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Biotic Fe(II) oxidation and nitrite reduction in autotrophic nitrate-reducing iron(II)-oxidizing cultures

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Nitrate reduction coupled to Fe(II) oxidation (NRFeOx) has been recognized as an environmentally important microbial process in natural ecosystems. During nitrate reduction, nitrite often accumulates resulting in abiotic Fe(II) oxidation (chemodenitrification), which is an energetically favorable reaction and contributes to N_2O formation and emission. Since Fe(II) is used as both energy and electron source for iron(II)-oxidizing bacteria that reduce nitrate and the following denitrification intermediates (NO_2^-, NO, N_2O), the competition between abiotic and biotic Fe(II) oxidation in the presence of nitrite becomes a critical factor. However, the kinetics of biotic Fe(II) oxidation and nitrite reduction in these cultures remain unclear due to the simultaneous occurrence of biotic and abiotic reactions.

In this study, we quantified nitrite reduction and Fe(II) oxidation in two autotrophic NRFeOx enrichment cultures KS and BP. First, we tested NO_2^- as the sole electron acceptor for biotic Fe(II) oxidation by incubating cultures KS and BP with nitrite and Fe(II). At a high NO_2^- concentration (3.3 mM), there was no significant difference in the rate and extent of Fe(II) oxidation and NO_2^- reduction between abiotic and biotic setups, suggesting that high NO_2^- concentrations may inhibit biotic reactions because of nitrite's toxic effect. In fact, when 1.7 mM NO_2^- was added to cultures that had been growing with 1 mM NO_3^- for 2 days, biotic Fe(II) oxidation and nitrate reduction almost stopped. In contrast, NRFeOx microorganisms grew at a lower NO_2^- concentration (0.68 mM). Under low-nitrite conditions, over 80% of 2 mM Fe(II) was oxidized in biotic setups, whereas only 23.3% of Fe(II) was oxidized in the abiotic setup. Furthermore, 57.6% and 95.5% nitrite were reduced in cultures KS and BP, respectively, while only 16.0% nitrite was reduced in the abiotic setup. Interestingly, although there was more biotic nitrite reduction than abiotic reduction, the N_2O production was similar. Overall, our data suggests that enzymes catalyze most Fe(II) oxidation and nitrite reduction at low nitrite concentrations and biotic processes do not significantly contribute to N_2O production. However, the biotic processes are inhibited at high nitrite concentrations, and as a consequence, abiotic Fe(II) oxidation dominates.