The Genetic Potential of Microbial Depolymerization of SPRUCE Peatland in Marcell Experimental Forest, MN

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Amino acid production by microbial extracellular proteases may be the critical step for amine production and release of bioavailable N from forest soil. Yet little is known about the abundance and diversity of the microbial extracellular protease encoding genes in the peatland ecosystems. To evaluate the genetic potential of microbial depolymerization in peatlands, four surface bog and fen peat samples (0-10 cm, two for each) and two deep bog peat samples (25-50 cm and 50-75 cm) were collected from the Spruce and Peatland Responses Under Climatic and Environmental Change (SPRUCE) experiment site, MN. Shotgun metagenomic sequencing was performed on all six samples and guided gene assembly was used to extrapolate microbial extracellular protease genes.\r\nWe found abundant and diverse proteases within peat metagenomes, comprising nine microbial extracellular protease gene groups and a total of 86,351 microbial extracellular protease genes. These assembled contigs encode for proteases belonging to metallo, aspartic, serine and unknown protease families. The overwhelming majority of genes resembled known proteases of bacterial origin, encompassing 84-95% of the identified proteases per metagenome, while the remaining proteases were similar to protease genes in fungi and archaea. Among identified proteases, aminopeptidase and trypsin encoding sequences were the most prevalent in all samples (approximately 52% and 17% of total assembled microbial extracellular protease genes, respectively). Further, genes similar to extracellular proteases associated with an uncharacterized bacterial protein family were the third most abundant (~16% of total assembled microbial extracellular protease genes). Microbial extracellular protease genes were most abundant and diverse in surface peatland. Approximately 15,025 microbial extracellular protease genes (from 676 species) were assembled from each 0-10 cm sample, compared to ~6,531 (from 286 species) from 25-50 cm and ~4,042 (from 156 species) from 50-75 cm. With increasing depth, the number of observed fungal extracellular protease genes and the number of unique fungal strains that host these genes decreased drastically below 25 cm, while similar phenomenon was observed on bacterial extracellular protease genes below 50 cm. Archaeal extracellular protease genes were more abundant in deep samples than surface samples. Our results indicate that peatland systems have a stratified microbial extracellular protease potential across geographies and depths. High abundance of bacterial extracellular protease genes also suggests that besides fungi, bacteria may play a substantial role in depolymerization of organic matter in peatland systems.