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Two novel micro fungus from Chhattisgarh, India: *Drechslera moracearum* and *Epicoccum apocyanacearum*

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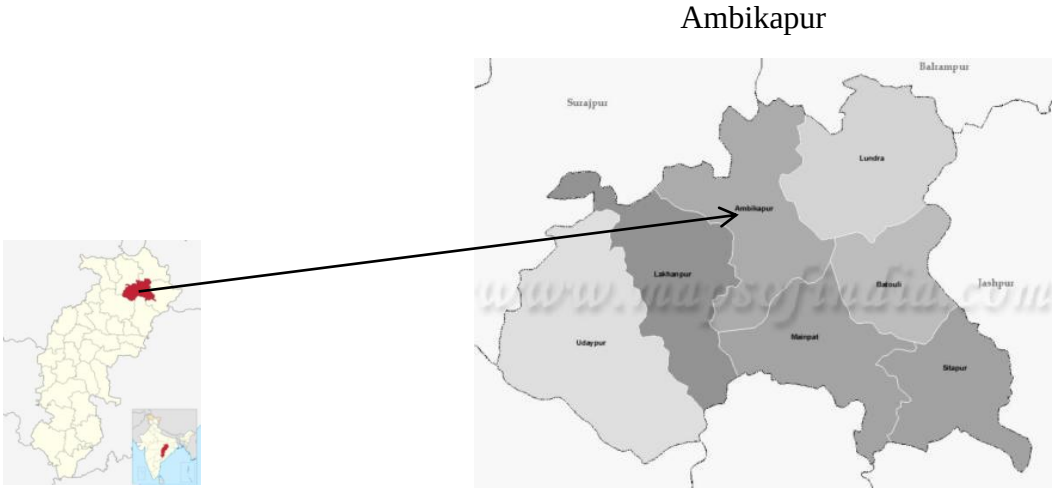
ABSTRACT
This study provides a detailed description of a novel fungus from Rice Bowl of India i.e Chhattisgarh. A survey was conducted to Ambikapur, north Chhattisgarh in India, a micro fungus was found in the month of January 2018. Most of the part of Chhattisgarh is untouched and underexplored for fungus and little work is done from the area and therefore is suitable for the emergence of novel fungal species. A velvety brown fungus was encountered and thus resulted in the collection of an undescribed novel fungus affecting living leaves of *Ficus racemosa* L. from Moraceae family. Symptoms appeared on both surface i.e upper and lower as brown to black colour. Morphological characters like stromata, mycelium, conidiophore, conidia, septation were described by microscopic image examination, morphological studies and scanning electron microscopic image analysis. The comparison with allied parallel taxa resulted as novel species of *Drechslera* i.e *Drechslera moracearum* sp. nov. The present investigation of fungus is the first report from Chhattisgarh, India. An undescribed novel fungus, affecting living leaves of *Nerium oleander* L. was collected. Based on morphological characters, microscopic images, SEM based analysis and comparison with other parallel taxa, a novel species of *Epicoccum* namely *Epicoccum apocyanacearum* has been described. Literature survey reveals no species of *Epicoccum* is reported on the host family as well as on plant *Nerium oleander* L. *Epicoccum poae* and *Epicoccum dendrobii* show some similarities with the proposed species alongside show differences. The critical investigation exposes that the present collection is utterly distinct in symptoms, dimension of larger conidia, wider mycelium and muriform conidia.

Introduction

Chhattisgarh is rich in floral and fauna diversity. The forest flora of Chhattisgarh has diversity of plants with little attention to microorganism, and specially no attention to fungal diversity (11). Huge number of fungal samples were collected from the forest flora and nearby areas of North Chhattisgarh i.e. Ambikapur and the plant samples collected is of great medicinal use. *Ficus racemosa* L. from Moraceae family is one of them with medicinal properties like stomachache, diabetes, diarrhea, piles, liver disorder, memory enhancing, inflammatory conditions, urinary diseases, skin disease, respiratory, antitussive, antipyretic, antimicrobial activity and hepatoprotective (1 & 2). The climate is very suitable for the growth of fungi with humidity. Regular visit (at alternate month) to forest was done for fungal collection. Greenish brown cottony eye-catching fungal symptoms on upper and lower side of leaves by conidia, conidiophore and mycelium was encountered and thus resulted in the collection of an

undescribed novel fungus affecting living leaves of *Ficus racemosa* L. from Moraceae family. Genus *Drechslera* was introduced by Ito in the year 1930. *Drechslera papendorfii* (8) and *Drechslera hawaiiensis* (8) demonstrates some resemblance of general generic chatacterstics whereas the present species shows a bit dissimilarity in appearance starting from symptomatology, very short conidiophores and longer conidia with more septation as compared to comparing species.

During the survey of Ghaghi forest, Ambikapur, Chhattisgarh massive fungal samples were collected on economic and medicinally important plant and one of them was *Nerium oleander* L. Regular survey of Ghaghi forest was conducted for fungal sample collection during every season. The number of fungi till date is likely 2.2 to 3.8 million fungal species, which is only 8% [10]. The proposed species is different by possessing wider mycelium and larger conidia from the comparing ones. It is also remarkable that the present species has clearly muriform conidia as against the tabular comparing species having only phragmosporous conidia. The present fungal species is never been previously disclosed, hence contributes to a novel fungal species. The proposed new taxon is compared with *Epicoccum poe* [14] (infecting *Poa annua* of Poaceae family), *Epicoccum dendrobii* [14] (infecting *Dendrobium fimbriatum* of Orchidaceae family while the proposed species falls under family Apocynaceae show some similarities with the proposed species alongside show differences. The critical investigation exposes that the present collection is utterly distinct. Epicoccum fungus is known for anti-nematode, antibacterial, antimicrobial, antifungal, anticancer, antialgal and anti diabetic activity [5,7,9]. *Epicoccum* fungus infecting *Nerium oleander* L. also has several medicinal values i.e. skin diseases, leprosy, eye diseases, heart, diaphoretic, gynaecological disorder, joint pain and ulcer [13].



Map 1: Map of Ambikapur

Materials and methods

Collection was done in every month between 2018 to 2021 at different forests, parks and nurseries of Ambikapur, Chhattisgarh, India. Equipment's like poly-bags, rubber-band, digital camera, magnifying glass, scissor, tags, marker and note book were carried during field (3 & 11). Infected leaves of *Ficus racemosa* L. and *Nerium oleander* L. was collected and kept with

naphthalene to avoid microbial attack and brought to laboratory for further investigation, slides were prepared by scratching infected leaves and mounted with lactophenol cotton blue (6 &12). Microscopic examination was done under Olympus CX2li trinocular microscope for morphological study and identification of fungus. Scanning Electron Microscopy (SEM) was also done under double beam FEI Nova nano SEM-450 which were chemically fixed and coated with gold (HiRes-Gold on Carbon) at DHSG University, Sagar, M.P. (4).
The dried fungal infected leaves were deposited at Ajrekar Mycological Herbarium- AMH, Pune, Maharashtra, India, One set of sample is deposited at Botany Department herbarium, Dr. Hari Singh Gour University, Sagar, M.P.

Result

***Drechslera moracearum* sp. nov.**

Taxonomy and Description

On living leaves of *Ficus racemosa* L. (Moraceae), Sanjay park, Ambikapur, Chhattisgarh, India, January 2018 leg. A. D. Khalkho, (Holotype, AMH- 10103, Isotype RAH 81).
Etymology: *moracearum* is derived from the name of host family.
Symptoms: Symptoms amphigenous, starting from leaf margin, brown to black. Colonies amphiphyllous, effuse, hairy, velvety, brownish black. Mycelium immersed. Stromata present, small to large, irregular, olivaceous brown to black, 30.38-71.20 x 31.66- 68.04 µm. Conidiophores macronematous, mononematous, mostly solitary rarely in loose fascicles (of two), straight to flexuous, unbranched, smooth, olivaceous brown, 0-2 septate, 27.44-47.95 x 3.64-8.86 µm, dark brownish black scar. Conidiogenous cells integrated, terminal, cylindrical, 10.18-12.10 x 3.92- 4.48 µm. Conidia solitary, straight to curved, cylindrical, rounded at ends, acropleurogenous, olivaceous brown, smooth, 1-8 pseudoseptate, 22.94-70 x 5.70-12 µm, hilum thickened, occasionally germ tube produced from both or single end.

Figure 1. Symptoms of *Drechslera moracearum* sp. nov. on *Ficus racemosa* (Holotype, AMH- 10103) (a) Symptom on upper surface (b) Symptom on lower surface

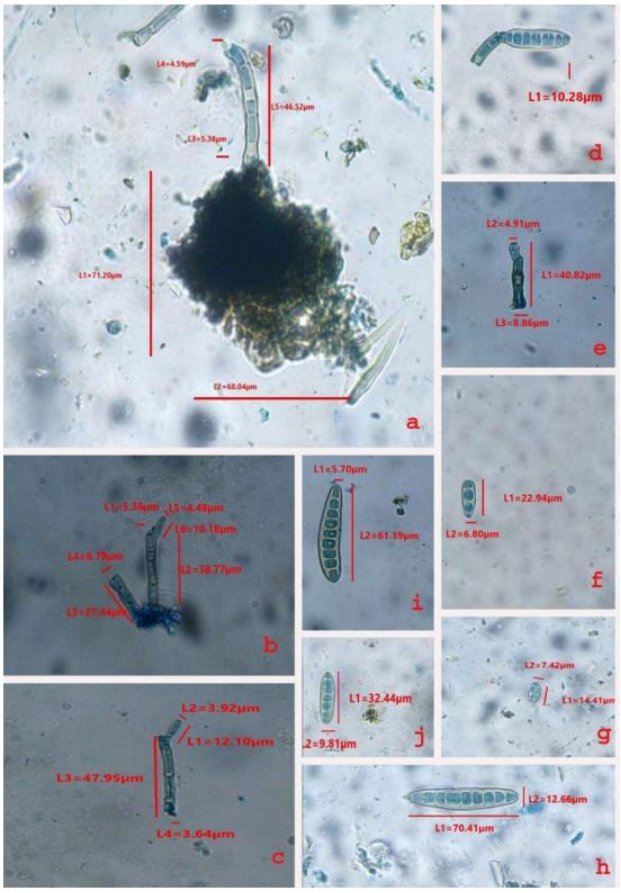


Figure 2. Microphotographs of *Drechslera moracearum* sp. nov. on *Ficus racemosa* (a) Conidiophore with stroma (b) Conidiophores with stroma (c,d) Attachment of conidium with conidiophore (e) Conidiophore (f-j) Different size and shape of Conidia.

***Epicoccum apocyanacearum* sp. nov.**

Taxonomy and Description

On living and dried leaves of *Nerium oleander* L. (Apocynaceae), Ghaghi forest, Ambikapur, Chhattisgarh, India January 2021 leg. A. D. Khalkho (Holotype-AMH- 10345, Isotype- 209)

Etymology:- ***apocyanacearum*** is derived from the name of host family.

Symptoms hypogenous, in patches, circular to irregular, black. Colonies hypophyllous, effuse, velvety, mostly confined in the centre, black. Sporodochia semiimmersed, pulvinate, black. Mycelium immersed. Stroma present, pulvinate. Conidiophores macronematous, mononematous, densely packed together covering the surface of stroma, unbranched to branched, short, straight to flexuous, olivaceous brown, smooth, 2.30-4.70 µm. Conidiogenous cells monoblastic, integrated, cylindrical. Conidia solitary, acrogenous, pyriform, olivaceous brown, sometimes with protuberant basal stalk cell, the younger conidia are mostly smooth while mature ones are rough walled, muriform but with the septa obscured in mature conidia by rough opaque wall, 8.50-31.50 µm.

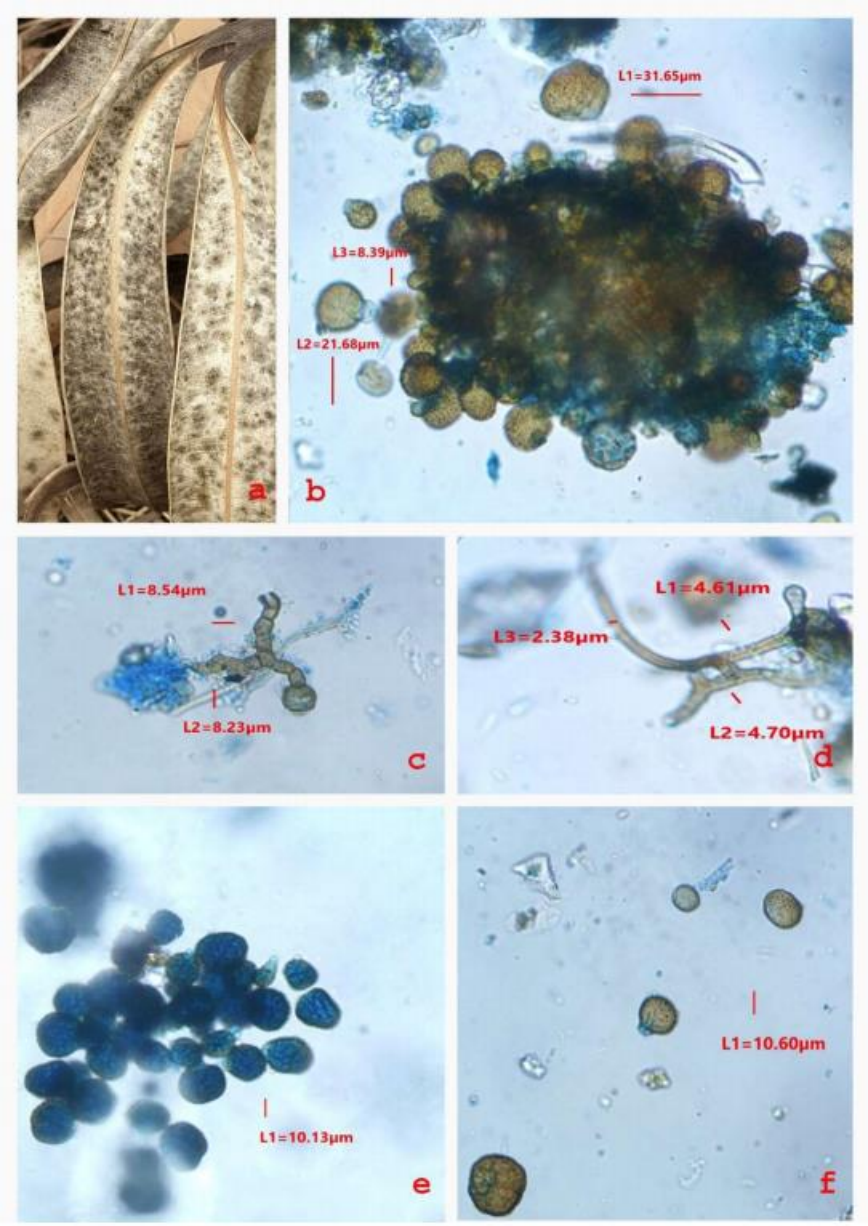


Figure 3- Symptoms and Microphotographs of *Epicoccum apocyanacearum* sp. nov. on *Nerium oleander* (Holotype-AMH-10345) (a) Symptoms on leaves (b) Sporodochia (c) Conidiophore with conidium (d) Mycelium (e) Bunch of conidia (f) Conidia.

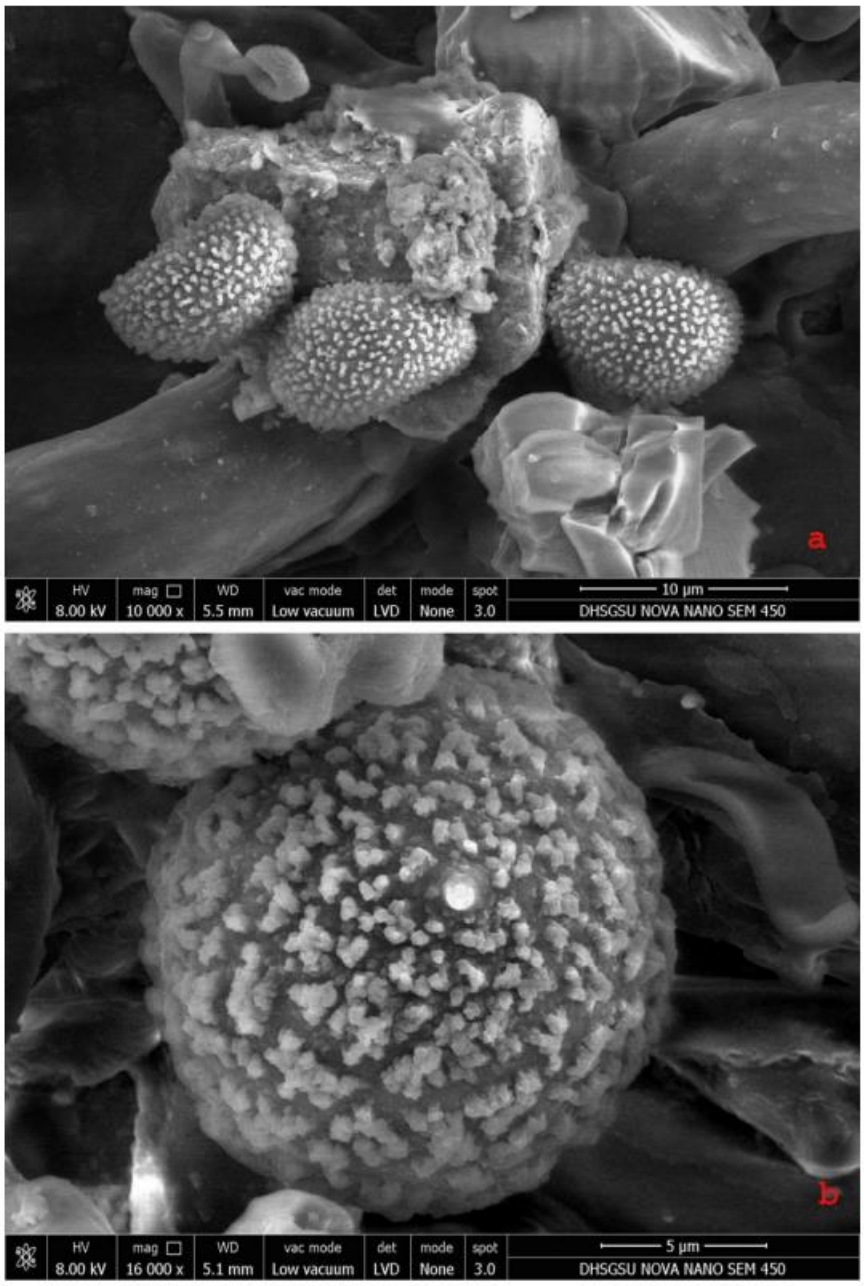


Figure- 4: SEM images of *Epicoccum apocyanacearum* sp. nov. on *Nerium oleander* (Holotype-AMH-10345) (a) Bunch of conidia (b) Conidium.

Discussion and conclusions

Of all the earlier described species of *Drechslera*, the present species falls comparable with *Drechslera papendorfii* (Ellis, 1971) and *Drechslera hawaiiensis* (Ellis, 1971) and without any doubt demonstrates some resemblance of general generic characteristics whereas the present species shows a bit dissimilarity in appearance starting from symptomatology, very short conidiophores and longer conidia with more septation as compared to comparing species of Literature survey reveals no species of *Drechslera* is previously reported on *Ficus racemosa* L. Holotype- AMH-10103. Therefore, due to the morphotaxonomic differences the present

species is justified for description and illustration as a new taxon of species rank i.e *Drechslera moracearun*, Holotype, AMH-10103.

After going through the literature, it was found that the proposed new taxon may be compared with *Epicoccum poe* (infecting *Poa annua* of Poaceae family), *Epicoccum dendrobii* (infecting *Dendrobium fimbriatum* of Orchidaceae family while the proposed species falls under family Apocynaceae. It was observed from the table that the proposed species is different by possessing wider mycelium and larger conidia by having 2.30-4.70 µm and 8.5031.50 µm dimension respectively from the comparing ones. It is also remarkable that the present species has clearly muriform conidia as against the tabular comparing species having only phragmosporous conidia. Therefore, the present species *Epicoccum apocyanacearum* is justified to place as new taxon of species rank.

It is also to be noted that the collection shows the intermixed symptom (of *Epicoccum* and *Cladosporium*). The colonies of *Epicoccum* are mostly surrounded by *Cladosporium.*

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