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DESIGN AND OPERATING STRATEGIES TO MINIMIZE BULKING BY ANOXIC-AEROBIC FILAMENTOUS ORGANISMS IN NUTRIENT REMOVAL ACTIVATED SLUDGE PLANTS

Report to the Water Research Commission

by

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SYNOPSIS

The objective of WRC project 775 is to test the principles of the Anoxic-Aerobic (AA) bulking hypothesis at large-scale activated sludge nutrient removal plants. The hypothesis was developed by the Water Research Group at the University of Cape Town to conceptually model the prolific development of a group of activated sludge filamentous organisms classified by Jenkins et al. (1984) as low F/M. As a consequence of the hypothesis, many of the filaments referred to as low F/M are now described by the term Anoxic-Aerobic (AA). The term AA describes more accurately the conditions required for proliferation of the specific filaments, namely Microthrix parvicella (M.p.), and types 0092, 0675, 0041, 0914, 1851 and 0803.

There has been some reluctance from scientists and engineers in the sanitary engineering profession to accept the hypothesis and apply its principles in the design and operation of nutrient removal activated sludge plants. This is probably a consequence of the development and testing of the hypothesis in laboratory-scale activated sludge systems only. To this point the only supportive evidence for the hypothesis from large-scale plants has been anecdotal in nature. The objective of this research project is to determine the applicability of the AA hypothesis to large-scale activated sludge plants and develop criteria for their design and operation in order to minimise filament proliferation.

Because it was not possible to conduct controlled experiments on large-scale plants, historical data from a number of plants was evaluated to determine the extent to which the biological response of the plant is in agreement with the hypothesis.

The consequences of the hypothesis proposed by Casey et al. (1993) were investigated in this project are that;

- high ammonia concentrations in plant influents can lead to poor sludge settleability,
- high nitrate concentrations in the anoxic zone can lead to poor sludge settleability,
- (3) aerobic mass fractions between 30 and 40 percent can result in poor sludge settleability.

The results of the investigation are described below:

Effect on filament proliferation of variation in the influent wastewater ammonia concentration

From plots of daily data of influent ammonia concentrations, it was not generally possible to draw conclusions as to the effect of the ammonia concentration on sludge settleability for the selected plants due to the extreme variation in the data both on a daily and seasonal basis.

Effect on filament proliferation of variation in the nitrate concentration in the anoxic zone

From plots of daily data of nitrate concentration at the end of the anoxic zone, it was not possible to draw conclusions as to its effect on sludge settleability due to the extreme variation in the data.

Effect on filament proliferation of variation in aerated and unaerated mass fractions

The value of system aerobic mass fraction was plotted against sludge settleability (as DSVI) for seven nutrient removal, activated sludge plants in the Gauteng region. A significant relationship was determined between the two parameters; for increasing aerobic mass fraction above about 45 percent, the sludge settleability improved significantly. The variation in system aerobic mass fraction was from 44 to 76 percent, and the variation in DSVI was from 61 to 162 ml/g. A factor that makes the findings somewhat more significant is that in evaluating the plants against each other, the plants conform to the hypothesis, despite the seven plants receiving wastewater from completely separate drainage areas and being operated by three different authorities.

Conclusions and Summary

From evaluation of the historical operating data and configurations of seven nutrient removal activated sludge plants in Gauteng, it can be concluded that:

- The filaments that dominate in the plants are those that are classified as AA
 to which the AA bulking hypothesis applies,
- (2) In comparing the sludge settleability and aerobic mass fractions of the plants, generally, plants with larger aerobic mass fractions had better overall sludge settleabilities.
- (3) A means of significantly controlling filament proliferation in nutrient removal activated sludge plants is to ensure that they have as large an aerobic mass fraction as possible without compromising the anoxic mass fraction required for denitrification.
- (4) The Daspoort data confirmed the relationship between SSVI and DSVI as SSVI = 0.69 DSVI.

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PROJECT OBJECTIVES

This report describes Water Research Commission Project No. 775, entitled "Development of strategies for amelioration of bulking by anoxic-aerobic filamentous organisms in nutrient removal activated sludge systems".

The objective of the project was to determine under full-scale plant operating conditions the applicability of the hypothesis proposed by Casey et al. (1993) for bulking by anoxic-aerobic filaments. A short discussion of the hypothesis is given in Section 3, along with a discussion of the application of the hypothesis to large-scale plants. A full discussion of the hypothesis is given by Casey et al. (1993) and the researchers also summarise laboratory-scale experimental work that supports the hypothesis.

The approach to the project has changed somewhat as it has progressed. Initially, the objective was to undertake a comprehensive survey of the operating conditions and plant characteristics of the majority of the municipal nitrogen (N), and nitrogen and phosphorus (P) removal wastewater treatment plants in the Gauteng region. From the survey it was hoped that sufficient historical operating and analytical data could be gathered, against which the hypothesis could be evaluated. It was intended that sufficient information would be gathered from the evaluation in order to propose design and operating criteria for the minimisation of bulking by AA filaments. However, from preliminary investigations it became apparent that sufficient analyses of nutrient concentrations and sludge settleability, and records of operational changes are made at only a few activated sludge plants in the Gauteng region. Therefore if the research project were to be undertaken to examine the aspects specific to the hypothesis, a dedicated sampling and analytical programme would be required. However, because the priority of a large wastewater treatment plant is to achieve a specific effluent quality, and because proper testing of the hypothesis would require the effluent quality to be compromised, it was not possible to alter operation of plants for the purposes of research.

Therefore it was decided to evaluate periods of historical data from the few plants that conduct regular analyses of nutrient (nitrogen in particular) concentrations in the influent and the effluent and which measure sludge settleability in an attempt to determine the applicability of the bulking hypothesis to full-scale activated sludge nutrient removal plants.

From this information, criteria have been determined that can assist in design of nutrient removal activated sludge plants to ensure minimal incidences of sludge bulking.

THE PHENOMENA OF BULKING

2.1 Background

The nitrification, denitrification, biological excess phosphorus removal (NDBEPR) system is given prime consideration whenever a new wastewater treatment plant is required in South Africa. This is a consequence of its ability to meet stringent nitrogen and phosphorus removal standards. However experience with ND and NDBEPR plants in South Africa and elsewhere has brought to light a behavioural response in the activated sludge that has a significant effect on the design and operation of the system. The systems are prone to produce sludges that settle poorly in the clarifier (secondary settling tank) where the sludge and treated effluent are separated by gravity sedimentation settling. The activated sludge biological culture that develops in ND and NDBEPR systems is diverse in organism type, function and physical characteristic. The organisms in activated sludge can be categorised on the basis of their morphological characteristics, either as filamentous or floc-forming.

The relative proportions of filamentous and floc-forming organisms largely determines the settleability of the sludge in the final stage of treatment, the clarifier. Absence of filamentous organisms gives rise to a sludge that settles rapidly but clarifies poorly, in that small sludge particles remain in the supernatant, a condition termed "pin-point floc" which results in loss of solids with the effluent. Presence of excessive filamentous organisms in sludge inhibits its settleability in the settling tank and it is referred to as a bulking sludge. A bulking sludge may result in gross sludge loss with the effluent i.e. secondary settling tank failure, under normal design operating conditions. For optimum settling tank performance, filaments should be present in sufficient numbers such that their growth within the floc provides the floc with structure, thereby enhancing the sludge clarification characteristics, but the filaments should not proliferate to the extent that filament bridging between the flocs, with associated diffuse floc-formation, inhibits sludge settleability. The correct proportion of filaments and floc-formers produces a rapidly settling sludge that also clarifies the surrounding mixed liquor.

Bulking sludges are common in nutrient removal activated sludge plants in South Africa. A survey during 1985 of 96 ND and NDBEPR plants showed that 32 percent of the plants had problems with filamentous bulking sludges; a further survey in 1987 of 26 NDBEPR plants showed that 82 percent had bulking problems caused by filamentous organisms.

In many of the existing NDBEPR plants, poor sludge settleability results in a considerable loss of treatment capacity. Controlling the proliferation of filamentous organisms, and thereby maintaining a good settling sludge in the plant would permit significantly higher wastewater flows (50 to 100%) to be treated in existing plants, resulting in large capital savings. The possibility of achieving such savings is the motivation behind the research into the causes and control of filamentous bulking in ND and NDBEPR systems.

2.2 Investigating the bulking phenomena

The discussion above has focused on bulking problems in ND and NDBEPR systems in South Africa, but the problem is not confined to these systems and country; it has been encountered worldwide in many different kinds of activated sludge system, including ones that do not have unaerated zones, i.e. completely aerobic systems. It has been found that a number of different filamentous organisms contribute to the bulking condition and considerable effort has been directed at identifying the conditions in the system that gives rise to proliferation of the different filamentous organism types. The filamentous organisms that give rise to bulking in aerobic systems are not the same as those in ND and NDBEPR plants. This, and other research led to the categorisation of the filaments in terms of the condition that apparently gives rise to their proliferation, viz. low dissolved oxygen (DO), low food/micro-organism (low F/M) ratio, septic wastewater, nutrient deficiency, and low pH. In terms of this categorisation, the filamentous organisms that proliferate in ND and NDBEPR systems belong mainly to the low F/M category. This categorisation seemed reasonable because the ND and NDBEPR plants invariably are low F/M (equivalently – long sludge age) plants.

As a consequence of the dominance of low F/M filaments causing the bulking problems in ND and NDBEPR plants, research was commenced to evaluate the strategy that was promoted for the control of the low F/M filaments. This strategy is based on the so-called selector effect.

A selector effect is induced in an activated sludge when a readily biodegradable COD (RBCOD) concentration gradient is established in the system by modifying the mixing regime of the system by introducing plug-flow conditions, compartmentalisation, or a selector reactor, the last being a small reactor receiving the sludge return and influent flow before the main aeration reactor. With these modifications, the floc-formers in the activated sludge acquire a high RBCOD uptake rate allowing them to successfully compete for RBCOD substrate against the filamentous organisms. However, when a selector (either aerobic or anoxic) was placed ahead of an intermittently aerated reactor mimicking an Orbal or Carousel nitrification-denitrification system, the selector effect was found to be not effective for controlling the proliferation of the low F/M filaments. Furthermore, anaerobic reactors in the NDBEPR systems, which in effect serve the same function as the selector reactor in that they also allow preferential uptake of RBCOD (or rather its fermentation by products such as volatile fatty acids) by floc-formers (in this case the polyP organisms responsible for the BEPR), also were not effective in controlling the low F/M filaments. Curiously, considering that these filaments were the so-called low F/M ones, continuous aeration (i.e. completely aerobic but maintaining the low F/M condition) was found to successfully control them, but the completely aerobic conditions were then not adequate for NDBEPR. It became apparent that the selector effect approach for understanding the low F/M filament bulking problems was no longer productive for guiding experiments into the causes and control of the filaments in this category. This set-back placed research into an exploratory phase requiring new research directions to be identified and pursued, in order to understand the causes and control of low F/M filament bulking.

The failure of the selector effect to control low F/M filament bulking in ND and NDBEPR systems prompted a wide-ranging exploratory programme into the causes of proliferation of these filaments in ND and NDBEPR systems. The programme resulted in development of the AA bulking model, which is described by Casey et al. (1993). The model is a fundamental biochemical/microbiological model that describes the competition between filamentous and floc-forming organisms and is referred to as the bulking model. The bulking model was developed from a fundamental conceptual biochemical model describing facultative heterotrophic behaviour and is described in detail in Section 3 below.

2.3 Parameters for measurement of sludge settleability

A number of parameters are used as a measure of activated sludge settleability. One of the best measures is the extent to which filaments protrude from the floc into the bulk liquid to form bridges between the flocs and this is referred to as the Total Extended Filament Length (TEFL). However because of the difficulty in undertaking the TEFL measure, other indirect measurements of the extent of filament proliferation are used. These are the Sludge Volume Index (SVI), the Diluted Sludge Volume Index (DSVI) and the Stirred Specific Volume Index (SSVI). A difficulty in comparing sludge settleability data from different plants is that different plant operators use different indices to measure sludge settleability. In order to compare sludge settleability data, the DSVI has been chosen as the standard indicator and SVI and SSVI data is equated to the DSVI parameter using the correlations of Pitman (1984) and Ekama and Marais (1986).

The DSVI parameter has been chosen as the standard for this study as a consequence of the findings of many workers investigating sludge settleability parameters. In their 1984 review, Ekama and Marais found that the SVI has attracted criticism because it:

- (1) is not independent of sludge concentration
- (2) is not independent of cylinder diameter and depth
- (3) is affected by gentle stirring
- (4) has no observable relation to rheological properties of sludge
- (5) has no relation to zone settling velocity
- (6) does not provide a good measure of the settling behaviour of the sludge.

However, for historical reasons, the SVI continues to be the method promoted by many plant operating authorities for measuring sludge settleability. The results from one of the plants examined in this study will demonstrate that a more consistent measure of sludge settleability can be achieved with the SSVI than with the SVI measure.

Lee et al. (1983) motivated for adopting the DSVI, because this parameter gave a significantly better correlation with TEFL than did the SVI. Also, it has a wider range of sensitivity with respect to TEFL over which sludges could be characterised. The SSVI has the advantage that for the same sludge, or sludges with similar settleability, reproducible results are obtained over a wide range of concentrations. White (1975) found that for most, but not all sludges he investigated, the SSVI was independent of initial sludge concentration. As a consequence, a standard concentration of 3,5 g/ ℓ was proposed for reporting SSVI data, and this parameter is known as the SSVI_{3,5}. Because a number of tests have to be conducted to interpolate to a mixed liquor suspended solids concentration of 3,5 g/ ℓ , the test is time consuming and as such has limited appeal to plant operators.

In an extensive review of the various sludge settleability parameters, Ozinsky and Ekama (1995) provide a summary of the most important work conducted by numerous researchers. The information from the review that is of most importance to this study are the relationships between SVI and DSVI (taken from Ekama and Marais 1986) and SSVI_{3,5} and SVI (taken from Pitman 1984). These relationships were used to generate DSVI data points from SSVI_{3,5} and SVI data points.

DESCRIPTION OF AA BULKING HYPOTHESIS AND ITS APPLICATION TO LARGE-SCALE NITROGEN AND NUTRIENT REMOVAL ACTIVATED SLUDGE PLANTS

3.1 Background to and description of AA Bulking Hypothesis

There were three steps involved in the development of the AA bulking hypothesis and Casey et al. (1993) explains the contribution of each of these. Firstly, a literature review was conducted of the major biochemical / microbiological pathways mediated by facultative organisms under separate aerobic and anoxic conditions and changes between these. Secondly, from the literature review, a conceptual model for facultative organism respiration was developed. Thirdly, the conceptual model was applied to filamentous and floc-forming organisms under anoxic and aerobic conditions and a biochemical / microbiological model (the bulking model) was developed to explain their behaviour. The bulking model provides a hypothesis for floc-former or AA filament dominance under different conditions. The hypothesis can be stated as follows:

In activated sludge systems, floc-formers and filaments compete for mutually growth-limiting substrate. Under continuous aerobic or continuous anoxic conditions, the floc-formers outcompete the filaments for substrate due to higher substrate utilisation rates and filament growth is restricted. In nitrification-denitrification (ND) and nitrification-denitrification biological excess phosphorus removal (NDBEPR) activated sludge systems, competition between filaments and floc-formers for mutually growth-limiting substrate is influenced by inhibition of the floc-formers' substrate utilisation under aerobic conditions. Under anoxic conditions, in utilisation of substrate the floc-formers execute the denitrification of nitrate through each of the denitrification intermediates to dinitrogen as follows:

In the absence of, or at low concentrations of readily biodegradable substrate, the intermediate, nitric oxide is accumulated intracellularly in the floc-formers. In the subsequent aerobic zone, intracellular nitric oxide inhibits the utilisation of oxygen by floc-forming organisms as a result of the interaction of nitric oxide with the enzymes that mediate aerobic respiration, specifically the cytochrome oxidase, cytochrome o. Under these conditions, floc-forming organisms are inhibited in aerobic respiration. The inhibition of aerobic respiration causes electrons to be redirected to the nitrite, nitric oxide, and nitrous oxide reductases for the reduction of the nitrogen oxide specific to these reductases (aerobic denitrification); this mechanism continues as long as nitrite remains available. Thus, under aerobic conditions a low concentration of nitrite contains the intracellular accumulation of nitric oxide (and thereby the inhibitory effect) and higher concentrations of nitrite exacerbate the inhibitory effect. The inhibition of aerobic respiration and the phenomenon of aerobic denitrification in floc-formers results in lower substrate utilisation rates at lower net energy yields.

In contrast to floc-formers, the filamentous organisms are nitrate reducers and execute only part of the denitrification pathway, i.e. the reduction of nitrate to nitrite as follows:

They do not accumulate nitric oxide and therefore are not inhibited in aerobic respiration in the subsequent aerobic zone. Thus in systems in which sludge is exposed to alternating anoxic-aerobic conditions with nitrite present in the anoxic zone, floc-formers are placed at a disadvantage in the competition for substrate under subsequent aerobic conditions and filaments gain an advantage. Given sufficient advantage, the filaments proliferate with time.

It was envisaged that with further work, the hypothesis would be developed into a form that could be used as a diagnostic tool to determine the propensity of activated sludge nutrient removal plants to produce bulking sludges. However, before this can be done, it must firstly be shown that the model is applicable to sludge settleability behaviour in large-scale BNR plants. Although the bulking model has been tested against the behaviour of laboratory-scale activated sludge plants, it is necessary to apply the model to large-scale plants.

The objective of the following section is to illustrate the manner in which the AA bulking hypothesis can be applied to large-scale BNR plants.

3.2 Application of the AA bulking hypothesis to large-scale plants

Although the model and the hypothesis present a fundamental description of the competition between filaments and floc-formers, there are four practical aspects concerned with large-scale plants to which the model and hypothesis can be applied. These are: (1) variation in the TKN/COD ratio of raw sewage, (2) variation in the nitrate and nitrite concentrations in the anoxic zone preceding the main aeration basin, and (3) variation in the aerobic and anoxic mass fractions. Described below are the predictions of the hypothesis related to changes in the three aspects above. It is these aspects that will be investigated during evaluation of the selected large-scale plants.

Variation in TKN/COD ratio in the influent sewage

Although not directly described by the hypothesis, from activated sludge kinetics, it can be assumed that increases in the TKN/COD ratio in the influent sewage would result in increases in the nitrate and nitrite concentration in the anoxic zone and decreases in the TKN/COD ratio will result in decreases in the same parameter. As noted in (2) below, the hypothesis proposes a relationship between the filament proliferation and the concentration of nitrate and nitrite in the anoxic zone. Therefore, the TKN/COD ratio is a parameter that can be used as an indicator of the potential for filament proliferation. Unfortunately at most of the plants analysed in the study the influent TKN is not measured and data only existed for the influent ammonia concentration. The variation of the ammonia concentration in the influent was therefore adopted as the parameter to be compared with the sludge settleability.

(2) Variation in nitrate and nitrite concentration in the anoxic zone immediately preceding the main aerobic zone

As stated in the hypothesis, floc-formers will be inhibited under aerobic conditions and filaments will proliferate if denitrification is incomplete in the anoxic zone preceding the main aerobic zone. In a large-scale plant this will be measured as nitrate and/or nitrite in the anoxic zone at concentrations of more than about $1 \text{ mgN}/\ell$.

(3) Variation in aerated and unaerated mass fractions

A major aspect of the hypothesis and model concerns the extent to which the aerated and unaerated mass fractions of the system affect the sludge settleability. The model predicts that poor sludge settleability will result from systems with an aerated mass fraction of between 30 and 40 percent and systems with aerated mass fractions larger than 40 percent and less than 30 percent will develop better sludge settleability. The effect of aerated mass fraction on sludge settleability is examined in Section 4.4 below and a fundamental explanation of the relationship between aerobic and anoxic respiration and aerated mass fraction is described in Section 5 below.

(4) Other areas related to sludge bulking

In addition to the above three aspects of the hypothesis, two other areas associated with filamentous organism sludge bulking were investigated, i.e.

- The type of filamentous organism found in N removal and N&P removal plants.
- (2) The relationship between DSVI and SSVI as sludge settleability parameters.

These areas were investigated not specifically to test the hypothesis, but to check whether these aspects which had been reported on in laboratory-scale systems would be applicable to full-scale systems.

ANALYSIS OF EIGHT LARGE-SCALE WASTEWATER TREATMENT PLANTS WITH REGARD TO THEIR POTENTIAL FOR BULKING

4.1 Selection of wastewater treatment plants for evaluation of bulking

As described in Section 1, it was intended that a wide-ranging survey would be conducted of activated sludge plants in the Gauteng region to determine the applicability of the hypothesis. However, it was realised that none of the plants conduct all the analyses required for an indepth evaluation of the model and only a few plants conducted sufficient analyses to undertake a superficial evaluation. Initially, eight plants were selected for the examination of the influence of wastewater characteristics, operational strategy, and system configuration on sludge settleability. These are Daspoort (operated by Pretoria Municipality), Waterval, Rynfield and Vlakplaats (operated by the East Rand Water Care Company [ERWAT]) and Goudkoppies, Bushkoppie, Olifantsvlei, and Northern Works (operated by the Greater Johannesburg Metropolitan Council [GJMC]).

The objective in analysing the influent wastewater characteristics, the operational strategy and the system configurations of the eight plants was to determine if one of the aspects listed in Section 3.2 above could be identified as having a major influence on sludge settleability.

4.2 Analysis of the data

Data for the plants was obtained from the operating authorities and analysed in relation to the requirements of this project.

In general the analysis of the data proved to be disappointing, mainly due to the following reasons:

- The sampling and analysis programmes on the works were orientated towards providing
 operating data rather than research data. As a result the data available was not always in a
 form which enabled meaningful conclusions to be drawn.
- Where data was available it often displayed such large daily variations that it was impossible to draw meaningful conclusions from it.
- At times the sampling and analysis protocol adopted were deficient which rendered the data unreliable.

Nevertheless an attempt was made to analyse the data and the conclusions drawn are described below.

4.2.1 Relationship between dominant filament type and reactor configuration

The historical filament identifications conducted at each of the four ERWAT wastewater treatment plants, i.e. Daspoort, Vlakplaats, Waterval, and Rynfield, were evaluated to determine the dominant filaments present. The historical data for each plant was extensive, and the identifications presented in Table 1 are a summary of the entire time period under examination. This time period varied for the plants but generally was a period of between one and two years. However, it is interesting that although there were changes in the type of filament dominant in any plant at any one time, generally, the same filament types remained dominant throughout the period investigated. The order in which the filaments are presented in the Table 1 represents their level of dominance in that plant. The dominant filaments present in the Johannesburg plants, ie Goudkoppie, Bushkoppie, Olifantsvlei and Northern Works are taken from Pitman (1996).

Table 1: Dominant filamentous organisms identified in eight activated sludge plants

Plant	Filament types present				
Rynfield WCW	M.p. / 0092 / 0041				
Vlakplaats	0092 / M.p. / 0041 / 1701				
Daspoort WCW	0092 / 0041 / M.p. / 0961 / 1701				
Waterval	N.lim. / 0041 / Begg. / Thiox. / 021N / 0092 / Noc. / M.p.				
Goudkoppies WWTW	$M.p. \rightarrow 1803 / M.p.$				
Bushkoppie WWTW	M.p. / 0092				
Olifantsvlei WWTW	1851 / 0041				
Northern WWTW (Unit 3)	$M.p. / 1803 \rightarrow 1803 / M.p.$				

The abbreviations used for the filaments are as follows: Microthrix parvicella (M.p.), Nostocoida Limicola (N.lim.), Thiothrix (Thiox.), Beggiatoa (Begg.) and Nocardia (Noc.). The filament identifications in which types 0092 and 0041 and M.p. were dominant are in agreement with the classification of Jenkins et al. (1984) which describes these filaments as low F/M and which have also been described by Casey et al. (1993) as developing under conditions in which the activated sludge is sequentially subjected to anoxic (and/or anaerobic) and aerobic conditions and consequently referred to as AA (anoxic-aerobic) filaments.

In particular, it is noteworthy that with the exception of Waterval, the types of filaments found to be dominant (i.e. type 0092, M.p. and types 1803, 1851 and 0041) are the filaments most often dominant in nitrogen and nutrient removal plants in South Africa, as indicated in the surveys conducted by Blackbeard et al. (1986, 1988). For Daspoort and Vlakplaats, the dominant filament was type 0092 with either M.p. or type 0041 as second or third most common. For Rynfield, M.p. was the dominant filament, with types 0092 and 0041 second and third most common.

As support for the hypothesis that M.p. proliferates with the presence of high nitrate concentrations in the anoxic zone, it has been found (Pitman, 1996) that for Goudkoppies and Northern works, M.p. was dominant in winter and type 1803 dominant in summer. This is indicated in Table 1 as an arrow between the filament groups. It is probable that under winter conditions, the rate of denitrification is somewhat reduced, and assuming complete nitrification in the system, an increased concentration of nitrate would be present in the noxic zone. In support of the above, Eikelboom (1994) reports that in large nutrient removal plants in Europe, the worst bulking conditions generally occur during the winter months. These findings support the AA bulking hypothesis which states that one of the conditions required for the proliferation of AA filaments is the presence of nitrate and/or nitrite at the end of the anoxic zone or period as described in Section 3.1 above.

The filaments found to be dominant at Waterval are those classified by Jenkins et al. (1984) as being associated with conditions of "low DO" and "septic wastewater". This finding correlates with the type of effluent treated at Waterval which contains a very high industrial fraction.

The analysis of the data as originally envisaged in Section 3.2 is discussed below.

4.2.2 Effect of influent ammonia concentration on sludge settleability

The relationship between the influent ammonia concentration and sludge settleability (DSVI) was analysed for those plants where the two parameters were measured regularly. The data analysed generally showed such a high degree of scatter that trends could not properly be interpreted. Only the data from Vlakplaats showed a discernible trend and is reported on here.

Vlakplaats

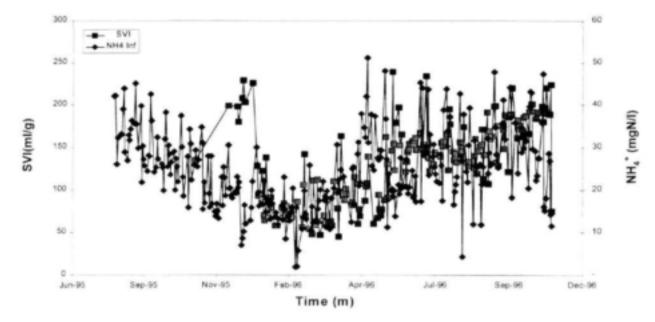


Figure 1: Vlakplaats: SVI and Influent Ammonia vs Time

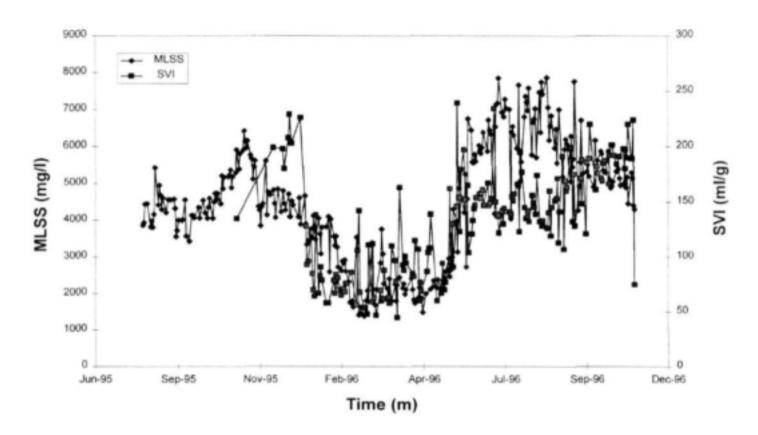


Figure 2: Vlakplaats: SVI and MLSS vs Time

It is apparent from Figure 1 that variation in ammonia concentration in the influent is reflected by similar variation in the sludge settleability. However it will be seen from Figure 2 that the MLSS concentration also follows the same trend as the SVI over the same period. It is therefore not conclusive as to whether the variation in SVI is a consequence of the concentration of nitrate produced in the system from nitrification and hence due to filament proliferation, or whether other factors influencing the MLSS concentration have caused the variation in SVI. This is a good illustration of the importance of measuring settleability in terms of DSVI rather than SVI because the former is effected by the MLSS concentration whereas the latter is not.

4.2.3 Effect of the nitrate and nitrite concentrations entering the aerobic zone from the anoxic zone

Of the plants investigated, only Vlakplaats and Rynfield had data available for the nitrate concentrations in the anoxic zone. In the absence of nitrate data in the anoxic zone at Daspoort, the variation of the nitrate concentration in the final effluent with the variation in SVI was analysed. The analysis of this data is discussed below:

Vlakplaats

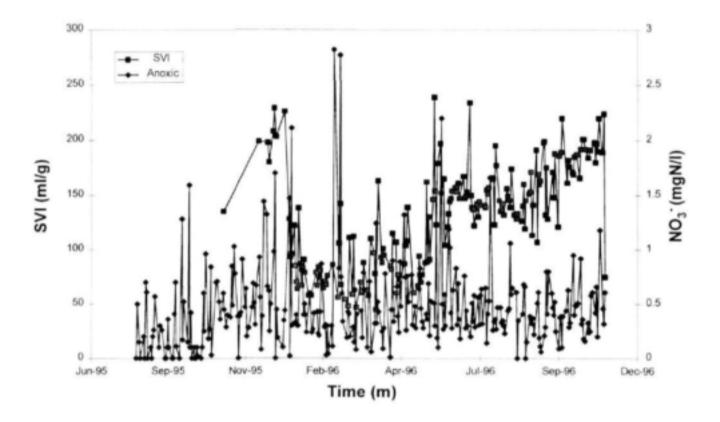


Figure 3: Vlakplaats: SVI and Nitrate in the Anoxic Zone vs Time

A plot of SVI with NO₃ concentration in the 2nd anoxic reactor is shown in Figure 3. Little information can be gained from this plot due to the excessive variation in value of the nitrate concentration. However the increase in SVI from April 1996 is clearly not caused by an increase in the nitrate concentration in the anoxic zone which remained steady at relatively low levels. This is contrary to the predictions of the hypothesis.

This data is therefore considered to be inconclusive with regard to proving the hypothesis.

Rynfield

Plots of SVI and nitrate concentration in the 2nd anoxic reactor are shown in Figures 4 and 5 for the North and South modules at Rynfield respectively. It is apparent that little information can be gained from the Northern Module data due to the high variation in the nitrate values.

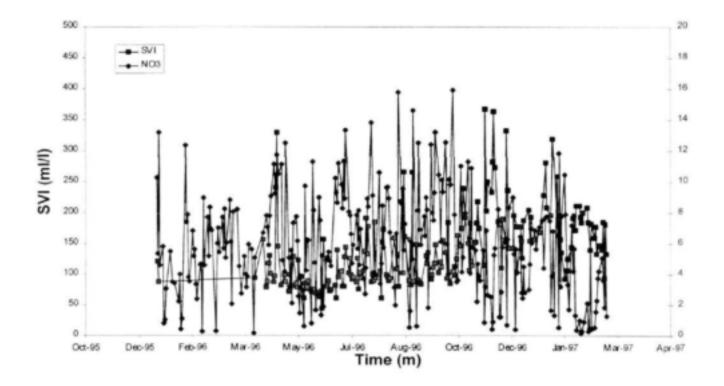


Figure 4: Rynfield North: SVI and Nitrate in the Anoxic Zone vs Time

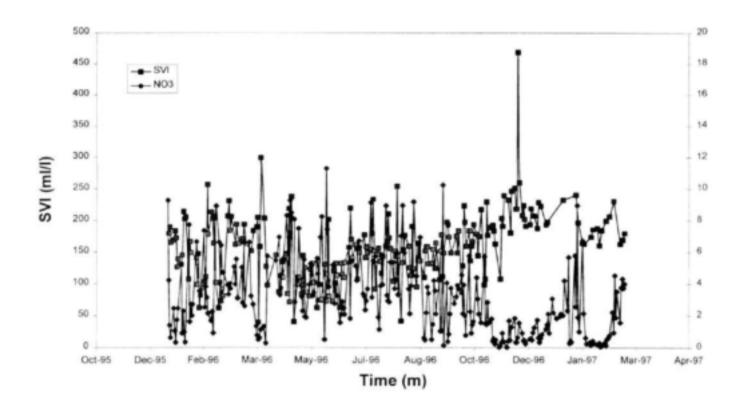


Figure 5: Rynfield South: SVI and Nitrate in the Anoxic Zone vs Time

The data for the Southern Module shows a similar trend to Vlakplaats in that the low nitrate concentrations in November and December 1996 appear to correspond to higher values of SVI which is contrary to the predictions of the hypothesis.

Daspoort

The most important parameter to compare with sludge settleability would be the concentration of NO₃ entering the aerobic zone, but this parameter is not measured as part of the routine daily analyses at Daspoort.

An analysis was however conducted into the correlation between the nitrate concentrations in the final effluent and sludge settleability at the Daspoort Works. The results of the analysis are discussed below.

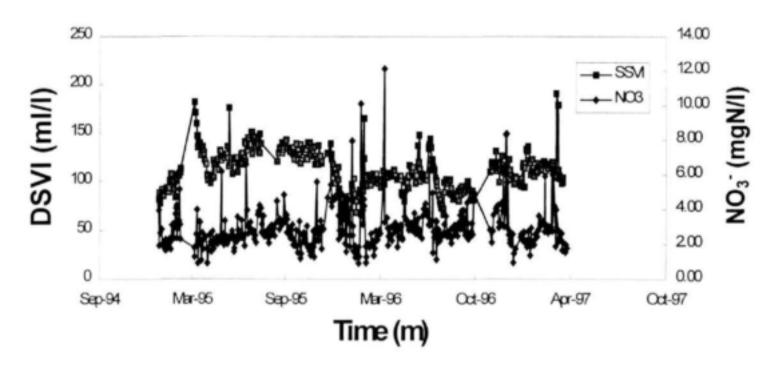


Figure 6 : Daspoort Module 9 : SVI and Effluent Nitrate vs Time



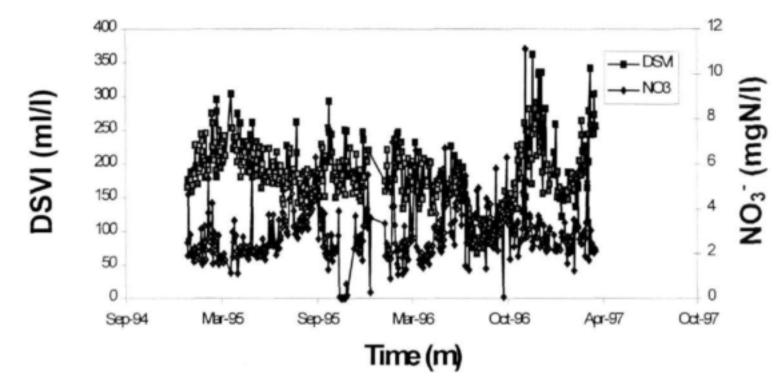


Figure 7: Daspoort Module 10: SVI and Effluent Nitrate vs Time

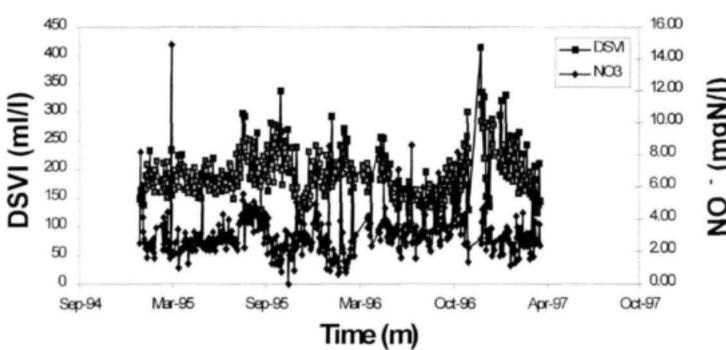


Figure 8: Daspoort Module 11: SVI and Effluent Nitrate vs Time

The Daspoort Wastewater Treatment Plant has three separate activated sludge modules which are referred to as Modules 9, 10 and 11. The process is operated as a 3-Stage Bardenpho Process. Settleability is measured using both the SVI and the DSVI parameters. Figures 6 to 8 respectively present plots of SVI and effluent NO₁ with time for modules 9 to 11. For Module 9, it is apparent that there is a definite relationship between the two parameters and the corresponding increase and decrease in SVI with increase and decrease in nitrate concentration is predicted by the AA bulking hypothesis. However, for Modules 10 and 11 it could be considered that the opposite effect is occurring, ie an increase in nitrate concentration results in a decrease in SVI and a decrease in nitrate concentration results in an increase in SVI. A similar result was noted for laboratory systems in which the aerobic mass fractions were changed from 33% (an aerobic mass fraction which has been shown to result in proliferation of AA filaments) to 65% (an aerobic mass fraction at which AA filaments do not proliferate). The concentration of nitrate in the effluent increased with increase in aerobic mass fraction due to reduced denitrification potential in the system. The historical operating protocol of the system was examined to determine if the changes in effluent NO3 concentration were due to historical changes being made to the aerobic mass fractions (eg. an additional aerator being turned on to ensure complete nitrification during winter). However this could not be established from the plant operating records, and so this data must also be regarded as inconclusive.

4.2.7 Effect of aerated/unaerated mass fractions on sludge settleability

The seven plants were evaluated to establish if there is any correlation between the aerated mass fraction of sludge within the process and sludge settleability. A problem in comparing sludge settleability in the seven plants is that each of the different operating authorities uses different parameters for measurement of sludge settleability, ie either SSVI, DSVI, or SVI. In order to compare the systems, each of the SVI and SSVI values was corrected to an equivalent DSVI value using the relationships developed by Pitman (1984) and Ekama and Marais (1986). While it is acknowledged that such corrections have inherent pitfalls, it was considered that the relationships would be suitable to at least provide indicative values of DSVI.

Table 2 is a summary of the activated sludge plant modules, the original sludge settleability data and the corrected DSVI data. The original data values are shown in regular font and the calculated values are italicised. At the Daspoort works, both SVI and DSVI were measured.

Table 2: Sludge settleability and aerobic mass fraction data for seven activated sludge nutrient removal plants in Gauteng

Plant	SSVI (m ℓ/g)	SVI (m l/g)	DSVI (mℓ/g)	Percentage Aerobic (%)	Period of Analysis
Goudkoppies			93	60	Jul 1997 - Jun 1998
Bushkoppie (Unit 1)			61	76	Jan 1998 - Dec 1998
Olifantsvlei (Unit 3)			61	58	Jan 1998 - Nov 1998
Northern Works (Unit 4)			74	63	Jan 1998 - Nov 1998
Rynfield (South)	74	156	113	50	Jan 1996 - Feb 1997
Vlakplaats (Module D)	69	132	104	54	Jan 1997 - Dec 1997
Daspoort (Module 9)		112	162	44	Jan 1995 - Mar 1997

Figure 9 is a plot of the DSVI values of the seven plants against Percentage Aerobic Mass Fraction of the configurations. It is apparent that a very strong relationship exists between the two parameters.

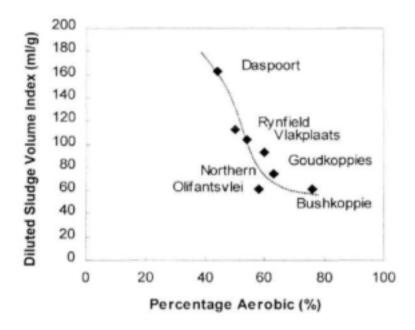


Figure 9: Plot of diluted sludge volume index with percentage aerobic mass for seven activated sludge nutrient removal plants in Gauteng

In experimental work to test the AA bulking hypothesis, Casey et al. (1993) noted a similar relationship for sludges developed in single reactor intermittently-aerated nitrogen removal systems. In that relationship, the poorest settling sludges developed when the system was operated at between 30 and 40 percent aerobic mass fraction, with improving sludge settleability values for systems with aerobic mass fractions greater and less than this range. In Figure 9, it is apparent that sludge settleability improves with increase in aerobic mass fraction, but it is not possible to comment on the effect of aerobic mass fractions less than 40 percent. No data points are shown for aerobic mass fractions less than 40 percent of the system and it is unlikely that a full-scale system would be designed or operated with an aerobic mass fraction in this range because of the implications for nitrification.

The relationship developed above can be explained on a biochemical/microbiological level by the AA bulking model and this is undertaken in Section 5 below.

4.3 Implications of the relationship between aerobic mass fraction and sludge settleability

Although the relationship between aerobic mass fraction and sludge settleability may seem simple in its application at both the design and operating stages, there are significant implications for system performance in increasing the aerobic mass fraction. Firstly, too large an aerobic mass fraction would result in incomplete denitrification, ie the denitrification potential of the anoxic zone is exceeded by the mass of nitrate to be denitrified. The consequence is that nitrate would be present in the effluent. Secondly, too high an aerobic mass fraction requires additional aeration, resulting in increased capital and operating costs. The increased operating costs are due to the increased power consumption from supply of air, but also because the nitrate is not being used as an electron acceptor for recovery of alkalinity consumed in the nitrification process.

4.4 Confirmation of the relationship between the diluted sludge volume index (DSVI) and the stirred specific volume index (SSVI)

In their 1986 paper relating sludge settleability and secondary settling tank design, Ekama and Marais established that the DSVI and SSVI relationship could be very roughly correlated by the relationship:

This is supported by Stofkoper and Trentelman (1982) who found the factor to be 0.65. In an attempt to determine the best measure of sludge settleability, the operators at Daspoort conducted daily SSVI and DSVI measurements on each of the three activated sludge modules. The data is plotted for the period Jan 1995 to Sept 1997 and is shown in Figure 10 for Module 9. Figure 10 demonstrates two outstanding features. Firstly, the plots of DSVI and SSVI follow the same trend with time, i.e. the SSVI values show similar increases and decreases with time as do the DSVI values. Secondly, the SSVI plot shows less variation between daily values than does the DSVI plot. This is a general finding from other research in the field and is discussed in Section 2.3.

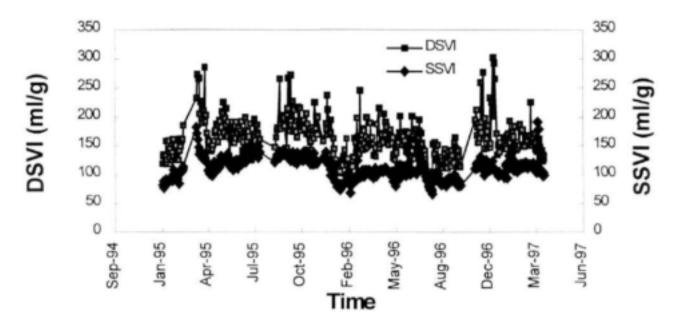


Figure 10: Daspoort Module 9: DSVI and SSVI vs Time

The ratio between SSVI and DSVI for the data shown in Figure 2 for Module 9 of the Daspoort plant is 0.69, a value similar to that given by Ekama and Marais (1986). Values of 0.68 and 0.66 were calculated for the SSVI/DSVI ratio for Modules 10 and 11 at Daspoort.

MICROBIOLOGICAL AND BIOCHEMICAL EXPLANANTION FOR THE EFFECT OF THE SIZE OF THE AEROBIC MASS FRACTION ON SLUDGE SETTLEABILITY

The objective of this section is to provide a microbiological/biochemical explanation for the role of the aerobic and anoxic zones in the competition between filamentous and floc-forming organisms. In a detailed description of the respiratory processes mediated by the two groups of organisms, Casey et al. (1993) discussed the competition between the filamentous and floc-forming organisms on a fundamental level. The objective in this section is to provide an easily understandable explanation for the manner by which the aerobic mass fraction affects the competition between filamentous and floc-forming organisms. This would provide wastewater treatment design engineers with a basis for design of the optimum aeration requirements that would ensure minimal filamentous organism growth.

5.1 The role of aerobic cytochromes and denitrification enzymes in aerobic and anoxic respiration

As a starting point, it is assumed that the organisms found in nutrient removal plants and which are referred to as AA filaments and floc-forming organisms are classified as heterotrophic facultative aerobic organisms. This classification describes their energy and carbon sources and their respiratory environment; i.e. both sets of organisms can use organic substances for energy and as a carbon source, and have the ability to utilise either nitrate or oxygen as an electron acceptor, depending on the availability of each. Although this classification has not been rigorously determined from microbiological testwork, the prolific growth of the organisms in an environment (wastewater treatment plant sludge) that alternates between anoxic and aerobic conditions would provide significant support for it.

To obtain energy to carry out metabolic processes, facultative organisms break down long chain organic molecules and pass the high-energy electrons between a number of sets of enzymes. After transfer, the energy associated with the electrons is reduced and the energy is used to produce and store a high-energy molecule (adenosine triphosphate, ATP) that can be used by the organism for cellular functions. The consequence is that the electrons become reduced in energy and need to be eliminated by the organism. Under aerobic conditions, facultative organisms use oxygen to dispose of the electrons (oxygen is referred to as the electron acceptor) and water is produced in the process. Under anoxic conditions, facultative organisms use nitrate as the electron acceptor and dinitrogen (N_2) is produced. The enzymes that transfer electrons to oxygen are known as cytochrome oxidases, i.e. cytochrome oxidase aa_3 and cytochrome oxidase o. The enzymes that transfer electrons to nitrate and then successively to each of the denitrification intermediates are known as the nitrogen oxide reductases.

Facultative organisms have the ability to switch between oxygen and nitrate as electron acceptors depending on the energetic return associated with the availability of each. Under

the aerobic conditions experienced in an activated sludge plant (DO $\approx 2 \text{ mgO}/\ell$), oxygen would be the favoured electron acceptor, but as the DO decreases below 1 mgO/ ℓ , oxygen becomes limiting, and if nitrate is available, the facultative organisms will use it as the preferred electron acceptor. This describes the essential aspects of the model for facultative heterotrophic organism respiration.

5.2 Consequences for sludge settleability of the heterotrophic organism respiration model

In summary, the model for facultative heterotrophic organism respiration, in combination with the model for AA filamentous organism bulking described in Section 3 is based on seven main tenets, viz.:

- Facultative heterotrophic organisms have sets of enzymes that allow the transfer of electrons to oxygen under aerobic conditions (cytochrome oxidases aa₃ and o) and nitrate under anoxic conditions (reductases).
- Unaerated conditions prevents the development of cytochrome oxidase aa₃ and enables the development of cytochrome oxidase o and the nitrogen oxide reductases.
- Aerated conditions prevent the development of the nitrogen oxide reductases and cytochrome oxidase o and promotes the development of cytochrome oxidase aa₃.
- Nitric oxide, an intermediate in the denitrification pathway, and produced intracelluarly by the facultative heterotrophic organisms interacts with cytochrome o (but not cytochrome aa₃) and prevents the transfer of electrons to oxygen under aerobic conditions.
- In terms of the organisms that affect settleability, activated sludge can be considered as comprising filamentous and floc-forming organisms, both of which are considered to be facultative heterotrophic organisms.
- The floc-forming organisms are hypothesised to denitrify in four steps; from nitrate (NO₃') to nitrite (NO₂'), to nitric oxide (NO), to nitrous oxide (N₂O) and finally to dinitrogen (N₂). The filamentous organisms are hypothesised to reduce nitrate to nitrite only and therefore do not produce the intermediate nitric oxide, which is inhibitory of aerobic respiration mediated by cytochrome oxidase o.
- The filamentous organisms are hypothesised to proliferate in activated sludge subjected to anoxic-aerobic sequences in which the anoxic zone is sufficiently large so that cytochrome o is the dominant oxidase, but sufficiently small such that not all of the nitrate is denitrified. Sufficiently high concentrations of nitrate during the anoxic zone or period usually would mean that nitric oxide is present in the cells of the flocforming organisms and would inhibit their aerobic respiration in the subsequent aerobic period or zone of the system. This set of conditions would result in the proliferation of filamentous organisms.

5.3 Effect of aerated and unaerated periods on the development of respiratory cytochromes and enzymes

It is assumed that in an activated sludge system that is operated as a completely aerobic process, organisms will develop that use oxygen only as electron acceptor. This was confirmed indirectly by Casey et al. (1993) who developed sludges in laboratory-scale systems under both completely aerated and completely unaerated conditions. The sludges developed under aerated conditions had very poor denitrification rates and the sludges developed under non-aerated conditions had high denitrification rates. When a sludge that was developed under completely aerobic conditions was changed to completely anoxic conditions, the sludge developed a high denitrification rate over a period of a sludge age. It can be concluded that either (1) the organisms that had developed under the completely aerobic conditions had been outcompeted under the completely anoxic conditions, or (2) the organisms that had developed under completely aerobic conditions had few denitrification enzymes, but developed them with time under the completely anoxic conditions. In many respects it is immaterial which of these events occurred because the entire heterotrophic sludge mass can be conceptualised as two organisms, a floc-forming mass and a filamentous mass each of which changes characteristics according to the environmental conditions to which it is exposed. Conversely, sludge developed under anoxic conditions demonstrated no reduction in oxygen uptake when exposed to aerobic conditions.

Therefore it can be concluded that aerated conditions adversely affect the development of the denitrification enzymes, but anoxic conditions do not adversely affect the development of the aerobic enzymes. In their literature review Casey et al. (1993) found that the above is not strictly true. The aerobic enzymes that develop under aerobic conditions (cytochrome aa₃) degrade with time under anoxic conditions, but another set of aerobic enzymes (cytochrome o) develop under anoxic conditions and allows the organisms to respire aerobically if they are exposed to oxygen after an extensive anoxic period.

A problem with cytochrome o that does not occur with cytochrome aa3 is that it becomes inactive when exposed to the intermediate products of denitrification, i.e. nitric oxide. Therefore under conditions in which the organism is switched from anoxic to aerobic conditions, nitric oxide that remains inside the organism from the denitrification of nitrate can interact with the cytochrome o and inhibit its transfer of electrons to oxygen. As described above, exposure of the organism to anoxic conditions results in degradation of the aerobic enzyme cytochrome aa3 and therefore it is only partially active during subsequent aerobic conditions. It should be remembered that although it is hypothesised that both the filaments and floc-formers produce cytochromes o and aa3, only the floc-formers would be inhibited under these conditions, because it is assumed that the filamentous organisms do not produce nitric oxide. Therefore it can be expected that as the length of exposure of the facultative organisms to anoxic conditions decreases, the extent of degradation of cytochrome aa₁ would decrease and therefore inhibition of aerobic respiration by the floc-formers would also decrease and they will become more competitive. This is what is demonstrated in Figure 9, which indicates that the sludge settleability improves with increase in system aerobic mass fraction.

CONCLUSIONS AND RECOMMENDATIONS

From a survey of seven nutrient removal activated sludge plants, it can be concluded that:

- (1) The filaments that dominate in the plants are those that are classified as AA to which the AA bulking hypothesis applies.
- (2) The Daspoort data confirmed the relationship between SSVI and DSVI as SSVI = 0.69 DSVI.
- (3) A major aspect has been identified that impacts on the proliferation of filamentous organism proliferation in nutrient removal activated sludge plants. In comparing the aerobic mass fractions and sludge settleabilities of seven nutrient removal activated sludge plants, it was found that generally, the higher the aerobic mass fraction of the system, the better the sludge settleability.
- (4) The hypothesis developed by Casey et al (1993) to explain filamentous organisms proliferation is supported by the finding described in (1) above.
- (5) Design engineers should incorporate flexibility into nutrient removal plants to increase the aerobic mass fraction of the process without compromising nitrogen removal, in order to minimise filamentous organism proliferation.

It is recommended that:

- (1) A study be conducted to compare (i) the capital and operating costs of increasing the aerobic mass fraction of the system through incorporation of additional aeration with (ii) the saving associated with reduced secondary settling tank surface area, based on the information found in this project.
- (2) Studies be conducted to determine a number of other facets of the AA bulking hypothesis at full scale, such as the effect on sludge settleability of changes in the influent TKN/COD ratio and the concentration of nitrate at the end of the anoxic zone.

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