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SCREENING, ISOLATION AND DOCUMENTATION OF PHARMACEUTICAL IMPORTANCE MACRO FUNGI OF KULIK RAIGANJ WILDLIFE SANCTUARY, WEST BENGAL, INDIA

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ABSTRACT :

Kulik Raiganj Wildlife Sanctuary is the second largest bird sanctuary of Asia situated on the bank of Kulik River at the close vicinity of Raiganj town of West Bengal, India. The Sanctuary is famous for Asian migratory Open Bill Stork (Anastomus oscitans) of international interest. The sanctuary has 224 species of angiosperm. There is no any documentation of macro fungal diversity of pharmaceutical importance so far. In my present study screening, examination and documentation of pharmaceutical importance macro fungi were performed since 2014 to 2016. It was found to grow about 50 different species of macro fungi of which 17 species of macro fungi are found to produce secondary metabolites of pharmaceutical importance such as Lentinus sp, Schizophyllum commune, Trametes versicolor, Ganoderma lucidum, Pleurotus ostreatus, Collybia dryophila*, Armillaria mellea, Cymatoderma elegans, Marasmius sp,* Amanita muscaria, Auricularia auricular, Sparassis crispa, Volvariella volvacea, Inonotus obliquus, Hericium erinaceus, *Lentinus edodes and Coprinus atramentarius. These macro fungi might be used as potential sources of bio-molecules for development of medicines of therapeutic significance for human welfare.*

KEYWORDS : *Pharmaceutical, Kulik Wildlife Sanctuary, Macro fungi, Open Bill Stork.*

INTRODUCTION

Kulik Wildlife Sanctuary is locally called as Raiganj Kulik forest and located at the close vicinity of Raiganj town of Uttar Dinajpur of the state West Bengal, India (**Map; Figure-A**). The sanctuary is famous for Asian migratory Open Bill Stork (*Anastomus oscitans*) arrived at the forest canopy during the month of June – July. At the time of nesting in the month of July - August, many trees were found to be wounded and form a thick forest floor/bed with fallen plant parts which offers macro fungi to grow on them (**Figure-B**). These macro fungi are the good source of nutrition rich heath food as well as source of secondary metabolites of pharmaceutical importance including antibacterial drug (Hur *et al.,* 2004; Ishikawa *et al*., 2005; Kalyoncu *et al*., 2010). Macro fungi are wood-decaying lignicolous organism mostly are belonging to the phyla Basidiomycota and Ascomycota and are easily noticed, collected and recognized in the field. It has been estimated that approximately 1.5 million fungal species exist on the biosphere and about 140,000 species belong to macro fungi. However, only 10% of them are explored and 16% are cultured artificially (Chang, Philip & Miles, 2004).

Since the discovery of the first antibiotic penicillin (derived from *Penicillium chrysogenum*) in 1928 by Alexander Fleming a real revolution in medicine with a new era of antibiotics have started, Later, the entire group of β-lactam antibiotics was discovered, followed by the Waxman`s discovery of streptomycin derived

from Streptomyces bacteria. It is estimated that about 12,000 antibiotics are known today. Approximately 55% are derived by Streptomyces, 11% by other Actinomycetes, 12% from other bacteria and 22% from filamentous fungi (Inouye *et al*., 2004). Today, antibiotic resistant pathogenic microbes are the serious problem of threats for humans. In the present study it was emphasis for documentation of macro fungi for pharmaceutical importance at Raiganj Wildlife Sanctuary. It is therefore necessary for screening new natural resources of undiscovered secondary metabolites of antimicrobial activities from the nature. Many macro fungi were reported as potential resources of secondary metabolites of therapeutic significance (Zeidman *et al.,* 2005; Giovaninni, 2006) and fungi belong to the phylum Basidiomycota gave the promising results. Fungal secondary metabolites and their derivatives play an important role in the development of medicines. Today, it is estimated that more than 150 natural compounds have been extracted from different macro fungi those are used in the treatment of infectious diseases. In this paper documentation of pharmaceutical importance macro fungal was done at Raignaj Kulik Wildlife Sanctuary with full potential of screening and examinations.

MATERIALS AND METHODS

Survey Area

Kulik Raiganj Wildlife Sanctuary is located on the bank of Kulik river at the close vicinity of Raiganj Town, West Bengal in the North Eastern region of India with total geographic area of 1.30 sq Km. It lies between 88°7' to 88°8' E and 25°39' to 25°40' N. During the present study regular surveys and collection were performed from October 2014 to May 2015 and October 2015 to May 2016 during the peak mushroom growing season.

Sample collection and identification

The Macro fungi from Kulik Raiganj Wildlife Sanctuary were collected in silver foil/collection boxes following the method of Jonathan (2002) and brought to the laboratory for identification. Mushrooms with leathery texture were preserved in 4% (v/v) formaldehyde solution and mushrooms with soft texture were preserved in 2% (v/v) formaldehyde solution and maintained as herbarium specimens. The collection was done by visiting the sanctuary from June to September in the years 2014–2016. The surveys for collection, screening and examination were limited to macro fungi only. The fruiting bodies encountered were photographed and their morphological and ecological features were determined. A portion of the collected macro fungal specimen were dried at 40-72°C using blowing hot air and kept for future references, characterization and documentation. The habitat, colour, size, shape, odour, morphology and spore print were studied prior to the preservation of the collected macro fungal specimen. Identification of the collected macro fungal was done by standard microscopic methods (Roy & De, 1996) and by studying the macroscopic and microscopic characters (David, 1986; Das, 2009; Philips, 2006). The macro fungal specimens were deposited in the Museum of Department of Botany, Raiganj University,West Bengal, India with the accession numbers.

RESULTS AND DISCUSSION

Macro fungi were morphologically and structurally different in shape, size, colour, pattern of growth and substrate. In my present study about fifty different types of macro fungi were found to grow on forest floor enriched with fallen plant parts at Raiganj Kulik forest. Out of 50 macro fungi, 17 species of macro fungi were found to have different secondary metabolites of pharmaceutical importance to cure different infectious diseases. The macro fungi producing secondary metabolites of pharmaceutical importance collected from Raiganj Wildlife Sanctuary are *Lentinus sp, Schizophyllum commune, Trametes versicolor, Coprinus sp, Ganoderma lucidum, Pleurotus ostreatus, Collybia dryophila, Armillaria mellea, Cymatoderma elegans, Marasmius sp*, *Amanita muscaria, Auricularia auricular, Sparassis crispa, Volvariell avolvacea, Inonotus obliquus, Hericium erinaceus* and *Coprinus atramentarius and* (**Figure-C, Table-1 and 2)**.

Daily consumption of foods supplemented with macro fungi of pharmaceutical importance properties is a cost effective way to achieve significant health benefits by preventing disease occurrence. This concept arises with the modern science on foods with pharmaceutical importance properties which improve health or reduce disease risk has been progressively gaining interest (Eussen *et al.,* 2011; Leal *et al.,* 2013). Thus, incorporation of macro fungi as a daily food or as a supplement, containing many nutrients and bioactive substances, can assist in maintaining more normal cellular and immune function which helps in well functioning of the body (Wachtel-Galor *et al.,* 2004; Han *et al.,* 2006; Cheung 2008; Borchers *et al.,* 2008; Bobovcak *et al.,* 2010; Jiang and Sliva 2010; Brennan *et al.,* 2012). Examples of the available dietary supplements, commercial products developed from medicinal mushrooms that claim to provide beneficial effects of mankind are shown in **Table-2**.

Therefore, the value of macro fungi as the dietary supplements as well as active secondary metabolites for medicinal importance grows day to day on the world market. They are very safe and considered as organic in nature useful in the daily diet, especially for people suffering from various diseases.

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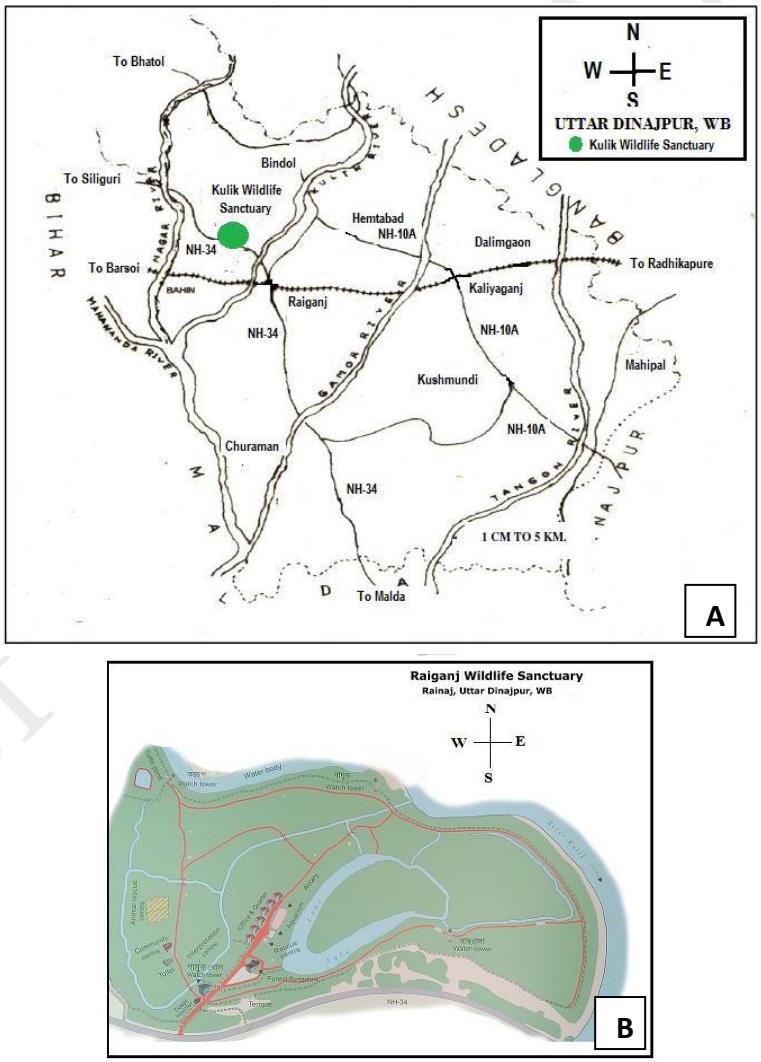

Map: A- Uttar Dinajpur District of West Bengal, India; B- Kulik Raiganj Wildlife Sanctuary at the close vicinity of Raiganj Town Raiganj, Uttar Dinajpur, WB.

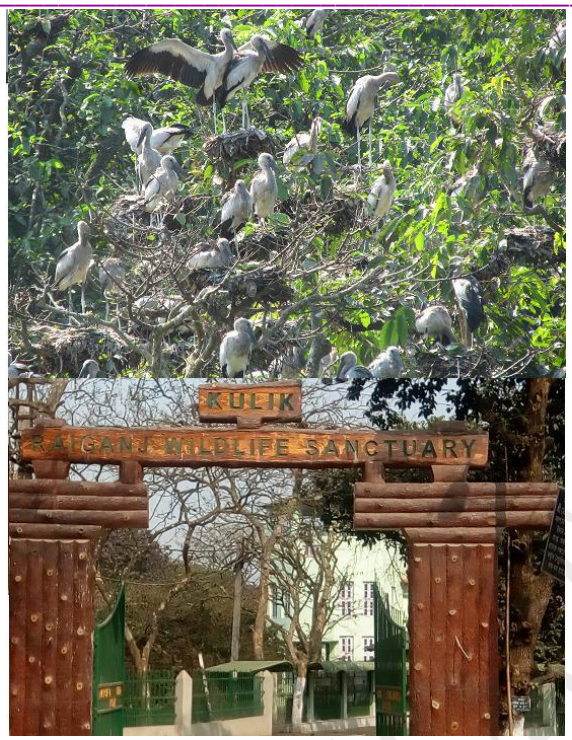

Figure-A: Kulik Raiganj Wildlife Sanctuary, Uttar Dinajpur, West Bengal, India

Figure-B: Asian migratory Open Bill Stork Night Heron (Anastomus oscitans) at Raiganj Wildlife Sanctuary:
1= Collecting branch of tree for their nesting; 2= Tree having some nests of Asian Open Bill Stork; 3= Eggs of Asian Open Bill Stork; 4= Fallen birds' nest made up with tree branches and leaves on the floor of forest

Table-1: Different Macro fungi grown on the forest bed of Raiganj Wildlife Sanctuary, West Bengal, India and their features and secondary metabolites of pharmaceutical importance

Macro fungi	***Secondary metabolites***	***Habit***	***Colour of fruiting body***	***Spore print***
Lentinus edodes (Berkeley) Pegler; Marasmiaceae	Lentinan	Grows on tree trunks and dead barks of trees	Colour light brown to darkish brown	White
Schizophyllum commune Fries; Schizophyllaceae	Schyzophyllan	Grows on branches of dead wood and cut timber	Whitish to greyish	White
Trametes versicolor (L.) Pilat; Polyporaceae	Krestin, proteoglycan	Grows on cut timber and fallen logs	Light browns to greenish brown	White
Hericium coralloides (Scop.) Pers; Hericiaceae	Galacto-Xyloglucan	Grows on dead trunks of trees	Creamy white	White
Coprinus comatus (O.F. Mull.)Pers.*;Agaricaceae*	Fucogalactan, hydrosoluble heteroglucan	Grows on dead and decaying leaves	Dark chocolate brown to almost black	Black
Ganoderma lucidum (Curtis) Kummer; Ganodermataceae	Ganodermin and Ganoderan	Grows on cut timber and fallen logs	light to dark chocolate brown	Reddish brown
Pleurotus ostreatus (Jacq. ex Fr.) P.Kumm.*; Pleurotaceae*	Pleurostrin Peptide, molecular weight of 7kDa	Grows on cut timber and fallen logs	Creamy white	White
Collybia dryophila (Bull.) P. Kumm;Tricholomataceae	Collybial	Grows on dead and decaying leaves	Whitish brown to dark brown	White
Armillaria mellea (Vahl) P.Kumm.; *Physalacriaceae*	Protoilludanes	Grows on dead and decaying leave	Yellowish	White
Cymatoderma elegans Jungh; Meruliaceae	Polysaccharides	Grows on dead trunks of trees	Whitish brown	White
Inonotus glomeratus (Peck) Murrill; Hymenochaetaceae	Xylo-Galactoglucan	Grows on dead trunks of trees	Whitish Brown	Yellow to brownish
Marasmius androsaceus (L.) Fr.; Marasmiaceae	Marasmanes	On dead and decaying logs	White	White
Amanita muscaria (L.) Lam.; Amanitaceae	Homoglucans	On dead and decaying logs	Brown	White
Auricularia auricula-judae (Bull.) Wettst.; Auriculariaceae	Homoglucans	On dead and decaying logs	Creamy grey to brown	White
Volvariella volvacea (Bul.) Singer; Pluteaceae	Homoglucans	Grows on dead and decaying leaves	Grayish brown	Brownish pink
Sparassis crispa (Wulfen) Fr.; Sparassidaceae	Homoglucans	On dead and decaying logs	White	White

***Figure-C: Different Macro fungi found during 2014-16 at Raiganj Wildlife Sanctuary, West Bengal, India*:** 1= *Lentinus edodes;* 2= *Schyzophyllum commune*; 3= *Trametes versicolor*, 4= *Hericium coralloides*; 5= *Coprinus comatus*; 6= *Ganoderma lucidum*; 7= *Pleurotus ostreatus*; 8= *Coprinus sp*; 9= *Collybia dryophila*; 10= *Armillaria mellea*; 11= *Cymatoderma elegans*; 12= *Inonotus sp*; 13= *Marasmius sp*; 14= *Amanita muscaria*; 15= *Auricularia auricular*, 16= *Volvariella volvacea*; 17= *Sparassis crispa*

Table-2: Different Macro fungi and their target organisms and biological activities

Macro fungi	***Target organism and Biological activity***	***References***
Lentinus edodes (Berkeley) Pegler; Marasmiaceae	Antifungal: *Candida albicans*, antibacterial: *Mycobacterium tuberculosis*, *Listeria monocytogenes*, *S. aureus*, *M. luteus, B. cereus* antiviral: Herpes simplex-a type 1	Stamets, 2002; Kitzberger et al., 2007; Chen & Seviour, 2007; Stamets, 2002
Schizophyllum commune Fries; Schizophyllaceae	Antifungal: *Candida albicans*, antibacterial: *S. aureus*.	Stamets, 2002
Trametes versicolor (L.) Pilat; Polyporaceae	Antifungal effect: *C. albicans*	Stamets, 2002; Kitzberger et al., 2007
Coprinus comatus (O.F. Mull.)Pers. *;Agaricaceae*	Showing antibacterial activity	Fan *et al*., 2006
Ganoderma lucidum (Curtis) Kummer; Ganodermataceae	Antifungal to phytopathogens *Botrytis cinerea*, *Fusarium oxysporum* and *Physalospora piricola*	Wang & Ng, 2006
Pleurotus ostreatus (Jacq. ex Fr.) P.Kumm.*; Pleurotaceae*	Antifungal effect : *Fusarium oxysporum*, *Mycosphaerella arachidicola* and *Physalospora piricola*	Chu *et al*., 2005
Collybia dryophila (Bull.) P. Kumm; Tricholomataceae	Low antifungal, high antibacterial *(Bacillus* sp.), high antiviral, cytotoxic, nonselective antibiotic, have antitumour polysaccharides, (1→3)-ß-glucan	Pacheco-Sanchez *et al.,* 2006;
Armillaria mellea (Vahl) P.Kumm.; *Physalacriaceae*	Prevent trombocite aggregation, cytotoxic, antimicrobial	Misiek and Hoffmeister, 2012
Cymatoderma elegans Jungh; Meruliaceae	Anticarcinogenic properties	Ohtsuka *et al.,*1973
Marasmius androsaceus (L.) Fr.; Marasmiaceae	Antibacterial less antifungal, cytotoxic and phytotoxic lower antibiotic and cytotoxic high antibacterial, antifungal & cytotoxic	Anke *et al*., 1981
Amanita muscaria (L.) Lam.; Amanitaceae	Antitumour polysaccharides	Kiho *et al*., 1992
Auricularia auricula-judae (Bull.) Wettst.; Auriculariaceae	Antitumour polysaccharides	Misaki and Kakuta, 1995
Sparassis crispa (Wulfen) Fr.;	Antitumour polysaccharides	Ohno *et al.,* 2000

Sparassidaceae		
Volvariella volvacea (Bul.) Singer; *Pluteaceae*	Antitumour polysaccharides	Kishida *et al., 1989*
Inonotus glomeratus (Peck) Murrill; Hymenochaetaceae	Antitumour polysaccharides	Mizuno *et al., 1999*
Hericium coralloides (Scop.) Pers; Hericiaceae	Antitumour polysaccharides	Mizuno *et al.*, 1992

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