
Diversity of Macrofungi (Mushroom) in the Fambong Loh Wildlife Sanctuary, Sikkim

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Fambong Loh Wildlife Sanctuary, Sikkim is an unexplored reserve forest when it comes to the diversity study on macrofungi. The present survey was carried with the background of ethno-mycological knowledge. The collected species were segregated into edible and medicinal and non-edible or unknown groups. Approximately 70 fungal samples belonging to 49 genera and 28 families were recorded, with division Basidiomycota constituting 94% and Ascomycota 6 % of the recorded species respectively, which was morphologically identified by following the standard keys and manuals. Few unknown samples needs further taxonomic characterization to report as new species. The present data will help in understanding the portion of fungal diversity common to a forest type and act as a baseline for potential future studies with respect to species richness, access forest health and ecological role. The overview idea is to reveal macrofungal diversity of Fambong Loh Wildlife Sanctuary and to create database for forest management for sustainable utilization of resource and to further explore on culturable species with economic value. The present study contributes in the existing situation of the wild mushrooms of Sikkim and putting emphasis on the ecological effects and sustainable forest management. The results could be a valuable document for the scientific community in domestication or cultivation of wild mushroom for nutritional and medicinal purpose in future.

Key words: Ascomycota, Basidiomycota, diversity, ectomycorrhiza, fungi, mushroom, Sikkim.

INTRODUCTION

Fungi are the second most species-rich organism group after the insects (Purvis 2000); hence, it is more challenging to complete the global fungal inventory, as compared to other organisms such as plants, more so because of its cryptic nature. An updated estimate of fungal diversity showed that the fungal species ranged from 2.2 to 3.8 million worldwide (Hawksworth and Luecking 2017). The estimates of 2.2 and 3.8 million fungal species could be described by the years 2210 and 2245, respectively. Macrofungi are the fungi that form macroscopic fruiting bodies, such as gilled fungi, jelly fungi, coral fungi, stinkhorns, bracket fungi, puffballs and bird's nest fungi, relatively easy to spot and identify (Richards and Murray 2002). This group of fungi has the longest history of diversity studies of any mycota. Macrofungal studies have interested scientists as well as for the general public due to their important role in human welfare, as

they serve as food, medicine, biocontrol agents, producers of bioactive compounds used in the pharmaceutical and many other industries (Ozturk *et al* 2003, Duarte *et al.*, 2006, Demirbas 2000; Kalac *et al.*, 2004). In folk medicine, history of using many different types of fungal extracts demonstrating immunostimulatory, anti-inflammatory and anticancer activity dates back to ancient Japan, China and other countries of the Far East. They produced more than 100 medicinal functions like antioxidant, anticancer, antidiabetic, antitumor, antiinflammatory, antiallergic, immunomodulating, cardiovascular protector, anticholesterolemic, antiviral, antibacterial, antiparasitic, antifungal, detoxification, and hepatoprotective effects (Chang andWasser,2012; Panda and Tayung, 2015).

Their role in the ecosystem as decomposers is well established (Deacon, 2006); but as mutualists and pathogens, their role still needs further clarity (Schmit and Mueller 2007). In the past, a parasitic fungus has been looked upon as biologically evil,

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but this view is rapidly changing as science progresses. Some species causing disease in plants produce minute quantities of medically significant bioactive compounds like anti-carcinogen Taxol, a proven treatment for breast cancer. A well-accepted theory proposes that fungi were instrumental in the evolution of the root system in plants and contributed to the success of Angiosperms. Plants with mycorrhizal fungal association can also resist diseases far better than those without. Most ecologists now recognize that a forest's health is directly related to the presence, abundance, and variety of mycorrhizal associations. Mycologists continue to unravel the unexplored and fascinating fungal biodiversity as many macro fungi are becoming extinct or facing threat of extinction because of habitat destruction and global climate change (Swapana *et al.*, 2008).

The state of Sikkim is situated in the Eastern Himalayan region, major biodiversity hotspots of the Indian subcontinent. The area experiences a heavy rainfall due to its proximity to the Bay of Bengal, average annual rainfall varies from 2000 mm at valleys to 4000 mm at the mountain ridges and humidity remains very high during the rainy season (85-97%). Fambong Lho Wildlife Sanctuary is situated in the East district of Sikkim which is around 52 sq km, with an altitudinal range between 1524-2749 m representing temperate biotic zone. Therefore macrofungi by establishing ectomycorrhizal associations with the forest trees are in abundance in this region. Since, mushrooms have ethnomycological uses, it is crucial to document and study the diversity, ecology of macrofungi of this region. Considering the importance of creating a databank on the naturally occurring organisms, including fungi, at the genomic, taxonomic and ecological levels, the work on diversity, taxonomy and ecology of the fungi will be the valuable contribution as very few studies has been carried in the region. With aim of producing a checklist of macrofungi for the area and also to document the traditional knowledge of mushrooms in the communities surrounding the reserve forest the present study of documentation and identification of mushrooms was undertaken.

MATERIALS AND METHODS

Study site

Fambong Lho Wildlife Sanctuary (FWS) sharing its boundaries from east to north and south district

of Sikkim lies between 27.3314° N latitude, and 88.6138° E longitude.

As the area is unexplored and blessed with diverse flora, fauna and probably millions of microbes. The sanctuary is very rich in natural vegetation and comprises of various dense forests with subtropical and temperate type. Some of the common tree species are *Symplocos*, *Castanopsis*, *Acer*, *Quercus*, *Machilus*, *Cinnamomum*, *Cupresses*, *Alnus*, *Cryptomeria*, *Engelherdia*, *Schima*, *Eurya*, *Viburnum*, *Michelia*, *Magnolia*, *Elaeocarpus*, *Abies*, *Pinus*, *Rhododendron*, *Betula*, *Juglans* etc. The sanctuary buffer zone approximately 9578 ha is highly influenced by agricultural activity in the lower reaches, but inaccessible slopes have dense mixed vegetation and also rich in diversity of ferns, mosses, orchids, bamboo, others herbs and shrubs. The FWS is a highly critical wildlife reserve because of its close proximity to human habitation and hydro power projects. In 1984 the Fambong Lho Wildlife Sanctuary was declared as Protected Area under Wildlife Protection Act, 1972 and was assigned to the Divisional Forest Officer of North and East district of Wildlife Division for overall management and development (Fig.1).

Macrofungal sampling and Identification

Sample collections were done frequently during the rainy season between (2018-2019) as during this time the humid and more continuous rain favours growth of diverse macrofungi, some of which were sighted during dry seasons too. Macrofungi exhibit pattern of diversity that is related to largely to substratum and host availability. Fresh specimens were photographed in the field, along with the substrate on which they were found or growing. The macroscopic characters like shape, size, colour, texture, odour, attachment of stipe, smell, habit and habitat was documented during the survey by filling specific annotation sheet. The specimens were wrapped in the aluminium foil and boxes which offers a good protection and brought to the laboratory where they were stored at 4°C, until further analysis. Tissue culture of few saprobic fungal species was carried out from fresh samples for isolation on different agar based media. The soft textured specimens were preserved in 2% formaldehyde and leathery textured were preserved in 4% formaldehyde and some were oven dried and preserved as dry preservation stored in sealed plastic bags with dehydrated silica

gel at room temperature for molecular identification if needed.

Each of the mushrooms was morphologically characterized and identification was done using guide books, monographs and different authentic keys (Wu et al 2010). Several descriptions articles and literatures were also consulted. Some online sites have also been used: www.mushroomexpert.com, www.mushroomobserver.com and www.mycobank.com.

Molecular characterization

The widely followed traditional morphologically based identifications have the following disadvantages; many of the macrofungi may appear to have similar morphological features, and thus, identifications based on a field guide become difficult for some samples. The DNA-based identification of macrofungi targeting the non-coding internal transcriber spacer region (ITS) of nuclear ribosomal rRNA, rapid identification of macrofungi down to the genus and species levels is possible (Osmundson et al 2013). The nucleic acid samples extracted from dried mushroom tissues by CTAB method. The amplified PCR products were differentiated using 2% agarose gel electrophoresis alongwith a ladder of 100bp and same PCR products were sequenced and confirmed from blast search of NCBI database for identification of fungi. For PCR amplification of the ITS locus, the primer pairs ITS4 and ITS5 were used and the primer sequences (ITS4 5'TCCTCCGCTTATTGATATGC3' and ITS5 5'GGAAGTAAAAGTCGTAACAAGG3') were those described by (White et al.1990). PCR products were sequenced by the SciGenom, Cochin, Kerala, India.

Ethnomycology

In order to generate baseline information, ethnomycological information was gathered from local people residing in the vicinity by semi-structured interview and also by showing the pictorial presentations. The utilization of different mushroom species for food and as medicine have been documented and confirmed by available literature; (Chang 2004, Das 2010, Wani et al.2010, Sharma and Atri 2014, Wangdi 2019).

RESULTS AND DISCUSSION

Diversity of Macrofungi

Species diversity in the natural environment of a region is one of the basic requirements to estimate the richness of species. The previous record on the survey of naturally occurring mushroom of Sikkim Himalayas listed 131 species and provided the taxonomic rank and nomenclature to 90 of them. In the present study, the sampling and collection was done throughout the year. Highest number of species were found during summer or rainy season, i.e between May to September, and frequently observed species are *Laetiporus*, *Termitomyces*, *Auricularia*, *Cyathus*, *Macrolepiota*, *Trametes*, *Pycnoporus*, *Aminita*, *Coltricia*, *Fistulina*, *Mycena*, *Polyporus*, *Xylaria*, *Boletus*, *Lycoperdon*, *Dacrypinax*, *Russula*, *Stereum*, *Lentinus*, *Xerocomellus*, *Maramillus*, *Termellia* etc. The species frequently found during autumn season between October-November are *Schizophyllum*, *Lycoperdon*, *Xylaria*, *Dedaleopsis*,

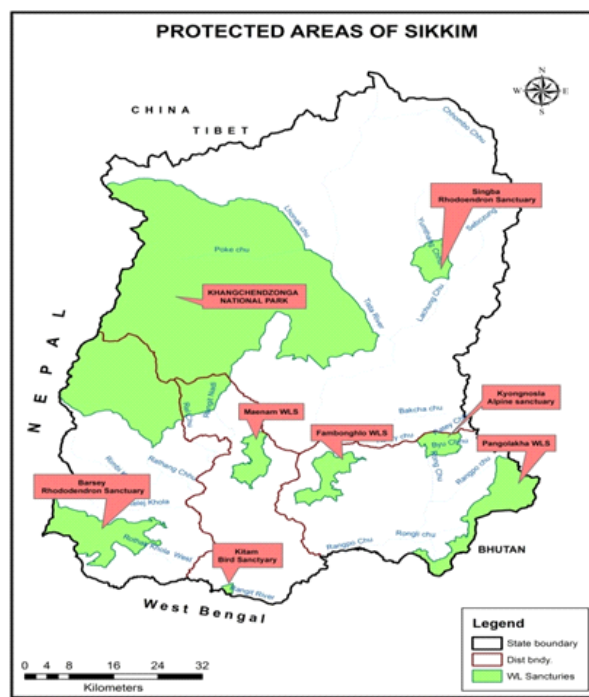


Fig. 1 : Study area Fambongluo Wildlife Sanctuary, Sikkim, India. *Stereum*, *Coprinellus Auricularia*, *Pleurotus Clavulina*, *Oligoporus*, *Trametes*, *Hypholoma*, *Ganoderma*, *Scleroderma* etc. In the winter i.e between December- February, species like *Porodaedalea*, *Fomes*, *Phellinus*, *Flammulina*, *Maramius*, *Cuprinellus* are common and lastly the species observed during spring between March-April are *Parasola*, *Morchella*. But *Polyporus* and few bracket or conks like *Fomes*, *Ganoderma*,

Pycnoporus can be seen throughout the seasons as their bodies are tough. In the current study, 70 species of macrofungi belonging to 49 genera and 28 families were recorded so far from Fambong Loh Wildlife Sanctuary, Sikkim; which belongs to the division Basidiomycota (94 %) and Ascomycota (6 %) respectively (Table 1 and 2).

Most of the mushroom samples were identified with the help of morphological studies following standard manuals, research articles and keys. But for some species the molecular identification was felt necessary for confirmation and identification up to the species level. The species identified through molecular characterization includes; *Polyporus guianensis*, *Grifola frondosa*, *Auricularia cornea*, *Morchella crassipes*, *Trametes sanguine*, *Auricularia cornea* and *Lentinula edodes* (Fig.2).

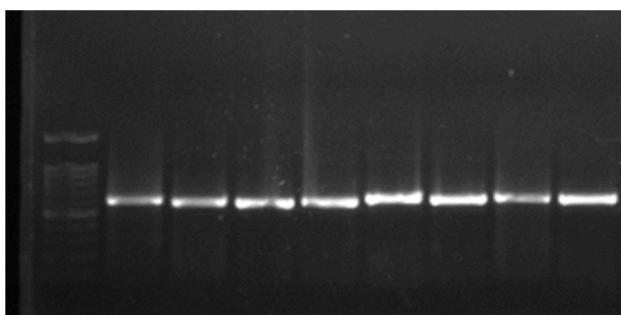


Fig. 2 : Gel electrophoresis of PCR products of 08 mushroom species

Ecological role

Most of the macrofungi growing on the forest floor are intimately linked to trees by **sympiosis**, they benefit each other by exchanging of nutrients. Most of the higher plants form mycorrhizal association with either endomycorrhizal or ectomycorrhizal fungi. In general, the macrofungus helps the tree to extract minerals and water from the soil; in exchange, the tree supplies the macrofungus with sugar compounds (carbohydrates). They cannot function independently, survival and conservation of both is significant for healthy forest ecosystem (Bonet, 2004). About 95 percent of the tree species occurring in tropical forests are purely endomycorrhizal. In the present study, the specimens were mostly belongs to saprophytic group, they are significant as wood decomposer and helps in nutrient cycling in forest ecosystem. The species like *Schizophyllum commune*, *Laetiporus sulphureus*, *Ganoderma lucidum*, *Trametes gibbose*, *T. versicolor*, *Porodaedalea*

pini, *Polyporus* spp., *Pholiota*, *Xylaria hypoxylon*, *Fistulina hepatics*, *Auricularia* spp., *Stereum* spp. are common. The Ectomycorrhizal (ECM) species like *Boletus* spp., *Russula* spp, *Lactarius* spp, *Lycoperdon* spp, *Xerocomellus* sp, *Clavulina* sp are found to be associated with tree species, such as *Castanopsis*, *Acer*, *Quercus*, *Machilus*, *Cinnamomum*, *Alnus*, *Cryptomeria*, *Schima*, *Viburnum*, *Michelia*, *Magnolia*, *Abies*, *Pinus* and parasitic fungi like *Asterophors parasitica* on decaying mushrooms was observed. Most dominant family was found to Polyporaceae with 24% representation from the total observed samples. The morphology of some selected mushrooms is depicted in Fig.3.

The industrial or suburban development threatens entire forests, and is unavoidable. But foresters may consider establishing of young seedlings in deserted landscape by inoculation with spore mass slurry of native mycorrhizal fungal species as they are more likely to adapt to the region than exotic species. This method has had the longest tradition of success in Europe. Therefore, understanding the diversity of species composition and richness in forest ecosystems is crucial for ecologists, biogeographers, conservationists, and forest managers for planning successful forest management policies.

Ethnomycology

From the focus group discussion and information obtained from the questionnaire and pictorial presentations, it was realized that many local communities were familiar with mushroom and its uses as food and medicine and for mythological purposes. The tree ethnic tribes of the state, the Lepcha, Bhutia and Nepali, generally calls mushroom in their native language as *Dobri*, *Syamo*, *Chew* respectively, with specific sub-tribe names, generally are named related to the texture and shape of the mushroom in their native dialect. The mushroom are generally consumes as a food and ingredient for medicinal uses. Most of edible mushroom like *Pleurotus ostreatus*, *Lentinula edodes*, *Pholiota microspora*, *Russula* spp, *Macrolepiota procera*, *Morchella* sp, *Fistulina hepatica*, *Termitomyces* spp, *Grifola frondosa* are freshly collected and consumed usually substituted for animal protein as side dish and some species like, *Tramella fuciformis*, *Laetiporus sulphureus*,

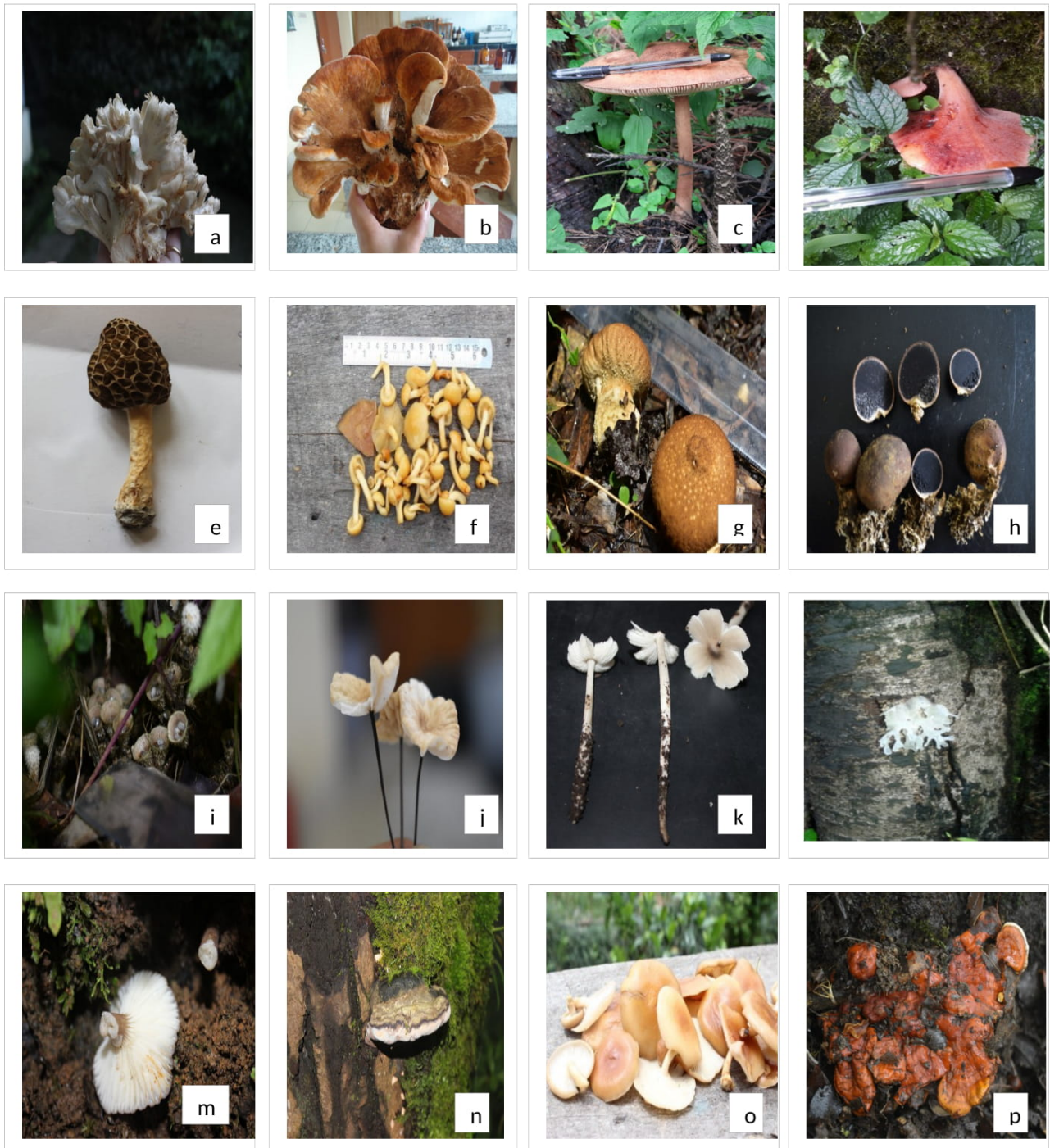


Fig. 3 : (a) *Grifola frondosa* (Dicks.) Gray (b) *Laetiporus sulphureus* (Bull.) Murrili (c) *Macrolepiotaprocera* (Scop.) Singer (d) *Fistulina hepatica* (Schaeff). (e) *Morchella crassipes* (L.) Pers. Fr (f) *Pholiota nameko* (Berk) Sacc. (g) *Lycoperdon perlatum* Pers, (h) *Scloderma citrinum* Pers (i) *Cyathus striatus* (Huds.) Wild. (j) *Maramius epiphyllus* (Pers.) Fr. (k) *Termitomyces medius* R. Heim & Grasse (l) *Tremella fuciformis* Berk (m) *Lactarius piperatus* (L.)Roussel (n) *Fomes fomentarius* Fr (Lr) (o) *Lentinula edode* (Berk.)Pegler (p) *Ganoderma lucidium* Karst.

Auricularia spp., *Ployporus guianensis* are sun dried and consumed during lean period. For the medicinal purposes, usually the bracket fungi like *Trametes* spp, *Laetiporus* sp., *Ganoderma* sp. *Pycnoporus* sp *Porodaedalea* sp were use as an ingredient by herbal medicine practitioners. The

traditional utilization knowledge they claimed that it was inherited from their elders in the community. The locals could distinguish between edible and non edible mushrooms and mushroom gathering is an important economic activity during the

Table 1 : Checklist of Macrofungi from Fambong Loh Wildlife Sanctuary, Sikkim, India.

SI no.	Scientific name	Family	SI no.	Scientific name	Family
Division Basidiomycota					
1.	<i>Clavulina rugosa</i> (Bull.) J. Schrot.	Clavulinaceae	2.	<i>Tremella fuciformis</i> Berk	Tremellaceae
3.	<i>Porodaedalea pini</i> (Brot.) Murrill	Hymenochaetaceae	4.	<i>Polyporus arcularis</i> (Batsch) Fries	Polyporaceae
5.	<i>Scleroderma citrinum</i> Pers.	Sclerodermataceae	6.	<i>Megacollybia platyphylla</i> (Batsch) Fries	Tricholomataceae
7.	<i>Mycena maculata</i> P. Karst.	Mycenaceae	8.	<i>Stereum complicata</i> (Fr.) Fr	Stereaceae
9.	<i>Oligoporus caesius</i> (Schrad.) Gilb. & Ryvarden	Fomitopsidaceae	10.	<i>Stereum hirsutum</i> (Wild.) Pers.	Stereaceae
11.	<i>Mycena inclinata</i> (Fr.) Quel.	Mycenaceae	12.	<i>Lycopedron perlatum</i> Pers.	Agaricaceae
13.	<i>Amanita constricta</i> Thiers & Ammirati	Amanitaceae	14.	<i>Phellinus</i> spp.	Hymenochataceae
15.	<i>Flammulina velutipes</i> (Curtis) Singer	Physalacriaceae	16.	<i>Macrolepiota procera</i> (Scop.) Singer	Agaricaceae
17.	<i>Trametes versicolor</i> (L.) Lloyd	Polyporaceae	18.	<i>Polyporus guianensis</i> Mont.	Polyporaceae
19.	<i>Asterophora parasitica</i> (Bull. ex Pers.) Singer	Lyophyllaceae	20.	<i>Crepidotus mollis</i> (Schaeff.) Staude	Inocybaceae
21.	<i>Hypholoma fasciculare</i> (Huds.:Fr) P. Kumm.	Strophariaceae	22.	<i>Auricularia cornea</i> Ehrenb.	Auriculariaceae
23.	<i>Fomes fomentarius</i> Fr (Lr)	Polyporaceae	24.	<i>Stereum sanguinolentum</i> (Alb. & Schwein)	Stereaceae
25.	<i>Ganoderma lucidium</i> Karst	Polyporaceae	26.	<i>Craterellus tubiformis</i> (Fr.) Quel	Cantharellaceae
27.	<i>Pycnoporus coccineus</i> (Fr.) Bondartsev & Singer	Polyporaceae	28.	<i>Russula cyanoxantha</i> (Schaeff.) Fr.	Russulaceae
29.	<i>Trametes hirsute</i> (Wulfen) Lloyd	Polyporaceae	30.	<i>Hohenbuehelia augustata</i> (Berk.) Sing.	Pleurotaceae
31.	<i>Pycnoporus canabarinus</i> (Jacq.) P. Karst	Polyporaceae	32.	<i>Boletus</i> spp.	Boletaceae
33.	<i>Parasola auricoma</i> (Pat) Redhead, Vilgalys & Hopple	Psathyrellaceae	34.	<i>Xerocomellus zelleri</i> (Murrill) Klofac	Boletaceae
35.	<i>Laetiporus sulphureus</i> (Bull.) Murrill	Polyporaceae	36.	<i>Tyromyces chioneus</i> (Fr.) P. Karst	Polyporaceae
37.	<i>Schizophyllum commune</i> Fries	Schizophyllaceae	38.	<i>Crepidotus</i> sp.	Crepidotaceae
39.	<i>Russula natarajanii</i> K. Das, J. R. Sharma & Atri	Russulaceae	40.	<i>Cyathus striatus</i> (Huds.) Willd.	Nidulariaceae
41.	<i>Trametes sanguine</i>	Polyporaceae	42.	<i>Sutorious luridiformis</i> (Rostk) Gelardi, Simonini & Vizzini	Boletaceae
43.	<i>Auricularia delicata</i> (Fr.) Henn.	Auriculariaceae	44.	<i>Fistulina hepatica</i> (Schaeff.) With.	Fistulinaceae
45.	<i>Stereum ostrea</i> <i>Stereum ostrea</i> (Blume & T. Nees) Fr	Stereaceae	46.	<i>Lactarius piperatus</i> (L.) Roussel	Russulaceae
47.	<i>Termitomyces medius</i> R. Heim & Grasse	Lyophyllaceae	48.	<i>Trametes gibbosa</i> (Pers.) Fr	Polyporaceae
49.	<i>Dadaleopsis confrosa</i> J. Schrot	Polyporaceae	50.	<i>Dacrypinax spathularia</i> (Schwein) G. W. Martin	Dacrymycetaceae
51.	<i>Trametes ochracea</i> (Pers.) Gilb. & Ryvarden	Polyporaceae	52.	<i>Marasmiellus candidus</i> (Fr.) Singer	omphalotaceae
53.	<i>Lentinus strigosus</i> Fr.	Polyporaceae	54.	<i>Auricularia -auricula judae</i> (Bull.) Quel	Auriculariaceae
55.	<i>Coltricia cinnamomea</i> (Jacq.) Murrill	Hymenochaetaceae	56.	<i>Grifola frondosa</i> (Dicks.) Gray	Meripilaceae
57.	<i>Maramius epiphyllus</i> (Pers.) Fr.	Maramiaceae	58.	<i>Pholiota microspora</i> (Berk.) Sacc.	Strophariaceae
59.	<i>Lentinus crinitus</i>	Polyporaceae	60.	<i>Lentinula edodes</i> (Berk.) Pegler	Omphalotaceae
61.	<i>Laccaria sp</i>	Hydnanginaceae	62.	<i>Ganoderma applanatum</i>	Ganodermataceae
Division Ascomycota					
63.	<i>Trichocoma paradoxa</i> Jungh	Trichocomaceae	64.	<i>Morchella crassipes</i> (L.) Pers. Fr	Morchellaceae
65.	<i>Xylaria hypoxylon</i> (L.) Grev.	Xylariaceae	66.	<i>Xylaria polymorpha</i> (Pers.) Grev.	Xylariaceae

Table 2 : Morphological description of selected macrofungi of FWS

Scientific name of Mushroom	Substrate/ Season	Description
<i>Clavulina rugosa</i> (Bull.) J. Schrot	Growing in cluster to scattered on growd below deciduous tree. August to December	Fruiting body long, slender or branches with blunt ends, rugged, white when young and yellowish with age.
<i>Porodaedale pini</i> (Brot.) Murril	Growing on hardwood. Throughout the year	Fruiting body fan-shaped, crasty, woody, upper surface velvety to hairy, lower surface dull with presence of concentric rings, reddish brown to dark brown.
<i>Oligoporus caesius</i> (Schrad.: Fr.) Gilbn. & Ryv.	Growing on decaying wood. Summer to fall	Fruiting body fan shaped, whitish to blue gray tint, hairy to glabrous
<i>Trichocoma paradoxa</i> Jungh.	Growing on decaying stump. Whole year	Fruiting body appears like shaving brush, sessile, non-cleistotheical stromata, spore pinkish.
<i>Amanita constricta</i> Thiers & Ammirati	Growing on soil solitary under hardwood Summer to fall	Fruiting cap oval to broad convex. Presence of distinct volva, presence of universal veil tissue white to grayish colour.
<i>Trametes vesicular</i> (L.) Llyod	Growing on decaying wood trunk May to December	Fruiting body growing in overlapping form, fan shaped, leathery texture, presence of concentric zones on upper surface with black and white color, lower surface creamy color.
<i>Hypholoma fasciculare</i> (Huds.: Fr.) P. Kumm	Growing on decaying wood Fall to winter	Fruiting body convex to umbonate, dry, edge with partial black vial remnants. Gills attachment free, spore brown or purplish brown;
<i>Fomes fomentarius</i> (L.) J. J. Kickx	Growing trunk of deciduous tree Throughout the year	Fruiting body sessile, hoof shaped, woody surface with zonation, brown or blackish brown, hard flesh.
<i>Ganoderma lucidium</i> (Sheng H. Wu, Y. Cao & Y. C. Dai)	Growing on trunk or base of conifers and hardwood as well Throughout the year	Fruiting body bracket or kidney, covered with shiny crust, cap yellow to dark red with zonation, stipe.
<i>Pycnoporus canabarinus</i> (Jacq.) P. Karst.	Growing on dead fallen trunk Summer and Autum	Fruiting body bracket fan shaped, sessile, hard, dark reddish orange, with faint zonation.
<i>Laetiporus sulphureus</i> (Bull.) Murrill	Growing on deaying wood of Prunus sp. March to August	Fruiting body irregular trumpet to somewhat fan shaped, emerging from same point, smooth velvety fleshy in texture.
<i>Macrolepiota procera</i> (Scop.) Singer	Growing solitary nearby pine tree. May to August	Cap oval when young later broadly compentulate turning flat, bump on centre, with fibrillose, frayed edge, gills creamy white later rusty brown and tall stipe.
<i>Auricularia comea</i> (Ehmb. ex Fries) S	Growing on decaying wood May to july	Fruiting body ear shaped, with very minute hairs on outside and wrinkle in inner surface. Deark brown purple brown.
<i>Xylaria hypoxylon</i> (L.) Grev.	Growing on decaying tree stump/branch May to December	Fruiting body cylindrical to subcylindrical, often branched, black lower surface with white upper surface.
<i>Polyporus guianensis</i> Mont.	Growing on decaying tree May to July	Fruiting body bracket, growing in overlapping in cluster, tough but thin, fibrilous, undersurface hexagonal pore, white in color.
<i>Coltricia cinnamomea</i> (Jacq.) Murr.	Growing on soil, June to September	Fruiting body, circular cap, vase shaped, zonation, cinnamon brown, dry to leathery texture.
<i>Cyathus striatus</i> (Huds.) Wild. S	Growing on decaying jute Throughout the year	Fruiting body vase shaped to bellshaped, with inside whitish grey peridioles. Fruiting body covered by minute dense hairs.
<i>Termitomyces medius</i> Heim & Grasse	Growing on soil in cluster June to August	Fruiting body white to gray in colour, conic cap and convex, cap umbo at centre stipe long tapering to base and inserted in soil.
<i>Lentinus strigosus</i> Fr.	Growing on decaying wood stump June to September	Fruiting body vase shape or trumpet shaped, margin inrolled, dense stiff hairs, purplish when young and later turns to dark brown in colour.
<i>Fistulina hepatica</i> (scharff.) With.	Growing on decaying hardwood June to September	Fruiting body liver or tong shaped, gelatinious, blood red when cut seems like beef streak.

<i>Scleroderma citrinum</i> Pers.	Growing on siul along with moss pad March to Early December	Fruiting body subglobose, body tough and hard, surface coarse, often cracked with brownish black colour spores.
<i>Mycena maculate</i> P.Karst	Growing on fallen decaying wood twigs. July to October	Fruiting body conic cap, cuticle smooth, flare margin with age, brown blackish brown colour, stipe blackish purple brown colour.
<i>Lycoperdon perlatum</i> Pers.	Growing on ground along with decaying litters. May to October	Fruiting body club shaped, white when young and tan brown, upper surface with spines with warts later open or burst at centre.
<i>Flammulina velutipes</i> (Curtis) Singer	Growing on stump roots September to May	Fruiting body growing on cluster, cap convex than flat, orange red to orange yellow in colour, stipe cylindrical and slightly tapering towards the base
<i>Asterophora parasitica</i> (Bull. ex Pers.) Singer	Saprobial or parasitic on <i>Russala</i> sp. or <i>Lactarius</i> sp	Fruiting body – cap convex, flat later depressed, creamy white, presence of outgrowth of new fruiting body from top, stipe cylindrical.
<i>Trametes hirsute</i> (Wulfen) Lloyd	Growing on dead wood stump June to December	Fruiting body bracket shaped to fan shaped, sessile, presence of zonation, fine hairs densely present, white to cream color.
<i>Pycnoporus canabarinus</i> (Jacq.) P.Karst	Growing on decaying fallen tree January to December	Fruiting body bracket shape to plate like, sometime overlapping, smooth to warty, dark orange red colour, pores minute and more darker brick red.
<i>Morchella. Crassipes</i> (L.) Pers. Fr	Growing on soil March to May	Fruiting body cone shape, cone with so many irregular ridges, ridges not so darker than pits, yellow to bark pale brown. Stem hollow thin with extended base.
<i>Parasola auricoma</i> (Pat) Redhead, Vilgalys & Hopple	Growing on soil along with wood chips March to may	Fruiting body ,cap convex to flat deeply grooved from nearby centre to margin, thin flesh, black gray in color. Brown nblack gills, stipe hollow.
<i>Schizophyllum commune</i> Fries	Growing on decaying wood Throughout the year	Fruiting body sessil, shell shaped, upper surfae rough with whitish to gray colour and inner surface with splits gills with tan color.

seasons, but unregulated foraging might deplete the diversity of the some edible mushroom.

Altogether from the collected samples the mushroom having potential as food is approximately 18 species, as a medicine and other biotechnological uses are 21 species and 25 species are unknown or inedible category. As mushrooms perform different work and serve to humans, nature, various microbes, plants, it has become evident to make a proper database. Tapping its potential in food, nutraceuticals and biotechnological fields should be another target to focus on before it vanishes from the nature. The diversity of macrofungi or mushrooms from Sikkim has a potential in terms of *in vitro* isolation of culturable strains like *Pleurotus* sp., *Pholiota* sp., *Grifola* sp., *Trametes* sp., *Xylaria* sp., *Schizophyllum* sp., *Termella* sp., *Lentinula* sp., *Ganoderma* sp. and *Lycoperdon* sp. etc., in future cultivation and domestication programme.

The sampling data collected within two years time clearly depicts high macrofungus or mushroom diversity in the region. The macrofungal data are the preliminary work and a part of fungal diversity study of FWS, along with diversity study of others

groups such as endophytes, soil and litter fungus. This work will serve as a reference database of Sikkim states treasure of mushroom diversity, potential resource as food, medicine and other industrial products; a reference document for the forest managers to include macrofungi biodiversity conservation in forest management policies.

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REFERENCES

- Bonet J, Fischer C and Colinas C. 2004. The relationship between forest age and aspect on the production of sporocarps of ectomycorrhizal fungi in *Pinus sylvestris* forests of the central Pyrenees. *For. Ecol. Manag.*, 203, 157–175.
- Chang S, Miles GP. 2004. Mushrooms: Cultivation, nutritional value, medicinal effects and environmental impact. CRC Press, USA, pp 436.
- Chang ST and Wasser SP. 2012. The role of culinary medicinal mushrooms on human welfare with a pyramid model for human health. *Int J Med Mushrooms*. 14: 93-134.
- Das K., 2010, Diversity and conservation of wild mushrooms in Sikkim with special reference to Barsey *Rhododendron* Sanctuary, NeBIO, 1(2), 1-13.

- Deacon J. 2006. Fungal biology. Fourth Edition. Malden: Blackwell Publishing Oxford.
- Demirbas A. 2000. Accumulation of heavy metals in some edible mushrooms from Turkey. *Food Chem* 68: 415-419
- Duarte S, Pascoal C, Cassio F and Barlocher F. 2006. Aquatic hyphomycete diversity and identity affect leaf litter decomposition in microcosms. *Oecologia*. 147:658-666.
- Hawksworth D.L., Kirk P.M., Sutton B.C. and Pegler D.N. 1995. Ainsorth and Bisby's dictionary of fungi. 624 pp
- Hawksworth, D. L. & Lücking, R. 2017. Fungal diversity revisited: 2.2 to 3.8 million species. In: *The Fungal Kingdom*, (eds J. Heitman, B. Howlett, P. Crous, E. Stukenbrock, T. James & N.A.R. Gow), pp. 79-95. Washington, DC: ASM Press. DOI: <https://doi.org/10.1128/microbiolspec.FUNK-0052-2016>.
- Kalac P, Svoboda L and Havilkova B.2004. Content of cadmium and mercury in edible mushrooms. *J. Appl. Biomed.* 2:15-20.
- Mueller, GM, Schmit JP, Leacock P R, Buyck B, Cifuentes J, Desjardin DE, Halling RE, Hjortstam K, Iturriaga T, Larsson KH, Lodge DJ, May TW, Minter D, Rajchenberg M, Redhead SA, Ryvarden L, Trappe JM, Watling R and Wu Q. (2007). Global diversity and distribution of macrofungi. *Biodiversity and Conservation* 16: 37-48.
- Osmundson TW, Robert VA, Schoch CL, Baker LJ, Smith A, Robich G. 2013. Filling Gaps in Biodiversity Knowledge for Macrofungi: Contributions and Assessment of an Herbarium Collection DNA Barcode Sequencing Project. *PLoS One.*; 8(4)1-8
- Ozturk C, Kasik G., Dogan H.H. and Aktas S. 2003. Macrofungi of Alanya district. *Turk J Bot* 27: 303.
- Panda MK and Tayung K. 2015. Documentation and ethnomedicinal knowledge on wild edible mushrooms among ethnic tribes of Northern Odisha, India. *Asian J Pharm Clin. Res.* Vol 8, Issue 4, 139-143
- Purvis A, Agapow PM, Gittleman JL and Mace GM. 2000. Nonrandom extinction risk and the loss of evolutionary history. *Science* 288, 328-330.
- Richards W and Murray D. 2002. Macrofungi of la Butte Creek, Fidler-Greywillow and Colin-Cornwall lakes Wildland Provincial Parks, Community development Parks and protected Areas division. Edmonton, Alberta. 33 pp.
- Sharma SK, Atri NS. 2014. Nutraceutical composition of wild species of genus *Lentinus* Fr. from Northern India. *CREAM*, 4: 11-32. DOI: 10.5943/cream/4/1/2.
- Swapana S, Syed A and Krishnappa M. 2008. Diversity of Macro Fungi in Semi Evergreen and Moist Deciduous Forests of Shimoga District-Karnatka, India. *Journal of Mycology and Plant Pathology*. 38, 21-26.
- Wangdi LP. 2019. Characterization and Nutritional Status of Wild Edible and Medicinal Mushrooms of Sikkim, *Int. J. Pure App. Biosci.* 7: 327-334. doi: <http://dx.doi.org/10.18782/2320-7051.7421>
- Wani BA, Bodha RH, Wani AH. 2010. Nutritional and medicinal importance of mushrooms. *Journal of Medicinal Plants Research* 4: 2598-2604.
- White T.J., Bruns T., Lee S., Taylor J.W. 1990. Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. PCR Protocols: A Guide to Methods and Applications. (Eds) Innis MA, Gelfand DH, Sninsky JJ, White T.J., New York: Academic Press Inc, 315-322.
- Wu, X., Yang, Z., Li, T. 2010, Fungi of Tropical China; Science Press: Beijing, China.
- Yu, F. Wang, X. Liu, P. 2002, Prospects of exploitation and utilization on edible fungi resource in Yunnan. *Chin. Wild Plant Resour.*, 21, 21-25.