Short Communication: 
Macrofungal diversity in Mt. Makiling Forest Reserve, Laguna, 
Philippines: with floristic update on roadside samples in Makiling 
Botanic Gardens (MBG)

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Abstract. Nacua AE, Pacis HJM, Manalo JR, Soriano CJM, Tosoc NRN, Padiroga R, Clemente KJE, Deocaris CC. 2018. Macrofungal diversity in Mt. Makiling Forest Reserve, Laguna, Philippines: with floristic update on roadside samples in Makiling Botanic Gardens (MBG). Biodiversitas 19: 1579-1585. The Mt. Makiling Forest Reserve (MMFR) stands as a highly biodiverse habitat and the only intact natural forest near Metro Manila, the Philippines. It is one of the 18 key centers of plant biodiversity and 32 key ecotourism sites in the Philippines. In monitoring the implementation plans for protecting MMFR, the information pertaining to the mushroom biodiversity across decades is important. Therefore, we aim to study mushroom as an indicator for biodiversity since there has been studies in the past 30 years on the macrofungi of MMFR which we summarized here along with ours. Sampling was done in August 2017 based on the transect line of 1000 m along roadsides of Makiling Botanic Gardens (MBG). The distribution of the sampling units was carried-out using random and stratified sampling. Our study describes 21 macrofungal taxa collected from MMFR. Of these, 20 taxa belong to Basidiomycota and only one belongs to Ascomycota. Polyporaceae was found as the most dominant macrofungi family (24%). There were six (6) species that are medicinal, and no poisonous species noted. There are eleven (11) species in this study which are unique records compared with previous studies done in the macrofungi of MMFR. This is the first study done comparing mushroom across 30 years on a reserved area. Information on these macrofungal flora across time serves as a reference for the currently existing conservation efforts and implementation of biodiversity-related policies in MMFR.

Keywords: Ascomycota, Basidiomycota, flora, macrofungi, Makiling

INTRODUCTION

Fungi comprises an astounding breadth of taxa and ecologies, from the mushroom-forming fungi to yeasts, rusts, smuts, mold, and other symbionts. There is an estimate of 1.5 million fungal species on Earth, of which only about 70,000 have been described. Yet despite this, limited information is available for most species as it is estimated to take 1,000 years to characterize the over 1 million still unidentified species (Hawksworth 2004, Mueller and Schmit 2007).

Referred to as mushrooms, macrofungi are frequently visible to the naked eye (> 1 cm in size). These eukaryotes possess morphologically distinct spore-bearing fruiting bodies and commonly appear with stalks. Of the four fungal phyla recognized, macrofungi belong to the Ascomycota and Basidiomycota with an estimated 53,000 to 110,000 species (Hibbett, Binder et al. 2007, Mueller and Schmit 2007). While macrofungi grow best during the rainy season, they can be found in almost all types of substrates, e.g., soil, grassy ground, rotten wood, decaying organic matter, etc. (López-Quintero, Straatsma et al. 2012).

The nutritional and medicinal properties of many macrofungi are well known and documented. Fruit bodies of about 200 mushroom species are being consumed throughout the world, preferably as a delicacy. They are generally regarded as healthy foods, poor in calories and in fat, but rich in crude fiber, proteins, chitin, vitamins, minerals and bioactive mycochemicals (Manzi, Gambelli et al. 1999, Kalač 2013). Many edible mushrooms are also valued in complementary and alternative medicine, including Lentinula edodes (shiitake mushroom), Grifola frondosa (maitake), Hericium erinaceus, Flammulina velutipes, Pleurotus ostreatus, and Tremella mesenterica (Sullivan, Smith et al. 2006). Limited local studies on macrofungi revealed several biological activities such as anti-hypertensive (Eguchi, Dulay et al. 2014), anti-microbial (Reyes, del Rosario et al. 2017, Gaylan, Estebal et al. 2018), anti-thrombotic (San Pedro, Mangansat et al. 2017).
As one-third of global fungal diversity is believed to exist in the tropical regions with macrofungi comprising 10% of that diversity (Mueller, Schmit et al. 2007), fungal biodiversity in the Philippines covers 3956 species and 818 genera based on old published enumeration from the classic studies done in the 1930s (Teodoro 1937) as well as the updated list in 1977 (Quimio and Capilit 1981). However, despite a strong historical foundation for mycology in the Philippines which dates back to as early as 1820 (Graff 1916), it has been noted that “there are only a few recent developments in terms of fungal survey to advance the field probably” which may be attributed to small number of mycologists in the country (Cumagun 2015).

The collection and identification of the macrofungi in the wild could be an enormous input to promote and conserve mushroom biodiversity. In our paper, we present a recent survey of the macrofungi in Mt. Makiling Forest Reserve (MMFR), a 4,000-hectare forest that is the major headwaters of the Laguna Lake vegetation in Southern Philippines. Mt. Makiling stands as a highly biodiverse habitat and the only intact natural forest near Metro Manila, and which according to a recent hydrologic modeling study, has its forest reserve affected by the adverse impacts of climate variability (Combalicer and Im 2012).

Mt. Makiling is one of the 18 key centers of plant biodiversity and 32 key ecotourism sites in the Philippines (Ranada 2013) and is recognized by the Food and Agriculture Organization of the United Nations as a model of “Exemplary Forest Management in Asia” (Durst et al. 2005). Despite a Philippine law in protecting MMFR by the UPLB, the economic growth in the region through exploitation of the rich natural resources base has been moving forward with environmental and social consequences. In monitoring the implementation plans for protecting MMFR, the information pertaining to the mushroom biodiversity across decades is important. Therefore, we are studying mushroom as an indicator for biodiversity since there has been studies in the past 30 years on the macrofungi of MMFR which we are summarizing here along with ours. The study is done to understand biodiversity and provide a trend of biodiversity across time.

**MATERIALS AND METHODS**

**Description of the sampling area**

Mt. Makiling rises to a maximum elevation of 1,130 m above sea level (asl) with more than 3,000 hectares of its forest reserve above 400 m asl. Its forest reserve is located at coordinates 14°08’N 121°12’E and is 65 km from Metro Manila, the Philippines. Connecting the political boundary of the provinces of Laguna and Batangas, the forest covers sections of the municipalities of Bay, Los Banos, and Calamba in Laguna, and Sto. Tomas in Batangas. Since 1990, the University of the Philippines-Los Baños (UPLB) has been granted by the Philippine government exclusive control, jurisdiction and administration of the forest reserve by virtue of the Republic Act 6967. Wet months are usually from May to December and drier periods are from January to April. The average annual rainfall is about 2.4 m. Mean annual temperatures range from 25.5-27.5°C. The coldest month is January and the warmest are in April and May. There has been no recorded eruption of Mt. Makiling in recent history but volcanism is still evident through its geothermal features, like mud spring and hot springs. The soil in the Forest Reserve is clay-loam derived from volcanic tuff and ash. High in organic matter, the soil is very fertile. Acidity is typical of rainforest soil, the pH ranging from 4.3 to 6.5. The location of the study could be seen in Figure 1.

Figure 1. A. Geographical satellite view of the study site in Mt. Makiling Forest Reserve, Philippines. B. 1000-meter transect line indicated by black arrowheads traversing from the road of Makiling Botanic Gardens towards the mountain area. (Figures was made using ©Scribble Maps)
Sample collection
Sampling was done in August 2017 based on the transect line of 1000 m from Makiling road to Botanic Garden of Mt. Makiling Forest Reserve. The distribution of the sampling units was carried-out using random and stratified sampling (de Vries 1986). Collection of macroscopic fungi was performed in the morning when temperature is favorable and fruiting bodies of the macrofungi are expanded. Fruiting bodies were carefully detached and stored in water-proof containers with moisture absorbent. The collected macrofungi specimens were preserved either in 2% (for fleshy) and 4% formaldehyde (for woody or hard). All the macrofungi were photographed in their natural habitat, collected, identified and classified. All samples were kept by the Bureau of Plant Industry (BPI) of the Republic of the Philippines.

Characterization and identification
Macrofungal specimens were characterized and identified based on standard procedures in Quimio (2001) and Tadiosa et al. (2012). The following criteria were used to describe and identify the collected specimens: (i) cap or pileus- size (cm), shape, color and color changes (upper and lower surface), margin, and firmness of basidiocarp; (ii) gills or lamellae-attachment to the stipe, thickness, forking, color and color changes, and orientation of hyphae in the inner gills; (iii) stipe or stalk- size (cm), diameter (cm), attachment or position, shape, and firmness; (iv) presence or absence of cystidia, basidia, and basidioles; (v) presence or absence of sterigmata; (vi) mycelium-if visible, presence or absence of cystidia, basidia, and basidioles; (vii) hyphae, relative size, presence or absence of septation and clamp connection; (viii) habitat (substrate) and (ix) edibility (Stamets and Chilton 1983, Angeles, Arna et al. 2016). Ecology, habitat and edibility status were also noted. Confirmation of initial identities of the specimens was done by consulting with an expert from the National Museum of the Philippines, Manila.

RESULTS AND DISCUSSION
Surveys of macrofungi has been reported by different researchers as it is estimated that about 60% of the known Philippine fungi are data derived from field surveys in Mt. Makiling Forest Reserve (Quimio and Capilit 1981, Quimio 1986, De Castro and Dulay 2015). The extensive bibliography on fungi in Mt. Makiling has provided essential data for the aims of this paper.

Our study describes 21 macrofungal taxa collected from Mt. Makiling Forest Reserve in August 2017 (Table 1). Of these, 20 taxa belong to Basidiomycota and only one belongs to Ascomycota. Furthermore, the samples were classified under 10 families and 17 genera. There were 15 samples identified up to species level, and 6 samples were identified only at the genus level. Polyporaceae with 5 species had the highest distribution in terms of species occurrence (24%), followed by Stereaceae with 3 species (14%), Agaricaceae, Fomitopsidaceae, Ganodermataceae, and Russulaceae each with 2 species (10%), and Sarcoscyphaceae, Bondarzewiaceae, Cantharellaceae, and Dacrymycetaceae each with only one species (5%). Panaeolus papilionaceus has not been assigned officially under a family, hence the term incertae sedis. The collected macrofungi are listed in Table 1 together with their morphological and ecological descriptions.

Moreover, our checklist also includes six (6) species of medicinal value – *Ganoderma lucidum*, *Ganoderma sinense*, *Trametes pubescens*, *Trametes versicolor*, *Stereum hirsutum*, and *Stereum ostrea*. Notably, *Ganoderma lucidum* has been recorded to contain antitumor polysaccharides such as (1→3)-β-glucan (Han et al. 1995) and proteoglycan (Baek et al. 2002) while *Trametes versicolor* shows cytoxic activity in restricting the proliferation of cancer cells in case of leukaemia and breast cancer due to polysaccharide-protein compounds (Lau et al. 2004). Some edible species are also found with no poisonous species noted.

The species diversity and distribution of macrofungi in this study may have been affected by some factors. One is the prevailing meteorological parameters. The study area exhibits type I climate which is characterized to have two marked distinct seasons - dry and wet. Our team and that of de Castro’s (De Castro and Dulay 2015) both surveyed in August (2013 and 2017) due to favorable growth conditions for the mushrooms. Mt. Makiling rains are usually frequent between June to September which is accompanied by the abundant collection of macroscopic fungi during field sampling. Heavy rains are known to cause mushrooms to grow and decompose its substrates quickly. Recent studies have provided evidence of climate-induced elevational shifts in fungal productivity and diversity (Parmesan and Yohe, 2003; Walther, 2010). Forest fungi have been known to colonize new areas faster than tree or shrub species (Bebber et al. 2013; Bebber et al. 2014) and hence can be used as proxies to monitor early climate change impacts on forest communities (Alday et al. 2017).

Another factor for the macrofungal diversity is the presence of diversity of vascular plants in the region (Schmit and Mueller 2007). Rare occurrence of other mushroom families may be caused by biological barriers such as restricted reproductive strategies that can be caused by substrate, nutrient, or atmospheric limitations. Mt. Makiling Forest Reserve is home to 225 families, 949 genera, and 2,038 species, 19 sub-species, 167 varieties, and many cultivars of flowering plants and ferns (Pancho 1983). Macrofungi are suitably occurring in varying habitats with the pronounced climate and presence of lignocellulosic agricultural wastes found in the forest. It must be noted that there was also a recent study by De Castro and Dulay (2015) documenting macrofungi in a multistorey agroforestry system of Mt. Makiling which is located opposite side of the mountain with reference to the sampling site of this study. Interestingly, records show distinct mushroom floral patterns in opposite areas of Mt. Makiling. For instance, *Russula virescens* has been
Table 1. Checklist of the 21 taxa (species or genus level) of macrofungi identified from the roadsides of Makiling Botanic Gardens of the Mt. Makiling Forest Reserve, the Philippines and their morphological and ecological descriptions

| Family            | Species (or genus level) | Cap            | Cap diameter (mm) | Gills   | Spore color | Ecology  | Habitat/Substrate | Edibility |
|-------------------|--------------------------|----------------|-------------------|---------|-------------|----------|-------------------|-----------|
| Ascomycota        | **Sarcoscyphaceae**      | *Cookeina speciosa* | infundibuliform   | 11      | smooth      | unidentified | saprotrophic      | decaying woods | edible     |
|                   |                          |               |                   |         |             |          |                   |           |
|                   | **Agaricaceae**          | *Coprinus sp.* | conical           | 63      | free        | black     | saprotrophic      | decaying leaves | edible     |
|                   |                          | *Lepiota cristata* | umbonate         | 12      | crowded     | deep red to reddish brown | saprotrophic | decaying leaves | inedible   |
|                   | **Bondarzewiaceae**      | *Heterobasidion annosum* | offset | 26      | lacking     | unidentified | parasitic | decaying woods | inedible   |
|                   | **Cantharellaceae**      | *Cantharellus sp.* | infundibuliform   | 74-82   | decurrent   | white cream | saprotrophic      | deciduous or mixed forest | edible     |
|                   | **Dacrymycetaceae**      | *Dacryopinax spathularia* | coral like   | 8       | lacking     | white     | saprotrophic      | decaying woods | inedible   |
|                   | **Fomitopsidaceae**      | *Daedalea sp.*  | offset            | 33      | teeth like  | white     | saprotrophic      | decaying woods | inedible   |
|                   |                          | *Fomitopsis sp.* | offset            | 42      | lacking     | white     | saprotrophic      | decaying woods | inedible   |
|                   | **Ganodermataceae**      | *Ganoderma lucidum* | offset           | 42 (primordia) | lacking | brown     | saprotrophic      | decaying woods | edible, medicinal |
|                   |                          | *Ganoderma sinense* | offset          | 18      | lacking     | brown     | saprotrophic      | decaying woods | inedible, medicinal |
|                   | **Polyporaceae**         | *Hexagonia sp.* | crustose          | 37      | lacking     | white     | saprotrophic      | decaying woods | inedible   |
|                   |                          | *Polystictus sp.* | flat             | 56      | lacking     | white     | saprotrophic, parasitic | decaying woods | inedible   |
|                   | **Lentinaceae**          | *Lentinus tigrinus* | infundibuliform   | 17      | decurrent   | white     | saprotrophic      | decaying woods, dried leaves | edible     |
|                   |                          | *Trametes pubescens* | offset        | 32      | lacking     | white     | saprotrophic      | decaying woods | inedible, medicinal |
|                   |                          | *Trametes versicolor* | offset       | 42      | lacking     | white     | saprotrophic      | decaying woods | edible, medicinal |
|                   | **Russulaceae**          | *Lactifluus piperatus* | infundibuliform | 23      | crowded     | white     | saprotrophic, mycorrhizal | decaying leaves | semi-edible |
|                   |                          | *Russula virescens* | flat to convex   | 23      | adnate      | white     | mycorrhizal       | deciduous or mixed forest | edible     |
| Basidiomycota     | **Stereaceae**           | *Stereum ostrea* | offset            | 19      | lacking     | white yellowish to brownish | saprotrophic | decaying woods | inedible, medicinal |
|                   | **Stereum hirsutum**     | *Stereum sp.*  | infundibuliform   | 16      | lacking     | white     | saprotrophic      | decaying woods | inedible, medicinal |
|                   | **incertae sedis**       | *Panaeolus papilionaceus* | campanulate | 9       | adnate      | black     | saprotrophic      | decaying leaves | edible     |
| Macrofungi Found in Different Areas or Habitats in Mt. Makiling Forest Reserve, Philippines |
|----------------------------------------|---------------------------------------------|----------------------------------------|---------------------------------------------|
| Roadside samples in MBG (This study 2018) | Multistorey Agroforestry Systems (De Castro & Dulay 2015) | Associated with Decaying Dipterocarps (Tadiosa et al. 2012; Tadiosa & Militante 2006) | Mt. Makiling (representative samples with majority collected from the foot of the mountain) (Quimio 2001) |
| Cantharellus sp. | Agaricus perfuscus | Auricularia auricula-judae | Agaricus sylvaticus |
| Cookeina speciosa | Auricularia auricula | Auricularia polytricha | Agaricus rhoadsii |
| Coprinus sp. | Auricularia polytricha | Auricularia sp. | Agaricus merillii |
| Dacryopinax spathularia | Boletinellus sp. | Corticium sp. | Agaricus perfuscus |
| Daedalea sp. | Cantharellus infundibuliformis | Daedalea ambiguа | Agaricus pocillator |
| Hexagonia sp. | Coprinus sp. | Fomes pachyphloaeus | Agaricus augustus |
| Fomitopsis sp. | Corticium sp. | Fomes senex | Agrocybe sp. |
| Ganoderma lucidum | Crepidotus herbarum | Ganoderma applanatum | Armillaria sp. |
| Ganoderma sinense | Ganoderma applanatum | Ganoderma lucidum | Amanita hemiphaba |
| Heterobasidion annosum | Geastrum triplex | Ganoderma sp. | Amanita angustilamelata |
| Lactifluus piperatus | Lentinus sajor-caju | Hexagonia tenuis | Cantharellus aureus |
| Polystictus sp. | Lentinus tigrinus | Hexagonia sp. | Cantharellus infundibuliformis |
| Lentinus tigrinus | Lepiota cristata | Hymenochaete tenuissima | Chlorophyllum molybdites |
| Lepiota speciosa | Marasmius ramealis | Lentinus sajor-caju | Clitocybe dealbata |
| Marasmius ramealis | Marasmius rotula | Lentinus tigrinus | Clitocybe gibba |
| Mycena alcalina | Mycena caerulea | Lentinus tigrinus | Collybia dryophila |
| Oudemansiella canarii | Mycena caerulea | Lepiota cristata | Collybia reinkeana |
| Panaeolus foenisecii | Mycena caerulea | Oudemansiella canarii | Conocybe tenera |
| Panaeolus papilionaceus | Cantharellus aureus | Chlorophyllum molybdites | Coprinus niveus |
| Schizophyllum commune | Cantharellus aureus | Clitocybe dealbata | Coprinus fimbriatus |
| Stereum ostrea | Cantharellus aureus | Clitocybe gibba | Coprinus flaviceps |
| Stereum hirsutum | Cantharellus aureus | Collybia dryophila | Cortinarius callices |
| Stereum sp. | Cantharellus aureus | Collybia reinkeana | Cortinarius sp. |
| Trametes pubescens | Cantharellus aureus | Conocybe tenera | Crepidotus herbarum |
| Trametes versicolor | Cantharellus aureus | Coprinus niveus | Entoloma lividum |
| Trametes pubescens | Cantharellus aureus | Coprinus fimbriatus | Gymnopilus sp. |
| Trametes versicolor | Trametes pubescens | Coprinus flaviceps | Hebeloma sp. |
| Trametes versicolor | Trametes pubescens | Cortinarius callices | Hygrocybe cinerea |
| Trametes versicolor | Trametes pubescens | Cortinarius sp. | Hygrophorus pratensis |
| Trametes versicolor | Trametes pubescens | Crepidotus herbarum | Lactarius sp. |
| Trametes versicolor | Trametes pubescens | Entoloma lividum | Lactarius porphyrosporus |
| Trametes versicolor | Trametes pubescens | Gyromitra esculenta | Lactarius sp. |
| Trametes versicolor | Trametes pubescens | Hebeloma sp. | Hygrocybe cinerea |
| Trametes versicolor | Trametes pubescens | Hygrophorus pratensis | Lactarius sp. |
| Trametes versicolor | Trametes pubescens | Lactarius sp. | Lactarius piperatus |
| Trametes versicolor | Trametes pubescens | Lentitubularia nigricans | Lentitubularia nigricans |
| Trametes versicolor | Trametes pubescens | Leucopaxillus giganteus | Leucopaxillus giganteus |
| Trametes versicolor | Trametes pubescens | Leucopaxillus giganteus | Lepiota arbutina |
| Trametes versicolor | Trametes pubescens | Leucopaxillus giganteus | Lepiota arbutina |
| Trametes versicolor | Trametes pubescens | Leucopaxillus giganteus | Lepiota arbutina |
| Trametes versicolor | Trametes pubescens | Leucopaxillus giganteus | Lepiota arbutina |
| Trametes versicolor | Trametes pubescens | Leucopaxillus giganteus | Lepiota arbutina |
| Trametes versicolor | Trametes pubescens | Leucopaxillus giganteus | Lepiota arbutina |
| Trametes versicolor | Trametes pubescens | Leucopaxillus giganteus | Lepiota arbutina |

n=21  n=20  n=27  n=71
distinctively identified in Makiling trail roads within UPLB upper campus. Members from the genus *Russula* is found to be an indicator of lead (Pb) on its fruiting body (Cayir et al. 2009). It can be associated with public utility vehicles accessing the upper campus of UPLB.

In total, eleven (11) species in this study is unique with reference to the records of De Castro and Daly (2015), Tadiosa et al. (2012), Tadiosa and Militante (2006), and Quimio (2001). Table 2 presents the comparison of species recorded by different authors with Quimio (2001) having the most number of species recorded representing the first effort in documenting macrofungal diversity in MMFR. In summary, this is the first study done comparing mushroom across 30 years on a reserved area. As a consequence, information on these macrofungal flora across time serves as a reference for the currently existing conservation efforts and implementation of biodiversity-related policies in MMFR.

In conclusion, a total of 21 macrofungi taxa (both genus and species level) belonging to 10 families and 17 genera had been recorded in roadside samples within the Makiling Botanic Gardens (MBG) vicinity at Mt. Makiling Forest Reserve. The study declares that *Polyporaceae* was found as the most dominant macrofungi family (24%). There were six (6) species that are medicinal, and no poisonous species noted. There are eleven (11) species in this study which are unique records compared with previous studies done in the macrofungi of MMFR. The researchers emphasize the need for the integration of local macrofungi researches into a general macroecological framework and in national biodiversity assessments in order to establish useful information to aid in harnessing the potential of these valued macrofungal resources.

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