

Ecological Diversity and Distribution of Noble Macro Fungi of Sal Dominated Forest of Central India with Special Reference to Agaricales

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Abstract: The present study was undertaken within a year from June 2012 to September 2013 in different sampling sites of Amarkantak Biosphere reserve forest, Anuppur district of central India. The vegetation and climatic conditions of Amarkantak possesses prime location in India for the hot spot biodiversity of macro-fungi. Although this region is still unexplored due to the unawareness and less attention towards this subject. The study was therefore done to explore the variable and diverse species of macro fungi which have economic and medicinal importance. Present study describing the diversity and distribution of different mushrooms in different habitats of Amarkantak forest of central India. Total 58 mushroom samples were collected belonging to different genera. Russulaceae and Amanitaceae were the most dominated families in this study. This study is the extended work of previous work done by the author. Preliminary work has been already published. Two medium were tested for the in vitro culture of mushroom mycelium. It was observed that PDA medium was the most suitable medium for agaricales.

Key Words: Amarkantak- Biosphere Reserve, Forest, Macro fungi, Diversity

I. INTRODUCTION

Sal forest is a type of forest which is mainly dominated by a single plant species, *Shorea robusta* which belongs to the category of 'Tropical Moist Deciduous Forest'.

Vegetation and distribution of *Shorea robusta* forests is influenced by the topography, geology, soil and climatic conditions. In India alone 13 million hectares, sal forest have broad distribution among all Dipterocarps. Bangladesh and Nepal together have over one million hectares of Sal Forest.

Ectomycorrhizal fungi can account for 25% or more of forest root biomass, representing a major below-ground structural component of fungal species diversity (Pande *et al.* 2004). Ectomycorrhizal associations are considered key factors in forest ecosystems for the survival and growth of trees supplying nutrients to host plants, particularly immobile nitrogen and phosphorus. Knowledge of the distribution and ecology of ectomycorrhizal fungi is important for conserving their diversity as well as for the selection of species for forest nurseries (Giachini *et al.* 2000). In forest ecosystems, the richness and diversity of ectomycorrhizal communities strongly contrasts with the low number of woody species. Scores of fungal species are commonly associated with a single tree, and heterogeneity amongst trees results in several hundred fungal species at stand scale (Dahlberg 2001; Horton *et al.* 2001; Jonsson *et al.* 2001).

The town of Amarkantak lies in the newly created district of Anuppur, in Madhya Pradesh. It is situated on the Maikal

Mountain range which links the Vindhya and Satpuda mountain ranges at about 1067 meters above mean sea level. Amarkantak region is a unique natural heritage. The temperate climate and equitable distribution of rain make Amarkantak an ideal plateau for dense forest. Climatically Amarkantak is temperate. Whilst the forest is sal dominated, there are associate species such as *Buchanania Lanza*, *Ougeinia Oojeinesis*, *Mallotus philipensis*, *Gardenia lalifolia*, *Anogeissus latifolia*, *Terminalia chebula*, *Bauhinia sp.*, *Grewia sp.*

The normal annual rainfall of the district is 1235.0 mm. The district receives maximum rainfall during south-west monsoon period from June to September. About 89.3 % of annual rainfall is received during monsoon season. Only 10.7 % of the annual rainfall occurs during non-monsoon period, from October to May. Thus maximum water available for ground water recharge is during south-west monsoon season.

The main objectives in this study were collection, identification, in-vitro culture of wild mushrooms and examining the possible changes in the associations between macro fungi and their hosts/substrates.

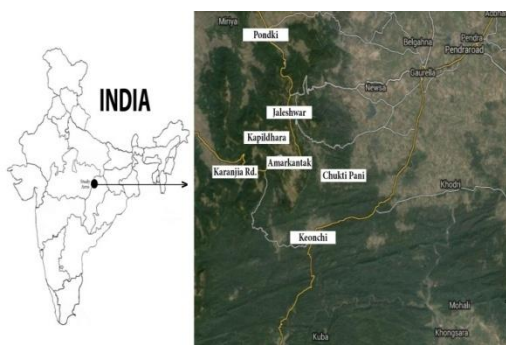


Fig. 1 Map showing the sampling sites In Amarkantak Biosphere Reserve Forest of Central India.

Table 1: Sampling sites in Sal dominated Amarkantak Forest of Central India

S.No.	Place	Longitude	Latitude
1	Amarkantak	22 ⁰ 41' 10.50" N	81 ⁰ 44' 31.35" E
2	Chuktipani	22 ⁰ 43' 2.172" N	81 ⁰ 45' 40.04" E
3	Jaleshwar	22 ⁰ 44' 19.62" N	81 ⁰ 46' 0.04" E
4	Karanjia Rd.	22 ⁰ 40' 31.44" N	81 ⁰ 42' 56.12" E
5	Kapildhara	22 ⁰ 42' 3.31" N	81 ⁰ 42' 11.93" E
6	Keonchi	22 ⁰ 37' 12.75" N	81 ⁰ 46' 37.88" E
7	Pondki	22 ⁰ 45' 38.81" N	81 ⁰ 44' 55.95" E

II. MATERIALS AND METHODS

The regular field trips were undertaken within a year from June 2012 to September 2013 in different sampling sites of Amarkantak Biosphere reserve forest, Anuppur district of central India to collect the samples of macro fungi. During collection of mushroom fungi extensive care has been taken to avoid any damage to the base and fragile parts of the samples. Macroscopic characters such as shape, size,

colour, colour difference with age, taste, odor, spore deposition of fresh specimens and its natural habitat was recorded with the season of appearance of their fruiting body. The specimens were kept in separate paper bags to avoid mixing. The photography was accomplished using digital camera. Each specimen was collected and labeled, indicating number, date of collection, locality and uses, and then brought to the laboratory for macro and micro investigations after that necessary micro chemical reactions were carried out to satisfy some probability factors. Collected specimens were dried, preserved in paper or polythene bags or in shape of herbarium sheets. Identification was made on the basis of critical observations of the specimens and by the use of relevant literature under the guidance of experts.

A. Collection of mushrooms

Equipments:

Use of large cool box or basket, so that specimens do not get squashed. Greaseproof paper sheets or bags and marker pens, to separate, identify and keep collections fresh (polythene bags provide warm, humid Trowel.

B. Identification-

Specimens were identified to their respective families, genera and species by consulting the available literature (Singer 1975, 1986; Jordan 1995, Rinaldi and Tyndalo 1974; Atri *et al.* 1992) Help of expert were also taken whenever required.

C. Isolation Of Mushroom Mycelium-

Potato Dextrose Agar Medium and malt extract agar medium were Prepared as per (Rahi, 2001, Upadhyay, 2004) for PDA medium Peeled Potato -200g, Dextrose 20 g, Agar-Agar 15g D/W 1000ml Malt Extract Agar Medium, Malt Extract-20g, Agar-Agar-15g, D/W 1000 ml.

D. Isolation of mushroom mycelium

It is done by tissue culture method under aseptic condition. Small piece of tissue of cap was inoculated in to the petridishes containing medium. After inoculation petridishes were kept for incubation at 28°C.

III. RESULTS

During the survey total 58 mushrooms samples were collected including edible as well as non edible out of which only 31 samples could be identified up to genus level. Spoiled samples were belonging to genus Russula. Most of them were infected

Table 2: Species and Family wise distribution of collected mushrooms from Amarkantak forest

Class	Order	Family	Genus	Species	Remarks
Homobasidiomycetes	Agaricales Boletales Russulales Lyophyllales	Agaricaceae	Agaricus	<i>Agaricus campestris</i>	edible
				<i>Agaricus bisporus</i>	edible
			<i>Leucoagaricus sp.</i>	edible	
			<i>Macrolapiota</i>	<i>Macrolapiota procera</i>	edible
		Amanitaceae	Amanita	<i>Milky white Amanita sp.</i>	Poisonous
		Boletaceae		<i>white Amanita sp.</i>	poisonous
				<i>Amanita veginata</i>	Edible but can not be eaten raw
				<i>Amanita pantherina</i>	Poisonous
				<i>Amanita caesarea</i>	poisonous
		<i>Boletus</i>	<i>Boletus edulis</i> <i>Xercomus chrysentron</i>	Edible Not edible	
		Russulaceae	Russula	<i>Suilus spraguei</i>	Not edible
				<i>Russula aquosq</i>	Edible
				<i>Russula solaris</i>	Edible
		Lyophyllaceae	Termitomyces	<i>Russula violacea</i>	Edible
	<i>Russula sp.</i>			Edible	
	<i>Termitomyces hemi</i>			Edible	
	<i>Termitomyces microcarpus</i>			Edible	
		<u>Schizophyllaceae</u>	<i>Schizophyllum</i>	<i>Schizophyllum sp.</i>	Not edible
	Ganodermatales	Ganodermataceae	Ganoderma	<i>Ganoderma lucidum</i>	medicinal
				<i>Ganoderma applanatum</i> <i>Ganoderma sp.</i>	Not known
				<i>Ganoderma sp.</i>	Not known
	Tricholomatales	Hygrophoraceae	<i>Nyctalis</i>	<i>Nictalis asterophora</i>	Not edible
	Licoperdales	Gastraceae	<i>Geastrum</i>	<i>Geastrum sps.</i>	edible
Cortinariales	Bolbitiaceae	<i>Panaeolus</i>	<i>Panaeolus foeniseii</i>	Poisonous	
			<i>Panaeolus semioratus</i>	poisonous	
Lycoperdales	Lycoperdaceae	<i>Lycoperdon</i>	<i>Lycoperdon sp.</i>	Not edible	
			<i>Lycoperdon sp.</i>	Not edible	
Sclerodematales	Sclerodermataceae	<i>Pisolithus</i>	<i>Pisolithus tinctorius</i>	medicinal	
Gastromycetes	Sclerodermatales	Sclerodermataceae	<i>Scleroderma</i>	<i>Scleroderma sp</i>	not edible

very soon with white insects because of this author was not able to perform microscopic studies. Out of These 31 only 12 could be identified up to species level. These 31 samples belongs to 8 order, 13 families and 14 genera. It was observed maximum mushroom diversity was observed during monsoon season which was represented by maximum families of Agaricales viz. Agaricaceae, Amanitaceae, Russulaceae, Boletaceae, Lyophyllaceae. During this survey Amanita belonging to Amanitaceae family, Russula belonging to Russulaceae, Boletus belonging to the family boletaceae were found to be predominant represented by 5 species, 4 species, and 3 species respectively Table 2. Both medium Potato Dextrose Agar medium and Malt

Extract Agar medium were tested for the growth of mushroom mycelium. It was observed that PDA favors the growth of mushroom mycelium of maximum species. Fig.3 Pure culture of *Nyctalis*, *Agaricus sp*, *Termitomyces sp.*, *Macrolapiota*, shown the maximum growth in PDA while other sp. like *Russula* and *Suilus spraguei* contaminated after 24 hrs of incubation. *Aspergillus sp.* was the main fungal contaminant in maximum mushroom mycelium. This is because of the climatic conditions of Amarkantak which favors the growth of this fungus. It was observed that after 24 hour of incubation one of the *Agaricus sp.* was always produced violet color pigment in Potato Dextrose Medium both in solid as well as liquid Figure 2 a and b. Biochemical study of This *Agaricus* sample is in progress. This mushroom sample was sent to the Agharkar Research Institute, Pune for the characterization but It could only identified up to genus level.

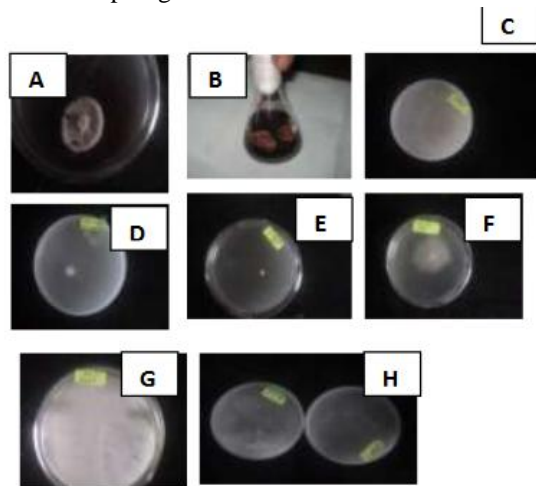


Fig. 2 *Agaricus sp.* In Potato Dextrose Agar Medium (A), In Potato dextrose broth (B) producing violet pigment in both solid and liquid medium. *Termitomyces hemi* (C) *Nyctalis* (D) *Agaricus campestris* (E) *Macrolapiota* (F) *Russula sp.*(G) *Termitomyces sp.*(H)*Agaricus sp.*

It was also observed that there was no growth of any mushroom mycelium in Malt Extract Medium. Among the microbiological media used in this study, it was indicated that PDA is more suitable media for mycelial growth of Agaricales

Table 3: In-vitro culture of some wild species of mushrooms of Amarkantak Biosphere Reserve forest of India

Code of the sample	Name of the species	Growth in mm in Potato Dextrose agar Medium(PDA)	Growth in mm in Malt extract Agar Medium
AA1	<i>Russula sp.</i>	Culture contaminated	No growth
AA2	<i>Nyctalis sp.</i>	Pure culture	No growth
AA4	<i>Agaricus campestris</i>	Pure culture	No growth
AA6	<i>Suilus spraguei</i>	contaminated	No growth
AA7	<i>Termitomyces microcarpus</i>	Pure culture	No growth
AA9	<i>Russula sp.</i>	contaminated	No growth
AA10	<i>Macrolapiota</i>	Pure culture	No growth
AA14	<i>Macrolapiota</i>	Pure culture	No growth
AA23	<i>Termitomyces hemi</i>	Pure culture	No growth

AA1	<i>Russula sp.</i>	Culture contaminated	No growth
AA2	<i>Nyctalis sp.</i>	Pure culture	No growth
AA4	<i>Agaricus campestris</i>	Pure culture	No growth
AA6	<i>Suilus spraguei</i>	contaminated	No growth
AA7	<i>Termitomyces microcarpus</i>	Pure culture	No growth
AA9	<i>Russula sp.</i>	contaminated	No growth
AA10	<i>Macrolapiota</i>	Pure culture	No growth
AA14	<i>Macrolapiota</i>	Pure culture	No growth
AA23	<i>Termitomyces hemi</i>	Pure culture	No growth

Table 3. Most *Russula* did not grow in culture or grew very slowly. Other prominent macrofungal genera include *Agaricus* (2sp.), *Termitomyces* (2sp.), *Leucoagaricus* (1sp.), *Boletus* (3sp.), *Shizopillium* (1), *Ganoderma* (2), *Nyctalis* (1) *Geastrum* (1), *Paneolus*(2), *Lycoperdon* (2), *Pisolithus* (1) *Scleroderma* (1) as represented in Table 2. Family Russulaceae dominated the mushroom flora represented by two genera *Amanita* and *Russula* with 5 and 4 sp. Respectively Table 2. Study of ecology revealed that ectomycorrhiza were dominated in this region followed by saprophytes and parasites Fig.3.

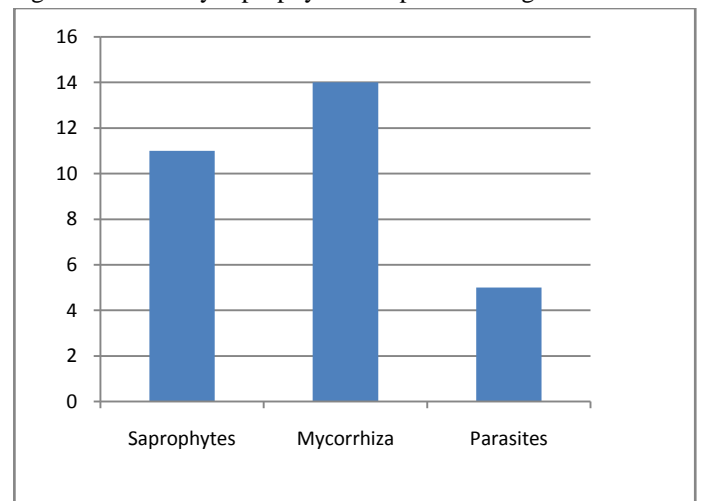


Fig. 3 Ecology of collected macro fungi.

In the year 2004-2008 including Madhya Pradesh and Chhattisgarh forests About 61 ectomycorrhizal mushroom species including 9 genera were identified (Sharma *et al* 2008) Out of 61, 38 were Agaricales

(61%), 11 Boletales (17%), 12 Sclerodermatales (19%). *Russula* was found to be the dominant genus inhabiting *Shorea robusta* forests. Similar results were obtained by author in the presented work.

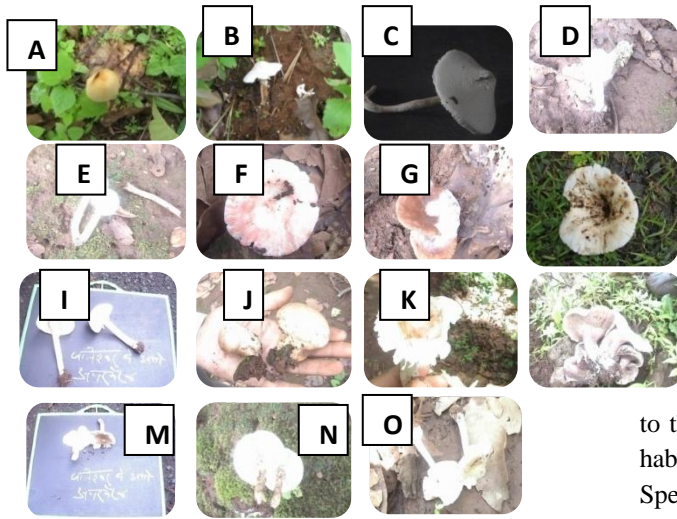


Fig. 4 Pictorial catalogue of wild edible mushrooms collected by Author during the study (A). *Paneolus* sp. (B). *Termitomyces* sp. (C). *Unknown* sp. (D). *Unknown* sp. (E). *Unknown* f-h *Russula* sp. (I). *Macrolapiota* sp. (J) *Pisolithus* sp. (K) *Russula* sp. (L) *Boletus* sp (M) *Russula* sp (N) *Amanita* sp. (O). *Amanita* sp

In forest ecosystem, ectomycorrhiza plays a significant role in tree regeneration and ecological function of several terrestrial ecosystems (Kennedy *et al.*, 2007; Agarwal and Sah, 2009; Kranabetter *et al.*, 2009). Turjaman *et al.* (2011) reports that *Boletus* sp. and *Scleroderma* sp. are capable of increasing the growth of *Shorea balangeran* on peat soil either during nursery or at field. *Shorea balangeran* (Dipterocarpaceae) is one of some types of trees which give significant wood produce grown on peat soil in Indonesia. In heath forests with podzolic soil type, organic decomposition runs slow due to being soaked in water, and because of its high acidic level, *Boletus*, *Entroma*, *Cortinarius*, *Amanita*, and *Tricholoma* are mostly found in such areas (Turjaman, 2013). (Pradeep and Vrinda 2010) agreed that genus *Russula* was generally found on soil surface (epigeous), and it grew solitarily, spreading to other areas such as under *Myristica malabarica* tree, *Vateria indica*, *Pongamia pinnata* and *Calophyllum apetalum*. (Morina *et al.* 1992) observed that the trees generally associated with ectomycorrhiza involved the family of Pinaceae, Fagaceae, Caesalpinaceae, Betulaceae, Dipeterocarpaceae, Myrtaceae, Casuarinaceae, dan Acaciaceae. Local people usually collect the mushrooms taken from the forest and are sold in local traditional markets. The abundance of this type of mushroom depends on climate (Chotimah *et al.*,

2013). Macroscopically, Ectomycorrhiza mushrooms are diverse in shape, size, colour, root and mantle surface geometry. Symbiosis between ectomycorrhiza and the tree or other plants results in carbohydrate needed such as sucrose and glucose. In reverse, mushroom mycelium could gain longer roots to reach further into soil. The carbohydrate is translocated from its source to root tissue and ectomycorrhiza (Pyasi *et al.*, 2013). The abundance of basidiocarp Russulaceae presumably related to the nutrient content of the

soil in the form of C - organic and P very high element. According to (Hernandez and Linera 2011), the abundance of macrofungi, including ectomycorrhiza and its distribution are related to the composition and structure of the tree colony in habitats with temperate and tropical climate. Specifically, in tropical regions, the existence of macro fungi and their genus distribution are related to precipitation and type of vegetation. The growth of ectomycorrhiza mushrooms is also influenced by environmental factors such as light intensity, temperature, humidity, soil fertility, aeration, and root exudate (Mardji, 2014). In forests with endemic, indigenous, and exotic trees of Kerala, more than 160 types of ectomycorrhiza and the family of Russulaceae were found (Pradeep and Vrinda, 2010). *Amanita* is categorized into the family of Amanitaceae, into which 500 species are included. Most of this genus is poisonous Muliyani *et al* (2014). Collected samples were also analyzed for their economic importance with help of tribal peoples living around Amatkantak. Out of 31 samples, 15 were poisonous, 13 were edible and 3 were of medicinal importance Table 2.

Climatic factors are thought to be the one of the main drivers of changes in fungal community structure. There is a strong body of literature showing that climate variables such as temperature and rainfall affect temporal patterns of various species of fungi (Zhang *et al.* 2005; Koide *et al.* 2007; Sato *et al.* 2012; A'Bear *et al.* 2013; Jarvis *et al.* 2013).

i) there will be differences in the rate of host range expansion and host shift between mycorrhizal and saprotrophic species, and ii) there are tendencies of any species in the dataset to shift host from their common hosts to another. In addition, part of this chapter has been published in (Gange *et al.* 2011) in which is an analysis of fungal fruiting of the common fungus *Auricularia, auricula-judae*, a species that is often cited as being mostly confined to one host. For mycorrhizal fungi, they depend on photosynthetically

fixed carbon produced by their associated trees and also, the physiological state of host trees may well drive the growth of these fungi (Egli 2011).

Of the common genera recorded in the dataset, most were dominated by mycorrhizal species compared to the saprotrophs. There were differences between these two trophic groups. Saprotrophic fungi decomposing wood and litter (Rayner & Boddy, 1988) obtain energy by degrading dead organic matter (Lindahl *et al.* 2007). On the other hand, mycorrhizal fungi obtain their energy from symbioses relationship with their host plants and in return providing their plant hosts with soil-derived nutrients (Smith & Read 1997; Lindahl *et al.* 2007). Similar results were also reported by (Lagana *et al.* 2002) and (Baptista *et al.* 2010) where the majority of their macrofungal species found were ectomycorrhizal species and the remaining were either saprotrophic and/or saprotrophic/parasitic. The high frequency of mycorrhizal species found in the records could be due to the age factor of the forests. (Dighton & Mason 1985) has shown that the number of ectomycorrhizal species present in a given ecosystem is dependent on the host plant's age. Moreover, research by (Luoma *et al.*, 1991; Keizer & Arnolds 1994 and Smith *et al.* 2002) again reinforce the fact that the number of ectomycorrhizal species could increase gradually with host plant age.

Saprotrophic fungi are key regulators of nutrient recycling in forest ecosystems and thus play an important role in decomposition, carbon sequestration and nutrient recycling processes in all terrestrial ecosystems (Griffith & Roderick 2008). According to (Bååth & Söderström 1979), up to 20% of the total amounts of nitrogen and phosphorus in a boreal forest soil may be incorporated into dead and active fungal mycelium. Moreover, (Frankland 1992) in her studies of fungal succession, found a declining number of saprotrophic fungal species when the resource became exhausted. Besides that, laboratory experiments have shown the ability of the mycelium of wood decaying fungi to take up inorganic phosphate from the soil and translocate it to high quality resource units (Boddy 1999; Lindahl *et al.* 2001). Meanwhile, (Jonathan & Fasidi 2001) have reported that calcium and magnesium were the best macro-elements while micro-elements (copper and zinc) enhanced optimum growth of an edible mushroom, *Psathyrella atroumbonata* in laboratory tests. Apart from resource availability, gases in the atmosphere may be another potential triggering factor for the formation of fruit bodies. Increased CO₂ concentration in the air was found to have a positive effect on mycelial growth in

the cultivation of *Pleurotus ostreatus*, *P. florida* and *P. eryngii* in laboratory experiments (Zadrazil 1975). Meanwhile, (Wallander & Nylund 1991) who studied the effects of excess nitrogen on ectomycorrhizal mycelium have suggested that nitrogen deposition from the atmosphere may damage the function of mycorrhizal fungi in the environment. They found that mycelial biomass increased rapidly when N was kept low and in balance with other nutrients, but showed no progress of mycelial growth when the N concentration was raised.

IV. CONCLUSION

On the basis of above findings it can be concluded that being rich in vegetation, it has rich diversity of mushrooms. Most of them till now not adequately documented so local and tribal peoples are not getting its socioeconomical advantage. Amarkantak forest can be a good source of potential edible and medicinal mushrooms. Identification and use of wild edible mushrooms play a vital role in enrichment of the social and economic life of tribals. Various bioactive substances like antifungal, antibacterial, antiviral, antioxidant and anticancerous etc. found in most of the mushrooms. Beside their consumption, the use of mushrooms in folk medicines also paves the way for upbringing the new industries. Further detailed studies on commercialization and conservation of popular wild edible macro fungi species in this Biosphere region will be profitable for economy.

V. ACKNOWLEDGEMENT

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