# Single stage biological nitrogen removal by nitritation and anaerobic ammonium oxidation in biofilm systems

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Abstract In full scale wastewater treatment plants with at times considerable deficits in the nitrogen balances, it could hitherto not be sufficiently explained which reactions are the cause of the nitrogen losses and which micro-organisms participate in the process. The single stage conversion of ammonium into gaseous end-products – which is henceforth referred to as deammonification – occurs particularly frequently in biofilm systems. In the meantime, one has succeeded to establish the deammonification processes in a continuous flow moving-bed pilot plant. In batch tests with the biofilm covered carriers, it was possible for the first time to examine the nitrogen conversion at the intact biofilm. Depending on the dissolved oxygen (DO) concentration, two autotrophic nitrogen converting reactions in the biofilm could be proven: one nitritation process under aerobic conditions and one anaerobic ammonium oxidation. With the anaerobic ammonium oxidation, ammonium as electron donor was converted with nitrite as electron acceptor. The end-product of this reaction was $N_2$. Ammonium and nitrite did react in a stoichiometrical ratio of 1:1.37, a ratio which has in the very same dimension been described for the ANAMMOX-process (1 : 1.31±0.06). Via the oxygen concentration in the surrounding medium, it was possible to control the ratio of nitritation and anaerobic ammonium oxidation in the nitrogen conversion of the biofilm. Both processes were evenly balanced at a DO concentration of 0.7 mg/l, so that it was possible to achieve a direct, almost complete elimination of ammonium without addition of nitrite. One part of the provided ammonium did participate in the nitritation, the other in the anaerobic ammonium oxidation. Through the aerobic ammonium oxidation into nitrite within the outer oxygen supplied layers of the biofilm, the reaction partner was produced for the anaerobic ammonium oxidation within the inner layers of the biofilm.
Keywords Biofilm; deammonification; nitritation; anaerobic ammonium oxidation; ANAMMOX

## Introduction

For problematic kinds of wastewater with a low COD/N ratio, such as landfill leachate, sludge liquor, or industrial wastewater, alternative metabolic pathways with autotrophic micro-organisms are of particular interest. Thus, it has already been proven that autotrophic nitrifiers of the *Nitrosomonas* genus are able to induce denitrification (Bock *et al.*, 1995). In nitrification tests with *Nitrosomonas eutropha* and *Nitrosomonas europea* with limited oxygen provision, one part of the electrons from the ammonium oxidation was no longer transferred to oxygen, but to nitrite. Oxidation and denitrification were running parallel, which led to a balance deficit in the sum of the dissolved nitrogen compounds. Anaerobic ammonium oxidation could be established for *Nitrosomonas* as well.

Anaerobic ammonium oxidation (ANAMMOX) has also been described for other autotrophic micro-organisms which had been enriched from a denitrifying fluidised bed reactor (Mulder *et al.*, 1995; Van de Graaf *et al.*, 1996). The ANAMMOX organisms are currently being identified. Further analyses have shown that through metabolic reactions of these organisms ammonium and nitrite can be converted to elementary nitrogen – with but a small production of nitrate. The consumption of ammonium and nitrite and the production of nitrate are in a relation of 1:1.31±0.06:0.22±0.02 (Van de Graaf *et al.*, 1996). The fundamental analyses to describe the ANAMMOX process were done with an enriched culture that had been grown within a synthetic medium. Initial tests with actual wastewater from

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sludge digestion allow for the assumption that the conversion rates of the ANAMMOX process are sufficiently high to permit its use in wastewater treatment technology (Strous *et al.*, 1997). The ANAMMOX process is inhibited by oxygen. As ammonium and nitrite have to be available as reaction partners, any utilisation of the process for wastewater treatment technology must be preceded by a preliminary step in which about 50% of the available ammonium is oxidized into nitrite.

Without being able to clearly define the reactions, the single stage nitrogen elimination in the biological contactor (BC) unit at Mechernich (Seyfried, 1987) was subsumed under the term "aerobic deammonification" (Hippen *et al.*, 1997, 1998). It was at first assumed that limited DO concentrations are necessary for the autotrophic (and possibly also for the heterotrophic) conversion of ammonium into $N_2$. Biomass removed from the disks of the BC showed nitrogen losses at an oxygen concentration of 1 mg/l, even after homogenising mechanically for destruction of "floc" structure (Helmer and Kunst, 1998). Nevertheless, it could not be excluded that even after homogenisation sufficiently large cell clusters remain which might enable anoxic reactions. Therefore, Helmer *et al.* (1999) introduced the term "aerobic/anoxic deammonification". Further examinations with batch tests (Helmer *et al.*, 1999) made apparent that – apart from ammonium – nitrite, but no organic substrate must be available to achieve deammonification. $N_2$ was identified as end product of the reaction. In microbiological analyses, large cell clusters of ammonium oxidisers were detected through the use of FISH (fluorescent in situ hybridization) with rRNA-targeted nucleic acid probes in biofilm areas with particularly high deammonifying activity (Helmer *et al.*, 1999). At this point in time, denitrification through autotrophic ammonium oxidisers with ammonium as electron donor and nitrite as electron acceptor was the most probable explanation for the observed nitrogen losses. The question whether what occurred was an aerobic denitrification or a denitrification in anoxic micro-zones could not be answered conclusively. Furthermore, it remained unclear how high the ratio of a classical heterotrophic denitrification must be estimated which can run with utilising the organic substrates dissolved from decaying biomass.

In the meantime, Hippen *et al.* (1999a) have succeeded to establish the aerobic/anoxic deammonification in a moving-bed pilot plant. As carriers, Kaldnes material was used. One line of the pilot plant was run with carriers which had initially been grown for 6–8 weeks with the deammonifying biomass of the BC unit at Mechernich. In the second line, deammonification could be established simply by the setting and adjusting of suitable operation conditions (Hippen *et al.*, 1999b). The pilot plant is fed with wastewater from sludge treatment of a municipal wastewater treatment plant, which is characterised by its high nitrogen concentrations. Through the successful operation of this pilot plant it became possible to further characterise the reactions of the aerobic/anoxic deammonification within the stacked biofilm, because the Kaldnes carriers could with their biofilm growth be used directly in the batch tests. For the tests, those carriers were selected which had been overgrown at Mechernich.

The batch tests were to provide answers to the following questions:

1. Does the aerobic/anoxic deammonification consist exclusively of autotrophic reactions?
2. Which impact does oxygen have on the reactions of the aerobic/anoxic deammonification?
3. Which reaction partners participate in the process of the aerobic/anoxic deammonification?

## Materials and methods

### Design and operation of the moving-bed pilot plant

The deammonifying pilot plant consists of a preliminary denitrification reactor with topped settlement tank and a nitrification stage consisting of three in-line reactors. Each reactor

Table 1 Operation parameters of the deammonifying moving-bed pilot plant

| Operation parameter | | Plant data |
|---|---|---|
| Hydraulic retention time per reactor | $t_R$ [h] | 8 |
| Influent flow rate | $Q_d$ [l/d] | 120 |
| Recirculation flow | $Q_{recycle}$ [l/d] | 120 |
| DO concentration | [mg/l] | ≤1 |
| pH-value | [–] | ≥8 |
| Temperature | [ºC] | 28 |

Table 2 Average performance data of the pilot plant in the operation phase with parallel batch tests

| | | Nitrification stage | | |
|---|---|---|---|---|
| Parameter | | Reactor 1 | Reactor 2 | Reactor 3 |
| Ammonium surface load | [g $NH_4$–N/$m^2 \cdot d$] | 11.9 | 9.9 | 7.8 |
| Ammonium surface degradation | [g $NH_4$–N/$m^2 \cdot d$] | 1.9 | 2.6 | 2.0 |
| Deammonification | [g $NH_4$–N/d] | 4.8 | 12.8 | 8.0 |

has a work capacity of 40 l; at the initiation of the plant (October, 1997), each was filled to 20% with overgrown Kaldnes material. According to Ødegaard *et al.* (1994) an effective specific growth surface of 100 $m^2$ per $m^3$ of reactor volume is reached in this way.

### Batch tests

The batch tests were run in two parallel reactors with a working volume of 1.8 l each. On test days, the carriers were taken from the reactor of the continuously working pilot plant that was selected respectively and put into batch reactors until a volumetric filling of 40% was reached. The reactors were equipped with slow stirring implements. The temperature was set at 28ºC, the pH-value to 8.0.

*Batch tests to prove the autotrophic character of the aerobic/anoxic deammonification.* The COD in sludge liquor must be regarded as hardly biodegradable, but it can still in principle be used as carbon source for heterotrophic micro-organisms. In order to exclude that organic carbon plays a role in the aerobic/anoxic deammonification, carriers were for reasons of comparison taken from the nitrification reactor 2 of the pilot plant in (a) sludge liquor, (b) mineral medium, and (c) mineral medium + acetate. The mineral medium contained (per l): 87.7 mg $KH_2PO_4$, 300 mg $MgSO_4 \cdot 7H_2O$, 5.06 g $NaHCO_3$. It contained the trace elements cobalt, manganese, zinc, copper, nickel, and iron in a concentration of 10 µg/l each. In approach (c), acetate was additionally dosed with a concentration of 250 mg/l. The ammonium concentration was at the beginning of the tests set to approximately 150 mg/l $NH_4$–N through the addition of sludge liquor in approach (a), or through addition of $NH_4Cl$ in approaches (b) and (c). The DO concentration was in all cases kept consistently at 0.7 mg/l.

*Batch tests on the impact of oxygen on the reactions of the aerobic/anoxic deammonification.* The nitrogen conversion was traced with carriers from all three nitrification reactors at DO concentrations of 0 mg/l, 0.7 mg/l, 2.0 mg/l and 5.0 mg/l. The carriers were taken in the mineral medium (see above). 150 mg/l $NH_4$–N were provided in the form of $NH_4Cl$ at the beginning of the tests.

*Batch tests to ascertain the reaction partners participating in the aerobic/anoxic deammonification*. In batch tests with biomass taken from the disks of the BC unit at Mechernich it had already become apparent that, apart from ammonium, nitrite is a major reaction partner in the aerobic/anoxic deammonification. Thus, further batch tests with carriers from the nitrification reactors 2 and 3 were run to examine the impact of an additional nitrite supply. For this, the carriers were again taken in the mineral medium (see above). Ammonium was provided at a concentration of 150 mg/l $NH_4$–N in the form of $NH_4Cl$, with the basic nitrite concentration having been set between 60 and 80 mg/l $NO_2$–N (addition in the form of $NaNO_2$). The impact of nitrite was checked in the anoxic environment, as well as with a DO concentration of 0.7 mg/l. Moreover, denitrification tests were run in which only nitrite with a concentration of 60 mg/l $NO_2$–N was provided, for comparison both with and without acetate as source of organic carbon. All batch tests were repeated with the provision of nitrate instead of nitrite.

*Balance of the nitrogen conversion in the anoxic batch test*. The batch reactors could be closed with a gas-tight lid, so that complete nitrogen balances at different points of time during any batch tests could be taken including the gaseous nitrogen compounds.

*Sampling during the batch tests.* Over a period of 5 hours, samples were taken over 30 minutes. The samples were filtered and analysed for $NH_4$–N, $NO_2$–N, $NO_3$–N, and COD. Dissolved organic nitrogen was analysed as TKN, taken gas samples were checked for $N_2O$ and $N_2$ in a gas chromatograph.

## Results and discussion

*Batch tests to prove the autotrophic character of the aerobic/anoxic deammonification.* Regardless of whether the batch tests were run in (a) sludge liquor, (b) mineral medium, or (c) mineral medium + acetate, a considerable decrease of the ammonium concentration without production of nitrite or nitrate could be observed during the 5 hour long test period. In approach (a) with sludge liquor, within 5 hours 73 mg/l of $NH_4$–N were removed. In approach (b) with a mineral medium, the $NH_4$–N conversion amounted to 66 mg/l, and in test (c) in mineral medium + acetate the result was 64 mg/l $NH_4$–N. Figure 1 shows as an example the nitrogen conversion in approach (b). In test (a), during the 5 hours 3.9 mg/l of COD were consumed. In approach (b) no further COD was provided, and no conversion could be measured, whereas in test (c) acetate was consumed, so that a COD decrease by 60 mg/l was recorded. Without addition of a carbon source, ammonium was directly converted at a rate of 13.3 mg/(l . h) $NH_4$–N. Neither the addition of hardly biodegradable carbon sources through sludge liquor, nor the addition of the readily biodegradable carbon source acetate could influence the conversion rate in any considerable manner.

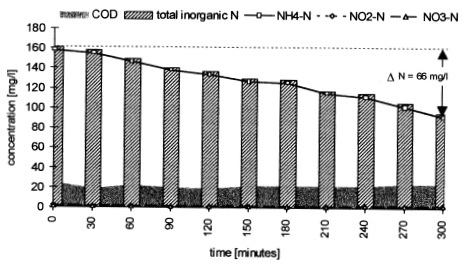


Figure 1 Conversion reactions in the batch test with carriers from reactor 2, taken in mineral medium (approach b) – DO concentration=0.7 mg/l

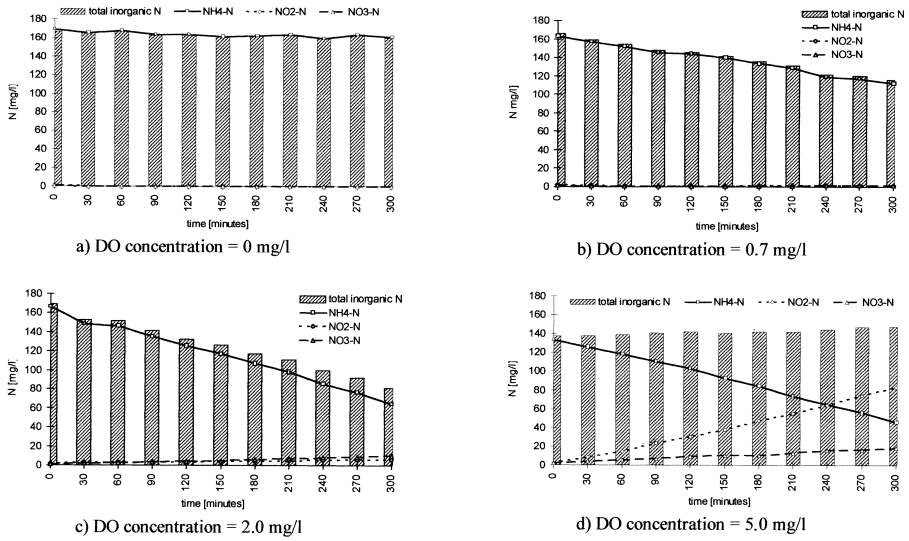


Figure 2 Nitrogen conversion reactions at different DO concentrations (carriers taken from the nitrification reactor 2 of the moving-bed pilot plant)

*Batch tests on the impact of oxygen on the reactions of the aerobic/anoxic deammonification.* In all batch tests without oxygen, no nitrogen conversion did occur if ammonium was the only nitrogen source, regardless of the origin of the carriers (Figure 2a for reactor 2). At increasing DO concentrations, the carriers of the three nitrification reactors showed different reactions. In the batch tests with carriers from *reactor 1*, a classical nitritation was measured (nitrification of ammonium up to nitrite). The conversion rate of the nitritation did linearly increase with the increasing oxygen supply.

In the batch tests with carriers from *reactor 2*, at a DO concentration of 0.7 mg/l an ammonium conversion of 10.1 g/(l . h) $NH_4$–N was measured, without any nitrite or nitrate amounts worth mentioning being produced as reaction products (Figure 2b). At a DO concentration of 2.0 mg/l, an accelerated ammonium conversion of 20.3 mg/(l . h) $NH_4$–N was recorded (Figure 2c), again with next to no production of either nitrite or nitrate. Only at a DO concentration of 5.0 mg/l did a classical nitritation happen (with a small ratio of nitratation), that is ammonium could completely be found again as nitrite (or nitrate) (Figure 2d). The largest amount of inorganic nitrogen was eliminated at a DO concentration of 2.0 mg/l. Ammonium was removed at a degree of 81.6%, the rest could be found again a nitrite or nitrate. At a DO concentration of 0.7 mg/l, the elimination of ammonium came off to a high degree – here, 98.9% were converted, without production of nitrite or nitrate. In the batch tests with carriers from reactor 3, the results were similar to those achieved in reactor 2.

*Batch tests to find out which reaction partners take part in the aerobic/anoxic deammonification.* As the reference literature describes reactions in which nitrite instead of oxygen can serve a electron acceptor, the impact of an additional supply of nitrite was analysed with carriers from the nitrification reactor 2 at a DO concentration of 0.7 mg/l. It became obvious that nitrite and ammonium were converted in parallel (Figure 3).

Given that after about half of the test period nitrite had been completely eliminated, a differentiated view at the first 120 minutes shows in comparison to the second test phase (minutes 120–200) the impact of nitrite on the aerobic/anoxic deammonification. The additional supply of nitrite accelerates the ammonium conversion rate in the first half of the test period by 50% compared to the second half with ammonium as only nitrogen source. The

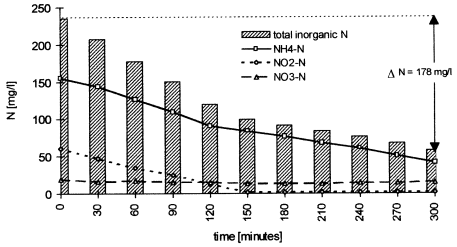


Figure 3 Nitrogen conversion reactions with initial addition of ammonium and nitrite (carriers taken from reactor 2 – DO concentration=0.7 mg/l)

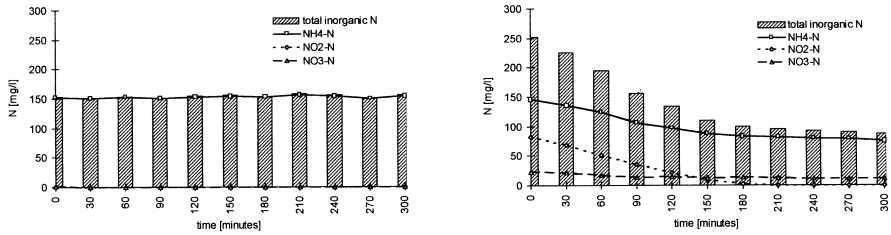


Figure 4 Nitrogen conversion reactions under aerobic or anoxic conditions, respectively (carriers taken from reactor 3 in a mineral medium)

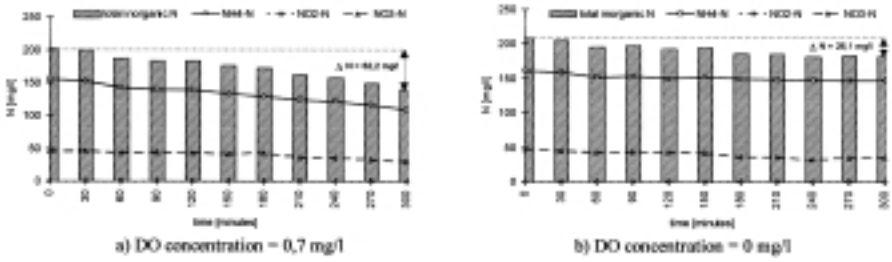


Figure 5 Nitrogen conversion reactions with initial provision of ammonium or nitrate, respectively (carrier taken from reactor 3 in a mineral medium)

accelerated ammonium conversion rate was 32.1 mg/(l . h) $NH_4$–N, compared to 16.5 mg/(l . h) $NH_4$–N without additional nitrite supply. The total amount of inorganic nitrogen eliminated during the five hour long test period was 75%. The nitrogen loss amounted to 97.7%.

Without oxygen as electron acceptor, in the batch test with provision of ammonium as only nitrogen source no reaction occurred (Figure 4a). In a comparable batch test with additional nitrite supply, however, it was possible to achieve during the first two hours an ammonium conversion rate of 24.4 mg/(l . h) $NH_4$–N, with a nitrogen loss of 97.9% (Figure 4b). During this period, nitrite was converted parallel to the ammonium at a rate of 30.8 mg/(l . h) $NO_2$–N. As soon as the nitrite was consumed completely, the reaction came to a standstill.

Anoxic conditions do on principle allow a classical heterotrophic denitrification of the provided nitrite as well, which, however, depends on the availability of a carbon source. The batch tests were run in a mineral medium without addition of COD. Still, there is the possibility of an endogenous denitrification by utilising storage substances of released COD from

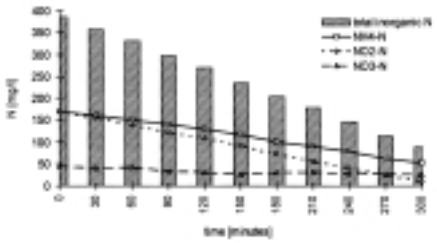

Figure 6 Nitrogen conversion reactions in the anoxic batch test with initial addition of ammonium and nitrite

Table 3 Elimination of inorganic nitrogen in denitrification tests with and without provision of substrate or ammonium

| | Elimination of inorganic nitrogen within 120 minutes [mg/l] | |
|---|---|---|
| Test approach | Provision: Nitrite | Provision: Ammonium + Nitrite |
| Without acetate | 17.9 | 118.3 |
| With acetate (250 mg/l) | 21.3 | 121.4 |

decaying biomass. In order to be able to estimate the ration of the denitrification within the nitrite conversion, denitrification tests were run without addition of a carbon source with provision of nitrite. The nitrite conversion happened at a rate of 3.7 mg/(l . h) $NO_2$–N, compared to 28.4 mg/(l . h) $NO_2$–N with additional ammonium addition at the beginning of the test. Even by additional dosing of acetate during the denitrification test, the nitrite conversion rate could be increased only to 4.9 mg/(l . h) $NO_2$–N. Table 3 shows a comparison of the elimination rates reached in the denitrification test with and without additional ammonium provision. It becomes apparent that an endogenous denitrification is responsible at the utmost for a ratio of 15% within the entire nitrogen conversion. Even if substrates are provided, the theoretically possible ratio of the denitrification rises only to 18%.

Numerous batch tests with nitrate instead of nitrite have shown that, in contrast to nitrite, nitrate can be used as reaction partner for deammonification only to a limited degree. Figure 5 shows the reaction courses in two batch tests with provision of ammonium and nitrate at a DO concentration of 0.7 mg/l and under anoxic conditions, respectively. Compared to the nitrite conversion rate in a comparable test at a DO concentration of 0.7 mg/l, namely 20.7 mg/(l . h) $NO_2$–N, the nitrate conversion rate in this case was 3.1 mg/(l . h) $NO_3$–N. In the anoxic test, the conversion rate reached with provision of nitrite was 28.4 mg/(l . h) $NO_2$–N; with nitrate, only 2.3 mg/(l . h). The amount of inorganic nitrogen of 62.2 mg/l $N_{inorg.}$, eliminated at a DO concentration of 0.7 mg/l is roughly equivalent to the amount which was removed when only ammonium was provided (Figure 1). The 26.1 mg/l $N_{inorg.}$ eliminated in the anoxic test are equivalent to the approximate endogenous denitrification potential (Table 3).

The stoichiometric ratio in which the reaction partners ammonium and nitrite were converted in the anoxic batch test was analysed in more detail at a provision of 170 mg/l of $NH_4$–N and $NO_2$–N each (Figure 6). The result was that the ratio between the ammonium conversion (116.6 mg/l), the nitrite conversion (160.3 mg/l), and a small nitrate conversion (18.6 mg/l) is 1:1.37:0.16. The balance of the nitrogen conversions in the anoxic batch test showed again that the end product of the deammonification is $N_2$. $N_2O$ was not produced at all.

## Conclusions

The operation of a deammonifying moving-bed plant made it possible via the use of overgrown carriers in batch tests to run detailed analyses of the microbial nitrogen conversion

reactions of an intact biofilm. Examinations at different DO concentrations make apparent that in dependence on the oxygen supply there are not only different reaction rates, but obviously also different reaction courses. In the nitrifying reactor 1, a nitrifying population with the capability to effect nitritation was found, which reacted to an improvement of the oxygen supply with a considerable increase in efficiency. In reactors 2 and 3, there was found – apart from the capability to effect nitritation – a clear potential for deammonification, that is ammonium could to a high degree be eliminated without the emergence of either nitrite or nitrate. In contrast to the nitritation, the deammonification reacted sensitively to increased DO concentrations: at a DO concentration of 0.7 mg/l, ammonium was eliminated almost completely at 98.9%, whereas with higher DO concentrations the production of nitrite or nitrate from ammonium increased.

The apparent increase of both the ammonium conversion rate and the elimination rate of inorganic nitrogen at low oxygen concentrations through additional dosing of nitrite allows for the assumption that nitrite can be used a electron acceptor for the oxidation of ammonium. Anoxic batch tests have shown that the oxidation of ammonium can be achieved independent of oxygen as long as nitrite is available as reaction partner to a sufficient degree. The end product of this anaerobic ammonium oxidation with nitrite as electron acceptor is $N_2$. At a DO concentration of 0.7 mg/l, however, the provision of ammonium only was sufficient to start the process of deammonification. Measurements of Schramm *et al.* (1997) with $O_2$-microelectrodes in nitrifying biofilms have shown that already at a biofilm thickness of 100–150 µm one has to reckon with anoxic zones within the lower layers. The respective diffusion depth of oxygen depends on the DO concentration in the surrounding medium. The biofilm on the Kaldnes carriers reached an average thickness of 400–500 µm. One can assume that at DO concentrations of 0.7 mg/l or 2.0 mg/l respectively, ammonium will be nitrified into nitrite in the outer aerobic layers, which thus produces the second reaction partner for an anaerobic ammonium oxidation. In the deeper biofilm layers which are not supplied with oxygen anymore, the micro-organisms oxidise ammonium anaerobically, with nitrite as electron acceptor. The higher the oxygen concentration in the surrounding medium, the deeper the oxygen will diffuse into the biofilm, so that the ratios of classical nitritation within the nitrogen conversion will increase.

Currently, two different groups of organisms with the capability for anaerobic ammonium oxidation are discussed in reference literature (cf. Chapter 2). Schmidt and Bock (1997) report of an anaerobic ammonium oxidation through *Nitrosomonas eutropha*. These micro-organisms oxidise ammonium when exposed to nitrogen dioxide, producing nitrite and NO. 40 to 60% of the produced nitrites are denitrified into $N_2$, with $N_2O$ emerging as intermediate product. The second group of organisms with the capability for anaerobic ammonium oxidation (ANAMMOX) are autotrophic bacteria, which hitherto have not been identified (Jetten and Van Loosdrecht, 1998). The anaerobic ammonium oxidation is described as having the same stoichiometric conversion ratio of ammonium and nitrite in the ANAMMOX process (1 : 1.31±0.06) as did occur in the anoxic batch tests presented here (1 : 1.37). Instead of the low nitrate conversion measured here, however, in the ANAMMOX process a small amount of nitrate is produced (Van de Graaf *et al.*, 1996). As in the batch tests there did exist an albeit rather low – endogenous denitrification potential in the mixed biocoenosis, it is conceivable that produced nitrate was converted directly, thus evading any measurements. In regard to the end product of the reaction – the emergence of $N_2$ without $N_2O$ as intermediate product – the anaerobic ammonium oxidation of the micro-organisms analysed here corresponds to the performance of the ANAMMOX organisms. In a study by Strous *et al.* (1997), it was possible with the ANAMMOX process being used to treat sludge liquor to reach a nitrogen conversion rate of 1.5 g N/(l . d). In the anoxic batch test (Figure 6), the achieved nitrogen elimination rate of 1.49 g N/(l . d) presented a potential of a similar dimension.

According to the latest findings, the aerobic/anoxic deammonification can be defined as a reaction in two steps: one part of the ammonium is taking part in a *nitritation*, the other in an *anaerobic ammonium oxidation* with obvious parallels to the ANAMMOX process. Ammonium and the nitrite produced during nitritation react into $N_2$. The fact that the biomass of the moving-bed pilot plant comes from the BC unit at Mechernich allows for the assumption that the reactions described here do also occur in the biological preliminary treatment stage at Mechernich.

For the use of the ANAMMOX process in wastewater technology, Jetten *et al.* (1997) propose a preliminary nitritation stage (SHARON Reactor) in order to produce nitrite as reaction partner for the anaerobic ammonium oxidation. The results presented here have conclusively proven for the first time that ammonium elimination through nitritation and anaerobic ammonium oxidation can be achieved as well in one biofilm system at low DO concentrations in the surrounding medium, provided that the biofilm is divided into an outer aerobic layer and an inner anoxic layer, so that ammonium oxidisers can settle preferably in the outer layer and organisms with the capability for anaerobic ammonium oxidation in the inner layer. Several current microbiological research projects aim to prove this hitherto only theoretical consideration. The current knowledge allows for the assumption that for wastewater with high nitrogen contents and low C/N ratio a complete nitrogen elimination can be achieved in one reactor. By an exact setting of the DO concentration it is possible to go for either the nitritation or the anaerobic ammonium oxidation as reaction. In a cycle with initially higher oxygen contents, it would be possible to produce at first mainly nitrite, which in the further course would at lower oxygen concentrations be used in the anaerobic ammonium oxidation. The nitrite concentration would be a possible control parameter for this single stage biological nitrogen removal.

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