

Detrital Input and Removal Treatment (DIRT) Network Soil Analysis in the UMBS

Michigan Field Site

by

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Abstract

Soil contains approximately 75% of the carbon pool on land - three times more than the amount stored in living plants and animals (Schlesinger 1999). Therefore, soils play a major role in maintaining a balanced global carbon cycle. Models of the soil carbon ecosystem assume a strong relationship between organic matter (litter) inputs and soil carbon accumulation, but there is little evidence for this assumption. To test the relationship between stored soil carbon and organic matter alterations, we used the DIRT (Detrital Input and Removal Treatment) Network on 27 plots of soil. The DIRT Network assesses how rates and sources of plant litter inputs influence accumulations or losses of organic matter in forest soils. The soil plots are located at the University of Michigan Biological Station (UMBS). Each plot had two depths, the first being a 0-10cm profile and the other a 10-20cm profile. The results showed that the 0-10cm depth trial had significant increases from the woody debris addition and significant decreases in the no-input exclusion. The 10-20cm depth trial showed no significant results across all trials. We concluded that the increase in stored soil carbon from applied woody debris was more likely because the woody debris takes longer to decompose rather than that the decomposition yielded more stored carbon. Too, we discussed that the lack of organic matter content from the no-input trial is representative of the decreased decomposition rate and lack of microbial interest.

Introduction

The soil carbon (C) cycle plays a key role in regulating the Earth's global temperature and climate by exchanging carbon dioxide in flux with the atmosphere. Within the soil, carbon is held in carbon stocks. Carbon stocks are the amount of carbon sequestered

from the atmosphere and are now stored within the forest ecosystem, mainly within living biomass and soil, and to a lesser extent in wood and litter. The carbon cycle assumes a strong relationship between organic matter (OM) inputs and soil organic carbon (SOC) accumulation in many ecological outlines. This assumption has little direct evidence of the relationship between additions and exclusions of organic matter and the stored soil organic carbon content.

Carbon levels in the soil are rapidly depleted from soil organic matter (SOM) decomposition. Soil organic matter is derived from decomposing plant detritus and microbial materials, modified by biotic and abiotic processes. Due to the activity of the microbial decomposer community and reduced amounts of litter via decomposition, there is a decrease in stored soil carbon. The factors that affect stored soil carbon stability and formation are not fully understood and further research is needed to determine the result of altering the amount and type of organic litter within soils. This experiment aims to deduce if changes in detrital organic matter inputs alter the soil's carbon stocks using the DIRT Network.

The Detrital Input and Removal Treatment (DIRT) Network assesses how rates and sources of plant litter inputs influence accumulations or losses of organic matter in forest soils. Within this experiment, the plant litter treatments include the addition and exclusion of organic plant litter and roots in a variety of ways per plot. Each plot contains a different treatment: control (no addition or exclusions of litter or roots), double litter (doubled annually), double woody litter (wood chips applied every other year), removed litter (removed annually in late fall), no litter inputs and removed roots, and the addition of double litter with additional fertilization. The double litter trials were

separated into two groups: fertilized and non-fertilized. The fertilized treatment received 30 kg N/ha/year as Ammonium Nitrate, applied in 3 equal amounts in spring, summer, and fall. The NH_4NO_3 for each aliquot for two plots was dissolved in deionized water in a 3L pesticide sprayer. These changes were made to each plot for long-term observation of organic carbon content within the soil to observe if the soil carbon concentration changes significantly.

This experiment attempts to answer the question; will increased detrital inputs in forested ecosystems result in significant increases in total soil C? I hypothesize that regardless of the factors contributing to each plot, the carbon content of the soil will not change significantly. I theorize my null hypothesis will be true due to soil priming effects with the microbial activity within the soil. Priming occurs when organic matter input into soils accelerates the decomposition of native soil carbon. In priming, inputs of C to soil enhance the microbial degradation of the C in soil that was already present, C that would have, otherwise, remained SOM. Too, increased plant residue inputs provide more substrate for soil microorganisms, resulting in a more active and more abundant microbial community. Therefore, plant litter provides more fuel for the microbes rather than more carbon storage capacity.

Methods

Site description

The location of this field site is within the University of Michigan Biological Station, United States. Michigan has a humid continental climate. Some parts of the state average low temperatures below freezing from December through February, and into early

March in the far northern parts. During the winter through the middle of February, the state is frequently subjected to heavy lake-effect snow. The state averages 30-40 inches of precipitation annually. Michigan is found in the temperate forest area. The principal forest trees include basswood, maple, elm, sassafras, butternut, walnut, poplar, hickory, oak, willow, pine, birch, beech, hemlock, witch hazel, tamarack, cedar, locust, dogwood, and ash. The dominant soils found in this area include spodosols, histols, and inceptisols. The University of Michigan Biological Station (UMBS) is located in the northernmost part of Michigan's lower peninsula (45°56'N 84°71'W). The UMBS DIRT study area is located on a high outwash plain which has been measuring CO₂ and water vapor fluxes year-round since 1999. Soils have low fertility, with total N stocks to 40 cm depth of 2000 kg ha⁻¹, an average in situ net N-mineralization rate of 42 kg N ha⁻¹ yr⁻¹, and <2% net nitrification (Nave et al. 2009). The mean annual temperature is 5.5° C (1942-2011) and the mean annual precipitation is 817 mm (including 294 cm snowfall).

The forest at the site is 90 to 100 years old and is dominated by mature bigtooth aspen (*Populus grandidentata*) which was established soon after regional forest harvesting and burning. Other canopy dominants and co-dominants include red maple (*Acer rubrum*), red oak (*Quercus rubra*), white birch (*Betula papyrifera*), Eastern white pine (*Pinus strobus*), trembling aspen (*Populus tremuloides*), sugar maple (*Acer saccharum*), and American beech (*Fagus grandifolia*). Forest age, composition, and disturbance history at this study site are representative of forests across the northern Great Lakes region, where aspen and birch-dominated hardwoods replaced pine-hemlock (*Tsuga canadensis*) forests following clearcutting and wildfires.

The UMBS DIRT site soil is a well-drained Haplorthod of the Rubicon series. The forest floor (Oe horizons) is 0-3 cm thick and overlies a bioturbated (by earthworm invasion) AO horizon of 1-3 cm held together by fine roots. AO horizons are underlain by an E

horizon of 10-15 cm and a Bs horizon of sand with occasional gravel. About 53% of the fine root mass is located within the upper 20 cm of the soil profile. Forest floor C mass is 5-15 Mg C ha⁻¹, and the mineral soil is ~95% sand and ~5% silt, with pH 4.5-5.5 in water (Gough et al. 2013).

Sample Collection and Handling

Sampling methods included taking two profiles including 0-10cm and 10-20cm increments per plot. There are 27 plots in total. The mineral soil increments were taken with an Oakfield core, yielding enough material for 100g composites per plot.

Bulk Soil Process

The bulk soil samples were dried for 48 hours using a drying oven. Each dry soil sample weighed between 39 and 43 grams. The samples were ground into fine dust. A small sample weighing 50 milligrams of the fine-ground soil was encapsulated into a square of foil. These steps were repeated for each soil sample, totaling 27 samples. The samples were run in an Elementar machine for the analysis of carbon and nitrogen concentrations.

The bulk soil process is not referenced in my research and does not apply to the findings below.

Density Fractionation Process (Lajtha and Pierson, n.d.)

The desired density for sodium polytungstate (SPT) in this experiment was 1.85 g/cm³. A 1000 ml 1.85 g/cm³ solution was created using 1051 grams of SPT per 799 ml of water.

Approximately 40.00 grams of the dried soil sample was added to a 250ml centrifuge tube and the mass was recorded. Fifty milliliters of the SPT solution were added to the centrifuge tube. The samples were shaken vigorously by hand for about two minutes, then set on the shake machine on the low setting for two hours. Every 30 minutes or so, the machine was stopped and shaken by hand for another two minutes. After the two hours passed, the centrifuge was balanced by weighing out each tube sample and identifying that all were very similar weights (within 1.5 grams). The samples were centrifuged at 3000rpm for ten minutes. The light fraction of the soil sample was aspirated. The addition of SPT was repeated to the same capacity for each sample and shaken for ten minutes on the shake machine on the low setting for each soil sample. After each round of centrifuging, the light fraction of the soil sample was aspirated. Then, these steps are repeated twice with water instead of the SPT solution. The separated heavy and light fractionation samples were dried in the drying oven for forty-eight hours. Each dry heavy fraction soil sample weighed approximately 30 to 40 grams. Each dry light fraction soil sample weighed approximately 0 to 7 grams. The separated samples were ground into a fine dust. A small sample weighing 50 milligrams of the fine-ground soil was encapsulated into a square of foil for each heavy and light fraction sample. These steps were repeated for each soil sample, totaling 54 samples per group (27 of the heavy, 27 of the light). The samples were run in an Elementar machine for the analysis of carbon and nitrogen concentrations.

Density fractionation of soil divides material by floatation or sedimentation in a solution according to particle density to physically separate soil organic matter into discrete fractions. Fractionation divides the soil organic matter into light and heavy fractions. The light fraction is composed of organic matter such as leaves, fine roots, and decaying

woody debris. Heavy fraction contains mineral clays, larger-sized soil particles, and rock. This process is important to the study because it allows the separation of free and obstructed organic matter without altering the carbon or nitrogen content.

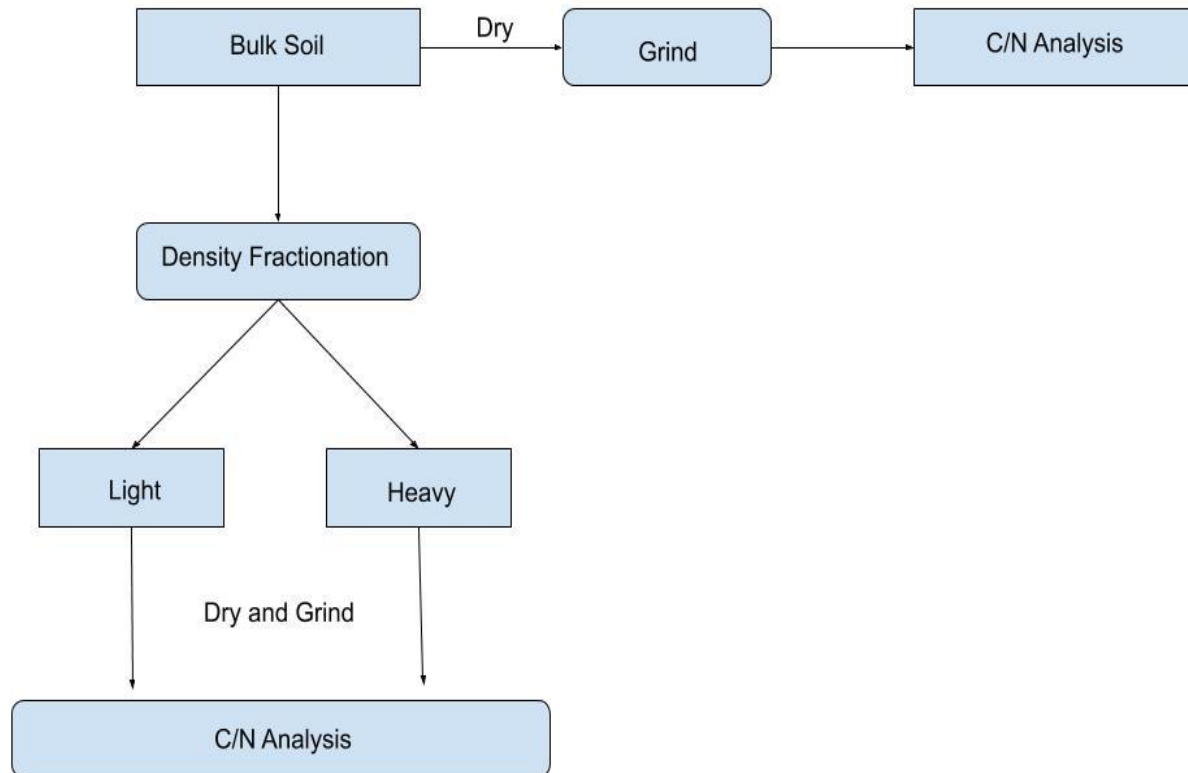


Fig. 1 Flow diagram of the process of soil carbon and nitrogen analysis.

Results

Total mean response to detrital manipulation 0-10cm

There were significant differences in total mean soil C among detrital treatments in the

0-10cm depth forested plots (Fig. 2). The total mean control C in milligrams per gram of bulk soil was 9.75 C mg/g. Comparatively to the control value, the no-input trial was statistically significantly lower than the control at a value of 6.95 C mg/g, meaning a percent decrease of 28.7%. The total mean no litter trial did not have statistically significant results compared to the control at a value of 8.98 C mg/g. The total mean woody litter addition trial significantly increased from the control at a value of 20.27 C mg/g and a percent increase of 107.9%. The total mean double litter fertilized treatment significantly increased by 36.1% at a value of 13.27 C mg/g. The total mean double litter non-fertilized treatment significantly increased by 45.9% at a value of 14.2 C mg/g.

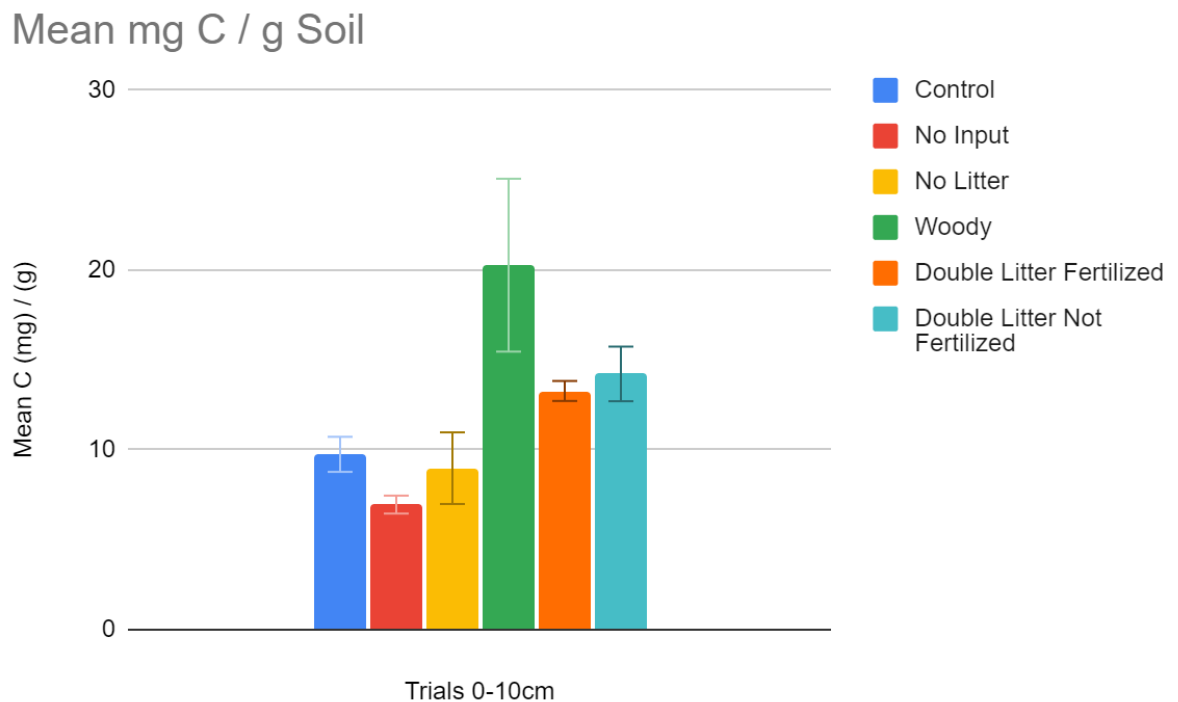


Fig. 2 Combined heavy and light fraction SOC concentrations in the Michigan DIRT Plots trials 0-10cm.

Heavy fraction mean response to detrital manipulation 0-10cm

There were significant differences in heavy fraction mean soil C among detrital treatments in the forested plots (Fig. 3). The heavy fraction mean control C in milligrams per gram of bulk soil was 2.92 C mg/g. Comparatively to the control value, the no-input trial was statistically significantly lower than the control at a value of 1.49 C mg/g, meaning a percent decrease of 48.8%. The heavy fraction mean no litter trial was significantly lower than the control at a value of 2.48 C mg/g and a percent decrease of 17.5%. The heavy fraction mean woody litter trial was significantly higher than the control value at 3.52 C mg/g and a percent increase of 20.4%. The heavy fraction mean double litter fertilized trial was slightly significantly lower at a value of 2.64 C mg/g and a percent decrease of 9.6%. The heavy fraction mean double litter non-fertilized trial was significantly lower at a value of 2.17 C mg/g and a percent decrease of 25.5%.

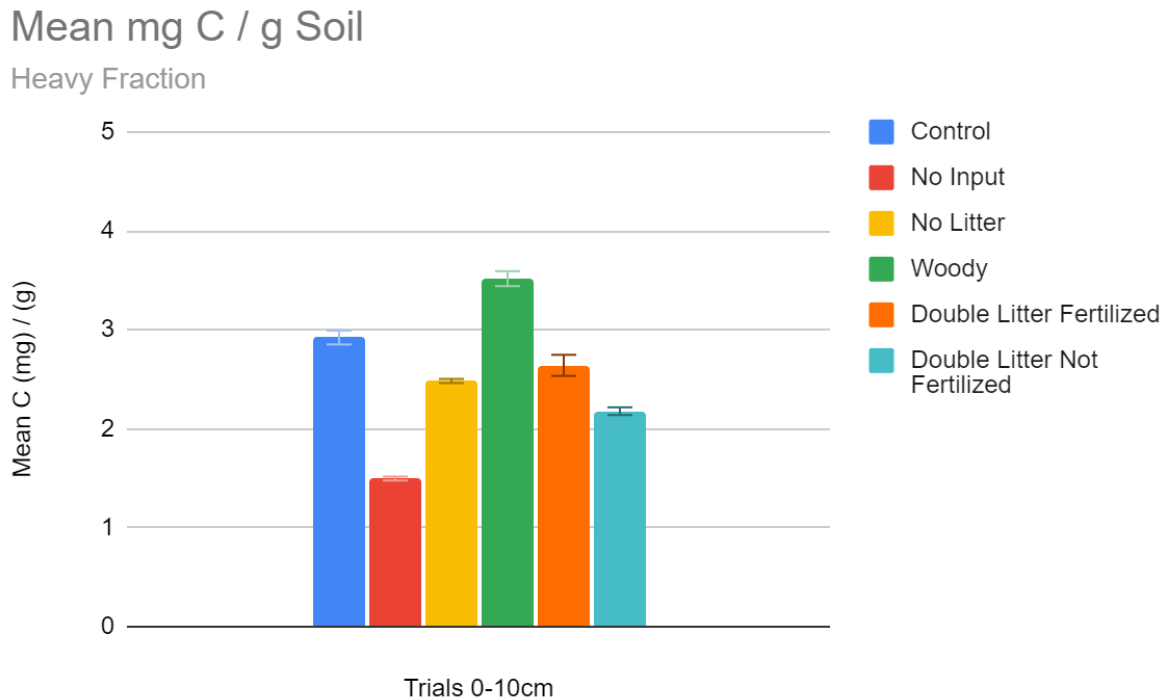


Fig. 3 Heavy Fraction SOC concentrations in the Michigan DIRT Plots trials 0-10cm.

Light fraction mean response to detrital manipulation 0-10cm

There were significant differences in light fraction mean soil C among detrital treatments in the forested plots (Fig. 4). The light fraction mean control C in milligrams per gram of bulk soil was 6.83 C mg/g. Comparatively to the control value, the no-input trial was very slightly significantly lower at a value of 5.46 C mg/g and a percent decrease of 20.1%. The light fraction mean no litter trial was not significantly different from the control at a value of 6.50 C mg/g. The light fraction mean woody litter trial had a significant increase at a value of 16.75 C mg/g and a percent increase of 145.4%. The light fraction double litter fertilized trial was significantly higher at a value of 10.63 C mg/g and a percent increase of 55.7%. The light fraction mean double litter non-fertilized trial was significantly higher at a value of 12.04 C mg/g and a percent increase of 76.4%.

Mean mg C / g Soil

Light Fraction

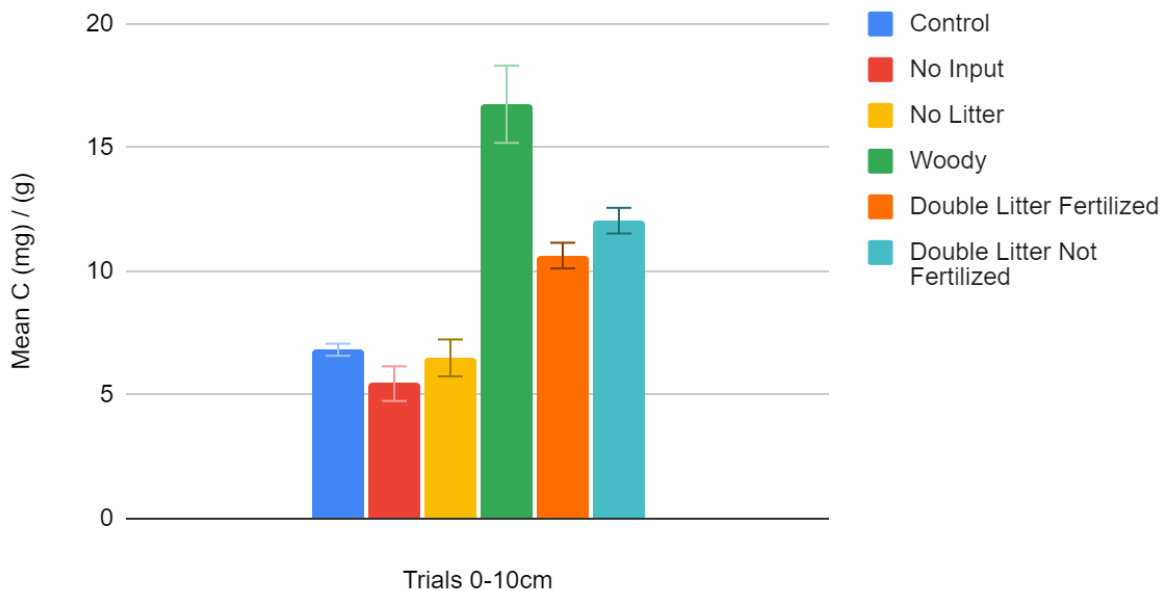


Fig. 4 Light Fraction SOC concentrations in the Michigan DIRT Plots trials 0-10cm.

Total mean response to detrital manipulation 10-20cm

There were no significant differences in total mean soil C among detrital treatments in the 10-20cm depth forested plots (Fig. 5). The total mean control C in milligrams per gram of bulk soil was 2.99 C mg/g. The total mean no input trial did not have statistically significant results compared to the control at a value of 6.15 C mg/g. The total mean no litter trial did not have statistically significant results compared to the control at a value of 2.94 C mg/g. The total mean woody litter trial did not have statistically significant results compared to the control at a value of 3.92 C mg/g. The total mean double litter fertilized trial did not have statistically significant results compared to the control at a value of 2.97 C mg/g. The total mean double litter non-fertilized trial did not have statistically significant results compared to the control at a value of 3.45 C mg/g.

Mean mg C / g Soil

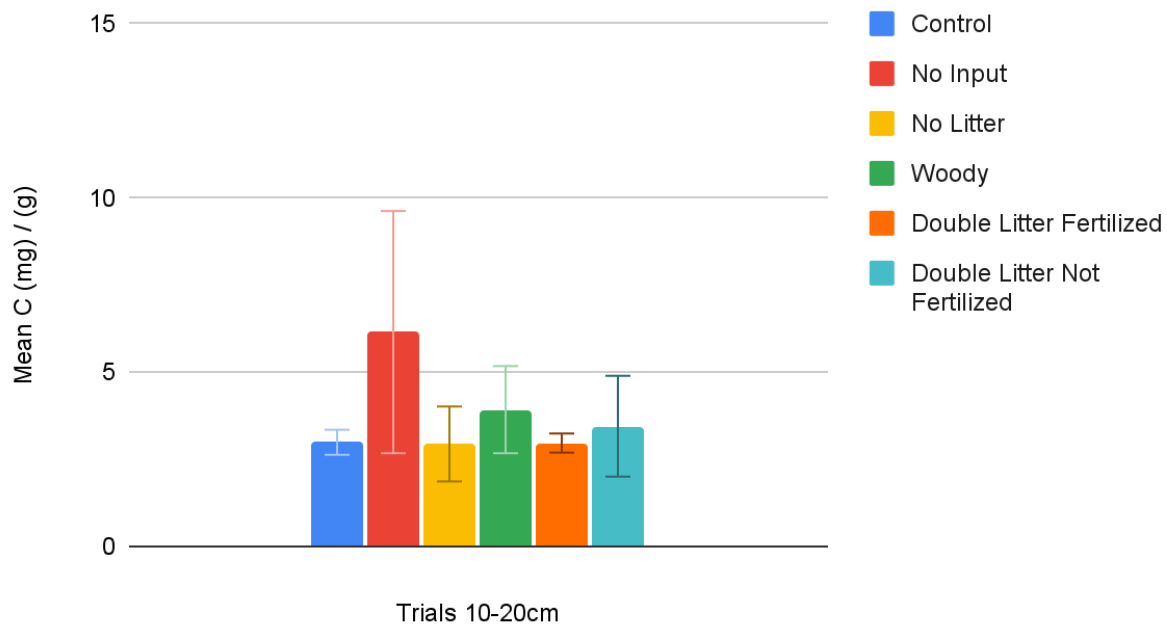


Fig. 5 Combined heavy and light fraction SOC concentrations in the Michigan DIRT Plots trials 10-20cm.

Heavy fraction mean response to detrital manipulation 10-20cm

There were significant differences in heavy fraction mean soil C among detrital treatments in the forested plots (Fig. 6). The heavy fraction mean control C in milligrams per gram of bulk soil was 2.99 C mg/g. The heavy fraction mean no input trial was not statistically significant from the control at a value of 0.99 C mg/g. The heavy fraction no litter trial was significantly higher than the control at a value of 1.82 C mg/g and a percent increase of 39.1%. The heavy fraction woody litter trial was not statistically

significant from the control at a value of 1.05 C mg/g. The heavy fraction double litter non-fertilized trial was not statistically significant from the control at a value of 1.28 C mg/g. The heavy fraction double litter fertilized was statistically significant from the control at a value of 0.75 C mg/g and a percent decrease of 42.8%.

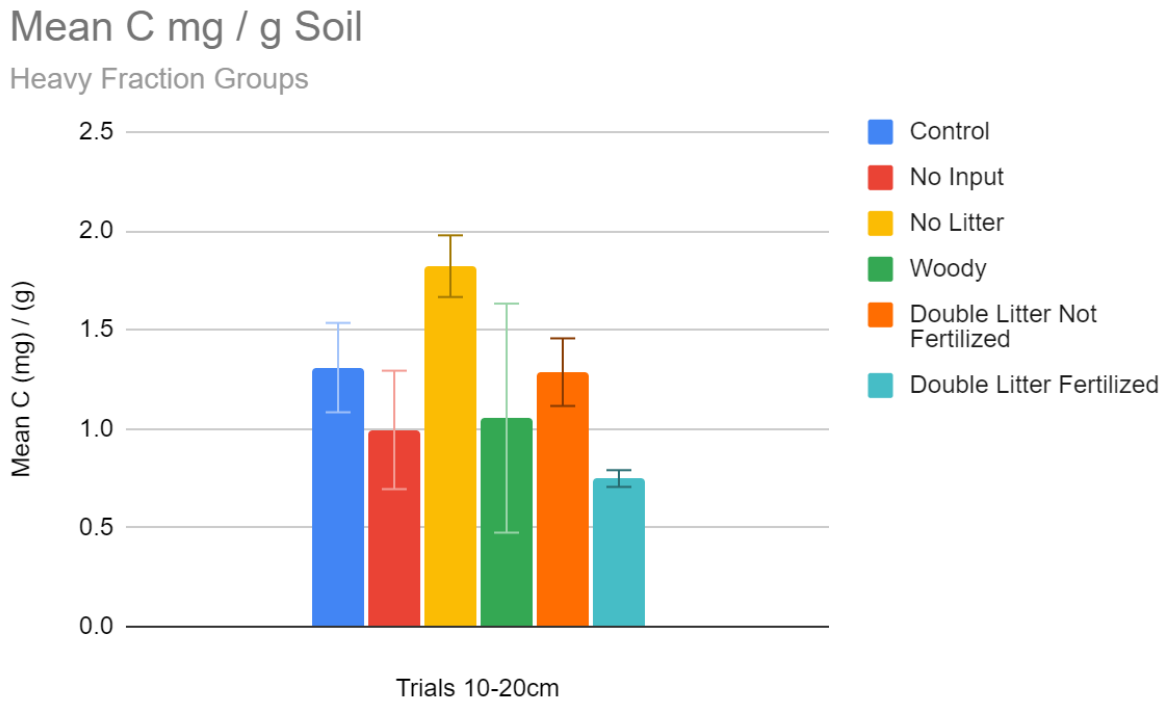


Fig. 6 Heavy Fraction SOC concentrations in the Michigan DIRT Plots trials 10-20cm.

Light fraction mean response to detrital manipulation 0-10cm

There were significant differences in light fraction mean soil C among detrital treatments in the forested plots (Fig. 6). The light fraction mean control C in milligrams per gram of bulk soil was 1.68 C mg/g. The light fraction mean no input trial was slightly statistically significant from the control at a value of 5.15 C mg/g and a percent increase of 207.27%. The light fraction no litter trial was not significant than the control at a value of 1.68 C

mg/g. The light fraction woody litter trial was not statistically significant from the control at a value of 2.87 C mg/g. The light fraction double litter non-fertilized trial could not be tested for statistical significance from the control at a value of 8.48 C mg/g because there was only one sample. The light fraction double litter fertilized was slightly statistically significant from the control at a value of 2.38 C mg/g and a percent increase of 41.9%.

Mean C mg / g Soil

Light Fraction Groups

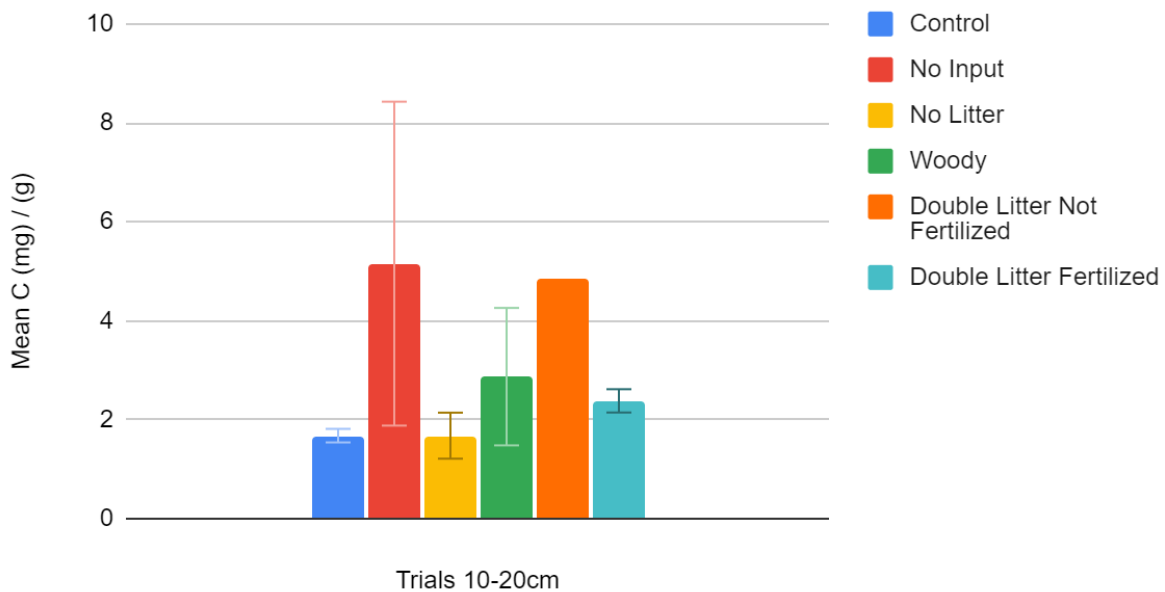


Fig. 7 Light Fraction SOC concentrations in the Michigan DIRT Plots trials 10-20cm.

The 0-10cm depth data are on a scale twice the value of the 10-20cm depth data as there is more carbon stored in the top 10cm of soil than the lower 20cm. The lower the soil profile layer, the less organic matter content, the less available carbon content. The heavy fraction samples consist of mostly sand and silt with little to no organic matter composition, resulting in far lower averages for carbon concentration. The light fraction samples contain most organic matter materials, either woody debris or other general

forest litter. This results in the light fraction samples having high carbon content.

Discussion

SOM is a major component of the global C cycle, containing more C than plant biomass and the atmosphere combined. Despite the key role of SOM in the global carbon cycle, interactions among the biological, chemical, and physical processes regulating SOM storage, accumulation, and stabilization are poorly understood (Lajtha et al. 2014). This study is important because identifying how different types of organic matter accumulations or exclusions alter carbon stocks gives implications for more sustainable farming practices and methods of keeping sequestered carbon in the soil rather than released into the atmosphere. The null hypothesis of this study is that regardless of the alterations made to the different plots, the carbon content would stay consistent with the numbers observed in the control plots. By observing the conclusions of this study, we find varied results.

In this study, I measured the carbon content in mg per gram bulk soil under trials that included additions or exclusions of organic matter to determine changes in carbon concentration. By analyzing the carbon content in each trial via a carbon and nitrogen Elemental machine, I've identified that, in the 0-10 cm depth trials, the addition of woody litter significantly increased the SOC compared to the control trial. Too, within the 0-10cm depth trials, the no-input trial significantly decreased in carbon content compared to the control trial. In the 10-20 cm depth trials, there were very little to no significant findings- aligning with the null hypothesis.

The 10-20cm depth trials did not undergo significant change. The carbon within the litter

takes a long period to fully dissolve into the deeper horizons of the soil profile. Once the carbon is integrated into the deeper horizons, it's more likely to stay than the carbon that is left near the topsoil. The element with the highest carbon content per gram of substrate is organic matter. Since organic matter is being applied to the topsoil, and the changes are measured in human lifespan years, the lack of significant changes in carbon content in the deeper horizon is not surprising.

Why do we see increased soil carbon with the addition of woody debris? I hypothesize that it is not because the highly fragmented wood became enhanced microbial biomass and stuck around in the soil. Woody debris contains lignin. The strong phenyl rings within lignin result in a slow decomposition process. Wood decomposes at a slower rate than other types of forest litter. Too, there is very little nitrogen in wood. Wood is not photosynthetic, so there are no enzymes within the woody debris that contributes to nitrogen content. The microorganisms within the soil are attracted to organic matter with higher nitrogen content and woody debris is low in nutrition, making it the last choice of organic matter among microbes. Overall, the null hypothesis is rejected for the 0-10cm depth trial and failed to reject for the 10-20cm depth trial.

Why is there decreased soil carbon with the exclusion of organic matter and roots? The lack of stored soil carbon content from the no-input trial is representative of a lack of organic matter. The microbial community feeds off of carbon, nitrogen, and other soil nutrients. Most of these nutrients come from organic matter. When there is not enough organic matter for a microbe to comfortably survive, they move on to greener pastures, literally.

In a journal from Lajtha et al. (2014), the addition of double litter has shown increases in SOC over 50 years of treatment in Wisconsin. This journal reports a significant decrease

in the no-input trial which aligns with the results found in my study. It's important to note that the soils within the Wisconsin study contain more organic matter, clay, and silt particles than the sandy soils of Michigan. In a journal from Lajtha et al. (2018), there was a significant decrease in SOC in the no-input and no litter at the HJ Andrews site. The evidence of decreased stored SOC in the no-input trials aligns with the alternative hypothesis that the changes to topsoil organic matter influence the stored SOC. I find this relationship to be consistent with other studies and my personal experience. It's important to note that while having zero organic matter input could result in lower stored carbon, the no litter treatment did not show the same significant results. The change between these two trials lies in what is being removed. In the no-input trial, there was an exclusion of both roots and organic matter. The no litter trial only excluded the organic matter. The baseline of these results yield that roots are an important factor in stored soil carbon. When a plant dies, the organic nutrients within the plant decompose back into the soil for soil microbes to ingest, same goes for the roots of the plant. Dead root material may be enough substance for the microbes to feed off, resulting in stored carbon levels relative to the control trial. Since the roots are in the soil substrate below the ground, the carbon is more likely to store and less likely to respire into the atmosphere as aboveground plant mass does.

I would like to revisit this experiment and practice the same methods on the soils in another 10 to 20 years. I feel that giving the 10-20cm depth soils more time to interact with the additions and exclusions of the study will yield significant results. I would hypothesize that the 10-20cm depth woody litter addition plots would end up having higher carbon content numbers because of the reasons listed above. Too, I would like to research the effect roots have on stored soil carbon.

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