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Can Membrane Inlet Mass Spectrometer Measure Short-term Denitrification Enzyme Activity and Denitrification Potentials of Soils?

M.I. Khalil and K.G. Richards

Teagasc Environment Research Centre, Johnstown Castle, Wexford, Ireland (ibrahim.khalil@teagasc.ie)

Denitrifier population size and potential activity combined with the relevant environmental factors regulate the rates of denitrification in terrestrial and aquatic ecosystems. Due to the high atmospheric background of di-nitrogen (N_2), denitrification enzyme activity (DEA) in soils is traditionally measured using the acetylene block or stable isotope techniques under non-limiting substrates and anaerobic/saturated conditions for periods from a few hours to several days so as to estimate denitrification potential (DP). This research investigated the estimation of DEA and DP by quantifying the N_2/Ar ratio changes in waters/sediments using membrane inlet mass spectrometry (MIMS). Two experiments were conducted with soils of A, B and C horizons collected from grazed grassland to obtain optimal NO_3^- and available carbon (C) rates. In experiment 1, 30 g soil (oven dry basis) followed by helium-flushed deionized water was taken in triplicate 160 mL glass bottles and sealed with rubber stoppers without any air entrapments. Then N as potassium nitrate (0 to 120 mg $NO_3 - N$ kg^{-1} soil) and readily available C as glucose (0 to 240 mg glucose-C) plus 30 mg $NO_3 - N$, kg^{-1} soil were amended. Laboratory incubation was performed in the dark at 21^{o}C under water to reduce the risk of N_2 contamination. After six hours, the treated water samples were transferred into 12 mL exetainers and kept under water at 4^{o}C before analysis using MIMS. The N_2/Ar ratios, representing DEA, varied between soil horizons and declined with decreasing soil depths. The maximum peak for N_2/Ar ratios were observed with the 30 mg $NO_3 - N$ kg^{-1} soil in all soil horizons and coupled with the 60 mg glucose-C kg^{-1} soil for C horizon, and 120 mg glucose-C kg^{-1} for A and B horizons. Experiment 2 was conducted to assess simulated unsaturated and saturated subsoil (C horizon) denitrification capacity ($NO_3 - N$only amendment), and DP (both C and N amendment) using the same methodology as experiment 1 and incubated for 3 days using groundwater. The optimal substrate rates (30 mg $NO_3 - N \pm$ 60 mg glucose-C, kg^{-1} dry soil) were used for this experiment. All treatments were in the dark at 12^{o}C under water to prevent N_2 contamination. The response of the unsaturated and saturated conditions to denitrification capacity (N only substrate) was identical. However, the denitrification capacity was significantly lower (0.54 mg N kg^{-1} soil d^{-1}), relating to the higher oxidative state, than the control (0.81 mg N kg^{-1} soil d^{-1}). In contrast, DP in the saturated subsoil was noticeably greater (2.19 mg N kg^{-1} soil d^{-1}) than in the unsaturated subsoil (0.92 mg N kg^{-1} soil d^{-1}) conditions. Results suggest that the DEA and DP of soils were mainly limited by the available C as an energy source for denitrifiers, and the amendment of glucose-C superseded the temporary increased oxidative state that occurred due to $NO_3 - N$ addition. N_2/Ar ratios measured using MIMS could be used as an alternate method to assay denitrification in soils but it requires further validation against other existing standard methods.