Bioremediation of Ammonia from Polluted Waste Waters- A Review

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Received September 20, 2014; Revised October 20, 2014; Accepted November 13, 2014

Abstract Global water pollution due to industries became a major concern now. Some industrial facilities generate ordinary domestic sewage that can be treated in municipal sewage systems whereas industries that generate waste waters high in conventional pollutants like ammonia need exclusive treatments for their removal from the effluents before discharging. Ammonia can be removed by means of physical, chemical, biological methods. Out of all the methods, bioremediation appears as a promising tool for ammonia pollution as it is a cost effective and converts ammonia into harmless dinitrogen gas. This paper mainly focuses on bioremediation methods available for the removal of ammonia from the industrial effluents and their drawbacks.

Keywords: ammonia pollution, biological methods, SND, Anammox, parameters

Cite This Article: Sheela. B, and Khasim Beebi. Sk, "Bioremediation of Ammonia from Polluted Waste Waters- A Review." *American Journal of Microbiological Research*, vol. 2, no. 6 (2014): 201-210. doi: 10.12691/ajmr-2-6-6.

1. Introduction

Ammonia is a compound of nitrogen and hydrogen with the formula of NH_3 . Ammonia is used either directly or indirectly in industries. Even though its wide use, it is caustic and hazardous in nature. Ammonia nitrogen discharges from municipal, industrial and agricultural wastewater sites, which subsequently mixes into water resources. The raw water with high ammonia concentration must be treated before it reaches the consumer and also the wastewater discharged into the receiving water bodies. The most widely used methods for ammonia removal from wastewaters are adsorption, chemical precipitation, reverse osmosis, air stripping, ion exchange, breakpoint chlorination and biological nitrification and denitrification (Metcalf and Eddy, 2003). Advanced oxidation processes (AOPs) such as direct photo oxidation (Wang. A. *et al*; 1994), ozone oxidation (Holgne. J. *et al*; 1978, Yang. M. *et al*; 1999), photocatalytic oxidation (Bravo. A. *et al*; 1993, Bonsen. E.M. *et al*; 1997, Gootzen. J.F.E *et al*., 1998, Zhu. X.; *et al*; 2005) electrochemical oxidation (Della monica. M. *et al*; 1980, Kim. K. *et al*; 2006). In oxidation process, the end product is nitrate which is more toxic to aquatic life. Each of these methods have disadvantages like most prominently, costs are too high, since a continuous input of expensive chemical reagents are required to maintain the operation of most AOPs systems. Conventional wastewater treatment technologies are expensive to build and also have associated operational and maintenance problems. For example, by air stripping method, it is

feasible to reduce ammonia level to 1 mg/L but special requirements of the process such as water temperature of over 15°C and carbonate deposition are disadvantages of this system. Thus both physic-chemical and biological technologies have been applied for the elimination of ammonia from wastewaters for long period (Bonmati and Flotats., 2002, Sugiyami *et al*; 2005). However, with the ever stringent discharge limits of ammonia and nitrate (50 mg/L $\text{NH}_4\text{-N}$, 10 mg/L $\text{NO}_3\text{-N}$) for inland surface waters, it is essential to remove them to permissible levels. Biotreatment, being cost effective, is normally adopted for wastewater treatments. There are many advantages of bioremediation over conventional methods. Here in this review overall biological methods used for ammonia removal and the factors which effects the ammonia removal from the polluted waters was discussed.

2. Biological Process for Ammonia-Nitrogen Removal

2.1. Nitrification/Denitrification

Biological autotrophic nitrification followed by heterotrophic denitrification has long been applied for municipal and industrial wastewaters. For application of industrial wastewaters it becomes very difficult as composition and characteristics of effluents varies from industry to industry and even day to day. Under strict aerobic conditions, complete nitrification is carried out in two sequential oxidative stages-ammonia is first converted to nitrite by ammonia oxidizing bacteria and this nitrite is again converted into nitrate by nitrite oxidizing bacteria.

Each of the oxidizing steps is carried out by different genera of bacteria which use ammonia or nitrite as electron acceptor, while carbon dioxide is used as carbon source. The most common genus of bacteria that carries out ammonia oxidation is *Nitrosomonas* species. Ammonia oxidizers are genetically diverse but related to each other and are found in the beta subdivision of proteobacteria. The nitrite oxidation is carried out by *Nitrospina*, *Nitrospina*, *Nitrococcus*, and *Nitrocystis*. The most known genus of nitrite oxidizer is *Nitrobacter* which is genetically closely related to alpha subdivision of proteobacteria (Rittmann B.E., 2001). In the subsequent process denitrification is performed by heterotrophic denitrifiers under anoxic conditions. The oxidized nitrogenous compounds (nitrite or nitrate) are reduced to gaseous nitrogen by heterotrophic microorganisms that use nitrite or nitrate instead of oxygen as electron acceptor and organic matter as carbon and energy. Denitrification is common among the gram negative bacteria such as *Pseudomonas*, *Alcaligenes*, *Paracoccus* and *Thiobacillus*. Some gram positive bacteria (like *Bacillus*) and a few halophilic archaeal microorganisms (*Haloferax denitrificans*) are able to denitrify (Kim et al., 2005, Cyplik et al., 2007). Denitrifiers are ubiquitous in nature (Szekeres et al., 2005) and several researchers cultivated them using mixed cultures taken from wastewater treatment plants as seed. Denitrification is a slow process particularly for industrial wastewaters that contains high concentrations of nitrate (Foglar et al., 2005) but the process gained huge interest as it is environment friendly and cost effective. Organic carbon sources used for heterotrophic denitrification include both solid and liquid form but mostly aqueous forms are used for wastewater treatment plants. Among the most common form of carbon source are methanol, ethanol and acetic acid which are used for wastewaters and drinking waters (Gomez et al., 2003, Osaka et al., 2008, Feleke et al., 2002). A combination of carbon source using methanol and acetic acid gave better results for nitrogen removal and also showed better settling of sludge compared to use methanol and acetic acid alone (Cho et al., 2004). A lot of research work is going on to speed up the denitrification process through different strategies in which better contact of nitrate with microorganisms in wastewaters was maintained such as packed bed reactors (Daniel et al., 2009), rotating biological contactors (Teixeira et al., 2005), membrane biofilm reactor (Tereda et al., 2006).

2.2. Simultaneous Nitrification and Denitrification

SND process starts with the partial nitrification of ammonia and to nitrite and subsequently continuous with a direct reduction nitrite to nitrogen gas (Jenicek et al., 2004, Lai et al., 2004). This process is operated in a single stage with identical operational conditions which reduces large reactor volumes and energy costs for recirculation that are required for aerobic and anoxic systems. Several research units have been proposed for SND process in a single reactor (Guo et al., 2005) and Zhang et al., introduced a flexible biofilm reactor with adjustable aerobic, buffer and anoxic zones with liquid circulation being dependent on aeration flow rate and both the experiments were successful providing proof for

possibility of SND in a single reactor. In SND process optimum pH can be maintained without the addition of acid or base as the optimum pH for nitrifiers and denitrifiers lies between 7 and 8.5 (Chang et al., 1999) and it also reduces cost for pH alterations. Polymeric beads, in which nitrifiers and denitrifiers were immobilized, were used to remove nitrogen and ammonia from wastewaters in a single step. Uemoto and saiki et al., 2000, investigated a novel immobilized cell bioreactor containing packed gel envelopes capable to carryout SND process. The packed gel envelopes consisted of two polymeric gel plates with an internal space for injecting electron donor for denitrification. Ammonia oxidizer *Nitrosomonas europaea* and a denitrifier namely *Paracoccus denitrificans* were co-immobilized in a plate gel. The immobilized *N.europaea* oxidizes ammonia to nitrite on the outer surface of the plate that was in aerobic contact with wastewater containing ammonia, immobilized *P.denitrificans* reduce nitrite to nitrogen gas on the inside of the plate that was in anaerobic contact with the electron donor. Additional aeration step was not required for this process as electrons were directly injected into the internal space between the plates. This resulted in increase in utilization of electron donor for denitrification process and decrease in sludge quantity was observed. In further attempt, a 30 packed gel envelopes were implemented for thermal power plant for the removal of nitrogen from ammonia containing desulfurization wastewater. The reactor achieved 95% nitrogen removal with continuous wastewater system and the outlet concentration was 9 mg N/L (Morita et al., 2008). Cao et al., 2004 constructed a shell and tube co-immobilized bioreactor in which PVA material was used as immobilizing media and it was stable even after 3 months of operation proving an excellent option for nitrogen removal from wastewaters. Sun.J et al., in 2011 compared PVA and sodium alginate with immobilized nitrifiers and found PVA showed better results compared to sodium alginate beads. Though sodium alginate showed poor mechanical strength its mass transfer capacity was excellent than PVA. Recent research in immobilization field showed PVA as suitable material for ammonia remediation work. Agglomeration is the big problem with PVA material but this can be resolved by incorporation with cellulosic polymers (Antczak.M and Galas.E in 2009).

2.3. Autotrophic Nitrification/Denitrification

Generally conventional biological nitrogen removal process is used for treating wastewaters with relatively low nitrogen concentrations (Total nitrogen concentrations less than 100 mgN/L). Mulder.A. (2003) stated that biological treatment by autotrophic nitrogen removal is to be preferred for concentrated wastewater streams with high ammonia concentrations in the range of 100-5000 mgN/L. Autotrophic denitrifiers utilize inorganic carbon compounds (carbon dioxide or bicarbonate) as a sole source of carbon. Autotrophic denitrification is more advantageous than heterotrophic denitrification due to low biomass yield, sludge production is very low resulting in the reduction of reactor clogging and easier post treatment (Rijn et al., 2006). A lot of research work has been done on elemental sulphur (Zeng et al., 2005, Wan et al., 2009)

and hydrogen (Mansell *et al.*, 2002, Rezaia *et al.*, 2007) as an electron donor for autotrophic denitrification. In general *Thiobacillus denitrificans* and *Thiomicrospira denitrificans* are the two most autotrophic denitrifiers commonly reported (Brettar *et al.*, 2006). In nature these microorganisms encounter both heterotrophic and autotrophic denitrification. In bench scale studies, nitrified wastewater was denitrified by *Thiobacillus denitrificans* in packed bed reactors containing elemental sulphur (Koenig *et al.*, 2001). The feasibility of autotrophic denitrification was investigated and it was accomplished that its applicability depends on costs of sulphur and methanol (Quan *et al.*, 2005). It was noted that autotrophic denitrification may be more stable than some heterotrophic systems during transient nitrate loadings. In heterotrophic systems addition of carbon source must be controlled to prevent over and under dosing but in case of autotrophic denitrification addition of elemental sulphur in excess may remain within the reactor in the solid form.

2.4. Nitrification /Denitrification via Nitrite Pathway

To meet ever stringent discharge standards, new technologies and application strategies for the removal of ammonia from wastewaters have been implemented. Partial nitrification to nitrite was reported to be technically feasible and economically favorable especially when wastewaters containing high ammonia concentrations with low C/N ratio are treated. Various novel biological processes such as short cut nitrification and denitrification, anaerobic ammonia oxidation, completely autotrophic removal over nitrite (CANON), oxygen limited autotrophic nitrification and denitrification (OLAND), have been developed (Verstraete and Philips 1998). Partial nitrification is a critical procedure as nitrite acts as an intermediary substrate (Philips *et al.*, 2002). Out all the process only SHARON process gained interest due to its successful operation in practical. Partial nitrification and denitrification via nitrite depends on the factor partial nitrification up to nitrite is followed by nitrite denitrification. Partial nitrification can be selectively obtained by inhibiting nitrite oxidizing through appropriate regulation of the systems DO concentration, SRT, pH, temperature, substrate concentration and load, operation and aeration pattern, and inhibitors. SHARON process is the first successful process in which nitrification and denitrification via nitrite as an intermediate has been achieved under stable conditions and operated in full-scale operation (Mudler *et al.*, 2001). Corral *et al.*, (2001) studied the effect of salts and acetate on the SHARON bioreactor treating ammonia rich wastewaters. The study demonstrated acetate concentrations above 0.3 mg TOC/L and salt concentration above 100 mM resulted inhibition of nitrification in ammonia rich wastewaters. However the operation conditions for SHARON, such as high temperature and high ammonium concentrations, limits its development and application (STOWA, 1995). This process can only be applied to stage on rejection water of sludge digestion and leachate of landfill site. To date only successful operation was achieved in sequence operation and a few was with continuous flow process (Peng *et al.*, 2003a).

2.5. Anammox Bacteria

The anammox process was first described in bioreactors of wastewater treatment plants (Van de graaf *et al.*, 1995, Mudler *et al.*, 1995) where ammonium reacts with nitrite to produce dinitrogen gas.

This process attained special attention as it is an efficient biological alternative to the conventional nitrification and denitrification for the nitrogen removal from wastewaters. In comparison to conventional nitrification and denitrification, Anammox bacteria requires no external carbon source, they take carbon dioxide from water, 50% less oxygen, therefore low operation costs (Tal *et al.*, 2006). These bacteria belong to the order planctomycetales. They are the most primitive group of bacteria and occupy the base of the bacterial tree. They are gram negative, uncultivable, first isolated through density gradient centrifugation (Van de graaf *et al.*, 1996, Strous *et al.*, 2002). Their distinctive phenotypic characteristics involve red color, budding production, crateriform structure on the cell surface, intracellular compartment, anammoxosome, intra cytoplasmic membrane containing ladder lane lipids (Van de graaf *et al.*, 1996, Linsey *et al.*, 2001, Sininghe Damste *et al.*, 2002). They have a coccoid shape with a diameter of less than 1 µm and generation time of 10-30 days (Van Niftrik *et al.*, 2004). The Anammox bacteria are characterized by their slow growth rate and low biomass yield (Strous *et al.*, 1998). they have relatively high affinity for the substrates. Nitrite is more toxic to them than ammonium in a pH range of 6.7-8.3 (Strous *et al.*, 1999b, Dapena mora *et al.*, 2007). The Anammox bacteria can with stand partial oxygen pressure lower than 0.5% air saturation. At high levels of dissolved oxygen concentrations (>18%), the biological reaction becomes irreversible (Strous *et al.*, 1997, Egli *et al.*, 2001). Table 1 show Anammox bacteria discovered up to date.

Table 1.

Genus	Species	Sources
Brocadia	Candidatus Brocadia anammoxidans (Strous <i>et al.</i> , 1999a)	Wastewater Wastewater
	Candidatus Brocadia fulgida (Kartal <i>et al.</i> , 2004)	
Kuenenia	Candidatus Kuenenia stuttgartiensis (Penton <i>et al.</i> , 2006)	Wastewater
Scalindua	Candidatus scalindua brodae (Schmid <i>et al.</i> , 2003)	Wastewater Wastewater Sea water
	Candidatus scalindua wagneri (Schmid <i>et al.</i> , 2003)	
	Candidatus scalindua sorokinii (Schmid <i>et al.</i> , 2003)	
Others	Candidatus jettenia asiatica (Tsushima <i>et al.</i> , 2007)	Not specified Synthetic wastewater
	Candidatus anammoxoglobus propionicus (Kartal <i>et al.</i> , 2007)	

2.6. Anammox Process

Industrial application of anammox process is still limited due its slow growth rate and low yield of anammox biomass. Even a slight loss of biomass during washout with the effluent could reinitiate the start up period. Due to this condition, it is highly recommended to use a reactor with high biomass retention capacity. Till now a large range of bioreactors were studied for the

cultivation of anammox bacteria which are fluidized bed reactors, SBR, gas lift reactors, fixed bed reactors, UASB reactors (wyffels *et al.*, 2004, strous *et al.*, 2002). Out of all the reactors sequencing batch reactor (SBR) was found to be suitable for anammox culturing for its efficient biomass retention, stability and reliability for its long term operation, simple operation, stable under substrate limiting condition and high nitrogen conversion (van dongen *et al.*, 2001, jetten *et al.*, 1993, strous *et al.*, 1998). Strous *et al.* (1997) conducted experiments with FBR and fixed bed reactor with sand and glass particles as supporting media for Anammox bacteria but could not prevent biomass loss floating sludge caused by entrapped gas bubbles. Dapena mora *et al.* stated that mechanical stirring in SBR could effectively solve the gas entrapped in the granules compared less abrasive stress in gas lift reactor and further more use of non-woven fibers can increase the biomass retention. It was proven in several studies that use of non-woven fibers demonstrated a short startup period and high nitrogen removal rates (Van niftrik *et al.*, 2004, Isaka *et al.*, 2006). Van der star (2008) stated that MBR is a powerful tool for anammox research as high production of almost pure anammox bacteria can be obtained avoiding the diffusion limitations within the flocs or granules. The efficiency of the anammox process in a bioreactor depends on the biomass retention capacity and availability of substrate concentration in wastewaters. The nitrite concentration is a crucial factor for growth of anammox biomass: a too low nitrite concentration leads to substrate limitation and too high nitrite concentration above 20 mgN/L can lead to inhibition of the process. Furthermore anammox is an anaerobic process; the absence of oxygen is an essential step for the startup of the reactor. To fasten up the startup period, Slikers *et al.* (2003) used inoculum from fully activated anammox sludge for SBR and their start up period was 14 days.

2.7. Partial Nitrification/Anammox

The increase in the costs of wastewater treatment systems are challenged by the novel microbial process like combination of anammox and partial nitrification process in one reactor namely CANON process (Complete Autotrophic Nitrogen removal Over Nitrite) (Third *et al.*, 2001). This combination is a promising new method of removing nitrogen from wastewaters with low C/N ratio and high ammonia concentrations (Ciudad *et al.*, 2005). Compared to conventional nitrification and denitrification process, more than 62.5% less oxygen demand and no organic carbon requirement in combined partial nitrification/anammox process (Feng *et al.*, 2007). In CANON systems, *Nitrosomonas* like ammonia oxidizing bacteria and *planctomycetales* like anammox bacteria perform two sequential reactions under oxygen limited conditions (Slikers *et al.*, 2002). The nitrifiers convert ammonia to nitrite and consume oxygen creating anaerobic conditions for anammox bacteria. The produced nitrite is utilized with the remainder of ammonium by anammox bacteria and converts it into nitrogen gas (Nielsen *et al.*, 2005). Typically in the CANON process the *Nitrosomonas* like bacteria are active in the outer aerobic region of biofilm and in aggregates, while anammox are active in inner anoxic zone. In this way anammox are protected from oxygen which is consumed

in the outer aerobic zone. Oxygen can inhibit anammox activity (Strous *et al.*, 2007). The CANON process has quite sensitive operational characteristics for DO, the nitrogen surface load, biofilm thickness and temperature (Hao *et al.*, 2004). The oxygen mass transfer efficiency from gas to the liquid phase and effective biomass retention are the two key factors for the operation of CANON system (Slikers *et al.*, 2003). Moreover autotrophic ammonia oxidizers grow slowly than heterotrophic bacteria with which they have to compete for oxygen and if the retention time is not long, they will be washed out of the reactor. The biomass concentration can be increased by recirculation of sludge after sedimentation but limited by the efficiency of the sedimentation of the vessel. To overcome these factors along with environmental factors, immobilization technique was adopted. This became an important confront in order to scale up CANON systems from laboratory to industrial application (Gonz *et al.*, 2007). With the immobilization, biomass can be retained in the reactor without washing out and also provides protection from toxic shocks and adverse temperatures. This helps in maintaining year round treatments (Morita *et al.*, 2007). Immobilized biomass can be divided into naturally attached biomass (Biofilm) and artificially immobilized biomass. Biofilm are widely used for wastewater treatment plants. To avoid particle settlement to the bottom of the reactor, membrane aerated biofilm reactors were developed in which hydrophobic, gas permeable membranes are used for bubble less oxygen transfer (Lapara *et al.*, 2006). Membrane aeration is more advantageous than conventional bubble gas transfer mechanism as gas transfer efficiencies are much higher. In MABR, the micro porous membrane carries two functions: oxygen gas supplemental material and carrier for immobilization media (brindle *et al.*, 1996). It was recently proved that thick membrane aerated biofilm can simultaneously provide favorable conditions for nitrification and denitrification within a single biofilm (Cole *et al.*, 2004). Gong *et al.*, (2007) developed a new MABR in which a micro porous carbon tube membrane was supported with non-woven fabrics and investigated its feasibility and performance of CANON type process in single stage autotrophic nitrogen removal to treat the synthetic ammonium rich wastewaters like anaerobic sludge liquid. This reactor supplied air through the micro porous carbon tube to the biofilm supported by the non-woven fabric. The system worked with partial nitrification and dissolved oxygen consumption in the inner layer and anammox in the outer layer of non-woven fabric realizing feasibility of autotrophic nitrogen in a single reactor. This study demonstrated MABR was an excellent option for single stage autotrophic nitrogen removal. For immobilization technique most commonly gel entrapment is used but natural and synthetic polymers are also been used. Rostron *et al.*, (2001) studied on comparison between kaldnes, linpor, PVA materials for high strength ammonia synthetic wastewaters (500-1000 mg/L) and demonstrated that PVA is an excellent immobilization media for high strength ammonia containing wastewaters. The immobilization media should support and fulfill various criteria's like photo transparency, retention of cellular viability, non toxicity and stability during culturing in medium. Usually immobilization techniques

are commonly used for immobilizing pure strains because mechanism of the strain can easily be understood (Hill *et al.*, 2008). Immobilization of activated sludge has also been reported (Isaka *et al.*, 2007). Yan *et al.*, (2009) studied partial nitrification and degradation of organics with an immobilized biomass in ammonium rich organic wastewaters. This served as a first step for anammox process with partial nitrification via nitrite. They used four immobilization materials: sodium carboxymethylcellulose, sodium alginate, polyvinyl alcohol and sodium alginate and chitosan for entrapping the biomass. Sodium alginate was selected as best material after comparing the partial denitrification rates and adsorption efficiency.

2.8. Factors Effecting Ammonia Removal Efficiency

2.8.1. Temperature

When treating sanitary landfill leachate containing an ammonia concentration of over 2200 mg N/L, were investigated at operating temperatures down to 10°C. When the operating temperature was decreased from 20-17°C, an approximate 15% decrease in denitrification was observed immediately without any noticeable effect on nitrification. With the temperatures below 24°C, nitrification also stopped and methanol loading rate was decreased progressively. At 100C the system suffered major nitrification and denitrification inhibition. Changes in the operation parameters such as decrease in influent ammonia concentration and methanol loading, with increase of temperature from 10-15°C did not improve the system performance (Cannavo *et al.*, 2004). Using pure cultures, the optimum temperature was 35°C for AOB and 38°C for NOB (Cameilia and Gunnel 2001). The SHARON process pre mentioned was operated at 35°C (Mudler *et al.*, 2001). Present reports show that high temperatures of 28-38°C is favorable for nitrogen removal via nitrite due to the fact that specific growth rate of AOB is higher than that of NOB. From the aspect of specific growth rate, only temperatures above 25°C are possible for ammonia oxidizers to effectively outcome the nitrite oxidizers (Brouwer *et al.*, 1996). Several authors found that the optimum temperatures for the operation of anammox process was around 30-40°C (strous *et al.*, 1999). But most of the operations with anammox process were carried at temperatures higher than 30°C (Imajo *et al.*, 2004). Recently it was proved that in a rotating biological contactor established with anammox was successfully operated at temperature around 20°C (Cema *et al.*, 2007). Rysgaard *et al.*, (2004) working with east and west coasts of Greenland, observed anammox activity between -2 and 30°C and optimum temperature being 12°C. Similar results were found by Dalsgaard and Thamdrup (2002) working with marine sediments from Skagerrak (Baltic-North sea). These results show that application of anammox process is restricted to effluents with temperatures around 30°C. Dosta *et al.*, (2008) evaluated the effects of moderately low temperatures on the stability of anammox process. First the short term effects of temperatures on anammox biomass were studied using batch tests and maximum activity was found at temperatures 35-40°C. Activity tests done at temperatures 45°C showed an irreversible loss due to the biomass lysis

and temperatures 30-15°C was selected for long term study. The system was successfully operated at temperature 18°C. When temperature was decreased to 15°C, nitrite accumulation was observed and system lost its stability.

2.8.2. pH

pH plays a key role in biological nitrification and denitrification. Most of the physiological activities of nitrifiers are between pH 7.3-8.0. Groeneweg *et al.*, 1994 studied on the effect of pH on ammonia oxidation of *Nitrosomonas europaea* and observed that maximum ammonia oxidation occurred at pH 6.7 and 7. If the pH drops less than 6.0 and more than 8.2 cease of nitrification process was observed (princic *et al.*, 1998). Walker *et al.*, (1979) isolated ammonia oxidizing autotrophic nitrifiers from acid soils of Bangladesh and Sri Lanka with pH 4-4.5 and found evidence of *Nitrosospira* nitrification in situ at pH 4.1. In 2008 Hill and Webster conducted pH tests on ATCC Strain 25978 *Nitrosomonas europaea* and determined the optimum pH was between 7 and 8 for this strain. Usually nitrification process is a pH sensitive and to maintain reasonable nitrification rates pH should be maintained from 7-7.2 (USEPA, 1993). Nitrification process also depends on COD loading rates. Presences of high COD levels suppress the nitrification rates in bioreactors. Peyton *et al.*, 2001 stated that complete nitrate reduction at 125 g/L of NaCl and at pH 9 was possible with ATCC strain 700597 *Halomonas campisalis* in brine solution regenerated from ion exchange resins with NaCl which contains high concentrations of nitrate which is unattainable to eliminate by conventional biological, chemical and physical processes. Compared to heterotrophic denitrification autotrophic denitrification consumes alkalinity and to control pH alkalinity should be supplied to the autotrophic denitrification. Mostly in research works sodium bicarbonate is used for alkalinity control. For wastewaters with low alkalinity, large amounts of sodium bicarbonate are required to carry out autotrophic denitrification process. An alternative for elemental sulphur particles is limestone (Koenig *et al.*, 2002).

2.9. DO Concentration

Biological nitrification and denitrification demands high rates of aeration as denitrifiers are facultative bacteria that require oxygen as their terminal electron acceptor. Due to this property they accept readily nitrogen oxides in absence of oxygen. In this aspect DO levels plays crucial role in nitrogen removal from industrial effluents. Cost of operation increases with aeration rates. High levels of DO can inhibit denitrification as the bacteria take oxygen over nitrite as terminal electron acceptor and this can influence the nitrogen removal rates (zumft *et al.*, 1997). Although it is necessary to enhance the activity of nitrifiers by providing high DO concentrations in a reactor but denitrifiers are affected. To avoid inhibitory effect on denitrification, operation of reactors at low DO levels should be selected. Experiments were performed on the activated sludge reactors with partial nitrification with different DO levels. Maximum nitrite accumulation was observed between 0.7-1.4 mg/L (Cuidad *et al.*, 2005 and Ruiz *et al.*, 2003). With these experiments, short term

effects of DO on partial nitrification in batch tests were studied (Cuidad *et al.*, 2005, Park *et al.*, 2004). Many researchers studied on SND process with low dissolved oxygen concentrations and the results revealed that SND is accessible with DO concentrations between 0.2-0.8 mg/L (Kwang ju *et al.*, 2006, Peng *et al.*, 2007). Guo *et al.* (2009) showed that at low DO concentrations AOB were the dominant nitrifying bacteria and none of the NOB were recovered from activated sludge.

2.10. Effect of Nitrate Concentration

High nitrate concentrations in wastewaters can inhibit denitrification process and led to termination of denitrification to form N_2O (Blackmer *et al.*, 1978). A few of the research work has been done on high nitrate wastewaters till now. Denitrification with high nitrate is a rather slow process as it requires enhancement of physical and biological parameters and process must be optimized. For high nitrate concentrations removal pre acclimatization of biomass is required. In support of this aspect Dhamole *et al.*, 2007 developed a process for degradation of high strength nitrate waste wherein the sludge from an effluent treatment plant of a fertilizer industry was acclimated to degrade nitrate waste as high as 9032 ppm- NO_3-N in a time period of 6 hours.

2.11. Salinity

The effect of high salt concentration on biological nitrification and denitrification has been previously studied (Campos *et al.*, 2002). High salt concentrations always have impact on nitrification and denitrification activities. It was reported that nitrification and denitrification activities were sustained by gradual acclimatization of fresh water sludge to high salt concentrations. Halophilic denitrifiers were isolated from long term acclimated sludge and higher denitrification rates were achieved with long term acclimated sludge (Yoshie *et al.*, 2006). Correl *et al.* (2002) studied on SHARON process in presence of salts and organic carbon and results demonstrated salt concentration above 100mM inhibited ammonia oxidation in SHARON bioreactor treating fish water effluents. Experiments on characterization of microbial community in an aerobic granular sludge reactor treating wastewaters showed AOB (*Nitrosomonas*) can sustain salt concentration of 33g/L and while NOB (*Nitrospira*) were severely affected (Bassin *et al.*, 2011). However partial nitrification/anammox process can be successfully applied for wide variety wastewaters like livestock wastewaters, landfill leachate, industrial wastewaters etc, but these waters contain high salt concentrations which were considered as an inhibition factors for biological nitrogen removal (van der Star *et al.*, 2007). Marine anammox bacteria belong to the genus *scalindua* have been detected in natural surroundings (Schmid *et al.*, 2007) and recently they were enriched in continuous culture system for more than one year (Van de Vossenberg *et al.*, 2008 and Nakajima *et al.*, 2008). Results from these studies suggest that anammox bacteria can inherently prefer high salt concentrations and live in high salt habitats and would be available for industrial purposes. Kartal *et al.*, 2006 enriched fresh water Anammox bacteria *Candidatus* *Kuenenia stuttgartiensis* in a high saline reactor with salt

concentrations of 30 g/L and showed stable growth. For acclimatization phase for salt tolerance Yang *et al.*, conducted lab scale experiments for 92 days in which reactor was fed with inorganic wastewaters with ammonia and nitrite with salt concentrations of 30 g/L and reactor showed a stable NRR of 4.5 ± 0.1 kg-Nm⁻³/day. This was the first report of high salt tolerance.

2.12. Biomass Concentration

Apart from the above mentioned factors which effect ammonia remediation biomass concentration is also another contributing factor. Autotrophic nitrifiers contribute less biomass when compared to heterotrophic nitrification. It is always an important aspect that bulk amount of biomass is more efficient than less biomass with good ammonia removal efficiency. This might be the biggest disadvantage for anammox bacteria as the growth rate for these bacteria are very slow. Dapena Mora *et al.*, used SBR to retain biomass of anammox and the results were successful with more ammonia removal. The highest removal rate of ammonia by anammox bacteria was reported by Sliekers *et al.*, in a gas lift reactor with granular sludge was 8.9 kgN/m³ d.

3. Conclusion

In this paper ammonia removal technologies are discussed in detail. Out of all technologies Anammox system was found to be feasible and extremely suitable for treatment of high concentrated ammonia containing industrial effluents. Anammox reduces additional carbon costs and aeration costs for industries unlike conventional nitrification and denitrification systems where operation at industrial level is more expensive. The main drawbacks for Anammox bacteria are their isolation in pure form and slow growth rate. Further research work is needed for Anammox isolation in pure form and for applicability of Anammox system for different industrial effluents.

References

- [1] Abeling, U.; Seyfried, C.F. Anaerobic-aerobic treatment of potato-starch wastewater. *Water science and Technology*, 28 (2): 165-176, 1993.
- [2] Ammonia in Ullmanns encyclopedia of Industrial Chemistry. Weinheim, Wiley-VCH, Max Appl, 2006.
- [3] Antczak, S.A., Galas, E. *Bacillus subtilis* cells immobilized in PVA cryogels. *Biomol Eng.* 17: 55-63, 2001.
- [4] Austermann-Haun, U.; Meyer, H.; Seyfried, C.; Rosen Winkel, K.H. Full scale experience with anaerobic/aerobic treatment plants in the food and beverage industry, *Water science and Technology*, 40 (1), 305-312, 1999.
- [5] Bassin, J.P.; Pronk, M.; Muyzer, G.; Kleerebezem, R.; Dezotti, M.; Van Loosdrecht, M.C.M. Effect of elevated salt concentrations on the aerobic granular sludge process: Linking microbial activity with microbial community structure. *Appl Environ Microbiol*, 77 (22): 7942-7953, 2011.
- [6] Blackmer, A.M.; Bremner, J.M. Inhibitory effect of nitrate on reduction of N_2O to N_2 by soil microorganisms. *Soil Biol Biochem*, 10: 187-191, 1978.
- [7] Bonmati, A.; Flotats, X. Air stripping of ammonia from pig slurry: characterization and feasibility as a pre- and post treatment to mesophilic anaerobic digestion. *Water manage*, 23 (3): 261-272, 2002.
- [8] Bensen, E.M.; Schroeter, S.; Jacobs, H.; Broekaert, J.A.C. photo catalytic degradation of ammonia with TiO_2 as photo catalyst in

- the laboratory and under the use of solar radiation. *Chemosphere*, 35, 1435-1445, 1997.
- [9] Bravo, A.; Garcia, J.; Domenech, X.; Peral, J. Some aspects of photo catalytic oxidation of ammonium ion by titanium dioxide. *J Chem Res*, 376-377, 1993.
 - [10] Brettar, I.; Labranz, M.; Flavie, S.; Botel, J.; Kousa, H.; Christen, R.; Hofle, M.G. Identification of *Thiomicrospira denitrificans*-like Epsilonproteobacterium as a catalyst for autotrophic denitrification in the central Baltic sea. *Appl Environ Microbiol*, 72: 1364-1372, 2006.
 - [11] Brindle, K.; Stephenson, T. The application of membrane biological reactors for the treatment of wastewaters. *Bio Technol Bioeng*, 49: 601-610, 1996.
 - [12] Brouwer, M.; Van Loosdrecht, M.C.M.; Heijnen, J.J. One reactor system for ammonium removal via nitrite. STOWA report, 96-01, Utrecht, Netherlands, (ISBN 90 74476 554).
 - [13] Camilla, G.; Gunnel, D. Development of nitrification inhibition assays using pure cultures of Nitrosomonas and Nitrobacter. *Water Res*, 35 (2): 433-440.
 - [14] Campos, J.L.; Mosquera-corral, A.; Saez, M.; Mendez, R.; Lema, J.M. Nitrification in saline wastewater with high ammonia concentration in an activated sludge unit. *Water Res*, 36: 2555-2560, 2002.
 - [15] Campos, J.L.; Mosquera-corral, A.; Saez, M.; Mendez, R.; Lema, J.M. Nitrification in saline wastewater with high ammonia concentration in an activated sludge unit. , 36: 2555-2560, 2002.
 - [16] Cannova, P.; Richaume, A.; Lafolie, F. Fate of nitrogen and carbon in the Vadose zone: insitu and laboratory measurements of seasonal variations in anaerobic respiratory and denitrifying activities. *Soil Biol Biochem*, 36: 463-478, 2004.
 - [17] Cema, G.; Wiszniowski, J.; Zebezyński, S.; Zablocka-Godłowska, E.; Raszka, A.; Surmacz-Gorska, J. Biological nitrogen removal from landfill leachate by deammonification assisted by heterotrophic denitrification in a rotating biological contactor (RBC). *Water Sci Technol*, 55: 35-42, 2007.
 - [18] Chang, Y.J.; Tseng, S.K. A double membrane system for simultaneous nitrification and denitrification in a single tank. *Lett Appl Microbiol*, 28: 453-456, 1999.
 - [19] Chemical engineering. News report on floral incident. Chemical engineering, July 1971, pp. 334. In Marshall, V.C. (1987) chemical Hazards. Ellis Horwood Ltd, ISBN 085312969X.
 - [20] Cho, E.; Molof, A. Effect of sequentially combining methanol and acetic acid on the performance of biological nitrogen and phosphorus removal. *J Environ Manage*, 73, 183-187, 2004.
 - [21] Chung, J.; Bae, W.; Lee, Y.; Ko, G.; Lee, S.; Park, S.; Investigation of the effect of free ammonia concentration upon leachate treatment by short cut biological nitrogen removal process in: IWA specialty symposium on strong Nitrogenous and Agro-wastewater. Vol. 1, Seoul, Korea, June 11-13, pp. 93-104, 2003.
 - [22] Ciudad, G.; Rubilar, O.; Munoz, P.; Ruiz, G.; Chamy, R.; Vergara, C.; Jaison, D. Partial nitrification of high ammonia concentration wastewater as a part of short cut biological nitrogen removal process. *Process Biochem*, 40: 1715-1719, 2005.
 - [23] Cole, A.C.; Semmens, M.J.; Lapara, T.M. Stratification of activity and bacterial community structure in biofilms grown on membranes transferring oxygen. *Appl Environ Microb*, 70: 1982-1989, 2004.
 - [24] Ciudad, G.; Rubilar, O.; Munoz, P.; Ruiz, G.; Chamy, R.; vergara, C.; Jeison, D. partial nitrification of high ammonia concentration wastewater as a part of short cut biological nitrogen removal process. *Process Biochem*, 40: 1715-1719, 2005.
 - [25] Ciudad, G.; Rubilar, O.; Munoz, P.; Ruiz, G.; Chamy, R.; vergara, C.; Jeison, D. partial nitrification of high ammonia concentration wastewater as a part of short cut biological nitrogen removal process. *Process Biochem*, 40: 1715-1719, 2005.
 - [26] Cyplik, P.; Grajek, W.; Merecik, R.; Kroliczak, P. Effect of macro/micro nutrients and carbon sources over the denitrification rate of haloferax denitrificans archaeon. *Enzyme Microbiol Technol*, 40, 212-220, 2007.
 - [27] Dalsgaard, T.; Thamdrup, B. Factors controlling anaerobic ammonium oxidation with nitrite in marine sediments. *Appl Environ Microbiol*, 68: 3802-3808, 2002.
 - [28] Daniel, L.M.; Pozzi, E.; Foresti, E.; Fabio Alexandre Chinalia, A. Removal of ammonium via simultaneous nitrification-denitrification nitrite short cut in a single packed bed batch reactor. *Bioresour Technol*, 100, 1100-1107, 2009.
 - [29] Dapena Mora, A.; Fernandez, I.; Campos, J.L.; Mosquera-corral, A.; Mendez, R.; Jetten, M.S.M. Evaluation of activity and inhibition effects on anammox process by batch tests based on the nitrogen gas production. *Enzyme Microbiol Technol*, 40 (4): 859-865, 2007.
 - [30] Della monica, M.; Agostizno, A.; Ceglie, A. An electrochemical sewage treatment process. *J Appl Electrochem*, 10, 527-533, 1980.
 - [31] Deng Petersen, P.; Jensen, K.; Lyngsie, P.; Hendrik, N.; Johansen. Nitrogen removal in industrial wastewater by nitration and denitration-3 years of experience, *Water science and Technology*, 47 (11): 181-188, 2003.
 - [32] Dhamole, P.B.; Nair, R.R.; D souza, S.F.; Lele, S.S. Gentrification of high strength wastewater. *Bioresour Technol*, 98: 247-252, 2007.
 - [33] Dosta, J.; Fernandez, I.; Vazquez-padin, J.R.; Mosquera-corral, A.; Campos, J.L.; Mata Alvarez, J.; Mendez, R. Short and long term effects of temperature on the anammox process. *J of Hazard Master*, 154: 688-693.
 - [34] Egli, K.; Fanger, U.; Alvarez, P.J.J.; Siegrist, H.; Van der meer, J.R.; Zehnder, A.J.B. Enrichment and characterization of an anammox bacterium from a rotating biological contactor treating ammonium rich leachate. *Arch Microbiol*, 175 (1): 198-207, 2001.
 - [35] Feleke, Z.; Sakakibara, Y. A bio electro chemical reactor coupled with adsorber for the removal of nitrate and inhibitory pesticide. *Water Res* 36, 3092-3102, 2002.
 - [36] Feng, Y.; Tseng, S.; Hsia, T.; Ho C chou, W. Partial nitrification of ammonium- rich wastewater as pretreatment for anaerobic ammonium oxidation (Anammox) using membrane aeration bioreactor. *J of Bio Sci Bioengin* 104: 182-187, 2007.
 - [37] Fliermans, C.B.; Bohlool, B.B.; Schmidt, E.L. Detection of Nitrobacter in natural habitats using fluorescent antibodies. *Appl Microbiol*, 27: 124-129, 1974.
 - [38] Foglar, L.; Briski, F.; Sipos, L.; Vukovic, M. High nitrate removal from synthetic wastewater with the mixed bacterial culture. *Bioresour Technol* 96, 874-888, 2005.
 - [39] Gill, K.I.; Choi, E. Nitrogen removal by recycle water, Nitration as an attractive for retrofit technologies in municipal wastewater treatment plant. *Water science and Technology*, 49 (5-6), 39-46, 2004.
 - [40] Gomez, M.A.; Galvaz, J.M.; Hontoria, E.; Gozalez-Lopez, J. Influence of concentration on biofilm bacterial composition from a denitrifying submerged filter used for contaminated ground water. *J Bio Sci Bioeng*, 95, 245-251, 2003.
 - [41] Gonz, Z.; Yang, F.; Liu, S.; Bao, H.; Hu, S.; Furukawa, K. Feasibility of a membrane- aerated biofilm reactor to achieve single-stage autotrophic nitrogen removal based on anammox. *Chemosphere*, 69: 776-784, 2007.
 - [42] Gonzalez, L.E.; Canizares, R.O.; Baena, S. Efficiency of ammonia and phosphorus removal from a Colombian agro industrial wastewater by the microalgae *Chlorella vulgaris* and *Scenedesmus dimorphus*. *Bioresour Technol*, 60: 259-262, 1997.
 - [43] Gootzen, J.F.E.; Wonders, A.H.; Visscher, W.; Van Santen, R.Q.; Van Veen, J.A.R.A. DEMS and cyclic voltammetry study of NH₃ oxidation on platinized platinum. *Electro chem. Acta*, 43 1852-1861, 1998.
 - [44] Groeneweg, J.; Sellner, B.; Tappe, W. Ammonia oxidation in Nitrosomonas at NH₃ concentrations near Km: effects of pH and temperature. *Water Res*, 28 (12): 2561-2566, 1994.
 - [45] Guo, H.; Zhou, J.; Su, J.; Zhang, Z. Integration of nitrification and denitrification in air lift bioreactor. *Biochem Engin J*, 23: 57-62, 2005.
 - [46] Guo, J.; Peng, Y.; Wang, S.; Zheng, Y.; Huang, H.; Wang, Z. Long term effects of dissolved oxygen on partial nitrification performance and microbial community structure. *Bioresour Technol*, 100: 2796-2802, 2009.
 - [47] Hao, X.D.; Van Loosdrecht, M.S.M. Model based-evaluation of COD influence on a partial nitrification-anammox biofilm (CANON) process. *Water Sci Technol*, 49: 83-90, 2004.
 - [48] Hill, B.C.; Khan, E.A. Comparative study of immobilized nitrifying and co-immobilized nitrifying and denitrifying bacteria for ammonia removal from sludge digester supernatant. *Water Air Soil poll*, 195: 23-33, 2008.
 - [49] Hill, D.R.; Webster, T. High pH inhibits nitrifying bacteria. American water works association, 21-22, 2008.
 - [50] Holgene, J.; Bader, H. Ozonation of water: Kinetics of oxidation of ammonia by ozone and hydroxyl radicals. *Environ Sci Technol*, 12, 79-84, 1978.
 - [51] Imajo, U.; Tokutomi, T.; Furukawa, K. Granulation of anammox microorganisms in up flow reactors. *Water Sci Technol*, 49: 155-163, 2004.

- [52] Isaka, K.; Sumino, T.; Tsuneda, S. High nitrogen removal performance at moderately low temperature utilizing anaerobic ammonium oxidation reactions. *J Bio Sci Bioeng*, 103; 486-490, 2007.
- [53] Isaka, K.; Date, Y.; Sumino, T.; Yoshie, S.; Tsuneda, S. Growth characteristics of anaerobic ammonium-oxidizing bacteria in an anaerobic filtered reactor. *Appl Microbiology and Biotechnology*, 70: 47-52, 2006.
- [54] Jenicek, P.; Svehla, P.; Zebranska, J.; Dohanyos, M. Factors affecting nitrogen removal by nitrification/denitrification. *Water science and Technology*, 49 (5-6): 73-79, 2004.
- [55] Jetten, M.S.M.; Strous, M.; Van de pas schoonen, K.T.; Schalk, J.; Van Dongen, U.G.J.M.; Van de Graaf, A.A.; Longmann, S.; Muyzer, G.; Van Loosdrecht, M.C.M.; Kunen, J.G. The anaerobic oxidation of ammonium. *FEMS Microbiology Reviews*, 22, 421-437, 1999.
- [56] Kalyuzhnyi, S.; Gladchenko, M. Sequenced anaerobic aerobic treatment of potato starch wastewater, *water science and Technology*, 28 (2): 165-176, 1993.
- [57] Kartal, B.; Koleva, M.; Arsov, R.; Vander star, W.; Jetten, M.S.M.; Strous, M. Adaptation of freshwater anammox population to high salinity wastewater. *J Bio Technol*, 126: 546-553, 2006.
- [58] Kartal, B.; Rattray, J.; Van Niftrik, I.A.; Van de vossenberg, J.; Schmid, M.I.; Webb, R.I.; Schouten, S.; Furest, J.A.; Damste, J.; Jetten, M.S.M.; Strous, M. *Candidatus "Anammoxoglobus propionicus"* a new propionate oxidizing species of anaerobic ammonium oxidizing bacteria. *Syst Appl Microbiol*, 30 (1): 39-49, 2007.
- [59] Kartal, B.; Van Niftrik, A.H.; Sliemers, O.; Schmid, M.C.; Schmidt, I.; Van de pas schoonen, K.; Cirpus, I.; Van de star, W.; Van Loosdrecht, M.; Abma, W. et al. Application eco-physiology and bio diversity of anaerobic ammonium-oxidizing bacteria. *Rev Environ Sci Bio/Technol*, 3 (3): 255-264.
- [60] Keller, J.; Subramaniam, K.; Gosswein, J.; Greenfield, P. Nutrient removal from industrial wastewater using single tank sequencing batch reactor. *Water science and Technology*, 35 (6): 137-144, 1997.
- [61] Kim, J.K.; Park, K.J.; Cho, K.S.; Nam, S.; Park, T.; Bajpai, R. Aerobic nitrification-denitrification by heterotrophic *Bacillus* strains. *Bioresour Technol*, 6, 1897-1906, 2005.
- [62] Kim, K.; Kim, Y.; Kim, I.; Park, G.; Lee, E. Electrochemical conversion characteristics of ammonia to nitrogen. *Water Res*, 40, 1431-1441.
- [63] Koenig, A.; Liu, L.H. Use of lime stone for pH control in autotrophic denitrification: continuous flow experiments in pilot scale packed bed reactors. *J Bio Technol*, 99: 161-171, 2002.
- [64] Kowalchuk, G.A.; Stephen, J.R. Ammonia oxidizing bacteria: a model for molecular microbial ecology. *Annu Rev Microbiol*, 55: 485-529, 2001.
- [65] Kroschwitz, I. and Howe-Grant, M. (Eds). Kirk-othmer encyclopedia of chemical technology, 4th edition, Vol 2: 638-691, 1995. John Wiley and sons Inc., New York, Chichester, Bristbane, Toronto, Singapore.
- [66] Kuai, L.; Verstraete, W. Ammonium removal by the Oxygen-Limited Autotrophic Nitrification-Denitrification system. *Appl Environ Microbiol*, 64 (11): 4500-4506, 1998.
- [67] Kwanyong, L.; Lee, C.G. Nitrogen removal from wastewaters by Microalgae without consuming organic carbon sources. *J Microbiol Biotechnol*, 12 (2): 979-985, 2002.
- [68] Lai, E.; Senkpiel, S.; Solly, D.; Keller, J. Nitrogen removal of high strength wastewater via nitrification/denitrification using a sequencing batch reactor. *Water Sci Technol*, 50, 27-33, 2004.
- [69] Lapara, J.M.; Cole, A.C.; Shanahan, J.W.; Semmens, M.J. The effect of organic carbon, ammoniacal-nitrogen, and oxygen partial pressure on the stratification of membrane aerated biofilms. *J Ind Microbiol Biotechnol*, 33: 315-323, 2006.
- [70] Lindsey, M.R.; Webb, R.I.; Strous, M.; Jetten, M.S.M.; Butler, M.K.; Forde, R.J.; Fuerst, J.A. Cell compartmentalization in planctomycetes: Novel types of structural organization for the bacterial cell. *Arch Microbiol*, 176 (6): 413-429, 2001.
- [71] Ma, L.; Sun, X.; Liang, X.; Cui, C. Biological nitrogen removal by nitrification-denitrification constructed rapid infiltration land system to treat municipal wastewater. *J Food Agri Environ*, 7 (3&8): 795-798, 2009.
- [72] Mansell, B.O.; Schroeder, E.D. Hydrogenotrophic denitrification in a micro porous membrane bioreactor. *Water Res*, 36: 4683-4690, 2002.
- [73] Marshall, V.C.; Major chemical hazards, Ellis Harwood Ltd, 1987, ISBN 085312969X.
- [74] Metcalf and Eddy. Wastewater engineering: treatment and reuse, 4th edition, Mc., Graw-Hill Co., New York, 2003.
- [75] Morita, M.; Kudo, N.; Uemoto, H.; Watanabe, A.; Shinozaki, H. protective effect of immobilized ammonia oxidizer and phenol-degrading bacteria on nitrification in ammonia and phenol containing wastewater. *Eng Life Sci* 7: 587-592, 2007.
- [76] Mulder, A.; Van de graaf, A.A.; Robertson, I.A.; Kunen, J.G. Anaerobic ammonia oxidation discovered in a denitrifying fluidized bed reactor. *FEMS Microbiol Ecol*, 16 (3): 177-184, 1995.
- [77] Mulder, J.W.; Van Loosdrecht, M.C.M.; Hellinga, C.; Van Kempen, R. Full scale application of the SHARON process for the treatment of rejection water of digested sludge dewatering. *Water Sci Technol*, 43 (11): 127-134, 2001.
- [78] Mulder, J.W.; Van Loosdrecht, M.C.M.; Hellinga, C.; Van Kempen, R. Full scale application of the SHARON process for the treatment of rejection water of digested sludge dewatering. *Water Sci Technol*, 43 (11): 127-134, 2001.
- [79] N. Sivaraman and Geeta sivaraman, Research news, *Current Science*, Vol. 84, No. 12, 25th June, 2003.
- [80] Nakajima, J.; Sakka, M.; Kimura, T.; Furukawa, K.; Sakka, K. enrichment of anammox bacteria from marine environment for the construction of a bioremediation reactor. *Appl microbiol Biotechnol*, 77: 1159-1166, 2008.
- [81] Nielson, M.; Bollmann, A.; Sliemers, O.; Jetten, M.S.M.; Schmid, M.; Strous, M.; Schmidt, I.; Larsen, L.H.; Nielson, L.P.; Revsbech, N. Kinetics, diffusional limitation and micro scale distribution of chemistry and organisms in a CANON reactor. *FEMS Microbiol Ecol*, 51: 247-256, 2005.
- [82] Obaja, D.; Mace, S.; Costa, J.; Sans, C.; Mata-Alvarez, J. Nitrification-denitrification batch reactor. *Bio resour Technol*, 87, 103-111, 2003.
- [83] Osaka, T.; Shirotani, K.; Yoshie, S.; Tsuneda, S. Effects of carbon source on denitrification efficiency and microbial community structure in a saline wastewater treatment process. *Water Res* 42, 3709-3718, 2008.
- [84] Park, H.D.; Noguera, D.R. Evaluating the effect of dissolved oxygen on ammonia-oxidizing bacterial communities in activated sludge. *Water Res*, 38: 3275-3286, 2004.
- [85] Peng, Y.Z.; Chen, T.; Tian, W.J. Nitrogen removal via nitrite at normal temperature in A/O process. *J Environ Sci Health*, part A, *Environ Sci Eng*, 39 (7): 1667-1680, 2003.
- [86] Penton, C.R.; Devol, A.H.; Tiedje, J.M. Molecular evidence for the broad distribution of anaerobic ammonium oxidizing bacteria in fresh water and marine sediments. *Appl Environ Microbiol*, 72 (10): 6829-6832, 2006.
- [87] Peyton, B.M.; Mormile, M.R.; Peterson, J.N. Nitrate reduction with *Halomonas campisalis*: kinetics of denitrification at pH 9 and 12.5% NaCl. *Water Res*, 35: 4237-4242, 2001.
- [88] Philips, S.; Laanbroek, H.J.; Verstraete, W. Origin causes and effects of increased nitrite concentrations in aquatic environments. *Reviews in environmental science and Biotechnology*, 1: 115-141, 2002.
- [89] Poo, K.; Jun, B.; Lee, S.; Woo, H.; Kim, C. Treatment of strong nitrogen swine wastewater at a full scale sequencing batch reactor. *Water science and Technology*, 49 (5-6): 315-323, 2004.
- [90] Princic, A.; Mahre, I.; Megusar, F.; Paul, E.A.; Tiedje, J.M. Effects of pH and oxygen and ammonium concentrations on the community structure of nitrifying bacteria from wastewater. *Appl Environ Microbiol*, 64 (10): 3584-3590, 1998.
- [91] Reginatto, V.; Teixeira, R.M.; Pereira, F.; Schmidell, W.; Furigo Jr, A.; Menes, R.; Etchebehere, C.; Soares, H.M. Anaerobic ammonium oxidation in a bioreactor treating slaughterhouse wastewater. *J Brazilian Chem Engin*, 22 (4): 593-600, 2005.
- [92] Rejn, J.V.; Tal, Y.; Schreier, H.J. Denitrification in recirculating systems: theory and applications. *Aquacul Eng*, 34: 364-376, 2006.
- [93] Reznia, B.; Olesz kiewi, Z.J.A.; Cicek, N. Hydrogen dependent denitrification of water in an anaerobic submerged membrane bioreactor coupled with a novel hydrogen delivery system. *Water Res*, 41: 1074-1080, 2007.
- [94] Risgaard, S.; Glud, R.N.; Risgaard-peterson, N.; Dalsgaard, T. Gentrification and anammox activity in Arctic marine sediments. *Limnol Oceanogr*, 49: 1493-1502, 2004.
- [95] Risgaard, peterson, N.; Langezaal, A.M.; Ingvarsen, S.; Schmid, M.C.; Jetten, M.S.M.; Op den camp, H.J. Metal evidence for

- complete denitrification in a benthic foraminifer. *Nature* 443:93-96, 2006.
- [96] Rittmann, B.E.; McCarty, P.L. Environmental biotechnology. Principles and applications 10020, McGraw-Hill, New York, NY; 2001.
- [97] Rostron, W.M.; Stuckey, O.C.; Young, A.A. Nitrification of high strength ammonia wastewaters: Comparative study of immobilization media. *Water Res*, 35:1169-1178.
- [98] Schmid, M.; Walsh, K.; Webb, R.; Rijpstra, W.I.C.; Van de pas schoonen, K.; Jan verbruggen, M.; Hill, T.; Muffet, B.; Furest, J.; Schouten, s.; et al. "*Candidatus scalindua Brodae*", sp nov., "*Candidatus scalindua wagneri*", sp nov., two new species of anaerobic ammonium oxidizing bacteria. *Syst Appl Microbiol*, 26 (4): 259-538, 2003.
- [99] Schmid, M.C.; Risgaard-peterson, N.; Van de vossenberg, J.; Kuypers, M.M.M.; Lavik, G.; Peterson, J.; Hulth, S.; Thamdrup, B.; Canfield, D.; Dalsgaard, T.; Rysgaard, S.; Sejr, M.M.; Strous, M.; de camp, H.J.M.O.; Jetten, M.S.M. Anaerobic ammonium oxidizing bacteria in marine environments: wide spread occurrence but low diversity. *Environ Microbiol*, 9: 1479-1484, 2007.
- [100] Schmidt, I.; Van spanning, R.J.M.; Jetten, M.S.M. Denitrification and ammonia oxidation by *Nitrosomonas europaea* wild-type, and NirK and NorB-deficient mutants. *Microbiol*, 150: 4107-4114, 2004.
- [101] Sezekeres, K.; Kiss, I.; Bejerano, T.T.; Soares, M.I.M. Hydrogen dependent denitrification in a two reactor bioelectro chemical system. *Water Res* 35, 715-719, 2001.
- [102] Sinnighe Damste, J.S.; Strous, M.; Rijpstra, W.I.C.; Hopmans, E.C.; Geenevasen, J.A.J.; Van Duin, A.C.T.; Van Niftrik, L.A.; Jetten, M.S.M. Linearly concentrated cyclobutane lipids from a dense bacterial membrane. *Nature*, 419(6908): 708-712, 2002.
- [103] Sliekers, A.O.; Derwort, N.; Gomez, J.L.C.; Strous, M.; Kuenen, J.G.; Jetten, M.S.M. Completely autotrophic removal of nitrogen over nitrite in a single reactor. *Water Res*, 36:2475-2482, 2002.
- [104] Sliekers, O.; Third, K.A.; Abma, W.; Kuenen, J.G.; Jetten, M.S.M. Canon and anammox in a gas lift reactor. *FEMS Microbiol Lett*, 339-344, 2003.
- [105] STOWA. Treatment of nitrogen rich return flows of sewage treatment plants. Evaluation of Dutch pilot plant research projects (in Dutch). STOWA report, pp 95-08, 1995.
- [106] Sun, J.; Liu, J.; Liu, Y.; Li, Z. Optimization of Entrapping Conditions of Nitrifying Bacteria and Selection of Entrapping Agent. Second International Conference on Environmental Science and Technology. IPCBEE. Vol.6. IACSIT press. Singapore, 2011.
- [107] Strous, M.; furest, J.A.; Kramer, E.H.M.; Logemann, S.; muyzer, G.; g Van de pas-schoonen, K.T.; Webb, R.; Kuenen, J.G.; Jetten, M.S.M. Missing lithotroph identified as planctomycete. *Nature*, 400 (6743): 446-449, 1999.
- [108] Strous, M.; Heijnen, J.J.; Kuenen, J.G.; Jetten, M.S.M.; and PQ. The sequencing batch reactor as a powerful tool for the study of slowly growing anaerobic ammonium oxidizing microorganisms. *Applied Microbiology and Biotechnology*, 50: 589-596, 1998.
- [109] Strous, M.; Heijnen, J.J.; Kuenen, J.G.; Jetten, M.S.M. The sequencing batch reactor as a powerful tool for the study of slowly growing anaerobic ammonium oxidizing microorganisms. *Applied Microbiology and Biotechnology*, 50, 589-596, 1998.
- [110] Strous, M.; Kuenen, J.G.; Fuerst, J.A.; Wagner, M.; Jetten, M.S.M. The anammox case-a new experimental manifesto for microbiological co-physiology. *Antonie Van Leeuwenhoek*, 81 (1/4): 693-702, 2002.
- [111] Strous, M.; Kuenen, J.G.; Jetten, M.S.M. Key physiology of anaerobic ammonium oxidation. *Appl Environ Microbiol*, 65 (7): 3248-3250, 1999.
- [112] Strous, M.; Van gerven, E.; Kuenen, J.G.; Jetten, M.S.M. effects of aerobic and micro aerobic conditions on anaerobic ammonium-oxidizing (Anammox) sludge. *Appl Microbiol*, 63 (6): 2446-3446, 1997.
- [113] Strous, M.; Van gerven, E.; Ping, Z.; Kuenen, J.G.; Jetten, M.S.M. Ammonium removal from concentrated waste streams with anaerobic ammonium oxidation (Anammox) process in different reactor configurations. *Water Research*, 31: 1955-1962, 1997.
- [114] Sugiyama, S.; Yokoyama, M.; Ishizuka, H.; Sotowa, K.I.; Tomida, T.; Shigemoto, N. Removal of aqueous ammonia with magnesium phosphates obtained from the ammonium elimination of magnesium ammonium phosphate. *J Colloid Interface Sci*, 292 (1): 133-138, 2005.
- [115] Tal, J.E.M.; Watts, J.; Schreir, H.J. Anaerobic ammonium oxidizing (Anammox) bacteria and associated activity in fixed-film biofilters of a marine recirculating aquaculture system. *Appl Environ Microbiol*, 72: 2896-2904, 2006.
- [116] Teixeira, P.; Oliveira, P. Denitrification in a closed rotating biological contactor. Effect of disk submergence. *Process Biochem*, 37, 345-349, 2001.
- [117] Terada, A.; Yamamoto, T.; Igarashi, R.; Tsuneda, S.; Hirata. Feasibility of a membrane-aerated biofilm reactor to achieve controllable nitrification. *Biochem Eng J*, 28, 123-130, 2006.
- [118] Third, K.A.; Olav Sliekers, A.; Kuenen, J.G.; Jetten, M.S.M. The CANON system (completely autotrophic removal of nitrogen over nitrite) under ammonium limitation: interaction and competition between three groups of bacteria. *Syst Appl Microbiol*, 24: 588-596, 2001.
- [119] Tsushmi, I.; Ogasawara, Y.; Kindaichi, T.; Satoh, H.; Okabe, S. Development of high rate anaerobic ammonium oxidizing (Anammox) biofilm reactors. *Water Res*, 41 (8): 1623-1634, 2007.
- [120] Uemoto, H.; Saiki, H. Nitrogen removal by tubular gel containing *Nitrosomonas europaea* and *Paracoccus denitrificans*. *Appl Environ Microbiol*, 62 (11): 4224-4228, 1996.
- [121] Uemoto, H.; Saiki, H. Nitrogen removal reactor using packed gel envelopes containing *Nitrosomonas europaea* and *Paracoccus denitrificans*. *Biotechnol Bioeng*, 67; 80-86, 2000.
- [122] Van de star, W.R.L.; Abma, W.R.; Blommers, D.; Mulder, J.W.; Tokutomi, T.; Strous, M.; Picioreanu, C.; Van Loosdercht, M.C.M. Startup reactors for anoxic ammonium oxidation: experiences from the first full scale anammox reactors in Rotterdam. *Water Res* 42: 4149-4163, 2007.
- [123] Van de star, W.R.L.; Miclea, A.I.; Van Dongen, U.G.J.M.; Muyzer, G.; Picioreanu, E.; Van Loosdrecht, M.C.M. The membrane bioreactor; a novel tool to grow anammox bacteria as free cells. *Biotechnology and Bio engineering*, 101:286-294, 2008.
- [124] Van de vossenberg, J.; Rattray, J.E.; Geerts, W.; Kartal, B.; Van niftrik, K.L.; Van donselaar, E.G.; Sinnighe Damste, J.S.; Strous, M.; Jetten, M.S.M. Enrichment and characterization of marine anammox bacteria associated with global nitrogen gas production. *Environ microbiol*, 10 (11): 3120-9, 2008.
- [125] Van dongen, U.; Jetten, M.; Jetten, M.S.M.; Van Loosdrecht, M.C.M. The SHARON Anammox process for treatment of ammonium rich wastewater. *Water Sci Technol*, 44 (1): 153-160, 2001.
- [126] Van niftrik, K.A.; Fuerst, J.A.; Damste, J.S.S.; Kuenen, J.G.; Jetten, M.S.M.; Strous, M.; and IK. The anammoxosome: an intra cytoplasmic compartment in anammox bacteria. *FEMS microbiology letter*, 233: 7-13, 2004.
- [127] Van Niftrik, L.A.; Sinnighe Damste, J.S.; Kuenen, J.G.; Jetten, M.S.M.; Strous, M. The anammoxosome an intracytoplasmic compartment in anammox bacteria. *FEMS Microbiology Letters*, 233: 7-13, 2004.
- [128] Verstraete, W.; Philips, S. Nitrification and denitrification process and technologies in new contexts. *Environ Pollution*, 102 (51): 717-726, 1998.
- [129] Wade, A.; Maher, B.; Lawrence, I.; Davis, N.; Zoppou, C.; Bell, C. Estimating the allowable ammonia concentrations in wastewater treatment plant discharge to ensure protection of aquatic biota. *Environ Technol*, 19 (7): 749-754, 1998.
- [130] Wan, D.; Liu, H.; Qu, J.; Lei, P.; Xiao, S.; Hou, Y. Using the combined bioelectro chemical and sulphur autotrophic denitrification system for ground water denitrification. *Bioresour Technol*, 100: 142-148, 2009.
- [131] Wang, A.; Edwards, J.G.; Davies, J.A. Photooxidation of ammonia with titania-based heterogeneous catalysts. *Sol Energy*, 52, 459-466, 1994.
- [132] Wyffels, S.; Boeckx, P.; Pynaert, K.; Zhang, D.; Van cleemput, O.; Chen, G.; Verstraete, W. Nitrogen removal from sludge reject water by a two stage oxygen limited autotrophic nitrification denitrification process. *Water Sci Technol*, 49 (5-6): 57-64, 2004.
- [133] Yan, Y.; Hu, Y.Y. Partial nitrification to nitrite for treating ammonium rich organic wastewater by immobilized biomass system. *Bioresour Technol*, 100: 2341-2347.
- [134] Yang, J.; Jhang, L.; Hira, D.; Fukuzaki, Y.; Furukawa, K. Anammox treatment of high salinity wastewater at ambient temperature.
- [135] Yang, M.; Uesugi, K.; Myoga, H. Ammonia reaction in a bubble column by ozonation in presence of bromide. *Water Res*, 33, 1911-1917, 1999.

- [136] Yoshie, S.; Ogawa, T.; Mokino, H.; Hirosawa, H.; Tsuneda, S.; Hirata, A. Characteristics of bacteria showing high denitrification activity in saline wastewater. *Lett Appl Microbiol*, 42: 277-283, 2006.
- [137] Zeng, H.; Zhang, T.C. Evaluation of kinetic parameters of a sulphur-limestone autotrophic denitrification biofilm process. *Water Res*, 39: 4941-4952, 2005.
- [138] Zhang, X.; Zhou, G.; Guo, H.; Qu, V.; Liu, G.; Zhao, L. Nitrogen removal performance in a novel combined biofilm reactor. *Process Biochem*, 42: 620-626, 2007.
- [139] Zhu, X.; Castleberry, S.R.; Nanny, M.A.; Butler, E.C. Effects of pH and catalyst concentration on photo oxidation of aqueous ammonia and nitrite in titanium dioxide suspensions. *Environ Sci Technol*, 39, 3784-3791, 2005.
- [140] Zumft, W.G. Cell biology and molecular basis of denitrification. *Microbial Mol Rev* 61: 533-616, 1997.