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Diversity and Ecological Roles of Macrofungi within UiTM Jengka, Pahang, Malaysia

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ABSTRACT. Macrofungi in urban areas are rarely explored, although their existence may significantly impact the ecosystem of these non-forested habitats. Therefore, early macrofungal documentation was carried out on the UiTM Pahang campus to record species diversity and their ecological roles in urban areas. Fruiting bodies of macrofungi were collected and documented from selected sites within the campus areas and were preserved as voucher specimens. Specimens were identified using main macro- and micromorphological key characters and other important features such as odour and colour change upon bruising. A total of 62 species representing two phyla and 23 families were collected in this study. Basidiomycota dominated the campus areas with 54 species, while Ascomycota appeared the least with only eight species. Numerous saprophytic fungi and a small number of mycorrhizal and parasitic fungi living within the campus areas were recovered. The majority of the macrofungi played a role as saprophytes (46 species; 74%), followed by mycorrhiza (14 species; 23%) and parasites (two species; 3%), displaying a balanced proportion of saprophytic and mycorrhizal fungi with extremely low parasitic fungi. The findings also clearly demonstrated that UiTMCPh is a good habitat for numerous groups of macrofungi and is highly favoured by saprophytic macrofungi. Considering the high diversity of macrofungal species from the current work, conducting extensive sampling and regular monitoring of macrofungi are necessary to enhance the conservation programs on the campus.

Keywords: Malaysia, Mycorrhiza, Parasitic saprophytic, Urban

INTRODUCTION

Macrofungi play essential ecological roles in their habitats and contribute to ecosystems' overall diversity and health. They act as natural decomposers (Crabtree et al., 2010; Kinge et al., 2017; Santamaria et al., 2023), facilitating nutrient cycling in ecosystems by breaking down organic matter such as fallen leaves, dead plants, and woody debris, which will then be released back into the soil and utilised by plants and other organisms. Macrofungi also form mutualistic relationships with the roots of certain plants (Sulzbacher et al., 2012; Helbert et al., 2019; Hermawan et al., 2020) by exchanging nutrients and water for sugar. In addition, certain macrofungi cause decay in healthy plants because of their parasitic behaviour (Tapwal, 2013). Contrasting to the symbiotic relationships observed in mycorrhizal macrofungi, parasitic macrofungi obtain nutrients from their host plants; in this process, they often cause harm or disease to the hosts. In addition to the above-mentioned roles, some macrofungi are beneficial for their

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nutritional and pharmaceutical properties, being consumed by many as food and traditional medicines (Enow Andrew et al., 2013; Degreef et al., 2016; Hyde et al., 2018; Samsudin and Abdullah, 2019).

Despite the diverse ecological roles and benefits of macrofungi, this remarkable group of organisms is currently threatened by multifaceted pressures, including those caused by human activities. Rapid development and urbanisation are considered the most threatening human activities because of their severe impact on macrofungi and their natural habitats. Their habitats experience major changes that usually possess several features, including parks, malls, residential areas, roads, health facilities, and many others. In addition, the environments of urban habitats are usually harsh, with extreme conditions such as compacted soil with limited volume, higher pH and temperature, reduced organic matter, and heavy metals (Lu et al., 2010; Skrbic et al., 2012). Endless land expansion causes a constant disturbance, which could lead to a long-term ecological imbalance. Due to this, urban habitat research is becoming more significant for biodiversity protection and, at the same time, to equalise conservation efforts and inevitable development in such areas. In Malaysia, most newly developed urban areas were converted from agricultural land use. These areas have experienced significant modifications, resulting in novel ecological niches with new species colonising the areas. Bandar Tun Razak Jengka, Maran is no exception to these modifications. This small town has become one of the successful stories of Malaysia's government projects to reduce inequality among its people. From an agricultural-based town, Bandar Tun Razak Jengka has transformed into urbanism. Urbanisation is economically beneficial for the area, but various unwelcome threats await. The main impacts of urbanisation are changes in land use and emissions, including air, noise, and water pollution. Due to the increased stress, urban environments like Bandar Tun Razak Jengka are unique and hostile to pre-existing organisms (Szabó et al., 2014), including macrofungi.

In the current work, Universiti Teknologi MARA Pahang Campus (UiTMCPh) was chosen as the site representing a fragment of urban habitat in Bandar Tun Razak Jengka. The campus and its vicinity consist of urban landscapes with buildings, avenues, gardens, and agricultural settings, especially oil palm and rubber plantations. Since campuses are considered disturbed areas, these areas may hypothetically support fewer macrofungi than forests. Previously, Tibuhwa (2011a) documented more than 61 species out of 676 fruiting bodies collected at the University of Dar es Salaam, Tanzania, with diverse ecological roles such as *Termitomyces* spp., *Agaricus* spp., *Chlorophyllum* spp., *Schizophyllum commune*, and *Ganoderma* spp. Csizmár et al. (2013), on the other hand, recorded 60 species from urban areas in Hungary consisting of 41 ectomycorrhizal fungi such as *Scleroderma* sp., *Inocybe* sp., and *Russula* sp. They also collected 19 saprophytic fungi, including *Agaricus* spp., *S. commune*, and *Pleurotus* sp., among others. Therefore, this study aimed to collect and identify the macrofungi found in the urban habitat of the UiTMCPh campus and determine their ecological roles, either as saprotrophs, mutualists, or parasites. Aside from that, there are likely novel or untapped species of macrofungi on campus that could be recorded for future reference. Such findings also benefit the new documentation of macrofungal species in Pahang.

METHODOLOGY

Study site and specimen collection

This study was conducted in UiTMCPh (Figure 1), which is situated in Bandar Tun Razak Jengka, Pahang (3°45'17" N, 102°33'43" E). Sampling series were carried out from January 2021 to October 2022 for optimal encounters with macrofungi. All the visible macrofungi in the university area were collected and documented. Fresh fruiting bodies of macrofungi were harvested with a scalpel, and wood-inhabiting macrofungi were collected using a bush knife. A paintbrush was used to remove dirt from the specimens. Collected macrofungi were temporarily stored in paper bags labelled with the specimen number, date, and location of the collection. The specimens were classified into saprophytes, mycorrhiza, or parasites based on their hosts or substrates *in situ.*

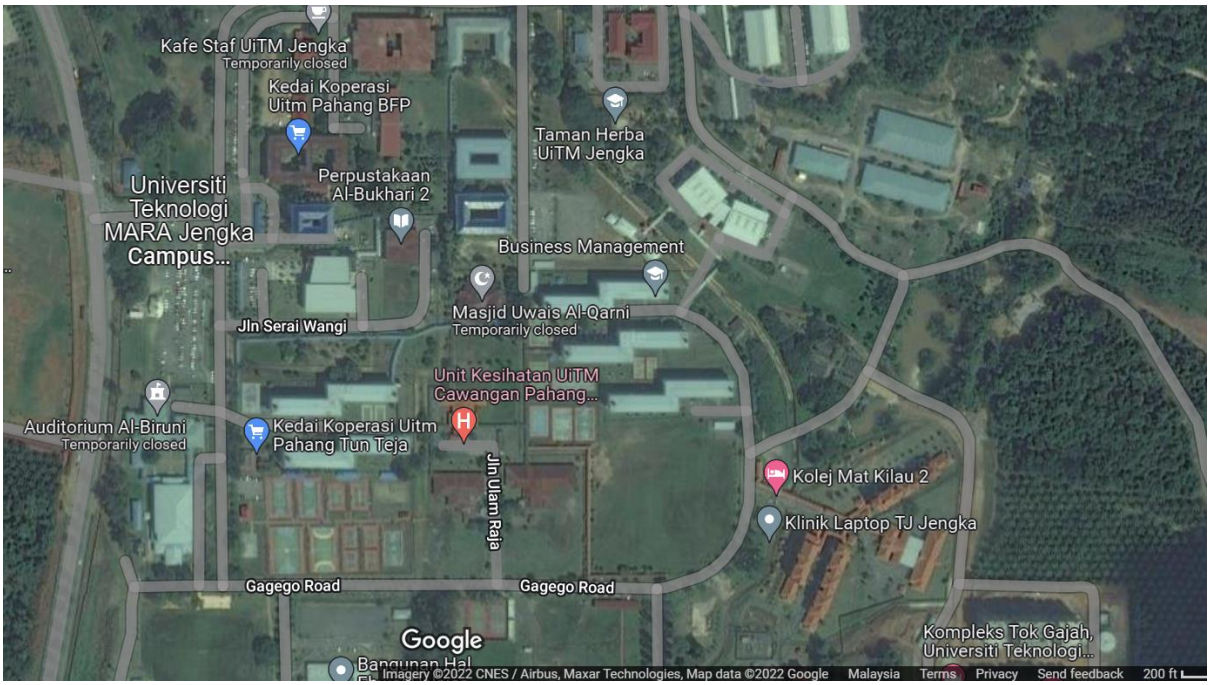

Figure 1. UiTMCPh campus area

Specimen identification and preservation

The specimens were photographed and documented from various views, including the specimens' side, top, and bottom (Aqilah et al., 2020). The shapes of fruiting bodies, cap colour, surface, edge, and margin (Parlucha et al., 2021) were also recorded. Hymenophore type (gilled, pores, or teeth) and attachment to the stipe (stem) were also observed and recorded. Other observed characteristics were stipe shape and colour, stipe surface, attachment position, stipe attachment type on the substrate, stipe cross-section, annulus (veil), and odour. The height and width of each macrofungal specimen were measured using a ruler (Leonard and Fechner, 2010). The specimens were then brought to the laboratory for further examination. Initial identification was based on the macroscopic morphology of the sporocarps, such as pileus, lamellae, stipe, the presence of annulus, and many others. Then, further observations were made on the microscopic characters of each collected specimen, such as spore ornamentation, asci, basidia, and cystidia, using a light microscope (Olympus, CX41). Specimens were identified to the species level (if possible) by

referring to multiple sources of literature (Phosri et al., 2004; O'Reilly, 2011; Ostry et al., 2011; Carbone et al., 2013; Hosen and Yang, 2013; Karun, 2015; Lee, 2017; Verma et al., 2018; Mohammad et al., 2019; Tsia and Mohammad, 2019; Tuah et al., 2019; Jaichaliaw et al., 2021) and two reliable websites (https://www.mushroomexpert.com/ and https://www.inaturalist.org/). Identified fungal specimens were dried using a commercial food dehydrator for 6-8 hours at 50°C. In general, larger samples were split in half or quartered from top to bottom before being placed in a dehydrator to prevent the sporocarp's inner part from decaying and prevent insect larvae's feeding activities. Meanwhile, smaller specimens were placed in a Petri dish during the drying process. After that, the specimens were stored in a plastic box for preservation.

RESULTS AND DISCUSSION

As an urban area, studies of macrofungi on university campuses are not many. However, appreciating the importance of biodiversity and its conservation, many institutions have shown interest in protecting and evaluating biodiversity at the campus level (Tibuhwa, 2011a; 2011b; Damian and Tibuhwa, 2014; Karun et al., 2018; Putra et al., 2020). Some of them plant avenue trees, or those that possess medicinal and nutritional values, as well as initiating gardens and arboretums for cultivating native, rare, and endangered tree species. Such landscapes generate substantial plant debris from woody leaf litter, old bark, and dead roots. All of these become the major substrates for the growth and perpetuation of macrofungi.

In this study, 62 species of macrofungi belonging to 26 families were collected (Table 1). Some specimens were identified up to the species level, and some were identified up to the genus level only due to the high similarities in their macro- and micromorphological characters. Saprophytes accounted for most groups (46, 74%), followed by mycorrhiza (14, 23%). The least number was noted for parasitic fungi, accounting for only two species (3%) (Figure 2).

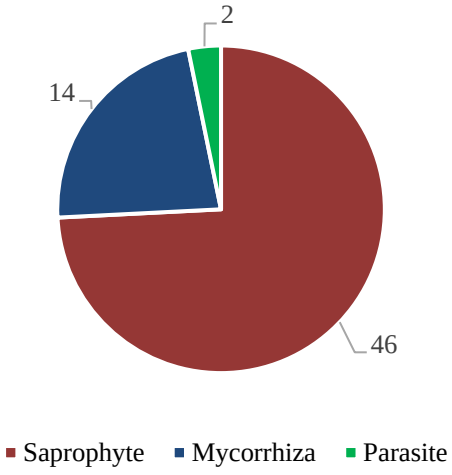

Figure 2. The total number of macrofungi within the UiTM Pahang campus based on their ecological roles

The findings matched those of studies in Southern Luzon, Philippines (Parlucha et al., 2021) and Northern Ethiopia (Alem et al., 2021), where both studies recorded saprophytes as the highest proportion of macrofungi collected. Saprophytic fungi are the most active decomposers of litter, contributing significantly to the carbon and nitrogen cycles as well as other soil nutrients (Chen et al., 2018). Therefore, the abundance of saprophytic fungi was common. Saprophytes are fungi that lack the ability to produce their nutrients. They feed on dead and decaying matter to survive. By nature, saprotrophs often grow on various substrates, primarily soil, wood, and leaf litter (Karun et al., 2018). All species from the phylum Ascomycota in this study were recovered from dead wood (Table 1). Meanwhile, saprophytes in the phylum Basidiomycota were derived from dead wood and soil. *Agaricus* and *Chlorophyllum* (Figure 3G) from Agariceae are great examples of saprophytic fungal genera that live in the soil (Table 1). Meanwhile, *Microporus* and *Trametes* (Figure 3E) from Polyporaceae are saprophytic fungal genera that live on dead wood and fallen branches. They are stiff and leathery in texture and can survive in low-moisture environments during the dry season (Parlucha et al., 2021). Saprotrophs are capable of degrading a variety of resistant organic substrates found in some land-use systems, such as "natural" and "planted" trees (Lynch and Thorn, 2006). UiTMCPh has many chopped-down trees and old branches, providing favourable environments for polyporoid fungi to grow, which explains the high number of these fungi within the campus (Table 1). This current finding fits well with a previous finding that saprotrophic macrofungi are more diverse in disturbed areas (Ye et al., 2019).

Mycorrhizal fungi recovered from the UiTM Pahang campus comprised 14 species (23%). Mycorrhiza, which means "fungus-root" in Latin, refers to the relationship between specific soil fungi and the fine roots of almost all forest plants (Molina, 1994). These fungi receive sugars from plants with greater access to nutrients and sometimes water from fungi (Dell, 2002). Also, the presence of mycorrhizal fungi improves the ability of plants to absorb nitrogen, phosphate, and potassium (Mohammadzadeh and Pirzad, 2021). Generally, the mycorrhizal fungi typically inhabited the forests. This was due to the fact that trees in disturbed or ornamental areas did not require the assistance of fungi since humans provided all the nutrients needed. In this study, several trees planted on the campus established associations with mycorrhizal fungi. For instance, species of the genus *Russula. Russula japonica*, *Russula* sp. 1, and *Russula* sp. 2 were found under the avenue tree, *Cinnamomum camphora* (Lauraceae) that has been planted alongside the road near the hostel buildings, meanwhile *Russula* sp. 3 was found under *Samanea saman* (Fagaceae) near nursery area. These findings are uncommon in Malaysia, particularly in Pahang, since there are no reports on the occurrence of *Russula* species associated with avenue trees available locally. However, fungi from the family Russulaceae were previously reported to be associated with plants from the family Orchidaceae, which included *Chamaegastrodia inverta* and under the canopies of *Areca catechu* (Pecoraro et al., 2020; Parlucha et al., 2021).

Among 14 mycorrhizal species, six were reported for the first time in Pahang, namely *Pisolithus* sp., *Astraeus hygrometricus Scleroderma* sp. 1, *R. japonica*, *Chanterellus* sp., *Tylopilus plumbeoviolaceus*, and *Inocybe* sp. (Figure 3). Studies of macrofungi in Pahang at Tasik Bera and Fraser's Hill have documented several mycorrhizal fungi, including *Boletus* spp., *Hemioporus* sp., *Pulveroboletus* sp., *T. nigropurpureus*, *Russula* spp., and *S. sinnamariense* (Zainuddin et al., 2010; Thi et al., 2011).

Table 1. Species diversity, host, and ecological roles of macrofungi found within UiTM Pahang campus

Phylum	Family	Species	Host	Roles
Ascomycota	Xylariaceae	*Xylaria hypoxylon*	Dead wood	Saprophyte
		Xylaria polymorpha	Dead wood	Saprophyte
		Xylaria sp.	Dead wood	Saprophyte
	Hypoxylaceae	*Daldinia concentrica*	Dead wood	Saprophyte
	Pezizazeae	*Peziza* sp.	Dead wood	Saprophyte
	Pyronemataceae	*Trichaleurina* sp.	Dead wood	Saprophyte
	Sarcoscyphaceae	*Cookeina sulcipes*	Dead wood	Saprophyte
		Cookeina tricholoma	Dead wood	Saprophyte
Basidiomycota	Schizophyllaceae	*Schizophyllum commune*	Dead wood	Saprophyte
	Mycenaceae	*Filoboletus manipularis*	Soil	Saprophyte
		Mycena sp.	Dead twigs	Saprophyte
	Marasmiaceae	*Marasmius haematocephalus*	Soil	Saprophyte
		Marasmius sp. 1	Soil	Saprophyte
		Marasmius sp. 2	Soil	Saprophyte
	Geastraceae	*Geastrum triplex*	Soil	Saprophyte
		Geastrum sp.	Soil	Saprophyte
	Polyporaceae	*Microporus xanthopus*	Dead wood	Saprophyte
		Pycnoporus sanguineus	Dead wood	Saprophyte
		Lentinus squarrosulus	Dead wood	Saprophyte
		Trametes elegans	Dead wood	Saprophyte
		Trametes versicolor	Dead wood	Saprophyte
		Trametes sp. 1	Dead wood	Saprophyte
	Auriculariaceae	*Earliella scabrosa*	Dead wood	Saprophyte
	Bolbitiaceae	*Auricularia auricula judae*	Dead wood	Saprophyte
	Dacrymycetaceae	*Conocybe apala*	Soil	Saprophyte
	Agaricaceae	*Dacryopinax spathularia*	Dead wood	Saprophyte
		Lepiota sp.	Soil	Saprophyte
		Agaricus thailandensis	Soil	Saprophyte
		Agaricus bernardii	Soil	Saprophyte
		Agaricus trisulphuratus	Soil	Saprophyte
		Agaricus endoxanthus	Soil	Saprophyte
		Agaricus sp. 1	Soil	Saprophyte
		Agaricus sp. 2	Soil	Saprophyte
		Agaricus sp. 3	Soil	Saprophyte
		Heinemannomyces	Soil	Saprophyte
		splendidissimus	Soil	Saprophyte
		Clarkeinda trachodes	Dead wood	Saprophyte
		Leucocoprinus cretaceous	Soil	Saprophyte
		Chlorophyllum molybdites		
	Nidulariaceae	*Cyathus striatus*	Dead wood	Saprophyte
	Phallaceae	*Mutinus bambusinus*	Soil	Saprophyte
		Phallus atrovolvatus	Soil	Saprophyte
		Phallus indusiatus	Soil	Saprophyte
	Pleurotaceae	*Pleurotus* sp.	Dead wood	Saprophyte
	Hygrophoraceae	*Hygrocybe conica*	Soil	Saprophyte
		Hygrocybe sp. 1	Soil	Saprophyte
		Hygrocybe sp. 2	Soil	Saprophyte
	Canterellaceae	*Cantherellus* sp.	*S. saman*	Mycorrhiza
	Sclerodermataceae	*Pisolithus* sp.	*S. saman, A.*	Mycorrhiza
		Scleroderma siannamariense	*mangium*	Mycorrhiza
		Scleroderma sp. 1	*Hibiscus rosa-*	Mycorrhiza
		Scleroderma sp. 2	*sinensis*	Mycorrhiza
			Unknown tree	
			C. camphora	
	Boletaceae	*Tylopilus* sp.	*A. mangium*	Mycorrhiza
	Tricholomataceae	*Clitocybe nuda*	*S. saman*	Mycorrhiza
	Inocybaceae	*Inocybe rimosa*	*C. camphora*	Mycorrhiza
		Inocybe sp.	*C. camphora*	Mycorrhiza

	Diplocystidiaceae	*Astraeus hygrometricus*	*C. camphora*	Mycorrhiza
	Russulaceae	*Russula japonica*	*C. camphora*	Mycorrhiza
		Russula sp. 1	*C. camphora*	Mycorrhiza
		Russula sp. 2	*C. camphora*	Mycorrhiza
		Russula sp. 3	*S. saman*	Mycorrhiza
	Ganodermataceae	*Ganoderma* sp. 1	Unknown tree	Parasite
		Ganoderma sp. 2	Unknown tree	Parasite
TOTAL	**26**	**62**	**-**	**-**

Pisolithus sp., which highly resembled *P. arhizus*, was found under two different host plants, which were *Acacia mangium* and *Elaeis guineensis*. Dating back, a species from the genus *Pisolithus* known as *P. aurantioscabrosus* was discovered in Negeri Sembilan under *Shorea macroprera* (Martin et al., 2002; Lee et al., 2012) and on the soil of Tasik Bera. Another *Pisolithus* species, *P. adbitus,* was discovered in Thailand under *Dipterocarpus alatus* (Kanchanaprayudh et al., 2003). Later, *P. indicus* was found to be associated with *Vateria indica*, a member of the family Dipterocarpaceae in a dipterocarp native forest in the Western Ghats of India (Reddy et al., 2005). It is widely known that species in the genus *Pisolithus* develop ectomycorrhizal connections with a variety of woody plants and is a global species that can be found in both tropical and temperate climates with a relatively broad host range (Reddy et al., 2005). Meanwhile, *A. hygrometricus* (Figure 3K), also a newly recorded species, formed a symbiosis with *Cinnamomum camphora*. The species was not far from *Russula* sp. 1 (Figure 3J), *Inocybe rimosa* (Figure 3L), and *Inocybe* sp. The interactions between the tree species and mycorrhizal fungi are unknown and warrant further investigations. Prior to this study, a smaller number of mycorrhizal fungi was expected, probably because the types of trees planted in UiTMCPh did not require interactions with ectomycorrhizal macrofungi. Adding on, geographical locations and climatic factors, including height, temperature, and rainfall, have an impact on ectomycorrhizal fungal diversity (Helbert et al., 2019). Among these mycorrhizal fungi, *Chanterellus* sp. (Figure 3I) was the least common species and infrequently found, of which the species was sighted only once throughout the study period. Constant soil disturbance and climate change may have affected its survivability at the site.

Parasitic fungi were the least common species recovered in this study, with only two species (5%), which were *Ganoderma* sp. 1 (Figure 3O) and *Ganoderma* sp. 2. Temperature seasonality, host abundance, and make-up of host species assemblages are among the factors that affect the diversity of parasitic fungal species in an area (Vacher et al., 2008). Parasitic fungi also require only specific nutrients on specific trees to grow. Therefore, a range of hosts has restricted the diversity of these types of fungi (Lodge et al., 1995). In general, *Ganoderma* species have a hybrid lifestyle of biotrophs and necrotrophs. First, the pathogenic species goes through a biotrophic phase in which the species initiates colonisation and infection on the intact host plant cells. Biotrophs focus on keeping their host cells intact for food intake (Bahari et al., 2018). The species then continues to the necrotrophic phase, where the cell wall of the plants is severely degraded, allowing the fungi to infect and survive saprotrophically (Chong et al., 2017; Bahari et al., 2018). Eventually, the infected trees died. The genus *Ganoderma* comprises more than 80 species worldwide. Many of these species are native to tropical climates (Kirk et al., 2008). *Ganoderma* species are the main causal agents of basal stem rot (BSR), a devastating disease in Southeast Asian (SEA) oil palm plantations, mainly in Malaysia and Indonesia (Paterson, 2019; Shokrollahi et al., 2021). The disease is caused by *G. boninense*, a white-

rot fungus that has been threatening the oil palm industry for over eight decades with no prominent cure (Paterson, 2019; Shokrollahi et al., 2021). In the present study, the hosts of *Ganoderma* species were not palm oil trees. Instead, the trees were *A. mangium* and an unidentified ornamental tree planted along the avenue of the UiTM Pahang campus. The first host died naturally due to infection by *Ganoderma* sp. 1, whereas the latter was mechanically removed for safety reasons because of its poor condition after being severely infected by *Ganoderma* sp. 2.

Twenty-six specimens in this study could not be identified up to the species level. Identifying macrofungi at the species level by morphology alone is challenging and a daunting task due to their broad characteristics and the high similarities they share. Furthermore, field guides and monographic publications for tropical macrofungi are scarce. Hence, the delineations of the fungal species could not be well executed. Molecular approaches using specific genes and regions such as ITS, TEF-1α, and LSU, to name a few, seem more reliable in identifying the specimens up to the species level. In addition, molecular approaches are rapid, uncomplicated, and less laborious than morphology-based identification. Such approaches would provide ample results and should be employed in the future.

Figure 3. Diversity of macrofungal species collected from selected locations in UiTM Pahang campus. **A**. *Cookeina sulcipes*. **B**. *Cyathus striatus*. **C**. *Marasmius haematocephalus*. **D**. *Calvatia cyathiformis.* **E**. *Trametes* sp. 1. **F**. *Filoboletus manipularis*. **G**. *Chlorophyllum molybdites*. **H**. *Geastrum triplex*. **I**. *Chanterellus* sp. **J**. *Russula* sp. 1. **K**. *Astraeus hygrometricus*. **L**. *Inocybe* sp. **M**. *Scleroderma* sp. 1. **N**. *Tylopilus plumbeoviolaceus*. **O**. *Ganoderma* sp. 1

CONCLUSION

A total of 62 macrofungi from two major phyla, Ascomycota and Basidiomycota, were obtained. They played three different ecological roles, namely saprophytes, mycorrhiza, and parasites, where saprophytes recorded the highest number of species (46 species), and parasites showed the least number of species, with only two species. Several trees formed mutual symbiotic relationships with several macrofungi, as 14 mycorrhizal fungi were found. The findings of this study demonstrate that UiTMCPh is a good habitat for numerous groups of macrofungi and a highly favoured habitat for saprophytic macrofungi. Although parasitic fungi within the campus area comprised only two species, concern should be given to both species because they are parasites, and their presence is considered harmful and threatening to the trees on the campus.

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AUTHOR CONTRIBUTIONS

Nur Amirah Sakinah Mat Shah Fee: Original content provider; **Nur Amalina Mohd. Izam**: Specimen collection; **Farah Ayuni Farinordin**: Specimen collection; **Nurul Farizah Azuddin**: Data validation and formatting; **Hafizi Rosli**: Data validation; **Nor Azliza Ismail**: Manuscript preparation

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COMPETING INTEREST

The authors declare that there are no conflicts of interest regarding the publication of this paper.

COMPLIANCE WITH ETHICAL STANDARDS

Not applicable.

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