TITLE:

BrdU Incorporation Study

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PHYSICAL LOCATION:

Oak Openings Preserve Metropark, Northwest Ohio

Latitude: 41.55

Longitude: -83.83

Stranahan Arboretum, Toledo, Ohio

Latitude: 41.70

Longitude: -83.67

DATA SET OVERVIEW:

This data set contributes information on soil enzyme activities; extractable nitrogen and phosphate; and microbial biomass. Data were collected from a 416 day incubation experiment including two treatments (sugar maple + sandy soil and white oak + sandy soil) and a no-litter control. These data are part of a BrdU incorporation study aimed at characterizing the active decomposer community at different stages of decay. Nucleotide analog labeling, such as BrdU incorporation, allows DNA to be isolated from active members of a microbial community through incorporation of a thymidine analog into newly synthesized DNA.

BACKGROUND:

<http://www.eeescience.utoledo.edu/Faculty/weintraub/Projects.htm>

SAMPLE COLLECTION:

*Leaf and Soil Collection*: Freshly senesced sugar maple (*Acer saccharum)* and white oak (*Quercus alba*)leaf litter was collected from the Stranahan Arboretum and Oak Openings Preserve near Toledo, Ohio using litter traps in the fall of 2008. Litter was air-dried and kept at constant humidity until incubation. Litter was ground into a fine powder using a Wiley Mill (20 mesh). A sandy soil (<0.5% Carbon) was collected from the Oak Openings Preserve located in Northwest Ohio. Soil was collected using a metal soil corer with a diameter of 5 cm to a depth of 5 cm. Soil was then taken immediately back to the lab and sieved (2 mm mesh) to remove coarse debris and organic matter and then homogenized by hand. Soil was stored under incubation conditions of 30% water holding capacity and 20 °C for at least 2 weeks.

*Incubation*: Twelve total jars were established at the beginning of the experiment. Four jars contained 10 g ground sugar maple litter and 100 g soil, four jars with 10 g oak litter and 100 g soil, and 4 -100 g soil only jars for a total of 12 jars. Jars were kept loosely covered (to minimize water loss, but allow gas exchange) in a dark incubator at 20 °C throughout the incubation. Subsamples from jars were harvested for soil enzyme activities; extractable nitrogen and phosphate; and microbial biomass (see below for descriptions) on days 0, 2, 54, 207, and 416. During harvests, soils were extracted with 0.5 M potassium sulfate (K₂SO₄). Extractions were performed by placing samples on an orbital shaker at ~120 rpm for 1 hour, and then vacuum filtering the mixture through Millipore APM 15 glass fiber filters. These extracts are labeled as non-fumigated.

Samples were harvested for microbial biomass. Microbial biomass carbon (MB-C) and microbial biomass nitrogen (MB-N) were quantified using a modification of the chloroform fumigation-extraction technique (Brookes et al. 1985; Scott-Denton et al. 2006). Following chloroform fumigation, soils were extracted as described above. These extracts are labeled as fumigated. Fumigated and non-fumigated extracts were then frozen until time of analysis.

DATA COLLECTION:

*Soil Nutrients*: Soil nutrients were analyzed using non-fumigated soil samples. Nitrogen analyses included nitrate and ammonium. Nitrate was analyzed using the method described by Doane & Horwath (2003). Final nitrate values are reported in μg NO3⁻ g-1 dry soil. Ammonium was analyzed using the method described by Rhine *et al*. (1998). Final ammonium values are reported in μg NH4+ g-1 dry soil. Phosphate analysis was conducted using the method described by D’Angelo *et al*. (2001). Final phosphate values are reported in μg PO43- g-1 dry soil.

*Microbial Biomass*: Microbial biomass C and microbial biomass N were analyzed using fumigated and non-fumigated soil extracts. Samples were analyzed for dissolved organic carbon (DOC) and total nitrogen (TN) using a Shimadzu total organic carbon (TOC-VCPN) analyzer equipped with a total nitrogen unit (Shimadzu Scientific Instruments Inc., Columbia, MD, USA). Microbial biomass was calculated by subtracting non-fumigated sample concentrations of total organic carbon or total nitrogen from fumigated sample concentrations of carbon and nitrogen. Final values are reported in μg C or N g-1 dry soil.

*Soil Enzyme Activities*: Hydrolytic and oxidative extracellular enzyme activities were analyzed using soil samples. Samples were analyzed for β-glucosidase (BG), N-acetyl-β-glucosaminidase (NAG), acid phosphatase (PHOS), peroxidase (PEROX) and phenol oxidase (PHENOX). BG produces glucose from the hydrolysis of cellulose oligomers; NAG, a chitinase, produces N-acetyl glucosamine from the hydrolysis of chitin derived oligomers; PHOS produces phosphate from the hydrolysis of phosphate monoesters such as sugar phosphates; PEROX is an oxidative enzyme involved in lignin degradation; PHENOX is an oxidative enzyme involved in lignin degradation. Hydrolytic enzyme activities (BG, NAG, and PHOS) were analyzed using the fluorometric assay described by Saiya-Cork *et al*. (2002). Oxidative enzymes (PHENOX and PEROX) were analyzed using the colorimetric assay described by Saiya-Cork *et al*. (2002).

NAMING CONVENTIONS:

The following refer to headers or terms used in the data spreadsheets:

*General Definitions*

day: day of incubation when measurements occurred

replicate: replicate number (n=4)

litter treatment: sugar maple + sand; oak + sand; sand only (control)

blank cells=no data available; cell value = 0 means no activity

*Harvests*

NH4: µg NH4-N g-1 soil from 0.5M K2SO4 extractions using microplate assay

NO3: µg NO3-N g-1 soil from 0.5M K2SO4 extractions using microplate assay

PO4: µg PO4-P g-1 soil from 0.5M K2SO4 extractions using microplate assay

BG: Beta glucosidase activity, nmol hr-1 g-1 soil (fluorescent enzyme assay protocol)

NAG: N-acetyl-beta-glucosaminidase activity, nmol hr-1 g-1 soil (fluorescent enzyme assay protocol)

PHOS: Phosphatase activity, nmol hr-1 g-1 soil (fluorescent enzyme assay protocol)

PHENOX-DOPA: Phenol oxidase activity (L-DOPA as substrate). µmol g-1 soil hr-1 (colorimetric microplate protocol)

PEROX: Peroxidase activity (L-DOPA as substrate). µmol g-1 soil hr-1 (colorimetric microplate protocol), Net Peroxidase reported (Perox-Phenox)

PHENOX-ABTS: Phenol oxidase activity (ABTS as substrate). µmol g-1 soil hr-1 (colorimetric microplate protocol)

MBC: microbial biomass C, ug C g-1 soil, measured in 0.5 M K2SO4 extract on Shimadzu analyzer

TOC: Total dissolved organic C, ug C g-1 soil, measured in 0.5 M K2SO4 extract on Shimadzu analyzer

TN: Total dissolved N, ug N g-1 soil, measured in 0.5 M K2SO4 extract on Shimadzu analyzer

MBN: microbial biomass N, ug N g-1 soil, measured in 0.5 M K2SO4 extract on Shimadzu analyzer

LINKS:

<http://www.eeescience.utoledo.edu/Faculty/weintraub/Projects.htm>

REFERENCES:

Brookes, P.C., Landman, A., Pruden, G., Jenkinson, D.S., 1985. Chloroform fumigation

and the release of soil nitrogen: a rapid direct extraction method to measure

microbial biomass nitrogen in soil. Soil Biology & Biochemistry 17, 837e842.

D'Angelo, E., Crutchfield, J., Vandiviere, M., 2001. Rapid, sensitive, microscale determination of phosphate in water and soil. J. Environ. Qual. 30, 2206-2209.

Doane, T.A., Horwath, W.R., 2003. Spectrophotometric determination of nitrate with a single reagent. Analytical Letters 36, 2713-2722.

Rhine, E.D., Sims, G.K., Mulvaney, R.L., Pratt, E.J., 1998. Improving the berthelot reaction for determining ammonium in soil extracts and water. Soil Sci Soc Am J 62, 473-480.

Saiya-Cork KR, Sinsabaugh RL, Zak DR, 2002. The effects of long term nitrogen deposition on extracellular enzyme activity in an Acer saccharum forest soil. Soil Biology and Biochemistry 34: 1309e1315.

Scott-Denton, L.E., Rosenstiel, T.N., Monson, R.K., 2006. Differential controls by climate and substrate over the heterotrophic and rhizospheric components of soil respiration. Global Change Biology 12, 205-216.