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**De novo biofilm community assembly from tap water source communities favors *Nitrotoga* over *Nitrospira* under elevated nitrite surface loading**

Marta Kinnunen, Arnaud Dechesne, Hans-Jørgen Albrechtsen, Barth F. Smets

Four main processes are considered to drive microbial community assembly: selection, drift, dispersal and speciation. These processes occur simultaneously, but the extent to which each process contributes to community assembly is unclear in natural communities.

We exposed a high-throughput flow-through biofilm system to continuous immigration from a tap water metacommunity while applying different nitrite surface loading rates. After 63 days of operation, we extracted biofilms and analyzed the community composition via Illumina MiSeq targeting the 16S rRNA gene. Previous studies have shown that *Nitrospira* is the dominant nitrite oxidizing genus in low nitrite environments. Hence, we postulated that by elevating the nitrite surface loading we would select for NOB with lower nitrite affinity than *Nitrospira*.

We observed different dominant NOB species under different loading rates. While in the metacommunity, *Nitrotoga* and *Nitrospira* were found at near equal abundances, in the biofilm community, elevated nitrite loading strongly selected for *Nitrotoga* over *Nitrospira*. The biofilms were also significantly different in their alpha-diversity ( $p < 0.001$ ) and beta-diversity, and the evenness and richness of the biofilm community decreased significantly ( $p = 0.004$ ) compared to the metacommunity. These observations indicate that the selection towards *Nitrotoga* and *Nitrospira* dominated community assembly under different nitrite loadings. Lastly, we compared our observations of community composition with that predicted by neutral community assembly model. The predictions did not match the community structure observed in the biofilms ( $p = 0.31$ ), providing further evidence of the importance of selection during community assembly.