Filamentous organism bulking in nutrient removal activated sludge systems. Paper 6: Review, evaluation and consolidation of results

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Abstract

The finding that the selector effect did not control bulking by low food to micro-organism ratio (F/M) filaments, concluded this research direction that was considered to hold promise for controlling low F/M filament proliferation, and placed this research back into an exploratory phase. In this paper, the information collected so far in the research programme is evaluated together with that published in the literature, in order to delineate new research directions aimed at solving the low F/M filament bulking problem. In the conclusions, a framework is established that provided guidance for the subsequent research.

List of symbols

COD	=	chemical oxygen demand
d	=	day
DO	=	dissolved oxygen (mg O/ℓ)
DSVI	=	diluted sludge volume index
F/M	=	food to micro-organism ratio
h	=	hour
K_s	=	half saturation coefficient (mgCOD/ℓ)
min	=	minute
MUCT	=	modified UCT
N	=	nitrogen
P	=	phosphorus
RBCOD	=	readily biodegradable COD
SBCOD	=	slowly biodegradable COD
TSS	=	total suspended solids
UCT	=	University of Cape Town
VFA	=	volatile fatty acids
μ_H	=	maximum specific growth rate of heterotrophs (/d)
µm	=	micro (10^{-6}) meter

Introduction

In the experimental research programme two approaches to bulking control in N and N & P removal systems were adopted: non-specific and specific. The non-specific control approach was evaluated because this approach is a useful emergency/temporary control measure, which can be quickly implemented and has a rapid effect. By following the procedure set out by Jenkins et al. (1984), chlorination was found to be successful for controlling the low F/M filament types 0092, *Microthrix parvicella* and type 0914 (Lakay et al., 1988). The DSVI was reduced from 230 mℓ/g to 48 mℓ/g over a period of 19 d and the biological N & P removal were not significantly adversely affected even at fairly high chlorine dosage rates [8 g Cl/(kg TSS·d)].

While successful, the problem with non-specific control methods is that they treat temporarily the symptoms of bulking but do not constitute a permanent cure - after dosing ceases the filaments inexorably regrow. With specific bulking control, the causes for the proliferation of the filaments are sought. By eliminating these through waste-water characteristic or system modification, the bulking problems caused by specific filamentous organism types are cured permanently.

Surveys of South African N and N & P removal plants indicated that the six most frequently dominant filaments are 0092, 0675, 0041, *M. parvicella*, 0914 and 1851 (Blackbeard et al., 1986; 1988). Four of these six filaments sort into the low F/M (or long sludge age) group of filaments. Interestingly although frequently dominant in laboratory-scale systems (Gabb et al., 1989) *Sphaerotilus natans* was not, and *Thiothrix* only rarely, identified as a dominant filament in South African full-scale plants.

Specific bulking control

The promoted specific bulking control method for low F/M filaments is system configuration modification so as to incorporate alternating or sequential feed-starve conditions into the system such as intermittent (batch) feeding; multi-reactor or plug flow conditions; or completely mixed systems including selector reactors (Jenkins et al., 1984). In the literature, it has been hypothesised that the mechanism whereby these systems apparently effect control over the low F/M filaments is that under high readily biodegradable COD (RBCOD) concentrations the floc formers have, or develop, a higher rate of RBCOD utilisation than the filamentous organisms and hence preferentially remove available RBCOD from the filamentous organisms. A sludge in which a high RBCOD uptake rate has been stimulated, is said to have acquired a "selector effect". This mechanism for controlling the low F/M filaments, and the systems in which they develop was investigated and described in Still et al. (1996); Ekama et al. (1996); and Gabb et al. (1996a; b). In this paper the conclusions and implications that emerged from this work are summarised and compared with other research conducted elsewhere.

1 Selector effect is stimulated under alternating feed-starve conditions under aerobic or anoxic conditions

The alternating feed-starve conditions imposed by intermittent feeding (once daily) to completely mixed reactor systems (batch fed), either fully aerobic or anoxic-aerobic; and aerobic selector reactors incorporated in continuously fed completely mixed systems,

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Received 10 March 1995; accepted in revised form 15 January 1996.

either with fully aerobic or intermittently aerated main reactors, stimulated in the mixed liquor a high readily biodegradable (or dissolved, <0.45 µm filtered) COD (RBCOD) uptake rate. The RBCOD uptake rate was 1.5 to 3 times higher than that in systems which did not have alternating feed-starve conditions, such as continuously fed completely mixed systems without selectors. If the conditions during which the RBCOD was taken up rapidly were aerobic, the high RBCOD uptake rate gave rise to an associated high initial oxygen uptake rate under batch conditions; if the conditions were anoxic, it gave rise to an associated high (initial) nitrate uptake rate under batch conditions.

The selector effect in a sludge could be stimulated (or lost) over a period less than a sludge age in long sludge age (> 20 d) systems by introducing (or eliminating) alternating feed-starve conditions. Acquisition of a selector effect by a sludge under alternating feed-starve conditions is in agreement with reported results in the literature (Still et al., 1996; Ekama et al., 1996).

2 Purely aerobic conditions apppear to ameliorate bulking by low F/M filaments

Low F/M filament bulking sludges (diluted sludge volume index, DSVI > 250 m*l*/g) containing, usually, in varying proportions 0092, *M. parvicella*, 0914, 0675, 1851 and 0041 filaments), from long sludge age full-scale (N removal) plants or laboratory scale N & P removal plants, when used to start up laboratory-scale long sludge age (> 15 d) activated sludge systems under fully aerobic conditions, invariably ceased bulking (DSVI < 80 m*l*/g) within a month whether or not the system stimulated a selector effect (Still et al., 1996; Ekama et al., 1996: Gabb et al., 1996a; b). Evidently, in long sludge age fully aerobic systems, the selector effect was irrelevant because the low F/M filament growth was suppressed both when the selector effect was present or absent.

3 Bulking caused by *Sphaerotilus natans*

In the fully aerobic, long sludge age (low F/M) systems, in which there was no selector effect, when bulking was observed it was not due to proliferation of low F/M filaments but due to *S. natans* and *Thiothrix* (Still et al., 1996; Ekama et al.,1996; Gabb et al.,1996a). According to Jenkins et al. (1984) *S. natans* sorts into the low DO group and *Thiothrix* into septic sewage or nutrient deficient groups. *S. natans* has not, and *Thiothrix* only rarely, been observed to cause bulking in full-scale long sludge age plants in South Africa.

4 *S. natans* bulking caused by seeding

While raising the DO concentration did control the *S. natans* bulking once (Still et al., 1996), regular and thorough cleaning of the influent feed lines eliminated the *S. natans* bulking problems in the laboratory systems every time (Still et al., 1996; Gabb et al., 1996a). From this it was concluded that *S. natans* proliferation in the laboratory systems was caused by seeding from *S. natans* attached growth on the influent feed line walls. This artifact may also have been present in many laboratory-scale studies throughout the world because numerous investigators have reported the proliferation of *S. natans* in their laboratory systems under a wide range of operating conditions (Gabb et al., 1989).

5 Selector effect controls *S. natans* and *Thiothrix*

Aerobic selectors and intermittent (batch) feeding conditions, which induce the selector effect, controlled the proliferation of *S. natans* and *Thiothrix*. This finding is in conformity with results reported in the literature. The success of the selector effect in controlling bulking by *S. natans* and *Thiothrix* in laboratory-scale low F/M *systems* possibly contributed to the notion that the selector effect also controls low F/M filament proliferation (Still et al., 1996).

According to the literature, control of *S. natans* growth by the selector effect takes place by competitive removal of soluble biodegradable COD (RBCOD) by floc formers. The selector effect results in the selection of floc formers with high RBCOD uptake rates, or adaptation of existing floc formers to acquire a high RBCOD uptake rate. The specific substrate uptake rate of *S. natans* is unlikely to change appreciably as it does not possess the diversity of the floc former population. Consequently the selected floc formers out-compete *S. natans* thereby effectively limiting substrate acquisition by *S. natans*, and curtailing their proliferation in the mixed liquor, a phenomenon known as kinetic selection (Wanner et al., 1987a; b). The acquisition of a selector effect by the sludge, manifested in terms of Chudoba's et al. (1973; 1974) selection criterion as an upward shift in the Monod curve of the floc-formers, is not due to an elimination of the slower growing filaments - a sludge already settling well (few filaments) when transferred from an aerobic completely mixed continuously fed system (no selector effect) to an aerobic intermittently fed fill and draw system, was found to acquire a selector effect without change in sludge settleability.

The acquisition of a selector effect by the sludge is not recognised by Chudoba's et al. (1973; 1974) selection criterion indicating that it requires modification. Instead of the Monod curves for floc-formers and filaments crossing over each other (see Fig. 1, Casey et al., 1995), Ekama and Marais (1986) suggested that with acquisition of the selector effect, the floc-former's Monod curve moves upward (higher μ_H and K_s) so that it lies above the filament's Monod curve even at very low soluble substrate concentrations. However, even this modified selection criterion cannot explain why low F/M filament bulking sludges invariably improved in sludge settleability (under fully aerobic conditions) in the absence of a selector effect.

6 Low F/M filaments proliferated in nutrient removal systems but not *S. natans* and *Thiothrix*

From 2 above, low F/M filaments did not proliferate in the laboratory-scale fully aerobic systems. In contrast, in N and N & P nutrient removal systems (i.e. systems with anoxic-aerobic or anaerobic-anoxic-aerobic zones which follow sequentially in usually single or multi reactors in series and incorporating appreciable unaerated sludge mass fractions ~ 50%) at full-scale, low F/M filaments proliferate and cause bulking problems. Indeed, of the laboratory systems operated in a parallel investigation to those of this one, the N & P removal systems were the only ones wherein the low F/M filaments proliferated (N removal systems were not operated). In these systems, *S. natans* did not, and *Thiothrix* only rarely[1] caused bulking, even when the feed lines were not regularly

[1] On one occasion when *Thiothrix* (I and II) caused bulking (DSVI ~ 150 m*l*/g) it was in a modified UCT system receiving sewage that was first passed through an acid fermentation reactor; the acid fermentation significantly increased the sulphide concentration of the influent and probably was the cause of the proliferation of the sulphur reducing *Thiothrix* filaments. Low F/M filaments were present also (0914, 0092, 0041, 0675) but were secondary in comparison to the *Thiothrix* spp. (Bagg et al., 1985) (Also see footnote 2 on page 149).

cleaned. This is in conformity with the full-scale plant filament surveys which showed that *S. natans* does not, and *Thiothrix* only rarely causes bulking.

7 Anaerobic reactors appear to ameliorate or totally suppress *S. natans* (and *Thiothrix*) bulking

From the absence of *S. natans* and *Thiothrix* in N & P removal systems, it would appear that the anaerobic reactor, which receives the influent flow, operates as a kind of selector reactor against *S. natans* (and possibly also *Thiothrix* and 021N) proliferation. This finds support from the laboratory experiments of Wanner et al. (1987a; b) who call this type of selection metabolic selection (as distinct from kinetic selection). This type of selection operates as follows: *S. natans* is an obligate aerobe (Mulder and Deinema, 1981) and only capable of metabolism in the aerobic reactor; in the anoxic reactor, the RBCOD is utilised by denitrifiers; in the anaerobic reactor, RBCOD is converted to VFA which together with the VFA from the influent, is taken up by polyphosphate accumulating organisms such as *Acinetobacter* spp. (Wentzel et al., 1985). Consequently in systems with anaerobic and/or anoxic reactors ahead of the aerobic reactor, very little RBCOD enters the aerobic reactor (Clayton et al., 1991) for growth of *S. natans*. In terms of this explanation, selectors, whether aerobic, anoxic or anaerobic, control *S. natans* proliferation by either removing RBCOD under conditions in which *S. natans* cannot function (anaerobic or anoxic reactors, i.e. metabolic selection) or stimulating high RBCOD uptake rates in floc-formers which then can compete successfully against *S. natans* (aerobic selectors, i.e. kinetic selection).

With regard to *Thiothrix* and 021N, these organisms are variously reported as obligate aerobic or facultative: If they are obligate aerobic, their proliferation is controlled in the same two ways as *S. natans* described above. If they are facultative, anaerobic reactors, and anoxic and aerobic selectors (though not anoxic reactors) should control their proliferation. The literature supports this conclusion; *Thiothrix* is controlled by anaerobic reactors (Wanner et al., 1987b), anoxic selectors (Shao, 1986; Shao and Jenkins, 1989) and aerobic selectors (Van Niekerk, 1985, Van Niekerk et al., 1988).

8 Evaluation of laboratory system artifacts possibly influences system behaviour

The appearance of *S. natans* in the laboratory systems as a consequence of seeding from the effluent feed line walls created the concern that other influences inadvertently incorporated in the laboratory-scale systems were possibly causing the decline in the DSVI and low F/M filaments in the laboratory systems started up with full-scale system sludge (see 2 above). To test this, three single reactor systems were started up with a low F/M filament bulking sludge from a laboratory N & P (Modified UCT) removal system and were fed the same waste water and operated at the same sludge age. Two of the systems were intermittently fed (once daily) while the third was continuously fed. One of the intermittently fed systems was anaerobic for the first 6 h after feeding and aerobic for 16 h, and finally settling for 2 h. The other intermittently fed system, and the continuously fed system, were maintained fully aerobic for 24 h. In the two fully aerobic systems (one intermittently fed, the other continuously fed) the DSVI declined steadily from a start-up value of around 200 to below 60 m*l*/g over a period of 2 to 3 sludge ages. Over the same period, the DSVI in the intermittently fed anaerobic/aerobic system and in the parent Modified UCT system remained high between 180 and 200 m*l*/g (Gabb et al., 1996a).

These experiments indicated that inadvertent laboratory system artifacts in all likelihood were not influencing the system behaviour because the same results as previously with full-scale N removal sludge were obtained, i.e. continuous aeration inhibits the growth of most of the low F/M filaments, in particular *M. parvicella*, 0092 and 0914 irrespective of whether the mixing regime is plug flow (intermittently fed) or completely mixed (continuously fed).

9 Selectors as a means of ameliorating bulking by low F/M filaments falls into question

Despite the promotion of selectors (also sometimes called contact zones) as a means of controlling filamentous bulking (including low F/M filaments), there is little unambiguous evidence of this for the low F/M filaments. Wheeler et al. (1983) describe a case where at full scale, bulking by type 0041 was ameliorated with an aerobic selector. Canler and Pujol (1993), Pujol and Canler (1994), and Kruit et al. (1994) cite cases where selectors have improved sludge settleability. However these improvements may not have been due to the selector effect *per se* but due to introducing additional aeration via the installed selector making the plant more aerobic. There are too many cases where selectors have not worked successfully in particular on anoxic/aerobic plants, to be sure that kinetic selection controls bulking by low F/M filaments (Eikelboom, 1994; Foot et al., 1994). Furthermore, because low F/M filaments proliferate in N & P removal systems, it is clear that metabolic selection certainly is unable to control proliferation of these filaments. As metabolic and kinetic selection are both based on the same principle, i.e. removal of RBCOD by floc formers at the expense of the filaments the claim[2] that kinetic selection, i.e. aerobic/anoxic selectors, controls low F/M filaments fell into question. Consequently developing laboratory systems in which low F/M filaments proliferate, other than N & P removal ones, became a matter of high priority. To do this attention was focused on N removal systems (in which sludge is exposed to alternating anoxic and aerobic conditions) because it was evident that low F/M filaments proliferated not only in full-scale N & P removal systems but also in full-scale N removal systems.

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[2] Albertson (1987), suggests that bulking in many South African nutrient removal systems arises because the anaerobic, anoxic and aerobic zones in these systems are in the form of single completely mixed reactors; if these zones were compartmentalised, bulking would be controlled because F/M gradients would be imposed in the various zones. He does not qualify which filaments are likely to be controlled by this method, but, from experimental evidence on laboratory-scale modified UCT systems, this approach is unlikely to be successful for controlling bulking by low F/M filaments: When the anaerobic zone with a mass fraction of 0.16 was compartmentalised into 4 equal sized completely mixed reactors, the DSVI increased steadily from 88 m*l*/g to 164 m*l*/g over a period of 3 sludge ages, caused by the proliferation of low F/M filaments 0092, 0914, *M. parvicella*, 0675, (Bagg et al., 1985); when the first anoxic zone with a mass fraction of 0.14 was in the form of a plug flow reactor, DSVIs over 200 m*l*/g were obtained for periods well in excess of 5 sludge ages, caused by the low F/M filaments 0092, 0041, *M. parvicella* and 0675 (Clayton et al., 1991). In the Kelowna plant (Canada), which is a completely compartmentalised nutrient removal system, DSVIs in the range of 200 m*l*/g are obtained. As Albertson suggests, Kelowna's high DSVIs may be due to the practice of acid fermenting primary sludge to augment biological P removal; if septic sewage filaments like *Thiothrix*, 021N or *Beggiatoa* cause the bulking, then this is an acceptable explanation, but if low F/M filaments cause the bulking it would mean that compartmentalisation does not control low F/M filaments.

10 Low F/M filaments proliferate in intermittent aeration systems

In an attempt to grow low F/M filaments in laboratory systems other than nutrient (N & P) removal ones, long sludge age single reactor continuously fed completely mixed systems with intermittent aeration (1 min air on, in a 10 min cycle with peak DO of 2.0 mg/ℓ) were set up to mimic Carousel and Orbal type plants. It was found that in such systems most of the low F/M filaments proliferated (DSVI > 300 mℓ/g), in particular *M. parvicella* and 0092 but also 0914, 0041, 0675 and 1851. In two such systems, switching one from intermittent aeration to continuous aeration caused a sharp decline in bulking (DSVI < 80 mℓ/g in under one sludge age) with a concomitant reduction in low F/M filaments while the other sustained low F/M filament proliferation. Switching back to intermittent aeration caused slow regrowth (3 to 4 sludge ages) of the low F/M filaments (DSVI > 300 mℓ/g). The experiment was repeated using the same two systems but with interchanged roles, the experimental becoming the control, and *vice versa*.

11 Aerobic selectors unable to control low F/M filament bulking

Having established that low F/M filaments proliferate in laboratory intermittent aeration systems as in similar ditch type full-scale plants, it became possible to check, by setting up an experimental and a control system, whether or not aerobic selectors control low F/M filaments. With a correctly sized selector installed on the experimental system it was found that the selector effect did not control most of the low F/M filaments; for more than 5 sludge ages the DSVI in both the control and the experimental systems remained above 250 mℓ/g. Switching the control system to continuous aeration (DO between 2 and 4 mg O/ℓ) caused the DSVI to decrease in less than a sludge age, with a concomitant decline in low F/M filaments, while the DSVI in the experimental (selector) system remained high.

12 Consistency of behaviour of anaerobic reactors and aerobic selectors

Having observed that aerobic selectors do not control bulking by low F/M filaments resolves the apparent inconsistency in behaviour of anaerobic reactors (metabolic selection) and aerobic selectors (kinetic selection). Neither anaerobic reactors, nor aerobic selectors (and anoxic selectors presumably also), and the preferential RBCOD removal by floc formers these stimulate, control bulking by low F/M filaments. Because removal of RBCOD by floc formers does not control low F/M filament proliferation, it would appear that the influent readily biodegradable COD does not play an important role in controlling bulking by low F/M filaments.

13 Low F/M filaments appear more sensitive to alternating unaerated/aerated conditions than to kinetic or metabolic selection pressures

From (6) and (11) above it would appear that many of the low F/M filaments proliferate in N & P removal systems, and continuously fed completely mixed systems with intermittent aeration whether or not these systems incorporate metabolic or kinetic selection. When these systems are made purely aerobic by continuous aeration, the filaments are reduced and low F/M filament bulking is ameliorated. It would appear that the presence of unaerated conditions is conducive to low F/M filament proliferation irrespective of the presence or not of metabolic or kinetic selection. However, factors to which no answer can be given at this stage are the effects of magnitude of unaerated mass fraction, retention time (actual or nominal) under unaerated conditions, the duration or frequency of the anoxic-aerobic cycles in intermittent aeration systems, and the concentrations of nitrate and DO during the anoxic and aerobic periods respectively.

Considerations for further research

The conclusions presented above provide convincing evidence that kinetic and metabolic selection do not control low F/M filament proliferation in intermittent aeration systems (like Carousel and Orbal plants) for N removal and multi-reactor anaerobic-anoxic-aerobic systems (like UCT and Bardenpho systems) for N & P removal. Recent full-scale experience in a number of European countries supports this conclusion [e.g. Kunst and Reins, 1994; Foot et al., 1994; Eikelboom, 1994; Ekama et al. (In prep)]. It would appear that the environmental conditions created for biological N removal, (the common feature of N and N & P removal systems) i.e. sequential unaerated-aerated zones, are conducive to proliferation of low F/M filaments; completely aerated systems cure bulking by low F/M filaments, but also preclude biological N removal. Consequently to effect specific control over the low F/M filaments, some environmental condition needs to be found that will lead to exclusion of the filaments but retention of the organisms effecting biological N (and P) removal. Research resulting in identification of these conditions is presented in Lakay et al. (In prep, a; b); Casey et al. (In prep, a; b; c).

The finding that kinetic and metabolic selection does not control bulking by low F/M filaments places research in this field back into an exploratory stage. As a consequence, the endeavour of further research will be to establish a framework for understanding the fundamental causes of low F/M filament bulking from which specific control methods can be devised. Taking due consideration of the conclusions drawn so far, the specific experiments of this endeavour would entail investigating the role or effect of the following on low F/M filament bulking:

- **Particulate SBCOD**: Because the RBCOD does not appear to play an important role in low F/M filament proliferation, it is expected that the SBCOD does play an important role because this COD is not significantly reduced by kinetic and metabolic selection. The effect of SBCOD can be examined in two ways: by developing a artificial waste water with defined RBCOD and SBCOD composition; and separating real sewage into its soluble and particulate components by membrane filtration. If it is found that SBCOD does influence low F/M filament proliferation, it should be noted that the influent is not the only source of this COD; in terms of the activated sludge kinetic models (Dold et al., 1980; 1991; Henze et al., 1987), it is also derived from organism death and lysis (Ekama and Marais, 1986).
- **Sludge age**: Determine minimum sludge age for low F/M filament proliferation and whether or not efficient biological nutrient removal is possible at this sludge age.
- **Magnitude of the aerobic mass fraction**: Because fully aerobic conditions and large aerobic mass fractions (> 75%) appear to ameliorate low F/M filament bulking, determine the aerobic mass fractions at which low F/M filament proliferation becomes significant. Do these filaments proliferate under fully anoxic conditions or do they require alternating anoxic-aerobic conditions?

- **Different ways of establishing anoxic-aerobic conditions**: Alternating anoxic-aerobic conditions in single reactor intermittent aeration and multi-reactor systems are different: In the former the frequency of alternation between anoxic and aerobic conditions is an order of magnitude greater and periods of low DO concentration between the aerobic and anoxic phases are much longer than in the latter.
- **Nitrate and DO concentrations**: Investigate the influence of the nitrate concentration during the anoxic period (or zone) and the DO concentration in the aerobic period (or zone).

Experimental work examining these aspects is described in Lakay et al. (In prep, a;b).

Acknowledgements

This research was financially supported by the Water Research Commission, the Foundation for Research Development and the University of Cape Town and is published with their permission.

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