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Fungal Associations in an Urban Forest

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Updated February 10, 2021

Fungal Associations in an Urban Forest

Jordan Leis

ABSTRACT

Fungal communities worldwide are scarcely understood in part due to their vast networks stretching underground or woven into the biomass of plant life. Even macrofungi which produce fruiting bodies colloquially called ‘mushrooms’ hold a transient lifestyle above soil. Urbanization has introduced another challenge towards studying fungi as highly disturbed soils and a lack of preferred host plant species can alter fungal community composition and prevalence. In the summer of 2020 the macrofungal community of Forest Park in Portland, Oregon was surveyed in addition to control sites in the Mount Hood National Forest. Fungi samples were identified by genus and when possible, species. Observations included nearest substrate, nearest vascular plant species, and GPS coordinates of the mushroom sample(s). Fungi samples were later identified as either saprophytic, mycorrhizal, or parasitic based on available literature. Our results of 648 confirmed (genus) fungi samples found that 90.8% were saprotrophs, 7.8% were ectomycorrhizal associated primarily with conifers, and that 1.4% were parasitic fungi. Shannon diversity and equitability scores showed that the national forest had a greater diversity of mycorrhizal fungi than the city and middle sections of Forest Park (P=0.0024), but that overall, there was little to no significant difference of the total fungal community composition across the urban forest compared to the national forest (P=0.1481). Further research is needed to identify additional members of the mycorrhizal community as to anticipate urbanization's effects, and how different types of mycorrhizal fungi improve urban forests and tree recruitment.

INTRODUCTION

Fungi are intricately entangled with all forms of life on land, and some are the original ecosystem engineers that allowed land plants to create favorable environments for animals. No ecosystem on land can survive without fungi (Eichhorn et al. 2003). There are over 200 known orders of fungi classified within 12 phyla and identified to six groups of major classification (Chang et al. 2021). The fungal kingdom is responsible for a host of ecological interactions including weathering rocks into soils, removal of pollutants and heavy metals from the environment, forming mutualistic and parasitic relationships with plants, influencing the atmosphere's composition, and as a source of nutrition and medicine (Lebel et al. 2010). One popular aspect of many fungi is the fruiting bodies some produce. These are usually above-ground reproductive structures that produce spores that ultimately grow into new mycelium - the body of the fungus. Fruiting bodies and mycelium are composed of interwoven mats of hyphae fungal cells (Eichhorn et al. 2003).

Studying fungi is challenging because most of their life cycle happens underground (Boddy et al. 2017). Mushrooms, the above-ground structures produced by mycelium, can be used as indicators of fungal diversity and activity below the soil. Mycelial networks connect specific fungi to plants or other mycelium, and as such they are important indicators of seral stages and ecosystem health in forests (Hood et al. 2005). Fungi that produce mushrooms fulfill a variety of ecological niches that increase stability of the environment. The vast majority of fungi are decomposers, although others form critical relationships with plant life and develop mutualistic associations between the kingdoms of Plantae and Fungi (Eichhorn et al. 2003).

Fungi and their Roles

All fungi are heterotrophic (organisms that consume other organisms to generate energy and vital nutrients) and exist as saprophytes, parasites, or mutualistic symbionts. Fungi are principal decomposers that are well adapted and prevalent in many environments (Chang et al. 2021 and Lebel et al. 2010). Unlike other eukaryotic organisms, fungi secrete digestive enzymes into the surrounding environment and absorb their food from the outside of their bodies. These hydrolytic enzymes catalyze large molecules, such as plant cellulose and lignin, into smaller subunits that are more easily absorbed by fungal hyphae. Because fungal cell walls are rigid, most absorption of nutrients occurs at the growing hyphal tips (Eichhorn et al. 2003). Fungi have the ability to digest almost all carbon-based substances and thus play a vital role in nutrient recycling by turning organic compounds back to biologically accessible compounds. Without decomposers, carbon, nitrogen, phosphorus, and other essential elements for life would be locked in organic forms in dead plant and animal matter. Decomposition of organic matter releases carbon dioxide into the atmosphere and returns vital nutrients such as nitrogen into the soil. This process recycles nutrients that can be reused by plants and animals. Fungi also can digest human products such as clothing, wax, jet fuel, wire insulation, and paint (Cain et al. 2014).

Parasitism is a not uncommon fungal method of obtaining nutrients. It often results in the decline or death of the host be it plant, animal, or other fungi (Cain et al. 2014). Fungal parasites absorb their nutrients from the cells of living hosts. Thirty percent of known fungi are parasitic heterotrophs or pathogens. Some fungal parasites are so specialized they require two or more host species to complete their life cycles. Fungal parasites can decimate tree populations, change the look and taste of macrofungi, or can go from being parasitic to saprotrophic once the host plant dies. Parasitic fungi, like saprotrophic and mycorrhizal ones, are key in the breakdown of organic matter and help to increase biological diversity in natural systems (Boddy et al. 2017).

Mycorrhiza is the single most important mutualistic symbiosis in the plant kingdom. This relationship occurs in most vascular plants with only flowering plants (Angiosperms) from the mustard family (*Brassicaceae*) and the sedge family (*Cyperaceae*) not forming mycorrhizal relationships with fungi (Eichhorn et al. 2003). Little is known about specific interactions between a host plant and its mycorrhizal partner(s). Conceptually it is understood that mycorrhizal fungi increase the host plant's root surface area, increasing the plant's uptake of water and essential nutrients from the soil. Studies have shown forest tree seedlings grown in sterile soil and then transplanted into grassland soil usually die of malnutrition because they are unlikely to form mycorrhiza. Mycorrhizal fungi also provide protection from pathogens and nematodes. In return the host plant provides plant carbohydrates and secondary metabolites to the fungi that they cannot produce on their own (Hood et al. 2005).

The special association between plants and fungi reduces the negative effects the host plant experiences during heat stress, times of drought, and pathogen attacks. Mycorrhizal fungi absorb nutrients from their host plant and in return the plant receives water and nutrients like nitrogen and phosphorus (Ammirati and Trudell 2009). Not all mycorrhizal associations are truly mutualistic with the plant and fungi receiving more or less equal amounts of nutrients or carbon, this is best described as biological market theory. Some plant roots preferentially supply more carbon to mycelium than the phosphorus they obtain. Another study showed fungi hoarding phosphorus in a flexible behavior that could rapidly change without an obvious reason (Bücking et al. 2014). Some mutualistic fungi are endophytes, ascomycetes that live within their plant hosts without causing harm. These fungal endophytes produce toxins that repel predators when they prey on the host plant's tissues and can help host plants in times of drought or in heavy metal poisoning (Cain et al. 2014).

History of Fungal and Plant Associations

Our ecological world is shaped by fungi; fungi were the first builders of their own ecosystems and paved the way for life to evolve onto land. Although fungi in the phylum Chytridiomycota (water molds) are considered true fungi, the first fungi to transition to land forfeited their chytrid flagellum and replaced it with branched-septate hyphae; these belong to the phylum Mucoromycotina (Cain et al. 2014; AFTOL 2021). Little is known about the earliest terrestrial fungi; though they most likely consisted of saprophytes (organisms that obtain energy from dead or decaying matter) and parasites. The early fungal colonizers weathered bedrock using strong hydrolytic enzymes they produced during metabolic digestion. Early plants such as liverworts, hornworts, and mosses colonized the emergent rocky soils, and the early plants and fungi interconnected in a symbiotic relationship. Those early vascular plants lacked root systems that would have increased their below-ground surface area enough to obtain sufficient mineral nutrients or water. Fungi, which lack chloroplasts, could not produce their own sugars, resulting in the benefits of these early associations (Eichhorn et al. 2003). Early non-vascular plants formed the first arbuscular mycorrhizal fungal (AMF) partnership some 400 million years ago in the early Devonian based on fossil evidence (Chang et al. 2021). Malloch and Pirozynski (1975) suggest the evolution of mycorrhizal fungi was a critical step for land plants as early soils were underdeveloped. Mycorrhizal connections would have facilitated the uptake of phosphorus and other vital nutrients for their plant hosts in exchange for plant carbohydrates (Cain et al. 2014).

It is believed that early plants and fungal life colonized land around the same time, roughly 470 million years ago during the Paleozoic era. Although, there is some evidence that single celled fungi may have made the transition before multicellular plants and fungi. Fungi present in the soil formed mutually beneficial relationships with nonvascular seedless land plants that increased the photosynthetic rate, carbon uptake, and growth of those plants. Fungi for millions of years served as root systems for plants before they evolved their own. The subphylum Glomeromycota developed as a monophyletic group that diverged from what was once the Zygomycota phylum and is a sister clade to the Dikarya (mushroom producing fungi). These Glomeromycota mycorrhizal fungi specifically form AMF associations with plant rhizoids and roots. The relationship also independently evolved later in Dikarya, in phylums Ascomycota (subphylum Pezizomycotina), Basidiomycota (subphylum Agaricomycotina), and

Mucoromycotina as ectomycorrhizal (ECM) associations (Cain et al. 2014; AFTOL 2021). ECM associations roughly 200 million years ago during the Jurassic after the origin of Pinaceae convergently evolved in at least 80 different lineages of Mucoromycotina, Ascomycota, and Basidiomycota. (Chang et al. 2021).

The early mycorrhizal associations persisted and are found now in almost every vascular land plant. Unlike the fungi in Glomeromycota where every member form mycorrhizal connection, not every species in Dikarya forms mycorrhizal relationships with plants. These fungi produce fruiting bodies colloquially called mushrooms. The mushroom structure forms from sexual reproduction and generates dikaryotic spores that undergo karyogamy to become diploid spores (Cain et al. 2014).

Mycorrhizal Associations

There are two types of mycorrhizal relationships: AMF that penetrate the root cells of the host plant and ECM that encircle the host plant's root cells. AMF generally are not host specific and all are within the Glomeromycota clade, which comprises 150 known species. Of all 65,000 fungal species in Glomeromycota, Ascomycota, and Basidiomycota only 10% are mycorrhizal (Lewis 2016). ECM fungi are characteristic of temperate trees and shrubs, mainly in the beech family (Fragaceae), the pine family (Pinaceae), the cottonwood family (Saliaceae), and the birch family (Betulaceae). ECM relationships also are found in tropical trees though fewer species of fungi are involved. Plants in timberline regions also have ECM. Species of pine (*Pinus)*, eucalyptus (*Eucalyptus*), and southern beech trees (*Nothofagus*) associated with ECM have increased resilience to cold/dry conditions that limit tree growth in those areas (Cain et al. 2014). These ECM fungi are most numerous in Basidiomycota and produce basidiocarp mushrooms, though some are ascomycetes including truffles *(Tuber*) and morels (*Morchella*) that produce ascocarp mushrooms. Approximately 5,000 macrofungi form ECM associations with some being generalists and others being highly specific with their host plant (Lebel et al. 2010).

Two specialized mycorrhizal associations are found in the heather family (Ericaceae) and orchid family (Orchidaceae), with the earliest fossil association with the heather family in the Cretacious. The Ericaceae, a large angiosperm family, includes huckleberry (*Vaccinium sp.*), cranberry (*Vaccinium sp*), rhododendron (*Rhododendron sp.*), and the inflorescent monotropes (*Monotropa sp.*) (Chang et al. 2021 and Eichhorn et al. 2003). ECM in the heather family releases hydrolytic enzymes into the soil that break down compounds and make elements like nitrogen readily available for their host plant. This adaptation allows plants in the Ericaceae family to be well adapted to the infertile and acidic soils where they are mostly found. Orchids in Orchidacea have a special relationship with mycorrhizal fungi. All orchids start out as heterotrophs and depend upon their fungal hosts as a source of carbon and nutrients early in their life cycles due to microscopic seeds that lack endosperm, the part of a seed that feeds the seedling during germination. Depending on the orchid species, this relationship sometimes reverses with the orchid becoming the source of carbon for its fungal association(s) (Argus et al. 2020).

Molecular studies have shown the formation of a symbiotic mycorrhizal relationship is dependent on certain genes being expressed by both the fungal and plant partners. Three specific plant genes, called "*sym*" genes, have been studied in angiosperms. They need to be expressed for mycorrhiza to become associated with the host plant. These genes have been found in major plant lineages beginning with liverworts (Cain et al. 2014). The genes have been conserved since the origin of land plants. One study transferred liverwort *sym* genes to a flowering plant mutant that lacked mycorrhizal connections; afterwards the mutant recovered its ability to form mycorrhizal associations when the *sym* genes were reintegrated (Bae and Mohanta 2015).

Mycoheterotrophs plants are either partially or completely achlorophyllous and thus don't fully rely on photosynthesis for their anabolic pathway. These plants can adapt a parasitic association with their symbiotic fungus or choose a more mutualistic approach where the plant relies on its fungal host for nutrients early in the plant's life cycle; and then become a source of carbon for the fungus after establishment (Bidartondo 2005). Mycoheterotrophic plants are very host-specific to AMF and ECM in addition to some saprophytic fungi. These plants are highly diverse and have co-evolved with fungi and can be found in the Orchidaceae and Monotropoideae families. Mycorrhizal networks harbored by photosynthetic plants can be tapped into by mycoheterotrophic plants for nutrients and energy requirements and to assist in establishing saprophytic fungi into the greater mycorrhizal symbiosis (Bidartondo et al. 2009). Little is known about the costs of establishing this kind of interaction on the fungus; or what kind of genetic or biomolecular factor allows mycoheterotrophy recognize and establish themselves with their hosts (Smith and Read 2010).

Urbanization and Fungi

In urban systems plants and soils enhance environmental quality and provide key ecosystem services, and ECM fungi improve plant and soil qualities (Francini et al. 2017). The global change brought on by human development has radically altered Earth's ecosystems and no such factor in global change is as well documented as fossil fuel combustion and deforestation’s contribution to atmospheric carbon dioxide levels. (Allen et al. 2001). The biological and ecological habitat requirements for fungi can be highly specialized for associations with host plants and plant litter for saprobes. Though due to a lack of knowledge and historical and current data on fungi in cities it is not well understood how fungi are adapting to urbanization. Though like many other human induced disturbances on the natural environment it is relevant to assume urbanisation is negatively affecting local fungal communities (Lebel et al. 2010). It is becoming more prevalent to study fungi in urban environments not only to satisfy human curiosity, but because it has become evident that the kingdom Fungi are diverse in the ecological roles they play, and how these interactions affect humans and our personalized environments. It hasn’t been till recent that technological advances in PCR and DNA-molecule detection procedures have allowed more comprehensive sampling of soil and air fungi present in samples, as well as analysis of plant organs for fungal traces (Garbelotto 2004 and Abdi et al. 2020).

Research on saprophytic fungi and the connection to soil nitrogen is uncommon, but it’s assumed that eutrophication of urban soils would influence saprophytic fungal communities (Lebel et al. 2010). Saprobic fungi are more likely to excel in urban environments due to their variability and favoritism for disturbed environments, excess inputs of nutrients, and human-facilitated dispersal (Cilliers et al. 2017). Many soil fungi are passed along in the commercial trade of plants and fertilizers; like the cosmopolitan death cap mushroom (*Amanita phalloides*) native to Europe. It was introduced to North America accidently and most likely from the transport of quark oak (*Quercus suber*) in Europe for commercial growth in the state of California as late as 1938 (Pringle and Vellinga 2006).

In a dataset compiled by Cillers et al. (2017) soil fungi tested in the USA, Finland, Hungary, and South Africa indicated diversity loss for this kingdom as evident by the urban filler effect and possible endemism. These effects are postulated to be caused by a variety of urban phenomena such as change in soil pH, nutrient composition, and a decline of host plant communities. ECM communities in northern cities situated in boreal and temperate biomes had a decline in Basidiomycota species abundance and richness, while in South Africa managed turf sites had higher ECM diversity. Urbanization reduces selectivity of host plants promoting ECM and AMF generalists, while reducing the competitive fitness of specialists that has resulted in non-nested mycorrhizal communities (Chen et al. 2021).

Our Study

In summer (May - August) of 2020 data regarding the presence of macrofungi in the Ascomycota and Basidiomycota were collected in and near Forest Park in Portland, Oregon and at control sites in the Mount Hood National Forest. In conjunction with counting fruiting body individuals and clusters, data was collected regarding the nearest vascular tree, shrub, and herbaceous plant species, the substrate, GPS coordinates, and whether the fungus was listed as saprotrophic, parasitic, or mycorrhizal. A citizen science platform, iNaturalist, was also developed to collect data year-round to compare trends in fungal composition in the summer months to those over the course of the full year. Urban forest ecosystems tend to have fewer species and lower diversity, and studies show urbanization affects the natural succession of forests (Broshot 2011). There are limited studies on how urbanization affects fungi (Lebel et al. 2010).

This study is part of a larger, long-term research project that is examining tree recruitment and mortality in an urban forest, Forest Park. Natural systems in urban environments are characterized as highly disturbed and fragmented ecosystems and thus are susceptible to invasion and proliferation of exotic species, while native populations are reduced (Bainard et al. 2010). Significantly fewer live trees and significantly more dead trees were found in 2003 than in 2003 in Forest Park. Recruitment of seedlings and saplings in 2003 was significantly lower in all areas of the park. In 2013 three control sites in the Mount Hood National Forest were added to the study, which has significantly more seedlings and saplings than any area near Portland (Broshot, personal communication) The loss of trees in the urban forest was not offset by increased diameter of live trees, suggesting natural self-thinning was not the cause of high tree mortality. It was postulated that climate change or pollution could be a cause of high mortality and low recruitment of trees in Forest Park (Broshot 2011).

One hypothesis for the lack of tree recruitment is a lack of mycorrhizal communities in the urban area from air pollutant deposition that has changed the fungal community composition.

Urbanization could affect the distribution of fungal species and communities and could change the composition of the natural community and its seral succession to one that favors urbanized saprotrophic fungi and non-host specific mycorrhizal fungi. A reduction in ECM fungi colonization and propagation has been observed in urban environments compared to less human impacted rural environments (Bainard et al. 2010; Stabler et al. 2001; Wiseman and Wells 2005). Urbanization increases concentrations of nitrogen and sulfur deposition that has a wide range of effects on soil chemistry. In the Pacific Northwest predominant winds carry air masses from the Pacific Ocean into metropolitan areas, where nitrogen pollution accumulates, and then continue into wilderness areas in the Cascade Mountain Range (Prather 2017). Nitrogen deposition has varied effects that can be positive or negative. According to the EPA (2019) excess nitrogen deposition has a myriad of effects including an increase in plant growth, decreased biodiversity, acidified soil, increased damages from pests, reduced rebound from frost events, and increased levels of nitrogen leaching into water bodies. In addition, soil eutrophication from nitrogen may reduce the likelihood of symbiotic ECM tradeoff dependency with hostplants (Allen et al. 2019).

METHODS

Site Description

Twenty-four permanent study sites each with three transects were established in Forest Park in 1993 with one additional site located in a remnant of mature forest NW of the park (the Ancient Forest Preserve). The urban forest is in northwest Portland, Oregon with an area of roughly 2,000 hectares and a length of 11 kilometers (Broshot 2011). The park was divided into three longitudinal sections along a perceived urban-to-rural land use gradient with urban development and industrial areas around the park's boundaries (Figure 1). Forest Park borders the Willamette River floodplain on the east side with the Tualatin Mountains to the west ranging between 20 and 335 meters above sea level. Soils found at sites are composed of Goble Silt Loam, Cascade Silt Loam, and Wauld Very Gravelly Loam. Ninety to 95% of the upland slopes exceed a 30% grade, resulting in much of the land having the potential for major landslides (Darling et al. 2011). The climate is a maritime environment with warm dry summers and cold wet winters in conjunction with mild temperatures (CSWCD 2020).

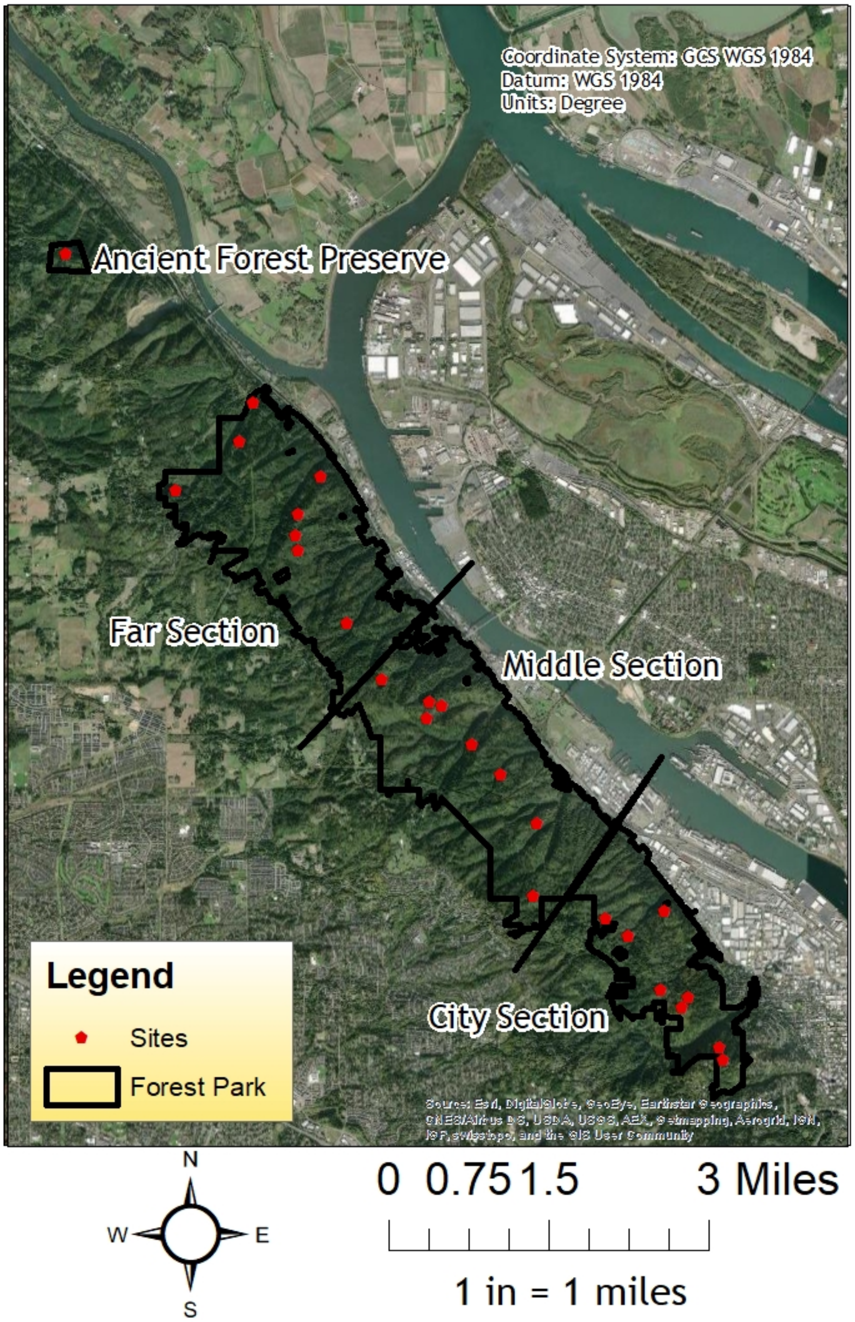


Figure 1. Map of Forest Park in Northwest Portland, Oregon. Locations of permanent sites and baby tree monitoring sites in the park and Old Growth (Ancient Forest Preserve) site. Map made by Jordan Leis using ESRI's ArcMap 10.8.

Three control sites were added to the study above Portland's air pollution plume in the Mount Hood National Forest in 2013 to determine if lack of recruitment of trees was just an urban phenomenon. These control sites were used to compare fungal community evenness and biodiversity to communities found in the urban forested environment. (Figure 2).

Source: Esri, Maxar, GeoEye, Earthstar Geographics, CNES/Airbus DS, USDA, USGS, AeroGRID, IGN, and the GIS User Community

Figure 2. Map of control sites located in Mt. Hood National Forest relative to Forest Park in Portland, Oregon. Map made by Jordan Leis using ESRI's ArcMap 10.8.

All study sites lie within the Douglas-fir (*Pseudotsuga menziesii*) - western hemlock (*Tsuga heterophylla*) vegetation zone that covers much of western Oregon and Washington. Western hemlock and western red cedar (*Thuja plicata*) are considered the climax species of this area, with Douglas-fir as a sub-climax species that dominates many forests due to its longevity of 500+ years (CSWCD 2020). The park's long history of 'disturbance' from fires and logging categorizes it as a second growth Douglas-fir forest. Continual frequent disturbances have led to bigleaf maple (*Acer macrophyllum*) and red alder *(Alnus rubra*), shade-intolerant deciduous species, dominating in many sections of Forest Park (Broshot 2011). Much of Forest Park consists of 50–100-year-old trees with a quarter of the urban forest covered by conifers and three-quarters by mixed coniferous-deciduous forest (CSWCD 2020).

Data Collection

We collected data from each permanent site in Forest Park, the Ancient Forest Preserve, and the control sites in the national forest. Mushrooms were identified (and a sample collected if not already known to species) in each of the three, 250m^2 quadrants at each site (Figure 3). In addition, mushroom samples were collected alongside trails to and from sites and were labeled based on the site to which they were closest. Each sample was tentatively identified in the field based on physical appearance to the most likely taxa; individual mushroom samples were collected and returned to the lab for spore prints and final identification to genus and species. For each mushroom, we noted the substrate (e.g., log, soil, duff), the identity of the nearest tree, shrub, and herbaceous species, and the GPS location taken. Soil moisture, ECp (electrical conductivity), and temperature were measured using a HH2 Moisture Meter from Delta-T Devices.

We used dichotomous keys and iNaturalist to identify fungi in the field and the lab (Ammirati and Trudell 2014; Kuo 2021; Miller 2021). In the lab unknown mushrooms were identified using spore prints and spore microscopy. Literature for which fungi are ECM with what vascular plant also was researched to sort mushroom samples into three categories: saprotrophic, parasitic, or ECM (Kuo 2021; Miller 2021). Species diversity index helps statistically quantify species richness and evenness in a community. We calculated Shannon's diversity index (H) and Shannon's equitability (E_H) to characterize Forest Park and the control sites fungal community (figure 3) (NIST 2016).

Shannon Diversity

$$H = -\sum_{i=1}^{S} p_i \ln p_i$$

Shannon Equitability

$$E_H = H / H_{\max} = H / \ln S$$

Figure 3. Equations used to calculate Shannon's diversity index (H) and Shannon's equitability (E_H)

iNaturalist is a collaborative project initiated by the California Academy of Sciences and National Geographic Society that shares in-site observations with other users and scientific data repositories (iNaturalist 2021). Using the website, we set up a project to record citizen science observations of fruiting body fungi in Forest Park and near our control sites from January 1st, 2020, to December 31st, 2020. Observations were sorted categorically by "Research grade" identifications to increase the likelihood of correct identification to genus.
Our project link: https://www.inaturalist.org/projects/urban-forest-mushroom-study

RESULTS

We found 47 genera of fungi in our 612 mushroom samples. The iNaturalist data included 52 species of fungi found in 505 samples. Thirteen genera of fungi found during the summer are known to form ECM associations (Kuo 2021; Miller 2021), and three genera have parasitic species (Kuo 2021) (Tables 1 and 2). 135 species of fungi were identified in our dataset with species from *Collybia* and *Mycena Coprinoid* (inky cap) being the most species-abundant groups. The iNaturalist data set found 139 species and the most species rich genus was *Stropharia* followed by *Collybia*, *Mycena*, and *Pholiota*. Eight genera were found in our dataset and not in iNaturalist: *Alnicola*, *Chroogomphus*, *Entoloma*, *Phaeocollybia*, *Phellodon*, *Pseudohydnum*, *Scleroderma*, and *Simocybe* (Table 1). iNaturalist identified 16 different genera of fungi than our own data set. This included five saprophytic, seven mycorrhizal, and three parasitic fungi. (Table 2).

Table 1. Fungal genera we identified May-August 2020 in the field. *denotes ECM and ^denotes parasitic associations.

Agaricus
Agrocybe
*Alnicola
*Amanita
Baeospora
Bolblitus
*Chroogomphus
*Clauvina / Ramaria
Clitocyboid
Collybia
Conocybe
Coprinellus
Coprinopsis
Coprinus
*Cortinarius
Crepidotus
Dacrymycetes
Entoloma
Galerina
Gymnopus
Gymnopilus
*Helvella
*Hygrophorus
^Hypomyces
*Inocybe
Lepiota
Lycoperdon
Mycena
Marasmius
Omphalinoid
Panaeolus
Peziza
*Phaeocollybia
Phellodon
Pholiota
Pluteus
^Polyporus
Psathyrella
^Pseudohydnum
Rhodocollybia
*Russula
*Scleroderma
Simocybe
Stropharia
*Thelephora
Tubaria
*Xerocomellus

Table 2. Fungi genera identified from iNaturalist citizen science observations January 2020 - December 2020 (iNaturalist 2021). * Denotes genus as ECM and ^ denotes genus as parasitic associations.

Agaricus
Agrocybe
*Amanita
^Armillaria
Astraeus
Baeospora
*Boletus
*Cantharellus
*Clavaria
*Clavulina / Ramaria
Clitocyboid
Collybia
Conocybe
Coprinellus
Coprinopsis
Coprinus
*Cortinarius
Crepidotus
Dacrymyces
Galerina
Gomphus / Turbinellus
Gymnopus
Gymnopilus
*Gyromitra / Helvella
^Hericium
*Hygrophorus
^Hypomyces
*Inocybe
*Laccaria
*Lactarius
Lepiota
Lichenomphalia
Lycoperdon
Marasmius
*Morchella/ Verpa
Mycena
Omphalinoid
Panaeolus
Peziza
Pholiota
Pluteus
^Polyporus
Psathyrella
Rhodocollybia
*Russula
Stropharia
*Suillus
*Thelephora
Tubaria
*Xerocomellus
Xylaria

Fungi known as Collyboid mushrooms were the most abundant (31.2%) and diverse (11.1%) group of mushrooms identified; the core genera of this group are *Collybia*, *Gymnopus*, *Baeospora*, and *Marasmius* (Kuo 2021). Other groups of fungi were the *Coprinoid* mushrooms (inky caps) with an abundance of 14.1% and made up 6.7% of fungal genera found. Closely related to inky caps are mushrooms in the genus *Psathyrella* with an abundance of 12.2% and composed 4.5% of the genera found. Fungi in the genus *Mycena* individual abundance was 8.9% and made up 10.3% of the genera found. We found a total of 612 mushroom samples at our sites during the summer of 2020. Of those samples, the vast majority 591 (90.8%) were saprotrophic, 48 (7.8%) were ECM fungi, and 9 (1.4%) were parasitic (Figure 4).

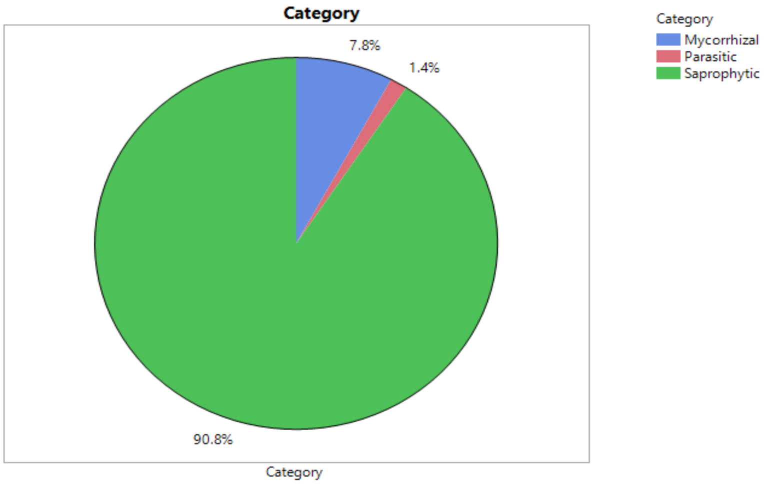
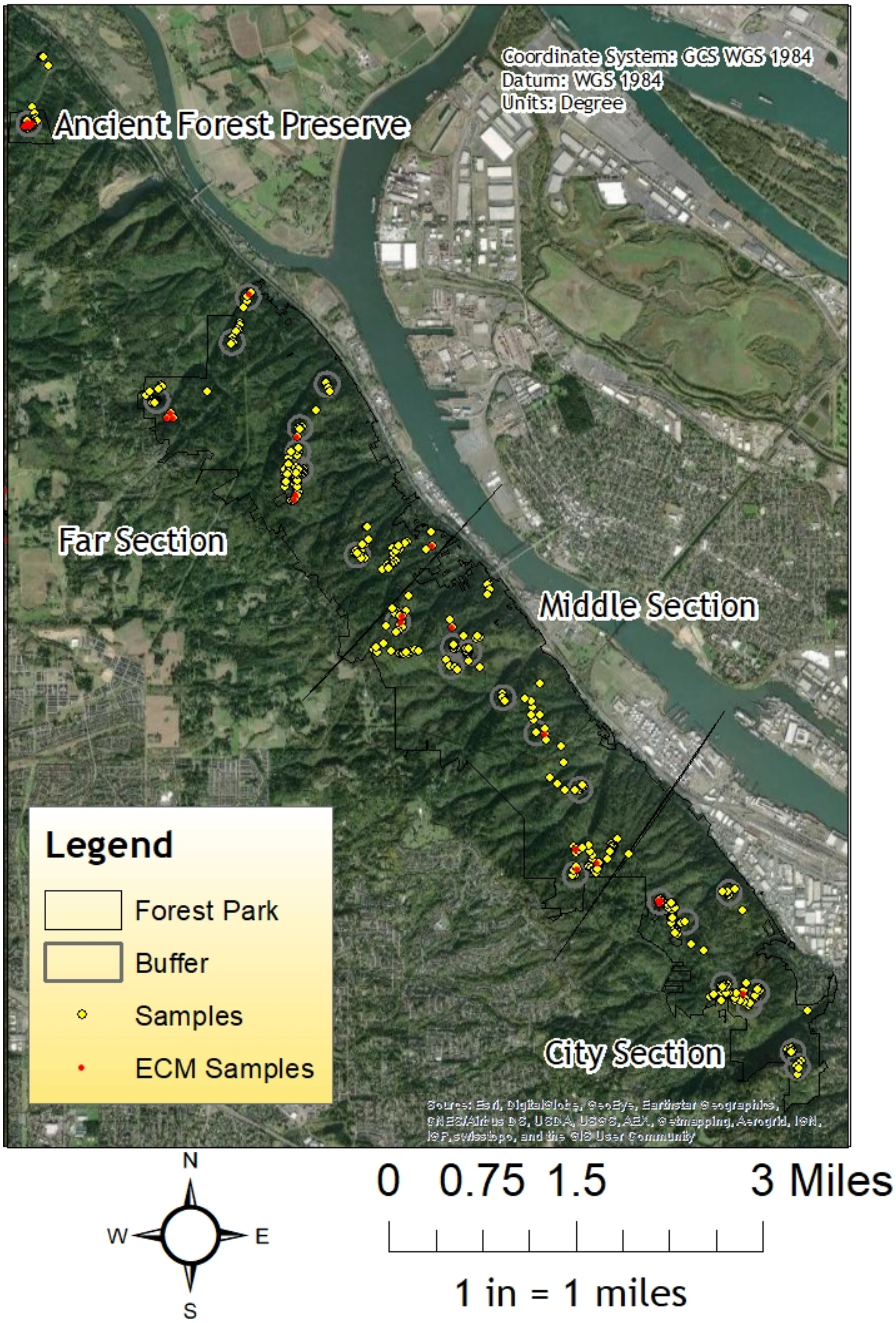


Figure 4. The percent (%) of saprotrophic, mycorrhizal, and parasitic fungi at all sites

Using ArcMap 10.8, I set up a 250.0 m^2 area buffered around each site to account for mushrooms ID'd around each site (Figure 5). In the city section of the park, 144 mushrooms were identified, five of which were ECM fungi. The middle section of the park, 156 mushrooms were identified with eight ECM fungi. The far section of the park had 195 mushroom identifications with eight ECM fungi. The Ancient Forest Reserve had 51 mushrooms identified with six ECM fungi (Table 3; Figure 5).



Source: Esri, DigitalGlobe, GeoEye, Earthstar Geographics, CNES/Airbus DS, USDA, USGS, AEX, Getmapping, Aerogrid, IGN, IGP, swisstopo, and the GIS User Community

Figure 5. Forest Park and all 546 mushroom samples recorded with ECM samples in red and saprobes and parasites as yellow. Buffer zones of $250.0m^2$ were allocated around each transect to make quadrats. Map made by Jordan Leis using ESRI's ArcMap 10.8.

Table 3. Sites and the section of the park with the total amount of mushrooms recorded at each site or towards the nearest site location.

Site Name	Park section	Total # of fungi found
1	City	9
2	Far	8
3	City	11
5	City	29
6	Far	9
7	Far	28
11	Middle	14
12	City	29
13	City	9
14	City	25
15	Far	70
17	Middle	14
18	Middle	16
19	Far	11
22	Far	15
23	Middle	3
24	Middle	11
28	City	23
30	City	11
32	Middle	56
36	Middle	14
37	Middle	1
39	Far	6
41	Far	8
50	Far	18
OG	Ancient Forest Preserve	51

The national forest control sites had 93 mushrooms identified, 21 of which were ECM fungi (Table 4).

Table 4. Control sites and the total number of mushrooms recorded at each site or nearest location.

Site Name	Section	Total # of fungi found
BB (Big Bottom)	national forest	12
RR (Roaring River)	national forest	53
RB (Rainbow)	national forest	28

We found no significant difference among the site means of total fungi. The mean for the ancient forest preserves old growth is higher than that of the mean for the national forest, although this is due to the difference in site samples as the ancient forest preserve only contains one site while the national forest has three (Figure 6).

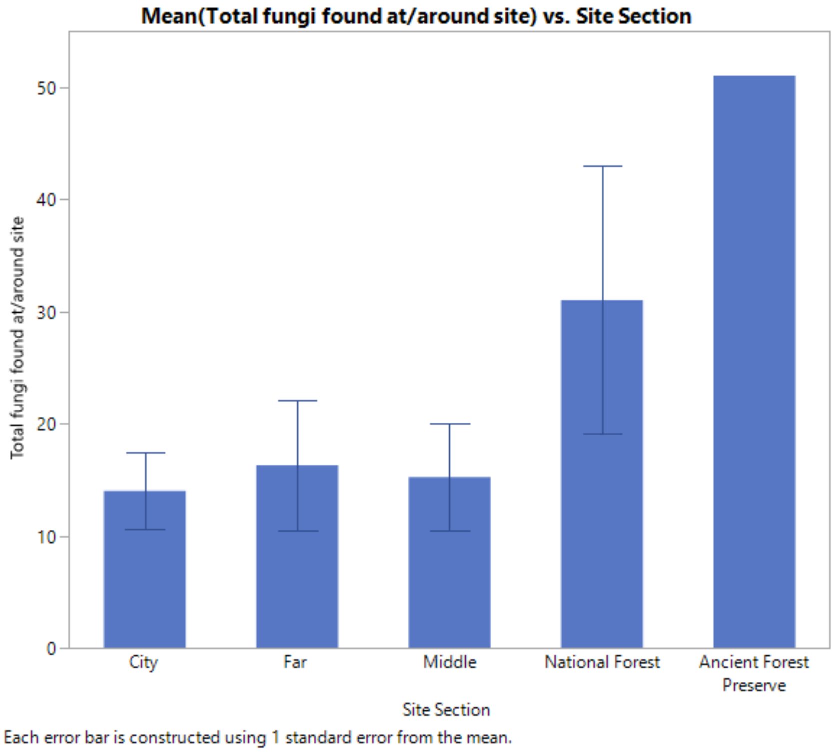


Figure 6. The mean number of fungi found at sites in Forest Park and the Mt. Hood National Forest with standard errors bars. An ANOVA resulted in a P=0.1481.

Examination and identification to which fungi were ECM and found a total of 48 ECM fungi between all sites. The national forest had the highest percent of mean ECM fungi (21.9%) compared to the other sections sampled, but the results were not significant according to the ANOVA and Tukey HSD statistical tests (P=0.0530) (Figure 7).

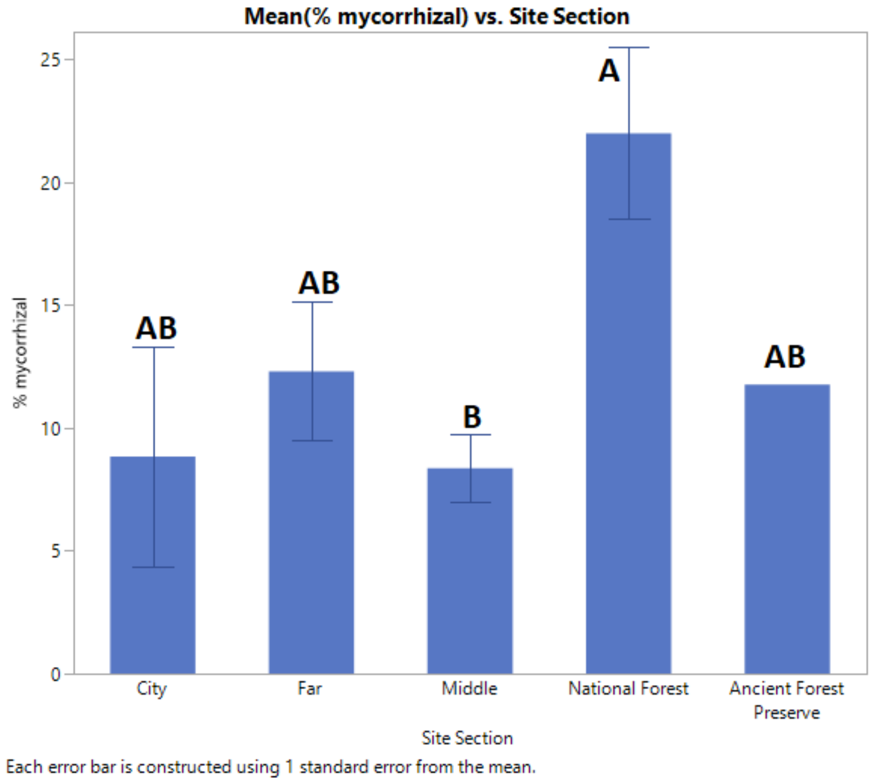


Figure 7. The mean percent (%) of ECM fungi found in different sections of Forest Park and the national forest with standard error bars. An ANOVA analysis resulted in a P=0.0530.

A X^2 analysis showed several fungal species had significant associations with nearby tree species (Table 5). *Amanita* mushrooms showed a high likelihood of associating with western hemlock, *Cortinarius* species were only associated with conifers, 50% of which were with western hemlock. *Inocybe* was the most abundant ECM fungi found (making up 47.62% of the ECM samples) and was associated with big leaf maple, Douglas-fir, western hemlock, and western red cedar. *Russula* fungi were most associated with Douglas-fir and less commonly with western hemlock.

Table 5. Contingency analysis of associated trees with mycorrhizal genera found in study sites. Pearson X^2 analysis revealed a P=0.0043 between ECM fungal genera and closely associated tree species present in Forest Park and the national forest.

Count Total % Col % Row % Cell Chi^2	Big leaf maple	Douglas fir	Pacific yew	Red alder	Western hemlock	Western red cedar	Total
Amanita	0 0.00 0.00 0.00 0.3571	0 0.00 0.00 0.00 0.9286	1 2.38 100.00 33.33 12.0714	0 0.00 0.00 0.00 0.2143	2 4.76 13.33 66.67 0.8048	0 0.00 0.00 0.00 0.3571	3
Cortinarius	0 0.00 0.00 0.00 0.7143	2 4.76 15.38 33.33 0.0110	0 0.00 0.00 0.00 0.1429	0 0.00 0.00 0.00 0.4286	3 7.14 20.00 50.00 0.3429	1 2.38 20.00 16.67 0.1143	6
Helvella	0 0.00 0.00 0.00 0.2381	1 2.38 7.69 50.00 0.2344	0 0.00 0.00 0.00 0.0476	0 0.00 0.00 0.00 0.1429	0 0.00 0.00 0.00 0.7143	1 2.38 20.00 50.00 2.4381	2
Inocybe	4 9.52 80.00 20.00 1.1010	5 11.90 38.46 25.00 0.2289	0 0.00 0.00 0.00 0.4762	0 0.00 0.00 0.00 1.4286	8 19.05 53.33 40.00 0.1029	3 7.14 60.00 15.00 0.1610	20
Naucoria	1 2.38 20.00 50.00 2.4381	0 0.00 0.00 0.00 0.6190	0 0.00 0.00 0.00 0.0476	1 2.38 33.33 50.00 5.1429	0 0.00 0.00 0.00 0.7143	0 0.00 0.00 0.00 0.2381	2
Russula	0 0.00 0.00 0.00 0.7143	4 9.52 30.77 66.67 2.4725	0 0.00 0.00 0.00 0.1429	0 0.00 0.00 0.00 0.4286	2 4.76 13.33 33.33 0.0095	0 0.00 0.00 0.00 0.7143	6
Xerocomellus	0 0.00 0.00 0.00 0.3571	1 2.38 7.69 33.33 0.0055	0 0.00 0.00 0.00 0.0714	2 4.76 66.67 66.67 14.8810	0 0.00 0.00 0.00 1.0714	0 0.00 0.00 0.00 0.3571	3
Total	5	13	1	3	15	5	42

Most ECM associations were with the coniferous species western hemlock (36%) and Douglas-fir (31%). The most abundant tree species in Forest Park, big leaf maple, was associated with only 12% of ECM fungi (Figure 8). Conversely saprobic fungi sampled associated more with deciduous species like big leaf maple (BLM) (45.2%) and red alder (9.59%) than coniferous douglas fir (17.5%), western hemlock (15.4%), and western red cedar (8.90%) (Figure 9).

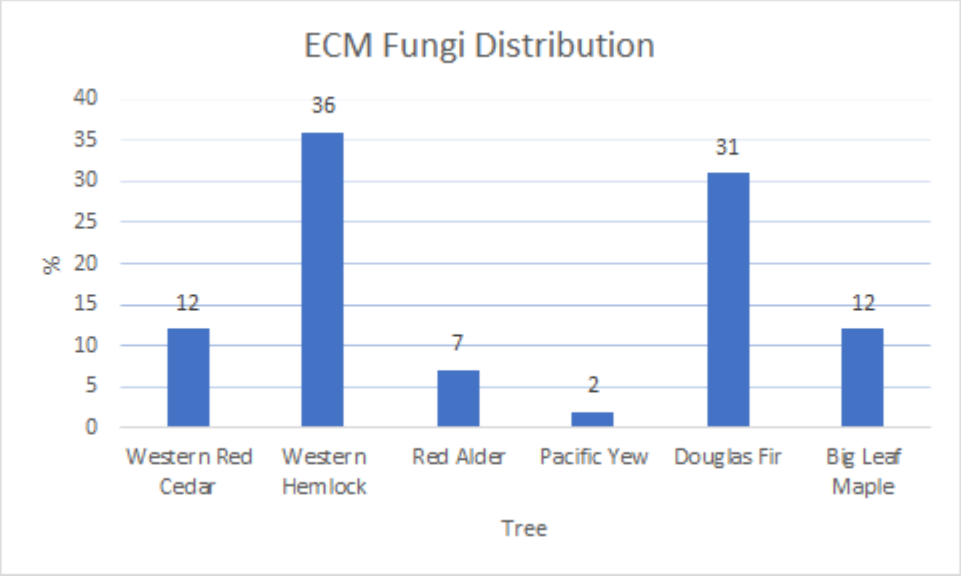


Figure 8. Distribution of ECM fungi with associated trees found in Forest Park and in the national forest.

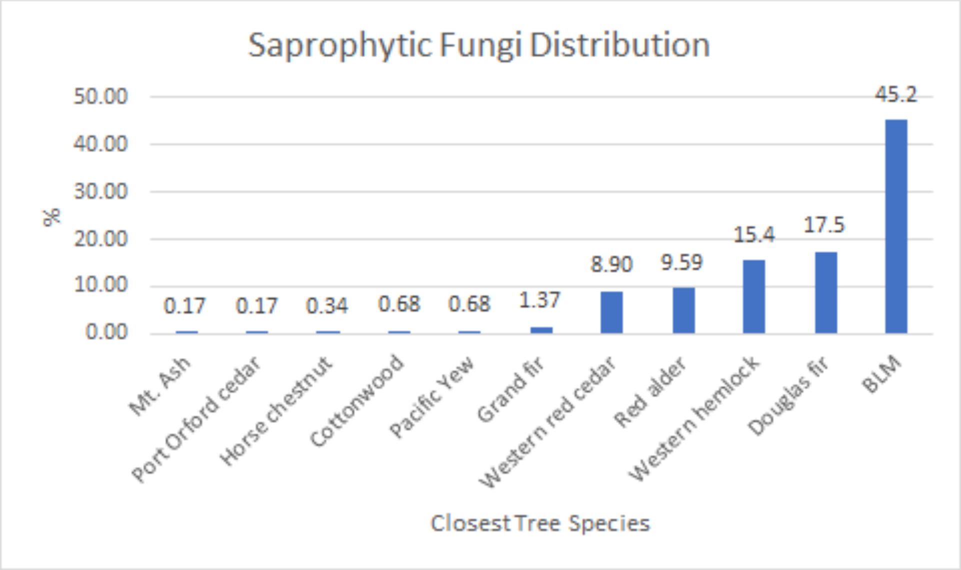


Figure 9. Distribution of saprophytic fungi with the closest tree found in Forest Park and in the national forest.

Sites in the national forest and far section of Forest Park had the greatest Shannon diversity scores from May-August, whereas the city section had the lowest diversity score, although these are not statistically significant (Table 6). The national forest, old growth, and far sections had the highest community evenness based on the Shannon equitability index. The city section of Forest Park had the greatest diversity according to the iNaturalist data and that the national forest had the greatest equitability. iNaturalist data contradicted data collected May through August and found that the national forest was less diverse than the urban forest.

Table 6. Shannon diversity index and equitability scores for sections of Forest Park and the national forest using data collected in the field and from the iNaturalist database.

Section	May-August Diversity Score	iNaturalist Diversity Score	May-August Shannon Equitability	iNaturalist Equitability Score
City	1.749	3.213	0.35	0.65
Middle	2.137	3.161	0.43	0.68
Far	2.669	3.168	0.51	0.58
Ancient Forest Preserve	1.984	-	0.50	-
National Forest	2.638	2.302	0.58	0.74

The raw diversity score per species of saprophytic and mycorrhizal (ECM) fungi were taken to find significant differences between the communities found in separate sections of Forest Park and the national forest. There was a significant difference in the mean genera diversity scores for ECM fungi in the national forest compared to ECM fungi in both the city and middle sections of Forest Park (Figure 10).

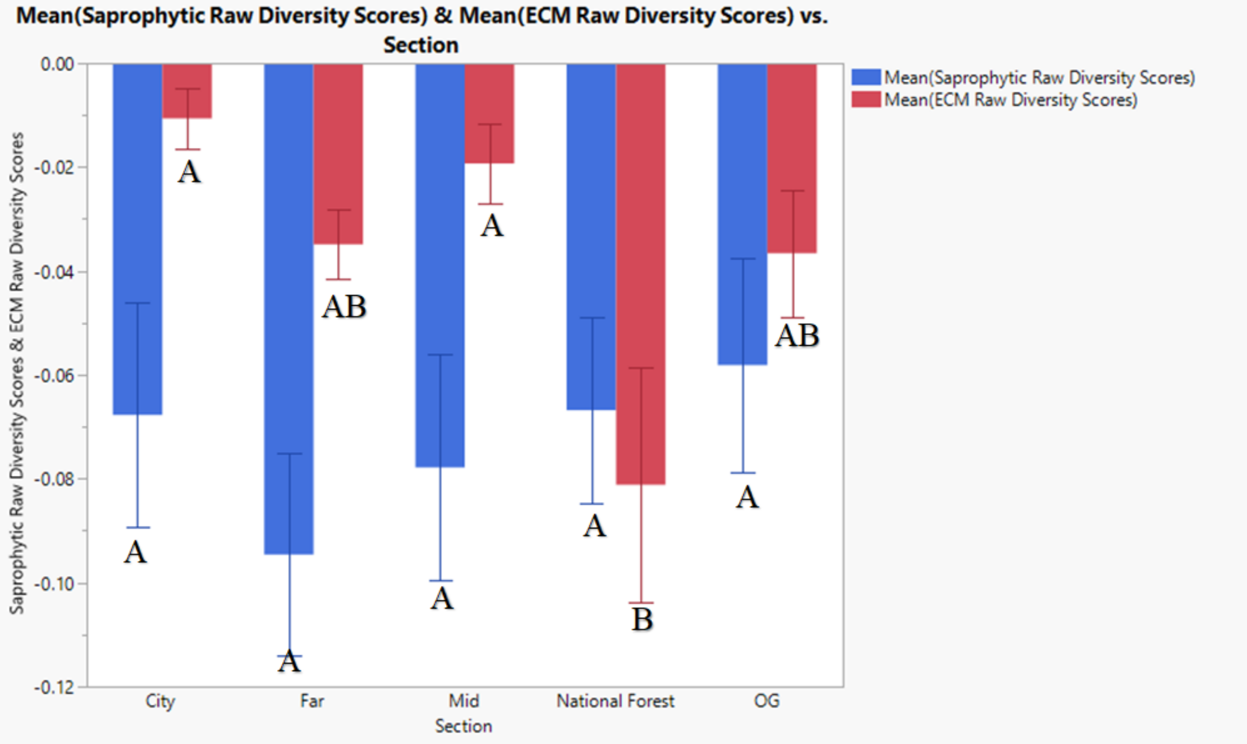


Figure 10. Mean diversity scores for fungi found in research sections sorted by saprophytic and mycorrhizal status. ANOVA analysis with a post-hoc tukey test resulted in a P=0.0024 (ECM) and P=0.7576 (Saprophytic) with standard error bars.

DISCUSSION

Fungal Community

There are no precise records of historical fungal populations in Portland other than the noted presence of choice edible mushrooms like common hardwood oysters (*Pleurotus ostreatus*), chicken of the woods (*Laetiporus sulphureus*), and lion's mane (*Hericium erinaceus*) (Darling et al. 2011). Our in-field data was collected during summer months and so our scope of the community was limited by that factor. For a greater community scope and to compare our list of fungal species found in Table 1., we incorporated citizen science data from iNaturalist that can be found in Table 2. It was anticipated that our iNaturalist project would provide more fungal genera than we had observed in the field especially during the fall and spring months when many fungi produce fruiting bodies with the abundance of moisture. Table 2. is a more accurate list of the variety of fungal mushroom producing species located in Forest Park. However, the iNaturalist data doesn't encapsulate community evenness and equitability of the fungal community in depth. The data we collected in the field may provide a fuller look at the summer fungal community, and compare specific substrates, plant associations, and detail more accurate identifications than iNaturalist users provide. iNaturalist data also may be biased. Diversity and equitability scores calculated using data from our iNaturalist project suggest that the city section of the park has greater fungal diversity than that in the far section, old growth, or national forest; but the equitability scores show the national forest had greater community evenness than Forest Park. Our data collected showed the city section with the lowest diversity and equitability, whereas the far and national forest had the greatest diversity and equitability. Figure 10 highlights this by showing a significant difference in the genera of ECM fungi located in the national forest compared to the city and middle sections of Forest Park. It clarifies that the far section and ancient forest preserve of Forest Park have similar genera diversity to the national forest. Some mushroom species are more easily found alongside trails and their appearances may be more interesting, inviting humans to snap a photo of and post online. The most common macrofungi found in Forest Park were in the genus *Gymnopus*, a group of woodland saprobes that are widely distributed throughout North America. Coprinoid mushrooms commonly referred to as inky caps are another group of saprophytes that are widespread across North America and are critical decomposers of wood, animal dung, grass, and leaf litter. *Psythrella candolleana* was a common occurrence found throughout Forest Park. This species is saprobic and typically occurs near dead hardwoods in woodlands but can also be found in lawns and pastures in urban and rural areas (Miller 2021). Fungi under the umbrella Coprinoid and in the genera of *Gymnopus* and *Psythrella* represented the bulk of fungi identified in Forest Park (57.5%) and can be categorized as cosmopolitan species as they are abundant in North America and thrive in highly disturbed urban environments (ANH 2011). Other described cosmopolitan mushrooms found in Forest Park though in lower abundance were species in the genera *Amanita*, *Agrocybe*, *Conocybe, Inocybe*, *Lepiota*, *Lycoperdon*, *Panaleous*, *Pholita*, and *Pluteus* (Kuo 2021; ANH 2011). Species in the genera *Amanita* and *Inocybe* are ECM fungi commonly found in urban areas, though *Inocybe* mushrooms can be both mycorrhizal and saprophytic depending on the species and environmental conditions (Kuo 2021).

Saprotrophic Fungi

In forest ecosystems saprotrophic fungi are vital decomposers of lignin, which makes up roughly 10-40% of woody plant tissue. The degraded residue is then available for a host of microbial life (Hobbie et al. 1998). Saprotrophs were the most common fungi found and several studies have shown that saprotrophic fungi have a competitive advantage over ECM fungi when nitrogen concentrations are higher in soils where decomposition rates were correlated with increasing nitrogen concentration (Peter et al. 2001; Lindahl et al. 2002). Urban soil sites tend to be saturated with nitrogen compounds due to nitrous oxides due to atmospheric deposition. This promotes saprotrophic dominance over ECM fungi in areas with high urbanization (Anonymous 2021). The relationship between saprotrophic and ECM fungi however also varies with forest age (Anderson et al. 2017, Gadd 2017, and Hood et al. 2005). Hobbie et al. (1998) found that rural forested stands with high concentrations of nitrogen and carbon isotopes had higher abundances of ECM fungi. The rate of urbanization also can affect saprotrophic fungi by altering chemical composition of soil duff, which is the primary substrate for many fungi. Wood decay fungi can be negatively impacted with reductions in the amount of down woody debris in various stages of decay that support species diversity and population viability (Lebel et al. 2010). As key regulators of nutrient cycling and soil O-horizon formation, saprotrophic fungi are vital in terrestrial ecosystems and cannot be replaced by fertilizers (Dighton 2007). In Forest Park changes in tree community composition have favored deciduous trees like big leaf maple and red alder (Broshot 2011) which could transform the species composition of saprophytic fungi towards a community of generalists or deciduous associated species over their less generalists and coniferous favoring species. After all, Forest Park and the national forest lie within the Douglas fir - western hemlock vegetation zone as the once climax community now more-so resembles a mid-age Douglas fir community (24.6% total acres) and hardwood topped by conifer community (41.7% total acres) (Darling et al. 2011).

Mycorrhizal Fungi

Of the 612 mushrooms collected, 48 were ECM fungi making up 7.8% of our sample size. We found 27 ECM fungi in Forest Park and the ancient forest preserve (25 sites) and 21 in the national forest (three sites). It was anticipated that the percent of fungi found to be ECM relative to saprophytes would be lower in Forest Park because the vegetation type there is dominated by hardwoods in many areas. Only 11.8% of the park is considered a mature or old growth vegetation type (Darling et al. 2011). Mature forests tend to be more dominated by mycorrhizal communities especially in less disturbed environments (Lebel et al. 2010). We found that the mean percent of mycorrhizal fungi were not significantly different among the sections of Forest Park. We did find that the National Forest had a significantly greater percent of ECM fungi than the middle section of Forest Park. Urban forests have been shown to have lower diversity and abundance of ECM fungi than rural forests, but that succession of these ECM communities is slower and continuously disturbed in urban environments from anthropogenic pressures (Bainard et al. 2010; Francini et al. 2017; Lebel et al. 2010).

The most abundant ECM genus was *Inocybe,* making up 47.62% of ECM fungi in Forest Park and the national forest. It is a large genus with over 1,000 species split between several subdivisions (AFTOL 2021). Some mushrooms in this genus are cosmopolitan ECM fungi widespread across North America like *Inocybe leptophylla* and *geophylla*, whereas others are more specialized and localized in ecoregions with specific habitat associations. Many are associated with both conifers and hardwoods but may also exist facultatively as a saprotroph (Kuo, 2021). Francini et al. (2017) found that *Inocybe* was the most dominant genus in urban parks with both young and old vegetation stands. Studies on mycorrhizae found that soils contaminated with heavy metals such as zinc and cadmium contained more abundant genera of *Inocybe*, *Russula*, and *Suillius* ECM (Gleason et al. 2017). Adaptive tolerance of some fungal species in soils with higher metal concentrations are necessary for plant community restoration and alleviation of these toxic compounds (Cain et al. 2014). Roughly 6 miles of the northwest Portland industrial zone borders Forest Park along with a long history of mismanagement and human intrusion that deposits nitrogenase and heavy-metal pollution (Broshot 2011 and Prather 2017). ECM fungi are easily damaged by acidic rain along with damaging the host plant and reducing fungal-plant carbon distribution (Lebel et al. 2010). ECM fungi produce small concentrations of organic acids which enhance weathering, dissolve heavy-metal bearing minerals, and mobilize phosphates in and out of biomass (Cain et al. 2014 and Gadd 2017).

Fungi in the genera *Cortinarius* and *Russula* were other abundant ECM fungi found in both Forest Park and the national forest. *Cortinarius* is the largest mushroom genus in the world but is limited in abundance due to specific ecological niches filled in association with host trees (Kuo 2021). The genus is not restricted to association with just conifers, but in Forest Park and the national forest, mushrooms in the genus *Cortinarius* were only associated with conifers such as Douglas-fir, western hemlock, and western red cedar, which are more prominent in later successional stage forests (Hood et al. 2005). *Russula* is another large genus with a variety of host trees and specific habitat requirements (Kuo 2021). All *Russula* found were associated with conifers. Both genera have adaptive rhizomorphs that prefer moist soils and are predicted to not be morphologically challenged from climate change (Talbot 2017). ECM fungi associated with Douglas-fir are often associated with western hemlock as well, though preference and other environmental factors can alter this (Hood et al. 2005). Nineteen percent of ECM fungi collected were associated with hardwoods in Forest Park and the national forest suggesting that some of the ECM community during summer associates with conifers. Douglas fir and western hemlock were the primary vegetation we found ECM associated with; historically that was the dominant vegetation of forests around Portland (CSWCD 2020).

ECM fungi colonize the roots of host trees, and all partners are dependent on one another for survival. Carbon, water, and nutrients flow through ECM networks and affect fungal and plant performance. They provide a competitive advantage to the plants involved (Bücking et al 2014). Carbon and nutrients flow symplastically and apoplectically through ECM networks, and soil and water can be redirected along water potential gradients (Lindahl et al. 2002). ECM networks are also vital in establishing recruitment of seedlings in primary succession. In already forested and secondary succession sites, seedlings can easily tap into mycorrhizal networks where plant-to-plant carbon transfer is most common in dark canopy forests (Nara 2015). Tree species in urban and rural parks in Ontario formed a tripartite association with AMF and ECM fungi, however ECM had significantly lower colonization rates in urban environments than AMF. This suggests that fungal communities in urban environments have a lower propagule abundance, and that lower colonization of ECM fungi occurs in highly disturbed urban soils (Bainard et al. 2010). Human modifications to soil pH, nutrient content, pollution, and lack of aeration are all possible factors (Boerner et al. 1996). A lack of mycorrhizal host species in urban environments may also play a role in decreased colonization as urban parks are often fragmented and disturbed and often dominated by invasive species that are unwilling or unable to form associations with local ECM communities (Bainard et al. 2010). Mycorrhizal networks across studies that show fungal networks' ability to facilitate interplant interactions by transferring photo assimilates from donor trees to seedlings and support natural regeneration of Douglas fir forests (Durall et al. 2009). ECM mutualistic fungi utilizing enzymatic decay processes to make biologically accessible nitrogen may also have implications in plant responses to rising CO_2 in poor organic nitrogen-containing soils (Arharya et al. 2021).

The degraded mutualism hypothesis proposed by Bever and Vogelsang (2009) states that invasive plants do not form vital AMF networks or are poorly colonized and alter the AMF community structure taking away vital beneficiaries to native plants. This follows in suit with a decrease in AMF abundance and/or efficacy can reduce community competition and alter biotic resistance of mycotrophic native populations (Bunn et al. 2015). The city of Portland lists one of Forest Parks management goals as “Intact native plant and animal communities with minimal disturbance from non-native species and invasive species populations controlled through management.” 30% of the park is statistically impacted by English ivy (*Hedra helix*) followed by clematis (*Clematis vitalba*), English holly (*Ilex aquifolium*), non-native laurel (*Prunus sp.*), English hawthorn (*Cratageus monogyna*), horse chestnut (*Aesculus hippocastanum*), and Himalayan blackberry (*Rubus bifrons*) (Darling et al. 2011). It’s postulated that due to the high diversity and occurrence of invasive and naturalized species in the urban forest this could influence the fungal community. So far there is little support that invasive species are harming AMF communities but there is some evidence from meta-analysis that AMF community composition changes slightly with naturalization of invasive forbs and grass; but that fungal colonization, plant-AMF responses, and community composition did not statistically change. This suggests that plant communities don’t shape AMF composition, but rather AMF are more likely influencing trajectories of invasion when exotic and native plants belong to differing functional groups. (Bunn et al. 2015).

RECOMMENDATIONS

Forest Park is a large urban forest with an area around 2,000 hectares and contains 80 miles of trails and roads. Being able to gather a more complete list of the ECM communities would require at least a year of surveying the park along trails and roads along with the 26 research sites and nine baby tree sites. This study was conducted over the time span of three months during the summer which greatly limits the species diversity that can be accounted for during Oregon’s dry summer. Using iNaturalist and other citizen science or identification groups online is a great tool for establishing a base list of mushrooms producing fungi and assisting with identification, and this paper establishes a more formalized basis of the fungal community in Forest Park. Additionally, continuous planting of saplings pre inoculated with generalist and ecosystem specific mycorrhizal spores (and beneficial soil bacteria) has proven more beneficial in establishment of new trees and the likelihood that those baby trees will survive into the future (Carrington et al. 2009).

In conjunction to more time doing in-field research, additional in-lab study conducting ITS-polymerase chain reaction (PCR) of soils and plant roots to identify fungal DNA strands and using BLAST to statistically match those sequences to known samples of ECM and AMF DNA or proteins. However, as soil fungi communities are well stratified and known to be diverse, specific PCR primers for ECM fungi are limited. Beeck et al. (2014) found that primer ITS86F_ITS4 had a 97% (±6%) PCR efficiency, ITS1F_ITS2 at 82% (±4%), and ITS3_ITS4 76% (±5%) PCR efficiency rate with standard error. These primers were tested in environmentally contaminated samples. New primers tested *in vitro* and *in silico* investigations by Sato et al. (2012) found 99% coverage for their DNA-primers and found statistically little taxonomic bias between Ascomycota and Basdiomycota amplified sequences.

Conjunction sampling of AMF soil communities would also be beneficial and supportive of any claims that diverse mycorrhizal connections are indicative of forest health. Unlike most soil fungi PCR analysis, and especially for ECM fungi, AMF genetic analysis has relied more-so on rRNA (rDNA) genes that are specific for fungi found in Glomeromycota clade that identify specific ribosomal subunits. The process of grinding host-plant roots into dilutions and nested PCR is tedious, while cloning and sequencing fragments of DNA using ribosomal small subunits is the most efficient but requires specific primers for specific groups of AMF (Beeck et al. 2014).

Soil sampling and composition analysis are important steps in deducing if eutrophication or soil pollutants are prevalent and affecting soil fungi communities. Nitrogen deposition over 35-60 kg ha^{-1} $year^{-1}$ determined by Kottke and Wallenda (1998) is sufficient for a loss of mycorrhizae on plant roots and sporocarp malformation. Though this study was done in European forests, in North America soils with low nutrient contents critical loads of nitrogen deposition range from 3.66-7.84 kg ha^{-1} $year^{-1}$ (Lebel et al. 2010). In the Pacific northwest studies have yet to determine the critical load for trees, understory vegetation, and mycorrhizal responses in marine west coast forests; yet a study in SE Alaska found that critical load range for mycorrhizal fungi was between 4-10 kg ha^{-1} year^{-1} bulk deposition (Blett et al. 2014). Studies in both Europe and the Pacific Northwest have found that ECM fungi decline, and saprophytic species richness remains unchanged when high N-containing soils have increased nitrogen deposition; suggesting that N pollution increases the ratio between saprobes and ECM fungi when nutrients are more available, which can be determined by looking at decomposition rates with nitrogen (Edmonds and Trudell 2004, Arnolds 1991, and Lindahl et al. 2002). However, it is 'low certainty' that cumulative ecological effects from prolonged atmospheric inorganic nitrogen deposition will adversely alter mycorrhizal fungi essential for plant growth and development (Blett et al. 2014). Our ratio of saprobes to ECM fungi was heavily in favor of saprophytic fungi with 7.8% of mushrooms sampled being ECM, and 13 of the 47 genera found (27.6%) being ECM fungi. The ratio may turn out to be an indicator of unwanted disturbance in urban communities where fragmented natural areas remain, and most notably where concentrations of anthropogenic pollutants are influencing soil and thus urban forest characteristics (Lebel et al. 2010).

ACKNOWLEDGMENTS

This project would not have been possible without the guidance and support of Dr. Nancy Broshot of Linfield University, who generously let me use data collected for her research regarding macrofungi communities in relation to the larger scope of tree recruitment and mortality of Forest Park in Portland, Oregon. In addition, Barbra Van-Ness, a former professor, and laboratory coordinator at Linfield University, offered her expertise in ArcGIS and assistance which allowed me to produce the map figures used in this project. To the Forest Park Conservancy, a non-profit organization dedicated to the stewardship and restoration of Forest Park. Lastly, I'd like to thank the team that brought together iNatrualist which allows everyday people to record an observation of any fauna or flora, share the observation on a vast online database, and includes comments and forums for findings to be discussed. Without that database compiling sample observations across Forest Park and in the Mt. Hood National Forest throughout a year would have been impossible.

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