**The impact of tornado and salvage-logging on ground beetle (Coleoptera: Carabidae) taxonomic and functional diversity**

**Research objectives**

The goal of this study is to understand the impacts of combined natural and anthropogenic forest disturbance on ground dwelling arthropods. In 2012, a tornado event knocked over canopy trees in a mature deciduous forest. After logging occurred on part of the tornado-affected area, we were curious how these combined disturbances would affect the invertebrates living on the forest floor.

**Part A:** We aim to measure the activity-abundance, species richness, and diversity indices of ground beetles (Carabidae) in each forest management treatment. We predicted that initially after the windthrow and salvage-logging (2015), that species richness would increase in the windthrow and salvage-logged areas, driven by the immigration of open-habitat species. Furthermore we predicted that in 2015 this immigration would be more pronounced in the salvage-logged treatment relative to the windthrow, due to the more complete canopy opening. However, because the salvage-logged treatment has less woody debris and altered understory compared to the unsalvaged windthrow, we predicted that species richness in the salvaged treatment would be lower than that of the windthrow by 2022.

In addition to documenting general trends in carabid abundance and alpha-diversity, we wanted to understand if the community composition of the carabids differs between each forest management treatment. One major question was what happens to the “forest specialist” ground beetles when a tornado and salvage-logging occur? Are these forest-adapted species completely absent from the windthrow and/or salvaged areas, and do they move back into the disturbed regions after a decade has passed? Conversely, do “open-habitat” ground beetles invade into windthrow and salvaged treatments? Furthermore, do these open-habitat beetles remain after a decade, or have they already left or perished due to the regrowth of trees?

**Part B:** We want to further understand the biology of the carabids found at Powdermill, and to do this we are attempting to use a ecomorphological trait approach. An ecomorphological trait is a morphological trait that tends to be found in species adapted to a certain environmental condition (Fountain-Jones, Baker, and Jordan 2015). The traits we are interested in are primarily regarding locomotion, sensory capabilities, and their interaction. Ground beetle adults can be categorized by their locomotion habits. There are wedge-pushers, climbers, and surface walkers. The wedge-pushers may be approximately cylindrical in body shape and have a longer hind trochanter. They are usually better at burrowing into soil to escape a predator or to seek out prey. They may spend a large amount of time pushing through soil or leaf litter. The climbers have proportionally longer legs and antennae and have a flatter body shape. The surface walkers may have a more hump-shaped abdomen, short hind trochanters, and a body which is not at all dorsoventrally flattened. Sensory capabilities of ground beetles include touch and gustation through antennae, as well as vision through eyes. Because the main method of locomotion influences what kind of sensory abilities are most important for beetles, we want to assess these too. For example, protruding eyes may be important for climbing beetles, but may cause problems for burrowing beetles which must push through substrate. Extremely long antennae could similarly cause problems for burrowing beetles. Of course, sensory abilities likely relate heavily to diel activity patterns, but almost all the carabids we collected in 2022 seem to be predominately nocturnal (exception: *Notiophilus aenius*) (Larochelle and Larivière 2003). Sensory abilities are also likely related to shade preferences (“forest specialist” vs. “open habitat”), which is a comparison we will be able to make.

Our goal is to compare the mean trait values found in each forest management treatment, with the idea of understanding how environmental differences between treatments would favor carabid species with certain traits, or certain syndromes of traits. We also want to determine whether the functional diversity of carabids differs between forest management treatments, which could indicate a greater variety of habitat niches.

**Methods**

Study site and tornado event

Research was conducted at Powdermill Nature Preserve in Rector, Westmoreland County, Pennsylvania. This is an area of temperate deciduous forest which serves as the field research station for the Carnegie Museum of Natural History. In June 2012, a tornado touched down, uprooting canopy trees in two areas, each about 120 × 480 m. The areas are located approximately at (40.14266, -79.27889) and (40.1447, -79.27491). These two areas are on the north- or northwest-facing slopes, which were dominated by maples (*Acer* spp.), tuliptree (*Liriodendron tulipifera*), black cherry (*Prunus serotina*), and other deciduous trees (Murphy et al. 2015), with an understory of predominately spicebush (*Lindera benzoin*) (Calinger et al. 2015). Before the tornado, this area had been forested since at least 1939 (Murphy et al. 2015).

Salvage logging

In summer through winter of 2013, half of each wind-disturbed area was salvage-logged using heavy machinery to remove the fallen and residual standing trees. In the southwest windthrow, the lower-elevation side of the windthrow was logged, while in the northeast windthrow, the higher-elevation side was logged. The operation of salvage logging on only half of each tornado-impacted area allowed us to compare arthropod communities between salvaged and un-salvaged forest.

Invertebrate sampling

In 2015 and 2022 (three and ten years post-tornado), ground-dwelling invertebrates were sampled as part of a larger research effort to understand the impacts of salvage-logging. Six transects were established across the disturbances, each with a site in windthrow (n=6), salvaged (n=6), and surrounding undisturbed forest (n=12). To capture invertebrates, one barrier pitfall trap was installed in each site and monitored every two weeks during the summer months. In 2015, traps were set up on May 27-28 and taken down on August 17 for a total of 6 collection intervals. In 2022, traps were set up on June 1-2 and taken down on September 6 for a total of 8 collection intervals. A pitfall trap consisted of two pairs of plastic cups (each pair having an inner 500 mL cup and an outer 1 L cup) which were placed into the ground so that the lip of the cup was flush with the ground surface. The two pairs of cups were placed 1 m from each other, and garden edging (Suncast® eco edge) was placed between them to create a barrier. Cups were filled 4 cm high with propylene glycol (recreational vehicle and marine antifreeze, Peak Company Old World Industries, Clear Lake, Texas) with a few drops of detergent added to prevent escapes. Masonite board (100 cm2) was placed at 3 cm above each cup to prevent overfilling due to rain. Steel landscaping cloth was secured over cups using 30 cm stakes in order to limit mammal disturbance. Traps were collected every 2 weeks by pouring the sample through a fine mesh strainer and placing the contents into 70% ethanol, before refilling the sample cups with propylene glycol for the following interval.

Annual temperature and rainfall

In 2015 in Donegal, PA (station USC00362183 ), winter temperatures (Jan-Feb) were about 20-40 F highs and 0-20 F lows (ranging down to -14 F in February), while summer temperatures were around 70-80 F highs and 50-70 F lows. At the beginning of the study period near May 27, cumulative precipitation (starting at January 1) was around 24 in., and heavy rainfall occurred in June but stopped by mid-July, so that by August 17 about 41 in. had accumulated.

In 2022 in Donegal, winter air temperatures (Jan-Feb) were about 30-40 F highs and 0-20 F lows (ranging down to -10 F in January), while summer temperatures were around 70-90 F highs and 50-60 F lows. When traps were set up on June 1-2, cumulative precipitation was about 25 in., and rainfall occurred steadily through the summer, so that about 36 in. had accumulated by mid-August. Thus, 2022 was slightly drier than 2015.

Environmental variables of the forest floor

Environmental variables of the forest floor relating to light availability, ground cover, vegetation height, and soil moisture were collected near each pitfall trap. Light availability was recorded using a spherical crown densiometer, which measures canopy openness. The densiometer was turned in each of the four cardinal directions and the number of squares with sky visible was recorded. Canopy openness was measured on June 9-10 and August 5, 2015, and on June 1-2, 2022. To record ground cover, we randomly selected two 1 m2 quadrats around the pitfall trap. Ground cover was estimated using the following categories: ground-level vegetation, leaf litter, bare ground, fine woody debris (<10 cm diameter at the large end), coarse woody debris (>= 10 cm diameter), and rocks. Ground cover estimates were collected monthly during the growing season. We additionally measured the average height of vegetation within the quadrats. Values from the two quadrats around each pitfall trap were averaged together. We measured soil moisture at three locations adjacent to each pitfall trap using a Dynamax Inc. (Houston, Texas) TH20 portable soil moisture meter with a Theta Probe ML2x sensor. Soil moisture measurements were taken biweekly when pitfall samples were collected. The three readings were averaged together for a single value at each plot-date combination.

Ground beetle identification

Beetles were counted and removed from the samples, and all ground beetles (Carabidae) were pinned or pointed. Ground beetles were identified to species using keys in (Lindroth 1961; Bousquet 2010; Freitag 1969; Bousquet and Messer 2010; Harden and Guarnieri 2017) and the nomenclature was verified using (Bousquet 2012).

Table 1. Ecomorphological traits measured in this study.

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| **Trait** | **Calculation** | **Connection to habitat** |
| Body length | **a** | Shorter body length was found for ground beetles caught in wind-disturbed forests, relative to undisturbed forests (Sklodowski and Garbalinska 2011). Body length is correlated with many other morphological traits (Barton et al. 2011). |
| Antenna length | **b/a** | Tactile hunter species, which rely on sense of touch more than vision, tend to have longer antennae (Bauer and Kredler 1993). Longer antenna length relative to body length was found for ground beetles caught under a tree, versus in the open (Barton et al. 2011). |
| Eye protrusion | **(c – d)/a** | A greater eye protrusion was found in a tree-climbing ground beetle, and it may allow partial overlap in the frontal visual field. However, greater eye protrusion might prevent a ground beetle from moving through thick vegetation or soil (Talarico et al. 2007). |
| Eye length | **e/a** | Diurnal ground beetle species and/or those adapted to open environments tend to rely on vision for predator avoidance or prey detection (Talarico et al. 2007). |
| Pronotum width | **f/a** | A proportionally wider pronotum can be found in robust-bodied beetles, which tend to be found within open habitats (Barton et al. 2011). |
| Abdomen width | **g/a** | Similar pattern to pronotum width (Barton et al. 2011). |
| Rear leg length | **h/a** | Open habitats seem to favor ground beetle species with shorter legs relative to body length (Barton et al. 2011). |
| Rear trochanter length | **i/a** | The rear trochanter connects to the femur of the rear leg. It is longer, on average, in species that push themselves through soil and underneath leaf litter. It is shorter in species that walk or run above the surface of the substrate (Talarico et al. 2007). |

Ground beetle activity-abundance

Because pitfall traps preferentially collect insects that are more active and mobile, the number of ground beetles caught in pitfalls is reported as activity-abundance rather than true abundance. Before analyzing activity-abundance, we first accounted for missing data. In 2022, mammals disturbed some of the pitfall traps, resulting in an occasional loss of trap catch. When the sample from one of the two sides of the barrier pitfall trap was lost, which occurred in 12 instances, we doubled the counts of species found at that plot during that interval. This resulted in an additional 13 beetles being added as a correction factor to the original count of 852 ground beetles in 2022 (3 beetles in forest, 8 in salvaged, 0 in windthrow). In 5 instances, which were all in the undisturbed forest plots, we lost both sides of the barrier pitfall sample. To correct for this, we standardized activity abundance. First, we summed the counts of ground beetles caught over the entire season. Then we divided the counts by the number of days each pitfall was operational (Sklodowski and Garbalinska 2011). This gave a measure of number of ground beetles caught per day at each plot.

We tested for differences in activity-abundance of ground beetles between windthrow, salvaged, and undisturbed forest using a linear mixed-effects model. Area (northeast area or southwest area) and transect nested within area were both included as random intercepts in the model to account for spatial structure. Residuals were tested for assumptions of normality and homogeneity of variances.

Taxonomic alpha-diversity measures

To verify if our sampling effort was sufficient to make estimates of species richness, we used species accumulation curves (Chao and Chiu 2016). We created species accumulation curves using the rarefaction method, which accumulates individuals rather than sites. This was implemented using the *specaccum* function in the R package ‘vegan’ (Oksanen, J. et al. 2024; R Core Team 2024).

To investigate the alpha-diversity at the plot level, we calculated measures of species richness, Shannon diversity, Simpson diversity, and Simpson evenness using the package “HillR” in R (Li 2018). Shannon diversity was calculated using Hill diversity with Hill number q=1. This form of Shannon diversity has a minimum of 0 and a maximum value of the species richness. Simpson diversity (Inverse Simpson Index) was calculated using the Hill diversity with Hill number q=2. Simpson evenness was calculated as the Inverse Simpson Index divided by the species richness. This metric measures the degree to which species have similar abundances.

To estimate the number of undetected species and thus estimate the true species richness of ground beetles, we used an asymptotic approach (Chao and Chiu 2016). We calculated the Chao1 estimator, which is a nonparametric estimator that gives a lower bound on the true species richness. This estimator incorporates the number of singletons and doubletons to estimate the number of undetected species and was implemented using the function “ChaoSpecies” using the R package “SpadeR” (Chao et al. 2016).

Taxonomic beta-diversity measures

Investigating beta-diversity answers the question: does the community composition of ground beetles differ between the wind-disturbed, salvaged, and undisturbed forest? Variation in community composition could be predominately *between* forest management treatments, or it could be predominately *within* each forest management treatment. We tested these possibilities using the Permutation-based Multivariate Analysis of Variance (PerMANOVA) and the Analysis of Multivariate Homogeneity of Group Dispersions methods, respectively. To implement these methods, we first calculated the distance in species-space between all pairwise combinations of our 24 plots. This was implemented using Bray-Curtis Dissimilarity with the *vegdist* function in the R package ‘vegan’ (Oksanen, J. et al. 2024). Because the inter-plot variability in total ground beetle catch was relatively low (σ / μ = 0.49), we did not perform any relativization prior to computing the distance matrix (McCune, Grace, and Urban 2002). PerMANOVA was conducted using the *adonis2* function in ‘vegan’ with 999 permutations. Beta-dispersion was implemented using the *betadisper* function in ‘vegan’. An Analysis of Variance (ANOVA) test was performed to test for differences in beta dispersion, and this was followed by pairwise tests using Tukey’s Honest significant difference test in the ‘stats’ package (R Core Team 2024).

The community composition of ground beetles was visualized using nonmetric multidimensional scaling (NMDS) using the ‘metaMDS’ function in ‘vegan’. We used a maximum of 500 random starts and a two-dimensional visualization. To verify a successful ordination, the Bray-Curtis dissimilarity between plots was graphed against the distance in ordination space using the *stressplot* function.

Idea: I’m also interested in which beetle species have positive spatial autocorrelation and which have negative spatial autocorrelation. For this I can use something called “Global Moran’s I”. Also, I think I should use presence/absence at each site rather than abundance.

Activity-abundance patterns of common species

For the 2022 collections, we individually analyzed any species of carabid where more than 30 individuals were collected. We tested for differences in activity-abundance between forest management treatments.

Morphological trait measurement

We selected a set of ecomorphological traits of beetles, meaning that these morphological traits have correlations with ecological variables (Fountain-Jones, Baker, and Jordan 2015). The traits we selected have been shown in previous studies to be related to environmental variables on the forest floor such as ground cover type and light availability (Table 1). For each species of ground beetle, we chose three male and three female individuals to measure trait values (Fountain-Jones, Baker, and Jordan 2015). These individuals were chosen in a way that attempted to encompass the intraspecific variation in body size. The individuals chosen for trait measurement were deposited as vouchers at the Ohio State Triplehorn Insect Collection (OSUC). Because the objective of this study was to capture interspecific variation in traits, we took the average of the six individuals to obtain mean trait values for each beetle species. Because we were interested in body proportions as opposed to absolute trait values, we analyzed traits after dividing by body length (Table 1). We used body length as a proxy for overall size of the beetle (Ribera et al. 2001).

Community-weighted mean traits

Functional alpha-diversity

Functional beta-diversity

**Results**