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Removal of ammonium and nitrate from cold inorganic mine water by fixed-bed biofilm reactors

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Abstract

Nitrogenous compounds, e.g. ammonium and nitrate, from various sources in extractive industry are often discharged with mine water to the aquatic environment. Ammonium and nitrate in blasting agents can dissolve in water from undetonated explosives and the nitrogenous compounds can negatively affect receiving water bodies. In this laboratory study, the removal of ammonium and nitrate from cold inorganic mine water was achieved by fixed-bed biofilm reactors at temperatures as low as 5 °C. Water from two underground mines was treated for ammonium removal in a nitrifying bioreactor. Ammonium concentration was from 2 to 83 mg NH_4^+–N/l. The sodium and chloride ion content was up to 0.8 and 2.2 g/l, respectively. At 5 °C, nitrification of up to 98% was reached at load of 0.33 g NH_4^+–N/m^2/d. The highest applied load was 2.42 g NH_4^+–N/m^2/d. The feed to the denitrifying bioreactor contained 12 to 86 mg NO_3^-–N/l. The anoxic methanol-fed denitrifying bioreactor reached up to 95% nitrate removal at loads as high as 0.91 kg NO_3^-–N/m^3/d and in combination with an anoxic unit a surface load of 4.26 g NO_3^-–N/m^2/d was applied at 5 °C. This is the first report on high-rate removal of ammonium and nitrate from cold inorganic mine water by fixed-bed biofilm reactors at low temperature.

Keywords: Bacteria; Biotechnology; Environmental; Mineral processing

1. Introduction

Ammonium and nitrate are usually discharged from mining operations to the aquatic environment. The main source of ammonium and nitrate in mine water originates from blasting agents such as ammonium nitrate fuel oil explosives (Forsberg and Åkerlund, 1999). Other sources of ammonium and nitrate in mine water are cyanide destruction, transformation of amines in flotation circuits, pH regulation agents, ammonium sulphate as eluent of uranium from ion exchange resins, ammonium hydroxide used in uranium precipitation, and ammonia used as lixiviant to recover copper and nickel in hydrometallurgical processes (EPA, 2003). In the European Union, no Best Available Technology has been defined for treatment of ammonium and nitrate in mine waters (EU, 2004). Several technologies exist for the removal of total nitrogen from water, including reverse osmosis (Awadalla and Kumar, 1994; Awadalla et al., 1994; Häyrynen et al., in press). The biological removal of total nitrogen by combined nitrification and denitrification is an established process for municipal and industrial wastewater treatment. Nitrification is the initial step of total nitrogen removal which proceeds by ammonium oxidation to nitrite (Eq. (1)) followed by nitrite oxidation to nitrate (Eq. (2)). Ammonia is oxidized by bacteria of the genera *Nitrosomonas* and *Nitrosospira* among others (Eq. (1))

$$NH_4^+ + 1.5O_2 \rightarrow NO_2^- + H_2O + 2H^+ \qquad \text{pH decrease} \tag{1}$$

Nitrite is oxidized by bacteria of the genera *Nitrobacter* and *Nitrospira* among others (Eq. (2))

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$$NO_2^- + 0.5O_2 \rightarrow NO_3^- \quad (2)$$

Biological denitrification proceeds by reduction of nitrate to dinitrogen gas by facultative, anaerobic, heterotrophic bacteria (Eq. (3))

$$2NO_3^- \rightarrow 2NO_2^- \rightarrow 2NO \rightarrow N_2O \rightarrow N_2 \quad (3)$$

For the denitrification in inorganic water, the heterotrophic bacteria require external carbon sources such as methanol (Eq. (4)). Methanol is often used, at a ration of 3 kg methanol/kg nitrate–nitrogen, due to its low cost and low excess biomass production (Tchobanoglous et al., 2003)

$$6NO_3^- + 5CH_3OH \rightarrow 3N_2 + 5CO_2 + 7H_2O + 6OH^- \quad (4)$$

The nitrification is affected by temperature (Chen et al., 2006) and nitrification rates double with 10 °C increase (Tchobanoglous et al., 2003). Denitrification is also strongly dependent on temperature, with denitrification rates doubling with every 4 °C increase (Given and Meyer, 1998). In municipal wastewater treatment plants, the total nitrogen removal performance is usually about 1 kg N/m^3/d (Tchobanoglous et al., 2003). The biological removal of ammonium and/or nitrate from mine water has been applied at a few mines in North America (Given and Meyer, 1998; Given et al., 1998; Reinsel and Plumb, 1999; Reinsel, 2001). Nitrification as part of biological cyanide destruction has been extensively studied (e.g. Akcil, 2003; White and Schnabel, 1998). However, the combined removal of ammonium and nitrate from water of mining and mineral processing plants has only been studied in suspended biomass systems as part of cyanide destruction. Since 1996, the Homestake's Nickel Plate mine in British Columbia, Canada, successfully applied a full-scale suspended sludge process for cyanide and thiocyanate removal from effluents of tailing facilities (Given and Meyer, 1998; Given et al., 1998). The biological destruction of cyanide and thiocyanate generates ammonium, which is biologically converted to dinitrogen gas via nitrification and denitrification. The bacteria involved in the nitrification and denitrification have long generation times and low cell mass yields. Therefore, in the applied suspended sludge process a high sludge age has to be maintained to achieve good performance results. The activated sludge process was designed for total nitrogen removal of about 20 mg N/l at flow rates of 0.7–1.1 m^3/min. The ammonium concentration is from 20 to 50 mg N/l and the nitrate content is from 100 to 130 mg N/l (Given and Meyer, 1998; Given et al., 1998). At Nickel Plate mine, nitrification requires temperature above 11 °C and the heating costs are about 5% of the total operational costs of about 0.01 euro/m^3. Methanol is used as a carbon source in the denitrification at a ratio of 3 kg methanol/kg nitrate–nitrogen and accounts for another 5% of treatment costs. However, reagents costs are only about 10% of the treatment costs with labor as the main costs of 40% (Given and Meyer, 1998; Given et al., 1998). Although plants treating water by suspended biomass processes perform successfully with organic wastewater, they are limited in treating dilute inorganic wastewaters. Reactors with biomass growing attached onto a carrier material, i.e. a biofilm, have several advantages over suspended biomass water treatment systems. These advantages are a relatively short treatment time, low maintenance costs, slow excess biomass generation with generally no waste biomass, and good resistance against low temperature, toxification and changing feed quality (pH, salinity, metals, biological and chemical oxygen demand content) (Rowan et al., 2003). The attachment property of bacteria to the carrier material in the biofilm reactor is a major parameter and minimizes biomass washout of slow-growing microorganisms at high flowrates. Further, biofilm reactors yield higher removal rates in smaller units. Attached bacteria usually show higher specific activity than suspended bacteria (Singh et al., 2006). Dictor et al. (1997) studied fixed-bed reactors for biological cyanide conversion to ammonium. So far laboratory studies have only applied synthetic mine effluents to study biological ammonium and nitrate removal at room temperature (e.g. Koren et al., 2000).

This study aims to: (i) establish and maintain nitrifying and denitrifying biofilms in bioreactors treating actual cold inorganic mine water, i.e. low in organic compounds, (ii) achieve high nitrogen removal rates in nitrifying and methanol-fed denitrifying biofilm reactors at temperature as low as 5 °C, and (iii) study the effect of two different mine waters on the performance of the biological nitrification.

2. Materials and methods

2.1. Bioreactor feed

Water was collected from the dewatering system of the ScanMining Ltd. Pahtavaara Gold Mine (mine I) and the Outokumpu Ltd. Kemi Chromite Mine (mine II) (Table 1). The two sites were chosen as representatives of underground mining in cold climate, i.e. Finnish Lapland. The mine water was sampled into 30 l or 220 l barrels and stored in the cold. The pH of the feed was about 8 (Table 1), which is favourable for the treatment of weakly buffered waters in nitrifying bioreactors (see Eq. (1)). During some tests on nitrification, the feed was at spiked with NH_4NO_3 to reach higher loading of the bioreactor. The nitrate content in the influent of the denitrifying bioreactor was increased by addition of HNO_3 and KNO_3 on day 301 and 307. The denitrifying bioreactor was supplemented with methanol as an external electron donor/carbon sources since the feed, i.e. mine water II, contained only minor amounts of organic carbon determined as chemical oxygen demand (COD_{Cr}) of 26.9 mg O_2/l. Furthermore, phosphate sources were supplemented to the feed of the denitrifying bioreactor after day 102.

2.2. Fixed-bed biofilm reactors

The nitrifying bioreactor combined a trickling filter, 12.8 dm^3, and a submerged fixed-bed reactor, 5.7 dm^3

Table 1
Composition of the water samples collected from dewatering system of two mines

Mine	pH	COD_{Cr} (mg O_2/l)	NO_3–N (mg/l)	NH_4–N (mg/l)	P (mg/l)	Cl (mg/l)	Li (mg/l)	Na (mg/l)	Mg (mg/l)	Al (mg/l)	S (mg/l)	K (mg/l)	Ca (mg/l)
I	8.3	8.3	17.5	24.2	0.08	15.1	0.01	25.17	18.20	<0.01	13.53	20.67	32.60
II	8.0	26.9	4.3	17.7	0.10	2251	0.03	772.13	112.03	0.36	83.56	132.33	467.60
	Mn (mg/l)	B (mg/l)	Fe (mg/l)	Co (mg/l)	Ni (mg/l)	Cu (mg/l)	Zn (mg/l)	As (mg/l)	Cd (mg/l)	Ba (mg/l)	Pb (mg/l)	Cr (mg/l)	Sr (mg/l)
I	0.03	0.10	0.01	0.01	<0.01	0.02	0.08	<0.01	<0.01	0.12	<0.01	<0.01	0.33
II	0.12	0.31	0.28	0.05	0.02	0.10	0.02	0.66	0.01	0.16	0.16	0.04	20.67

(Fig. 1). The column, height 1.2 m and diameter 0.14 m, was aerated from the bottom with air. The nitrifying bioreactor contained Hufo 200 plastic carrier disks with a total surface area of 1.9 m^2 and 50 g of lignite coke to support biofilm growth. From the effluent of the nitrifying bioreactor biomass and solids were retained in a clarifier, 1.7 dm^3. Clarified water was fed to the top of an anoxic unit designed as submerged fixed-bed reactor, 2.7 l, filled with Hufo 200 carriers with a total surface area of 0.5 m^2. The denitrifying bioreactor was a submerged fixed-bed of 6.8 dm^3 containing F2 polymer carriers with total surface of 1.1 m^2. The denitrifying bioreactor was operated in upflow mode (Fig. 1). The nitrifying bioreactor was started eight months prior to the denitrifying bioreactor. While mine water II was treated in the nitrifying bioreactor, its effluent was collected into a 200 l barrel for subsequent denitrification. The denitrifying bioreactor was fed with the effluent from the nitrifying bioreactor. The first batch of effluent from the nitrifying bioreactor was stored for one month prior to start-up of the denitrifying bioreactor. During storage denitrification was not occurring due to lack of organic carbon in mine water II. Nitrifying microorganisms are slow-growing, therefore the operating temperature for nitrification was step-wise decreased from 14 to 5 °C. The nitrifying bioreactor was maintained for 54 days at 14 °C followed by 43 days at 12 °C and finally 385 days at 5 °C, in total 482 days. The denitrifying bioreactor was operated for 337 days at 5 °C.

2.3. Inocula

The nitrifying reactor was inoculated with cultures from four nitrifying bioreactors: (1) Laboratory-scale nitrifying bioreactor treating synthetic wastewater of low BOD and high ammonia for one year at room temperature. (2) Laboratory-scale nitrifying bioreactor inoculated with sludge from a fishpond maintained with ammonium rich water, 50–100 mg/l, for one year at 8 °C. (3) Full-scale bioreactor treating cold municipal landfill in arctic climate. (4) Laboratory-scale nitrifying bioreactor treating landfill leachate, 1500–2000 mg/l NH_4^+–N for three months at 15–20 °C. The denitrifying bioreactor was inoculated with a mixed population of cold-tolerante methylotrophic denitrifying bacteria developed from activated sludge (Helsinki wastewater treatment plant) (Zaitsev et al., 2007). The mixed culture had earlier shown excellent performance in full-scale denitrification of cold landfill leachate (unpublished results) and biodegradation of methyl *tert*-butyl ether at low temperature (Zaitsev et al., 2007). The mixed culture had been adapted to methanol as external carbon source over a period of two months in the laboratory.

2.4. Analyses

Ammonium, nitrite, nitrate, total-phosphorus, chloride and COD_{Cr} concentrations were colorimetrically

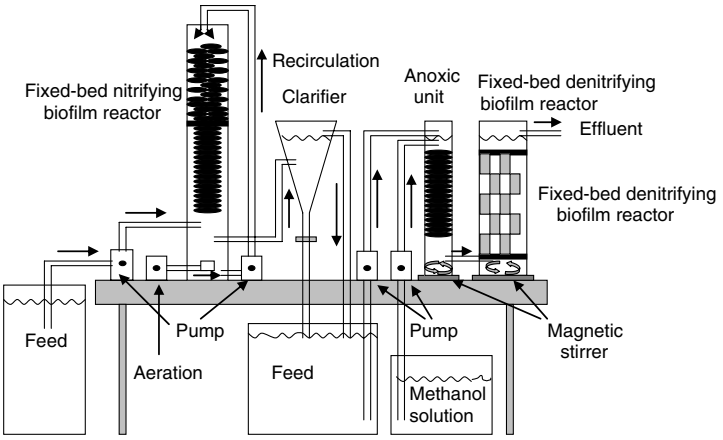

Fig. 1. Schematic of the experimental set-up with nitrifying and denitrifying fixed-bed biofilm reactors.

determined based on the method indophenol blue, sulfanilic acid, 2,6-dimethylphenol, phosphormolybdenum blue, iron(III)thiocyanate and chromosulphuric acid oxidation, respectively (LCK standard test kits, HACH Dr. Lange). The elemental composition of water samples was measured by inductively coupled plasma atomic emission spectroscopy (Thermo Jarrell Ash model ICAP 1100). The pH was measured with a SCHOTT model CG 842 pH meter.

3. Results and discussion

Removal of ammonium and nitrate from actual cold inorganic mine water was studied using fixed-bed biofilm reactors. The nitrifying bioreactor achieved high ammonium removal at high load and low temperature without pH adjustment or addition of external phosphorus sources. The effect of temperature and load on the nitrification was studied in a bench-scale bioreactor (Fig. 2). The ammonium and nitrate concentration in the influent and effluent of the nitrifying bioreactor were as shown in Fig. 3a and b. The concentrations of nitrogenous compounds in influent and effluent varied because real mine water was used, which was sampled at several occasions (Fig. 3b). Since nitrifying bacteria are sensitive to low temperatures the nitrification bioreactor was started by recirculation of mine water I for three days at 14 °C. Within a week of operation nitrification was achieved to 88%. The good growth and attachment of biomass in the nitrifying bioreactor is remarkable since the inorganic mine water contained only traces of organic compounds to support biofilm formation (Table 1). During phase II, 12 °C, and phase III, 5 °C, the feed was spiked with NH_4NO_3 to reach higher loading of the bioreactor. Nitrite concentrations were generally below 1 mg NO_2^-–N/l and only increased up to 6 mg NO_2^-–N/l in phase II. A step-wise decrease of the temperature resulted in temporary decrease of nitrification performance at 5 °C (Fig. 2). However, during phase III the nitrification was as high as 98% at a load of 0.10 kg NH_4^+–N/m^3/d or 0.33 g NH_4^+–N/m^2/d at 5 °C (Fig. 2). The latter units for load and removal rates (g N/m^2/d) best describes the performance of bioreactors with attached biomass. The nitrification performance at such high surface loads, 0.33 g NH_4^+–N/m^2/d, is similar to other nitrifying biofilm reactors (Tchobanoglous et al., 2003). At 5 °C, the highest measured removal rate was 0.54 g NH_4^+–N/m^2/d which is one third of the removal rate of 1.55 g NH_4^+–N/m^2/d in a nitrifying fixed-bed biofilm reactor at 8 °C (Zhu and Chen, 2002). The nitrifying bioreactor was maintained by internal recirculation of mine water II during phase IV. In phase V the feed was mine water II with an elevated sodium and chloride content while the temperature was kept at 5 °C. The elevated content of sodium and chloride in mine water II compared with mine water I is likely to have contributed to decrease in nitrification performance. The ammonium load was always kept less than 0.2 NH_4^+–N/m^3/d to reach nitrification up to 99%. Campos et al. (2002) showed that sodium ions rather than chloride or sulphate inhibits nitrification. Sodium ion concentrations were only up to 772 mg/l, thus well below the concentration of 5750 mg/l for total inhibition of nitrification as given by Campos et al. (2002). However, mine water II contained about 30 times more sodium ions than mine water I and about 21 mg/l of strontium (Table 1), of which the latter might have caused partial inhibition of the nitrifying biofilm.

The removal of nitrate after the nitrifying bioreactor was achieved in a denitrifying bioreactor. The nitrate concentrations in the influent and effluent of the denitrifying bioreactor were as shown in Fig. 4. Denitrification was minor during the first eleven weeks of operation and addition of phosphate after day 102 improved nitrate removal (Fig. 5). However, denitrification remained unstable during the first six months of the experiment, likely due to slow growth of methylotrophic denitrifying bacteria at low temperature. The shorthand of methanol compared to ethanol as an external carbon source is its lower support of growth of denitrifying bacteria (Rusten et al., 1996). Similar results were reported by Hiebert (1998) for a pilot-scale denitrifying fixed-bed bioreactor, feed 25 mg NO_3^-–N/l at 26 l/min, at Mineral Hill Mine in Montana. Even after one year of operation, the denitrification with methanol was unstable with 31–65% at 6 °C (Hiebert, 1998).

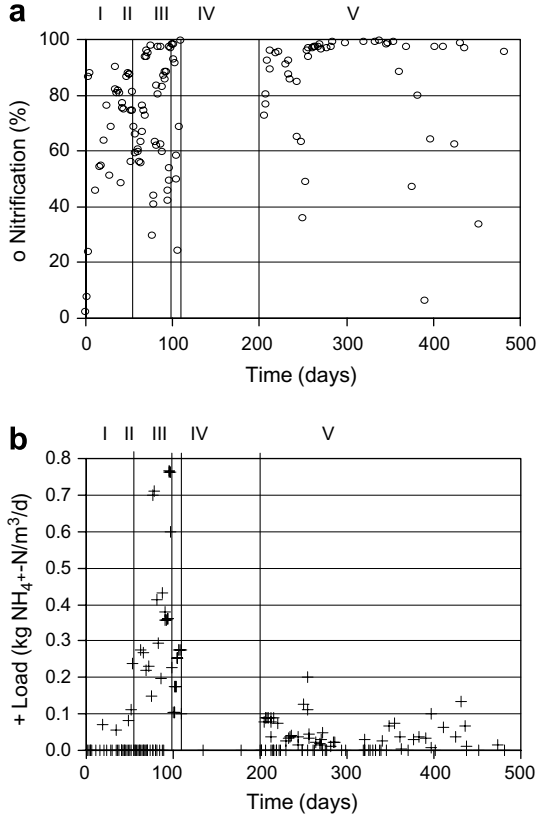

Fig. 2. (a, b) Nitrification performance at different load and temperature. The feed during the test works phase I to III was mine water I, phase IV to V mine water II. The temperature was as follows: phase I + 14 °C, II + 12 °C, III–VII + 5 °C.

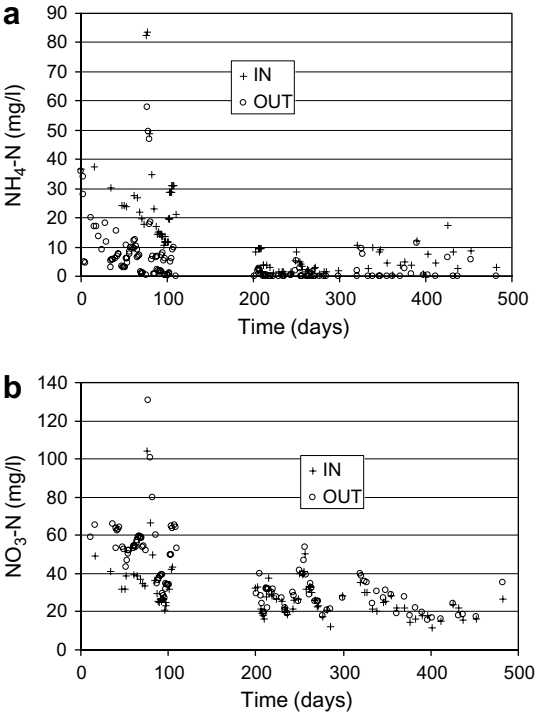

Fig. 3. (a, b) Ammonium and nitrate concentration in the influent and effluent of the nitrifying bioreactor.

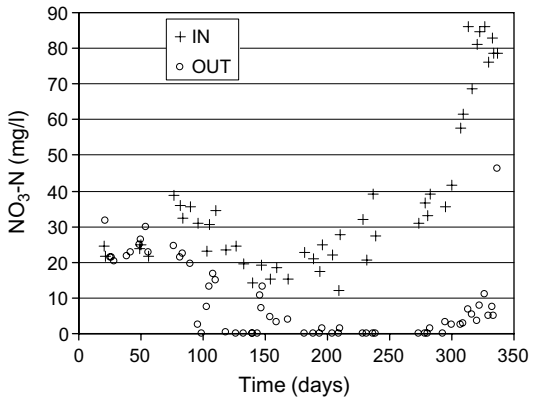

Fig. 4. Nitrate concentration in the influent (mine water II) and effluent of the denitrifying bioreactor.

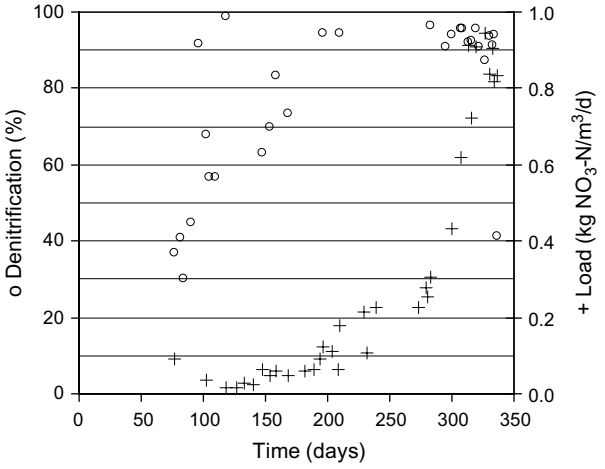

Fig. 5. Denitrification performance with increasing load at 5 °C.

Nitrate concentrations in the influent were increased after 300 days to study denitrification performance at higher load (Fig. 4). Denitrification with up to 95% was stable at loads of up to 0.91 kg NO_3^-–N/m^3/d or 4.26 g NO_3^-–N/m^2/d at 5 °C (Fig. 5). Higher loads could not be tested due to shortage of feed. Since the objective of the denitrification bioreactor was to reach good performance at high load and low temperature, minimization of COD content in the effluent was not attempted. The COD_{Cr}-value of the effluent was from 33 to 14500 mg O_2/l. The achieved denitrification rate of 4.1 g NO_3^-–N/m^2/d with methanol is significantly higher than reported denitrification rates of suspended biofilm reactors fed with methanol or ethanol of 1 or 2 g NO_3^-–N/m^2/d, respectively, at 5 °C (Rusten et al., 1996). Welander and Mattiasson (2003) reached denitrification rates of 1.6 g NO_3^-–N/m^2/d in bioreactor fed with a mixture of sodium acetate, peptone and yeast extract at 7 °C. Rusten et al. (1996) also reported denitrification rates of 1.5 g NO_3^-–N/m^2/d at 3 °C and 1.6 g NO_3^-–N/m^2/d which were much lower than the results presented here. At East Boulder Mine and Stillwater Mine in Montana, USA, removal of nitrate has been achieved by full-scale low maintenance denitrifying fixed-bed bioreactors (Reinsel and Plumb, 1999;

Reinsel, 2001). The removal of nitrate in slightly alkaline mine water proceeded at temperature as low as 2 °C (Reinsel and Plumb, 1999). However, these fixed-bed bioreactors operate at low nitrogen load. At the Stillwater Mine the denitrifying bioreactor has a treatment capacity of 4.2 m^3/min and operating costs have been stated to be in the range of 0.08–0.20 euro/m^3 (Reinsel, 2001).

In this study, nitrifying and methylotrophic denitrifying biofilms were stable against washout during periods of up to 482 and 337 days, respectively. This is the first report on high-rate removal of ammonium and nitrate from cold inorganic mine water by biofilm reactors.

4. Conclusions

Nitrifying biofilms can be established in reactors treating inorganic mine water at low temperature without external carbon or phosphorus sources. The activity of defined nitrifying and methylotrophic denitrifying biofilms can be maintained during long-term operation for more than one year. Denitrifying biofilms require long incubation times at low temperature to reach stable operation. However, excellent denitrification is achievable with methanol as an external carbon source and supplementation of phosphate. Nitrifying biofilms are strongly affected by temperature and feed salinity. Nitrification of $\leqslant$99% at load of $\leqslant$0.77 kg NH_4^+–N/m^3/d is possible at 12 °C. Nitrification >99% can be achieved at temperature as low as 5 °C, as can be denitrification of $\leqslant$95% at load of $\leqslant$0.91 kg NO_3^-–N/m^3/d and 5 °C. Thus, nitrification and denitrifiation rates are similar to operating biofilm process for organic wastewater treatment. Thus, fixed-bed biofilm reactors have potential to remove ammonium and nitrate also from mine water.

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