Keynote Speech

Fungi, an Overlooked Component of Sri Lanka's Biodiversity and Recent Advances in Mycology

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Abstract

Sri Lanka has a rich biodiversity but compare to other disciplines, understanding of fungi is less. Current studies on fungi mainly based on DNA sequences analyses thus it is important to revisit old species reported from Sri Lanka. And subject to morpho-molecular analyses. Missing species of fungi are predicted to be mainly in tropics, biodiversity hotspots and little-explored habitats. Hence, it is essential to implement collecting programs and carry out analyses. This type of studies will be helpful to establish correct generic boundaries, resolve species complexes and polyphyletic taxa, provide taxonomic placements. It is essential to update the existing checklists according to recent name changes (Art. 59.1). Taxonomists are encouraged to deposit putative cultures at culture collections and deposit herbarium specimens at least two well-known Fungaria. Establishing and maintaining a culture collection in Sri Lanka is also necessary while implement new collaborative programs with other leading scientific groups in other countries. Cultivation and domestication of wild edible mushrooms can be promoted after comprehensive survey.

Introduction

Fungi are one of the most diverse groups of eukaryotes and widely distributed worldwide. They occur as different life modes (*viz.* saprobes, pathogens (animal and plant), endophytes, lichenicolous, fungicolous, insectivorous etc.) and play important roles in different ecosystems such as mineral cycling, decomposing litter, causing diseases etc. Furthermore, some fungi are used in industries including bakery, pharmaceutical, and bioremediation. Approximately, 144,000 species of fungi have been identified (Willis 2018), but Hawksworth and Lücking (2017) estimated that it would be between 2.2 - 3.8 million species on earth. Therefore, only 3%-8% species are known to science (Hawksworth and Lücking 2017). Wijayawardene et al. (2020) accepted 19 phyla in kingdom fungi. This result was based on current knowledge comprising traditional, morphology based classification and modern DNA based classification. Recent studies by Tedersoo et al. (2018), Wijayawardene et al. (2018, 2020) accepted Rozellomycota (including Microsporidia) as a new lineage in fungi. Some groups which was treated as fungi in traditional taxonomy, have been excluded from kingdom fungi based on DNA based phylogenetic studies (e.g. Myxomycota).

Understanding of floral and faunal diversity in Sri Lanka is higher than of fungi and other microorganisms. 'Information available on Sri Lankan fungi is scattered. There are difficulties in estimating exact numbers due to synonyms used and duplicate entries' (Karunarathne et al. 2012) and lacking sequence data, proper protologue, herbarium deposits, cultures and collection details. There were not any significant surveys been carried out except for plant pathogenic fungi (Adikaram et al. 2001, 2013) and macro fungi (Karunarathna et al. 2011). However, still a large number of species has to be discovered based on both morpho-molecular analyses.

Identifying the species

In traditional taxonomy, species have been identified and described based on only morphological characters (Sutton 1980; Sivanesan 1984). Thus, morphological characters such

as ascomata, asci and ascospores in sexual fungi, conidiomata, conidiophores, conidiogenesis and conidia in asexual fungi have been mainly used to describe a species. However, since 1990, DNA based taxonomic studies play the vital role in mycology (White et al. 1990) along with morphological studies. Currently, mycologists widely use DNA sequences to establish new species, resolve species complexes and polyphyletic taxa, establish generic boundaries, provide classification and link sexual asexual morphs. Moreover, epitypification of old species is another hot topic which is popular among mycologists.

However, all the significant studies carried out in Sri Lanka are mainly based on morphological characters (Berkeley and Broome 1871, 1873, 1877; Petch 1906, 1909, 1910, 19121, b, 1916, 1917, 1918, 1919, 1920; Petch and Bisby 1950; Coomaraswamy 1979, 1981; Coomaraswamy and De Fonseka 1981; Coomaraswamy and Kumarasingham 1988). Almost all these taxa preserved as only herbarium materials and provided drawings (mainly for mushrooms). Thus, revisiting old species is a challenge as some herbarium materials are in poor condition or lost. Moreover, for the identification and classification of asexual fungi, taxonomists strongly suggest to use DNA sequences (Wijayawardene et al. 2019) as they show morphological plasticity. Thus, recollecting old species is important to determine whether those species were identified correctly.

Furthermore, some pathogenic species have been revealed as species complexes (i.e. morphologically similar but phylogenetically distinct) 'which means that species differentiation is problematic when relying only on morphological characters' (Phillips et al. 2008, 2012; Wijayawardene et al. 2017b) (e.g. *Colletotrichum acutatum fide* Damm et al. 2012; *Colletotrichum destructivum fide* Damm et al. 2014). Thus, it is important to recollect pathogenic species reported from Sri Lanka and carry out multi-gene phylogenetic analyses to determine the correct species. In quarantine, this type of study will be a critical step to update the checklists of pathogenic species.

DNA based classification

Since DNA sequences are the main source for phylogenetic and evolution studies, mycologists try to generate cultures from taxa they study. Single spore isolation is the most popular technique used by mycologists to isolate saprobes and pathogens (Chomnunti et al. 2014). Putative cultures are used to extract DNA. Different protocols are used to extract DNA from the mycelia (e.g. Kit). PCR technique is used to amplify the extracted DNA fragments and products are utilized for sequencing.

Generated sequences are widely used to determine the phylogenetic placements of the taxa. This is the best and most reliable method to clarify the classification of asexual genera. In traditional taxonomy, asexual fungi have been placed in subdivision Deuteromycotina (Ainsworth 1966). However, this classification is highly artificial thus DNA based studies are essential to reveal the correct taxonomic placements (Verkley et al. 2004; de Gruyter et al. 2009, 2013; Wijayawardene et al. 2016). Moreover, a large number of asexual genera are listed as 'orphan genera' (i.e. without family or higher ranks) in Wijayawardene et al. (2017a). Therefore, re-collecting old species is an important topic among mycologists.

As the known historic taxa from Sri Lanka lacking sequence data, it is highly recommended to recollect and subject to carryout DNA based phylogenetic studies. Some taxa are morphologically similar but polyphyletic in phylogeny (Wijayawardene et al. 2014b). Hence, without DNA sequence analyses, it is not a wise decision to determine the familial placements.

Nomenclature

Saccardo (1904) proposed the dual system of fungal nomenclature (i.e. asexual and sexual morphs with different names) 'as a solution to the confused situation of asexual and sexual

morphs faced by taxonomists at the time' (Wijayawardene et al. 2014a). However, mycologists link sexual-asexual morphs based on the culture studies, occurrence of taxa on same host. Nevertheless, DNA based taxonomic studies of pleomorphic genera, provide accurate and reliable links between two morphs (or three morphs when presence of synasexual morphs). Since, two taxa are linked, it was proposed to end the dual system of fungal nomenclature (Hawksworth 2012). Several studies with the involvement of mycologists, pathologists, proposed one name for pleomorphic genera (Rossman et al. 2013, 2015a, b, 2016; Wijayawardene et al. 2014; Réblová et al. 2016).

It is important to update the current lists of fungi in Sri Lanka after the name changes. Most of the taxa important in quarantine are subjected to name changes thus it is essential update (e.g. *Phomopsis* was suppressed under *Diaporthe*).

Importance of promoting mycological studies in SL

Sri Lanka has been listed as a biodiversity hotspot but compare to the studies in other disciplines, the number of researches in mycology are less in number. Hawksworth and Lucking (2017) suggested to carry out more research works in tropical regions and biodiversity hotspots to reveal missing species. Moreover, Hawksworth and Lucking (2017) concluded that plant: fungi ratio as 1: 8.1. Currently there are 3154 flowering plants and 366 ferns have been recorded (Wijesundara pers. com.) thus approximately 28,568 fungal species could be reported. However, only c. 2000 species are known (Karunarathe et al. 2012). Hence it is essential to carry out more studies to reveal missing taxa from different habitats. Some life modes such as fungicolous, lichenicolous have not been properly studied thus it is important to implement new researches to identify these species as well.

Domestication of wild mushrooms is one of the most popular topics among mycologists who are studying mushrooms in tropical regions. Gathering the knowledge of local people on wild edible mushrooms and experimenting to cultivate them in farms can be promoted in Sri Lanka. This type of projects need to be collaborated with experts in other countries.

Limitations and overcome them

Current mycological studies highly depend on DNA sequences analyses. Hence it is essential to get pure cultures from or extract DNA direct from the fruiting structures (i.e. ascomata or conidiomata). However, obtaining DNA from cultures is highly recommended for cultivable species and deposit the cultures at reputed culture collections. Moreover, depositing herbarium materials with the fungal species at well-known Fungaria is also mandatory. However, these practices (such as sending cultures abroad and sending specimens) are restricted from the legal background in Sri Lanka thus it is essential to establish culture collections in the island. And implementing a collaborative program with other scientific communities or institutes to exchange knowledge is also important.

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