

## Phage diversity and activity associated with seasonal changes in a model montane soil ecosystem

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**Project Goals:** This project aims at characterizing the diversity and dynamics of phage communities in a montane meadow soil across a full year, from beneath the snowpack during winter, through snowmelt and plant growth in spring, and to plant senescence in the fall. Leveraging paired metagenomes and metatranscriptomes, a holistic view of the activity and turnover of the local viral community will be assembled to explore the potential roles and impacts of different viral groups on microbiome diversity and biogeochemical cycles across ecological regimes.

**Abstract Text:** Viruses are key elements of all microbiomes on Earth, shaping microbial community composition and structure while influencing host cell metabolism during infection. Recent viral ecology studies powered by ‘omics approaches have provided a thorough description and investigation of the virosphere in many ecosystems, but have been challenging to apply to the incredibly diverse soil viruses, so that the exploration of global soil viral diversity is still only getting started<sup>1</sup>. In addition to establishing a global census of viruses across soil types and locations, one of the major outstanding questions in soil viral ecology is the characterization of soil virus dynamics across time, both in terms of diversity and activity. At the East River, CO, watershed, soil microbiomes respond to dramatic changes in subsurface conditions through snowfall, snowmelt, plant re-emergence, and plant senescence. The snowmelt period in particular results in a large crash of microbial biomass and shifts in community composition<sup>2</sup>. To characterize phage diversity, dynamics, and potential ecosystem impacts during this period we leverage 44 paired metagenomes and metatranscriptomes sampled across a whole year in this montane meadow ecosystem soil. A combined assembly approach enabled by the recently developed MetaHipMer tool<sup>3</sup> yielded >3,800 DNA phage populations whose abundance and activity could be followed through the entire year. Overall, these DNA phages were associated with the main bacterial groups present, around one third were reliably predicted as temperate, and most were consistently detected through the year albeit with low relative abundance. Transcriptional activity could be detected for both abundant and rare DNA phages, but consisted most often in the expression of one or two individual genes, and was thus more compatible with either self-regulation of temperate phages or host cell manipulation rather than active phage replication, since the latter would be associated with widespread expression of a large fraction of the phage genes. This suggests that a substantial portion of the DNA phage community in this ecosystem may reside in their host cell for prolonged period of time without actively replicating, but maintaining a basal level of expression for key phage-encoded genes.

Meanwhile, an unexpectedly abundant and diverse community of RNA phages could be detected in the same samples totaling >3,500 distinct populations, which in contrast to the DNA phages displayed a high turnover rate between time points, and included a substantial portion which were predicted as actively replicating (~10–30%). Taken together, the picture starting to emerge from these data is one of a relatively stable DNA phage community with sporadic activity and primary impact likely stemming from host cell manipulation during latent infections, alongside a more dynamic and active RNA phage community which may be responsible for a substantial fraction of the viral lysis in these microbiomes. More broadly, these observations of contrasted diversity and activity for DNA and RNA phages across seasons in a montane soil ecosystem highlights the need to expand our understanding of the eco-evolutionary drivers of phage diversity and activity in different soil types, especially for RNA phages, and demonstrates how multi ‘omics approaches can provide a unique data framework to start such characterization at both local and global scales.

### References:

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