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Denitrification as the dominant N-elimination process in hydrothermal vents of the Juan de Fuca Ridge, North-East Pacific Ocean

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Nitrogen (N) is an essential macronutrient for all organisms. Therefore, oceanic N sinks, removing bio-available (or fixed) N, ultimately affect primary productivity. The relative importance of the two main N-elimination pathways, i.e. denitrification and anaerobic ammonium oxidation (anammox) in different oceanic environments and in the global ocean is still a matter of debate. Little is known about metabolic processes and bacterially-mediated N-cycle dynamics occurring in the subsurface biosphere of hydrothermal vent systems. Rates of major N-elimination processes have never directly been quantified in diffuse vent fluids. In this study, we measured rates of major fixed N-elimination pathways (denitrification, anammox) and dissimilative nitrate reduction to ammonium (DNRA) in hydrothermal vent fluids at 12 different sites on the Juan de Fuca Ridge using ^{15}N-label incubations. We also measured the isotopic composition of dissolved inorganic nitrogen (DIN) (nitrate and ammonium) and N_2O concentrations, an intermediate product of denitrification. Bacteria, Archaea and SUP05 (a group belonging to the gamma subclass of Proteobacteria that mediates chemolithotrophic denitrification) SSU rRNA gene copy numbers were determined from DNA extracted from Sterivex-filtered HV fluids using q-PCR (quantitative polymerase chain reaction) assays. All samples were collected during cruises in the Northeast Pacific Ocean onboard the R/V Atlantis in August 2008 and June 2009. Elevated nitrate δ^{15}N and δ^{18}O in the high-Mg^{2+}, low-T vent fluids associated with a $[NO_3^-]$ decrease is indicative of dissimilatory (denitrification) or assimilatory (nitrate uptake) nitrate consumption. Denitrification rates in hydrothermal vent fluids at Axial Volcano and Endeavour Segment were high and variable between sites, ranging from $\sim$1 to 1040 nM/day. $[N_2O]$ ranged between $\sim$0-430 nM, with no observable correlation with denitrification rates. Anammox rates were below $\sim$5 nM/day at 3 sites and not detectable at all other sites. DNRA rates ranged from $\sim$0 to 200 nM/day. These results suggest that denitrification is by far the dominant N sink in hydrothermal vent fluids of the Juan de Fuca Ridge. Furthermore, q-PCR results showed that nitrate reduction associated with sulfide oxidation is likely an important N-process in hydrothermal vent systems, with bacteria of the SUP05 cluster representing up to 38% of the total bacterial population. On-going phylogenetic analysis of the denitrifier functional genes *nirS and nirK*, encoding for the nitrite reductase enzyme, together with q-PCR, will allow us to better characterize the diversity and abundance of sub-seafloor denitrifier populations.