

ABSTRACT

Nitrous oxide (N₂O) is an unwanted byproduct from biological nitrogen removal in wastewater treatment, because it is a strong green house gas and major stratospheric ozone depletion substance. N₂O emissions from biological nitrogen removal can contribute substantially to the carbon dioxide (CO₂)-equivalent footprint of wastewater treatment. The majority of these N₂O are produced by ammonia oxidizing bacteria (AOB) and denitrifiers via nitrifier nitrification (NN), nitrifier denitrification (ND), and heterotrophic denitrification (HD) pathways. Many efforts have been done to characterize the production of N₂O in wastewater treatment system. However, the fundamental knowledge about pathways and controls of N₂O production is limited, and mainly challenged by difficulties in distinguishing these different pathways in a complex microbial community. In this thesis, batch incubation studies, lab-scale investigations and full-scale monitoring of the dynamics of N₂O production are combined with the ¹⁵N/¹⁸O stable isotope labeling approach to provide a better understanding of N₂O production mechanisms.

In **Chapter 2** pathways of N₂O production and their dependence on environmental parameters (DO, NO₂⁻ and NH₄⁺) in nitrification-anammox biomass were investigated in short-term incubations. Under low NO₂⁻ condition, NN was the dominant production pathway of N₂O at 0.2 mg L⁻¹ DO. However, ND was induced by external NO₂⁻ addition and became as major N₂O production pathway with 0.7 mM NO₂⁻ addition.

Dynamics of N₂O in a lab-scale partial nitrification reactor was studied by online dissolved N₂O monitoring combined with ¹⁵N stable isotope labeling (apply ¹⁵NH₄⁺/¹⁵NO₂⁻ to the reactor) to reveal the mechanisms of N₂O production (described in **Chapter 3**). N₂O production was completely dominated by ND.

Chapter 4 focuses on full-scale plant. A novel ¹⁵N/¹⁸O dual labeling method was established to study the dynamics of N₂O production pathways in a full-scale activated sludge plant. The results showed that the oxic phase was the major source of N₂O production with more than 5 times higher accumulation rates than in the anoxic phase. NN and ND contributed roughly equal at 3 mg L⁻¹ DO, while ND was the dominante source at 1mg L⁻¹ DO.

The studies demonstrated that ¹⁵N/¹⁸O stable isotope labelling is a robust approach to distinguish different N₂O production pathways in biological nitrogen removal plants, and it can contribute to the development of operational strategies to minimize N₂O emissions. All three pathways (NN, ND, and HD) were active in nitrogen removal processes, each responding differently to changes in the operational conditions. NN can be a significant N₂O source in nitrification-anammox and activated sludge system. It can contribute with up to ~100% of the total N₂O production. However, ND is the predominate source of N₂O in the partial nitrification process, due to high NO₂⁻ concentrations together with low concentrations of DO. HD was a minor source of N₂O under oxic conditions in all tested nitrogen removal processes, while promoted when DO was absent or organic carbon was present. The relative importance of NN and ND to total N₂O production was mainly determined by DO level and NO₂⁻ concentration.