

Impact of Forest Fragmentation on Ectomycorrhizal Species Health Surrounding Blacksburg

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Introduction

Beneath almost all living plants today exists an intricate subterranean world fueling variegation, speciation, and communication (Brundrett, 2009). This ecosystem is referred to as the rhizosphere, made up of bacteria, nematodes, fungi, and plant parts all interacting in the soil (Pinton et al. 2007). A healthy rhizobiome has been found to foster codependent soil-microorganism relationships that both directly and indirectly impact each layer of atmospheric nutrient-cycling, especially among forested communities (Flores-Rentería et al., 2018). This idea couples well with the concept of forest fragmentation, the separation of interrelated species and disruption among functional structures supporting forest populations. Together both ideas are proving to be immensely impactful concepts in the analysis and maintenance of adaptive networks among plant species (Gorzalak et al., 2016).

The chemical networks facilitated through root interactions in the rhizosphere are fundamental in shaping the ecosystem above (Pinton et al. 2007). Forests are known to be areas where species effectively communicate threats amongst each other through specialized root networks (Bennett et al. 2017). Exactly just what level of connection mycelial networks provide between interrelated plant species is still a topic of discussion among biologists and one this study aims to narrow through the lense of forest communities. Well-established parent trees, characterized by high levels of related neighboring offspring, have been found to send nutrients to each other through their fungal networks (Bennett et al. 2017). These trees, often well-aged “mother” trees, will develop highly efficient root symbioses with specialist fungi to directly influence neighboring plants, both positively and negatively, depending on the levels of stressors to the plant’s environment (Bennett et al. 2017) .

Edge populations, areas with higher rates of stressors to the rhizobiome, are often found to result in higher competition rates among impacted plants. Communities of specific trees are often unable to effectively establish symbioses particular to their species then ultimately suffer invasion by less specialist species (Bainard et al. 2011). Healthy forest ecosystems are characterized by populations of both edge and interior community types, with interior plants serving vastly different structural roles in the community than interrupted populations of the same species (Laurance et al. 2007). In addition to competition among predators, highly stressed trees are expected to compete for surface area, for exposure to pollinators, and defend themselves against pathogens, which is far less effectively done in interrupted, stressed ecosystems (Hoffmeister et al. 2005) . Competition among fungal species in the rhizosphere becomes highly variable when considering that multiple stressors affect important symbiotic fungal capacity to establish (Bainard et al. 2011)

Over 86% of all flowering land plants form mycorrhizal relationships of some kind, utilizing the fungi’s decomposition activities to access nutrients in exchange for photosynthesized sugars through the roots (Brundrett, 2019). These relationships have recently

become the subject of much discussion to climatologists and forest ecologists as mycorrhizae have been found to interconnect related species and foster chemical communication (Bennett et al., 2017) across vast distances (Weile et al., 2016). As optimal soil conditions for these structures exist virtually nowhere but lab simulated conditions, most plants have also adapted methods of nutrient foraging in addition to chemical transmission through these symbiotic relationships in the earth (Weile et al., 2016). Mycorrhizal relationships are not always mutualistic, some with a higher tendency to become parasitic towards the plant host than others, dependent on which species are interacting in what environmental conditions.

Arbuscular mycorrhizal (AM) trees need enough space from genetically similar species to establish themselves, correlating well with their ability to vary root proliferation as a means of nutrient foraging (Weile et al., 2016). In contrast, ectomycorrhizal (EM) trees are more successful among related species, only thriving when established trees can support them through the rhizosphere (Pinton et al. 2007). Competition through edge effects on the forest communities in addition to competition with less particular fungal species for the same resources is causing gradual shifts in forest community types (Jo et al. 2018) and this study aims to correlate forest fragmentation to fungal abundance surrounding genetically impactful ectomycorrhizal trees.

It is hypothesized that transects measured within forest fragments with a 50% or higher wooded area will demonstrate greater amounts of visible ectomycorrhizal fungal growth than those with <50% surrounding forest community due to greater potential for communication among trees in unbroken forest. In addition to visible ectomycorrhizal fungal variation, other correlations indirectly affected by forest health will support the hypothesis that more visible growth correlates with less fragmentation.

Methods

Study Organisms:Fungi

As the organism of interest, ectomycorrhizal fungal specimens from Families: Amantiaceae, Gomphidaceae and Genus members *Russula*, *Lactarius*, and *Leucangium* are the main subject of this study. These fungi are the focal point and primary objective in deciding survey locations for when choosing transect trees. These target fungi have extended seasons where fruiting is known to be observed, frequently from mid-summer to late autumn (Miller et al. . This survey assess any late season fruiting bodies/signs of terrestrial fungal presence as indicative of established fungal networks, as species observed pair with particular host trees, only fruiting when 100% colonized and thriving in rhizosphere.

Study Organisms:Trees

The temperate forests of Appalachia studied are renowned for their native diversity, but as climatic shifts allow for ease of non-native invasion (Jo et al. 2018), a preference for Conifers and Oaks was placed in data collection. Large trees (Circumference >1.5 m) was a secondary distinguishing factor.

Field Procedures

Transects of 5-m-by-5-m taken with each large ectomycorrhizal tree located at the center of the measured area. Among the 7 sites surveyed, all surveys are taken within the same 48 hours to control for highly variable environmental conditions at the end of the fruiting season. Composite soil samples collected from the base of the tree, at the site of the largest fungal

colony or 1m from the base, and at the edge of the transect with a trowel. Soil samples kept 45 F until subsequent testing for soil pH, soil Phosphorus, and soil Nitrogen. Both host trees and fungi have preferential ranges of each, thus soil pH was measured with both a conductivity probe and tested in the lab with a Rapidsoil Test Kit, accounting for the root pH variable, which is comprised of field measurements, and soil pH, which tests the composite soil samples. An emphasis on minimal soil disturbance is taken into account during data collection. Circumference of study tree is observed and recorded using a meter tape, with species of uncertainty compared using the iNaturalist app.

7 forested sites were surveyed with varying percentages of surrounding forest within a 1 kilometer radius of each base tree surveyed. GIS tasks and values for altitude were calculated using Google Earth.

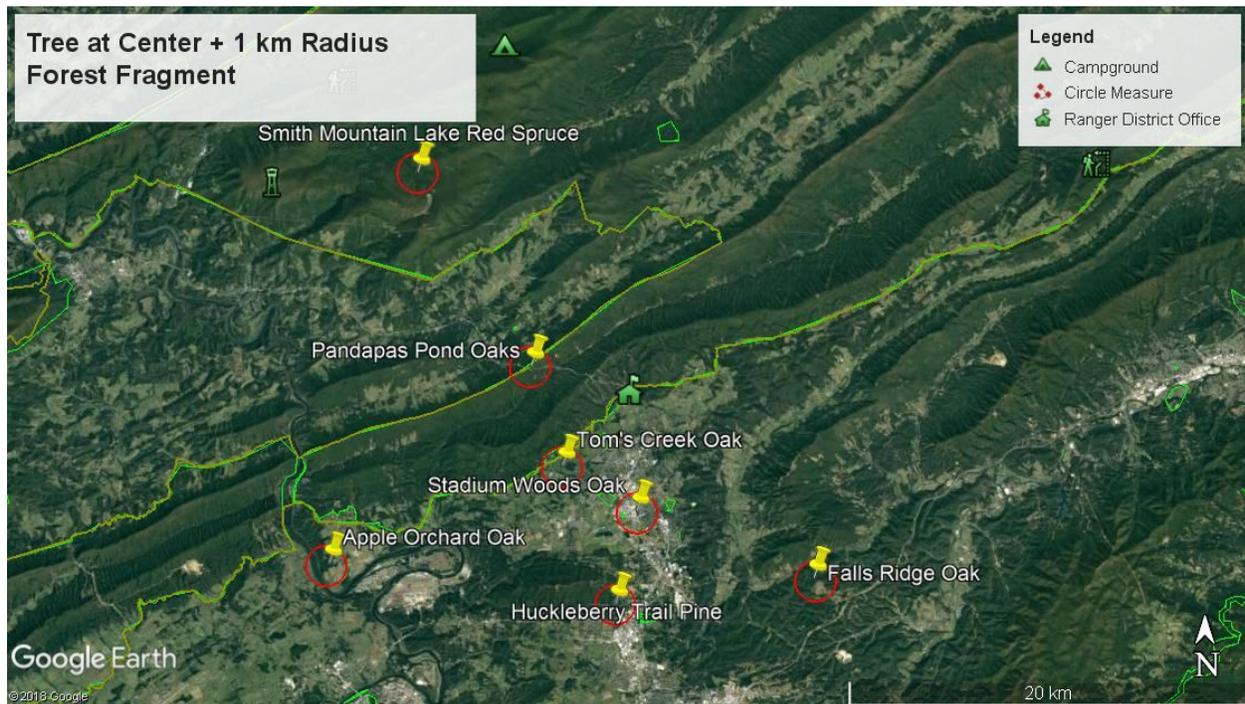


Figure 1: Map of sample sites with 1 km radius included.

Data Analysis

To test whether forest fragmentation impacted fungi abundance, we used a linear model with percent forest cover in a 1km radius as the predictor and total fungal abundance as the response. The significance of forest cover as a predictor was assessed using t-statistics. In addition, we used the same approach to test how forest fragmentation impacted other variables, including soil pH, root pH, variation in root pH, and variation in altitude. All statistical analyses were performed through R-studio.

Results

Number of Fungi:

The amount of forest cover surrounding a target tree had no effect on the number of fungi observed ($t = 0.570$, $p = 0.59$; $R^2 = -0.1267$; Fig. 2).

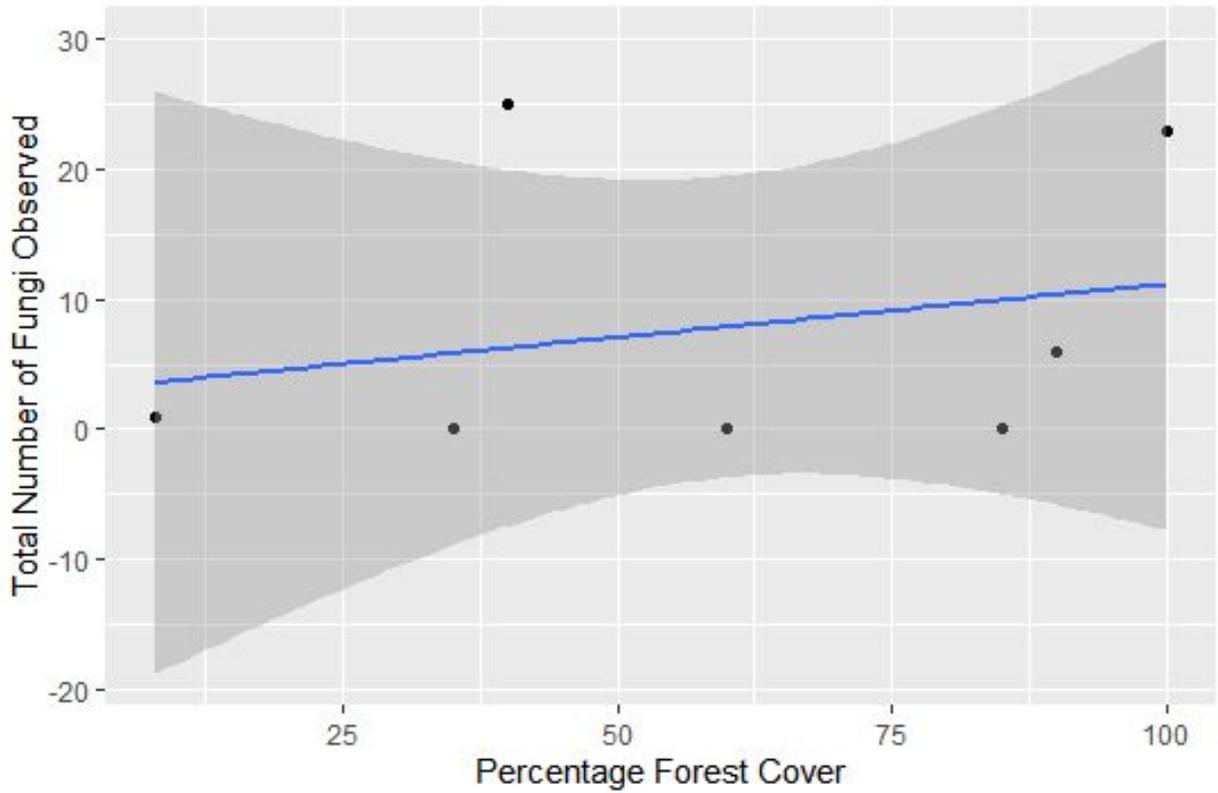


Figure 2: Percentage of forest surrounding transect area in relation to the number of visible fungi observed at the transect.

Average Root pH:

The amount of forest cover surrounding a target tree had no effect on the average pH of soil near the tree roots ($t = -0.475$, $p = 0.65$, $R^2 = -0.1482$; Fig. 3).

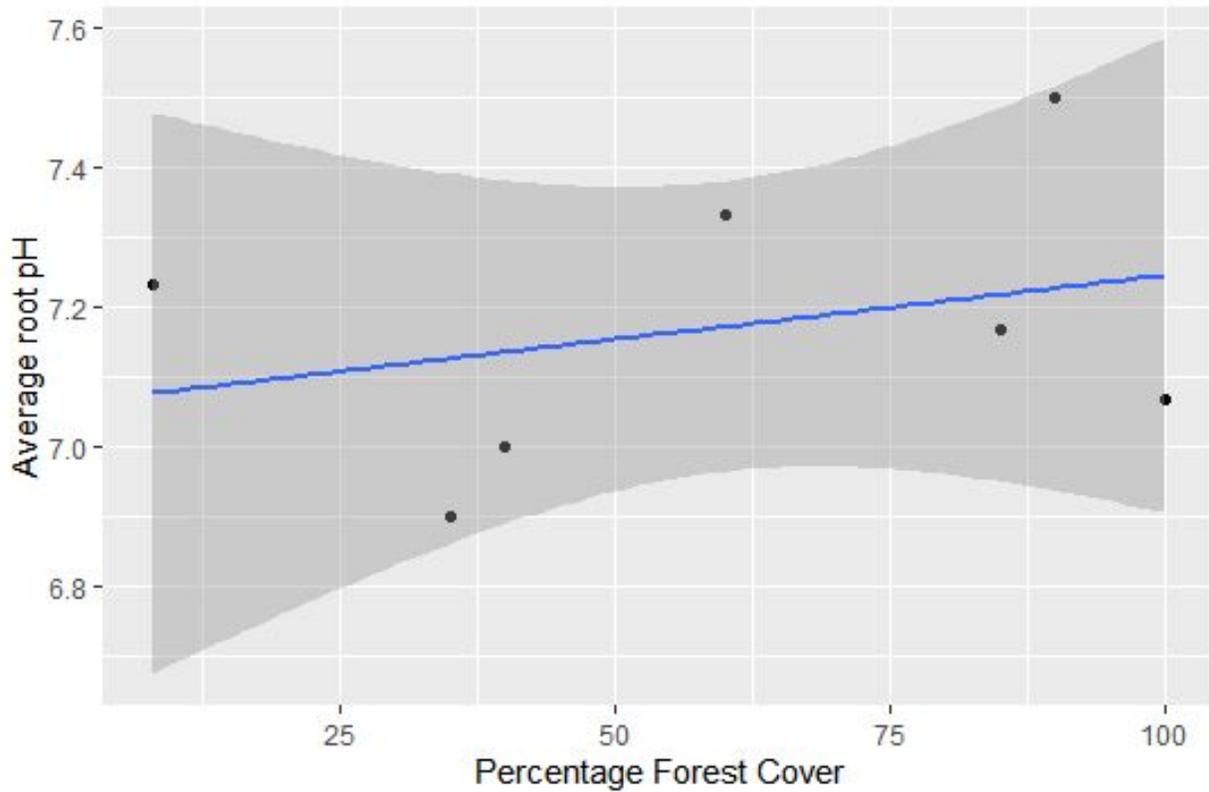


Figure 3: Average root pH compared with the percentage of forested area per transect

Soil pH:

The amount of forest cover surrounding a target tree had no effect on the pH of soil measured per fragment ($t = -1.067$, $p = 0.33$, $R^2 = 0.02252$; Fig. 4).

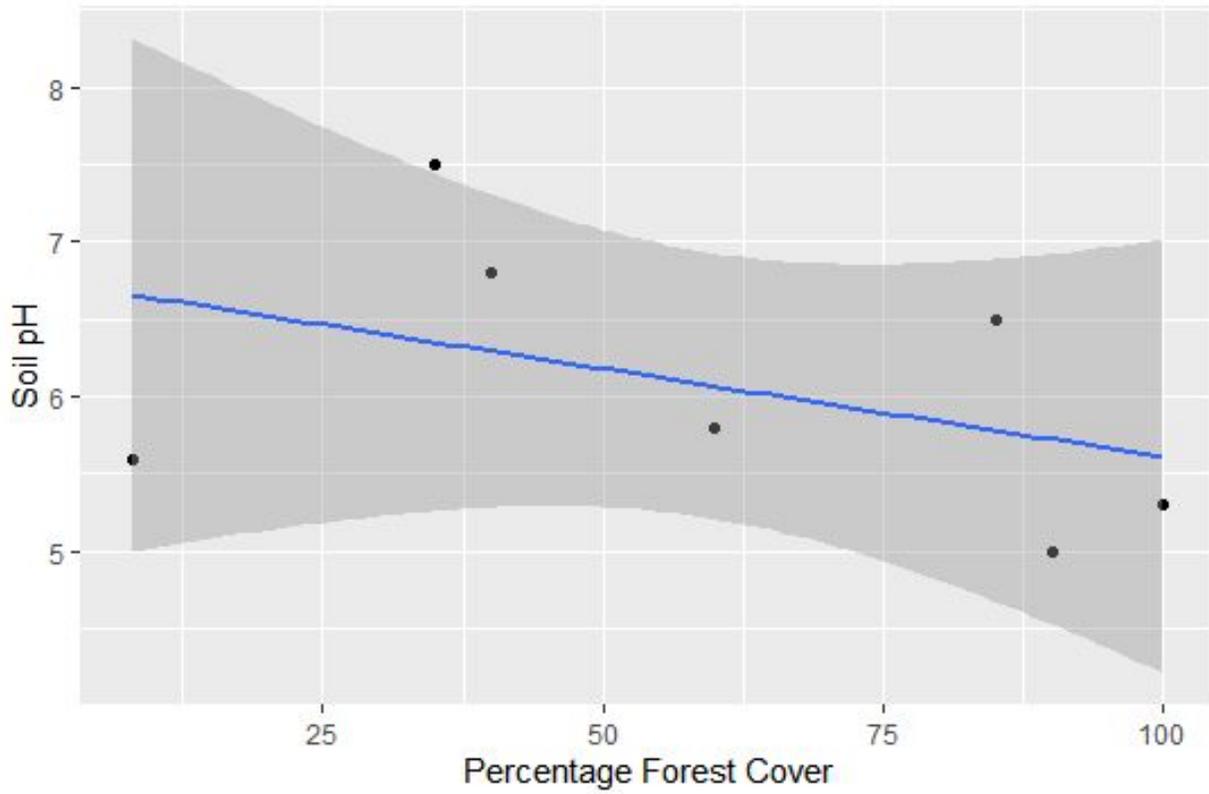


Figure 4: Percentage Forest cover in relation to soil pH of transect

Standard Deviation between Root pH:

The amount of forest cover surrounding a target tree had no observable effect on the standard deviation in pH values measured in soil from near the tree roots ($t = -1.101$, $p = 0.321$, $R^2 = 0.03417$; Fig. 5).

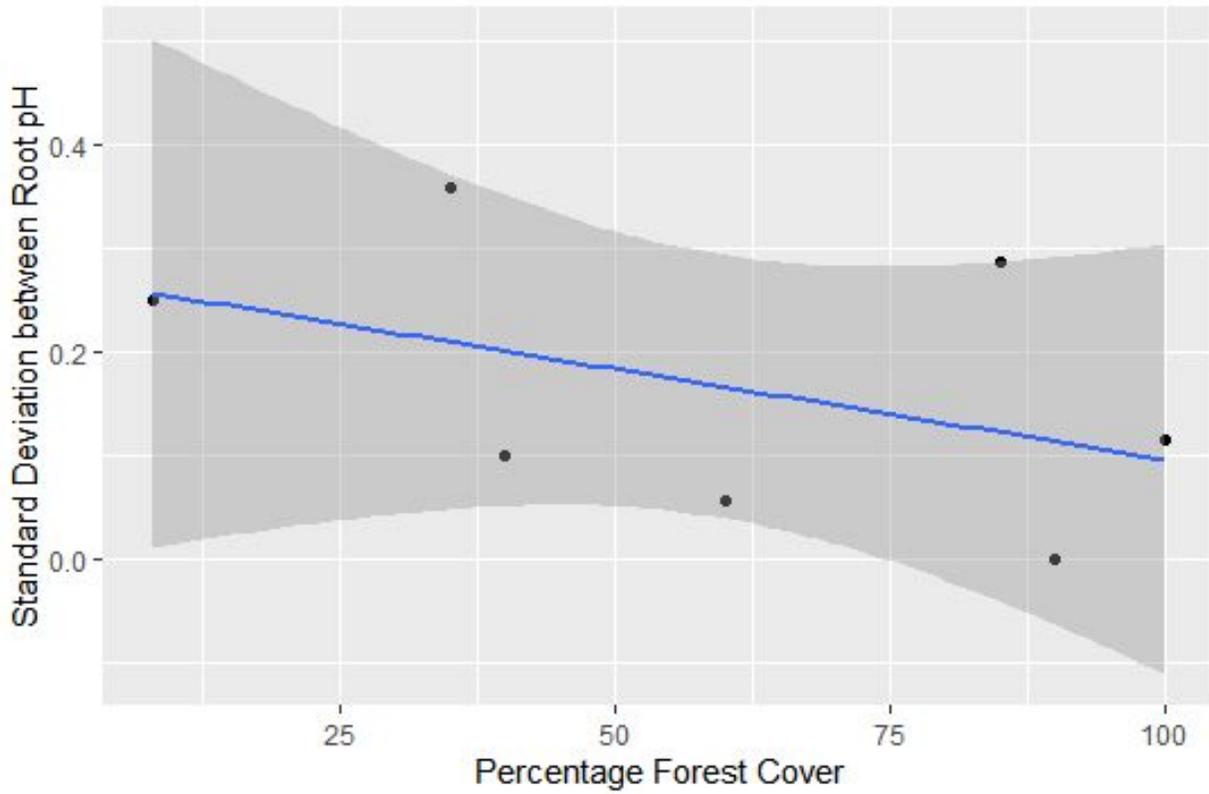


Figure 5: Standard deviation between root pH levels contrasted with percentage forest cover.

Variability in Altitude:

The amount of forest cover surrounding a target tree represents a strong predictor in the variation of altitude per fragment ($t = 8.207$, $p = 0.00043$, $R^2 = 0.917$; Fig. 6).

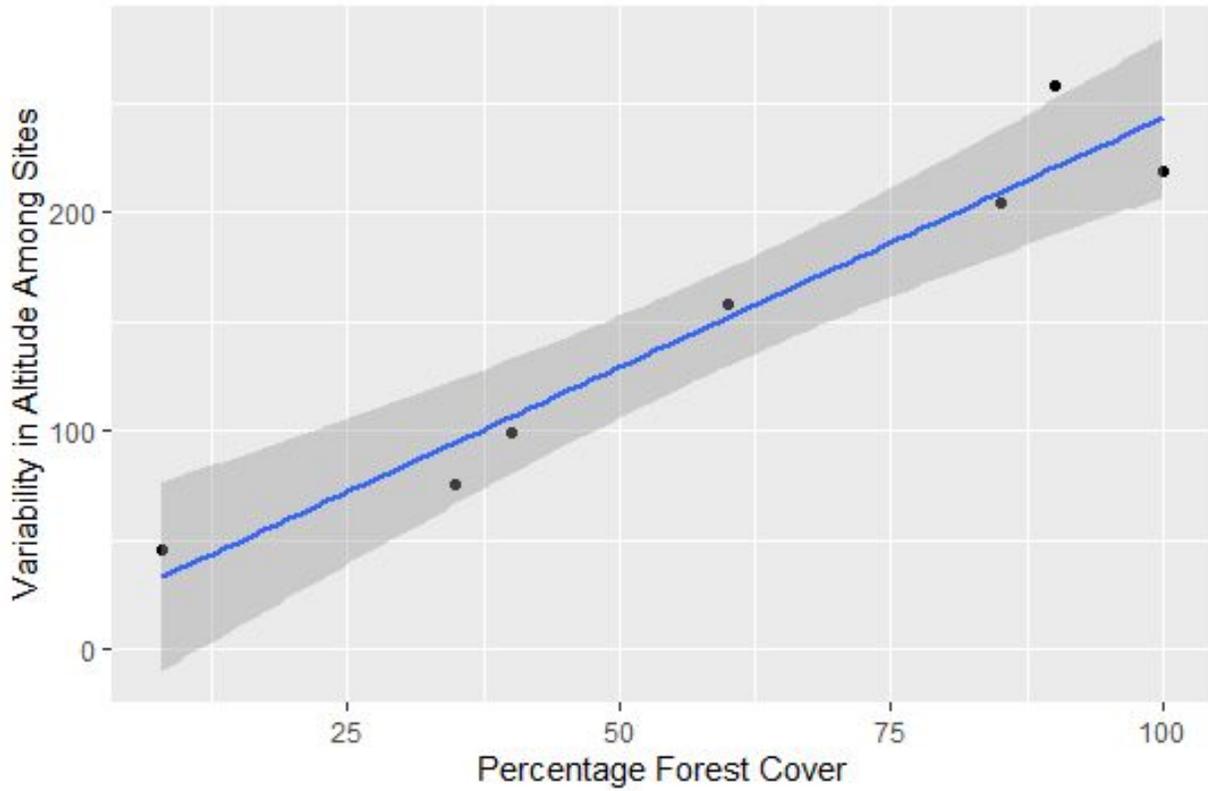


Figure 6: Variability in altitude (m) per forest fragment compared to the amount of forest per fragment

Discussion

This study should be viewed in its entirety as a pilot exploration into examining mycorrhizal relationships with as little damage to the rhizobiome as possible. The hypothesis that a definitive correlation between fragmentation and visible fungal growth is related to fragment size is rejected by this study. The sample size of trees to forest fragments was small in excess due to constraints sampling within similar enough weather conditions.

The immense number of alternative factors that impact forest health are vast, so tying visible fungal health and fragmentation in this study led to a rejection of the tested hypothesis.

The relatively large correlation between fragmentation and variation in altitude per site can easily be attributed to the frequent lack of development done on land that is more challenging to work. Land with intense variability in elevation often remains underdeveloped and unfragmented. The statistical implications of these correlative affects emphasize the importance of maintaining mountainous forested community health, but do not drastically inform the goals of this study.

Few studies have attempted to look at forest community interactions through visible fungal growth. The lack of correlational data between the majority of the variables studied suggests that further data collection to increase sampling sizes will enhance the impact each carries. Studies of these specimens over time could provide fascinating insight into overall productivity of the transects as seasonal effects take hold.

Studies into a codependent relationship between forest decline and mycorrhizal health are underway, with climatologists eager to further explore the potential significance of mycorrhizal presence among plant communities (Sapsford et al. 2017). Climactic controls of decomposition have been found as a leading driver in the global biogeography of forest-tree symbioses, intensely linked to atmospheric carbon and nitrogen sequestration (Steidinger et al. 2019). For humans to thrive with the plants on this planet in the future, an understanding of how ectomycorrhizal fungi are involved is proving essential.

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