

The Impact of Carrion Insects on Human Impacted Soil

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Abstract

Carrion plays an important role in maintaining ecosystems and providing resources for different forms of life, returning key nutrients to the environment and serving as a food source for animals. While vertebrates who consume carrion are well studied, invertebrates are not as well studied, primarily carrion insects. With the decline of mammalian scavengers due to human impact and removal of large vertebrate carcasses from ecosystems, there is a need to understand how effective insects are as scavengers, as they are common on carrion sources. In this experiment, different carrion insects were placed into containers containing poor soil, and after 3 weeks were removed and seeds were planted into the soil as a bioassay, where the root weight fraction was calculated and used to determine the quality of the soil. The results showed that carrion insects have varying influences on soil nutrients, with carrion beetles having more impact compared to flies and carrion alone.

Introduction

Carrion is an essential source of nutrients in any ecosystem, forming from dead organisms and providing much-needed nitrogen, phosphorus, calcium, and micronutrients (Benbow et al., 2016). The decomposition of carrion plays an important role in nutrient cycling. In an ecosystem, carrion decomposition that occurs in one location can create nutrient hotspots- “islands of fertility”- that can remain active in the

soil years after the death of the organism (Danell and Brathen, 2002). This source can come from large vertebrates like bison and elk to smaller organisms like passerine birds and small rodents. However, the impact large vertebrate carrion has on soil, along with other various animals from different trophic levels, has been well-studied (see Barton, 2016), but there are still knowledge gaps on the impacts of small vertebrate carrion, what is cycled through ecosystems, and how various invertebrate and other smaller organisms aid the cycling process (Barton et al. 2013). For example, insects play an important role in the decomposition of large and small deceased vertebrate organisms, yet it is poorly understood which insects benefit the most from carrion sources and how the nutrients are moved by insects (Ilardi et al., 2021). In a recent study by Ilardi, it was discovered that *Nicrophorus vespilloides* played a role in the regulation of nutrient cycling in soils, yet it is still not clear how other carrion beetles affect soil, or what other ways these carrion insects can affect soils. It is understood that when there is a carrion source and decomposition taking place insects will often be present, but just how much this affects the nutrient content on the soil where the carrion source is located, along with which insects return the most nutrients, still needs further study.

Carrion sources and animals that use them provide significant ecosystem services to humans. An important function that they play is their ability to keep ecosystems healthy, which humans benefit from as that is where we get all of the materials that allow us to function (Santangeli et al., 2019). Carrion animals specifically also help with lowering disease transmission, as they remove dead bodies that were infected with a disease, removing disease hotspots that would affect other animals (Patterson et al., 2022). For example, in areas where vulture populations were significantly lowered,

disease transmission increased between humans and wild animals, as well as increased human and animal conflict, such as wild dog attacks (Santangeli). Vultures lowered disease transmission due to having a specialized gut that is able to keep them from getting sick and spreading diseases. In the absence of vultures the carcasses, they would have consumed attracted mammalian scavengers who would become infected with diseases, such as rabies, and spread them to humans (Santangeli).

One method to determine soil fertility is to conduct a bioassay. A bioassay is conducted by using an organism to determine what chemicals or nutrients are present in the soil. For example, a growing plant can be used to determine if there are chemicals present that affect growth by measuring growth, and weight, and comparing plants grown in the soil in question with known soils (Neal, 2021). A bioassay can also be used to determine what nutrients are available to aid plant growth, by comparing plants thought to be impacted areas with plants with access to nutrients. In Denslow (et. al, 1987), it was determined that soil from a Costa Rican lowland rainforest lacked key nutrients, which were discovered in the planting of shrubs and comparing the shrubs to ones that were in fertilized soil. The size and weight of the rainforest shrubs were also smaller compared to the fertilized shrub, and nutrient analyses confirmed the rainforest shrubs were stunted. Along with this, the roots of the bioassay plants can help determine how fertile the soil is. Nutrient uptake for plants greatly depends on the geometry of their roots, focusing on root growth if they are in infertile soils (Fitter, 1986). A plant putting more energy into root growth than shoot growth is a common indicator of poor soil nutrition and can be measured by calculating root weight fraction (RWF) by dividing the dry root weight by the total dry plant weight (Fitter, 1986).

The goal of this experiment was to determine if carrion sources consumed by different carrion insects return different amounts of nutrients to the soil. I also developed a plant growth bioassay to indicate soil fertility by using the root weight fraction (RWF) and different soil types. This will be an experiment on its own, the pilot experiment, where I used soil that had a carrion source and beetles and soil with nothing added to it. I hypothesize that the grass grown on the soil with the beetles and carrion will have the lowest RWF value. For the second experiment, three different types of carrion insects were studied and compared with one another, as well as compared with soil that had no carrion or no carrion insect presence. Due to their carcass burying behavior, I hypothesized that *Nicrophorus investigator* would return a higher quantity of nutrients to the soil, especially when compared with soil where carrion remained on the surface and was utilized by other carrion insects.

Methods

Study organisms:

The carrion insects involved were two carrion beetle species in the Family Silphidae: *Nicrophorus investigator* (Subfamily Nicrophorinae: burying beetle), and *Thanatophilus lapponica* (Sub-family Silphinae), and the common species of blowflies or bottleflies (Family Calliphoridae, blue and green bottleflies). The beetles were collected from traps set up in Gothic, Colorado (Rocky Mountain Biological Laboratory) around the GRC and in the aspen forest north of Ore House. (see MAP, figure 1).

Carrion sources used were *Peromyscus maniculatus* carcasses salvaged from Gothic buildings (trapped by residents, donated to the project). Carcass salvage was

permitted by the Colorado Parks and Wildlife Mammalian Scientific Collection License 22TRO403 to R. Smith. Two carcasses were used in each treatment container, with a combined mass of ~30 g.

Soil source and decomposition treatments:

The soil was collected from the area behind the Gothic Research Center (GRC) where plants have hardly grown for the past 10 years after disturbance from the construction of the GRC. The soil was sifted (¼ inch metal screen) to remove large rocks and debris before being placed in experiment containers. After the soil was placed in 50 plastic containers the carrion treatment of each container began. The first soil treatment had no manipulation, just soil by itself in the container. The 2nd treatment was soil with just the carrion source (two dead *Peromyscus*, *see above*) laid on top of the soil and allowed to decompose without the presence of any carrion insect but had access to air and sunlight. The 3rd treatment was the carrion source and soil placed in a container that would allow flies to enter and leave. The 4th treatment had two breeding pairs of the black beetle (*Thanatophilus lapponica*) that were not allowed to leave the container, a total of 4 beetles in the container. The 5th treatment was *Nicrophorus investigator*, and each container had one breeding pair that raised a brood in the container. After three weeks all insects and larvae were removed from the container and the soil was prepared for the bioassay.

Soil fertility bioassay (plant growth):

While waiting for the insects to complete their breeding cycle and consume their carrion source, a pilot project was conducted to determine the achievability of the plant

growth bioassay. The purpose of the pilot study was to determine which plant growth measures worked the best to determine how nutrient rich or poor the soil samples were. I used soil from a container that housed a breeding pair of *Nicrophorus investigator* that had completed their breeding cycle, expecting the soil to be high-nutrient. I collected 26 soil samples and placed them into plastic growing tubes, with paper towels at the base to limit soil loss. I filled another 26 tubes with soil collected from the same site but did not have a burying beetle or carrion source contained in it, acting as a low-nutrient control. I pre-soaked commercial oat seeds for 24-36 hours to allow for imbibition and jump-start germination. I planted the seeds individually in each tube and watered them with distilled water as needed (2-5 x day). Each day I noted germination and/or measured the shoot height (cm). The pilot experiment measured: plant growth rate (daily growth in shoot length, over 8 days), final plant root and shoot length on the day of harvest, total plant dry mass (g), and the dry mass of the root separately. The dry mass was obtained by drying the entire seedling at 60C for about 48 hours, weighing the entire plant, severing the root, and then weighing the root separately. I calculated the Root Weight Fraction (RWF) as: $\text{root mass (g)}/\text{total plant mass (g)}$.

When the carrion insects had completed their development I collected two soil samples from each container (2 samples each from the 10 replicates for each of the 5 treatments) for a total of 100 planting tubes. I also filled a few extra tubes to use to monitor soil moisture levels. As before, I planted 1 pre-soaked oat seed into each tube and measured daily plant height for 11 days, collected the final height, the dry mass of the total plant, and the dry mass of the root. From this data rate of growth and RWF

were collected. As with the previous bioassay, the plants were watered daily with enough distilled water to prevent overwatering, and a paper towel was placed on the bottom of the tube to prevent any soil from being lost. Similar data to the first bioassay was collected as well, minus the final root length due to time constraints. Rate of growth, final shoot length, and dry plant weight were collected from the carrion insect bioassay.

Results

Pulled Data

Some data points were pulled due to seeds not sprouting. As this was most likely an issue with the seed rather than the soil these data points were removed as they could not be used to measure the soil quality of the soil they were planted in.

Pilot Experiment

The grass grown in the carrion-treated soil grew significantly faster (measured by cm/day) and therefore, was taller on the day of harvest (cm) than the grass grown in the control, as seen in figures 1A & 2A (ANOVAs 1B & 2B). The mean root weight fraction for the grass grown in the carrion-treated soil was significantly lower than the mean for the untreated soil, indicating less root growth (Figure 3A and ANOVA 3B).

Carrion Insects Treatments

The plant growth bioassay showed a significant effect of treatment on Root Weight Fraction (RWF). Figure 4A & 4B show the mean of each treatment. There were three levels of nutrients in the soil: the two carrion beetles species added the most nutrients to the soil, carrion alone and flies were next, and, the soil without carrion

added the least. There was no significant effect of treatment on the final shoot length (cm) or shoot growth rate (cm/day) (as seen in figures 5A & 6A).

Discussion

The pilot bioassay showed that RWF is a good measure of soil nutrients, as there was a significant difference between RWF in the soil that had the presence of carrion insects and the control soil (no carrion). This bioassay was then used in this study to detect how different insects play a role in carcass decomposition and return limiting nutrients to the soil. My hypothesis for the pilot experiment was supported, as the grass grown in the beetle soil had significantly lower RWF values compared to the control. The RWF shows how energy was spent by the plants, with a higher RWF value indicating growth was focused on the roots to find more nutrients in the soil before the plant can focus on shoot growth. Another way to determine this is by the root architecture, as mentioned earlier. Root architecture looks at the geometry of the roots, with more branching and longer roots signaling poor nutrients and the plant's focus to find more resources to sustain itself.

In the experiment on the effects of how different insects process carrion, I found differences among the treatments. I predicted that seedlings grown in soil that had *N. investigator* would have the lowest RWF value, and they did- but *T. lapponica*, the other carrion beetle had a similar impact on plant growth. While both beetles lay their young on carcasses for them to consume, they both have very distinct rearing styles that could have affected the nutrient levels of their soils. *T. lapponica* adult beetles and their larvae did not totally consume the carcass and tended to spread the carcass around,

while *N. investigator* adults buried it all in one spot and they and their larvae consumed most of it. Flies may have been less effective because the adults fed on carcasses and were then free to leave, and the maggots did not enter the soil very deeply, leaving the carcass on the surface. The carcass on the surface was consumed primarily microorganisms, although some small flies entered the container (resulting from a weasel opening the lid of the containers). The carcasses in this treatment dried, but did not get mixed into the soil.

In the experiment comparing different insects feeding on carrion, there was no significant difference between final shoot length and rate of growth, therefore these measures did not help indicate the nutrient quality of the soil. The strong effect seen in the pilot experiment was most likely due to the longer time (8 days) allowed for shoot growth. In the experiment, only 5 days were available after germination before the plants were harvested and dried. In the future, I would recommend a minimum of 8-10 days after germination to measure shoot and root growth. Time also should also be a heavier factor in future experiments, as the length of how long the carrion source is left on or in the soil also affect how nutritious the soil is, as each insect left behind parts of the carcass that were not eaten in the three weeks. Leaving the carcass in the container, whether it be a month or a year, can potentially change how much nutrients are put back in the soil.

Understanding how carrion insects break down carcasses and how this affects the soil where the carcass is located (creating islets of fertility) is important as many vertebrate carrion sources are small. With the decline of vertebrate scavengers due to human activity, insects may have a more important role. A better understanding of the

nutrients put back by insects can potentially restore impacted soils that are in desperate need of key nutrients. Instead of using synthetic fertilizer natural parts of the ecosystem (small vertebrate carrion and carrion insects) can be utilized and managed to help ecosystems return to a healthy state.

Figure 1A

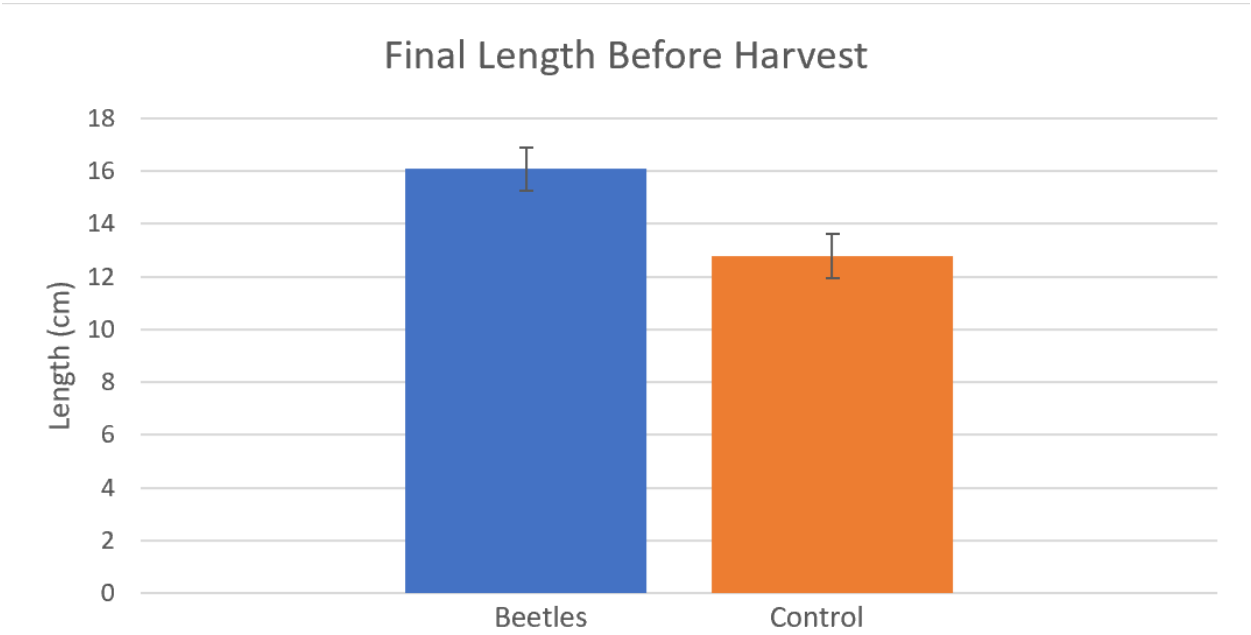


Figure 1B

Anova: Single Factor						
SUMMARY						
Groups	Count	Sum	Average	Variance		
Beetles	26	401.8	15.45385	25.19138		
Control	25	294	11.76	27.12833		
ANOVA						
Source of Variation	SS	df	MS	F	P-value	F crit
Between Groups	173.9005	1	173.9005	6.652634	0.012953	4.038393
Within Groups	1280.865	49	26.14009			
Total	1454.765	50				

Figure 2A

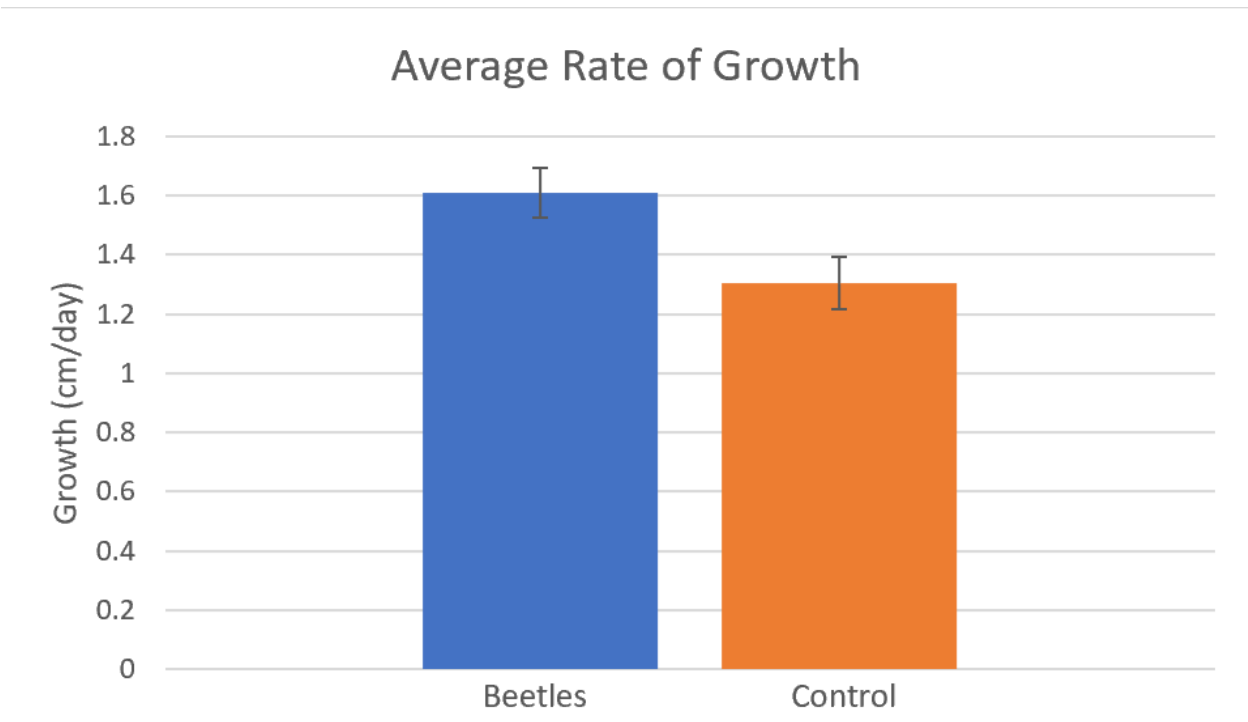


Figure 2B

Anova: Single Factor						
SUMMARY						
Groups	Count	Sum	Average	Variance		
Beetles	25	40.25476	1.61019	0.182479		
Control	23	29.98929	1.303882	0.18444		
ANOVA						
Source of Variation	SS	df	MS	F	P-value	F crit
Between Groups	1.123944	1	1.123944	6.127813	0.017051	4.051749
Within Groups	8.437173	46	0.183417			
Total	9.561117	47				

Figure 3A

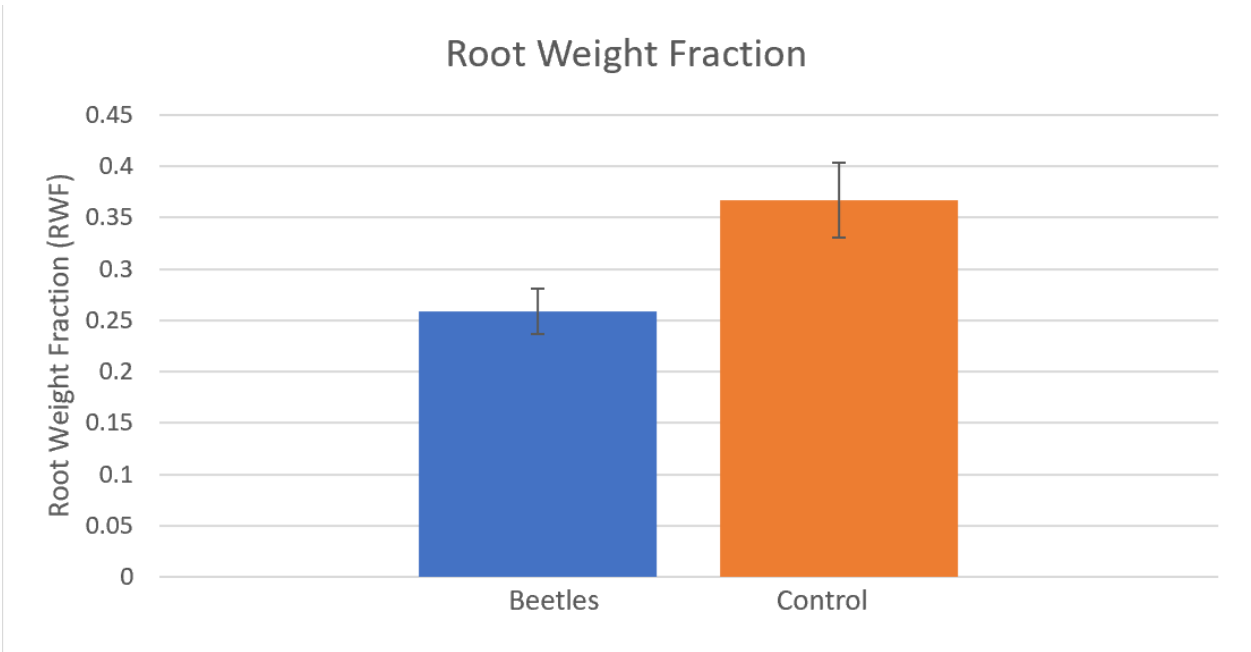


Figure 3B

Anova: Single Factor						
SUMMARY						
Groups	Count	Sum	Average	Variance		
Control	23	8.443473	0.367108	0.030105		
Beetles	24	6.209044	0.25871	0.01203		
ANOVA						
Source of Variation	SS	df	MS	F	P-value	F crit
Between Groups	0.138	1	0.138	6.613393	0.013496	4.056612
Within Groups	0.939002	45	0.020867			
Total	1.077002	46				

Figure 4A

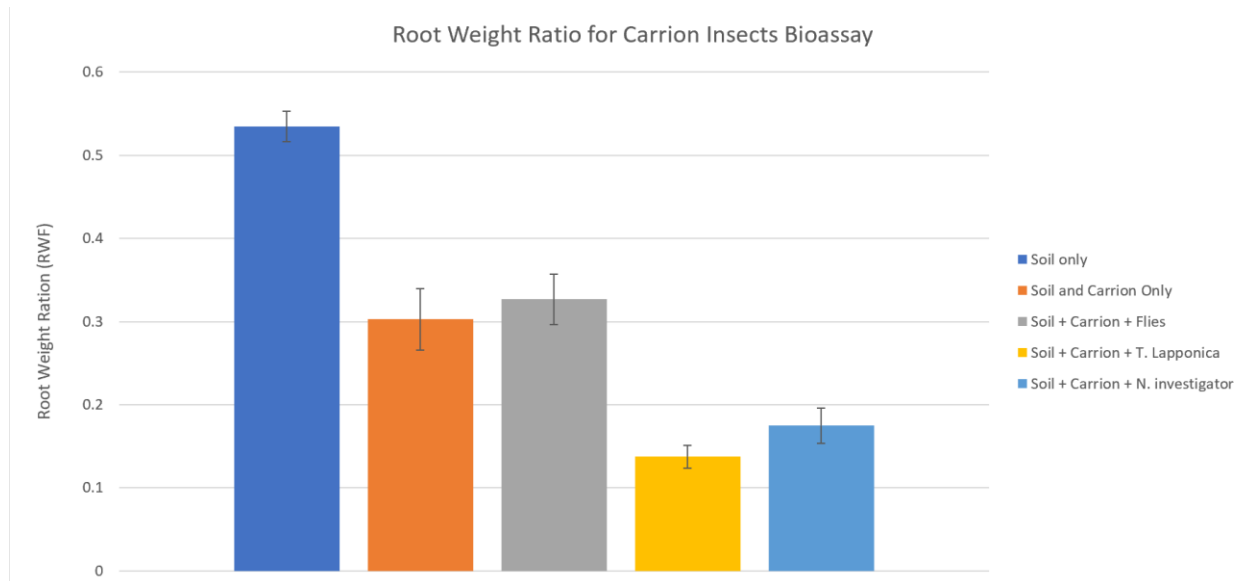
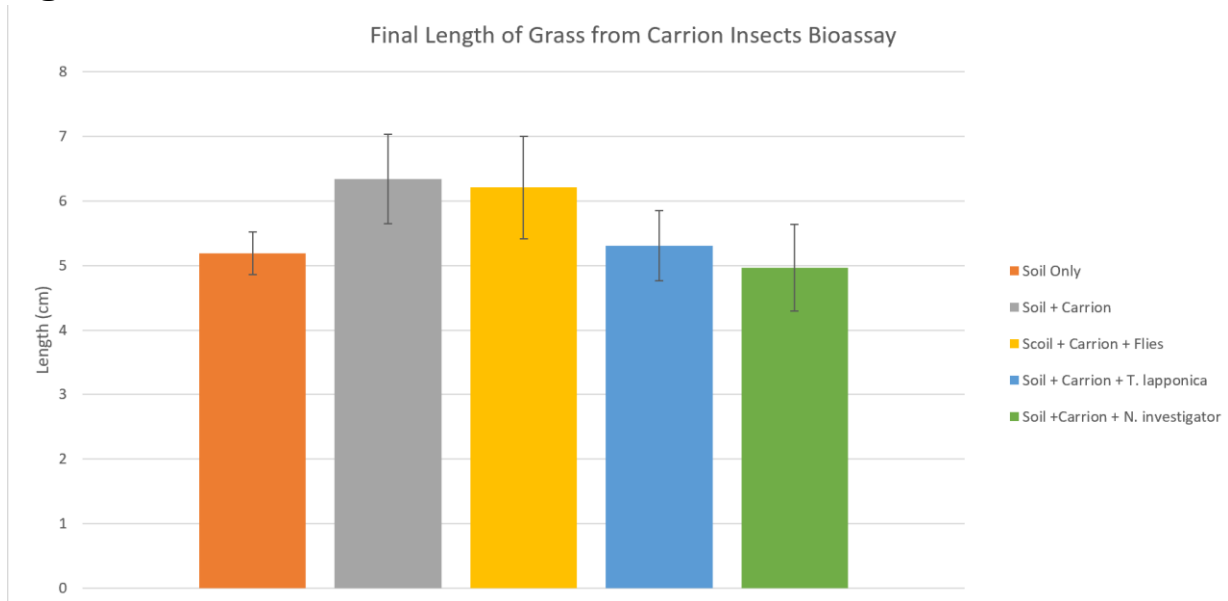


Figure 4B

Anova: Single Factor						
SUMMARY						
Groups	Count	Sum	Average	Variance		
Soil only	19	10.15579	0.534515	0.006276		
Soil and Carrion Only	14	4.235706	0.30255	0.019238		
Soil + Carrion + Flies	17	5.553494	0.326676	0.015801		
Soil + Carrion + T. Lapponica	12	1.649283	0.13744	0.00235		
Soil + Carrion + N. investigator	11	1.92075	0.174614	0.004852		
ANOVA						
Source of Variation	SS	df	MS	F	P-value	F crit
Between Groups	1.511456	4	0.377864	37.22608	1.81E-16	2.506621
Within Groups	0.690235	68	0.010151			
Total	2.201692	72				

Figure 5A**Figure 5B**

SUMMARY						
Groups	Count	Sum	Average	Variance		
Soil Only	10	51.9	5.19	1.087667		
Soil + Carrion	10	63.4	6.34	4.809333		
Scoil + Carrion + Flies	10	62.1	6.21	6.283778		
Soil + Carrion + <i>T. lapponica</i>	8	42.45	5.30625	2.351027		
Soil +Carrion + <i>N. investigator</i>	8	39.7	4.9625	3.605536		
ANOVA						
Source of Variation	SS	df	MS	F	P-value	F crit
Between Groups	14.7369	4	3.684225	0.998218	0.419526	2.599969
Within Groups	151.3229	41	3.690803			
Total	166.0598	45				

Figure 6A

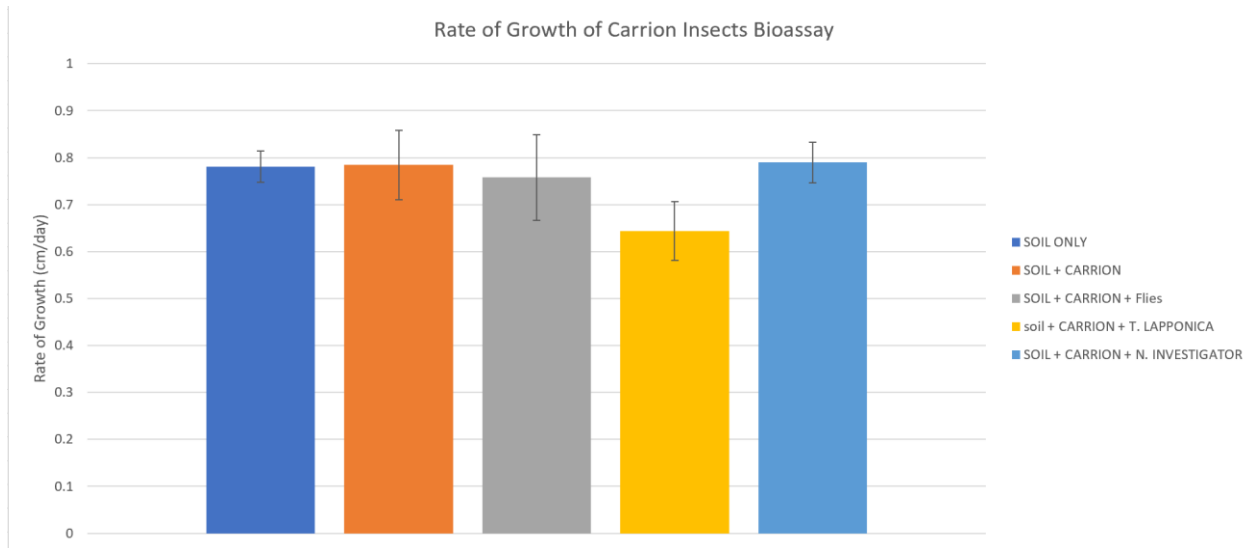


Figure 6B

Anova: Single Factor						
SUMMARY						
Groups	Count	Sum	Average	Variance		
SOIL ONLY	10	7.81232	0.781232	0.010975		
SOIL + CARRION	10	7.84195	0.784195	0.053687		
SOIL + CARRION + Flies	10	7.578912	0.757891	0.08263		
soil + CARRION + T. LAPPONICA	8	5.150159	0.64377	0.031189		
SOIL + CARRION + N. INVESTIGATOR	8	6.315728	0.789466	0.015069		
ANOVA						
Source of Variation	SS	df	MS	F	P-value	F crit
Between Groups	0.123948	4	0.030987	0.770242	0.550846	2.599969
Within Groups	1.649436	41	0.04023			
Total	1.773384	45				

Citations

Barton, P.S., (2016). The Role of Carrion in Ecosystems. In *Carrion Ecology, Evolution, and Their Application* (pp. 273-286). CRC Press.

Barton, P.S., Cunningham, S.A., Lindenmayer, D.B. and Manning, A.D., 2013. The role of carrion in maintaining biodiversity and ecological processes in terrestrial ecosystems. *Oecologia*, 171(4), pp.761-772.

Benbow, M.E., Tomberlin, J.K., and Tarone A.M. (2016). Introduction to Carrion Ecology, Evolution, and Their Applications. In M.E. Benbow, J.K. Tomberlin and A.M. Tarone *Carrion Ecology, Evolution, and Their Application* (pp. 3). CRC Press.

Crawley, M.J., editor, *Plant Ecology* 2nd edition (1997). Blackwell Science.

Danell, K., Berteaux, D. and Bråthen, K.A., 2002. Effect of muskox carcasses on nitrogen concentration in tundra vegetation. *The Arctic*, pp.389-392.

Denslow, J.S., Vitousek, P.M. and Schultz, J.C., 1987. Bioassays of nutrient limitation in a tropical rain forest soil. *Oecologia*, 74(3), pp.370-376.

Ilardi, M.O., Cotter, S.C., Hammer, E.C., Riddell, G. and Caruso, T., 2021. Scavenging beetles control The temporal response of soil communities to carrion decomposition. *Functional Ecology*, 35(9), pp.2033-2044.

Patterson, J. R., DeVault, T. L., & Beasley, J. C. (2022). Integrating terrestrial scavenging ecology into contemporary wildlife conservation and management. *Ecology and Evolution*, 12(7), e9122.

Santangeli, A., Girardello, M., Buechley, E., Botha, A., Minin, E.D. and Moilanen, A. (2019), Priority areas for conservation of Old World vultures. *Conservation Biology*, 33:1056-1065