DR. RASHMI DUBEY
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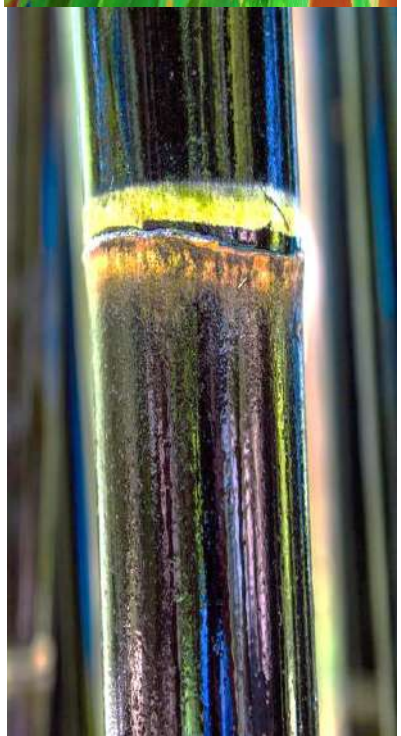
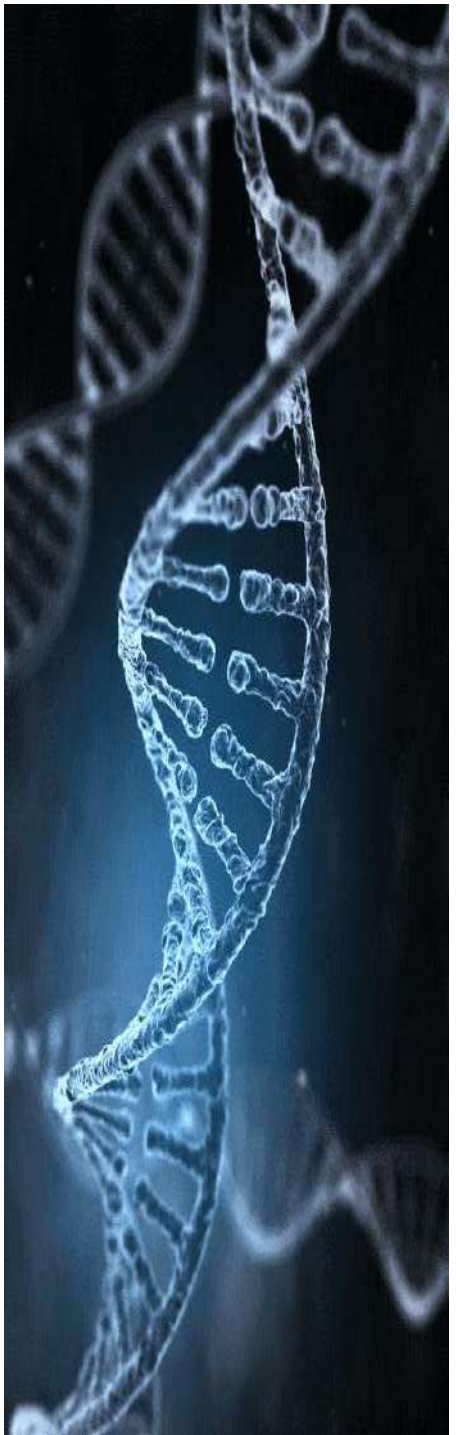


ANNUAL PROGRESS REPORT

(2020 –21)

Bambusicolous Fungi of Goa

(01.04.2020-31.03.2024)



CONTEXTUAL OF THE PROJECT

Bamboo, are currently classified in a subfamily *Bambusoideae* within the extensive grass family *Poaceae*.

As per FSI 2019 report there are about **90 genera comprising of 1,200 species** of Bamboo (worldwide).

India is the world's second largest cultivator of Bamboo after China, **with 125 indigenous and 11 exotic species** of bamboo from 23 genera and spread over 13.96 million hectares of land (FSI, 2019).

Bamboo contributes significantly to the social, economic & ecological development of any region. It is a universally used plant and contributes to subsistence needs of more than 2.5 billion people.

Bamboo also plays an important role in carbon sequestration, bio-diversity and soil moisture conservation

Bambusicolous fungi, which embodies fungi growing on any bamboo substrates, including leaves, culms, branches, sheathes, rhizomes, and roots (Kevin D. Hyde 2000).

The fungal associations have diverse habits- as saprobes, pathogens and also as symbiont

About **170 species of bamboo** are infected by Fungal diseases (Mahanan 1997). The genera of bamboo with the highest numbers of fungi recorded globally are ***Arundinaria, Bambusa, Dendrocalamus*** *sp.,* ***Phyllostachys*** **and** ***Sasa*** **sp.** *Gigantochloa* sp., *Melacona* sp., *Ochlandra* sp., *Oxytenanthera* sp., *Phyllostachys* sp., *Schizostachyum* sp., *Thamnocalamus* sp., *Thyrsostachys* sp. *Bambusa and Dendrocalamus* in particular have been found to support a high fungal diversity.

Potential fungal diseases affecting the productivity of Bamboo includes rot of emerging, and growing culms, bamboo blight, thread blight, witches broom, little leaf diseases, leaf rust, foliage blight, seedling leaf blight, leaf spots and basal rot.

DIVERSITY

China and Taiwan - Kuai 1996 reported 190 pathogenic bambusicolous fungi in Mainland China and Taiwan.

Hong Kong- Zhou etal., 2000. reviewed 189 species belonging to 75 genera of bamboo fungi from main land China 79 species of 58 genera from Hong Kong

In Japan fewer than 86 bamboo taxa was reported.

A list of diseases on bamboo is provided by Boa (1967), while Eriksson and Yue in 1988 examined all ascomycetes described as new species from bamboo.

In India Bamboo diseases studies were made by Mohanan & Liese (1990) and Mohanan (2002) Hosagoudar 1985, 1994, 2002, 2005, 2013

PATHOGENICITY

FOLIAGE DISEASE - Ascomycota showed the highest record of the foliage disease incidence of about 64.99%.

Inflorescence Disease: Mainly by Ustilaginales (50 %)

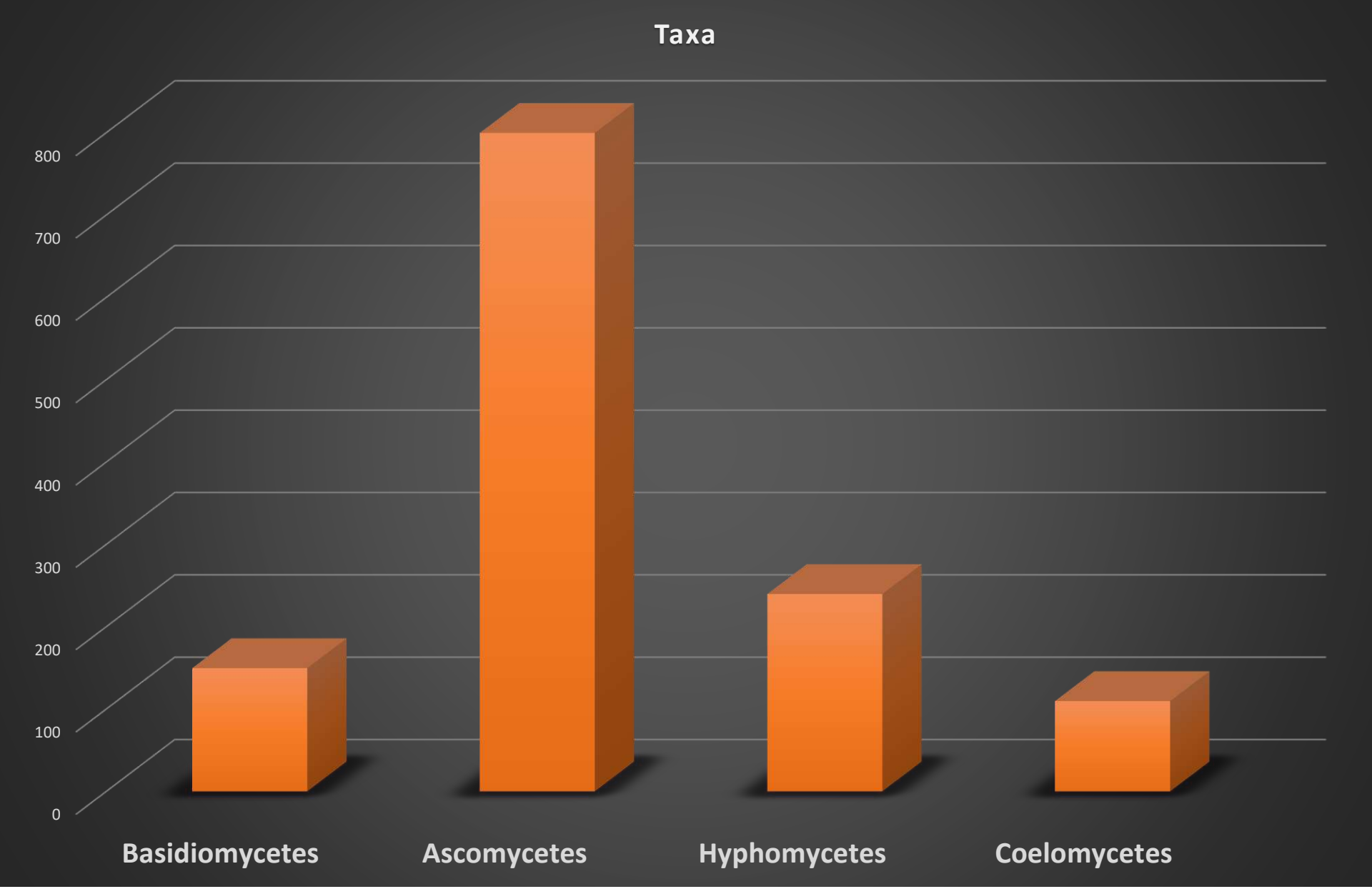
CULM DISEASES was highest by Basidiomycota (26.25%) Coelomycota and Myxomycetes

ROOT AND RHIZOME –Order Polyporales (42.86%)

BENEFICIAL IMPACTS

Some bambusicolous fungi are also medicinal.

Engleromyces goetzii Henn., *Hypocrella bambusae* (Berk. & Broome) Sacc. and *Shiraia bambusicola* Henn. *Dictyophora indusiata* (Vent.) Desv are used in traditional Chinese medicines to treat various human diseases (Ying *et al.*, 1987).



Worldwide 1300 species of Bambusicolous fungi have been reported (Dong-Qin Dai, et al. 2018)

WHY ?

1. Cultivation of economically important bamboos is often threatened by fungal infection and diseases which eventually result in serious damages on bamboo cultivation.
2. The complex lifestyle of bamboo species which encompasses fast growth, giant height, often growing in difficult terrain, limits investigation and hinders awareness on bamboo pathology
3. Most bamboo species are in the wild and not domesticated for phytopathological scrutiny
4. No consolidate reports are there on the bambusicolous fungi complex using morphological characters and Molecular systematics to identify the taxa in India.
5. Out of 1300 spp. reported only 180 spp. are sequenced
6. Phylogenetic analyses of bamboo fungi need more study and should focus on protein genes.

THUS, It is recommended that fresh specimens needs to be collected, and the existing species should be designated as reference specimens.

DURATION	01.04.2020-31.03.2024
OBJECTIVES	1. To explore the diversity of bambusicolous fungi of Goa. 2. Morphological identification of the bambusicolous fungal species along with Scanning Electron Microscopic studies. 3.Isolation and Molecular characterisation of bambusicolous fungal species. 4. To evaluate the validity of bambusicolous fungal taxa and clarify their phylogenetic relationships by Multigene sequencing. (the combined sequences of data with multi-genes (LSU (large subunit rDNA), SSU (small subunit rDNA), TEF (translation elongation factor 1-α gene region), and beta-tubulin). 5.Cataloguing, preservation and maintenance of fungal germplasm.

OUTCOME

Thus this work will strength the existing data base of mycofloristics, taxonomy, molecular systematics, conservation and utilization of Bambusicolous fungi at National and International Level. Thus, Functional genomics methodologies will provide an opportunity for a comprehensive understanding of complex of Bambusicolous Fungi. The studies will also be helpful to the farmers in solving certain Pathological issues

Study area

Goa is a part the Konkan, which is a ridge rising up to the Western Ghats of India.

It lies between the latitudes 14°53’54” N and 15°40’00” N and longitudes 73°40’33” E and 74°20’13” E.

Encompasses an area of 3,702 km^2 (1,429 sq mi). Equatorial forest cover in state stands at 1,424 km^2 (549.81 sq mi).

The state of Goa ranks seventh for Bamboo availability in India. 25.61 % per cent of total forest area of Goa is covered by bamboo forests which covers 0.03-million-hectare area(As per Selvan (2018).

Protected areas : Bondla Wildlife Sanctuary, Mahadei Wildlife Sanctuary, Bhagwan Mahavir Wild Life Sanctuary, Cotigao Wildlife Sanctuary, Netravali Wildlife Sanctuary, Chorla Ghat, Chandreshwar hills, Dr.Salim Ali Bird Sanctuary, Mollem NP Morpirla forests region having a good vegetation of Bamboo sp.
The main bamboo are the species of *Dendrocalamus and Bambusa* ·

Location of Goa in India

Forest areas

TABLE 4.2 State/ UT wise Number of species of Trees, Shrubs and Herbs

Table Showing Number of Species observed during the Rapid Assessment of Biodiversity					
S. No.	StateName	Trees	Shrubs	Herbs	Total Number of Plant Species
		No of Species	No of Species	No of Species	
1.	Andhra Pradesh	242	64	58	364
2.	Arunachal Pradesh	110	435	192	737
3.	Assam	143	149	153	445
4.	Bihar	113	42	52	207
5.	Chhatisgarh	129	48	50	227
6.	Delhi	16	11	36	63
7.	Goa	118	50	38	206
8.	Gujarat	102	37	73	212
9.	Haryana	45	43	50	138

TABLE 4.5 State/ UT wise and Forest type wise Shannon-Wiener Index for Herbs

S. No.	State Name	1-Tropical Wet Evergreen Forests	2-Tropical Semi-Evergreen Forests	3-Tropical Moist Deciduous Forests	4-Littoral and Swamp Forests	5-Tropical Dry Deciduous Forests	6-Tropical Thorn Forests
1.	Andhra Pradesh			2.89	*	2.63	2.25
2.	Arunachal Pradesh	2.99	4.05	3.09			
3.	Assam	3.16	3.47	2.85	2.38	2.82	
4.	Bihar		2.85	2.02	2.72	1.21	
5.	Chhattisgarh			2.59		2.61	
6.	Delhi					*	3.38
7.	Goa	2.61	2.28	1.83	0.67	0.41	
8.	Gujarat			2.02	1.80	3.30	2.58

TABLE 8.3 State/UT wise Distribution of Bamboo Area in Recorded Forest Area

(in sq km)

S. No.	State/UTs	Bamboo bearing area	Bamboo bearing area as per ISFR 2017	Change in area with respect to ISFR 2017
1.	Andhra Pradesh	7,003	7,578	-575
2.	Arunachal Pradesh	14,981	15,125	-144
3.	Assam	10,525	8,955	1,570
4.	Bihar	1,136	1,004	132
5.	Chhattisgarh	11,255	11,060	195
6.	Goa	418	382	36
7.	Gujarat	3,393	3,544	-151
8.	Haryana	72	21	51
9.	Himachal Pradesh	650	540	110
10.	Jharkhand	4,123	4,470	-347
11.	Karnataka	10,181	10,442	-261
12.	Kerala	2,849	3,484	-635
13.	Madhya Pradesh	20,867	18,167	2,700
14.	Maharashtra	15,408	15,927	-519
15.	Manipur	9,903	10,687	-784
16.	Meghalaya	5,410	5,943	-533
17.	Mizoram	3,476	3,267	209
18.	Nagaland	4,284	6,025	-1,741
19.	Odisha	11,827	12,109	-282
20.	Punjab	255	44	211
21.	Rajasthan	1,874	1,976	-102
22.	Sikkim	1,176	553	623
23.	Tamil Nadu	4,357	4,154	203

Field Tours	No Field Tours were undertaken Although few samples were collected during the field tours to Goa under SERB (external funding project) project in January 2020 and work was started.



Salim Ali Wildlife Sanctuary

Mhadei Wildlife Sanctuary

Gululem, Mhadei WLS

Surla, Mhadei WLS

Kudal, Mhadei WLS

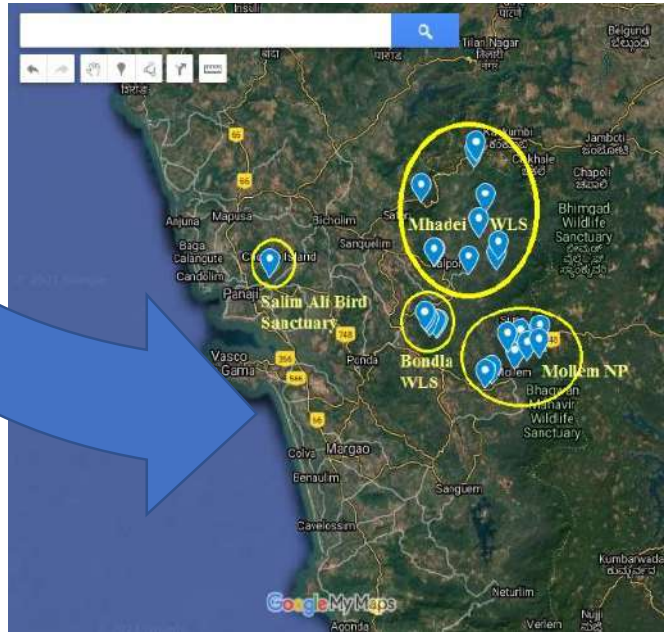
Honda, Mhadei WLS

Saverdem, Mhadei WLS

Satrem, Mhadei WLS

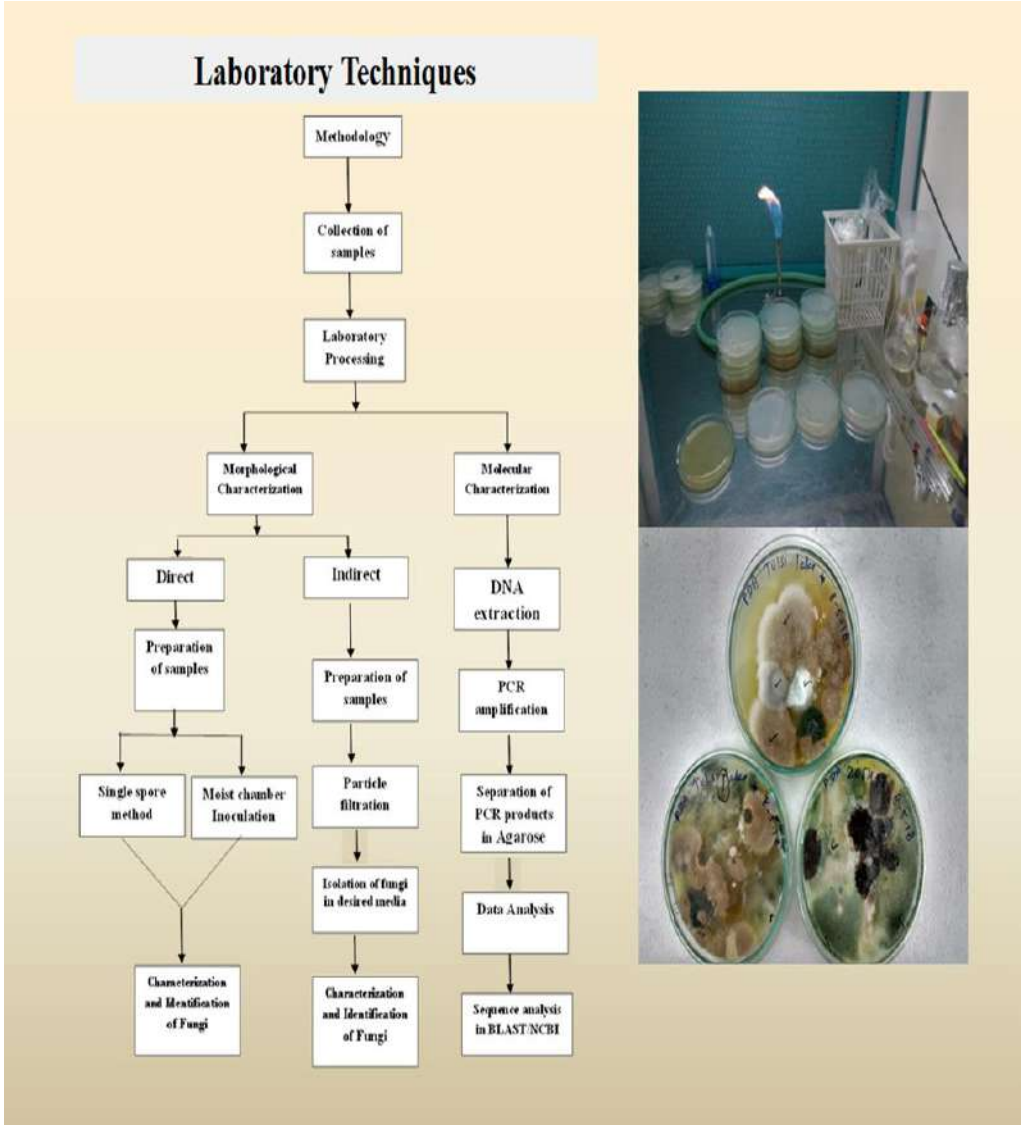
Goa-Karnataka Border, Mhadei

Karanzel, Mhadei WLS



QGIS Map showing the areas covered

Methodology

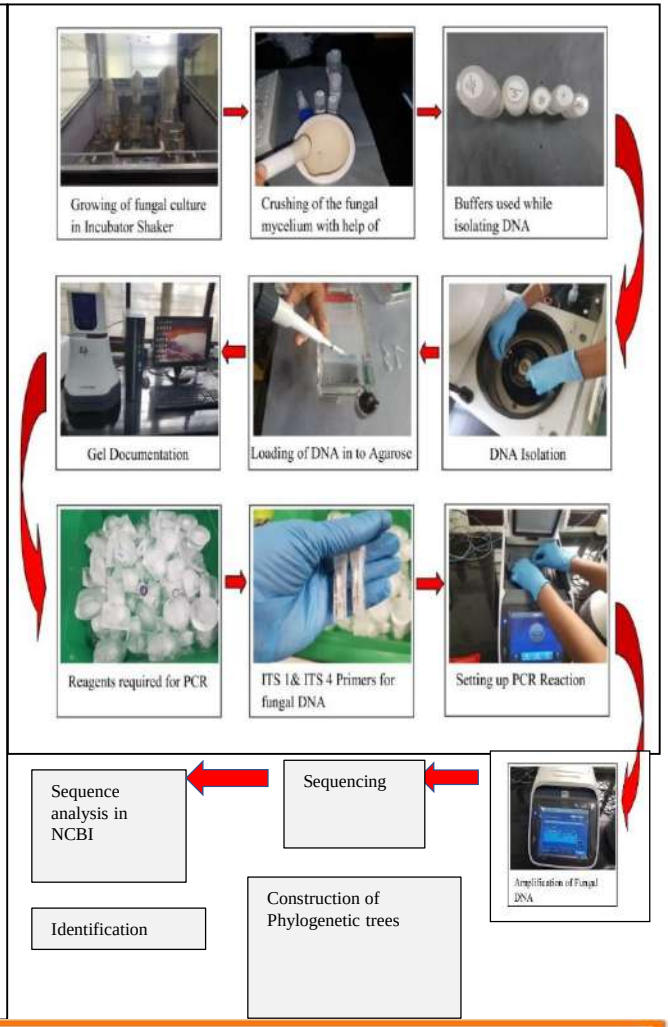


Outline of Methodology

Different parts of Bamboo infested with Fungi

LABORATORY ANALYSIS

Fig.3 Particle filtration method



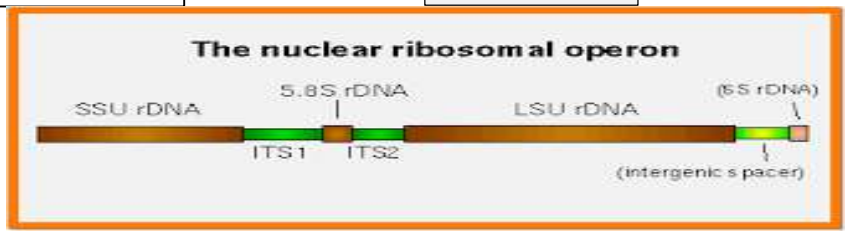


Fig.5 Molecular laboratory at BSI, WRC, Pune

Outline of Work accomplished in 2020-21

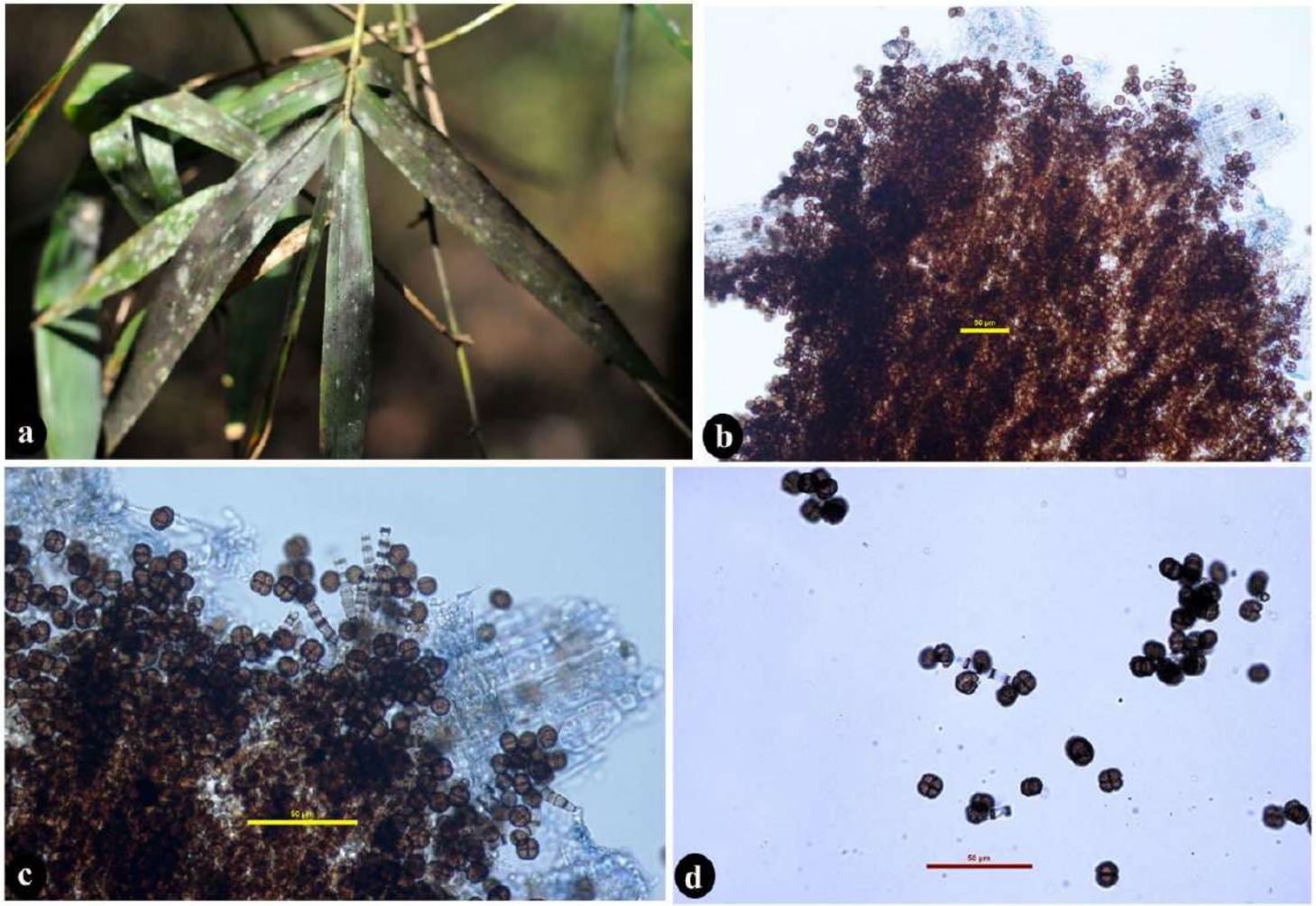
Field Tours	Two field tours were proposed this year, but field tour was not materialised, due to crisis of Funds in DTE. Although 2-3 field tours will be undertaken this year to equilibriums the project work. Although few samples were collected during the field tours to Goa under SERB (external funding project) project in January 2020 and work was started.
Herbarium Consultation Tour	National Fungal Culture Collection Centre (NFCC), Agarkar Research Institute, Pune and University of Pune on 23.12.2020 and 24.12.2020.
Literature survey	Literature survey was conducted for Bambusicolous fungi from different sources.
Number of specimens collected:	22 samples
Number of Fungal specimens identified	21 spp.
No. of fungal specimens brought in Culture	16 samples cultures were cultured and subcultures on Malt Extract Agar, Potato carrot Agar media, Oat meal Agar
No. of photo micrographs taken during this period	More than 100
Scanning Electron Microscopic	07 sp.

List of Fungal species documented 2020 - 21

S.no	Name	Taxonomic position	Host (Bamboo)	Collection site
1	*Acrodictys bambusicola* M.B. Ellis	Pezizomycotina	Dead Bamboo	Dr. Salim Ali Bird Sanctuary,Goa
2	*Beltraniella spiralis* Piroz. & S.D. Patil, 1966	Beltraniaceae	Dead Bamboo	Mhadei WLS, Goa
3	*Corynespora cassiicola* (Berk. & M.A. Curtis) C.T. Wei 1950-	Corynesporacaceae	Leaves	Bondla WLS
4	*Chaetosphaerulina lignicola*	Tubeufiaceae	Bamboo stem	Mhadei WLS, Goa
5	Diatrype loranthi Tend. 1971	Diatrypaceae	Bamboo stem	Mhadei WLS, Goa
6	*Fusarium proliferatum* (Matsush.) Gerlach & Nirenberg (1982)	Nectriaceae	Leaves	Dr. Salim Ali Bird Sanctuary, Goa
7	*Fusarium chlamydosporum* Wollenw. & Reinking, 1925	Nectriaceae	Leaf	Mhadei WLS, Goa
8	*Fusarium incarnatum* (Desm.) Sacc., (1886)	Nectriaceae	Leaves	Dr. Salim Ali Bird WLS, Goa
9	*Exserohilum elongatum* Hern. -Restr. & Crous 2018-.	Pleosporaceae	Dead Bamboo stem	Bondla WLS
10	*Kamalomyces bambusicola* Y.Z. Lu & K.D. Hyde, 2017	Dothideomycetes	Dead bamboo stem	Dr. Salim Ali Bird Sanctuary,Goa
11	*Lasiodiplodia theobromae* (Pat.) Griffon & Maubl., (1909),	Botryosphaeriaceae	Leaves	Dr. Salim Ali Bird WLS, Goa

S.no	Name	Taxonomic position	Host (Bamboo) parts	Collection site
12	*Monodictys putredinis* (Wallr.) S. Hughes,	Pezizomycotina	bamboo stem	Dr. Salim Ali Bird Sanctuary,Goa
13	*Nigrospora sphaerica* (Sacc.) E.W. Mason, (1927)	Sordariomycetes	Leaves	Mhadei WLS, Goa
14	*Paradictyoarthrinium diffractum* Matsush. 1996	Paradictyoarthriniaceae	Dry Bamboo stem	Mhadei WLS, Goa
15	*Pithomyces ellisii* V.G. Rao & Chary 1972-	Didymellaceae	Dry Bamboo stem	Bondla WLS
16	*Phoma herbarum Westend. 1852*	Didymellaceae	Leaves	Dr. Salim Ali Bird Sanctuary, Goa
17	*Sporidesmium ehrenbergii* M.B. Ellis 1958	Amorphothecaceae	Dry Bamboo stem	Bondla WLS
18	*Sporidesmium* sp.	Dothideomycetes	Dry Bamboo stem	Dr. Salim Ali Bird Sanctuary, Goa
19	*Trichoderma asperellum* (1999),	Hypocreaceae	Leaves	Mhadei WLS, Goa
20	*Aspergillus melleus*	Aspergillaceae	Leaves	Mhadei WLS, Goa
21	*Dictyoarthrinium sacchari* (J.A. Stev.) Damon. *Bull. Torrey bot. Club* 80: 164 (1953).	Apiosporaceae	Leaves	Mhadei WLS, Goa

Photo micrographs of fungi reported



Dictyoarthrinium sacchari (a) Infected leaves of *Dendrocalamus strictus* (Roxb.) Nees. (b-c) Conidiophores with Conidia (d) Conidia. [Scale bar: (b-d) = 50 µm]

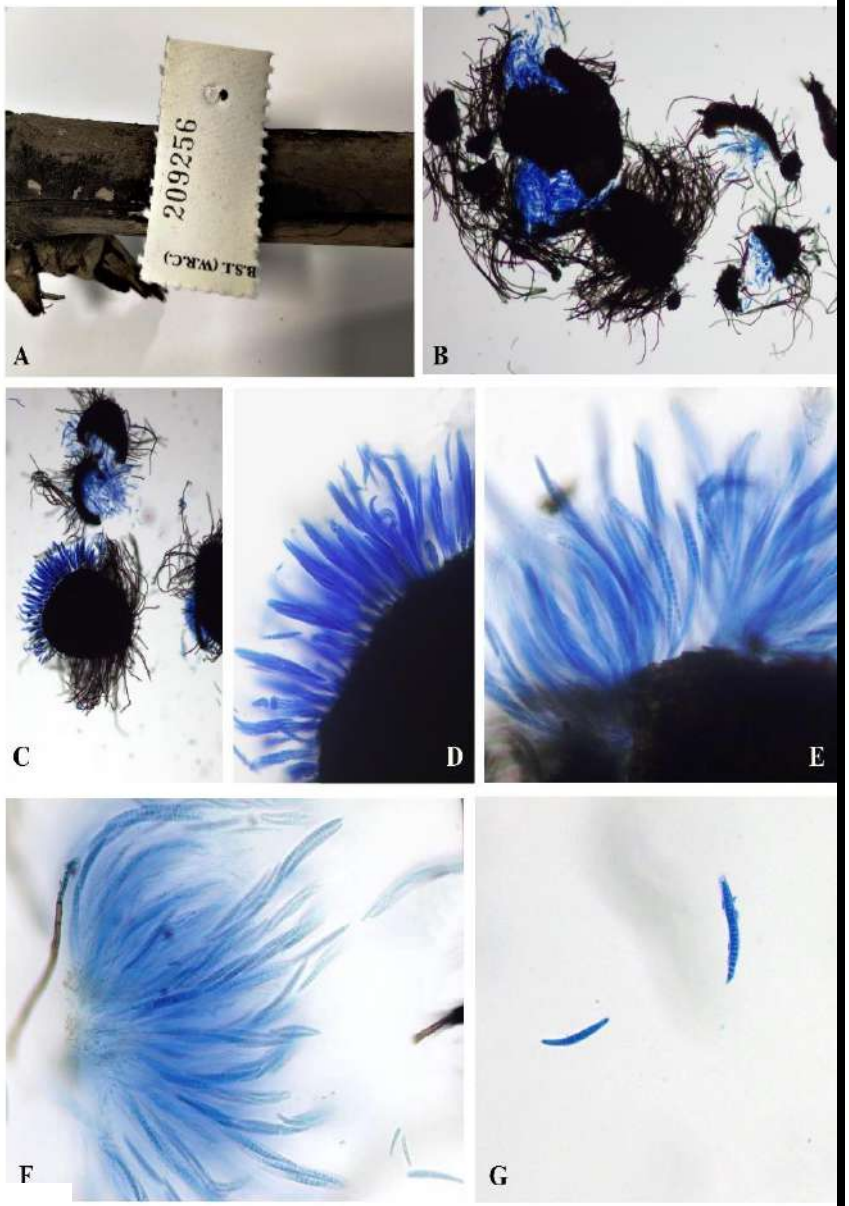


Fig. 52. *Kamalomyces bambusicola* : A. Dead bamboo stem; B-C Asci enveloped in subiculum; D-F Asci with ascospores ; G. Ascospores.

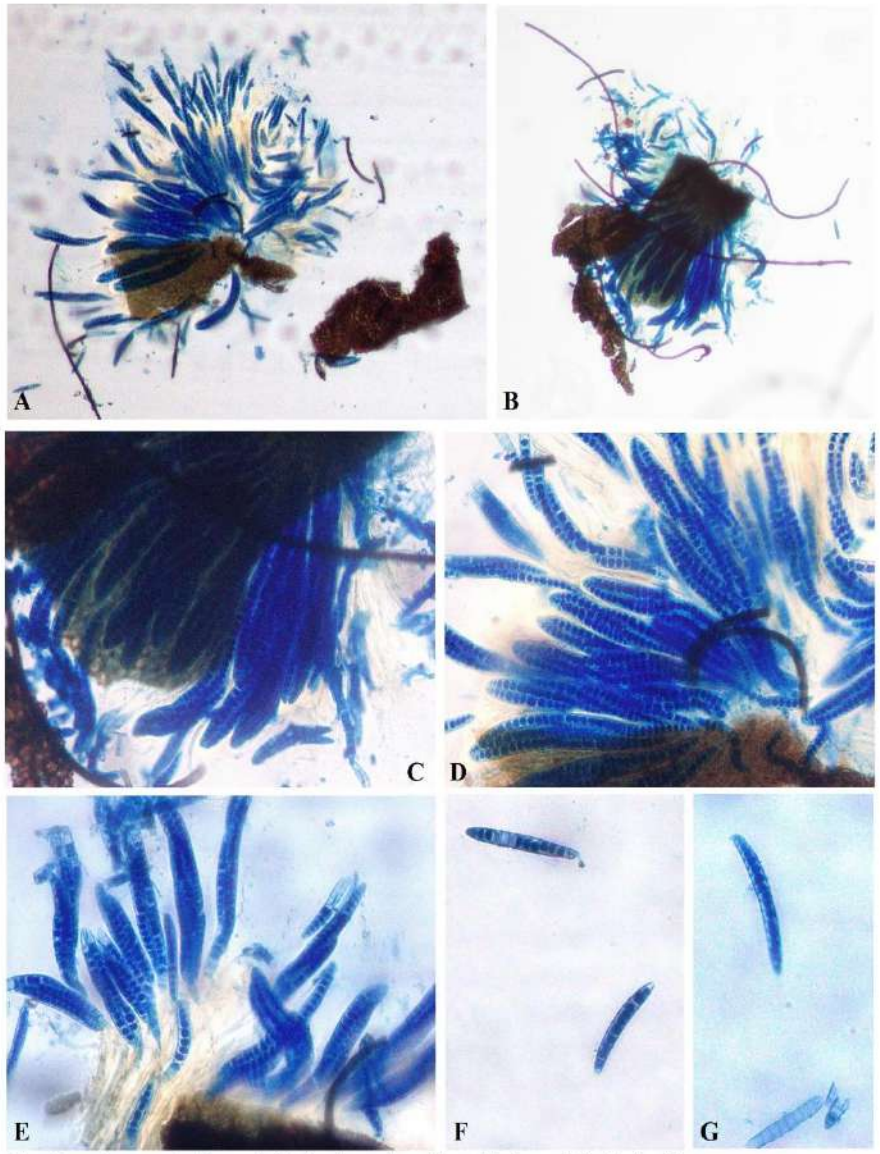


Fig. *Kamalomyces indicus* : A-B. Asci enveloped in subiculum; C-E. Asci with ascospores; F-G. Ascospores.

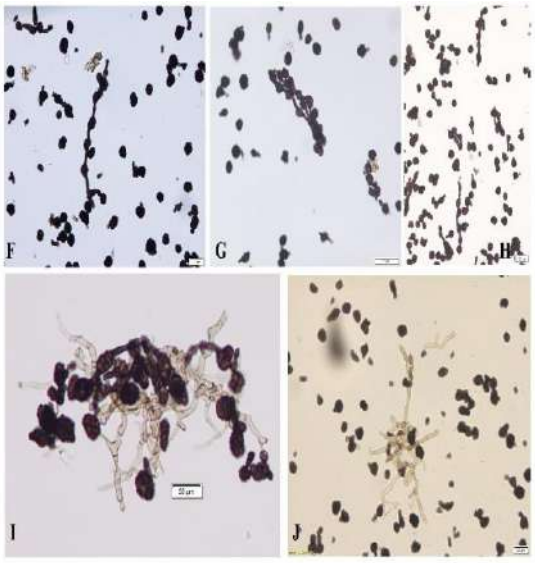
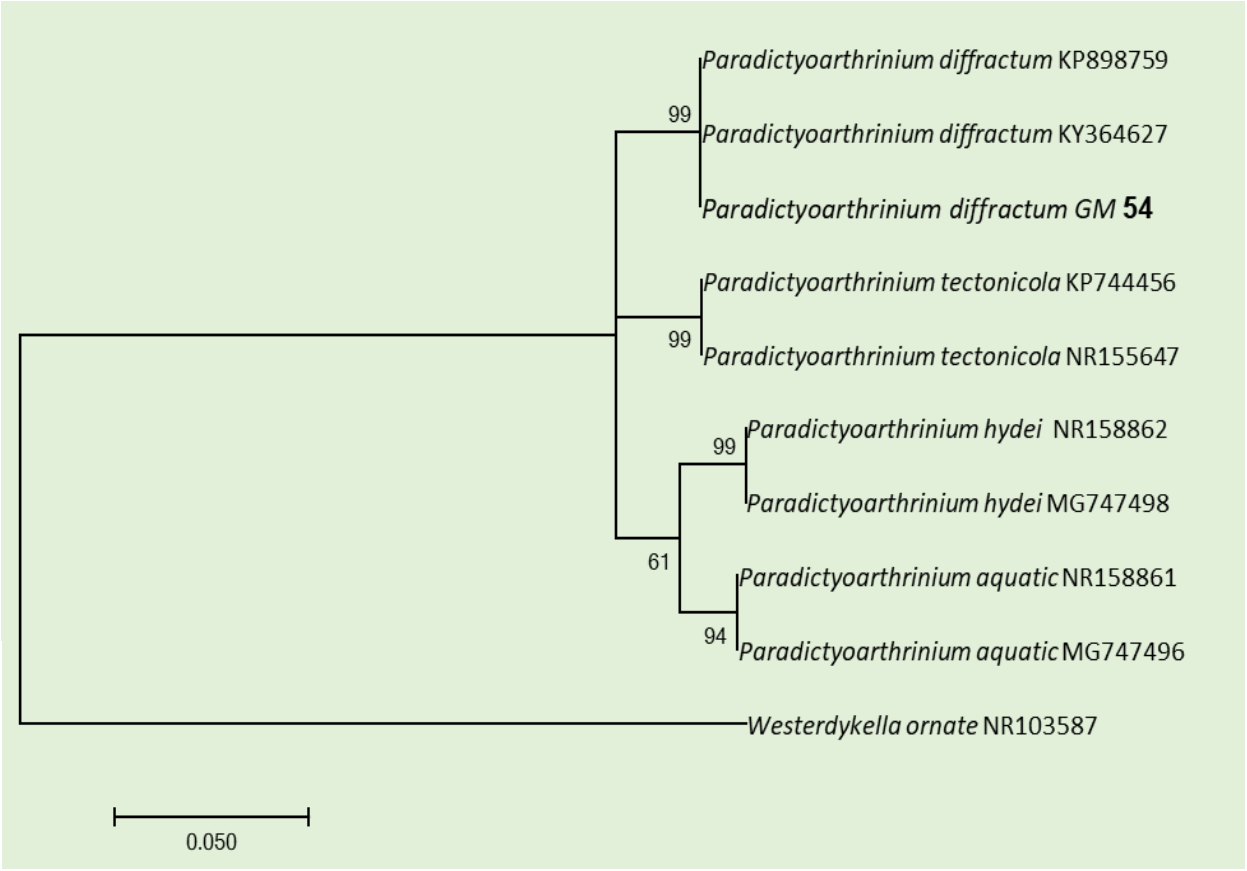


Fig. *Paradictyoarthrinium diffractum*



Molecular Phylogenetic analysis of our strain GM 54 by Maximum Likelihood method

The evolutionary history was inferred by using the Maximum Likelihood method based on the Kimura 2-parameter model [1]. The tree with the highest log likelihood (-1081.4574) is shown. The percentage of trees in which the associated taxa clustered together is shown next to the branches. Initial tree(s) for the heuristic search were obtained automatically by applying the Maximum Parsimony method. A discrete Gaμma distribution was used to model evolutionary rate differences among sites (5 categories (+G, parameter = 0.2042)). The tree is drawn to scale, with branch lengths measured in the number of substitutions per site. The analysis involved 10 nucleotide sequences. There were a total of 454 positions in the final dataset. Evolutionary analyses were conducted in MEGA7 [2].

Paradictyoarthrinium diffractum Matsush. 1996

Corynespora cassicola

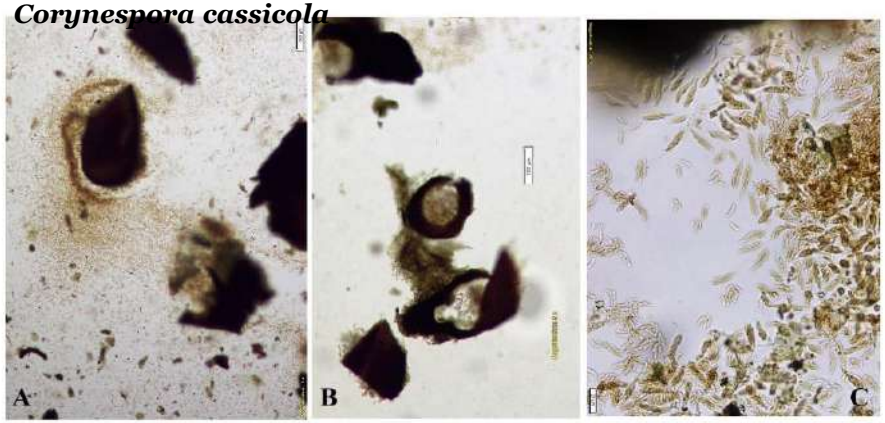


Fig. ***Diatrype loranthii*** (A-C) : A-B. Opened perithecia showing asci and ascospores; C. Asci and Ascospores;

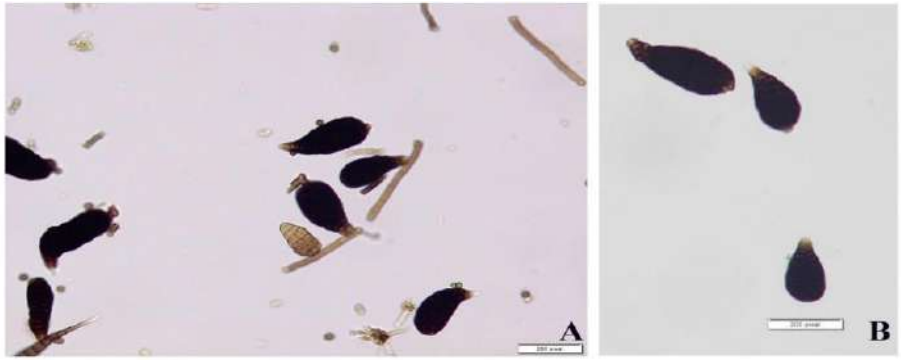
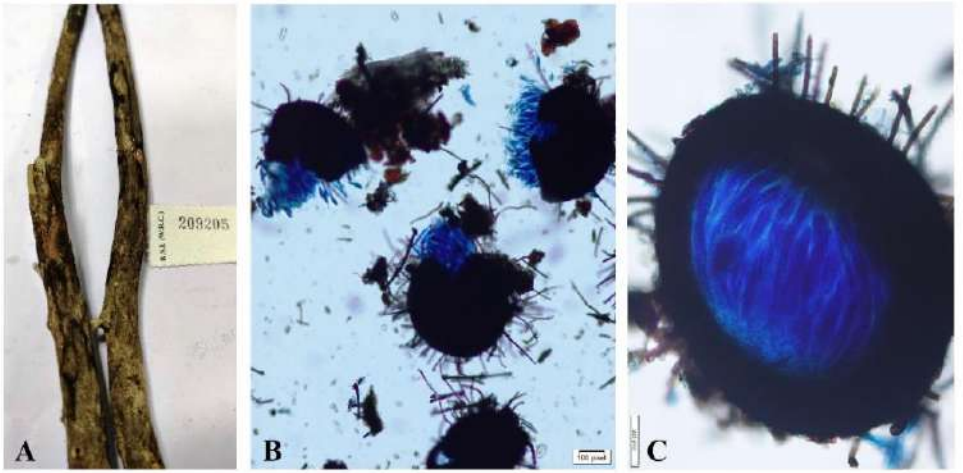
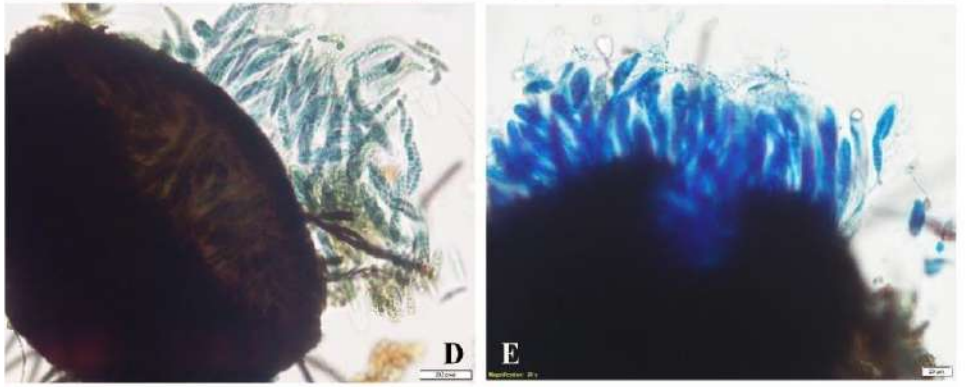


Fig. ***Acrodictys bambusicola***





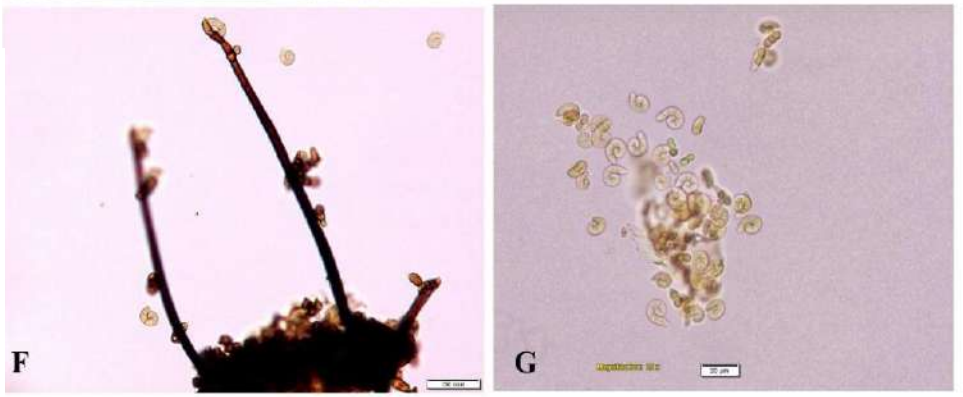


Fig. 22 ***Chaetosphaerulina bambusae*** : A. Dead bamboo twigs; B. Colony of Ascomata; C. Section of Ascomata; D-E. Ruptured Ascomata; F-G. Anamorphic stage (conidiophores and conidia).

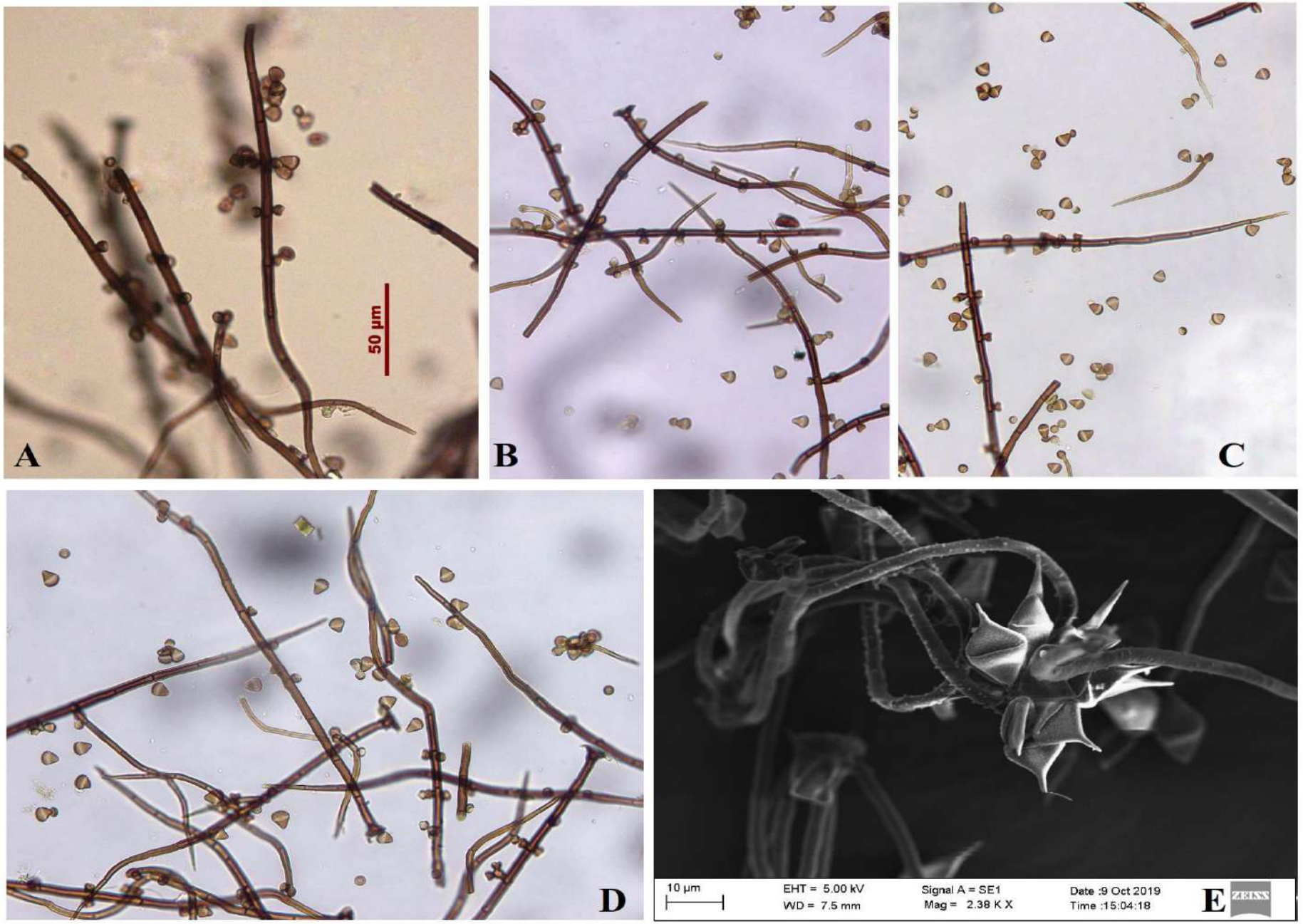
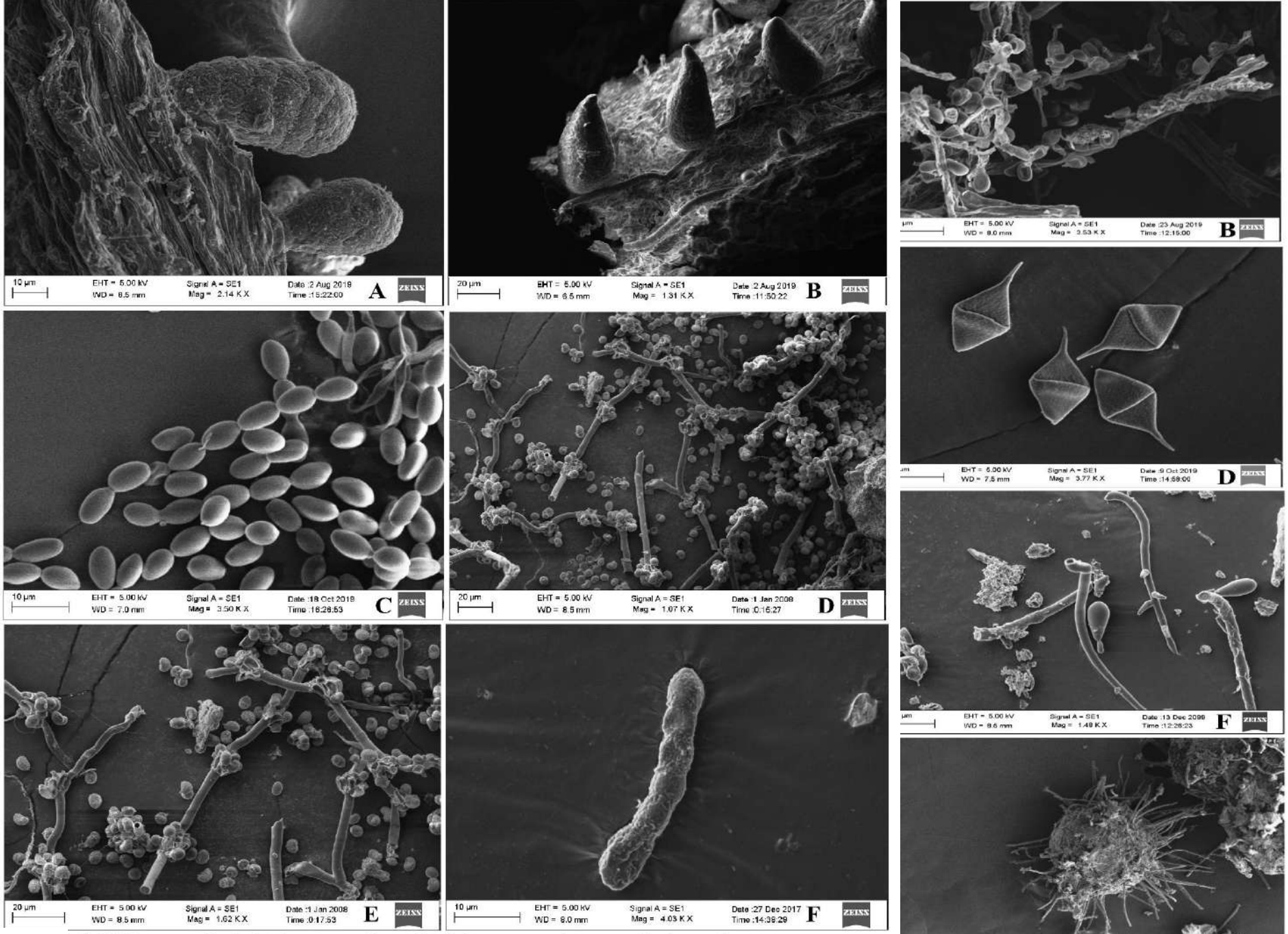
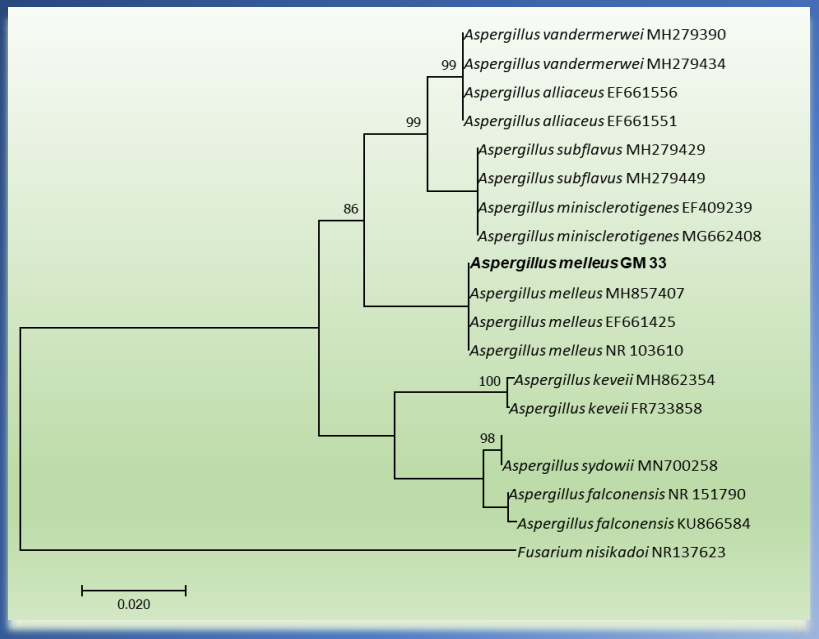


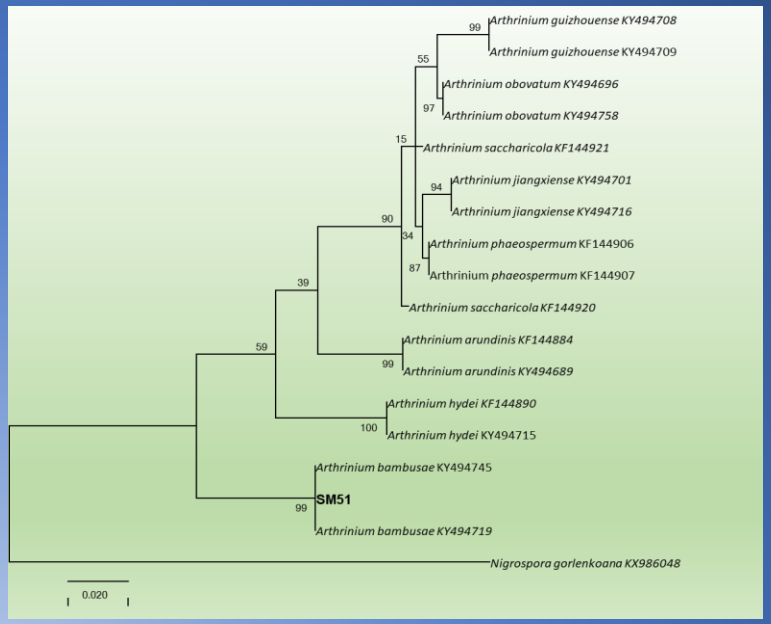
Fig. ***Beltraniella spiralis*** : A-C. Conidiophores with setae and spirally arranged conidia; D. Turbinate conidia; E. SEM image.



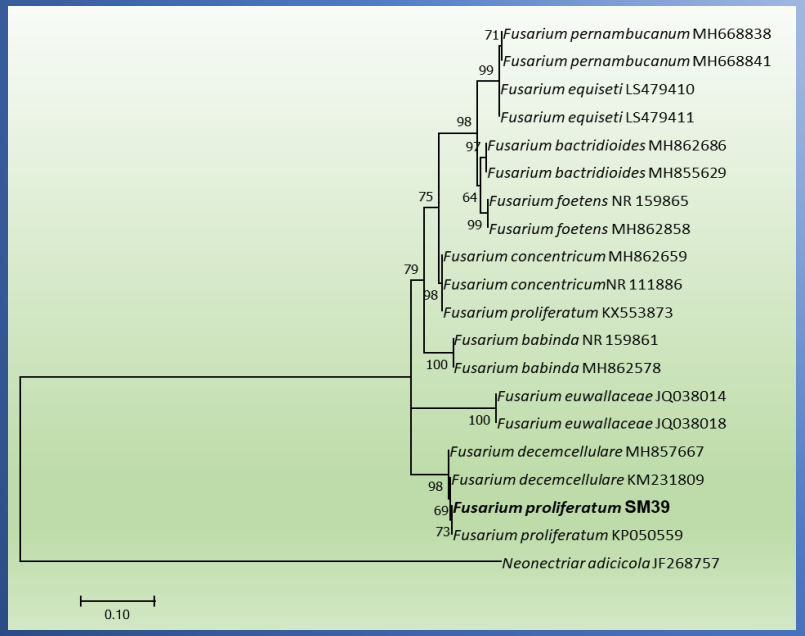
. SEM Images of - A. *Pithomyces ellisii;* B. *Pithomyces pulvinatus*; C. *Sorocybe resinae*; D. *Stachybotrys charatum*; E. *Stachybotrys echinatus*; F. *Taeniolella breviuscula*.



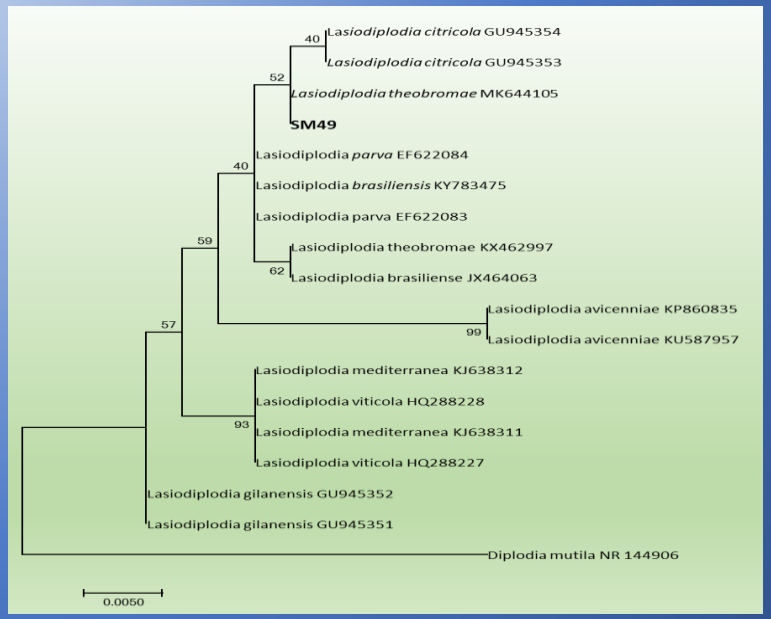
Molecular Phylogeny of *Aspergillus melleus*



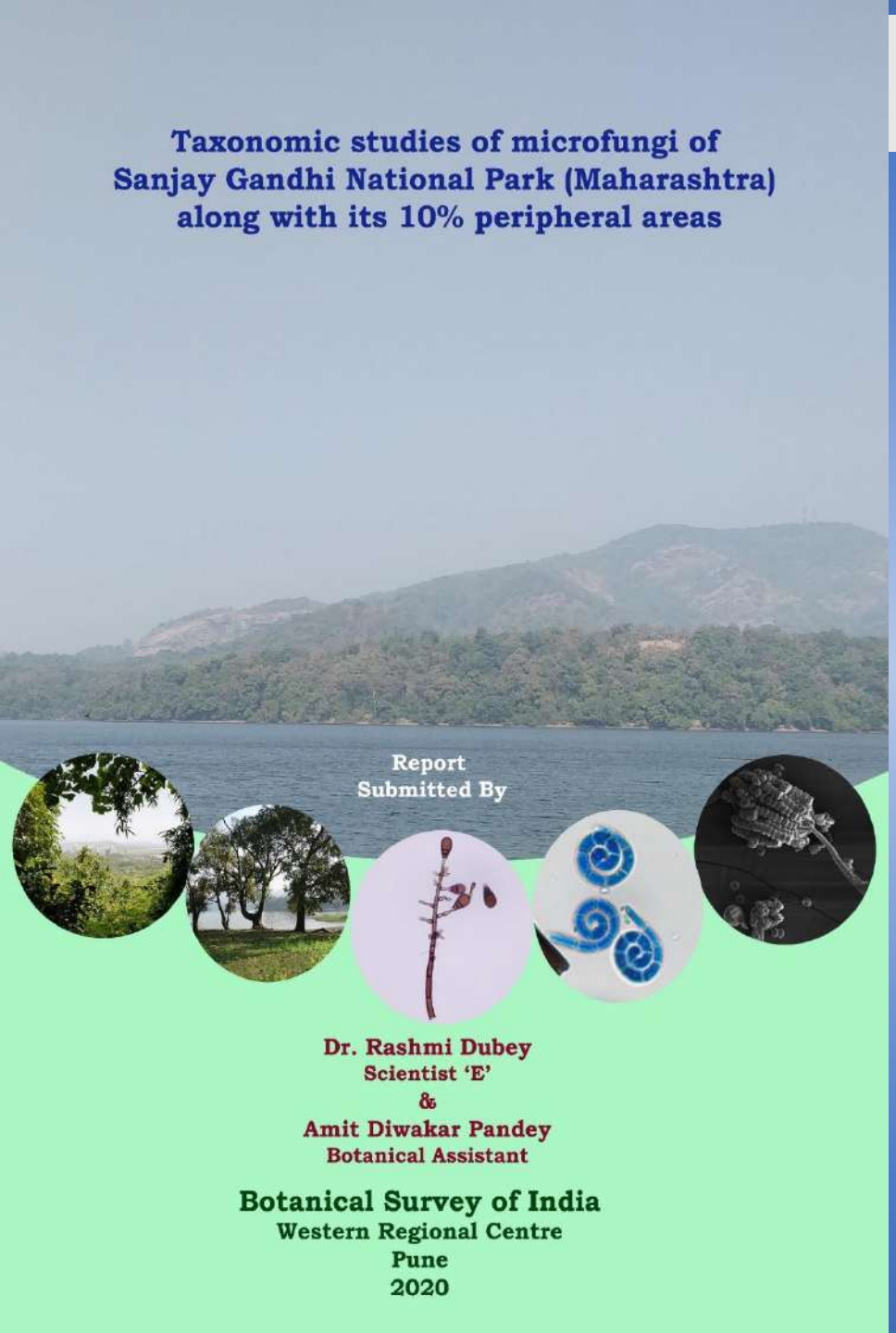
Molecular Phylogeny of *Arthrinium bambusae*



Molecular Phylogeny of *Fusarium proliferatum*



Molecular Phylogeny of *Lasiodiplodia theobromae*



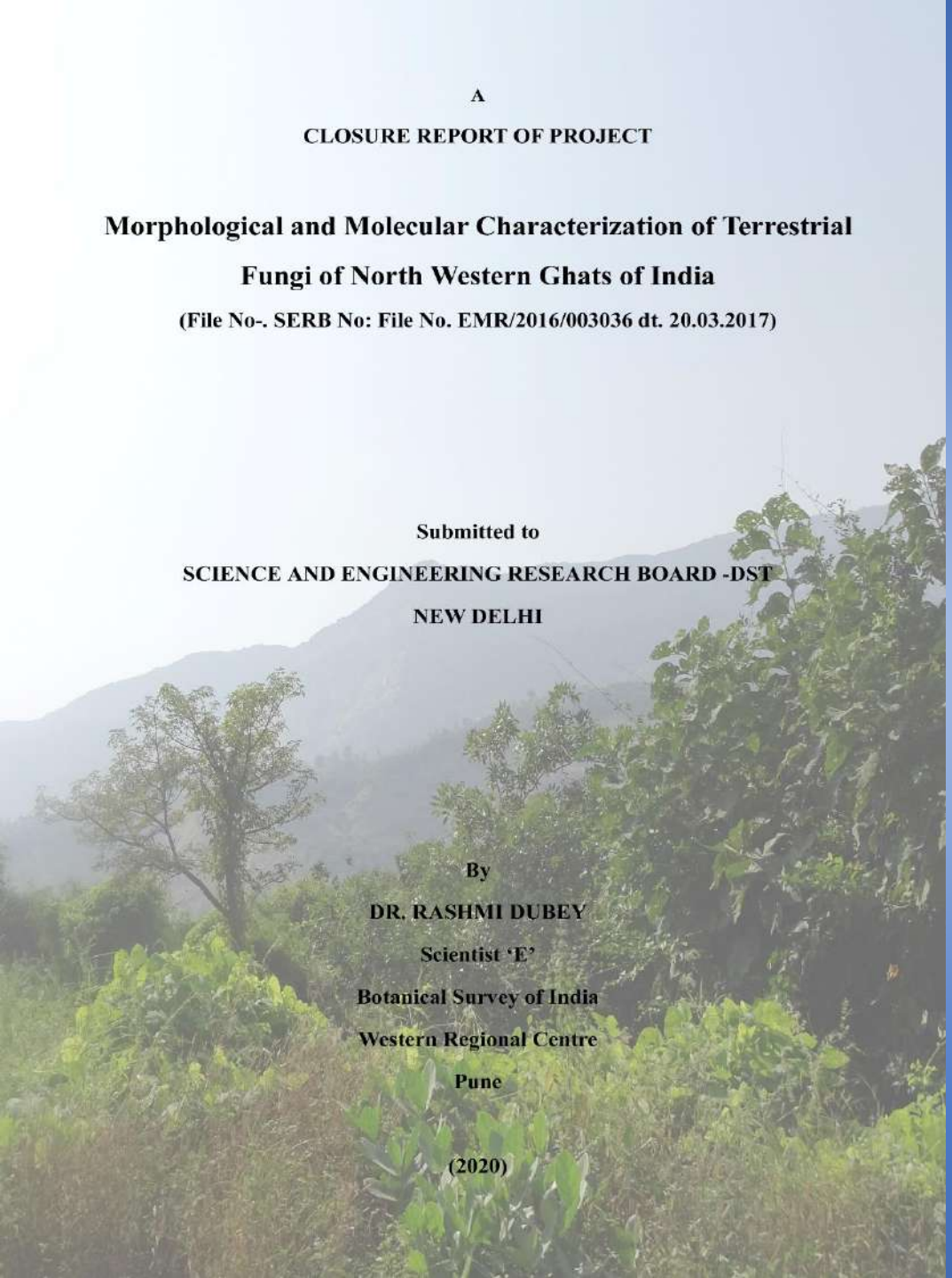
The report was submitted to Director, BSI on 10.09.2020.

In all 334 fungal species (186 litter fungi, 43 foliicolous fungi, 77 soil fungi and 28 water fungal species) was documented.

Scanning Electron Microscopic studies was also conducted for 59 interesting species. Molecular phylogeny was conducted for 39 fungal species. Sanjay Gandhi National Park evidenced as a type location for seven (07) new species and sixteen new records of fungi to India.

07 new species and 16 new records to fungi of India

1.*Brevistachys yeoorensis* sp.nov.
2.*Elotespora mumbiansis* sp.nov.
3.*Heteroconium tulisianensis* sp.nov.
4.*Janetia heterospora* sp.nov.
5.*Mycoenterolobium borivaliensis* sp.nov.
6.*Pseudoacrodictys lignicola* sp.nov.
7.*Sporidesmiella thaneiensis* sp.nov.



The report was submitted to SERB DST on April 2021

In all 256 fungal species Scanning Electron Microscopic studies was also conducted for 46 interesting species.

Molecular phylogeny was conducted for 42 fungal species

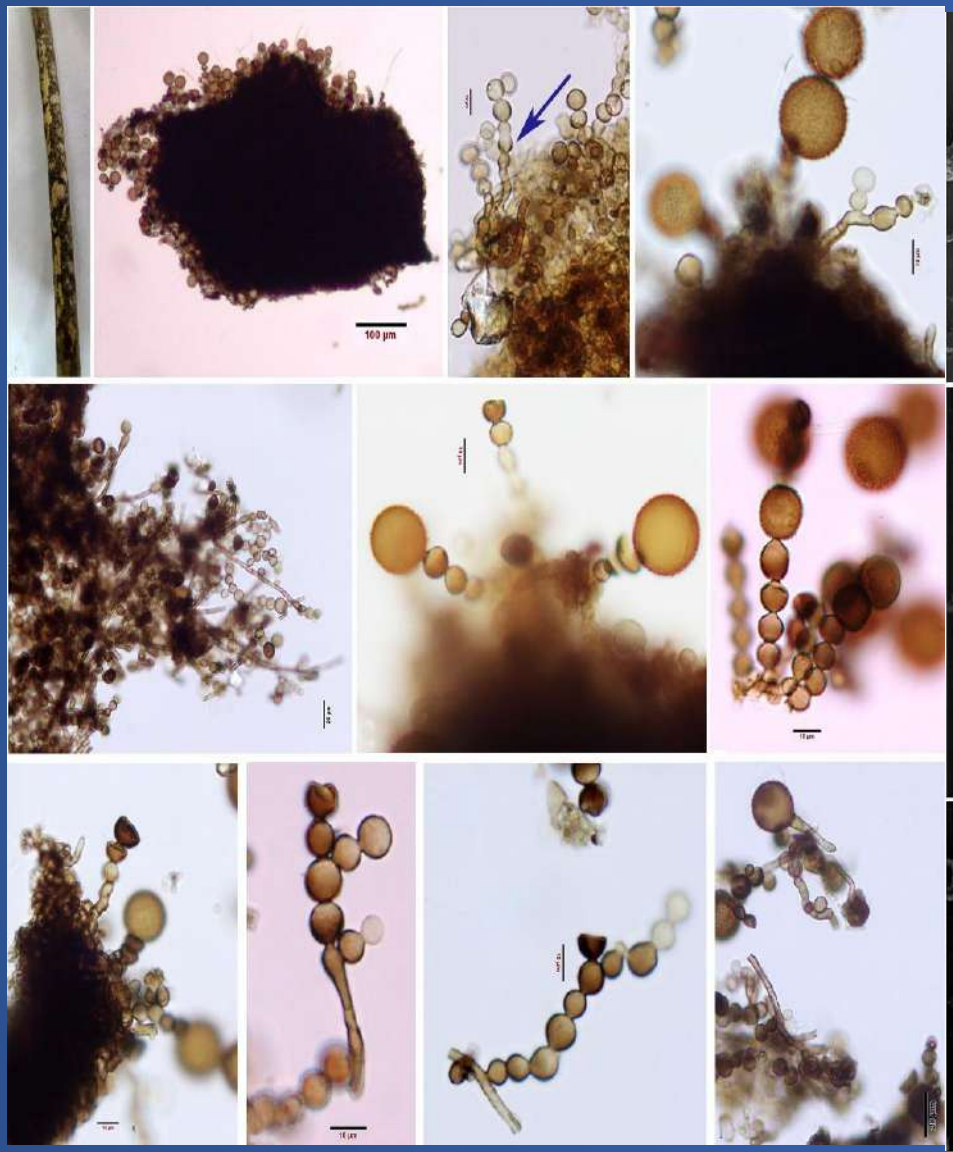
One New genus

03 new species and 07 new records to fungi of India

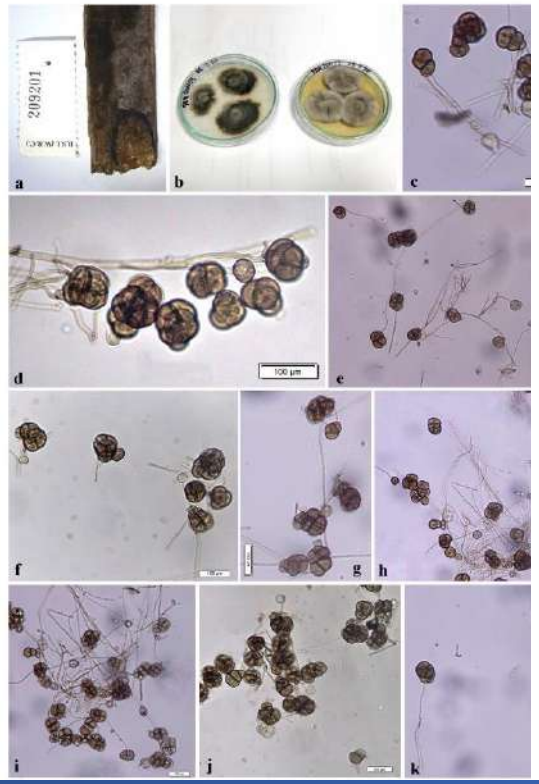
New Findings (2020-21)

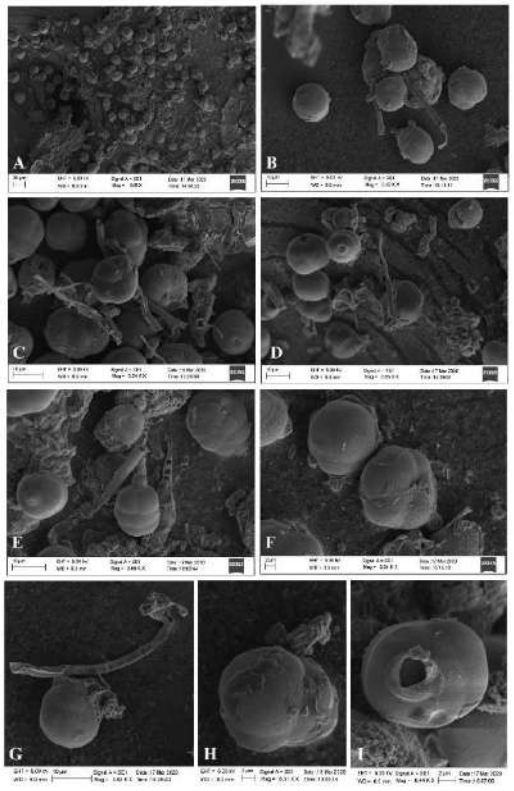
NEW GENUS PUBLISHED :01			
1.	***Lonavalomyces indicus* Gen et sp.nov. Dubey 2020**	**Pezizomycotina, Ascomycota**	**Lonavala (MH)**
NEW SPECIES PUBLISHED: 02			
2.	***Mycoenterolobium borivaliensis* sp. nov. Dubey & Pandey 2020**	**Pezizomycotina, Ascomycota**	**Sanjay Gandhi National Park, Mumbai**
3.	***Conlarium indicum* sp. nov Dubey & Manikpuri 2021**	**Conlariaceae, Ascomycota**	**Sawantwadi (MH)**
NEW RECORDS TO INDIA :02			
1	***Didymocrea leucaenae* Jayasiri et al. (2019)**	**Didymosphaeriaceae, Ascomycota**	**Amboli Ghat (MH)**
2.	***Lasiodiplodia mahajangana* Begoude, Jol. Roux & Slippers,2009**	**Botryosphaeriaceae, Ascomycota**	**Sanjay Gandhi National Park, (MH)**

NEW FINDINGS



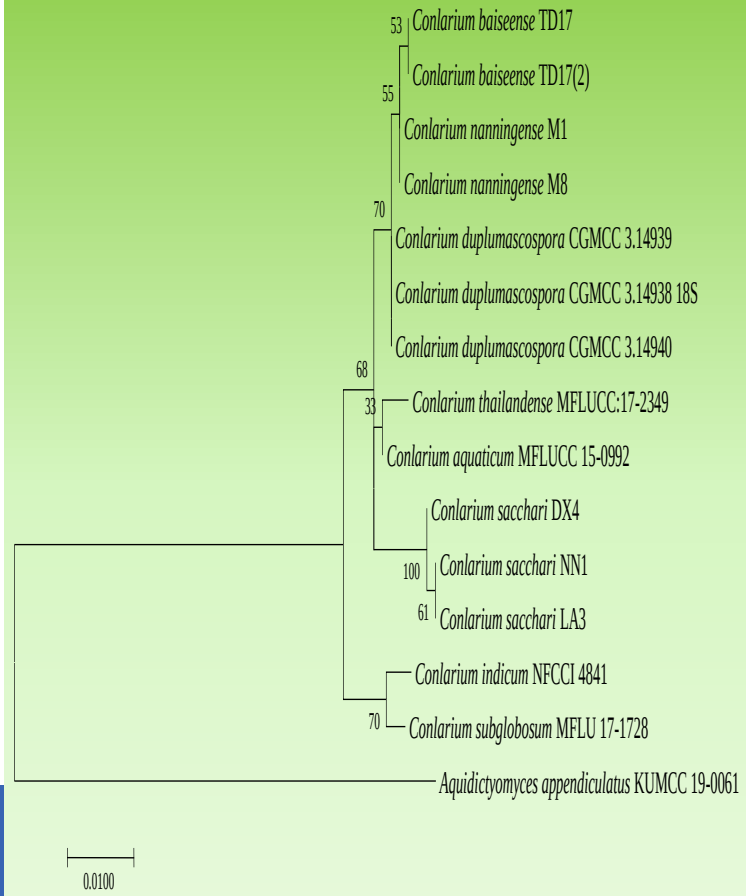
***Lonavalomyces indicus* Gen.et.sp.nov. Dubey 2021**



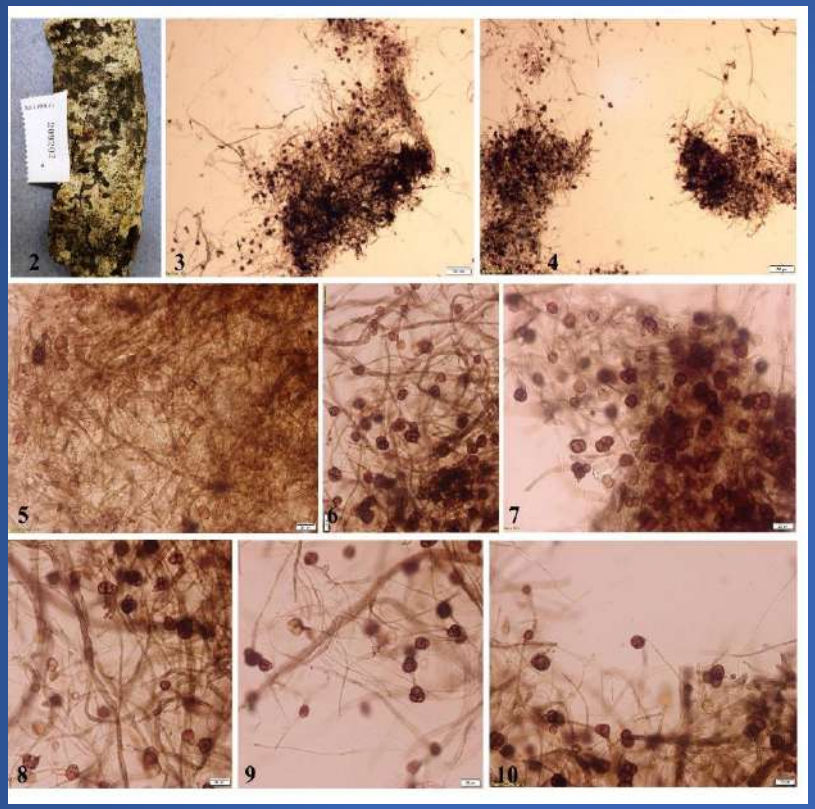


***Conlarium indicum* (NFCCI 4841)** – A Colonies on host surface. B Colony morphology on PCA and PDA. C-K Mature conidia. Scale bars: C-K=100 µm,

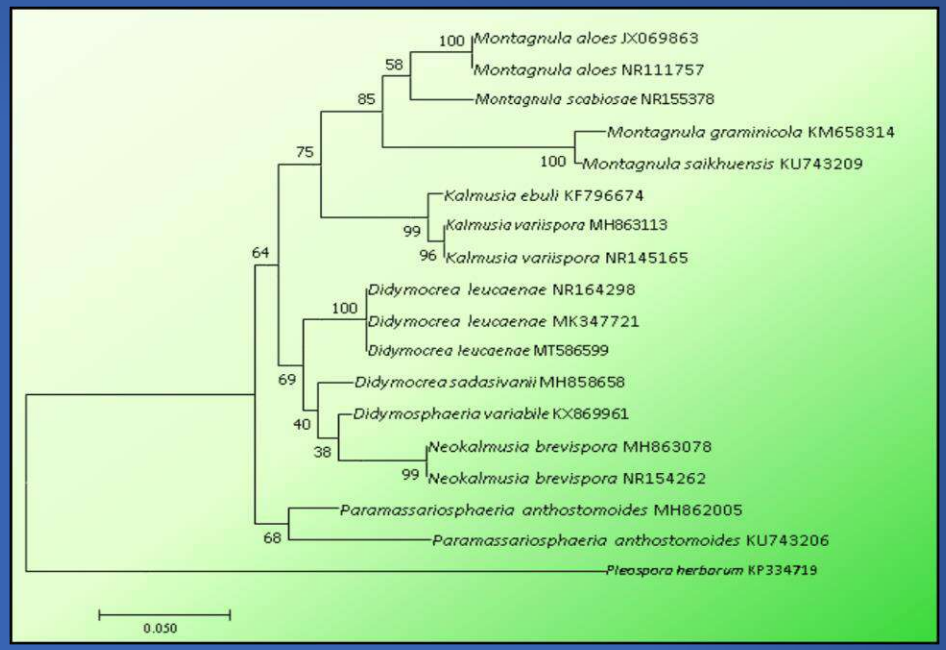
***Conlarium indicum* (NFCCI 4841)** –A-I Scanning Electron Microscopic Images.



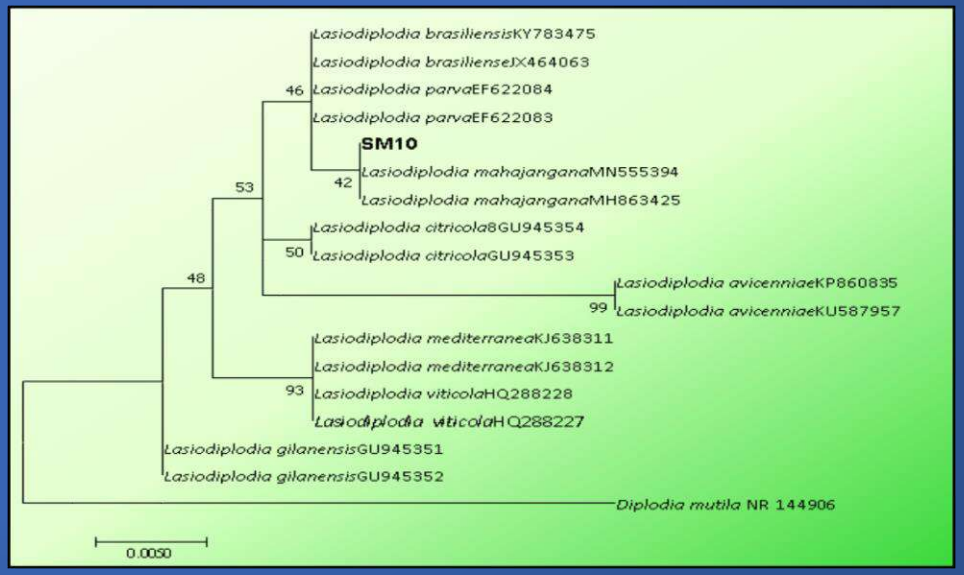
Molecular Phylogeny by maximum likelihood method



Didymocrea leucaenae **(MT586599-ex-type). 2 Colonies on Host surface. 3, 4, 5 Hyphal coils and anastomosing hyphae with conidiophores and conidia. 6,7,8, 9,10 Conidia.**



Molecular Phylogenetic analysis of *Didymocrea leucaenae* : (A new record to India)



Molecular Phylogenetic analysis of *Lasiodiplodia mahajangana* (A new record to India)

Research paper published

- **Research papers published: 04**

1. **DUBEY, R.** 2020. *Lonavalomyces* - A New Anamorphic Ascomycetes Genus reported from Lonavala, Western Ghats of India. *Journal on New Biological Reports.* 9(3) 316 – 320 (2020).

2. **DUBEY, R.** & AMIT D. PANDEY 2020. *Mycoenterolobium borivaliensis* sp. nov. (Pleosporomycetidae, Dothideomycetes) from reported from India. *Journal on New Biological Reports:* 9(3) 312 – 315.

3. SENGUPTA S. AND **R. DUBEY** 2021**:** Taxol producing fungi: a critical review in experimental aspects. *J. Mycopathol.Res.* 59 (1): 1-9.

4. **DUBEY, R.** and MANIKPURI S. 2021. *Conlarium indicum:* A novel fungus from Western Ghats of India.(Current Research in Environmental & Applied Mycology (Journal of Fungal Biology) 11(1), 112-118

RESEARCH PAPERS ACCEPTED/ COMMUNICATED

1. DUBEY, R. (2020). *Didymocrea leucaenae*. Jones & Hyde: A New record to Mycoflora of India. *Mycoasia* (Accepted and likely to be published in May 2021).
2. SHREYA SENGUPTA CHATTERJEE AND DUBEY, R. (2020). *Sadasivania biligiriensis*- a new hyphomycetous fungus from Western Ghats, India" in CREAM. (Accepted)
3. DUBEY, R., (2020). *Janetia heterospora* sp. nov– An unusual anamorphic fungi from Protected areas of Maharashtra communicated in *J.Mycopathological Research* (Accepted)
4. DUBEY, R. and Amit D. Pandey (2021). Enumeration & Statistical Analysis of Biodiversity of Microfungi of Sanjay Gandhi National Park and Its 10% Peripheral Area, Maharashtra (India)- Mycoasia (communicated)
5. DUBEY, R. and Amit D. Pandey (2021). Enumeration and Statistical Analysis of Foliicolous Fungal Biodiversity of Western Ghats of Desh Region of Maharashtra, India – *J.Mycopathological Research* (Communicated)
6. SHREYA SENGUPTA CHATTERJEE AND DUBEY, R. (2020). *Bilgiriella indica* gen et sp.nov- A new hyaline synematous fungus from hills of Biligiri Rangaswamy Temple Wildlife sanctuary, Karnataka, India- *Asian Journal of Mycology.*
7. DUBEY, R. (2019). Taxonomic studies of Floristic Microfungi of North Western Ghats of India to *Journal of Indian Botanical Society*.
8. DUBEY, R. AND S. SENGUPTA CHATTERJEE AND PANDEY, D AMIT." 2020. बिलगिरी वन्यजीव अभयारण्य, कर्नाटक की कवक सम्पदा communicated to Vanaspativani. (Accepted).

ANNUAL ACTION PLAN (2021-22)

Q1	Q2	Q3	Q4
Collection of literature from different sources.	**One Field tour to Bhagwan Mahaveer WLS and Mollem National Park,** Goa and its adjoining areas for collection samples of bambusicolous fungi	• Isolation, identification and preservation of bambusicolous fungi samples collected in the previous tour. • Scanning Electron Microscopic studies of important fungal species. • Molecular sequencing of rare and interesting species. • One Herbarium Consultation tour to Indian Institute of Science, Bangalore	• **One Field tour to Mahadei WLS, Bondla Wildlife Sanctuary** and its adjoining areas for collection samples of Bambusicolous fungi. • Isolation, identification and preservation of bambusicolous fungi samples collected in the tour. • Scanning Electron Microscopic studies of Molecular sequencing of rare and interesting species.

THANK YOU