# **RECENT DEVELOPMENTS IN BIOLOGICAL NUTRIENT REMOVAL**

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# ABSTRACT

Biological nitrogen (N) and phosphorus (P) removal from municipal wastewater with the activated sludge (AS) system has been the preferred technology for the last 40 years. While several questions remain to be answered for more consistent, reliable and stable performance for enhanced biological P removal (EBPR), recent developments in this technology have focused on (i) increasing capacity and reducing plant space footprint and (ii) improving N removal. To increase capacity and reduce AS system space, (a) integrated fixed-film activated sludge (IFAS), (b) external nitrification, (b) membrane and (c) aerobic granulation BNR systems have been developed. With IFAS, fixed media are added to the aerobic activated sludge reactor to make nitrification independent of the suspended AS sludge age. With external nitrification, nitrification is achieved in a side-stream fixed media reactor, which removes the size defining nitrification process from the suspended AS system and halves its sludge age, improves sludge settleability and increases capacity. With membranes, secondary settling tanks are replaced with in-reactor membranes for solid-liquid separation. With aerobic granulation, the activated sludge is controlled to form fast settling granules comprising heterotrophs, nitrifiers, denitrifiers and phosphorus accumulating organisms (PAOs) in a sequencing batch (SBR) type reactor - the granules not only settle fast but also the SBR type operation removes the need for secondary settling tanks allowing higher reactor solids concentrations and hence smaller reactors. To improve N removal methods are being developed to (i) short-circuit nitrification-denitrification (ND) by preventing nitrate formation and enforcing ND over nitrite - this requires less oxygen and organics than ND over nitrate allowing lower N concentrations to be achieved for the same influent organics concentration and oxygen supply, and (ii) encouraging the growth of Anammox bacteria in the activated sludge which remove N autotrophically by combining ammonia and nitrite to form nitrogen gas - this halves oxygen demand for nitrification and requires no organics. These recent developments in BNR technology are briefly reviewed in this paper.

# **1. INTRODUCTION**

The size, footprint and energy consumption of the activated sludge (AS) system is governed by the requirement of the system to remove nitrogen - if nitrogen does not need to be removed by nitrification-denitrification (ND), for example when 100% source separation of urine is practised, the AS system could be much smaller and consume much less energy (1). The sludge age of the biological nutrient removal (BNR) AS system is governed by the slowest growing organisms in the system (2). When nitrogen removal is required, these are the autotrophic nitrifiers, which nitrify ammonia to nitrate. So the sludge age of the BNRAS system needs to be longer than the minimum required for the nitrifiers to be sustained in it. Furthermore, the effluent ammonia concentration, being a dissolved constituent is strongly affected by influent ammonia cyclic flow and load conditions. The longer the sludge age is beyond the minimum from nitrification, the greater is the attenuation in effluent ammonia concentration relative to influent ammonia cyclic flow and load variation (3).

Once the sludge is selected to ensure efficient nitrification, the influent organic (COD) and inorganic (ISS) loads fix the mass of sludge (TSS) in the biological reactor and the oxygen demand. The longer the sludge age, the greater the mass of sludge in the reactor and the higher the oxygen demand (4). The volume of the reactor and the surface area of the secondary settling tanks are then determined by selecting the reactor TSS concentration that minimizes the combined cost of biological reactor and secondary settling tank for a selected sludge settleability (5). While the nitrifiers add to the total oxygen demand by the system, they have a negligible effect on the reactor TSS concentration - the nitrifier biomass makes up less than 2% of the reactor TSS (3). So the impact of the nitrifiers is that they dictate the sludge age and thereafter the organic removal and sludge settleability dictate the AS system size. If nitrification can be achieved at shorter sludge ages and the solid-liquid separation be made less sensitive to sludge settleability, then the BNRAS system can be significantly reduced in size (or capacity increased for an existing system). The developments in BNR technology over the past 30 years have all focused on looking for different ways of getting around these two issues and have resulted in some remarkable discoveries and inventions, some of which are still on-going today. For example, (i) integrated fixed-film activated sludge (IFAS), (ii) external nitrification (iii) membrane (MBR) solid liquid separation and (iv) aerobic granulation BNR systems have been developed. To improve N removal, methods are being developed to (v) enhance the nitrite shunt, which "short-circuits" ND by suppressing nitrate formation and forcing ND over nitrite and (vi) encourage the growth in the BNR reactor of Anammox bacteria, which remove N autotrophically by combining ammonia and nitrite to form nitrogen gas. These six inventions and developments have been made for non-saline water "aerobic" activated sludge systems and are briefly described in this paper. Other novel inventions and developments, such as the SANI system for saline seawater treatment arising from seawater toilet flushing, which offers major reductions sludge production and oxygen demand (6), are not discussed. No doubt, many more discoveries and inventions for fresh and saline wastewater treatment still will be made and developed in future.

# 2. INTEGRATED FIXED-FILM ACTIVATED SLUDGE (IFAS) SYSTEMS

To reduce the sludge age required for nitrification, static or moving fixed media such as solid AccuFAS<sup>TM</sup> or Bio-Blok<sup>TM</sup>, suspended rope Ringlace<sup>TM</sup> or moving bed Kaldness<sup>TM</sup> carriers are added to the aerobic reactor (7), (8), (9). Such systems are called integrated fixed-film activated sludge (IFAS). The nitrifiers grow on the fixed media establishing a population permanently resident in the aerobic reactor. These nitrifiers are not subject to either the unaerated sludge mass fraction (fraction of sludge mass in reactor not aerated) or the suspended mixed liquor sludge age so that the system sludge age can be reduced. Such a reduction in system sludge age is particularly beneficial for low temperature wastewaters (10-15°C).

The overall objective of IFAS is to increase the treatment capacity and nitrification performance of the existing suspended AS system by adding fixed media to it instead of extending the plant with additional reactors. Fixed film systems are well known for biological treatment of ammonia (and dissolved organics), particularly in cold climates because the biomass mediating the bioprocesses on the fixed media are retained in the system and not removed via the waste sludge. Hence, during the cold wastewater temperature operation, the majority of the ammonia oxidizing bacteria is found on the media and good nitrification performance is maintained by the system even though the system (suspended AS) sludge age of the system is shorter than the minimum required for nitrification.

Generally, the static media are placed above the bubble aeration system so that the bulk liquid can make its way through the media providing contact of the wastewater constituents with the biomass on media. Free floating media (like Kaldness) are generally small plastic buoyant media which are placed in a reactor and move freely throughout the entire aeration basin volume. Since these media move freely in the reactor, screens are required to retain them in the reactor so that they do not escape with the effluent. The approach velocity of the media to the screens is very important and must be sufficiently low to prevent them accumulating on the screens and keep them moving around the reactor.

There are several fullscale IFAS systems in operation. The suspended medium solids retention time (SRT or sludge age) versus wastewater temperature of these plants are shown in Figure 1 (10). All of these plants are operating well below the minimum suspended medium SRT for nitrification recommended by the ATV (Abwasser Technischen Vereinigung) 131 guideline (blue line). The red line in Figure 1 represents the average nitrification performance of these IFAS plants. If these plants were conventional suspended medium AS systems, then the maximum specific growth rate of nitrifiers at 20°C ( $\mu_{Am20}$ ) and temperature sensitivity coefficient ( $\theta_{\mu}$ ) in the minimum sludge age for nitrification ( $R_{sm}$ ) equation  $R_{sm}=1/\{\mu_{Am20} (\theta_{\mu})^{(T-20)} - b_{A20}(1.03)^{(T-20)}\}$  that best fits the red line in Figure 1 are  $\mu_{Am20} = 1.10$  /d and  $\theta_{\mu} = 1.143$ . This temperature sensitivity is quite close to that used for nitrification in suspended medium AS, i.e.  $\theta_{\mu} = 1.123$  (3), but the  $\mu_{Am20}$  of 1.10 /d is much higher, at least double that used for nitrification in suspended medium AS (the ATV blue line in Figure 2 has best  $\mu_{Am20} = 0.545$  /d and  $\theta_u = 1.148$ ). This indicates that the fixed media have at least halved the minimum system sludge age for nitrification, which makes a significant volume saving for the activated sludge reactor. Although the DO concentration in IFAS reactors is required to be high (5-6 mgO/l) for effluent ammonia concentration below 0.5 mgN/l, the oxygen transfer rate is increased by the presence of the fixed media which offsets some of the aeration energy required by the high DO.



#### SRT Vs Temperature

**Figure 1**: System (suspended AS) solids retention time (SRT) versus wastewater temperature ( $T^{\circ}C$ ) for 15 IFAS biological nutrient removal wastewater treatment plants (WWTP) [from (10)]. The red and blue lines give respectively (i) the average SRT vs T relationship for the 15 plants and (ii) the ATV suspended AS SRT vs T guideline for nitrification.

Some media surface specific nitrification rates  $(r_n)$  are reported in the literature. Zimmermann (11) found  $r_n = r_{nmax}$  [1-exp(-k.L<sub>n</sub>)] where  $r_{nmax} = 1.30$  gFSA-N/(m<sup>2</sup>.d) and k=0.93. L<sub>n</sub> is the ammonia loading rate in gFSA-N/(m<sup>2</sup>.d) and ranged between 0.44 and 1.65 gFSA-N/(m<sup>2</sup>.d). Their rates were measured at a DO concentration of 5 mgO/l and temperature of 15°C. Rusten (12) gives a linear increase in  $r_n$  with increase in DO concentration, increasing from 0.60 gFSA-N/(m<sup>2</sup>.d) at 2 mgO/l to 2.1 gFSA-N/(m<sup>2</sup>.d) at 8 mgO/l at 15°C, zero organic loading and residual ammonia concentration >2.5 mgN/l. Di Trapani (13) observed  $r_n$  at 5.0 mgO/l, 14°C and 3.4 days sludge of 0.92 gFSA-N/(m<sup>2</sup>.d). To achieve low effluent ammonia concentrations of around 0.5 mgN/l, the ammonia removal rate ( $r_n$ ), and hence also the ammonia loading rate ( $L_n$ ), are significantly lower, i.e.  $r_n$  is around 0.5 gFSA-N/(m<sup>2</sup>.d). Also, the nitrification rate ( $r_n$ ) decreases with increasing organic loading rate. Rusten (12) give a value of around 0.30 gFSA-N/(m<sup>2</sup>.d) at 8 mgO/l, 15°C and 5 gBOD/(m<sup>2</sup>.d).

Placing the media in middle section of the aerobic reactor has several advantages: Significant organics removal will have already taken place, the ammonia concentration is highest in early stages of the reactor favoring the nitrification capacity of the attached biomass, the DO may be reduced in the last compartment of the aerobic reactor so less DO is recycled back to the anoxic reactor, low intensity of mixing in the last compartment improves flocculation, and the last compartment is seeded with nitrifiers from the media increasing the suspended AS nitrification in the last compartment.

#### 3. EXTERNAL NITRIFICATION BIOLOGICAL NUTRIENT REMOVAL SYSTEMS

By achieving nitrification independently of the BNRAS mixed liquor, the system sludge age can be reduced from the usual 10 to 15d to less than half, around 5 to 8 days. The reduction in sludge age increases the wastewater (WW) treatment capacity of the system by some 50% or, alternatively, reduces the biological reactor volume requirement per M $\ell$  WW treated by about 1/3rd, without negatively impacting either biological N or P removal: In fact, a reduction in sludge age increases both biological N and P removal per mass organic load (14) and this would be particularly beneficial for low temperature wastewaters (10-15°C). Because nitrification is no longer required, the aerobic mass fraction is governed by the P uptake process, for which aerobic mass fractions can be smaller than for nitrification.

External nitrification can be achieved at wastewater treatment plants (WWTPs) where old trickling filter (TF) plants have been extended with a BNRAS system or by adding nitrifying trickling filters to an existing BNRAS plant (15). There are many WWTPs with old TFs. Often at these WWTPs, to retain the benefit of the old TF, a proportion of influent WW is passed through the TF and the effluent (see Figure 2) is either (i) discharged to the BNRAS system for biological N and P removal (16) - this removes organics, the "fuel" for N and P removal and therefore decreases N and P removal, or (ii) is chemically treated to precipitate the P before discharge to the BNRAS system. This not only adds cost, but also reduces the alkalinity of the water and does not decrease the N load on the BNRAS system.

A significantly better system is obtained if the nitrification process is transferred to the TF and all the WW flow discharged to the BNRAS system (15) (17) (Figure 3). A side-stream of mixed liquor is taken from the end of the anaerobic zone and passed through internal secondary settling tanks to remove the AS solids. The underflow sludge is discharged to the beginning of the anoxic zone and the overflow is passed onto the TF for nitrification. The nitrified TF effluent is then discharged to the anoxic zone for denitrification. In this way the TF assists the BNRAS system in its weakness, i.e. nitrification, rather than taking away from it's strength, i.e. organics driven biological N and P removal. Furthermore, the oxygen demand in the aerobic reactor is markedly reduced because nitrification no longer takes place there. Indeed, not only is the nitrification oxygen demand obtained "free" outside the BNRAS system, but also the oxygen demand in the BNRAS

system, by about 1/3rd. In fact, with external nitrification, the reduction in oxygen demand in the BNRAS system is much greater than when 1/3rd of the WW is bypassed to the trickling filter as in existing TF/BNRAS systems (Figure 2). Therefore, by changing the TF to a nitrifying system as in Figure 3, the treatment capacity of the BNRAS plant is increased without having to increase aeration capacity and N&P removal are achieved on the full WW flow. If a TF plant is not available, it is possible to include plastic fixed media systems, the cost of which may be offset by the increase in WW treatment capacity.



*Figures 2 (left) and 3 (right):* At WWTPs with both activated sludge and trickling filters, common split wastewater flow use of trickling filters (Figure 2, left) and external nitrification use of tricking filters (Figure 3, right).

At short sludge ages and small aerobic mass fractions, nitrifiers would not ordinarily be supported in the BNRAS system. However, nitrifiers are not completely excluded from the BNRAS system because they are seeded into the system from the TF effluent. Therefore, nitrification in the aerobic reactor still takes place and the nitrate concentration in the aerobic reactor is governed by the ammonia concentration that enters it. Provided the TF nitrifies well (17), this nitrate concentration is mainly from the ammonia which bypasses the TF via the internal settling tank underflow, and therefore will be relatively low. If the TF does not nitrify well and the residual ammonia concentration from it is high, then, if sufficient nitrifiers are present in the aerobic reactor, the nitrate concentration will be high, with the result that a significant nitrate concentration will be present in the underflow from the final settling tank. To protect the BEPR against this potential nitrate ingress to the anaerobic reactor, a pre-anoxic reactor is placed in the underflow to denitrify the nitrate (Figure 3). If sufficient nitrifiers are not present in the aerobic reactor, then the ammonia concentration will only be partially nitrified with the result that return sludge nitrate concentration will be relatively low, but the effluent TKN concentration will be high, the concentration of the nitrification efficiency of the TF.

Tertiary nitrifying trickling filters (TNTFs), which are employed for nitrification only and negligible organic material removal, are fairly common in the USA (18). While certain problems with macro fauna (snails, worms, larvae and flies), which reduce nitrification rates, have been encountered, high removals of ammonia have been economically achieved in TNTFs (19) (20) (21). This has also been found to be the case for rock media TFs (17). Therefore, implementing the external nitrification scheme (Figure 3) is entirely feasible.

Despite the significant differences in technology, it is interesting that the specific surface nitrification rates in ventilated TNTF systems, viz. around 1.0 gFSA/( $m^2$ .d) for plastic media (20) and 0.86 gFSA-N/( $m^2$ .d) for rock media (17), are of a similar magnitude as those in IFAS systems (see 2 above). The significant reduction in nitrification rate with increasing organic load in IFAS systems is also observed in NTFs.

# 4. MEMBRANE BIOLOGICAL NUTRIENT REMOVAL SYSTEMS

Effective solid-liquid separation in suspended medium biological wastewater treatment (WWT) systems is an essential step in the process, because it has a major influence on effluent quality - in fact, SSTs are expected to achieve a 99.5% suspended solids removal to maintain an effluent suspended solids concentration < 20 mgSS/ $\ell$ . While conventionally solid-liquid separation in activated sludge systems has been accomplished in SSTs, membranes are an increasingly attractive alternative due to decreasing costs, improved performance and life, moderate maintenance and control, smaller foot print WWTP and improved effluent quality for reuse.

Membrane solid-liquid separation in place of sedimentation in SSTs offers several advantages for the activated sludge (AS) system in general, and biological nutrient removal (BNR) ones in particular (22): (i) Insensitivity to sludge settleability and filamentous bulking; this is a significant advantage as BNR systems notoriously produce rather poor settling sludges (DSVI~150 ml/g) when aerobic mass fractions are low (< 60%). (ii) Insensitivity to AS flocculation characteristics and hydraulic shear in the reactor; membranes retain all solids, which may include free swimming bacteria depending on pore size. (iii) SSTs are not required, a WWTP footprint reduction. (iv) Very high reactor concentrations of 12 to 18 gTSS/l (1.2 to 1.8%) resulting in reduced reactor volumes compared with conventional BNR systems with SSTs (further footprint reduction). (v) Variable anaerobic, anoxic and aerobic mass fractions (within a range) by varying the inter-reactor recycle ratios (see below), allowing flexibility of the biological N and P removal as dictated by influent wastewater characteristics and discharge effluent N and P concentrations. (vi) Production of high quality effluent with reduced tertiary treatment for industrial or horticultural reuse. (vii) Possibly obviate waste AS thickening when reactor concentrations are at the high end of the range for optimal membrane operation.



Figures 4 and 5: Common types of membranes used in wastewater treatment: Flat sheet (left) and hollow fibre (right).

Generally there are two types of membranes applied in activated sludge, flat sheet (FS, Kubota), which are 6mm wide hollow rectangular panels covered with membrane sheets, and

hollow fibre (HF, Zenon), which are 2m long small (1mm) diameter tubes. Both are outside to inside membranes. Flat sheet membranes are submerged vertically in the AS reactor in units comprising 50, 100, 150 or 200 panels (Figure 4). They operate under low trans-membrane pressures (100 to 800mm water) and do not require pumping. Hollow fibre membranes are also submerged but usually placed in a separate reactor and are "vacuum" pumped to create a trans-membrane pressure (Figure 5).

Fouling of the membranes is reduced by coarse bubble aeration, which, in the case of the flat sheet membranes, also supplies some or all of the oxygen required for aeration of the AS. For the hollow fibre membranes separate aeration is provided for the activated sludge reactor (fine bubble) and membrane reactor (course bubble). To increase the scour effectiveness of the air and increase the critical flux, the solids (TSS) concentration in the reactor can be high in the 12 to 18 gTSS/ $\ell$ . This is advantageous because it reduces reactor volume and increases flux (saves capital cost) but is also disadvantageous because it strongly decreases the oxygen transfer rate in the activated sludge reactor (increases running cost) (23). This is the crux of the issue with MBR AS - finding an appropriate reactor concentration which minimizes total cost (capital + running). Flat sheet membrane units can also be placed one above the other in "double storey" stacks which increases the membrane surface area per unit reactor volume and air flow.

To size a membrane BNR system, the aerobic reactor volume and aeration requirements of the activated sludge system are imposed on those of the membranes (22). The biological reactor volume is governed by (i) organic and inorganic loads, (ii) sludge age, (iii) selected reactor MLSS concentration, (iv) wastewater characteristics and (v) incorporation of primary sedimentation or not. The membrane surface area and hence the volume required to accommodate these membranes in the aerobic reactor, is governed by the peak wet weather flow (PWWF,  $Q_{PWWF}$ ) and the critical flux (m<sup>3</sup> effluent per m<sup>2</sup> membrane area per day, j<sub>crit</sub>) through the membranes. For normal municipal wastewater, flat sheet membranes have a short term (at PWWF) and long term (balanced flow) critical operational fluxes ( $j_{max}$ ) of 1.0 and 0.7 m<sup>3</sup>/(m<sup>2</sup>.d) respectively. In order to minimize membrane fouling, coarse bubble aeration is supplied below the membranes, which, together with the high TSS concentration, creates an effective scour over the membranes surfaces. This coarse bubble aeration also supplies some, or all, of the peak biological oxygen requirement. If the oxygen supplied by the membrane aeration system is insufficient to meet the peak biological oxygen demand, the aerobic reactor has to be enlarged to accommodate additional fine bubble aeration to supply the biological oxygen demand deficit. Therefore two criteria govern the volume of the aerobic reactor; (i) at low influent COD concentration, the volume required to accommodate the membranes and (ii) at high influent COD concentration, the volume required to transfer the peak biological oxygen demand when the scour of the membrane units do not meet this demand. Accurate aeration information is therefore essential to correctly size the reactor. The impact of these two criteria on the sludge age and volume requirements of flat sheet membrane BNR systems with different anaerobic, anoxic and aerobic mass fractions in comparison with conventional BNR systems with the same mass fractions was evaluated by Ramphao (22) for three cases, i.e. diurnal flow and load variation with a PWWF to average dry weather flow (ADWF) ratio ( $f_{0}$ ) of 2:1 with single (Case 1) and double (Case 2) storey membrane layouts in the aerobic reactor and an influent flow balanced case with a single storey membrane layout (Case 3). These three cases were evaluated treating for normal raw and settled municipal wastewater.

A uniform distribution of sludge mass in membrane BNR systems will not occur, even in systems with a single recycle flow from the aerobic to the zone receiving the influent flow. For example, changing an anoxic - aerobic reactor modified Ludzack - Ettinger (MLE) ND system or a UCT system with SSTs to membrane solid-liquid separation systems (Figures 6 and 7) will change the distribution of the sludge mass in the systems, the magnitude of change depending on the magnitude of the recycle ratios. The equations linking the volume and sludge mass fractions of common BNR configurations are given by (22).



*Figures 6 and 7: Modified Ludzack Ettinger (MLE) ND (left) and University of Cape Town (UCT) ND and EBPR (right) system configurations with membranes solid-liquid separation.* 

A significant advantage of multi- zone BNR systems with membranes in the aerobic reactor and fixed volumes for the anaerobic, anoxic and aerobic zones (i.e. fixed volume fractions) is that the mass fractions can be varied (within a range) by varying the inter-reactor recycle ratios. For example, in a UCT system (Fig 4) with anaerobic, anoxic and aerobic zone volume fractions of 0.25, 0.35 and 0.40 and an r-recycle ratio from the anoxic to the anaerobic zones of 1:1, the anaerobic, anoxic and aerobic zone mass fractions can be varied from 0 to 0.131, 0 to 0.366 and 1 to 0.503 respectively by changing the a-recycle ratio from 0:1 to 5:1 (Figure 8). Increasing the a-recycle ratio concomitantly increases the nitrate load on the anoxic reactor thereby increasing the denitrification and N removal as the anoxic mass fraction increases. Increasing the r-recycle ratio increases the anaerobic mass fraction (at the expense of the other two zone mass fractions) and increases (not proportionally) the P removal. This zone mass fraction flexibility is a significant advantage of membrane BNR systems over conventional BNR systems with SSTs because it allows changing the mass fractions to optimize biological N and P removal in conformity with influent wastewater characteristics and the effluent N and P concentrations required.



Figure 8: Anaerobic, anoxic and aerobic mass fraction versus mixed liquor a-recycle ratio for fixed r-recycle ratio of 1:1 and anaerobic, anoxic and aerobic volume fractions of 0.25, 0.35 and 0.40 respectively for a UCT ND EBPR system (Figure 7).

If required, the performance of membrane BNR systems can be simulated with current BNR activated sludge simulation models such as IWA ASM Nos 1, 2 and 2d - ND (24) and BEPR (25), (26) by returning the SST underflow into the aerobic zone from which the SST feed flow exits. Such simulations require *a priori* information on the reactor and zone volumes and recycle flows, which can be determined with the usual steady state design procedures (14) (27) (28). The only kinetic rate that was found to be influenced by reactor TSS concentration was the maximum specific growth rate of the nitrifiers ( $\mu_{AmT}$ ), which decreased with increasing TSS concentration relative to a parallel BNR system at low TSS concentration (4 gTSS/ $\ell$ ) (29). However, unlike conventional BNR systems with SSTs, the maximum specific growth rate of the nitrifiers ( $\mu_{AmT}$ ) does not usually define the size of the membrane BNR reactor at high TSS concentration (22) -

sludge ages need to be longer than for conventional systems to generate sufficient TSS mass in the reactor if operated at elevated reactor TSS concentration.

Generally, the longer the sludge age of the membrane system, the greater the biological reactor volume as a percentage of the equivalent conventional system volume at 15d sludge age and 4 gTSS/*l* aerobic zone concentration. Although ND EBPR systems generate more sludge (10-20%) and utilize less oxygen (5-10%) than ND systems (27) (3) and therefore require shorter sludge ages, the difference is not large enough to make a significant difference to the volume of the membrane ND EBPR system as a percentage of the equivalent conventional system volume. The aerobic mass fraction (f<sub>maer</sub>) has the greatest impact on the sludge age of the system and therefore also the membrane reactor volume as percentage of the equivalent conventional system volume - the lower the aerobic mass fraction, the longer the sludge age and the greater the percentage volume. For f<sub>maer</sub><0.50 and low wastewater strengths, the volume of the membrane BNR system increases above 50% of the equivalent conventional system volume, which erodes some of the volume reduction advantage of using membranes. Influent flow balancing significantly reduces the sludge age of the membrane BNR system and hence also the percentage of the equivalent conventional system volume. Double storey membrane units (without flow balancing) also reduce the sludge age and the percentage volume of the equivalent conventional system, but not as significantly as influent flow balancing.

Combining membranes and SSTs for solid-liquid separation in the same BNR system is possible in a side stream membrane reactor with a low sludge return flow (20%) to the reactor to harvest a high grade effluent from the BNR reactor for re-use. Also, by wasting activated sludge directly from such a side stream reactor obviates waste sludge thickening but the costs of this would have to be compared with other thickening systems such as dissolved air flotation. Gravity thickening of P rich waste activate sludge (WAS) is counter-productive because it results in the return to the influent of an overflow with a very high P concentration due to P release – MBR or flotation thickening keep the WAS aerobic and the P in the PAO during thickening.

Installing membranes for solid-liquid separation into biological nutrient removal (BNR) activated sludge (AS) systems makes a profound difference not only to the design of the BNR system itself, but also to the approach to design of the whole wastewater treatment plant (WWTP).

#### 5. AEROBIC GRANULATION ACTIVTED SLUDGE (AGAS) SYSTEMS

Sludge settleability remains one of the most unpredictable, capricious and least controllable parameters of the suspended activated sludge system that governs the size of the secondary settling tanks (SST) (5). While significant advances have been made in understanding the causes and control of filamentous organisms, such as control of specific filament organism group proliferation with kinetic and metabolic selection (30) (31) (32) (33) or with non-specific control with toxicants like chlorine or ozone (30), the problem of filamentous organism proliferation (bulking) resulting in poor sludge settleability is far from solved. However, other developments such as membrane solid-liquid separation (MBR, see 3 above) and aerobic granular activated sludge (AGAS) essentially eliminate the sensitivity of the suspended AS system to sludge settleability. Full biological nutrient (N&P) removal is included in AGAS - in fact, the growth of phosphorus accumulating organisms (PAO) is essential to effective aerobic granulation (34) (35) (36).

The physical classification of the aerobic granules is (i) a minimum diameter of 0.21mm, and (ii) a Sludge Volume Index (SVI) at 5 minutes closely equal to that at 30 minutes. A photo of typical aerobic granules is given in Figure 9.



Figure 9 (left): Aerobic granules - Ede WWTP; Figure 10 (right): The Nereda® AGAS cycle.

The AGAS is a sequencing batch (SBR) operated system with three cycle steps (Figure 10): (i) Fill and Draw - due to the fast settling velocity of the granules, wastewater can be fed into the reactor without significantly expanding the concentrated sludge bed on the reactor floor after the settling phase. This feature allows a plug flow feed which displaces the effluent at the top of the reactor like a settling tank weir overflow. Due to the high concentration of the settled sludge bed, any residual nitrate in it is rapidly denitrified creating anaerobic conditions in the bed. The feed into the bottom of the reactor into the "anaerobic" settled sludge bed on the floor results in conversion of readily biodegradable organics to volatile fatty acids and selects for PAOs. (ii) Aeration - during this step all the usual BNR biological reactions take place simultaneously. This is possible due to an oxygen gradient inside the granule, resulting in an aerobic outer shell and a core that is anoxic/anaerobic (Figure 11). Nitrifying bacteria and PAO grow in the outer shell due the "high" DO concentration from the bulk liquid resulting in nitrification of ammonia to nitrite/nitrate and P uptake. The nitrite/nitrate is denitrified to nitrogen gas in the inner part of the granule by heterotrophic and autotrophic denitrifiers. Due to the deep penetration of substrate into the granule during the feed phase, sufficient stored organics for denitrification are available in the core during aeration. Also, as a result of long-term anoxic conditions in the granule, Anammox and nitrite shunt bioprocesses are able to take place (see 7 below). (iii) Settling - to separate the treated wastewater and the granules, only a short settling time is required due to the very fast settling velocity of the granules. The reduced settling time minimises 'idle time' in the reactor (37).

In 1993 the Delft University of Technology (TUD) commenced the development of AGAS. The milestones in this development are: (i) 1993: Commenced research at TUD. (ii) Since 1998: Co-operation between RoyalHaskoningDHV and TUD. (iii) 2000-2002: Successful feasibility study. (iv) 2002: Stable granulation at lab scale. (v) 2003-2005: Pilot scale research commenced at Ede WWTP. (vi) 2004: 1st Industrial Application - Cheese processing factory. (vii) 2005: Launched Nereda® and the National Nereda programme (NNP). (viii) 2006: Pilot scale research commenced at Aalsmeer WWTP. (ix) 2006: 2nd Industrial Application - Food Industry. (x) 2006: Started NNOP - Dutch National Nereda Ontwikkelings (Development) Programme, a partnership between STOWa (Dutch Foundation for Applied Water Research), STW (Foundation for Applied Science of Netherlands), TUD and RoyalHaskoningDHV. (xi) 2007: Pilot Research Epe, Hoensbroek and Dinxperlo WWTPs. (xii) 2007: Full Scale Industrial Chemical Industry (conversion of SBR tanks). (xiii) 2008: Full Scale Demonstration Domestic in Portugal (Freilas) and South Africa (Gansbaai). (xiv) 2008: Pilot Scale Testing Brewery wastewater (post UASB). (xv) 2010: Epe plant under construction. (xvi) 2010-2013: Wemmershoek full scale, South Africa, 2x1800m<sup>3</sup> reactors. (xvii) 2010-2013: Vroomshoop full scale, Netherlands, 2x2400m<sup>3</sup> reactors. (xviii) 2011-2013: Dinxperlo full scale, Netherlands, 2x1800m<sup>3</sup> reactors. (xix) 2012: Garmerwolde full scale, Netherlands, 2x9500m<sup>3</sup> reactors. (xx) 2012-2013: Ryki full scale, Poland, 2x2500m<sup>3</sup> reactors. (38)



*Figure 11:* Schematic representation of the layered structure of aerobic granules, enabling simultaneous nitrogen and phosphate removal during the aeration step.

In comparison with the conventional AS, the benefits of the AGAS are: (i) All the BNR bioprocesses take place in the same reactor, including settling, making wide ranging process configurations possible by manipulating the process conditions in the reactor - the process configuration is not "cast in concrete" (ii) A mixed liquor sludge concentration of 8 000-10 000mg/l (compared with 3000 – 6000mg/l) allowing for smaller reactor size. (iii) A granule settling rate of 8 -12m/h (compared to 0.5 - 1.5m). (iv) No separate settling tanks required (compared with 1m<sup>2</sup>/m<sup>3</sup> flow/hr) which results in a much reduced plant footprint. (v) No recycle pumps (compared to a mixed liquor a-recycle of 3 – 5xADWF and an underflow s-recycle of 1xADWF) which results in energy savings and reduced maintenance. (vi) No mixers in the anaerobic and anoxic reactors once again reducing energy consumption and a reduction in maintenance. (vii) A footprint approximately 1/3rd of a conventional activated sludge plant, allowing plant upgrades at existing works where space is limited obviating the need to relocate the plant with possible associated additional costs. (viii) Significant savings in both capital and operation and maintenance costs with significant improvement in energy efficiency. (ix) The process configuration is flexible because the "size" of the anaerobic, anoxic and aerobic "zones" are determined not by concrete divisions, but by a PLC controller which controls the cycle times and aeration rate so that these system parameters can be adjusted to obtain optimum system condition for BNR. (x) The sludge handling characteristics of granular sludge is superior to suspended activated sludge (38).

#### 6. NITRITE SHUNT AND ANAMMOX

Nitrogen removal is accomplished in wastewater treatment by nitrification and denitrification (ND). Nitrification is mediated by two groups of autotrophic nitrifying organisms: (i) the first, generically called the ammonia oxidizing bacteria (AOB), utilize ammonia as electron donor and oxygen as electron acceptor and produce nitrite, and (ii) the second, generally called nitrite oxidizing bacteria (NOB), utilize nitrite as electron donor and oxygen as electron acceptor and produce nitrate. In denitrification, most facultative heterotrophic organisms utilize organics as electron donor and nitrate as electron acceptor and produce nitrogen gas. Some facultative heterotrophs perform only the first step, viz. denitrify nitrate to nitrite, while others perform only the second step, viz. denitrify nitrite to nitrogen gas. Generally very little nitrite accumulation takes

place in activated sludge and practically all nitrate formed by nitrifiers is denitrifying by facultative heterotrophs to nitrogen gas. This nitrogen removal cycle is shown schematically in Figure 12.



**Figure 12:** Nitrogen cycle in AS with the oxidation state (electron state) of the various reduced and oxidized nitrogen compounds on the vertical axis (Nitrite shunt – dark green, Anammox - red, oxygen input - blue, organics input - purple.

A generalised complete CHONPS, COD and charge mass balanced stoichiometry for a wide range of bioprocesses has been developed by (39), *inter alia*, methanogenesis, sulphidogenesis, autotrophic denitrification and also for the nitrification and denitrification (both in one and two steps) and anammox bioprocesses relevant to this paper (Figure 12). This generalized stoichiometry is not given in this paper, but the results relevant to the bioprocesses in Figure 12 are. In Table 1 are given stoichiometric values for the autotrophic nitrification bioprocesses #3, #3a, #3b, the heterotrophic denitrification bioprocesses #6, 6a, 6b utilizing methanol (CH<sub>3</sub>OH) as organic substrate, the anammox bioprocess #7 and aerobic heterotrophic bioprocesses #4 utilizing methanol as organic substrate for (1) biomass net yield (E), i.e. fraction of substrate electron donating capacity (EDC) becoming biomass (anabolism) and fraction (1-E) passed to the electron acceptor (catabolism), of facultative heterotrophs, nitrifiers (AOB and NOB), anammox and aerobic heterotrophs of 0.40, 0.15, 0.05 and 0.50 respectively, and (2) a generic biomass composition of all bacterial species of  $C_1H_{1.6811}O_{0.4480}N_{0.1655}P_0S_0$ , which is obtained from COD, C, H, O ,N, P and S mass ratios (g/gVSS) of 1.481, 0.518, 0.0726, 0.3094, 0.100, 0.00 and 0.00 respectively.

From this stoichiometry, the oxygen requirements for the two steps of nitrification are 2.92 gO/gN ammonia transformed to nitrite and 0.97 gO/gN nitrite transformed to nitrate. The organics (COD) requirement for the two steps of denitrification are 1.91 gCOD/gN nitrate transformed to nitrite and 2.86 gCOD/gN nitrite transformed to nitrogen gas. So if all the N removal takes place over nitrite instead of over nitrate, the oxygen saving would be 25% and the methanol saving would be 40%. These are considerable savings, so significant research effort is being made to find ways to discourage the growth of NOB and encourage the growth of Anammox bacteria in N removal activated sludge systems.

**Table 1:** Numerical values of the bioprocess stoichiometry terms for 1 mmol/l reactant substrate for ammonia, nitrite and methanol (x=1.00, y=4, z=1, a=0, b=0, c=0, ch=0) and biomass (k=1.00, l=1.6811, m=0.4480, n=0.1655, p=0.0, s=0.0, ch=0, which are obtained from the mass ratios of  $f_{cv} = 1.500 \text{ gCOD/gVSS}$ ,  $f_c = 0.500 \text{ gC/gVSS}$ ,  $f_n=0.0 \text{ gN/gVSS}$ ,  $f_p=0.0 \text{ gP/gVSS}$ ,  $f_s=0.0 \text{ gS/gVSS}$  and  $f_{ch}=0$  for methanol and  $f_{cv} = 1.481 \text{ gCOD/gVSS}$ ,  $f_c = 0.518 \text{ gC/gVSS}$ ,  $f_n=0.100 \text{ gN/gVSS}$ ,  $f_p=0.0 \text{ gP/gVSS}$  for biomass and net yield coefficients (E) = 0.05 for anaerobic (bioprocess #7), 0.15 for autotrophic aerobic (#3, #3a, #3b), 0.40 for heterotrophic denitrification (#6, #6a, #6b) and 0.50 for aerobic heterotrophic growth (#4). Note that in Table 1 the bioprocess and compound numbering from Ekama (39) has been retained.

Compounds			1	4	8	9	13	14	15	16	17	18	23
Bioprocess		Units	Organics	Biomass	$O_2$	$\mathrm{NH_4}^+$	NO <sub>3</sub> <sup>-</sup>	NO <sub>2</sub> <sup>-</sup>	$N_2$	H <sub>2</sub> O	HCO <sub>3</sub> <sup>-</sup>	CO <sub>2</sub>	Alk
3	Nitrification (NH <sub>4</sub> <sup>+</sup> to NO <sub>3</sub> <sup>-</sup> )	mmol/l mg/l <sup>a</sup>	-	0.267 6.20	-1.625 -52.00	-1.00 -14.00	0.956 13.38	-	-	2.753 49.56	-1.956 -23.47	1.688 20.26	-1.956 -97.79
3a	Nitrification $(NH_4^+ \text{ to } NO_2^-)$	mmol/l mg/l <sup>a</sup>	-	0.203 4.70	-1.232 -39.43	-1.00 -14.00	-	0.966 13.53	-	2.813 50.63	-1.966 -23.60	1.764 21.16	-1.966 -98.32
3b	Nitrification (NO <sub>2</sub> <sup>-</sup> to NO <sub>3</sub> <sup>-</sup> )	mmol/l mg/l <sup>a</sup>	-	0.0676 1.57	-0.411 -13.14	-0.0112 -0.16	0.9664 13.53	-0.9664 -13.53	-	-0.0289 -0.52	-0.0112 -0.13	-0.0564 -0.68	-0.0112 -0.56
4	Aerobic Heterotrophic	mmol/l mg/l <sup>a</sup>	-1.00 -48.00	0.6995 16.21	-0.75 -24.00	-0.116 -1.62		-		1.7014 30.63	-0.1158 -1.39	0.4162 4.99	-0.1158 -5.79
6	Hetero. denit. $(NO_3^{-} to N_2)$	mmol/l mg/l <sup>a</sup>	-1.389 -66.67	0.7773 18.01	-	-0.1286 -1.80	-1.00 -14.00	-	0.50 14.00	1.9460 35.03	0.8714 10.46	-0.260 -3.12	0.8714 43.57
6a	Hetero. denit. $(NO_3^- \text{ to } NO_2^-)$	mmol/l mg/l <sup>a</sup>	-0.556 -26.67	0.311 7.20	-	-0.0514 -0.72	-1.00 -14.00	1.00 14.00	-	0.9784 17.61	-0.0514 -0.62	0.2961 3.55	-0.0514 -2.57
6b	Hetero. denit. $(NO_2^- to N_2)$	mmol/l mg/l <sup>a</sup>	-0.833 -40.00	0.4664 10.80	-	-0.0772 -1.08	-	-1.00 -14.00	0.50 14.00	0.9676 17.42	0.9228 11.07	0.556 -6.67	0.9228 46.14
7	Anammox	mmol/l mg/l <sup>a</sup>		0.0356 0.82	-	-1.0232 -14.32	-	-0.9664 -13.53	0.992 27.77	2.045 36.81	-0.057 -0.68	0.0212 0.25	-0.057 -2.84

<sup>a</sup> The mass units for the compounds are: 1- mgCOD/l; 4 - mgVSS/l; 8 - mgO/l; 9, 13, 14, 15 - mgN/l; 16 -mgH<sub>2</sub>O/l; 17, 18 -mgC/l; 23 - mg/l as CaCO<sub>3</sub>.

Anaerobic ammonia oxidizing (Anammox) bacteria were first discovered about 20 years ago (40) - for a history of its discovery see (41). These bacteria utilize ammonia as electron donor and nitrite as electron acceptor to form nitrogen gas. They have been found in the back anaerobic part of biofilm systems (42) and old rock media trickling filters (43). This process has been called several different names in the literature, such as Oxygen Limited Aerobic Nitrification Denitrification (OLAND), deammonification (DEMON) and Completely Autotrophic Nitrogen removal Over Nitrite (CANON) (44). Although Anammox bacteria are anaerobic bacteria and very slow growers, they have also been found in activated sludge systems, which is remarkable considering the generally aerobic conditions of activated sludge. Interestingly, these bacteria are slightly denser than activated sludge and so accumulate in the denser and faster settling fraction of activated sludge (45). This has led to the use of hydro-cyclones on waste activated sludge streams, wasting only the light fraction of activated sludge and returning the denser fraction to the reactor (46). In this way the Anammox bacteria are retained, accumulate in the activated sludge and can make a considerable contribution to the N removal. This saves both oxygen and organics - N removal by Anammox bacteria requires only about 1.42 gO/gN removed and zero organics consumption (Figure 12) allowing N removal wastewater treatment plants to become energy selfsufficient.

Simultaneous nitrification- denitrification (SND) at low aerobic reactor DO has been observed for many years in many WWTPs. In instances where the low DO operation did not compromise nitrification, it was welcomed as additional N removal. It was believed to take place in the usual way over nitrate (Figure 12). However, increasingly evidence is coming to light that at least some of this N removal is taking place over nitrite, called nitritation/denitritation or nitrite shunt. Exploitation of nitrite shunt is still limited in BNR plants because knowledge of design, control and operational conditions which stimulate it are not well known yet. To achieve nitrite-shunt in the mainstream WWTP requires suppression of the NOB activity. Under "normal" WWTP conditions, the NOB are faster growers than the AOB and is the reason why nitrification kinetics in many AS models is based on the AOB maximum specific growth rate ( $\mu_{Am20}$ ) converting ammonia to nitrate in one step. Finding the conditions resulting in suppression of NOB is a growing research topic. While conditions for AOB proliferation and NOB suppression are well known for reject water treatment (47), these conditions cannot easily be replicated in mainstream WWTPs. Compounding the difficulty, there is currently conflicting information on the role of DO on NOB suppression to stimulate nitrite-shunt: Low DO suppresses NOB (48) while high DO was found to favor AOB over NOB in other mainstream studies (49) (50) (46) (51). Jimenez (52) describes a two stage anaerobic (25%)-aerobic (75%) Phoredox (or A/O) plant (Southwest WWTP in St Petersburg, Florida, USA) treating an influent wastewater with a COD/TKN ratio of 7:1 and temperature between 23 and 30°C at low aerobic reactor DO (0.4 to 0.1 mgO/l). This plant achieves a low effluent total inorganic N (2-4 mgN/l) and low Ortho-P (0.1 mgOP/l). Specific nitrification [mgFSA-N/(gVSS.h)] and denitrification (mgNO<sub>3</sub>-N/(gVSS.h)] rate tests revealed that the NOB were significantly suppressed due to the low DO operation and that nitrite-shunt occurs at the plant. Phosphorus release and uptake tests indicated that the low DO operation (and high temperature) did not adversely affect the biological P removal.

#### 7. CONCLUSIONS

The drive to intensify the activated sludge (AS) system so that it requires less space and consumes less energy without compromising delivery of a high quality treated effluent has led to some remarkable inventions and developments in biological nutrient removal over the past two decades. The main focus of these inventions and developments are to (i) maintain nitrifiers in the system at short sludge ages (Type A), (ii) make the system less sensitive to the capricious sludge

settleability (Type B) and (iii) remove more nitrogen with less oxygen (energy) and organics (Type C). Six of these inventions and developments have been briefly described in this paper, viz. (i) the integrated fixed-film activated sludge (IFAS) system (Type A), (ii) external nitrification (Type A), (iii) membrane solid liquid separation (Type B), (iv) aerobic granulation BNR systems (Type B), (v) short-circuiting nitrification-denitrification (ND) by preventing nitrate formation and enforcing ND over only nitrite (nitrite-shunt) (Type C) and (vi) and encouraging the growth of Anammox bacteria in the activated sludge (Type C). With IFAS, fixed media are placed in the aerobic reactor (internal) to make nitrification independent of the suspended AS sludge age. With external nitrification, nitrification is achieved in a side-stream fixed media reactor, which removes the size defining nitrification process from the suspended AS system. Both these systems (Type A) halve the suspended AS system sludge age and so increases wastewater treatment capacity. With membranes, secondary settling tanks are replaced with in-reactor membranes for solid-liquid separation. With aerobic granulation, the activated sludge is controlled to form fast settling granules comprising heterotrophs, nitrifiers, denitrifiers and phosphorus accumulating organisms (PAOs) in a sequencing batch (SBR) type reactor. With membranes and granules (Type B), secondary settling tanks are no longer required making a major reduction in wastewater treatment plant (WWTP) size. Short-circuiting ND by suppressing nitrite oxidizing organisms and enforcing ND over nitrite and/or encouraging the growth of Anammox bacteria in the activated sludge which remove N autotrophically by combining ammonia and nitrite to form nitrogen gas, requires significantly less oxygen and organics (Type C) than ND over nitrate. Of these inventions and developments, it is the author's opinion that (iv) granulation and (vi) encouraging the growth of Anammox bacteria in the activated sludge, hold the most promise because these appear easiest to achieve in practice at the lowest cost.

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