Research Article

Macrofungi in Some Forests of Telangana State, India

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The fruiting bodies of macrofungi were collected from some forests, fences, waste fields, timber depots of Telangana state during rainy season. This is an attempt to give a broad picture of diversity of macrofungi belonging to the class Basidiomycetes in some forest areas of Telangana region. A total number of 50 fruiting bodies were collected and cultured and among them only ten were identified based on their macroscopic features and molecular identification since they showed good lignolytic activity.

1. Introduction

The total forest cover in India according to the latest Forest Report 2013 is 69.8 million hectares and this constitutes 20.64% of the geographic area. The state of Telangana is one of the progressive states in the southern part of India with an area of 114,840 sq.km, while the recorded forest area is 29242.08 sq.km. The annual rainfall is between 900 to 1500 mm in northern Telangana and 700 to 900 mm in southern Telangana, from the southwest monsoons and it is a semiarid area and has a predominantly hot and dry climate. The Basidiomycota is the second largest phylum of kingdom Fungi, with approximately 23,000 species [1] including many of the common macroscopic forest fungi (e.g., mushrooms, shelf fungi). Mushroom is a general term utilised mostly for the fruiting body of the macrofungi, belongs to Ascomycota and Basidiomycota, and represents only a short reproductive stage in their life cycle. Investigations on the taxonomy and diversity of macrofungi are gaining importance, as many macrofungi are facing a threat of extinction due to environmental destruction. Wild mushrooms have deep biological and economical impact. Information on mushroom diversity is important because of its major functions in natural and organised eco-schemes as ectomycorrhizal fungi. The fungal group becomes a significant component for reforestation programs. Furthermore, they are significant as nourishment source for human beings and animals [2]. The macrofungi were also used as a bioindicator of environmental quality [3]. Most of the fleshy and gilled macrofungi were prevalent in the rainy times of the year as this time is favourable for their output, since there is ample moisture, favourable warmth, relative humidity, and sunshine, which furthermore aids the macrofungi in the decomposition of dead organic tissue. The early dry time of the year collection was predominated by the polypores since there is decline in rainfall and relative humidity, boost in warmth, and sunshine and most of the fleshy macrofungi will not withstand these conditions. During rainy season, there is abundant growth of several kinds of Basidiomycetes. Many fungal species groups do not produce visible fruit bodies or other species-specific characteristics, or these characteristics are extremely rare and cannot be detected in traditional surveys; hence these can be studied using molecular methods [4–6]. Further, Basidiomycetes are considered to be a very interesting group of fungi with their exceptional adjustment abilities and as natural lignocellulose degraders with different ecological groups such as white rot, brown rot, and leaf litter fungi [7]. Many kinds of macrofungi are not edible but possess variety of pharmaceutical and medicinal qualities. A fraction of complete fungal wealth has been subjected to scientific scrutiny and mycologists continue to unravel the unexplored and hidden wealth; one-third of fungal diversity of the globe exists in India and of this only 50% is characterized until now [8]. About 10,000 species within the overall fungal estimates of 1.5 million belong to
this group. Mushrooms alone are represented by about 41,000 species, of which approximately 850 species are recorded from India [9]. Some of the wild edible mushrooms have been reported from South West India [10]. The first listed Indian Fungi was published [11] and that was revised by Vasudev [12] and extra additional lists appeared in between culminating with the fungi of India (Bilgrami et al. [13]).

The present communication is the collection, culture, identification, and preservation of the macrofungi in some forests of Telangana region. This study adds extra information to the present knowledge on the data of diversity of fungi in Telangana region and also to understand their bioprospects.

2. Materials and Methods

2.1. Study Area. The fruiting bodies of white rot fungi were collected in rainy season from the forest of Etunagaram, Bhadrachalam, Kothagudem, Narsampet, Ilandhu, Khammam, Bodhan, and Nirmal of Telangana region.

2.2. Culturing and Preservation. The collected fruit bodies were cultured for their mycelial growth as per the method suggested [14]. A small piece of fruiting body was dipped in 0.01% mercuric chloride to remove surface contamination and washed several times with distilled water to remove the traces of mercuric chloride and transferred aseptically on to 3% malt extract agar [15] slants and was incubated for 5–7 days. The mycelium collected from the growing edge of those slants was transferred on to new malt extract agar slants and incubated further 5–7 days to obtain pure culture. Pure cultures were subcultured on malt extract agar slants and plates every fortnight.

2.3. Characterization of Macrofungi. Based on macroscopic features, namely, size, shape, sporocarp nature, color, pore print, margin of pileus, characters of lamella, spacing of gills, characters of the stipe, and the presence or absence of veins on stipes, the fungi were identified, http://en.wikipedia.org/wiki/Template:Mycomorphbox.

2.4. Identification of Macrofungi

2.4.1. Macroscopic Identification. Seven mycological characters useful in tentative identification of mushrooms are hymenium type, cap shape, gills, stipe character, color of the spore print, ecological type, and edibility.

2.4.2. Molecular Identification. Based on their lignolytic activity (data not shown here) ten species of wild macrofungi were selected for confirmation of their identities using molecular methods (see Figure 1). Collected fruiting bodies are initially cultured on malt agar (MEA), and the genomic DNA of the macrofungi was extracted using modified CTAB method [16]. The genomic DNA of the macrofungi was then subjected to PCR to amplify the ITS regions of the nuclear ribosomal DNA using two primers: ITS 1 (5'TCCGTAGGTGAA CCTTGCGG3') and ITS 4 (5'TCCTCCGCTTTAGATGC3') [17]. The PCR products were then purified using QIAGEN purification kit following the manufacturer's instructions and the purified PCR products were sent to SciGenom, Cochin, for outdoor DNA sequencing. Related gene sequences for each of the macrofungal specimens were obtained from NCBI GenBank and then automatically aligned using ClustalW program incorporated in BioEdit v. 7.1.9 [18]. Manual sequence alignments were then performed using Bioedit to allow maximum sequence similarity. Finally, phylogenetic tree was constructed based on maximum parsimony using PAUP v. 4.0 b10 [19].

3. Results

The fruiting bodies of white rot fungi were collected in rainy season from few forest areas of Telangana state (Table 1). The collected fruit bodies were cultured until the pure cultures were obtained. The lists of identified wild mushrooms were depicted in Tables 2 and 3. The fungi that belong to Polyporales are more in number than other orders. Schizophyllum radiatum belong to the order Agaricales. All the collected fungi were saprotrophic and were not edible. Based on seven mycological characters they were characterized. The climatic conditions in Telangana are congenial for prevalence of macrofungi and hence recorded maximum genera. During the systematic surveys at different forests of Telangana, total 50 mushroom samples were collected and these samples belong to different genera out of which only 10 mushroom samples were identified based on their lignolytic activity (data not shown here) and were identified basing on the molecular identification and macroscopic characters. All collected mushrooms have been deposited in the department herbarium with accession numbers. The ten identified mushrooms by molecular level are Trametes versicolor (GenBank Accession number HP953984), Pycnoporus cinnabarinus strain SYBC–L14, Daedaleopsis flavidula strain 5A, Trametes elegans voucher, Trametes hirsuta, Fomitopsis feei (GenBank Accession number AY 513271), Trametes gibbosa (GenBank Accession number AY351924.1), Trametes elegans GenBank (Accession number AY351925.1), Ganoderma lucidum, and Schizophyllum radiatum (GenBank Accession number HE863742.1).

4. Discussion

In Basidiomycotina more than 2000 species of edible mushrooms are reported in the publications from different components of the world. People all over Asian countries in the twentieth century know that mushrooms are important biosource of novel secondary metabolites. In India, the alternative systems of medicine utilize the curative properties of mushrooms. Secondary metabolites of these mushrooms are chemically diverse and possess a broad spectrum of biological activities, which are explored in traditional medicines [20]. In India, several mushrooms have been reported as medicinal mushrooms which have antioxidant, antimicrobial, and anti-inflammatory activity with antitumor
Figure 1: Photographs of identified macrofungi.
**Table 1: List of macrofungi collected in some forests of Telangana state.**

| Site Number | Site name                      | Strain name                  | Source         | Altitude                  |
|-------------|--------------------------------|------------------------------|----------------|---------------------------|
|             | Etarnagaram forest             | *Abortiporus biennis*        | Ground-dwelling| 18° 20' 26" N, 80° 25' 45" E |
|             |                                | *Agaricus xanthodermus*      | Hardwood       |                           |
|             |                                | *Cantharellus subalbidus*    | Hardwood       |                           |
|             |                                | *Tyromyces sp.*              | Tree           |                           |
|             |                                | *Gleophyllum sp.*            | Tree           |                           |
|             |                                | *Gomphus sp.*                | Tree           |                           |
|             |                                | *Trametes pubescens*         | Dead wood      |                           |
|             |                                | *Trametes elegans*           | Wood log       |                           |
|             |                                | *Pycnoporus cinnabarinus*    | Wood log       |                           |
|             |                                | *Trametes hirsuta*           | Wood log       |                           |
|             |                                | *Oligoporus sp.*             | Wood log       |                           |
|             |                                | *Schizophyllum radiatum*     | Wood log       |                           |
|             |                                | *Trichaptum*                 | Wood log       |                           |
|             |                                | *Trametes versicolor*        | Wood log       |                           |
|             |                                | *Trametes gibbosa*           | Wood log       |                           |
|             |                                | *Fomitopsis feii*            | Tree           |                           |
|             | Etarnagaram Timber Depot       |                              |                |                           |
| 1           |                                | *Auricularia auricula*       | Wood log       |                           |
|             |                                | *Clitocybe philophylla*      | Tree           |                           |
|             |                                | *Tremella foliacea*          | Fallen branches|                           |
|             |                                | *Tremella hirsuta*           | Dead wood      |                           |
|             |                                | *Daedaleopsis flavida*       | Fallen branches| 17° 40' 12" N, 80° 52' 48" E |
|             |                                | *Ganoderma lucidum*          | Wood log       |                           |
|             |                                | *Stereum hirsutum*           | Dead wood      |                           |
|             |                                | *Trametes betulina*          | Wood log       |                           |
|             |                                | *Ganoderma sp.*              | Wood log       |                           |
|             |                                | *Inonotus hispidus*          | Tree           |                           |
| 2           | Bhadrachalam forest            | *Phellinus igniarius*        | Tree           | 18° 40' 12" N, 77° 54' 0" E |
|             |                                | *Tremella aurantia*          | Wood log       |                           |
|             |                                | *Tremella ostrea*            | Tree barks     |                           |
|             |                                | *Dichomitus sp.*             | Wood log       |                           |
|             |                                | *Trametes sp.*               | Wood log       |                           |
| 3           | Bodhan                         | *Ganoderma applanatum*       | Tree           |                           |
|             |                                | *Amauroderma*                | Wood log       |                           |
|             |                                | *Trametes ochracea*          | Wood log       | 19° 6' 0" N, 78° 21' 0" E |
|             |                                | *Trametes elegans*           | Wood log       |                           |
|             |                                | *Pycnoporus coccineus*       | Wood log       |                           |
| 4           | Nirmal                         | *Pleurotus sp.*              | Wood log       |                           |
|             |                                | *Trametes sp.*               | Wood log       |                           |
| 5           | Illandu Timber Depot           | *Daedaleopsis confragosa*    | Wood log       | 17° 36' 0" N, 80° 19' 48" E |
|             |                                | *Pycnoporus sanguineus*      | Dead wood      |                           |
|             |                                | *Phyllotopsis*               | Wood log       |                           |
Table 1: Continued.

| Site Number | Site name      | Strain name       | Source       | Altitude                  |
|-------------|----------------|-------------------|--------------|---------------------------|
| 6           | Kothagudem     | Trametes sp.      | Wood log     | 17° 33' 0'' N, 80° 37' 48'' E |
|             |                | Ganoderma sp.     | Wood log     |                           |
| 7           | Narsampet      | Daedaleopsis sp.  | Wood log     | 17° 55' 35.02'' N, 79° 53' 48.99'' E |
|             |                | Trametes sp.      | Wood log     |                           |
|             |                | Pycnoporus sp.    | Wood log     |                           |
| 8           | Khammam        | Schizophyllum commune | Wood log | 17° 15' 0'' N, 80° 9' 0'' E |

Table 2: Classification of identified wild mushrooms.

| Division              | Class         | Order       | Family       | Genus            | Species          | Place          |
|-----------------------|---------------|-------------|--------------|------------------|------------------|----------------|
| Basidiomycota         | Agaricomycetes| Polyporales | Polyporaceae | Trametes         | versicolor       | Etunagaram     |
|                       |               |             |              | Trametes         | hirsuta          | Etunagaram     |
|                       |               |             |              | Ganoderma        | elegans voucher  | Etunagaram     |
|                       |               |             |              | Daedaleopsis     | lucidum          | Etunagaram     |
|                       |               |             |              | Trametes         | flavida          | Etunagaram     |
|                       |               |             |              | Trametes         | gibboa           | Etunagaram     |
|                       |               |             |              | Pycnoporus       | cinnabarinus     | Etunagaram     |
|                       |               |             |              | Fomitopsis       | feet             | Etunagaram     |
| Basidiomycota         | Basidiomycetes| Polyporales | Polyporaceae | Schizophyllum     | radiatum         | Khammam        |
|                       |               |             |              | commune          |                  |                |

and other properties [21]. Study on mushrooms in South India such as Tamil Nadu, Kerala, Karnataka, and Andhra Pradesh was neglected as regards to studies on agarics until 1975 [22]. All these collected white rot fungi were rich in medical importance. *Ganoderma lucidum* is well known to promote health and longevity, lowers the risk of cancer and heart disease, and boosts the immune system [23]. Polysaccharides from mushrooms as anticancer agents, other constituents exhibiting antioxidants and anti-hyper-tensive, cholesterol-lowering, liver protection, anti-biotic, anti-inflammatory, anti-diabetic, antiviral, and antimicrobial-like activities, have overtly primed its potential as dietary supplements [24]. Some of the Indian mushrooms which are having the medicinal properties are *Agaricus bisporus* [25], *Astraeus hygrometricus* [26], *Volvariella bombycina* [27], *Ramaria formosa* [28], *Pleurotus sajor-caju* [29], *Pleurotus pulmonarius* [30], *Pleurotus ostreatus* [31], *Phellinus rimosus* [32, 33], *Lycoperdon perlatum* [34], *Lentinus tuber-regium* [35], and *Lentinus squarrosulus* [36]. One of the authors [37] worked on *Schizophyllum radiatum* and reported the potentials of extracellular biosynthesis of silver nanoparticles, their characterization, and antimicrobial activity on gram-positive and gram-negative bacteria. The *S. radiatum* was also studied for its antimicrobial and antioxidantive properties under submerged fermentation. The supernatant of the seed media obtained after separating the mycelia has been used for the synthesis of silver nanoparticles, whereas the species of *Fomitopsis feei* were recorded in decolorization of triphenyl methane dyes, namely, bromophenol blue, basic fuchsin, methyl violet, methyl green, ethyl violet, and malachite green [38]. This report clearly explains that the bioremediation by utilising fungal organisms was advised to be the cost-effective and eco-friendly method of decolorization of effluents released from the dye industries. The genus *Trametes* and its species have many immunomodulatory and anticancer effects [39]. The genus of *Daedaleopsis* has antibiotic, antihyper-sensitive, and antitumour properties [40].

A constant monitoring and collection, identification, and preservation of the wild mushrooms are the need of the hour to explore and make use of the bioprospects of the diversified macrofungal species. This is a preliminary and basic work carried out to locate and identify the bioprospects of the existing macrofungi which will pave the way for understanding an elaborate study on this aspect.

5. Conclusion

Amongst the vast number of living forms very little amount of attention has been paid to conservation of fungal biodiversity. Due to loss of natural habitats, soil and air pollution expansion of monocropping and loss of genetic diversity many fungal species are on threat. For the smooth working of this terrestrial environment, the preservation of mushroom differences is discriminating. Keeping in view this gigantic mushroom treasure it is the high time to completely save this biodiversity. A few mushrooms are known to be the
Table 3: Mycological characters of identified wild mushrooms.

| Name                     | Hymenium type         | Cap shape           | Which gills | Stipe character | Spore print colour | Ecological type      | Edibility               |
|--------------------------|-----------------------|---------------------|-------------|-----------------|--------------------|----------------------|-------------------------|
| *T. versicolor*          | Pores                 | Offset or distinct  | Decurrent   | —               | White to yellow    | Saprotrophic          | Edible but unpalatable |
| *T. hirsuta*             | Pores                 | Distinct            | Decurrent   | Bare            | Cream              | Saprotrophic          | Inedible               |
| *T. elegans voucher*     | Pores and gills       | No distinct cap     | Decurrent   | —               | White              | Saprotrophic          | Inedible               |
| *Ganoderma lucidum*      | Pores                 | Offset or indistinct| Irregular   | Bare            | Brown              | Saprotrophic          | Edible                 |
| *Daedaleopsis flavida*   | Pores                 | No distinct cap     | Decurrent   | —               | White              | Saprotrophic          | Inedible               |
| *T. gibbosa*             | Pores                 | Offset              | Decurrent   | Bare            | White to cream     | Saprotrophic          | Inedible               |
| *T. elegans*             | Pores                 | Distinct            | Decurrent   | Bare            | Cream              | Saprotrophic          | Inedible               |
| *Pycnoporus cinnabarinus*| Pores                 | Distinct            | Decurrent   | Bare            | Cream              | Saprotrophic          | Inedible               |
| *Fomitopsis fæi*         | Pores                 | No distinct cap     | Decurrent   | —               | Brown              | Saprotrophic          | Inedible               |
| *Schizophyllum communæ*  | Pores                 | Offset or indistinct| Decurrent   | —               | White              | Saprotrophic          | Inedible               |

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