

## Cometabolic Degradation of Insensitive Munitions Constituents during Nitrification

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**Background/Objectives.** As insensitive munitions (IM) enter the military supply chain the interactions of IM with environmentally relevant metabolic pathways will determine the natural attenuation capacity of explosives impacted areas on military training ranges. The most commonly investigated cases of biological transformation for munitions constituents are determined in terms of the prevailing electron accepting conditions (Hoferkamp and Weber, 2006) or the most abundant electron donor (Fahrenfeld, Pruden, and Widdowson, 2015). Nitrification is an important environmental process for the stepwise oxidation of ammonium to nitrite and nitrate. The first step in this process is the production of nitrite by ammonia oxidizing bacteria (AOB). The intermediate step in ammonia oxidation is the formation of hydroxylamine by an ammonium monooxygenase enzyme (AMO). The AMO enzyme is known to oxidize compounds other than its target leading to a possible cometabolic transformation pathway for nitrogenous compounds including MCs. This project seeks to establish the possible extent of cometabolic transformation of IM during nitrification. Of special interest were two IM constituents with high water solubilities, nitrotriazolone (NTO) and nitroguanidine (NQ).

**Approach/Activities.** A continuous nitrifying culture was established in a 5-L suspended growth reactor with an integrated settling chamber. The system was maintained at a steady state nitrification rate of 40 gN/m<sup>3</sup>/d with an activity level of 8.08 gN/gVSS/d. The system was challenged with sequential levels of NTO or NQ, and the mixed liquor and effluent concentrations were monitored. Total system mass balances were determined by liquid chromatography (HPLC) for IM constituents and ion chromatography (IC) for ammonium, nitrite, and nitrate. Effluent data were plotted against a mixed reactor model with a reaction term to determine the mass balance across the reactor, and steady state transformation was calculated by minimizing the residuals squared. The mass loading rate was varied to observe the kinetic rate response of nitrification associated transformation.

**Results/Lessons Learned.** The removal of NQ by the suspended nitrifying culture increased along with the mixed liquor concentration. Total NQ transformation rates between 13 and 143 mg NQ/L/d were observed. NTO exhibited a removal rate of 42 mg NTO/L/d at a mixed liquor concentration of 1,456 mg/L. Additional studies are being undertaken to identify the possible transformation products of nitrification dependent cometabolism. Depending on overall mass loading rates, nitrification may provide an important transformation pathway for IM constituents in the near surface soil environment.