



# Mammal Mediated Dispersal of Mycorrhizal Fungi: Using Microscopy as a Method for Quantifying Diet and Fungal Richness

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## Background and Objectives

- Small mammal species have diverse diets composed of plants, insects, and fungi.



A. Fungal spores and tissue B. Insect piece C. Plant tissue

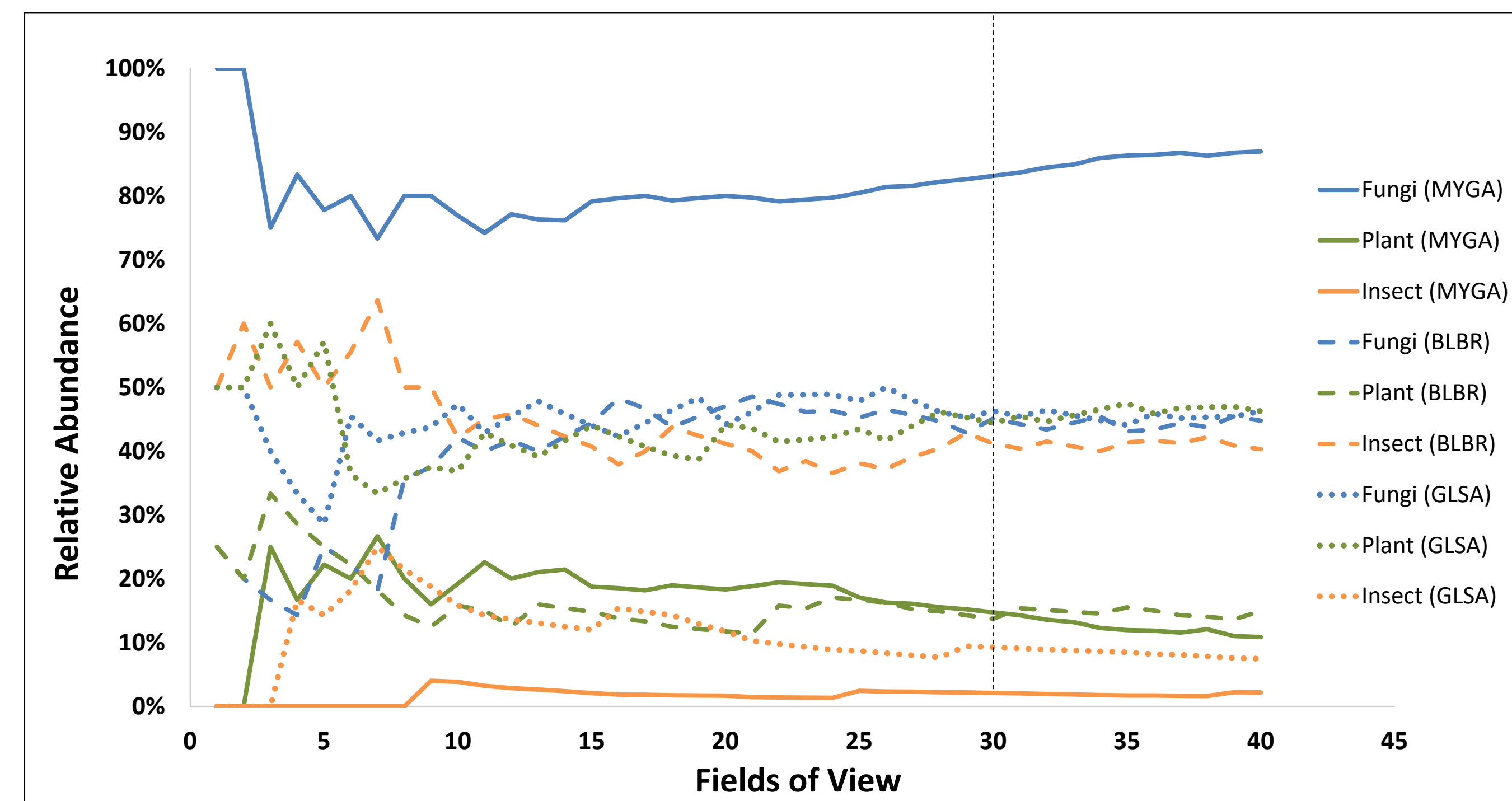
- Small mammals help initiate mycorrhizal fungal networks by consuming fungi and dispersing spores through their scat.
- Mycorrhizal fungal networks play important roles in ecosystem health including soil aggregation, carbon sequestration, and tree establishment.
- Mammal mediated spore dispersal can be a critical step in reestablishing fungal networks following forest stand disturbances such as volcanic eruptions, wildfire, and commercial logging.

## Objectives

- Develop effective protocol for quantifying diet composition using microscopy.
- Determine diet composition and fungal consumption for four small mammal species: Red-backed Vole (*Myodes gapperi*), Short-tailed Shrew (*Blarina brevicauda*), White-footed Mouse (*Peromyscus leucopus*), and Deer Mouse (*Peromyscus maniculatus*).

## Methods

- Small mammals were captured in the Chippewa National Forest in the fall of 2020. Fecal material was collected from the gastrointestinal tracts to be analyzed for diet quantification. We treated the samples with ethanol, potassium hydroxide, and Lactophenol Cotton Blue (LCPB). LCPB stains chitin, an organic polymer present in fungus and insect exoskeletons.
- We viewed the samples under a compound microscope at 100x magnification. Diet components were identified by obvious morphological features and the presence or absence of stain. Plotting fields of view versus relative abundance of diet components allowed us to determine that 30 fields of view were sufficient to ensure that diet composition was estimated accurately.

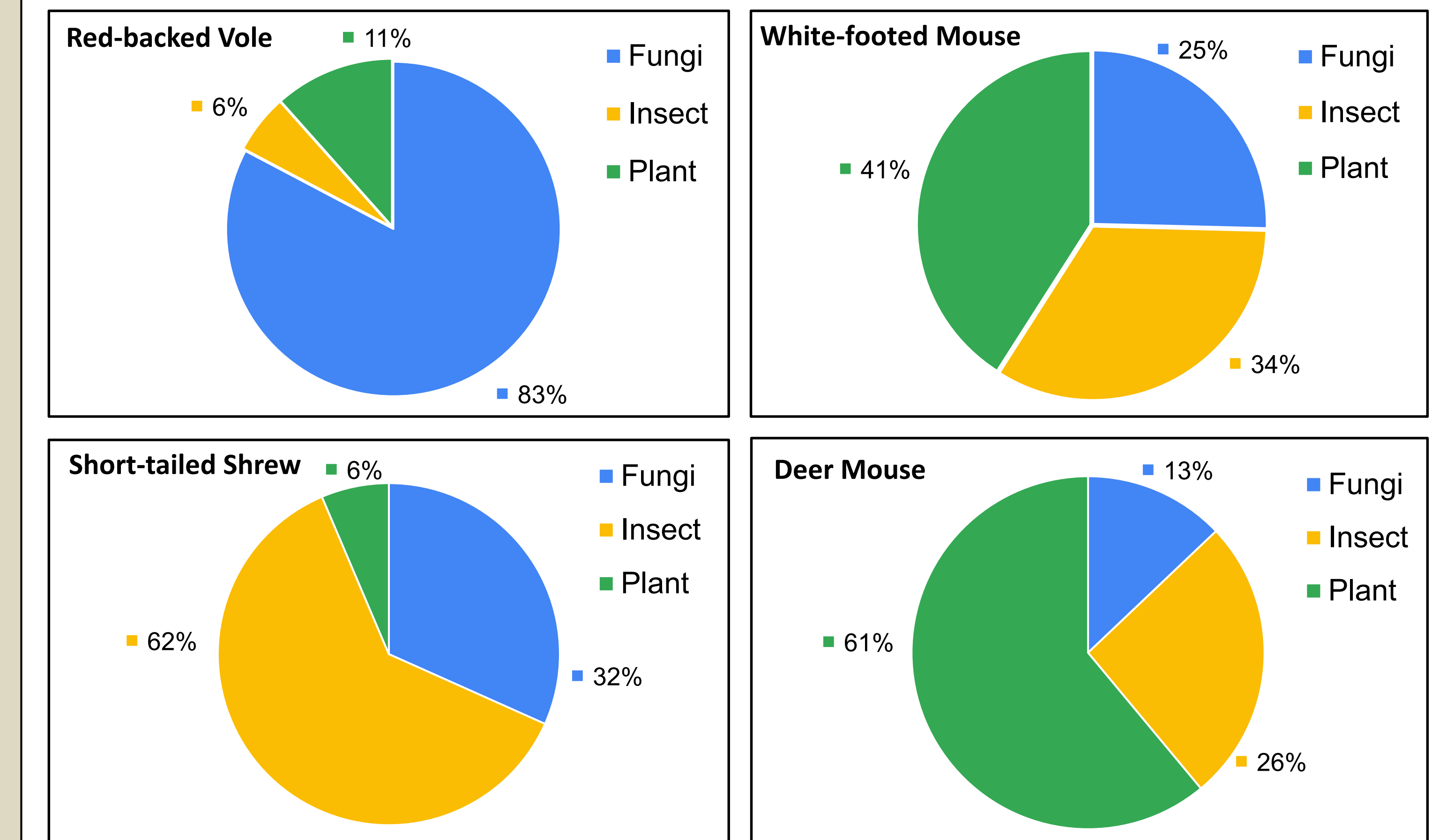


Graph made using test samples from *Myodes gapperi* (MYGA), *Blarina brevicauda* (BLBR), and *Glaucomys sabrinus* (GLSA)

- The total number of each diet component (fungi, insect, plant) were summed and divided by the total number of items to determine the relative proportion of each component in the bulk diet.

## Preliminary Results

Average diet composition by species:



- On average, red-backed voles have the highest fungal abundance in their diets, making up 83%
- Short-tailed shrews primarily eat insects, with fungi making up 32% of their diet.
- Both species of mice primarily eat plants and insects with fungi making up the smallest portion of their diets.

## Next Steps

- Complete diet quantification with a larger sample size. Compare diets based on study site and cover type (coniferous vs deciduous).
- Perform statistical analysis to determine significant differences in diet composition between species.
- Compare other methods of diet quantification, including genetic analysis of specimen tissues and stable isotope analysis.

## Acknowledgements and References

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- Elliott, T.F., et al. "Mammalian Mycophagy: A Global Review of Ecosystem Interactions between Mammals and Fungi." *Fungal Systematics and Evolution*, vol. 9, no. 1, 2022, pp. 99–159.
- Rillig, Matthias C., and Daniel L. Mummey. "Mycorrhizas and Soil Structure." *New Phytologist*, vol. 171, no. 1, 2006, pp. 41–53.
- Sharma, Mahaveer P., and Jeffrey S. Buyer. "Comparison of Biochemical and Microscopic Methods for Quantification of Arbuscular Mycorrhizal Fungi in Soil and Roots." *Applied Soil Ecology*, vol. 95, 2015, pp. 86–89
- Stephens, Ryan B., and Rebecca J. Rowe. "The Underappreciated Role of Rodent Generalists in Fungal Spore Dispersal Networks." *Ecology*, vol. 101, no. 4, 2020

## Preliminary Results

Species	Sample	Total Fungi	Total Insect	Total Plants	Total Items
Red-backed Vole	1	75	6	4	85
	2	61	3	8	72
	3	130	5	5	140
	4	139	5	6	150
	5	32	6	20	58
Short-tailed Shrew	1	4	41	0	45
	2	15	43	1	59
	3	28	33	8	69
	4	7	40	8	55
	5	34	12	2	48
White-footed Mouse	1	18	32	46	96
	2	3	26	24	53
	3	9	21	55	85
	4	133	9	10	152
	5	3	37	27	67
Deer Mouse	1	9	29	75	113
	2	25	21	47	93
	3	6	24	55	85
	4	22	45	74	141
	5	9	28	91	128