DENITRIFICATION OF SURFACE AND GROUND WATER APPLYING AUTOHYDROGENOTROPHIC BACTERIA IN A NOVEL UPFLOW BIO-ELECTROCHEMICAL REACTOR (UBER)

Shahin Ghafari*, Prof. Masitah Hasan, Prof. Mohamed Kheireddine Aroua

Department of Chemical Engineering, Faculty of Engineering, University of Malaya,
50603 Kuala Lumpur, Malaysia
*E-mail: shghafari@gmail.com

ABSTRACT

Nitrates in different surface and groundwater resources became a worldwide problem which raised concerns due to severe impacts on human and animal health. Although diverse abiotic methods were reported to remove nitrate from water, they fail to entirely treat nitrate. Biological denitrification is the only treatment method capable to reduce inorganic nitrate compounds to harmless nitrogen gas. Autohydrogenotrophic denitrifiers were applied in this study to prevent any problematic outcomes of heterotrophs' metabolisms raised from organic carbon source. A novel upflow bio–electrochemical reactor (UBER) was designed and employed to generate in-situ hydrogen and fulfill denitrifying process at cathode zone. Granular activated carbon (GAC) utilized to play a dual role as biocarrier and cathode material. Simple but effective design of this reactor helped to easily control the operating parameters such as electric current and flowrate. Furthermore, employing a circulating system, this reactor succeeded to cope with accumulation of nitrite in the cathode zone, which is known as a common and problematic phenomenon inhibiting the reaction.

Key words:
Autohydrogenotrophic denitrification; water; nitrate; Bio–electrochemical; UBER

INTRODUCTION

Nitrate (NO_3^-) and nitrite (NO_2^-) in various sources of surface and groundwater represent a significant risk to human health. Nitrate by itself is harmless, but it is a proven precursor of the toxic nitrite. Nitrite inside the digestive system of the consumer can cause methaemoglobinemia, (blue baby syndrome), offspring mortality, decreases weight gain as well as conversion to carcinogenic nitrosamine which can also cause hypertension. Accordingly, various agencies created standards for nitrate and nitrite in drinking water to minimize these health risks and nitrite had been more restrictedly limited: the United States Environmental Protection Agency (USEPA) has set maximum contaminant levels of 10 mg NO_3^-–N/L and 1.0 mg NO_2^-–N/L; The World Health Organization (WHO) and the European Economic Community (EEC) have set standards of 11.3 mg NO_3^-–N/L and 0.03 mg NO_2^-–N/L (Ghafari et al., 2008; 2009a).

Authors previously discussed about different abiotic and biotic techniques applied to remove nitrate from water streams, and also highlighted advantages associated with autohydrogenotrophic treatment using bio–electrochemical method (Ghafari et al., 2008). Autohydrogenotrophic denitrification takes place in two successive steps as shown below, if sufficient electron donor (H_2) and carbon source (bicarbonate/CO_2) is available for the microorganisms, and the favorable conditions (e.g. pH and HRT) had been prepared.

$$2NO_3^- + 2H_2 \longrightarrow 2NO_2^- + 2H_2O \qquad\qquad (1)$$

$$2NO_2^- + 3H_2 \longrightarrow N_2 + 2H_2O + 2OH^- \qquad\qquad (2)$$

In a former study, denitrifiers were successfully acclimatized, and effect of pH and bicarbonate concentration on their function was surveyed in batch experiments (Ghafari et al., 2009a, b). Prior to this study, the novel UBER showed a reasonably good removal on nitrate; however nitrite accumulation was encountered (Ghafari et al., 2009c). Rise of pH was diagnosed to be the main reason for such a failure. Therefore, the UBER was equipped with a circulating system which enabled the reactor to control pH. Superiorities of this novel UBER over the previous reactors are: simple design of this reactor, easy control of treatment, and its perfect treatment after being equipped with a pumparound system.

MATERIALS AND METHODS

Two sets of enclosed 2.1 L Plexiglas cylindrical UBER, as illustrated in Figure 1, were used in this study to provide a desirable anoxic environment by means of electric current at the cathode zone. Granular activated carbon (GAC) was used as cathode material to form the cathode zone with 17 cm height. The anode was a stainless steel mesh placed 1cm below the outlet level and was attached to the cap. A programmable DC power supply (IPS–3202, RS Components, England) provided the electric current. Four inlet ports were installed on the wall of the cylindrical column at 1cm above the bottom and one outlet port located at 25 cm from the bottom making a 5 cm head space. A sample ports (SP) was installed at height of 20 cm. Commercial palm shell GAC (supplied by Bravo Green Sdn. Bhd., Sarawak, Malaysia) with grain size range of 1–2 mm was used as the solid support and cathode materials in the UBER. Total volume of cathode zone was 1.2 L, and considering 66.5% GAC porosity, the void volume was 800mL. The acclimatized autohydrogenotrophic denitrifying bacteria obtained from the former study (Ghafari et al., 2009b) were used as the inoculums. Hydrogen gas and sodium bicarbonate were used as electron donor and inorganic carbon source, respectively, for growth and adaptation of bacteria enabling them of treating 20 mg NO_3^-–N/L/d. The acclimated mixed culture liquor was pumped into the UBER through 4 inlets of the reactor and was circulated until all the bacteria were trapped in the porous GAC cathode zone and a transparent supernatant was left. Biofilm formed gradually throughout the experiments and a dark grey color covered the activated carbon granules within one month. The synthetic contaminated water was prepared fresh by dissolving 0.122 g $NaNO_3$/L, 1.250 g $NaHCO_3$/L, 0.650 g K_2HPO_4/L, 0.170 g KH_2PO_4/L, 0.1 g

MgSO$_4$/L, 0.027 g CaCl$_2$/L in deionized water. The original pH of this solution was about 8 and it was adjusted to 7 using CO$_2$ (gas was bubbled through a commercial gas diffuser) before being injected to the UBER.
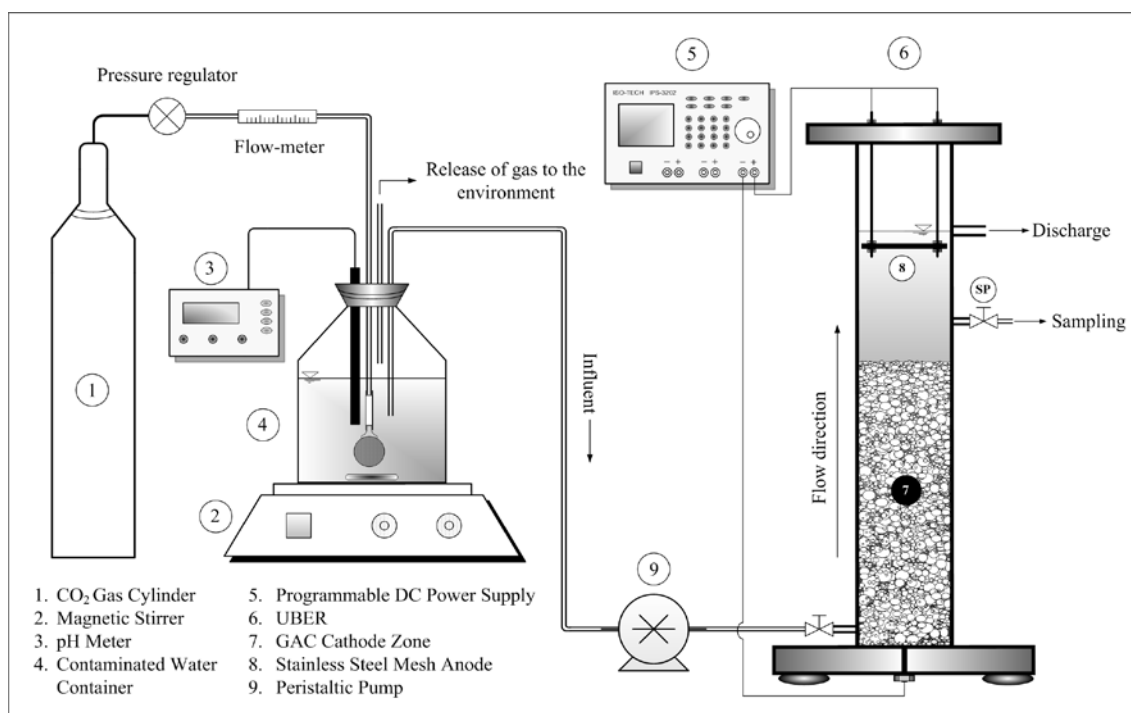

Fig. 1: Schematic representation of the experimental setup (Ghafari et al., 2009c)

RESULTS AND DISCUSSION

Obtained results from a former study (Ghafari et al., 2009c) revealed the developed UBER fails to properly treat the nitrate, and treatment process lead to nitrite accumulation in effluent. Figure 2 represents the pertinent results. This predicament was indicated to be due to either incompetent denitrifiers or pH rise in cathode zone due to three factors: pH–increasing nature of GAC, produced hydroxyl ion in the cathode zone through electrolysis of water, and production of hydroxyl ion through denitrification process. Therefore, to tackle the problem, two major courses of action were taken: (i) Employing stronger denitrifiers to achieve a proper population of autohydrogenotrophs capable of a faster degradation of nitrite than nitrate (Ghafari et al., 2009b), (ii) modification of the UBER in order to control pH in the favourable range (i.e. 7 to 8).
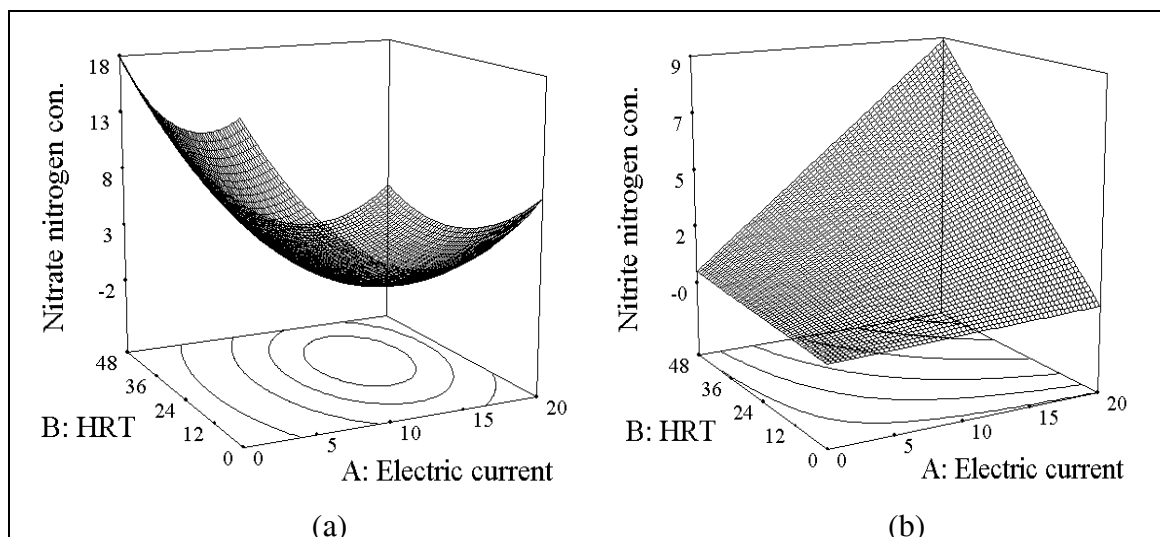
Fig. 2: Response surface plots for nitrate (a) and nitrite concentration in the treated water from UBER applying different electric currents and HRTs (Ghafari et al., 2009c)

Figure 3 shows that complete denitrification attained in the modified UBER as a result of altering two parameters at the same time: electric current (I) and circulation flow rate (F_c). Hydrogen was boosted by means of increase in electric current, and subsequent pH rise could be successfully controlled by manipulating the F_c. Continuous denitrification in UBERs commenced applying HRT=24h and I=15 mA, which are reasonably close to the previously achieved optimum conditions (HRT=28 h and I=14 mA) by Ghafari et al. (2009c). Experiments initiated applying electric current but pumparound system was off. Within eight days, nitrate concentration dropped to about 13 mg NO_3^-–N/L but pH rose up to around 9.3. Circulation was then commenced applying flowrate 0.5 mL/min with no CO_2 sparging. Results showed pH still increased but more gently. This was due to circulating the treated water with alleviated pH, because this water contained H^+ generated on the anode as it is taken from an outlet installed above the anode level. Nevertheless, H^+ was not enough to drop the pH to satisfactory level. Denitrification was also improved by keeping on the circulation and concentrations about 10.63 mg NO_3^-–N/L was detected in the treated water after 6 days on the 14th day.

With the purpose of putting an end on the ascending trend of pH, CO_2 was sparged into the circulated treated water (in the CO_2 sparging bottle) on the 14th day and a significant change was observed in both pH and nitrate concentration on the very next day. Afterwards, carbon dioxide was being bubbled incessantly all over the experiments to the end. After 4 days, pH laid on a constant value at 7.8 and nitrate concentration dropped to 7.15 mg NO_3^-–N/L. At this time point, pH was in the desirable range and denitrification process succeeded to meet the guideline value with no nitrite in the treated water. Thus far, no nitrite was detected, although pH values higher than the safe range were experienced.
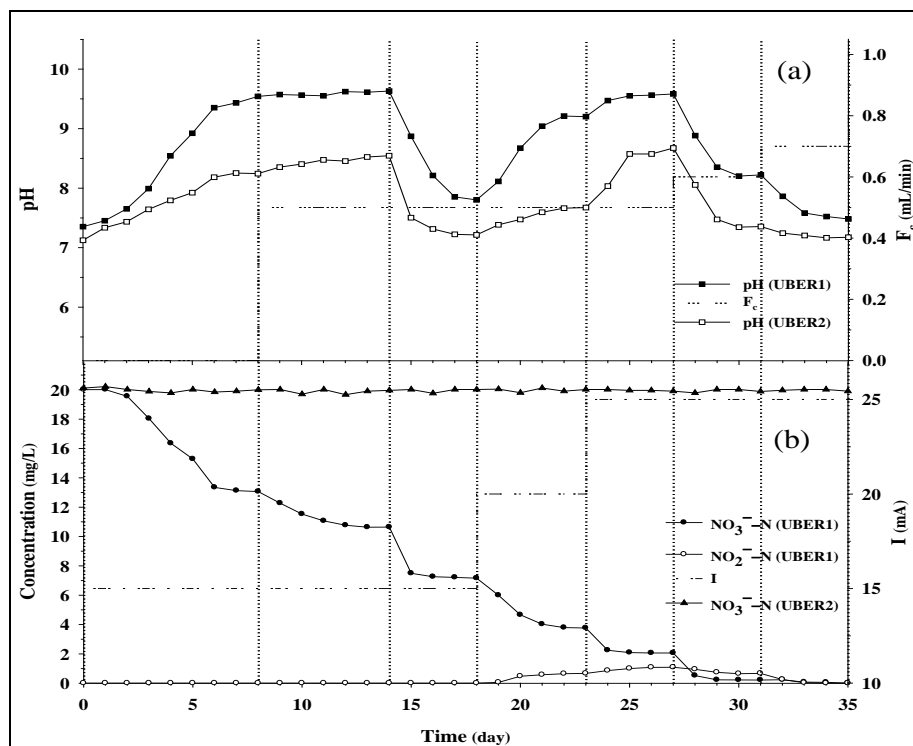
Fig. 3: Simultaneous variation of circulating flowrate (a) and electric current (b), and their effects on pH values (a) and nitrate/nitrite concentrations (b) of the treated water in the modified UBERs when HRT was 24 h

Experiments were conducted at higher electric currents aiming to reach a treated water entirely free from nitrogen oxide, and also further investigation on the ability of pumparound system for control of pH. Accordingly electric current increased to 25 mA in two steps. Firstly, UBER was operated for five days with I=20 mA and F_c=0.5 mL/min. As a result, pH rose to 9.2 and nitrite was observed at concentrations about 0.66 mg NO_2^-–N/L, but further depletion in nitrate concentration up to 3.75 mg NO_3^-–N/L was obtained. Electric current was then increased to 25 mA, which could supply more hydrogen and also could extremely shift the pH to more alkaline region. As a result, pH reached 9.6 and concentration of nitrate and nitrite were respectively, 2.05 mg NO_3^-–N/L and 1.08 mg NO_2^-–N/L on the 27th day. To alleviate the pH at the same electric current, circulation flowrate as the pH regulator was increased to 0.6 mL/min, and it could make a pH drop to 8.2 after 4 days when nitrate and nitrite in the treated water were 0.21 mg NO_3^-–N/L and 0.66 mg NO_2^-–N/L, respectively. With the same electric current, F_c was increased again to 0.7 mL/min. Consequently, pH fell to about 7.5, and complete treatment of nitrate with no trace of nitrite was achieved.

CONCLUSION

This study represented a research for the continuous denitrification in a novel UBER operated to generate in–situ hydrogen for the autohydrogenotrophic denitrifying bacteria immobilized on the GACs which also played the role of solid support for

microorganisms. Results from former investigation showed that nitrite was accumulated in concentrations above the MCL (0.1 mg NO_2^-–N/L) because of pH rise in the cathode zone. Hence, the UBER was improved with a pumparound system aiming to control pH in the cathode zone, and also more competent microbes were employed. Experiments carried out at higher electric currents which could exacerbate pH rise in the GAC bed; however, results showed that pumparound system succeeded to control the pH in the safe range by means of manipulating the circulation flowrate, and consequently, complete denitrification was attained at F_c 0.7 mL/min when HRT and I were respectively, 24 h and 25 mA.

ACKNOWLEDGEMENTS

REFERENCES

Ghafari, S, Hasan, M & Aroua, MK 2008, 'Bio–electrochemical removal of nitrate from water and wastewater–A review', *Bioresource Technology,* vol. 99, no.10, pp. 3965–3974.

Ghafari, S, Hasan, M & Aroua, MK 2009a, 'Effect of carbon dioxide and bicarbonate as inorganic carbon sources on growth and adaptation of autohydrogenotrophic denitrifying bacteria', *Journal of Hazardous Materials*, vol. 162, no.2–3, pp. 1507–1513.

Ghafari, S, Hasan, M & Aroua, MK 2009b, 'Improvement of autohydrogenotrophic nitrite reduction rate through optimization of pH and bicarbonate dose in a batch experiment', *Journal of Bioscience and Bioengineering*, vol. 107, no. 3, pp. 275–280.

Ghafari, S, Hasan, M & Aroua, MK 2009c, 'Nitrate remediation in an upflow bi–electrochemical reactor (UBER) using palm shell activated carbon as the cathode materials', *Electrochimica Acta*, vol. 54, no. 17, pp. 4164–4171.