | 3.9 | 7.7 | 105 | 160 | 171 |
| 26/8 | 9 | 44 | 0.6 | 15.4 | 5.7 | 7.7 | 104 | 162 | 97 |
| 2/9 | 3 | 34 | <0.4 | 13.8 | 5.2 | 7.9 | 102 | 166 | 110 |
| 9/9 | 3 | 55 | <0.4 | 14.0 | 6.5 | 7.8 | 101 | 159 | 104 |
| 16/9 | 12 | 52 | 0.4 | 9.7 | 6.6 | 7.7 | 101 | 167 | 114 |
| 24/9 | ND | ND | 1.8 | 6.5 | 6.2 | 7.9 | 101 | 171 | 121 |
| 30/9 | 6 | 40 | <0.4 | 3.1 | 6.8 | 7.5 | 124 | 208 | 57 |
| 7/10 | 6 | 37 | <0.4 | 17.8 | 5.9 | 7.1 | 111 | 224 | 56 |
| 14/10 | 4 | 54 | 1.2 | 9.9 | 7.7 | 7.8 | 106 | 203 | 108 |
| 21/10 | 4 | 45 | <0.4 | 5.9 | 8.0 | 7.9 | 98 | 181 | 108 |
| 28/10 | 6 | 30 | <0.4 | 3.3 | 5.0 | 8.0 | 178 | 416 | 122 |
| 4/11 | 16 | 58 | <0.4 | 16.7 | 6.8 | 7.7 | 108 | 203 | 75 |
| 11/11 | 3 | 42 | <0.4 | 17.6 | 8.5 | 7.6 | 127 | 253 | 86 |
| 18/11 | 9 | 56 | 2.2 | 16.9 | 9.0 | 7.5 | 104 | 200 | 84 |
| 25/11 | 12 | 47 | 0.4 | 17.9 | 8.5 | 7.5 | 97 | 178 | 75 |

| CLARIFIER 2 (EFFLUENT) | | | | | | | | | |
|------------------------|----|-----|------|------|-----|-----|-----|-----|-----|
| Date: | SS | COD | AMM | NN | OP | PH | CON | CL | ALK |
| 3/6 | 13 | 58 | 1.1 | 0.8 | 7.6 | 7.5 | 88 | 155 | 122 |
| 10/6 | 16 | 38 | <0.4 | 6.1 | 3.5 | 7.3 | 82 | 129 | 125 |
| 17/6 | 4 | 33 | 0.7 | 0.7 | 7.1 | 7.6 | 89 | 143 | 185 |
| 24/6 | 3 | 40 | 0.9 | 0.7 | 5.9 | 7.6 | 88 | 149 | 126 |
| 1/7 | 10 | 41 | <0.4 | 6.9 | 4.1 | 7.8 | 89 | 139 | 129 |
| 8/7 | 7 | 44 | 0.8 | 6.0 | 5.3 | 7.6 | 90 | 150 | 119 |
| 15/7 | 12 | 33 | 3.2 | 0.3 | 1.1 | 7.6 | 100 | 173 | 154 |
| 22/7 | 3 | 44 | 3.1 | 0.9 | 4.8 | 7.6 | 97 | 164 | 148 |
| 29/7 | 3 | 30 | <0.4 | 14.8 | 0.2 | 7.7 | 100 | 153 | 142 |
| 5/8 | 13 | 40 | <0.4 | 13.3 | 0.2 | 7.5 | 108 | 177 | 145 |
| 12/8 | 5 | 48 | <0.4 | 7.5 | 3.0 | 7.8 | 97 | 157 | 154 |
| 19/8 | 10 | 47 | <0.4 | 9.9 | 5.4 | 7.8 | 105 | 166 | 129 |
| 26/8 | 4 | 29 | <0.4 | 7.2 | 3.8 | 7.3 | 93 | 146 | 124 |
| 2/9 | 2 | 34 | 1.5 | 0.3 | 5.3 | 8.0 | 100 | 171 | 155 |
| 9/9 | 2 | 37 | <0.4 | 10.7 | 5.7 | 7.9 | 100 | 159 | 114 |
| 16/9 | 7 | 37 | 0.4 | 4.3 | 6.2 | 8.0 | 100 | 171 | 130 |
| 24/9 | ND | ND | 1.6 | 0.3 | 4.9 | 7.9 | 100 | 177 | 133 |
| 30/9 | 4 | 36 | 0.5 | 1.4 | 4.8 | 7.9 | 118 | 205 | 128 |
| 7/10 | 7 | 26 | <0.4 | 10.9 | 5.2 | 7.7 | 110 | 223 | 80 |
| 14/10 | 3 | 43 | 0.6 | 7.9 | 7.2 | 7.9 | 106 | 206 | 109 |
| 21/10 | 4 | 41 | <0.4 | 1.5 | 7.9 | 7.8 | 97 | 182 | 119 |
| 28/10 | 6 | 37 | <0.4 | 6.1 | 6.2 | 8.0 | 174 | 401 | 109 |
| 4/11 | 7 | 40 | 0.4 | 3.6 | 6.8 | 7.9 | 108 | 214 | 119 |
| 11/11 | 3 | 45 | 0.9 | 8.4 | 7.6 | 7.9 | 123 | 246 | 110 |
| 18/11 | 2 | 45 | 0.7 | 2.0 | 6.4 | 7.8 | 100 | 204 | 126 |
| 25/11 | 10 | 37 | 0.8 | 1.0 | 7.3 | 7.9 | 95 | 177 | 127 |

- AGE: Sludge age (days)
 ALK: Alkalinity (CaCO₃ mg/L)
 AMM: Ammonia (N mg/L)
 CL: Chloride (Cl mg/L)
 COD: Chemical oxygen demand (mg COD/L)
 CON: Conductivity (mS/m)
 DSVI: Dissolved sludge volume index (ml/g)
 ERR: Result error
 Flow: Flow (ML/day) for entire plant
 ND: Not determined
 NN: Nitrate and nitrite (N mg/L)
 OP: ortho-phosphate (P mg/L)
 RASF: RAS flow rate (s) (kL/day)
 RASSS: RAS suspended solids (mg/L)
 SETS: Settleable solids (30 min ml/L)
 SS: Suspended solids (105°C mg/L)
 SVI: Sludge volume index (ml/g)
 SWR: Sludge wasting rate (kL/day)
 TKN: Total Kjeldahl nitrogen (N mg/L)
 TP: Total phosphorus (P mg/L)
 VSS: Volatile suspended solids (mg/L)

Figures in red are anomalies and not transcription errors from the laboratory printouts.

| ANNEXURE 4.1.7 RAW DATE FROM MITCHELLS PLAIN WWTP | | | | | | | | | | | |
|---------------------------------------------------|------|-----|------|------|-----|------|------|-----|-----|-----|-----|
| PST C-F (INFLUENT) | | | | | | | | | | | |
| Date: | Flow | SS | COD | TKN | AMM | TP | OP | PH | CL | ALK | CON |
| 4/6 | 40 | 254 | 577 | 93.1 | 81 | 13.0 | 10.7 | 7.2 | 78 | 357 | 109 |
| 11/6 | 29 | 148 | 542 | 72.8 | 54 | 11.2 | 8.8 | 7.1 | 71 | 263 | 90 |
| 18/6 | 34 | 262 | 768 | 101 | 85 | 16.2 | 13.1 | 7.3 | 92 | 363 | 114 |
| 25/6 | 34 | 168 | 1307 | 74.1 | 66 | 13.9 | 11.4 | 7.1 | 78 | 281 | 100 |
| 2/7 | 28 | 235 | 748 | 93.0 | 90 | 14.3 | 12.3 | 6.9 | 90 | 377 | 115 |
| 9/7 | ND | 268 | 892 | 105 | 83 | 17.5 | 14.7 | 7.0 | 89 | 371 | 122 |
| 17/7 | 34 | 244 | 867 | 99.3 | 92 | 12.5 | 11.0 | 6.8 | 91 | 394 | 118 |
| 23/7 | 34 | 243 | 870 | 99.2 | 72 | 16.6 | 13.5 | 6.8 | 100 | 337 | 121 |
| 30/7 | 39 | 235 | 707 | 98.4 | 88 | 14.3 | 11.5 | 7.1 | 94 | 399 | 134 |
| 6/8 | 40 | 254 | 778 | 76.5 | 49 | 12.4 | 9.8 | 6.6 | 85 | 305 | 100 |
| 13/8 | 42 | 220 | 791 | 101 | 78 | 16.9 | 13.5 | 6.9 | 84 | 378 | 122 |
| 20/8 | 37 | 373 | 824 | 102 | 74 | 12.5 | 10.1 | 7.0 | 94 | 373 | 120 |
| 27/8 | ND | 338 | 853 | 96.2 | 77 | 13.7 | 11.2 | 7.0 | 71 | 348 | 114 |
| 3/9 | 38 | 300 | 990 | 74.6 | 50 | 19.2 | 15.4 | 7.0 | 85 | 308 | 107 |
| 10/9 | ND | 272 | 871 | 77.6 | 55 | 12.6 | 9.7 | 6.8 | 83 | 293 | 104 |
| 17/9 | ND | 206 | 638 | ND | 84 | 11.7 | 10.6 | 7.1 | 81 | 366 | 110 |
| 24/9 | 34 | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| 1/10 | 34 | 175 | 602 | 74.6 | 70 | 12.2 | 10.3 | 7.3 | 86 | 318 | 119 |
| 8/10 | 33 | 216 | 679 | 86.9 | 72 | 11.2 | 9.6 | 7.3 | 87 | 326 | 107 |
| 15/10 | 34 | 140 | 604 | 94.0 | 78 | 13.5 | 12.0 | 7.3 | 91 | 351 | 113 |
| 22/10 | ND | 164 | 540 | 80.6 | 70 | 10.7 | 9.2 | 7.4 | 88 | 320 | 106 |
| 29/10 | ND | 184 | 662 | 101 | 83 | 14.2 | 12.2 | 7.2 | 88 | 357 | 116 |
| 5/11 | 31 | 267 | 669 | 79.9 | 58 | 10.8 | 9.2 | 6.9 | 75 | 287 | 95 |
| 12/11 | ND | 204 | 830 | 91.0 | 71 | 14.7 | 13.4 | 7.1 | 90 | 324 | 111 |
| 19/11 | ND | 240 | 873 | 80.9 | 63 | 18.0 | 15.5 | 6.8 | 84 | 308 | 106 |
| 26/11 | ND | 172 | 634 | 104 | 90 | 14.9 | 12.6 | 7.5 | 87 | 361 | 117 |

| PST GH (INFLUENT) | | | | | | | | | | | |
|-------------------|------|-----|------|------|-----|------|------|-----|-----|-----|-----|
| Date: | Flow | SS | COD | TKN | AMM | TP | OP | PH | CL | ALK | CON |
| 4/6 | 40 | 264 | 747 | 119 | 98 | 14.8 | 11.7 | 7.1 | 85 | 408 | 124 |
| 11/6 | 29 | 360 | 945 | 79.9 | 53 | 13.6 | 10.2 | 6.6 | 80 | 274 | 91 |
| 18/6 | 34 | 874 | 1684 | 91.6 | 61 | 18.1 | 12.8 | 6.7 | 87 | 299 | 97 |
| 25/6 | 34 | 474 | 1178 | 66.8 | 48 | 12.1 | 9.4 | 6.4 | 73 | 239 | 82 |
| 2/7 | 28 | 255 | 719 | 111 | 98 | 14.4 | 12.2 | 7.1 | 90 | 394 | 129 |
| 9/7 | ND | 384 | 1158 | 85.9 | 56 | 19.6 | 15.3 | 6.7 | 77 | 296 | 104 |
| 17/7 | 34 | 356 | 888 | 112 | 83 | 12.5 | 10.5 | 6.6 | 94 | 375 | 122 |
| 23/7 | 34 | 392 | 1199 | 103 | 63 | 16.7 | 12.7 | 6.5 | 108 | 311 | 115 |
| 30/7 | 39 | 227 | 662 | - | 89 | 14.6 | 12.0 | 7.2 | 94 | 411 | 136 |
| 6/8 | 40 | 338 | 876 | 72.0 | 46 | 11.5 | 8.9 | 6.4 | 84 | 283 | 95 |
| 13/8 | 42 | 192 | 727 | 105 | 83 | 16.1 | 13.3 | 7.0 | 86 | 402 | 126 |
| 20/8 | 37 | 416 | 874 | 120 | 92 | 13.8 | 11.3 | 7.1 | 88 | 406 | 131 |
| 27/8 | ND | 290 | 754 | 106 | 79 | 14.2 | 11.4 | 7.2 | 77 | 352 | 119 |
| 3/9 | 38 | 228 | 1025 | 71.6 | 49 | 18.9 | 14.7 | 6.9 | 81 | 290 | 105 |
| 10/9 | ND | 449 | 864 | 117 | 91 | 13.6 | 6.3 | 7.0 | 85 | 352 | 127 |
| 17/9 | ND | 645 | 1437 | 111 | 91 | 14.9 | 11.6 | 7.1 | 82 | 381 | 128 |
| 24/9 | 34 | ND- | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| 1/10 | 34 | 352 | 952 | 97.7 | 80 | 12.8 | 10.7 | 6.8 | 85 | 361 | 128 |
| 8/10 | 33 | 472 | 1002 | 107 | 80 | 12.7 | 10.2 | 6.9 | 85 | 355 | 113 |
| 15/10 | 34 | 534 | 1295 | 110 | 80 | 15.5 | 12.7 | 6.7 | 86 | 362 | 116 |
| 22/10 | ND | 688 | 1028 | 113 | 94 | 13.5 | 11.5 | 6.8 | 89 | 388 | 123 |
| 29/10 | ND | 416 | 1122 | 139 | 110 | 17.1 | 13.6 | 7.0 | 91 | 396 | 127 |
| 5/11 | 31 | 357 | 945 | 102 | 77 | 12.0 | 10.1 | 6.9 | 77 | 322 | 108 |
| 12/11 | ND | 500 | 1123 | 96.5 | 74 | 15.0 | 12.8 | 6.9 | 84 | 323 | 109 |
| 19/11 | ND | 368 | 1039 | 74.8 | 64 | 17.4 | 15.3 | 6.9 | 91 | 306 | 106 |
| 26/11 | ND | 705 | 1371 | 119 | 89 | 17.9 | 14.0 | 7.0 | 88 | 358 | 118 |

| MIXED LIQUOR REACTOR C | | | | | | | | |
|------------------------|------|------|------|-----|------|-----|-----|-----|
| Date | SETS | SS | VSS | SVI | DSVI | PH | SWR | AGE |
| 4/6 | 870 | 4070 | 3445 | 214 | 138 | 6.7 | 105 | 21 |
| 11/6 | 870 | 4335 | ND | 201 | 120 | 6.5 | 99 | 29 |
| 18/6 | 770 | 3780 | ND | 204 | 116 | 6.6 | 100 | 29 |
| 25/6 | 900 | 5490 | ND | 164 | 102 | 6.4 | 100 | 23 |
| 2/7 | 650 | 4100 | 3500 | 159 | 98 | 6.7 | 100 | 26 |
| 9/7 | 580 | 4280 | ND | 136 | 94 | 6.7 | ND | ND |
| 17/7 | 660 | 4890 | ND | 135 | 82 | 6.6 | ND | ND |
| 23/7 | 540 | 3720 | ND | 145 | 86 | 6.5 | ND | ND |
| 30/7 | 530 | 4250 | ND | 125 | 85 | 6.8 | ND | ND |
| 6/8 | 700 | 4710 | 3860 | 149 | 85 | 6.7 | ND | ND |
| 13/8 | 750 | 4520 | ND | 162 | 87 | 6.6 | ND | ND |
| 20/8 | 650 | 4900 | ND | 133 | 90 | 6.8 | ND | ND |
| 27/8 | 670 | 4910 | ND | 136 | 90 | 6.8 | ND | ND |
| 3/9 | 720 | 5900 | ND | 122 | 75 | 6.8 | 100 | 28 |
| 10/9 | 810 | 5590 | ND | 145 | 86 | 6.8 | ND | ND |
| 17/9 | 770 | 5150 | ND | 150 | 93 | 6.9 | ND | ND |
| 24/9 | 550 | 4900 | ND | 112 | 90 | 6.8 | 100 | ND |
| 1/10 | 430 | 3300 | 2830 | 130 | 97 | 6.9 | 100 | 26 |
| 8/10 | 420 | 4180 | ND | 100 | 96 | 6.9 | 100 | 28 |
| 15/10 | 700 | 4840 | ND | 145 | 83 | 6.9 | 100 | 28 |
| 22/10 | 650 | 4160 | ND | 156 | 87 | 6.8 | ND | ND |
| 29/10 | 1000 | 4210 | ND | 238 | 86 | 6.9 | ND | ND |
| 5/11 | 300 | 3570 | 3080 | 84 | 78 | 6.8 | ND | ND |
| 12/11 | 520 | 4910 | ND | 106 | 65 | 6.8 | ND | ND |
| 19/11 | 760 | 5780 | ND | 131 | 76 | 6.7 | ND | ND |
| 26/11 | 740 | 7570 | ND | 98 | 58 | 6.8 | ND | ND |

| MIXED LIQUOR REACTOR G | | | | | | | | |
|------------------------|------|-------|-------|-----|------|-----|-----|-----|
| Date | SETS | SS | VSS | SVI | DSVI | PH | SWR | AGE |
| 4/6 | 760 | 9920 | 8535 | 77 | 52 | 6.5 | 65 | 70 |
| 11/6 | 870 | 11395 | ND | 76 | 49 | 6.5 | 23 | ND |
| 18/6 | 870 | 11770 | ND | 74 | 51 | 6.5 | 200 | 30 |
| 25/6 | 820 | 8970 | ND | 91 | 67 | 6.4 | 200 | 30 |
| 2/7 | 910 | 12660 | 10960 | 72 | 54 | 6.7 | 200 | 30 |
| 9/7 | 890 | 12900 | ND | 69 | 46 | 6.6 | ND | ND |
| 17/7 | 960 | 13300 | ND | 72 | 51 | 6.4 | ND | ND |
| 23/7 | 750 | 9990 | ND | 75 | 52 | 6.5 | ND | ND |
| 30/7 | 690 | 9510 | ND | 73 | 50 | 6.7 | 400 | 15 |
| 6/8 | 520 | 8130 | 6690 | 64 | 54 | 6.6 | 400 | 15 |
| 13/8 | 680 | 9730 | ND | 70 | 45 | 6.6 | ND | ND |
| 20/8 | 610 | 9770 | ND | 62 | 45 | 6.8 | ND | ND |
| 27/8 | 490 | 8330 | ND | 59 | 48 | 6.8 | ND | ND |
| 3/9 | 640 | 7990 | ND | 80 | 50 | 6.9 | 300 | 20 |
| 10/9 | 650 | 9490 | ND | 69 | 46 | 6.8 | ND | ND |
| 17/9 | 570 | 9510 | ND | 60 | 50 | 6.8 | ND | ND |
| 24/9 | 740 | 11610 | ND | 64 | 45 | 6.7 | 200 | ND |
| 1/10 | 840 | 11590 | 9900 | 73 | 52 | 6.6 | 200 | 30 |
| 8/10 | 600 | 9370 | ND | 64 | 47 | 6.9 | 200 | 30 |
| 15/10 | 860 | 9790 | ND | 88 | 65 | 6.9 | 100 | 59 |
| 22/10 | 890 | 12930 | ND | 69 | 46 | 6.7 | ND | ND |
| 29/10 | 780 | 12950 | ND | 60 | 43 | 6.7 | ND | ND |
| 5/11 | 780 | 12380 | 10630 | 63 | 45 | 6.7 | ND | ND |
| 12/11 | 660 | 11040 | ND | 60 | 40 | 6.7 | ND | ND |
| 19/11 | 720 | 11920 | ND | 60 | 37 | 6.7 | ND | ND |
| 26/11 | 740 | 10510 | ND | 71 | 46 | 6.7 | ND | ND |

| CLARIFIER CD (EFFLUENT) | | | | | | | | | |
|-------------------------|----|-----|------|------|------|-----|----|-----|-----|
| Date: | SS | COD | AMM | NN | OP | PH | CL | Alk | CON |
| 4/6 | 23 | 51 | <0.4 | 8.4 | 11.7 | 6.7 | 82 | 82 | 62 |
| 11/6 | 7 | 23 | <0.4 | 8.1 | 12.0 | 6.4 | 76 | 55 | 55 |
| 18/6 | 3 | 30 | <0.4 | 16.7 | 9.0 | 6.6 | 88 | 48 | 63 |
| 25/6 | 29 | 55 | 6.6 | 6.9 | 4.6 | 6.6 | 79 | 93 | 60 |
| 2/7 | 15 | 41 | <0.4 | 9.6 | 6.6 | 6.7 | 87 | 69 | 66 |
| 9/7 | 7 | 37 | <0.4 | 9.1 | 0.5 | 6.8 | 86 | 85 | 64 |
| 17/7 | 9 | 33 | <0.4 | 6.6 | 0.1 | 6.8 | 91 | 87 | 68 |
| 23/7 | 8 | 30 | 0.4 | 11.7 | 2.9 | 6.6 | 94 | 64 | 72 |
| 30/7 | 9 | 44 | <0.4 | 8.5 | 1.0 | 6.8 | 90 | 99 | 71 |
| 6/8 | 10 | 43 | <0.4 | 8.6 | 2.3 | 6.5 | 93 | 107 | 72 |
| 13/8 | 6 | 36 | <0.4 | 7.0 | 1.5 | 6.7 | 84 | 109 | 67 |
| 20/8 | 7 | 40 | <0.4 | 6.7 | 3.7 | 7.1 | 88 | 102 | 71 |
| 27/8 | 9 | 37 | <0.4 | 7.4 | 6.1 | 6.9 | 73 | 88 | 65 |
| 3/9 | 8 | 22 | 1.0 | 6.5 | 4.1 | 7.1 | 76 | 109 | 67 |
| 10/9 | 11 | 51 | 0.2 | 3.1 | 1.6 | 7.0 | 80 | 108 | 64 |
| 17/9 | 9 | 48 | 0.6 | 5.8 | 5.8 | 7.1 | 81 | 100 | 67 |
| 24/9 | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| 1/10 | 10 | 30 | <0.4 | 11.5 | 9.7 | 6.9 | 84 | 72 | 75 |
| 8/10 | 8 | 44 | 0.4 | 12.1 | 9.1 | 6.9 | 93 | 71 | 67 |
| 15/10 | 8 | 25 | <0.4 | 13.5 | 9.9 | 7.0 | 85 | 61 | 65 |
| 22/10 | 13 | 37 | <0.4 | 7.9 | 6.2 | 6.9 | 84 | 82 | 64 |
| 29/10 | 6 | 30 | <0.4 | 9.4 | 10.3 | 7.1 | 82 | 79 | 64 |
| 5/11 | 8 | 34 | <0.4 | 8.4 | 7.0 | 7.2 | 85 | 92 | 65 |
| 12/11 | 12 | 49 | 0.5 | 8.9 | 11.8 | 7.0 | 86 | 85 | 66 |
| 19/11 | 8 | 37 | 3.2 | 6.1 | 8.2 | 6.8 | 85 | 113 | 68 |
| 26/11 | 6 | 30 | 0.8 | 11.0 | 9.9 | 7.0 | 81 | 81 | 64 |

| CLARIFIER GH (EFFLUENT) | | | | | | | | | |
|-------------------------|----|-----|------|------|------|-----|----|-----|-----|
| Date: | SS | COD | AMM | NN | OP | PH | CL | Alk | CON |
| 4/6 | 29 | 61 | 0.4 | 11.8 | 6.6 | 6.7 | 80 | 81 | 63 |
| 11/6 | 7 | 37 | 0.4 | 10.0 | 3.7 | 6.6 | 76 | 64 | 55 |
| 18/6 | 11 | 51 | 0.4 | 14.4 | 9.0 | 6.8 | 89 | 72 | 65 |
| 25/6 | 12 | 50 | <0.4 | 14.5 | 4.9 | 6.6 | 84 | 80 | 64 |
| 2/7 | 20 | 55 | <0.4 | 10.9 | 5.8 | 6.7 | 86 | 76 | 66 |
| 9/7 | 45 | 104 | 0.6 | 10.0 | 3.1 | 6.9 | 85 | 98 | 67 |
| 17/7 | 14 | 44 | 0.3 | 7.6 | 6.5 | 6.8 | 91 | 82 | 69 |
| 23/7 | 34 | 62 | <0.4 | 14.5 | 8.9 | 6.5 | 96 | 59 | 74 |
| 30/7 | 9 | 37 | <0.4 | 13.0 | 5.5 | 6.8 | 90 | 76 | 72 |
| 6/8 | 12 | 47 | <0.4 | 15.5 | 6.1 | 6.5 | 93 | 83 | 74 |
| 13/8 | 2 | 47 | <0.4 | 13.1 | 4.5 | 6.7 | 86 | 83 | 71 |
| 20/8 | 8 | 51 | 0.4 | 17.0 | 7.4 | 7.0 | 87 | 70 | 75 |
| 27/8 | 6 | 44 | <0.4 | 16.6 | 6.9 | 6.9 | 75 | 50 | 67 |
| 3/9 | 7 | 40 | <0.4 | 20.7 | 8.7 | 6.9 | 77 | 61 | 71 |
| 10/9 | 11 | 58 | <0.4 | 12.7 | 4.4 | 7.0 | 78 | 61 | 69 |
| 17/9 | 15 | 51 | <0.4 | 13.4 | 3.4 | 7.1 | 82 | 78 | 70 |
| 24/9 | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| 1/10 | 14 | 44 | <0.4 | 14.4 | 2.8 | 7.0 | 83 | 85 | 77 |
| 8/10 | 8 | 48 | <0.4 | 18.9 | 6.9 | 7.2 | 90 | 59 | 70 |
| 15/10 | 7 | 46 | 29 | 0.1 | 4.8 | 7.5 | 84 | 224 | 86 |
| 22/10 | 22 | 48 | 0.8 | 3.0 | 5.8 | 7.1 | 84 | 122 | 67 |
| 29/10 | 26 | 52 | 0.4 | 15.2 | 9.0 | 7.0 | 83 | 75 | 67 |
| 5/11 | 15 | 45 | 0.4 | 14.5 | 12.1 | 7.1 | 84 | 72 | 68 |
| 12/11 | 30 | 67 | 0.5 | 15.1 | 13.1 | 6.8 | 86 | 69 | 71 |
| 19/11 | 14 | 52 | 0.4 | 23.0 | 12.0 | 6.7 | 87 | 56 | 70 |
| 26/11 | 22 | 65 | 0.4 | 13.9 | 9.4 | 7.0 | 81 | 88 | 66 |

AGE: Sludge age (days)
ALK: Alkalinity (CaCO₃ mg/L)
AMM: Ammonia (N mg/L)
CL: Chloride (Cl mg/L)
COD: Chemical oxygen demand (mg COD/L)
CON: Conductivity (mS/m)
DSVI: Dissolved sludge volume index (ml/g)
Flow: Flow (ML/day) for entire plant
ND: Not determined
NN: Nitrate and nitrite (N mg/L)
OP: ortho-phosphate (P mg/L)
SETS: Settleable solids (30 min ml/L)
SS: Suspended solids (105°C mg/L)
SVI: Sludge volume index (ml/g)
SWR: Sludge wasting rate (kL/day)
TKN: Total Kjeldahl nitrogen (N mg/L)
TP: Total phosphorus (P mg/L)
VSS: Volatile suspended solids (mg/L)

Figures in red are anomalies and not transcription errors from the laboratory printouts.

ANNEXURE 4.1.8 RAW DATA FROM PAROW WWTP

RAW INFLUENT (NO PST)

| Date: | Flow | SS | SETS | COD | TKN | AMM | TP | OP | PH | CON | CL | ALK |
|-------|------|-----|------|------|------|-----|------|-----|-----|-----|-----|-----|
| 6/6 | 1.0 | 336 | 14.0 | 754 | 65.9 | 58 | 16.1 | 9.2 | 7.5 | 75 | 134 | 271 |
| 13/6 | 1.0 | 453 | 27.0 | 793 | 83.2 | 73 | 16.3 | 7.8 | 8.2 | 120 | 147 | 309 |
| 20/6 | 1.0 | 708 | 27.0 | 1180 | 118 | 70 | 18.6 | 7.9 | 7.7 | 120 | 151 | 286 |
| 27/6 | ND | 360 | 22.0 | 778 | 87.8 | 59 | 16.1 | 7.3 | 8.2 | 121 | 149 | 274 |
| 4/7 | ND | 353 | 17.0 | 688 | 81.7 | 58 | 12.9 | 6.8 | 8.2 | 132 | 177 | 274 |
| 11/7 | ND | 400 | 22.0 | 704 | 84.2 | 61 | 13.0 | 6.8 | 7.9 | 143 | 209 | 285 |
| 18/7 | ND | 384 | 20.0 | 707 | 95.4 | 70 | 12.4 | 7.1 | 8.2 | 118 | 140 | 275 |
| 25/7 | 1.1 | 473 | 21.0 | 855 | ND | 67 | ND | 7.2 | 8.2 | 132 | 169 | 287 |
| 1/8 | 1.2 | 408 | 27.0 | 699 | 80.7 | 62 | 12.1 | 6.8 | 8.1 | 136 | 178 | 285 |
| 8/8 | ND | 324 | 15.0 | 695 | ND | 29 | ND | 5.8 | 7.5 | 107 | 164 | 216 |
| 15/8 | ND | 252 | 18.0 | 557 | 75.1 | 47 | 10.1 | 6.0 | 7.9 | 123 | 164 | 274 |
| 22/8 | ND | 423 | 23.0 | 760 | 96.4 | 69 | 12.4 | 7.2 | 8.4 | 129 | 164 | 309 |
| 29/8 | 1.1 | 484 | 20.0 | 669 | 76.8 | 57 | 12.4 | 6.5 | 8.3 | 129 | 183 | 281 |
| 5/9 | 1.1 | 360 | 22.0 | 698 | 71.2 | 50 | 13.4 | 6.3 | 7.8 | 115 | 151 | 259 |
| 12/9 | 1.2 | 297 | 22.0 | 584 | 54.3 | 39 | 11.9 | 5.7 | 8.0 | 115 | 158 | 238 |
| 26/9 | ND | 452 | 22.0 | 838 | 95.6 | 69 | 12.7 | 7.5 | 8.1 | 140 | 204 | 299 |
| 3/10 | 1.1 | 456 | 22.0 | 893 | 96.2 | 64 | 13.7 | 7.3 | 8.3 | 120 | 164 | 281 |
| 10/10 | 1.0 | 440 | 20.0 | 872 | 104 | 74 | ND | 8.3 | 8.4 | 123 | 154 | 309 |
| 17/10 | 1.0 | 296 | 20.0 | 802 | 83.2 | 56 | 15.0 | 8.0 | 8.2 | 107 | 132 | 254 |
| 24/10 | 1.0 | 380 | 18.0 | 679 | 86.5 | 66 | 13.5 | 7.3 | 8.2 | 116 | 152 | 262 |
| 1/11 | 1.0 | 620 | 29.0 | 1008 | 97.6 | 69 | 17.1 | 9.5 | 7.7 | 118 | 137 | 335 |
| 8/11 | 1.0 | 504 | 24.0 | 940 | 112 | 82 | 14.8 | 9.0 | 8.3 | 123 | 149 | 311 |
| 14/11 | 1.0 | 523 | 23.0 | 862 | 102 | 73 | 14.6 | 8.3 | 7.9 | 114 | 134 | 294 |
| 21/11 | 1.1 | 925 | 24.0 | 1969 | 125 | 66 | 21.3 | 7.7 | 8.2 | 117 | 152 | 268 |
| 28/11 | 1.0 | 503 | 25.0 | 938 | 96.2 | 73 | 15.2 | 9.1 | 8.1 | 117 | 149 | 305 |

MIXED LIQUOR

| Date | SETS | SS | VSS | SVI | DSVI | PH | RASSS |
|-------|------|------|------|-----|------|-----|-------|
| 6/6 | 970 | 4510 | 3885 | 215 | 151 | 7.0 | 6560 |
| 13/6 | 1000 | 5740 | ND | 174 | 160 | 7.1 | 7790 |
| 20/6 | 990 | 5620 | ND | 176 | ND | 7.0 | 11310 |
| 27/6 | 1000 | 6420 | ND | 156 | 156 | 6.7 | 7180 |
| 4/7 | 1000 | 5620 | 4570 | 178 | 164 | 6.5 | 7500 |
| 11/7 | 970 | 4850 | ND | 200 | 173 | 6.7 | 5620 |
| 18/7 | 980 | 4650 | ND | 211 | 163 | 6.3 | 5860 |
| 25/7 | 920 | 4300 | ND | 214 | 149 | 6.4 | ND |
| 1/8 | 920 | 3870 | ND | 238 | 165 | 6.7 | 5330 |
| 8/8 | 960 | 3410 | ND | 282 | 223 | 6.8 | ND |
| 15/8 | 970 | 4230 | ND | 229 | 189 | 6.6 | 4600 |
| 22/8 | 980 | 5050 | ND | 194 | 198 | 6.9 | 7400 |
| 29/8 | 990 | 5920 | ND | 167 | 135 | 7.0 | 6200 |
| 5/9 | 1000 | 5780 | 4835 | 173 | 104 | 7.0 | 9280 |
| 12/9 | 1000 | 5690 | ND | 176 | 225 | 7.1 | 8710 |
| 26/9 | 980 | 4630 | ND | 212 | 216 | 7.1 | 6570 |
| 3/10 | 1000 | 5190 | 4250 | 193 | 247 | 7.1 | 5930 |
| 10/10 | 1000 | 5290 | ND | 189 | ND | 6.9 | 7680 |
| 17/10 | 1000 | 4560 | ND | 219 | 228 | 7.0 | 7280 |
| 24/10 | 1000 | 4740 | ND | 211 | 253 | 6.9 | 6060 |
| 1/11 | 1000 | 4500 | ND | 222 | 222 | 6.8 | 6980 |
| 8/11 | 1000 | 5420 | 4530 | 185 | 236 | 7.0 | 6180 |
| 14/11 | 1000 | 5200 | ND | 192 | 292 | 6.9 | 6640 |
| 21/11 | 1000 | 5555 | ND | 180 | 360 | 6.9 | 5635 |
| 28/11 | 1000 | 4820 | ND | 208 | 332 | 6.9 | 5400 |

| CLARIFIER (EFFLUENT) | | | | | | | | | | |
|----------------------|----|-----|------|------|------|-----|-----|-----|-----|--|
| Date: | SS | COD | AMM | NN | OP | PH | CON | CL | ALK | |
| 6/6 | 6 | 40 | 1.5 | 7.0 | 6.6 | 7.0 | 99 | 204 | 84 | |
| 13/6 | 5 | 33 | 15 | <0.1 | 7.1 | 7.5 | 123 | 240 | 179 | |
| 20/6 | 1 | 48 | 12 | 0.7 | 0.5 | 7.4 | 112 | 219 | 171 | |
| 27/6 | 2 | 58 | 0.4 | 1.9 | 1.8 | 7.2 | 110 | 229 | 112 | |
| 4/7 | 4 | 45 | <0.4 | 17.6 | 8.3 | 7.1 | 126 | 238 | 66 | |
| 11/7 | 5 | 29 | <0.4 | 23.3 | 6.9 | 6.9 | 133 | 270 | 50 | |
| 18/7 | 3 | 34 | <0.4 | 20.6 | 6.7 | 6.8 | 118 | 240 | 60 | |
| 25/7 | ND | ND | 0.4 | 16.3 | 7.4 | 7.1 | 102 | 194 | 52 | |
| 1/8 | 8 | 55 | <0.4 | 16.6 | 5.9 | 7.0 | 139 | 276 | 90 | |
| 8/8 | ND | ND | ND | ND | ND | ND | ND | ND | ND | |
| 15/8 | 5 | 44 | 0.6 | 21.5 | 6.4 | 6.6 | 125 | 239 | 70 | |
| 22/8 | 5 | 62 | <0.4 | 19.6 | 6.1 | 7.1 | 129 | 262 | 80 | |
| 29/8 | 3 | 55 | 7.0 | 0.8 | 1.1 | 7.4 | 132 | 267 | 180 | |
| 5/9 | 4 | 33 | 9.2 | 0.3 | 0.9 | 7.5 | 130 | 254 | 183 | |
| 12/9 | 6 | 40 | 17 | 0.1 | 0.7 | 7.6 | 130 | 246 | 196 | |
| 26/9 | 6 | 48 | 17 | 0.5 | 2.7 | 7.5 | 124 | 234 | 184 | |
| 3/10 | 5 | 40 | 5.0 | 2.5 | 0.4 | 7.4 | 105 | 214 | 134 | |
| 10/10 | 2 | 41 | 14 | 0.1 | 4.2 | 7.4 | 114 | 219 | 174 | |
| 17/10 | 4 | 33 | 11 | 1.5 | 1.9 | 7.4 | 111 | 210 | 152 | |
| 24/10 | 4 | 45 | 5.2 | 1.6 | 7.3 | 7.3 | 104 | 201 | 126 | |
| 1/11 | 6 | 55 | 23 | 0.1 | 4.5 | 7.6 | 116 | 200 | 191 | |
| 8/11 | 2 | 57 | 12 | 0.4 | 0.8 | 7.5 | 107 | 206 | 162 | |
| 14/11 | 2 | 30 | 0.5 | 3.3 | 8.2 | 7.4 | 98 | 202 | 109 | |
| 21/11 | 3 | 41 | <0.4 | 5.9 | 10.1 | 7.3 | 92 | 183 | 95 | |
| 28/11 | 6 | 44 | <0.4 | 9.4 | 10.1 | 7.2 | 97 | 199 | 88 | |

- AGE: Sludge age (days)
 ALK: Alkalinity (CaCO₃ mg/L)
 AMM: Ammonia (N mg/L)
 CL: Chloride (Cl⁻ mg/L)
 COD: Chemical oxygen demand (mg COD/L)
 CON: Conductivity (mS/m)
 DSVI: Dissolved sludge volume index (ml/g)
 Flow: Flow (ML/day) for entire plant
 ND: Not determined
 NN: Nitrate and nitrite (N mg/L)
 OP: ortho-phosphate (P mg/L)
 RASSS: RAS suspended solids (mg/L)
 SETS: Settleable solids (30 min ml/L)
 SS: Suspended solids (105°C mg/L)
 SVI: Sludge volume index (ml/g)
 TKN: Total Kjeldahl nitrogen (N mg/L)
 TP: Total phosphorus (P mg/L)
 VSS: Volatile suspended solids (mg/L)

Figures in red are anomalies and not transcription errors from the laboratory printouts.

ANNEXURE 1.9 RAW DATE FROM POTSDAM WWTP

PST A (INFLUENT)

| Date: | RAW FLOW | PSTA FLOW | SS | COD | TKN | AMM | TP | OP | PH | CL | ALK | CON |
|-------|----------|-----------|-----|-----|------|-----|------|------|-----|-----|-----|-----|
| 5/6 | 36.9 | 17.2 | 192 | 676 | 60.8 | 40 | 11.8 | 8.5 | 6.6 | 161 | 288 | 124 |
| 12/6 | 37.5 | 16.8 | 156 | 610 | 50.7 | 38 | 11.2 | 8.4 | 6.7 | 168 | 272 | 121 |
| 19/6 | 36.8 | 16.4 | 164 | 599 | 57.7 | 40 | 10.8 | 8.5 | 6.8 | 166 | 291 | 120 |
| 26/6 | 34.3 | 21.9 | 152 | 475 | 47.3 | 31 | 9.2 | 6.4 | 6.7 | 135 | 215 | 99 |
| 3/7 | 36.5 | 17.9 | 121 | 579 | 53.5 | 38 | 10.0 | 8.2 | 7.1 | 171 | 285 | 125 |
| 10/7 | 34.1 | 19.4 | 142 | 588 | 56.6 | 41 | 10.4 | 8.6 | 6.7 | 192 | 275 | 135 |
| 17/7 | 32.5 | 19.3 | 162 | 616 | 61.9 | 43 | 10.9 | 8.6 | 6.7 | 184 | 262 | 132 |
| 24/7 | 32.7 | 19.0 | 122 | 540 | 55.3 | 35 | 8.8 | 6.3 | 6.8 | 163 | 271 | 119 |
| 31/7 | 36.4 | 21.0 | 126 | 519 | 45.6 | 32 | 10.3 | 8.1 | 6.7 | 198 | 258 | 132 |
| 7/8 | 33.7 | 20.1 | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| 14/8 | 33.6 | 20.4 | 80 | 460 | 45.8 | 29 | 7.9 | 5.1 | 6.8 | 184 | 235 | 121 |
| 21/8 | 33.2 | 19.4 | 120 | 545 | 44.9 | 31 | 8.1 | 6.4 | 6.8 | 177 | 228 | 124 |
| 28/8 | 37.4 | 22.6 | 132 | 577 | 51.1 | 33 | 10.4 | 8.0 | 6.9 | 182 | 278 | 136 |
| 4/9 | 35.3 | 19.8 | 172 | 692 | 57.2 | 39 | 10.5 | 8.3 | 7.0 | 177 | 294 | 132 |
| 11/9 | ND | ND | 174 | 612 | 58.2 | 39 | 11.3 | 9.1 | 6.9 | 205 | 267 | 140 |
| 18/9 | 40.0 | 16.6 | 128 | 609 | 50.0 | 38 | 10.4 | 8.2 | 6.9 | 187 | 255 | 133 |
| 25/9 | 30.0 | 15.6 | 140 | 531 | 59.7 | 43 | 9.9 | 8.3 | 7.0 | 170 | 280 | 128 |
| 2/10 | 30.6 | 17.7 | 114 | 536 | 51.9 | 43 | 10.3 | 9.5 | 6.8 | 170 | 285 | 122 |
| 9/10 | 31.3 | 18.0 | 140 | 677 | 65.2 | 44 | 10.8 | 8.4 | 6.9 | 162 | 292 | 104 |
| 16/10 | 36.5 | 18.0 | 150 | 700 | 63.2 | 45 | 12.2 | 9.5 | 7.0 | 187 | 296 | 130 |
| 23/10 | 32.5 | 16.5 | 172 | 556 | 56.2 | 44 | 8.8 | 6.9 | 6.9 | 182 | 264 | 124 |
| 30/10 | 35.7 | 14.4 | 142 | 604 | 60.0 | 44 | 11.6 | 9.4 | 7.0 | 163 | 309 | 122 |
| 6/11 | 37.3 | 12.8 | 154 | 646 | 60.5 | 45 | 11.3 | 9.5 | 7.0 | 163 | 275 | 123 |
| 13/11 | ND | ND | 186 | 823 | 61.3 | 47 | 15.1 | 13.1 | 6.7 | 156 | 306 | 121 |
| 20/11 | ND | ND | 226 | 710 | 61.7 | 46 | 12.6 | 10.6 | 6.9 | 161 | 283 | 118 |
| 27/11 | 31.4 | 16.2 | 152 | 621 | ND | 47 | ND | 9.8 | 6.9 | 157 | 286 | 120 |

MIXED LIQUOR COMBINED E AND W REACTORS

| Date | SETS | SS | VSS | SVI | DSVI | PH | SWR | RASF | AGE | RASSS |
|-------|------|------|------|-----|------|-----|------|------|-----|-------|
| 5/6 | 980 | 6180 | 5005 | 159 | 136 | 6.6 | 994 | ND | 8 | 9410 |
| 12/6 | 980 | 6900 | ND | 142 | 139 | 6.7 | ND | ND | ND | 7630 |
| 19/6 | 990 | 6830 | ND | 145 | 146 | 6.8 | ND | ND | ND | 10210 |
| 26/6 | 1000 | 6840 | ND | 146 | 146 | 6.6 | ND | ND | ND | 10280 |
| 3/7 | 1000 | 7960 | 6270 | 126 | 75 | 6.8 | ND | ND | ND | 10900 |
| 10/7 | 1000 | 7980 | ND | 125 | 160 | 6.8 | 1126 | ND | 8 | 11210 |
| 17/7 | 1000 | 7070 | ND | 141 | 141 | 6.6 | 1061 | ND | 8 | 10330 |
| 24/7 | 1000 | 7210 | ND | 139 | ND | 6.6 | 991 | ND | 9 | 10850 |
| 31/7 | 1000 | 6800 | ND | 147 | 153 | 6.9 | 1148 | ND | 8 | 10590 |
| 7/8 | 970 | 6490 | ND | 150 | 150 | 6.9 | 1262 | ND | ND | ND |
| 14/8 | 1000 | 6660 | ND | 150 | 150 | 6.6 | 676 | ND | 11 | 11170 |
| 21/8 | 1000 | 6410 | ND | 156 | 156 | 6.7 | 1219 | ND | 7 | 10510 |
| 28/8 | 1000 | 6960 | ND | 144 | ND | 6.9 | 855 | ND | 9 | 10230 |
| 4/9 | 1000 | 6790 | 5510 | 147 | ND | 7.0 | 1547 | ND | 6 | 10340 |
| 11/9 | 1000 | 6380 | ND | 157 | 144 | 6.9 | ND | ND | ND | 8990 |
| 18/9 | 970 | 6660 | ND | 146 | 138 | 6.7 | 1437 | ND | 6 | 10630 |
| 25/9 | 970 | 7600 | ND | 128 | 116 | 6.7 | 1081 | ND | 8 | 9660 |
| 2/10 | 990 | 8050 | 6500 | 123 | 109 | 6.5 | 1186 | ND | 8 | 10190 |
| 9/10 | 980 | 7930 | ND | 124 | 106 | 6.8 | 1596 | ND | 6 | 12630 |
| 16/10 | 990 | 8390 | ND | 118 | 105 | 6.8 | 1556 | ND | 6 | 14990 |
| 23/10 | 970 | 8390 | ND | 116 | 105 | 6.8 | 638 | ND | 13 | 13430 |
| 30/10 | 1000 | 9860 | ND | 101 | 101 | 6.8 | 656 | ND | 13 | 12250 |
| 6/11 | 980 | 9040 | 7100 | 108 | 93 | 6.9 | 1483 | ND | 6 | 12240 |
| 13/11 | 970 | 8610 | ND | 113 | 102 | 6.9 | ND | ND | ND | 12840 |
| 20/11 | 970 | 8615 | ND | 113 | 98 | 6.8 | ND | ND | ND | 11315 |
| 27/11 | 960 | 8680 | ND | 111 | 101 | 6.8 | 1515 | ND | 6 | 11220 |

| CLARIFIER A (EFFLUENT) | | | | | | | | | | |
|------------------------|----|-----|------|-----|-----|-----|-----|-----|-----|--|
| Date: | SS | COD | AMM | NN | OP | PH | CL | Alk | CON | |
| 5/6 | 25 | 69 | <0.4 | 6.0 | 0.2 | 7.3 | 143 | 143 | 88 | |
| 12/6 | 15 | 44 | <0.4 | 2.7 | 0.2 | 7.3 | 140 | 119 | 85 | |
| 19/6 | 23 | 52 | <0.4 | 3.5 | 0.3 | 7.2 | 158 | 153 | 91 | |
| 26/6 | 19 | 55 | <0.4 | 2.4 | 0.4 | 7.1 | 147 | 128 | 88 | |
| 3/7 | 9 | 41 | <0.4 | 3.0 | 0.4 | 7.5 | 168 | 159 | 100 | |
| 10/7 | 37 | 92 | <0.4 | 1.2 | 1.5 | 7.2 | 167 | 181 | 107 | |
| 17/7 | 15 | 76 | 0.5 | 4.6 | 0.5 | 7.2 | 163 | 121 | 102 | |
| 24/7 | 42 | 76 | 0.5 | 2.9 | 1.4 | 7.3 | 156 | 129 | 91 | |
| 31/7 | 34 | 180 | <0.4 | 3.6 | 0.6 | 7.1 | 182 | 148 | 106 | |
| 7/8 | ND | ND | ND | ND | ND | ND | ND | ND | ND | |
| 14/8 | 16 | 55 | 0.6 | 3.6 | 1.2 | 7.2 | 186 | 125 | 106 | |
| 21/8 | 12 | 55 | <0.4 | 4.1 | 1.2 | 7.9 | 170 | 115 | 104 | |
| 28/8 | 12 | 54 | <0.4 | 2.7 | 0.8 | 7.8 | 171 | 154 | 106 | |
| 4/9 | 22 | 55 | <0.4 | 3.3 | 0.4 | 7.9 | 183 | 158 | 106 | |
| 11/9 | 59 | 123 | <0.4 | 6.0 | 0.2 | 7.7 | 177 | 110 | 106 | |
| 18/9 | 30 | 97 | <0.4 | 4.6 | 0.5 | 7.9 | 171 | 122 | 101 | |
| 25/9 | 10 | 62 | <0.4 | 6.4 | 0.7 | 7.8 | 167 | 123 | 97 | |
| 2/10 | 11 | 51 | <0.4 | 2.9 | 1.2 | 7.4 | 170 | 149 | 96 | |
| 9/10 | 5 | 44 | 1.0 | 2.6 | 3.5 | 7.9 | 153 | 160 | 95 | |
| 16/10 | 7 | 40 | <0.4 | 2.0 | 0.4 | 7.9 | 170 | 151 | 98 | |
| 23/10 | 10 | 69 | 1.4 | 2.0 | 1.3 | 7.8 | 182 | 139 | 99 | |
| 30/10 | 6 | 51 | <0.4 | 1.3 | 1.0 | 7.9 | 167 | 139 | 96 | |
| 6/11 | 8 | 41 | <0.4 | 3.7 | 0.4 | 7.9 | 159 | 145 | 93 | |
| 13/11 | 6 | 42 | <0.4 | 1.7 | 4.6 | 7.9 | 160 | 161 | 93 | |
| 20/11 | 10 | 66 | 0.4 | 3.1 | 3.9 | 8.1 | 156 | 142 | 92 | |
| 27/11 | 9 | 55 | 0.6 | 1.2 | 5.4 | 8.2 | 158 | 157 | 92 | |

AGE: Sludge age (days)
 ALK: Alkalinity (CaCO₃ mg/L)
 AMM: Ammonia (N mg/L)
 CL: Chloride (Cl⁻ mg/L)
 COD: Chemical oxygen demand (mg COD/L)
 CON: Conductivity (mS/m)
 DSVI: Dissolved sludge volume index (ml/g)
 Flow: Flow (ML/day) for entire plant
 ND: Not determined
 NN: Nitrate and nitrite (N mg/L)
 OP: ortho-phosphate (P mg/L)
 RASF: RAS flow rate (s) (kL/day)
 RASSS: RAS suspended solids (mg/L)
 SETS: Settleable solids (30 min ml/L)
 SS: Suspended solids (105°C mg/L)
 SVI: Sludge volume index (ml/g)
 SWR: Sludge wasting rate (kL/day)
 TKN: Total Kjeldahl nitrogen (N mg/L)
 TP: Total phosphorus (P mg/L)
 VSS: Volatile suspended solids (mg/L)

Figures in red are anomalies and not transcription errors from the laboratory printouts.

ANNEXURE 1.10 RAW DATE FROM WESFLEUR INDUSTRIAL WWTP

RAW INFLOW (NO PST)

| Date: | Flow | SS | VSS | SETS | COD | TKN | AMM | TP | OP | PH | CON | CL | ALK |
|-------|------|-----|-----|------|------|------|-----|------|-----|-----|-----|-----|-----|
| 5/6 | 4.3 | 206 | ND | 6.0 | 814 | 48.6 | 26 | 8.5 | 5.7 | 7.2 | 171 | 307 | 279 |
| 12/6 | 3.9 | 627 | ND | 8.0 | 1417 | 73.5 | 27 | 10.5 | 4.3 | 7.7 | 243 | 295 | 353 |
| 19/6 | 4.2 | 284 | ND | 12.0 | 1062 | 59.9 | 34 | 10.9 | 6.8 | 7.3 | 170 | 306 | 277 |
| 26/6 | 4.0 | 366 | ND | 12.0 | 1013 | 53.5 | 32 | 6.3 | 3.9 | 7.4 | 187 | 313 | 233 |
| 3/7 | 4.1 | 322 | ND | 8.0 | 981 | 51.9 | 30 | 10.0 | 6.5 | 7.5 | 194 | 402 | 280 |
| 10/7 | 4.1 | 356 | ND | 13.0 | 849 | 52.8 | 29 | 10.5 | 5.9 | 7.4 | 198 | 354 | 265 |
| 17/7 | 6.0 | 248 | ND | ND | 767 | 57.5 | 42 | 8.7 | 7.1 | 7.0 | 168 | 270 | 244 |
| 24/7 | ND | 324 | ND | 9.0 | 940 | 54.2 | 29 | 7.5 | 4.5 | 7.2 | 165 | 125 | 249 |
| 31/7 | 4.3 | 334 | ND | 12.0 | 1037 | 46.5 | 28 | 8.9 | 5.4 | 7.5 | 178 | 299 | 267 |
| 7/8 | 4.2 | 303 | ND | 10.0 | 952 | ND | 28 | ND | 4.9 | 7.4 | 193 | 355 | 265 |
| 14/8 | 3.9 | 192 | ND | 5.0 | 1070 | 44.8 | 30 | 6.7 | 4.4 | 7.5 | 213 | 264 | 516 |
| 21/8 | 4.3 | 273 | ND | 4.0 | 1138 | 54.2 | 24 | 9.3 | 5.0 | 7.7 | 267 | 324 | 331 |
| 28/8 | 4.4 | 216 | ND | 6.0 | 811 | 48.7 | 30 | 8.3 | 6.2 | 7.5 | 181 | 279 | 248 |
| 4/9 | 4.2 | 294 | 290 | 12.0 | 1002 | 56.3 | 29 | 8.9 | 5.5 | 7.2 | 179 | 317 | 267 |
| 11/9 | 4.0 | 284 | ND | 2.0 | 984 | 56.9 | 28 | 8.5 | 4.1 | 7.9 | 236 | 401 | 310 |
| 18/9 | 4.3 | 230 | ND | 5.0 | 733 | 44.3 | 26 | 8.0 | 5.8 | 7.4 | 212 | 397 | 290 |
| 25/9 | 3.7 | 148 | ND | 2.0 | 990 | 48.8 | 36 | 7.1 | 5.8 | 7.1 | 212 | 304 | 439 |
| 2/10 | 3.8 | 204 | 188 | 3.0 | 885 | 42.2 | 26 | 7.8 | 5.5 | 7.0 | 195 | 371 | 279 |
| 9/10 | 3.9 | 308 | ND | 10.0 | 1074 | 51.2 | 30 | 9.2 | 6.2 | 7.2 | 191 | 344 | 277 |
| 16/10 | 4.2 | 442 | ND | 12.0 | 949 | 76.2 | 47 | 9.1 | 5.8 | 7.5 | 192 | 313 | 260 |
| 23/10 | 4.1 | 500 | ND | 15.0 | 1368 | 70.9 | 46 | 9.5 | 5.4 | 7.3 | 161 | 254 | 247 |
| 30/10 | 4.2 | 576 | ND | 18.0 | 1438 | 59.5 | 27 | 12.2 | 7.1 | 7.1 | 154 | 264 | 298 |
| 6/11 | 4.1 | 664 | 556 | 25.0 | 1539 | 69.1 | 33 | 12.2 | 7.2 | 7.2 | 185 | 325 | 270 |
| 13/11 | 4.2 | 384 | ND | 7.0 | 896 | 46.4 | 30 | 8.1 | 5.3 | 7.1 | 196 | 406 | 343 |
| 20/11 | 4.1 | 178 | ND | 2.0 | 691 | 42.4 | 35 | 7.3 | 6.7 | 7.2 | 178 | 319 | 233 |
| 27/11 | 4.2 | 420 | ND | 12.0 | 1129 | 89.8 | 65 | 11.0 | 6.9 | 7.4 | 160 | 400 | 212 |

MIXED LIQUOR INDUSTRIAL BIOREACTOR

| Date | SETS | SS | VSS | SVI | DSVI | PH | SWR | AGE | RASS |
|-------|------|------|------|-----|------|-----|-----|-----|-------|
| 5/6 | 960 | 7080 | 6115 | 136 | 107 | 7.1 | 91 | 59 | 11960 |
| 12/6 | 960 | 6410 | ND | 150 | 112 | 7.1 | 179 | 32 | 14510 |
| 19/6 | 900 | 6360 | ND | 142 | 88 | 7.0 | 241 | 25 | 10970 |
| 26/6 | 890 | 5780 | ND | 154 | 90 | 7.0 | 102 | 52 | 12310 |
| 3/7 | 880 | 6060 | 5190 | 143 | 91 | 7.1 | 127 | 46 | 10100 |
| 10/7 | 920 | 6730 | ND | 137 | 95 | 7.2 | 98 | 58 | 9730 |
| 17/7 | 890 | 6860 | ND | 130 | 76 | 6.5 | 139 | 38 | 12620 |
| 24/7 | 880 | 6860 | ND | 128 | 93 | 6.7 | ND | ND | 10920 |
| 31/7 | 850 | 6950 | ND | 122 | 104 | 7.0 | 132 | 45 | 10400 |
| 7/8 | 940 | 6770 | ND | 139 | 100 | 6.9 | 87 | ND | ND |
| 14/8 | 920 | 7270 | ND | 127 | 77 | 6.9 | 113 | 52 | 12240 |
| 21/8 | 940 | 7020 | ND | 134 | 80 | 6.9 | 122 | 46 | 14110 |
| 28/8 | 900 | 7050 | ND | 128 | 85 | 7.0 | 112 | 49 | 10930 |
| 4/9 | 860 | 6100 | 5600 | 141 | 85 | 7.2 | 217 | 28 | 10340 |
| 11/9 | 860 | 5850 | ND | 147 | 89 | 7.1 | 175 | 34 | 10170 |
| 18/9 | 780 | 6010 | ND | 130 | 73 | 7.0 | 134 | 43 | 10940 |
| 25/9 | 470 | 6440 | ND | 73 | 50 | 6.9 | 165 | 34 | 8450 |
| 2/10 | 640 | 6100 | 5240 | 105 | 59 | 7.1 | 119 | 47 | 9860 |
| 9/10 | 670 | 6850 | ND | 98 | 58 | 7.0 | 81 | 68 | 10790 |
| 16/10 | 600 | 6090 | ND | 99 | 59 | 6.9 | 266 | 23 | 8810 |
| 23/10 | 490 | 5440 | ND | 90 | 59 | 7.1 | 279 | 22 | 6540 |
| 30/10 | 430 | 5400 | ND | 80 | 52 | 6.9 | 144 | 42 | 8480 |
| 6/11 | 500 | 5970 | 5010 | 84 | 60 | 7.1 | 120 | 50 | 8100 |
| 13/11 | 650 | 5660 | ND | 115 | 71 | 7.3 | 128 | 43 | 8960 |
| 20/11 | 650 | 6425 | ND | 101 | 69 | 6.9 | 172 | 36 | 9365 |
| 27/11 | 810 | 6210 | ND | 130 | 77 | 7.0 | 105 | 56 | 9940 |

| CLARIFIER I (INFLUENT) | | | | | | | | | | | |
|------------------------|----|-----|------|------|------|-----|-----|-----|-----|-----|--|
| Date: | SS | COD | FCOD | AMM | NN | OP | PH | CON | CL | ALK | |
| 5/6 | 28 | 104 | ND | 9.5 | 0.2 | 4.4 | 7.6 | 182 | 277 | 278 | |
| 12/6 | 39 | 112 | ND | <0.4 | 30.7 | 0.2 | 7.3 | 160 | 264 | 136 | |
| 19/6 | 25 | 67 | ND | <0.4 | 14.7 | 0.2 | 7.3 | 156 | 297 | 163 | |
| 26/6 | 32 | 100 | 66 | <0.4 | 14.7 | 1.7 | 7.4 | 185 | 345 | 162 | |
| 3/7 | 20 | 70 | ND | <0.4 | 14.4 | 2.2 | 7.4 | 175 | 375 | 111 | |
| 10/7 | 21 | 98 | ND | 0.5 | 2.7 | 3.9 | 7.7 | 210 | 324 | 374 | |
| 17/7 | 32 | 88 | ND | 3.3 | 11.9 | 1.0 | 7.0 | 128 | 232 | 92 | |
| 24/7 | 15 | 66 | 48 | 0.4 | 14.2 | 4.8 | 7.1 | 151 | 284 | 99 | |
| 31/7 | 15 | 73 | ND | <0.4 | 11.7 | 0.2 | 7.1 | 166 | 316 | 142 | |
| 7/8 | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | |
| 14/8 | 17 | 69 | ND | 0.5 | 19.0 | 0.2 | 7.4 | 169 | 351 | 140 | |
| 21/8 | 27 | 78 | 49 | <0.4 | 15.0 | 0.2 | 7.1 | 147 | 259 | 77 | |
| 28/8 | 29 | 79 | ND | <0.4 | 15.7 | 2.4 | 7.3 | 167 | 281 | 90 | |
| 4/9 | 16 | 69 | 51 | <0.4 | 2.5 | 3.6 | 7.6 | 181 | 354 | 153 | |
| 11/9 | 18 | 70 | ND | <0.4 | 12.2 | 0.9 | 7.5 | 194 | 407 | 140 | |
| 18/9 | 19 | 99 | ND | <0.4 | 13.5 | 1.2 | 7.6 | 222 | 434 | 142 | |
| 25/9 | 40 | 98 | ND | <0.4 | 18.2 | 8.7 | 7.1 | 144 | 270 | 67 | |
| 2/10 | 28 | 122 | ND | 26 | 0.7 | 5.5 | 7.7 | 199 | 332 | 279 | |
| 9/10 | 23 | 86 | ND | <0.4 | 14.7 | 0.2 | 7.5 | 189 | 361 | 126 | |
| 16/10 | 14 | 76 | ND | <0.4 | 12.8 | 0.5 | 7.5 | 167 | 345 | 105 | |
| 23/10 | 12 | 98 | ND | <0.4 | 10.3 | 0.4 | 7.5 | 171 | 350 | 126 | |
| 30/10 | 12 | 63 | ND | 0.4 | 12.1 | 0.2 | 7.4 | 155 | 306 | 108 | |
| 6/11 | 12 | 78 | ND | 0.4 | 12.8 | 0.2 | 7.4 | 184 | 382 | 102 | |
| 13/11 | 26 | 133 | ND | 1.2 | 1.4 | 8.9 | 7.8 | 199 | 383 | 296 | |
| 20/11 | 12 | 62 | 55 | 0.4 | 17.1 | 0.4 | 7.4 | 156 | 301 | 106 | |
| 27/11 | 13 | 73 | ND | <0.4 | 13.7 | 0.1 | 7.4 | 162 | 335 | 122 | |

- AGE: Sludge age (days)
- ALK: Alkalinity (CaCO₃ mg/L)
- AMM: Ammonia (N mg/L)
- CL: Chloride (Cl⁻ mg/L)
- COD: Chemical oxygen demand (mg COD/L)
- CON: Conductivity (mS/m)
- DSVI: Dissolved sludge volume index (ml/g)
- FCOD: Filtered chemical oxygen demand
- Flow: Flow (ML/day) for entire plant
- ND: Not determined
- NN: Nitrate and nitrite (N mg/L)
- OP: ortho-phosphate (P mg/L)
- RASF: RAS flow rate (s) (kL/day)
- RASSS: RAS suspended solids (mg/L)
- SETS: Settleable solids (30 min ml/L)
- SS: Suspended solids (105°C mg/L)
- SVI: Sludge volume index (ml/g)
- SWR: Sludge wasting rate (kL/day)
- TKN: Total Kjeldahl nitrogen (N mg/L)
- TP: Total phosphorus (P mg/L)
- VSS: Volatile suspended solids (mg/L)

Figures in red are anomalies and not transcription errors from the laboratory printouts.

ANNEXURE 4.11 RAW DATE FROM WILDEVOELVLEI WWTP

RAW INFLUENT (NO PST)

| Date: | Flow | SS | VSS | SETS | COD | TKN | AMM | TP | OP | CL | PH | CON | ALK |
|-------|------|-----|-----|------|-----|------|-----|------|------|-----|-----|-----|-----|
| 4/6 | 10.2 | 400 | 328 | 27.0 | 790 | 67.9 | 47 | 12.8 | 8.6 | 103 | 7.3 | 101 | 260 |
| 11/6 | 15.8 | 228 | ND | 11.0 | 355 | 42.2 | 26 | 7.0 | 4.2 | 124 | 7.3 | 93 | 20 |
| 18/6 | ND | 250 | ND | 11.0 | 554 | 72.1 | 39 | 11.0 | 8.4 | 135 | 7.4 | 106 | 247 |
| 25/6 | ND | 406 | ND | 15.0 | 732 | 55.2 | 31 | 10.4 | 7.1 | 112 | 7.3 | 95 | 218 |
| 2/7 | 19.0 | 220 | 192 | 9.0 | 533 | 46.3 | 30 | 10.8 | 7.9 | 138 | 7.4 | 108 | 229 |
| 9/7 | 15.3 | 312 | ND | 14.0 | 679 | 60.7 | 37 | 12.2 | 9.0 | 135 | 7.4 | 113 | 249 |
| 17/7 | ND | 350 | ND | 12.0 | 595 | 57.8 | 43 | 8.4 | 9.8 | 135 | 7.3 | 99 | 296 |
| 23/7 | ND | 303 | ND | 14.0 | 712 | 63.2 | 38 | 12.7 | 10.0 | 125 | 7.2 | 100 | 245 |
| 30/7 | 22.7 | 175 | ND | 9.0 | 411 | 32.5 | 21 | 8.9 | 5.7 | 150 | 7.4 | 106 | 220 |
| 6/8 | 19/6 | 210 | 167 | 7.0 | 369 | 36.4 | 20 | 5.7 | 3.4 | 102 | 7.4 | 79 | 175 |
| 13/8 | ND | 160 | ND | 10.0 | 520 | 45.9 | 28 | 9.2 | 6.7 | 135 | 7.3 | 106 | 258 |
| 20/8 | 18.5 | 211 | ND | 10.0 | 566 | 50.0 | 28 | 11.1 | 8.1 | 131 | 7.6 | 108 | 231 |
| 27/8 | 17.1 | 280 | ND | 9.0 | 636 | 53.4 | 30 | 8.7 | 6.8 | 131 | 7.5 | 109 | 247 |
| 3/9 | 17.1 | 178 | ND | 7.0 | 421 | 36.4 | 24 | 8.0 | 6.1 | 91 | 7.6 | 83 | 192 |
| 10/9 | 16.2 | 344 | ND | 13.0 | 679 | 62.1 | 44 | 14.1 | 10.6 | 120 | 7.4 | 113 | 273 |
| 17/9 | ND | 325 | ND | 8.0 | 576 | 46.8 | 32 | 10.7 | 8.3 | 98 | 7.5 | 94 | 217 |
| 24/9 | ND | 296 | ND | 11.0 | 547 | 88.9 | 60 | 9.0 | 6.3 | 163 | 8.2 | 135 | 206 |
| 1/10 | 12.9 | 456 | 376 | 14.0 | 823 | 60.1 | 40 | 14.3 | 11.2 | 116 | 7.3 | 116 | 260 |
| 8/10 | ND | 364 | ND | 12.0 | 679 | 64.4 | 45 | 12.5 | 9.7 | 127 | 7.5 | 110 | 275 |
| 15/10 | ND | 277 | ND | 13.0 | 688 | ND | 40 | 14.1 | 11.3 | 117 | 7.4 | 101 | 250 |
| 22/10 | ND | 300 | ND | 15.0 | 684 | 59.3 | 43 | 11.5 | 9.2 | 130 | 7.3 | 106 | 263 |
| 29/10 | ND | 384 | ND | 19.0 | 713 | 76.6 | 55 | 14.1 | 10.9 | 129 | 7.5 | 113 | 286 |
| 5/11 | ND | 297 | 187 | 11.0 | 804 | 61.9 | 40 | 11.8 | 9.8 | 114 | 7.4 | 104 | 249 |
| 12/11 | ND | 192 | ND | 7.0 | 564 | 59.1 | 42 | 12.2 | 10.8 | 130 | 7.5 | 110 | 254 |
| 19/11 | ND | 328 | ND | 16.0 | 787 | 54.6 | 36 | 14.5 | 12.9 | 107 | 7.3 | 96 | 249 |
| 26/11 | 10.9 | 367 | ND | 14.0 | 747 | 98.6 | 74 | 11.7 | 7.6 | 146 | 7.9 | 129 | 330 |

MIXED LIQUOR BIOREACTOR A

| Date | SETS | SS | VSS | SVI | DSVI | PH | SWR | AGE | RASSS | RASF |
|-------|------|------|------|-----|------|-----|-----|-----|-------|--------|
| 4/6 | 920 | 6200 | 5140 | 148 | 116 | 6.6 | ND | ND | 10840 | ND |
| 11/6 | 930 | 5450 | ND | 171 | 110 | 6.7 | ND | ND | 15690 | ND |
| 18/6 | 910 | 4925 | ND | 185 | 114 | 6.8 | ND | ND | 10215 | ND |
| 25/6 | 760 | 3660 | ND | 208 | 109 | 6.6 | ND | ND | 18410 | ND |
| 2/7 | 770 | 4240 | 3490 | 182 | 113 | 6.8 | 63 | 63 | 10760 | ND |
| 9/7 | 970 | 6250 | ND | 155 | 122 | 6.7 | 133 | 51 | 7100 | ND |
| 17/7 | 950 | 5630 | ND | 169 | 121 | 6.5 | ND | ND | 8340 | ND |
| 23/7 | 970 | 6560 | ND | 148 | 122 | 6.6 | ND | ND | 10290 | ND |
| 30/7 | 900 | 5525 | ND | 163 | 145 | 6.8 | 131 | 51 | 11555 | ND |
| 6/8 | 810 | 4500 | ND | 180 | 142 | 6.9 | 236 | 27 | 12440 | ND |
| 13/8 | 970 | 5400 | ND | 180 | 141 | 6.9 | ND | ND | 11730 | ND |
| 20/8 | 960 | 5700 | ND | 168 | 147 | 6.6 | 275 | 26 | 8920 | ND |
| 27/8 | 1000 | 5340 | ND | 187 | 157 | 6.9 | 333 | 21 | 6520 | ND |
| 3/9 | 950 | 4630 | ND | 205 | 147 | 7.1 | 182 | 37 | 7820 | 10MI/d |
| 10/9 | 950 | 5070 | ND | 187 | 142 | 6.9 | 278 | 25 | 6060 | 13 |
| 17/9 | 760 | 3330 | ND | 228 | 144 | 7.1 | ND | ND | 6180 | ND |
| 24/9 | 940 | 5110 | ND | 184 | 141 | 7.0 | ND | ND | ND | ND |
| 1/10 | 550 | 2600 | 2130 | 212 | 139 | 7.0 | 151 | 39 | 8340 | 182 |
| 8/10 | 940 | 4880 | ND | 193 | 148 | 6.9 | ND | ND | 6530 | ND |
| 15/10 | 950 | 4930 | ND | 193 | 138 | 6.9 | ND | ND | 7800 | ND |
| 22/10 | 820 | 3570 | ND | 230 | 123 | 6.9 | ND | ND | 7900 | ND |
| 29/10 | 920 | 4980 | ND | 185 | 120 | 7.0 | ND | ND | 7310 | ND |
| 5/11 | 380 | 2200 | 1860 | 173 | 127 | 7.1 | ND | ND | 6910 | ND |
| 12/11 | 880 | 3660 | ND | 240 | 142 | 6.9 | ND | ND | 8080 | ND |
| 19/11 | 930 | 4480 | ND | 208 | 134 | 6.9 | ND | ND | 5540 | ND |
| 26/11 | 930 | 4230 | ND | 220 | 151 | 7.0 | 339 | 21 | 5240 | ND |

| MIXED LIQUOR BIOREACTOR B | | | | | | | | | | |
|---------------------------|------|------|------|-----|------|-----|-----|-----|-------|------|
| Date | SETS | SS | VSS | SVI | DSVI | PH | SWR | AGE | RASSS | RASF |
| 4/6 | 1000 | 6110 | 4990 | 164 | 164 | 6.6 | ND | ND | 7630 | ND |
| 11/6 | 390 | 2050 | ND | 190 | 137 | 6.7 | ND | ND | 5470 | ND |
| 18/6 | 950 | 4105 | ND | 231 | 156 | 6.8 | ND | ND | 4535 | ND |
| 25/6 | 770 | 2990 | ND | 258 | 160 | 6.7 | ND | ND | 4110 | ND |
| 2/7 | 1000 | 5680 | 4610 | 176 | 176 | 6.7 | 65 | 74 | 7230 | ND |
| 9/7 | 1000 | 6530 | ND | 153 | 153 | 6.6 | 151 | 46 | 7900 | ND |
| 17/7 | 1000 | 7480 | ND | 134 | 171 | 6.4 | ND | ND | 8740 | ND |
| 23/7 | 460 | 2490 | ND | 185 | 161 | 6.9 | ND | ND | 11850 | ND |
| 30/7 | 970 | 5215 | ND | 186 | 161 | 6.7 | 221 | 30 | 9605 | ND |
| 6/8 | 970 | 5880 | ND | 165 | 156 | 6.7 | 286 | 21 | 7260 | ND |
| 13/8 | 800 | 4310 | ND | 186 | ND | 6.8 | ND | ND | 19770 | ND |
| 20/8 | 940 | 3970 | ND | 237 | 161 | 6.6 | 360 | 20 | 8420 | ND |
| 27/8 | 950 | 3960 | ND | 240 | 182 | 6.8 | 377 | 19 | 6200 | 11 |
| 3/9 | 930 | 3990 | ND | 233 | 170 | 7.0 | 225 | 30 | 5940 | ND |
| 10/9 | 970 | 6200 | ND | 156 | 142 | 6.7 | 225 | 30 | 8910 | ND |
| 17/9 | 770 | 5960 | ND | 129 | 121 | 6.8 | ND | ND | 15000 | ND |
| 24/9 | 940 | 5450 | ND | 172 | 125 | 7.0 | ND | ND | ND | ND |
| 1/10 | 940 | 5570 | 4490 | 169 | 129 | 6.8 | 13 | 169 | 6060 | 36 |
| 8/10 | 950 | 5480 | ND | 173 | 124 | 6.8 | ND | ND | 10070 | ND |
| 15/10 | 980 | 6670 | ND | 147 | 132 | 6.6 | ND | ND | 9710 | ND |
| 22/10 | 970 | 6760 | ND | 143 | 118 | 6.7 | ND | ND | 9740 | ND |
| 29/10 | 820 | 4930 | ND | 166 | 97 | 6.9 | ND | ND | 10350 | ND |
| 5/11 | 980 | 7230 | 6020 | 135 | 121 | 6.9 | ND | ND | 7190 | ND |
| 12/11 | 730 | 3360 | ND | 217 | 119 | 6.8 | ND | ND | 6480 | ND |
| 19/11 | 1000 | 8230 | ND | 122 | 107 | 6.8 | ND | ND | 12140 | ND |
| 26/11 | 960 | 6570 | ND | 146 | 116 | 7.0 | ND | ND | 12950 | ND |

| CLARIFIER A (EFFLUENT) | | | | | | | | | |
|------------------------|----|-----|------|------|------|-----|-----|-----|-----|
| Date: | SS | COD | AMM | NN | OP | CL | PH | CON | ALK |
| 4/6 | 8 | 33 | 11 | <0.1 | 1.7 | 117 | 7.3 | 80 | 160 |
| 11/6 | 6 | 30 | <0.4 | 7.9 | 1.8 | 136 | 6.9 | 81 | 95 |
| 18/6 | 4 | 41 | <0.4 | 3.3 | 8.5 | 142 | 7.1 | 75 | 113 |
| 25/6 | 10 | 47 | <0.4 | 1.3 | 6.8 | 130 | 7.0 | 79 | 114 |
| 2/7 | 12 | 41 | <0.4 | 12.4 | 5.8 | 151 | 6.9 | 91 | 86 |
| 9/7 | 4 | 33 | 1.3 | 8.8 | 5.9 | 147 | 7.2 | 91 | 99 |
| 17/7 | 5 | 33 | 0.7 | 8.5 | 7.3 | 143 | 7.1 | 88 | 96 |
| 23/7 | 6 | 37 | <0.4 | 8.4 | 8.2 | 144 | 6.9 | 90 | 88 |
| 30/7 | 3 | 51 | <0.4 | 8.9 | 4.6 | 161 | 7.1 | 98 | 121 |
| 6/8 | 9 | 33 | <0.4 | 10.4 | 5.1 | 158 | 7.2 | 94 | 110 |
| 13/8 | 2 | 36 | 0.4 | 9.6 | 4.5 | 149 | 7.2 | 93 | 127 |
| 20/8 | 2 | 51 | <0.4 | 9.5 | 4.8 | 151 | 7.3 | 96 | 110 |
| 27/8 | 4 | 30 | <0.4 | 9.5 | 4.2 | 140 | 7.3 | 92 | 102 |
| 3/9 | 4 | 30 | <0.4 | 10.7 | 4.6 | 137 | 7.4 | 91 | 106 |
| 10/9 | 4 | 51 | 0.1 | 9.4 | 5.5 | 136 | 7.3 | 88 | 102 |
| 17/9 | 3 | 30 | <0.4 | 11.5 | 6.1 | 132 | 7.3 | 88 | 87 |
| 24/9 | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| 1/10 | 7 | 33 | <0.4 | 4.9 | 5.4 | 125 | 7.3 | 91 | 103 |
| 8/10 | 3 | 41 | <0.4 | 8.1 | 5.8 | 126 | 7.3 | 79 | 91 |
| 15/10 | 15 | 35 | <0.4 | 7.0 | 7.2 | 127 | 7.3 | 81 | 94 |
| 22/10 | 3 | 34 | <0.4 | 6.7 | 8.3 | 120 | 7.2 | 77 | 90 |
| 29/10 | 3 | 19 | <0.4 | 8.1 | 8.7 | 129 | 7.3 | 79 | 84 |
| 5/11 | 4 | 30 | <0.4 | 2.3 | 4.3 | 114 | 7.5 | 73 | 108 |
| 12/11 | 4 | 38 | 0.7 | 6.2 | 7.2 | 123 | 7.2 | 76 | 97 |
| 19/11 | 4 | 37 | <0.4 | 13.6 | 9.8 | 121 | 7.3 | 79 | 67 |
| 26/11 | 6 | 30 | <0.4 | 13.2 | 10.3 | 116 | 7.2 | 76 | 71 |

| CLARIFIER B (EFFLUENT) | | | | | | | | | | |
|------------------------|----|-----|-----|------|-----|-----|-----|-----|-----|--|
| Date: | SS | COD | AMM | NN | OP | CL | PH | CON | ALK | |
| 4/6 | 7 | 30 | 0.6 | 4.2 | 5.9 | 115 | 7.1 | 74 | 107 | |
| 11/6 | 10 | 34 | 0.4 | 5.8 | 2.4 | 143 | 7.0 | 84 | 111 | |
| 18/6 | 5 | 34 | 1.8 | 2.7 | 1.0 | 142 | 7.2 | 83 | 128 | |
| 25/6 | 21 | 70 | 1.2 | 2.3 | 1.4 | 125 | 7.0 | 77 | 117 | |
| 2/7 | 10 | 44 | ND | 4.0 | 2.7 | 150 | 7.1 | 89 | 124 | |
| 9/7 | 4 | 40 | 0.5 | 3.3 | 2.6 | 149 | 7.3 | 89 | 126 | |
| 17/7 | 5 | 37 | 1.1 | 1.5 | 4.0 | 146 | 7.2 | 86 | 125 | |
| 23/7 | 10 | 44 | 0.7 | 9.2 | 2.4 | 133 | 7.0 | 83 | 91 | |
| 30/7 | 5 | 50 | ND | 7.9 | 2.7 | 160 | 7.2 | 97 | 128 | |
| 6/8 | 18 | 47 | ND | 6.4 | 2.9 | 156 | 7.3 | 93 | 128 | |
| 13/8 | 14 | 75 | 9.8 | 5.6 | 5.5 | 148 | 7.2 | 98 | 168 | |
| 20/8 | 3 | 51 | ND | 8.5 | 1.0 | 151 | 6.9 | 91 | 114 | |
| 27/8 | 4 | 40 | ND | 5.7 | 2.6 | 134 | 7.4 | 89 | 117 | |
| 3/9 | 5 | 37 | ND | 7.2 | 3.5 | 136 | 7.6 | 89 | 118 | |
| 10/9 | 6 | 44 | 0.1 | 6.7 | 3.7 | 135 | 7.4 | 87 | 116 | |
| 17/9 | 6 | 33 | ND | 12.8 | 4.9 | 133 | 7.5 | 88 | 89 | |
| 24/9 | ND | ND | ND | ND | ND | ND | ND | ND | ND | |
| 1/10 | 13 | 40 | ND | 9.4 | 3.7 | 127 | 7.4 | 90 | 91 | |
| 8/10 | 1 | 37 | ND | 10.4 | 4.8 | 129 | 7.3 | 78 | 86 | |
| 15/10 | 6 | 32 | 1.0 | 0.6 | 3.3 | 120 | 7.4 | 79 | 114 | |
| 22/10 | 5 | 37 | 3.6 | 1.8 | 7.0 | 120 | 7.3 | 79 | 126 | |
| 29/10 | 6 | 30 | 5.4 | 0.2 | 9.1 | 128 | 7.5 | 80 | 136 | |
| 5/11 | 8 | 37 | 12 | <0.4 | 4.3 | 114 | 7.5 | 73 | 108 | |
| 12/11 | 17 | 60 | 24 | 0.5 | 8.4 | 122 | 7.4 | 95 | 201 | |
| 19/11 | 23 | 70 | 33 | 0.4 | 7.4 | 120 | 7.5 | 99 | 229 | |
| 26/11 | 22 | 61 | 14 | 0.4 | 0.6 | 112 | 7.4 | 79 | 174 | |

- AGE: Sludge age (days)
- ALK: Alkalinity (CaCO₃ mg/L)
- AMM: Ammonia (N mg/L)
- CL: Chloride (Cl mg/L)
- COD: Chemical oxygen demand (mg COD/L)
- CON: Conductivity (mS/m)
- DSVI: Dissolved sludge volume index (ml/g)
- Flow: Flow (ML/day) for entire plant
- ND: Not determined
- NN: Nitrate and nitrite (N mg/L)
- OP: ortho-phosphate (P mg/L)
- RASF: RAS flow rate (s) (ML/day)
- RASSS: RAS suspended solids (mg/L)
- SETS: Settleable solids (30 min ml/L)
- SS: Suspended solids (105°C mg/L)
- SVI: Sludge volume index (ml/g)
- SWR: Sludge wasting rate (kL/day)
- TKN: Total Kjeldahl nitrogen (N mg/L)
- TP: Total phosphorus (P mg/L)
- VSS: Volatile suspended solids (mg/L)

Figures in red are anomalies and not transcription errors from the laboratory printouts.