

# Isotopic Fractionation Associated to Nitrate Reduction by Zero Valent Iron

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## INTRODUCTION

One of the methodologies developed in recent years to induce nitrate attenuation is the use of permeable reactive barriers (PRB). This in situ remediation technique involves the interception of groundwater flow to remove contaminants by physical, chemical or biological processes. In the case of nitrate pollution, heterotrophic or autotrophic denitrification can be induced by using organic or inorganic substrates, respectively. On the other hand, several PRB filled with Zero-Valent Iron (ZVI) have been installed to remediate groundwater polluted with chlorinated solvents (Wilkin et al., 2014) or nitrate (Gu et al., 2002).

The following reaction pathway has been proposed as the dominant one during abiotic nitrate reduction by ZVI (Yang and Lee, 2005):



The use of Zero-Valent Iron to reduce nitrate can lead to extremely reducing conditions that can induce ammonium production (Liou et al., 2005), which is more toxic than nitrate itself. The application of ZVI in reducing nitrate is therefore limited by ammonium production and the requirement for adequate pH control. However, no clear reaction pathway has been determined yet and other studies suggest that nitrite and nitrogen gas could be other possible products of nitrate reduction by ZVI (Chen, et al., 2005).

As the abiotic reduction of nitrate by ZVI occurs, residual nitrate might be enriched in heavy N and O isotopes following the Rayleigh equation. Therefore, isotopic analyses of dissolved nitrate might permit evaluating the extent of nitrate attenuation by ZVI and distinguishing degradation from other processes at contaminated sites. The oxygen and nitrogen isotope fractionations associated to nitrate

reduction by ZVI have not been reported in the literature. The potential of the in situ measurement of the isotopic composition of nitrate for assessing the effectiveness of ZVI-PRBs for removing nitrate has yet to be evaluated.

## OBJECTIVES

The main aim of this study is to carry out lab experiments to determine nitrogen and oxygen isotope fractionations ( $\epsilon\text{N}$  and  $\epsilon\text{O}$ ) associated with the ZVI-driven nitrate reduction reaction in order to investigate the potential of isotope analyses to assess and quantify the performance of a ZVI-PRB at contaminated sites.

## MATERIAL AND METHODS

### Experimental set-up

Batch experiments were carried out in 40mL bottles. Each bottle contained 6.75 mg of cast iron and 35 mL of a nitrate-containing aqueous solution. Three different solutions were used: MilliQ water (pH=5.5.), a pH 4 HCl 0.1M solution and groundwater sampled upflow of a ZVI-PRB installed in a DNAPLs polluted site (Audi-Miró et al., 2013). In this site, high nitrate concentrations (up to 215 mg/L) were found upstream of the barrier, whereas nitrate content downflow of the barrier ranged between 1 and 150 mg/L, suggesting the occurrence of nitrate reduction processes. All the synthetic solutions contained 120 mg/L of nitrate, whereas the groundwater nitrate content was 180 mg/L. All the experiments were performed in duplicate under anaerobic conditions in a sterilized and anaerobic glove box with  $\text{N}_2$  at  $28 \pm 2^\circ\text{C}$ .

The specific surface area of the cast iron determined by  $\text{N}_2$  gas adsorption (BET method) was  $0.704 \pm 0.005 \text{ m}^2/\text{g}$ . Prior to the experiment the Fe was acid-cleaned with 1N degassed HCl and dried

inside the glove box. After preparation inside the glove box, bottles were immediately covered with aluminium foil to avoid oxidation due to light and were rotated on a horizontal roller table at 60 rpm. Experimental runs lasted 8 days. Concentration of N species, pH and,  $\delta^{15}\text{N}$  and  $\delta^{18}\text{O}$  of dissolved nitrate were measured.

## RESULTS AND DISCUSSION

Consumption of nitrate over time was observed in all the batch experiments (Fig. 1). Almost complete consumption of nitrate occurred in 8 days.

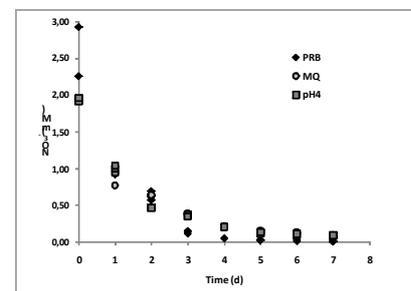
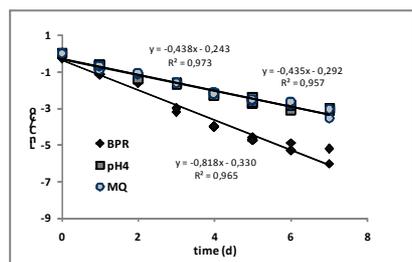


Fig 1. Consumption of nitrate concentration over time in representative batch experiments. The error bars show the standard deviation for duplicate experiments.

The reaction was assumed to be of pseudo first order and the pseudo-first-order rate constants,  $k'$ , were calculated from the slope of the regression lines of the  $\ln C/C_0$  vs. time graph (Fig. 2). Nitrate reduction rate in the experiments with groundwater (PRB experiments) was faster ( $k'=0.9 \text{ d}^{-1}$ ) than in the experiments with synthetic solutions (MQ and pH4 experiments,  $k'=0.5 \text{ d}^{-1}$ ), suggesting additional  $\text{NO}_3^-$  degradation by autochthonous bacteria. Denitrifying bacteria may greatly increase the rate and extent of nitrate reduction in the reducing zone of a ZVI-PRB. The combination of ZVI-driven nitrate reduction with denitrification by heterotrophic denitrifying bacteria has been proved at lab scale, resulting in an increase in the nitrate reduction rate and in a decrease in the ammonium

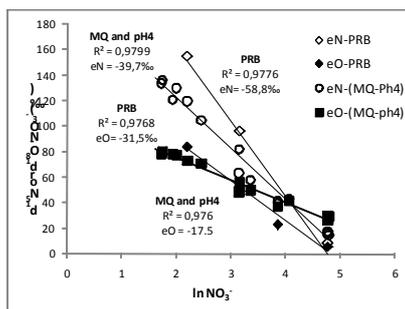
release (An et al., 2009). In the present experiments, additional autotrophic denitrification by oxidation of cathodic hydrogen from anaerobic ZVI corrosion (Till et al., 1998) could be proposed, although further research is required. At both pH values tested in the experiments with synthetic solutions (4 and 5.5) similar nitrate reduction rates were obtained, suggesting that at the tested conditions nitrate reduction was independent of pH.



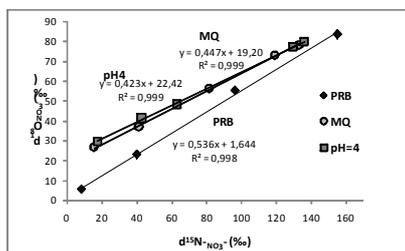
**Fig 2.** Evolution of  $\ln(C/C_0)$  over time. The error bars show the standard deviation for duplicate experiments. Linear regression lines and high correlation values ( $R^2$ ) evidenced pseudo-first order kinetics.

All experiments show a considerable enrichment in both  $^{15}\text{N}$  and  $^{18}\text{O}$  in the remaining nitrate over the course of the experiments. In the PRB experiment  $\delta^{15}\text{N}$  increased from +8.2‰ to +154.9‰, whereas  $\delta^{18}\text{O}$  increased from +5.4‰ to +83.5‰ (Fig. 3). Similarly, the  $\delta^{15}\text{N}\text{-NO}_3^-$  in the MQ and pH4 experiments increased from +15.6 and +17.5‰ to +133.9 and +135.9‰, respectively.  $\delta^{18}\text{O}$  values increased from +26.5 and +29.6 ‰ to +78.2 and 79.9‰, respectively. The N and O isotopic enrichment factors in the experiments were calculated from the slope of the regression lines that fit the data of the natural logarithm of nitrate concentration vs.  $\delta^{15}\text{N}$  or  $\delta^{18}\text{O}\text{NO}_3^-$ , respectively (Fig.3). The nitrogen isotopic enrichment factor ( $\epsilon\text{N}$ ) was -58.8‰ for the PRB experiments and -39.7‰ for the MQ and pH4 experiments. The oxygen isotopic enrichment factor ( $\epsilon\text{O}$ ) was -31.5‰ and -17.5‰, respectively.

In all the experiments  $\delta^{15}\text{N}$  showed a linear relationship with  $\delta^{18}\text{O}$ , with slopes between 0.42 and 0.54, yielding  $\epsilon\text{N}/\epsilon\text{O}$  ratios of 1.9 for the PRB experiment and 2.3 for the MQ and pH4 experiments (Fig. 4). This slight difference between  $\epsilon\text{N}/\epsilon\text{O}$  values in the PRB experiments and in the experiments with purely abiotic solutions suggests that the use of this relationship could allow to distinguish additional degradation of nitrate by the action of bacteria autochthonous to the aquifer. Further research is required.

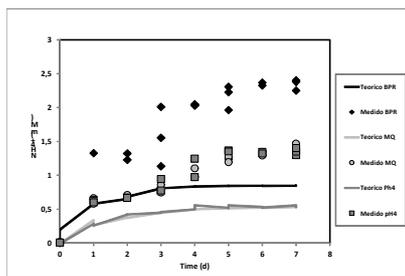


**Fig 3.**  $\delta^{15}\text{N}$  vs.  $\delta^{18}\text{O}\text{NO}_3^-$  for batch experiments. The error bars show the standard deviation for replicate measurements.



**Fig 4.**  $\delta^{15}\text{N}$  vs.  $\delta^{18}\text{O}\text{NO}_3^-$  for batch experiments. The error bars show the standard deviation for replicate measurements.

In all the experiments ammonium was produced whereas insignificant nitrite contents were detected. Nitrogen mass balance in the current experiments shows that ammonium accounts for about 85-95% and 60-80% of the reaction products in the PRB and the MQ and pH4 experiments, respectively (Fig. 5). This fact suggests that gaseous N species should account for the balance.



**Fig 5.**  $\text{NH}_4^+$  concentration vs. time. The evolution of theoretical (assuming complete conversion of nitrate to ammonium following Eq. 1) and measured ammonium values is shown.

### CONCLUSIONS

Zero-Valent Iron induced nitrate reduction in laboratory batch experiments. Nitrate was consumed in less than 8 days, releasing ammonium to the solution. The nitrate reduction and ammonium production rates were independent of pH at the tested pH range. In all the experiments, the mass balance of nitrogen aqueous species suggested production of  $\text{N}_2$  (g) as proposed by other authors (Yang and Lee, 2005). Further research is needed

to confirm this hypothesis.

Nitrogen and oxygen isotope fractionations ( $\epsilon\text{N}$  and  $\epsilon\text{O}$ ) associated with the ZVI-driven nitrate reduction reaction were calculated.

### ACKNOWLEDGMENTS

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### REFERENCES

An, Y., Li, T. L., Jin, Z. H., Dong, M. Y., Li, Q. Q. (2009): Decreasing ammonium generation using hydrogenotrophic bacteria in the process of nitrate reduction by nanoscale zero-valent iron. *Sci. Total Environ.*, **407**, 5465-5470.

Audi-Miró, C., Cretnik, S., Torrentó, C., Rosell, M., Palau, J., Shouakar-Stash, O., Elsner, M., Otero, N., Soler, A. (2013): Chlorine, hydrogen and carbon multi isotopic approach to assess natural attenuation and ZVI-PRB induced degradation in a DNAPLs contaminated site. *Central European Geology*, **56**, 202-204.

Chen, Y. -M., Li, C. -W., Chen, S. -S. (2005): Fluidized zero valent iron bed reactor for nitrate removal. *Chemosphere*, **59**, 753-759.

Gu, B., Watson, D. B., Wu, L., Phillips, D. H., White, D. C., Zhou, J. (2002): Microbiological characteristics in a zero-valent iron reactive barrier. *Environ. Monit. Assess.*, **77**, 293-309.

Liou, Y. H., Lo, S. L., Lin, C. J., Hu, C. Y., Kuan, W. H., Weng, S. C. (2005): Methods for accelerating nitrate reduction using zerovalent iron at near-neutral pH: Effects of  $\text{H}_2$ -reducing pretreatment and copper deposition. *Environ. Sci. Technol.*, **39** (24), 9643-9648.

Till, B. A., Weathers, L. J., Alvarez, P. J. J. (1998): Fe(0)-supported autotrophic denitrification. *Environ. Sci. Technol.*, **32**, 634-639.

Wilkin, R. T., Acree, S. D., Ross, R. R., Puls, R. W., Lee, T. R., Woods, L. L. (2014): Fifteen-year assessment of a permeable reactive barrier for treatment of chromate and trichloroethylene in groundwater. *Sci. Total Environ.*, **468-469**, 186-194.

Yang, G. C. C., Lee, H.-L. (2005): Chemical reduction of nitrate by nanosized iron: kinetics and pathways. *Water Research*, **39** (5), 884-894.