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Cover: Life and death in one night - wolf hunting the hare. Mixed media—gouache, acrylics, pen & colour pencils. © Dupati Poojitha.

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COMMUNICATION

On the *Maravalia echinulata* (Niessl ex Rabenh.) Ono (Pucciniales: Chaconiaceae) with reference to its host range and distribution

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Abstract: *Maravalia echinulata* (Niessl ex Rabenh.) Ono has been rediscovered from West Bengal, India, after its last report in 1931. Sections of infected host leaves were cut, stained in cotton blue, and mounted in lactophenol. All five spore forms of this macrocyclic autoecious rust fungus are described in detail, with notes on its world distribution, distribution in India, and host range. Pycnia were found to be amphigenous, whereas aecia, uredinia, and telia were hypophyllous. Pycnia subcuticular, globose with flexuous hyphae belonging to type 4 of Hiratsuka & Sato (1982) containing chains of spermatia. Aeciospores are spiny, and catenulate, forming long chains connected by hyaline disjunctor cells. Urediniospores pedicellate, spiny, hyaline, and thin-walled when young, but brown and thick-walled when mature, intermingled with paraphyses. Basidium cylindric with a pedicel, 4-celled, tetrasterigmatic; basidiospores globose. Discrepancies in descriptions of microscopic characters by different authors with regard to the present observations have also been discussed.

Keywords: India, macrocyclic, rediscovery, rust fungus, *Scopella echinulata*, spore forms, West Bengal.

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Author details: During this study, Mr. Sayantan Jash was a postgraduate student of Botany with keen interest in the field of mycology and plant pathology. Now he is working on the systematics of fungi as a junior research fellow at Botanical Survey of India. Dr. Asit Baran De is a retired associate professor of Botany with a great contribution to fungal research, especially fungal taxonomy. He has described eight taxa new to science. Dr. De is one of the authors of the very popular book 'Polyporaceae of India'.

Author contributions: All authors have contributed equally throughout the study.

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INTRODUCTION

Uromyces echinulatus Niessl ex Rabenhorst was first described in 1881 (Rabenhorst 1881), which parasitized *Bassia latifolia* Roxb. (= *Madhuca latifolia* Macbr.). It was described based on the collection made by Dr. S. Kruz from the Royal Botanic Garden, Calcutta (now Acharya Jagadish Chandra Bose Indian Botanic Garden, Shibpur, Howrah, West Bengal, India). In 1939, it was transferred to the genus *Scopella* Mains (Mains 1939) and then in 1984 to the genus *Maravalia* Arth. (Ono 1984). Therefore, the valid name of the species is *Maravalia echinulata* (Niessl ex Rabenh.) Ono.

From literature and also from available herbarium specimens, it is evident that the world distribution of *Maravalia echinulata* is restricted to only five Asian countries, namely India (Ono 1984), Myanmar (Thaung 2005), Nepal (IMI 189185), Pakistan (Ahmad 1956), and Sri Lanka (Spaulding 1961). Its global distribution has been presented in an outline map of the world (Figure 1).

Butler & Bisby (1931) mentioned that Mitra collected this fungus from West Bengal, India. But this report was not substantiated by any reference or herbarium specimen. It has been rediscovered from West Bengal in 2019 by the present authors, and was found to grow on the leaves of *Madhuca latifolia*. This is the report of this fungus from the state of West Bengal about 90 years after its last report (Butler & Bisby 1931). From the literature (Mains 1939; Cummins 1950; Thirumalachar 1950; Verma 2015), it is evident that there is much controversy regarding its characters.

In the present study, the characteristics of *M. echinulata* originating from West Bengal is described, with special emphasis on its spore types and microscopic features. The host range and distribution of this fungus have also been discussed.

MATERIAL AND METHODS

During a survey conducted in West Bengal (October 2019–February 2020) to record naturally occurring host plants of different rust fungi, some infected leaves of *Madhuca latifolia* were found in some localities of Bardhaman town in Purba Bardhaman District. The infected leaves of different plants were collected separately on 20 November 2019. Thin sections of infected leaves were cut by using sharp blades. Staining of the sections was done with cotton blue and mounted in lactophenol. After staining, some sections were teased apart with sharp needles and then mounted in lactophenol. Microscopic observations were made under ×600 and ×1,500 magnifications of a Nikon Ti-U inverted microscope. To determine the range of spore sizes, 20 spores of each type were measured. Voucher specimens were properly processed and then deposited in the herbarium of the Department of Microbiology, The University of Burdwan (BURD), Bardhaman, West Bengal, India. Identification of the fungus was done based on the descriptions provided by different literatures, including Mains (1939), Cummins (1950), and Ono (1984). The holdings of *Maravalia echinulata* were also studied from different internationally recognized herbaria, including Royal Botanic Garden, Kew (K), The New York Botanical Garden (NY), Meise Botanic Garden (BR), New Zealand Fungal Herbarium (PDD), University of Michigan (MICH), and University of Minnesota (MIN), to record its host range and distribution.

RESULTS

Field observation

The fungus was regularly seen in the field infecting leaves of *Madhuca latifolia* as very small dark brown somewhat elevated pustules. Dark pustules were surrounded by irregular pale-green to yellowish halo zones (Image 1). The infection started in October and remained up to February. The survey showed that all the trees in the study area were infected.

Taxonomy

Each infected leaf collected from different trees showed all of the five spore forms, confirming the species as macrocyclic and autoecious. The species is described based on the present collection, which is as follows:

***Maravalia echinulata* (Raben.) Ono, Mycologia 76(5): 924. 1984.** (Image 2 & 3)

Uromyces echinulatus Niessl ex Raben., Hedwigia 20: 149. 1881.

Scopella echinulata (Raben.) Mains [as (Niessl) Mains], Ann. Mycol. 37: 58. 1939.

Pycnia (Image 2a) amphigenous, sub-cuticular, lenticular to hemispheric, 40.0–50.0 × 80.0–120.0 µm; spermatiophores producing chains of spermatia, spermatia (Image 2b) globose to elliptical, hyaline, thin- walled, 1.5–2.0 × 1.5–1.8 µm; flexuous hyphae long and hyaline. Aecia hypophyllous, sub-epidermal, more or less concentrically and densely grouped with

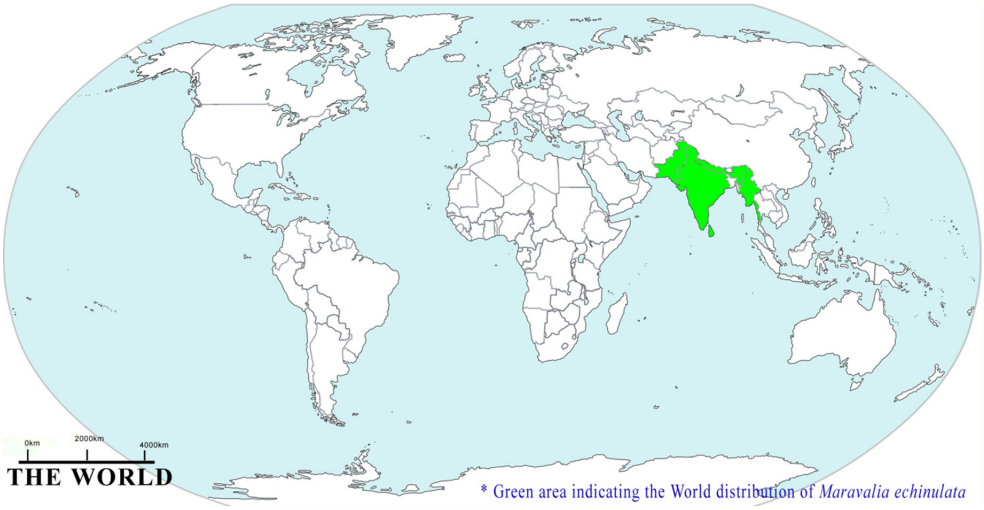

Figure 1. World distribution of *Maravalia echinulata*.

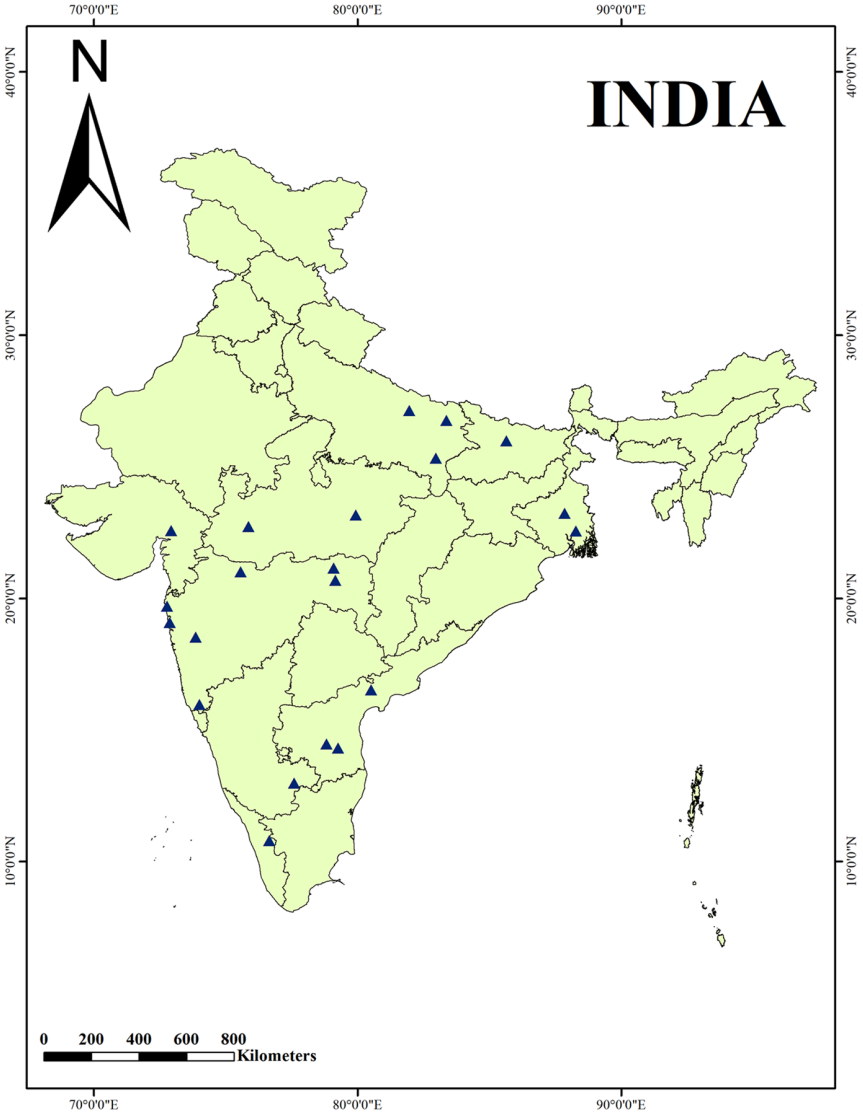

Figure 2. Map showing distribution of *Maravalia echinulata* in India (blue triangles indicate the localities).

pycnia, becoming confluent, causing reddish brown spots epiphyllously, about 0.2–1.0 mm in diameter, individual sori of about 0.1–0.2 mm diameter, chocolate-brown, pulverulent; aeciospores forming chains (Image 2c,d) connected by hyaline disjunctors, asymmetrical, triangular in face view, obovate or oblong-elliptic in lateral view, 26.0–32.0 × 31.0–42.0 µm, hyaline and thin-walled when young but when mature dark cinnamon to chestnut-brown, coarsely echinulate above (Image 2e), spore wall 2.0–3.0 µm thick. Uredinia hypophyllous, sub-epidermal, very similar to aecia but more scattered and chestnut-brown; urediniospores (Image 2f,g) similar to aeciospores in shape and size, sometimes containing oil droplets; spore wall densely covered by spines but with smooth area on one side near the base; the urediniospores borne on long pedicels, pedicels hyaline, up to 28.0 µm long. Telia hypophyllous sub-epidermal, more or less rounded or ovoid, 0.2–0.5 mm in diameter, crowded in irregular groups or often scattered singly, yellowish or pale brownish; teliospores (Image 2h,i,j) obovoid or sub-angular, apex broadly rounded to sub-truncate, pedicellate, 22.0–27.0 × 34.0–50.0 µm; outer spore wall hyaline, very thin, about 0.5 µm, covered with spines, whereas inner spore wall brownish, up to 1.5 µm thick; pedicels hyaline, up to 30.0 µm long. Teliospores intermingled with numerous paraphyses, teliospores and paraphyses arising from highly developed hyaline to yellowish thin- to slightly thick-walled cylindrical basal cells (Image 2k). Paraphyses (Image 2l) cyllindric to clavate, hyaline, aseptate, longer than pedicellate teliospores. Basidia cyllindric with a hyaline pedicel, 4-celled, tetrasterigmatic, tetrasporous (Image 2m,n); sterigmata short, hyaline bearing single basidiospore on each; basidiospores globose, hyaline, thin-walled.

Mycelia hyaline, thin-walled to slightly thick-walled, simple-septate, up to 5.0 µm wide, intercellular (Image

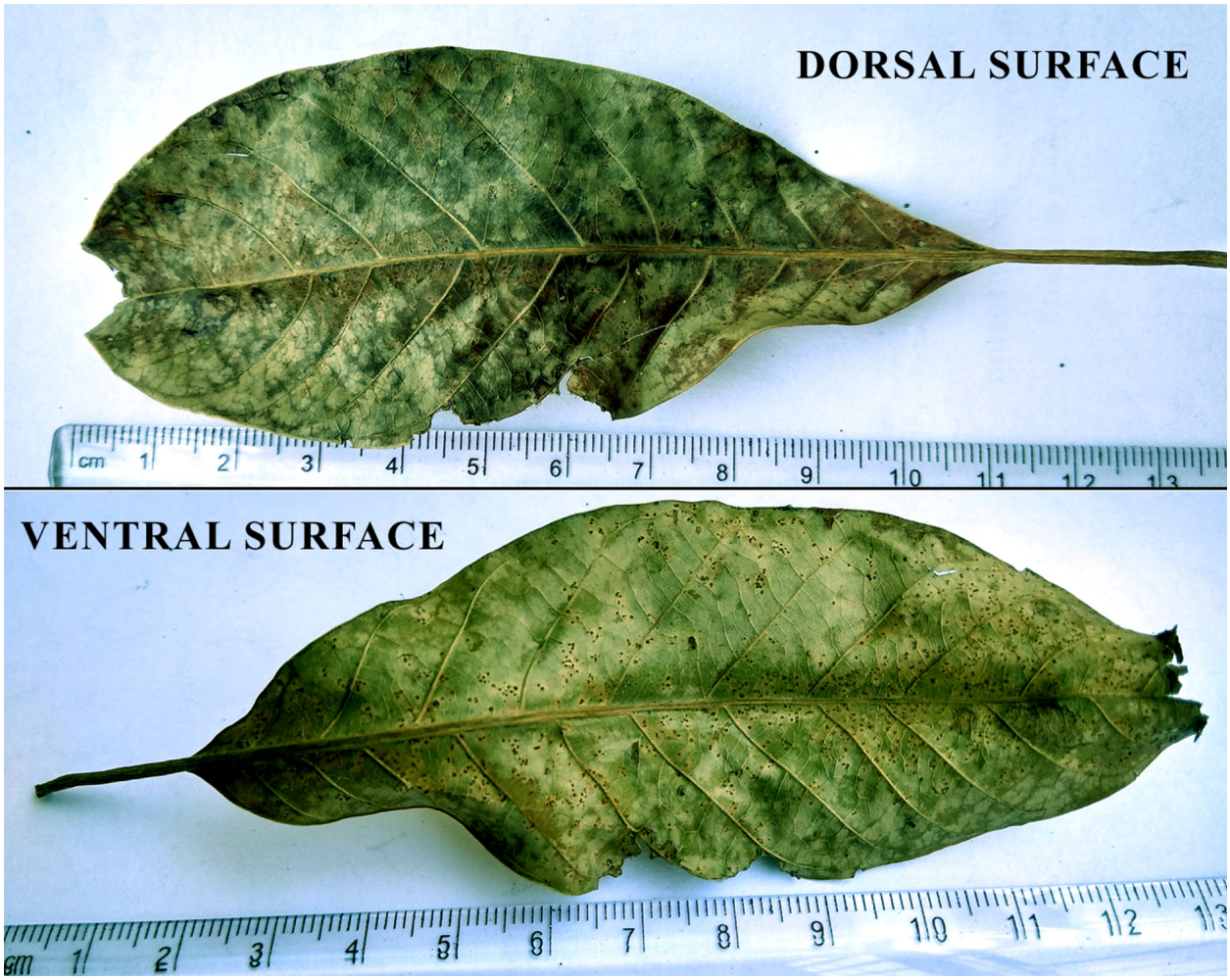

Image 1. Infected leaf of *Madhuca latifolia* with dark brownish pustules surrounded by irregular pale-green to yellowish halo zones. Scale as per photograph. © Sayantan Jash & Asit Baran De

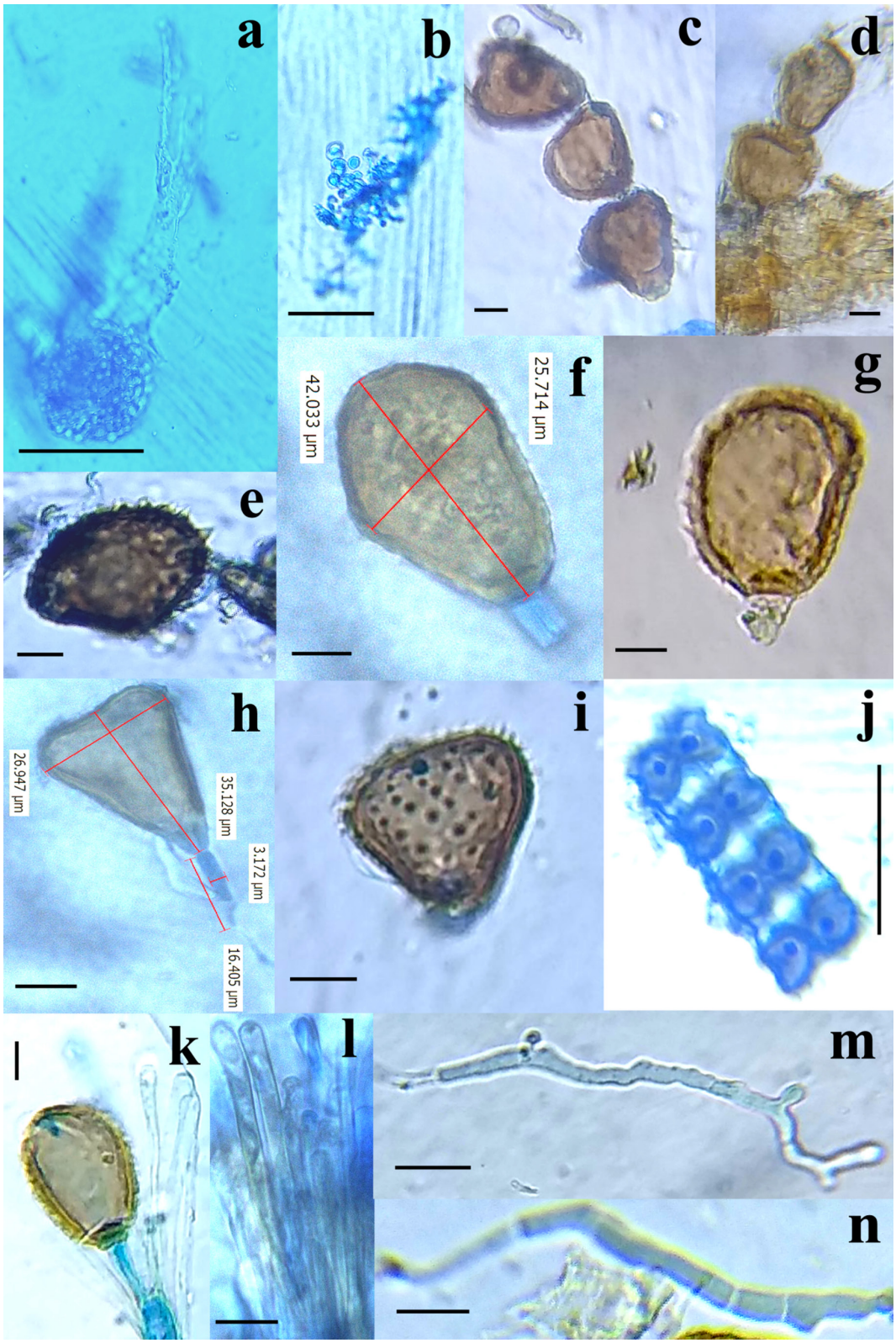

Image 2. Different spore forms: a—pycnium with a spermatium attached on flexuous hypha | b—spermatia | c—chain of three aeciospores connected by disjunctor cells | d—chain of two aeciospores connected with host tissue | e—aeciospore with spiny outer wall | f & g—urediniospore | h—pedicellate teliospore | i—'reticulate-spinulose' teliospore | j—spines of teliospore arising from the lumina surrounded by muri | k—pedicellate teliospore and paraphyses arising from a large cylindrical basal cell | l—paraphyses | m—successive basipetal development of basidiospore on 4-celled tetrasterigmatic basidium | n—4-celled basidium with the initiation of basidiospore on sterigma of its terminal cell. Bar: a = 50 µm; b–n = 10 µm. © Sayantan Jash & Asit Baran De.

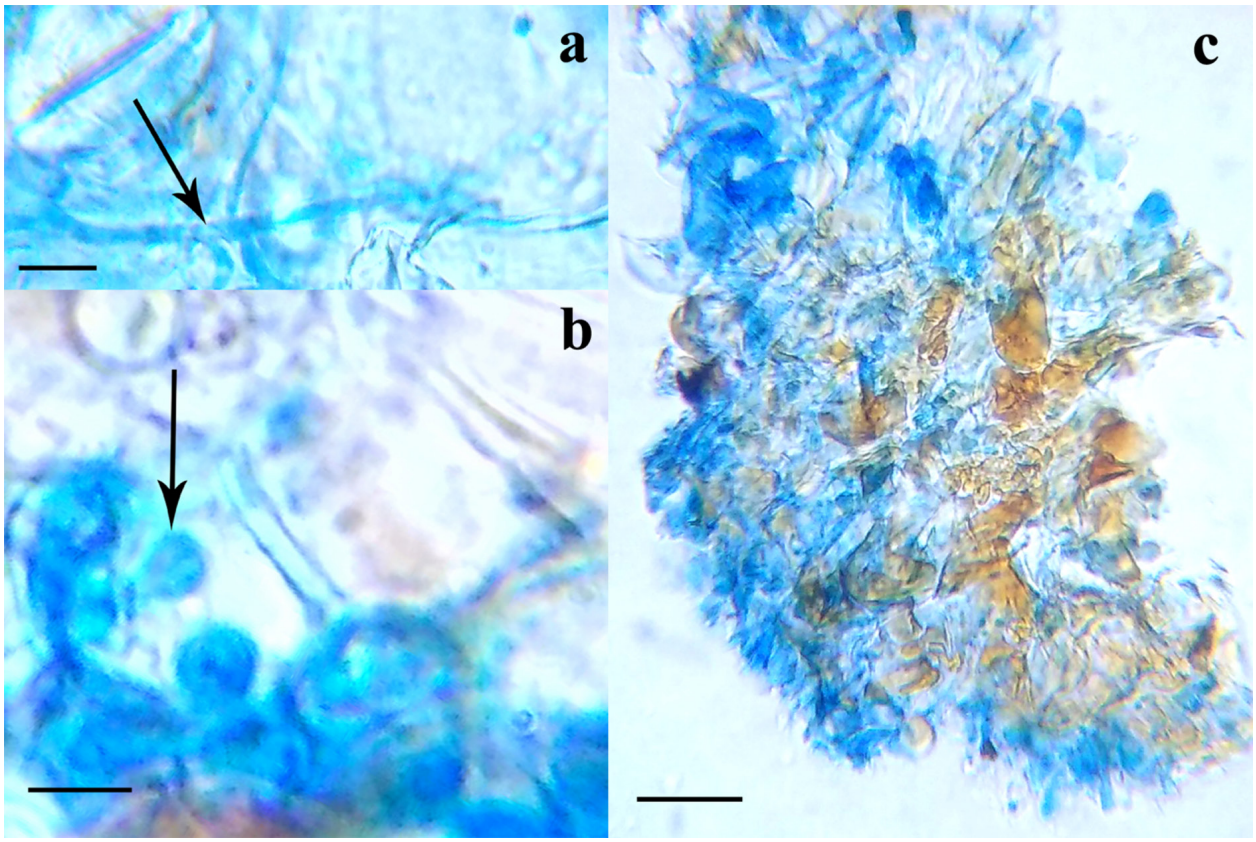

Image 3. Tissues of the infected leaf showing: a—intercellular hyphae of the pathogen | b—intracellular bulbous haustorium | c—post-inflectional gummy secretion by the host. Bar = 10 µm. © Sayantan Jash & Asit Baran De.

3a), but some are intracellular, forming hyaline, thin-walled and bulbous haustoria (Image 3b). Abundant gummy substances (Image 3c) were secreted by the host plant at the post infection stage to defend itself from further invasion by this pathogen.

Specimens examined

On leaves of *Madhuca latifolia* (Roxb.) Macbr., Leg. A.B. De & S. Jash; 20 November 2019; Bardhaman (23.233 ^{0}N, 87.863 ^{0}E, 30 m altitude), Purba Bardhaman, West Bengal, India—BURD MCBH1939, BURD MCBH1940, BURD MCBH1941.

Collections: INDIA: Bihar – Pusa, 21 December 1914, leg. P.C. Kar (PDD 14053); locality not mentioned, 05 February 1986, leg. J.N. Sinha (IMI 314115). Gujarat – locality not mentioned, March 1984, leg. not mentioned (IMI 284368). Karnataka – Bangalore, 1 March 1946, leg. M.J. Thirumalachar (WIS-f-0080583); 3 August 1946, leg. M.J. Thirumalachar (MICH 295078). Madhya Pradesh – locality not mentioned, 23 April 1980, leg. V.R. Neelay (IMI 248182); 21 October 1986, leg. K.S. Khan (IMI 311371). Maharashtra – Palghar, 22 February 1912, leg. H.M. Chebber (MICH 295080); Poona, Maharashtra Assoc. for Cult. Sci., 19 March 1965, leg. M.N. Kamat (IMI 112556); Sirsi, September 1912, leg. G.S. Kulkarni (PDD 9797). Uttar Pradesh – Gorakhpur, 23 February 1984, leg. P. Narayan (IMI 283997); 22 October 1984, leg. not mentioned (IMI 290974); locality not mentioned, 05 March 1980, leg. R.P. Verma (IMI 246910). West Bengal – Bardhaman, 20 November 2019, leg. A.B. De & S. Jash (BUR MCBH1939, BUR MCBH1940, BUR MCBH1941); Calcutta, H. Botanico, – s.d., leg. Dr. S. Kruz 3551 (K-M000187873, K-M000187874, NY 00046133, NY 00046134, UC 1886570); s.d., leg. G. Rabenhorst s.n. (BR5020078034467).

MYANMAR: Mandalay, 28 November 1974, leg. M.M. Thaung (LAM 220082, UC 1886570); s.d., leg. M.M. Thaung (IMI 161589).

NEPAL: locality not mentioned, 18th October 1974, leg. K.L. Manandhar (IMI 189185, IMI 189192).

SRI LANKA: Peradeniya, 16 June 1908, leg. T. Petch (K-M000187875); August 1912, leg. T. Petch (BR5020078035471, K-M000187876, MICH 295079, MICH 296112, MIN 1228326, MIN 1228327, MIN 1267657, NY 03432410, WIS-f-0085023); locality not mentioned, s.d., leg. T. Ptech (IMI 67265).

DISCUSSION

Although *Maravalia echinulata* (Niessl ex Rabenh.) Ono has been found to cause rust disease in five Asian countries (India, Myanmar, Nepal, Pakistan and Sri Lanka), it mostly affects its hosts growing in India except its northern and northeastern parts (Figure 2). Different localities of its occurrence in India are presented in Table 1 along with its hosts recorded by different workers.

From Table 1 it is evident that *Maravalia echinulata* (Niessl ex Rabenh.) Ono mostly grows on two species of *Madhuca* Ham. ex Gmel., namely *Madhuca latifolia* (Roxb.) Macbr. and *Madhuca longifolia* (Koenig ex L.) Macbr. Rabenhorst (1881) described the species based on a specimen infecting the leaves of *Bassia latifolia* Roxb. Several authors, including Spaulding (1961), Sathe (1969), Narayan & Kamal (1985), and Bilgrami et al. (1991) reported *Madhuca indica* Gmel. as its host. But *Bassia latifolia* and *Madhuca indica* are just synonymous with *Madhuca latifolia*. Mains (1939) and Sathe (1969) mentioned the occurrence of this rust fungus on *Bassia longifolia*, which is currently accepted as *Madhuca longifolia*. Thirumalachar (1950) mentioned *Bassia bourdilli* [correct name is *Madhuca bourdillonii* (Gamble) H.J.Lam] as a new host collected from Mysore (now Karnataka, India). In 1986, Dr. C.R. Patil collected the rust fungi on leaves of *Sideroxylon tomentosum* Roxb. from Amboli (Maharashtra) and M.S. Patil reported it as a new host of *Maravalia echinulata* (Patil 1991). So, this fungus has, so far, been found to parasitize only four host species, namely, *M. bourdillonii*, *M. latifolia*, *M. longifolia*, and *S. tomentosum*. From all these reports, it can be stated that the host range of this fungus is strictly restricted to the angiospermic family Sapotaceae to date.

As regards characters of its different spore forms, there are great controversies. According to Thirumalachar (1950), its pycnium is conoid without conspicuous ostiolar paraphyses. But from the present observation, it is evident that the pycnium of *M. echinulata* is globose with flexuous hyphae and, therefore, belongs to type 4 of Hiratsuka & Sato (1982), not type 5 or 7.

Aeciospores were stated to be pedicellate and uredinoid type (Thirumalachar 1950), but in our observation they are catenulate, forming long chains connected by hyaline disjunctor cells.

Teliospores have been stated to be hyaline and smooth (Mains 1939; Cummins 1950), but very thin-walled according to Mains (1939) and slightly thick-walled according to Cummins (1950). Mains (1939) and Cummins (1940) also found that teliospores of this species arise from highly developed cylindric basal cells. Similar observations were made by the authors, who also observed spiny teliospores as hyaline and thin-walled when young, but brown and thick-walled when mature. Similar types of brown, thick-walled teliospores with a spiny outer wall have been observed by Rabenhorst (1881) and Pathak et al. (2015).

Teliospores are intermingled with numerous paraphyses, which are cylindrical to clavate, hyaline, thin-walled and much longer than pedicillate teliospores. Although Thaung (2005) has mentioned numerous fine

Table 1. Different localities and hosts of *Maravalia echinulata* growing in India.

State	Locality	Host	References
Andhra Pradesh	Amaravati	*Madhuca longifolia*	Hosagoudar 2013
	Cuddapah	*Madhuca latifolia*	Cummins 1950
	Penagaluru	*Madhuca latifolia*	Cummins 1950
Bihar	Pusa	*Madhuca latifolia*	Butler & Bisby 1931
Gujarat	Anand	*Madhuca latifolia*	Jamaluddin et al. 2004
Karnataka	Bangalore	*Madhuca latifolia* and *Madhuca bourdillonii*	Thirumalachar 1950
Kerala	Palghat	*Madhuca latifolia*	Bilgrami et al. 1991
Madhya Pradesh	Indore	*Madhuca latifolia*	Pathak et al. 2015
	Jabalpur	*Madhuca latifolia*	Verma 2015
Maharashtra	Amboli	*Sideroxylon tomentosum*	Patil 1991
	Jalgaon	*Madhuca latifolia*	Firdousi 2020
	Mumbai	*Madhuca longifolia*	Sathe 1969
	Nagpur	*Madhuca latifolia*	Parandekar 1964
	Palghar	*Madhuca latifolia*	MICH 295080
	Poona	*Madhuca latifolia* and *Madhuca longifolia*	IMI 112556, Sathe 1969
	Sirsi	*Madhuca latifolia*	PDD 9797
Uttar Pradesh	Benaras	*Madhuca latifolia*	Payak 1949
	Gonda	*Madhuca latifolia*	Narayan & Kamal 1985
	Gorakhpur	*Madhuca latifolia*	IMI 283997, IMI 290974
West Bengal	Bardhaman	*Madhuca latifolia*	Present study
	Howrah	*Madhuca latifolia*	Rabenhorst 1881, Mains 1939, Ono 1984

paraphyses in *Maravalia echinulata*, most of the authors had probably ignored this character in their description.

A gap in the secondary wall above the hilum of urediniospores and teliospores has been observed, which indicates the existence of continuous cytoplasmic communication of these spores with their pedicels through the passage in the secondary wall.

Spines of aeciospores, urediniospores and teliospores are formed in the centre of lumina bordered by muri. These spores of *Maravalia echinulata*, therefore, may be better described as reticulate-spinulose rather than just reticulate or just spinulose.

Ono (1984) reported that each metabasidium forms one short, apical sterigma, and from this sterigma, globose to subglobose basidiospores are formed successively. Thus, the basidiospores occur in clumps at the apex of the metabasidium. Thirumalachar (1950) had reported another type of metabasidium divided into four cells and arranged in a linear fashion. No sterigmata was reported. Moreover, the spore formation was described by directly rounding off of basidial cells. In the present observation, basidium was found with a linear row of four cells. Typical short hyaline sterigmata were found to develop from each cell bearing basidiospore on each. According to Ono (1984), the basidiospore of the terminal cell of basidium is the largest one and becomes successively smaller towards the basal cell.

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