



Characterization and potential impacts of active nitrifier host–virus interactions in soil

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While the complexity of prokaryotic communities in soil is relatively well understood, we are currently ignorant of the role of viruses in influencing their ecology. Viruses infect every living organism and affect rates of biogeochemical processes by killing active cells via cell lysis, or augmenting function through the transfer of auxiliary metabolic genes. The microbially-mediated process of nitrification is central to nitrogen losses and emissions of the greenhouse gas nitrous oxide, contributing to global warming and stratospheric ozone depletion, and viruses may impact this central step of the global N cycle. However, challenges remain in identifying active interactions with specific host or functional groups and virus populations within structurally complex and diverse soil environments. To address this, discrete active interactions between individual hosts and viruses in soil microcosms were examined using the transfer of assimilated carbon from autotrophic prokaryotes to viruses. Microcosms were established with amendments of urea to fuel nitrification and ^{13}C carbon dioxide, followed by DNA stable-isotope probing (SIP) combined with metagenomic analyses. Hybrid analysis of GC mol% fractionation and ^{13}C -DNA-SIP resulted in the identification of active ammonia oxidizing archaea (AOA) hosts and viruses only. To enable characterization of viruses actively infecting all active ammonia- and nitrite oxidizing bacteria (AOB, NOB), a second approach was used where incubations were performed using filtration of virus-like particles to increase the recovery of virus metagenomes. This included differential inhibition of specific nitrifier groups to alleviate competition and potentially increase the abundance of viruses infecting non-inhibited groups. Applications of DMPP or octyne were added to preferentially inhibit AOB and encourage growth of AOA, or acetylene to inhibit all ammonia oxidizing activity as a negative control. This approach dramatically increased the recovery of high-quality virus contigs, with 225 contigs associated with those infecting AOA, AOB or NOB including 69 complete or near-complete genomes. Viruses of ammonia-oxidizers contained auxiliary metabolic genes involved in central metabolic pathways, and analysis of viral hallmark genes revealed that they were distinct from previously cultivated viruses. Results demonstrate that virus infection of nitrifiers and propagation of viruses is a dynamic process during soil nitrification, providing insights into potential impacts of viruses on a specific functional process in soil.