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Rajiv Ranjan

Department of Botany, M.L.K. (P.G.) College, Balrampur, Uttar Pradesh, India *Pseudocercospora eugeniae-heynianae* sp. nov. on *Eugenia heyneana* from Tilkoma forest, Gorakhpur, U.P., India

Rajiv Ranjan

Abstract

During survey for the foliicolous fungi from diversified habitats of Tilkoma Forest, Gorakhpur U.P. India, we came across an important plant of the locality, Eugenia heyneana (Family-Myrtaceae) commonly known as Kath Jamun is a large shrub or small tree, up to 6 m tall, with branchlets obscurely four-edged. Leaves are simple, opposite, estipulate; leaf-stalk 4-5 mm long, slender, grooved above, hairless; blade 6-10 x 1.3-3 cm, oblong or elliptic, base narrow, tip blunt or flat, margin entire, leathery, hairless, pale beneath, glandular dotted; lateral nerves many parallel, close, but slightly irregular, very slender, slightly prominent on both sides, looped at the margin forming intramarginal nerve, intercostal net veined, slender prominent as lateral nerves. Flowers are bisexual, small, stalk less, white, in lateral, at branch-ends or rarely in leaf-axils cymes; sepal-cup tube 2 x 3.5 mm, top-shaped, no thick disc; petals calyptrate, 3 mm across; stamens many, free, bent inwards at the middle when in bud; filaments 3 mm long; ovary 2-celled; ovules many; style 1; stigma simple. Fruit is a berry, obovoid, 1-1.5 x 0.5-0.7 cm, crowned with the cup-like sepal-cup limb. Kath Jamun is native to India. On critical study the living leaves were found to be infected with Pseudocercospora. Since it has also been customary for plant pathologists and mycologists to describe as new any Cercospora or Pseudocercospora found a host for the first time (Ellis, 1971), this undescribed taxa has been described and illustrated as Pseudocercospora eugeniae-heyneana e sp. nov. Rajiv Ranjan. The review of available literatures reveals that there has been no record of this fungus from India on this host so for. Therefore, this host of the new species is a new record to Indian mycoflora from Tilkoma Forest, Gorakhpur U.P. India.

Keywords: Foliicolous fungi, *Pseudocercospora*, Gorakhpur, morphotaxonomic treatment, camera lucida

Introduction

The leaves provide a very suitable habitat for the growth & development of fungal pathogen by providing ample surface area and nutrient supply. Such leaf inhabiting fungi are known as foliicolous and the invaded area of the leaf appears as leaf spot or leaf lesion. Taxonomic studies of such fungal forms have been generally considered as only of academic interest but the taxonomic treatment of a fungal organism in the first requirement for any studies concerning its biology. Correct identification of a fungus absolutely free from ambiguities is vital for its employment in applied disciplines. In fact without being equipped for ascertaining the correct identity of a fungal pathogen all studies concerning its phytopathological aspects would be misleading. The weed and forest plants serve as reservoirs of leaf spot pathogens which on getting opportunity may spread to agriculture and horticulture plants. Eastern Uttar Pradesh is also well diversified for Foliicolous fungi. Keeping this in view we surveyed the locality of Tilkoma Forest, Gorakhpur during January 1999 to December 2001.

The Foliicolous Fungi causes huge losses every year in different parts of the world. The fungal pathogens producing leaf spots infect a large variety of hosts including most of the crops, forests and other plants. The destruction caused by these enemies of leaves is a serious problem before us. The focus of this research is identification & documentation of foliicolous fungi which will assist in the discovery of new fungicides and ideas to overcome from the severity of these enemies of nature as well as in the protection of floral diversity from the infection of these pathogens and also in the conservation of valuable flora of the area.

Correspondence: Rajiv Ranjan Department of Botany, M.L.K. (P.G.) College, Balrampur, Uttar Pradesh, India

Materials and Methods

The climatic condition favors the growth of various types of phanerogamic vegetation along with seasonal and annual crops and other plants. With a view to study the foliicolous fungi in their natural habitat, frequent collection trips will be arranged. The following articles would be required for collecting foliicolous fungi-collection containers, hand lens, pruning scissor or secateurs, light plant pressures, blotting paper, paper envelop, field note book etc.

Laboratory processing and preliminary examination:

Preparations

- (a) Photograph of both host and pathogen will be taken.
- (b) Scrap mount: If the organisms are superficially attached with the host tissue scrap mounts are made by a sharp razor or scalpel.
- (c) Collodion Preparation: A drop of collodion solution is applied to a colony on the leaf. The fungus gets embedded entirely and the dried film is peeled off readily from the host surface. Removal of colliding by acetone on a glass slide gives undisturbed preparation.
- (d) Squash preparation: The fruiting body is mounted, cleared and examined. Then the preparation is tapped vigorously and reheated. In this way the fruiting body is broken and content is released.
- (e) Hand cut Section preparation: A hand cut section of infected tissue is made with sharp razor to study immersed or semi-immersed fungi. Section cutting for host parasite interaction / relation.

Staining and Mounting

For routine microscopic study in the lab temporary slides are made in different type of stains and mountants according to nature of fungal forms involved.

- (a) Lacto- phenol cotton blue: The lacto-phenol mounting fluid is used for mounting-colored fungi. For locating cytoplasm, septa, guttules other structures and hyaline forms 0.05-0.01% cotton blue is added.
- (b) **Poly- vinyl Alcohol:** Benson, 1969 is used in routine staining and mounting.
- (c) Lacto-fuchsin: By this cell walls are stained more clearly, rapidly and with more suitable color specially for photography [Carmichael., 1955] ^[15]. Slides prepared in mountants are sealed with wax or commercial good quality nail polish and are stored for further study.

Camera Lucida: Drawings will be made of the distinctly different taxa of generic or species rank so as to show the morpho taxonomic features of vital importance.

Morpho taxonomic treatment: Hitherto undescribed forms of foliar fungi will be executed with the help of present literature and expertise available at hand.

- New taxon will be described in English or Latin or both as and when required.
- Material (holotypes) will be deposited in recognized Herbaria for accession no.

During collection, infected leaf samples were taken in separate polythene bags. Suitable mounts of surface scrapping and hand cut sections were prepared from infected portions of the leaf samples. Slides were prepared in cottonblue lacto phenol mixture & were examined. Camera Lucida drawing were made and the morpho-taxonomic determination of taxa was done using available literature and with the help of resident's expertise available. The fungal taxa were identified using microscopic preparation.

Result and Discussion

The author surveyed during January 1999 to December, 2001 in diversified habitats of Tilkoma Forest, Gorakhpur U.P. India for the collection, study and documentation of the leaf spot micro fungi infecting variety of the angiosperms has resulted in abundant gathering of the fungal specimens. During survey the author came across an important plant of the locality, *Eugenia heyneana* (Family-Myrtaceae) commonly known as Kath Jamun is a large shrub or small tree, up to 6 m tall, with branchlets obscurely four-edged. Leaves are simple, opposite, estipulate; leaf-stalk 4-5 mm long, slender, grooved above, hairless; blade 6-10 x 1.3-3 cm, oblong or elliptic, base narrow, tip blunt or flat, margin entire, leathery, hairless, pale beneath, glandular dotted; lateral nerves many parallel, close, but slightly irregular, very slender, slightly prominent on both sides, looped at the margin forming intramarginal nerve, intercostal net veined, slender prominent as lateral nerves. Flowers are bisexual, small, stalk less, white, in lateral, at branch-ends or rarely in leaf-axils cymes; sepal-cup tube 2 x 3.5 mm, top-shaped, no thick disc; petals calyptrate, 3 mm across; stamens many, free, bent inwards at the middle when in bud; filaments 3 mm long; ovary 2-celled; ovules many; style 1; stigma simple. Fruit is a berry, obovoid, 1-1.5 x 0.5-0.7 cm, crowned with the cup-like sepal-cup limb. Kath Jamun is native to India. On critical study the living leaves were found to be infected with *Pseudocercospora*. Since it has also been customary for plant pathologists and mycologists to describe as new any Cercospora or Pseudocercospora found on a host for the first time (Ellis, 1971)^[22], this undescribed taxa has been described and illustrated as Pseudocercospora eugeniae-heynianaesp. nov. Rajiv Ranjan. The holotype specimen has been submitted in HCIO, IARI New Delhi for allotment of accession number. The Accession no. is 43862.

Pseudocercospora eugeniae- heyniannae Rajiv Ranjan sp. nov.

Maculae: Hypotenuse, irregulars.

Coloniae: Hypophylla, effuse, cinerae.

Mycelium: Hyphae immersae, racemose, septate.

Stromata: Pseudoparenchymatous, immerse, fuscoolivaceae 16.0-36.0 μm. diam.

Cellulaeconidiogeniae: Integratae, polyblasticae, sympodiales, denticulatae

Conidia: Holoblastica, solitaria, sicca, acropleurogenosa, non ramose, pallidevelolivacae, cylindratavelobclavato cylindrical, recta velleniterflexuosa, laevia, 1-5 transverse septate, apiceacuta et basiobconico truncate valsubtruncata, hilo non incrassato, 14.0-52.5 \times 3.5-6.0 µm In foliisvivis *Eugenia eheynianae* Duthie. (Myrtacearum), Dec. 1998; Tilkoma Forest, Gorakhpur, leg. R.R. Srivastava, M.L.K./ R.R- 1060 holotypus, HCIO-43862 isotypus

Infection spots: Hypophyllous, irregular

Colonies: Hypophyllous, effuse, dull grayish

Mycelium: Hyphae immersed, branched, septate, hyaline

Stromata: Developed, pseudoparenchymatous, immersed, dark olivaceous, 16.0-36.0 µm diam.

Conidiophores: Fasciculate, 2-8in each fascicle, macronematous, mononematous, septate, mostly erect to slightly flexuous, originating from stroma, not gerniculate, smooth-walled unbranched, dark olivaceous, $26.0-84.5 \times 3.5-6.5 \mu m$

Conidiogenous cells: Integrated, polyblastic, sympodial, denticulate

Conidia: Holoblastic, solitary, dry, acropleurogenous, unbranched, light to moderate olivaceous, cylindrical to obclavato-cylindrical, erect to slightly flexuous, smooth-walled, 1-5 transversely septate, apices acute to obtuse with obconico-truncate to subtruncate bases, hila unthickened, $14.0-52.5-6.0 \mu m$.

On living leaves of *Eugenia heyneana* Duthie (Mytaceae), Dec. 1998; Tilkoma Forest, Gorakhpur, leg. R.R. Srivastava, M.L.K./ R.R.-1060 holotype, HCIO-43862 isotype

A survey of literature (Table) shows that two species of *Pseudocercospora* have been described on different host genera of the host family, viz. *P. eucalyptorum* (Crous *et al.*, 1989) and *P. eucalypti* (Hsieh & Goh, 1990). However, *Cercosporaeugeniae* Rangel has been described on the same host genus but on a different host species viz. *Eugenia javanica* (Chupp, 1953). This fungus is not a true *Cercospora* and it appears to be better accommodated in *Pseudocercospora*. Our collection is, therefore, comparable with *C. eugeniae*. A comparative account of the morphotaxonomic features of *C. eugeniae*nad the present collection is given in table 1 as follows:

Table 1: Comparison of Morphotaxonomic Features of P. eugeniae-heynianae with C. eugeniae

Species	Stroma	Conidiophore	Conidia
C. eugenia Rangel	Dark brown,	Sub-hyaline to very pale olivaceous brown,	Sub-hyaline to very pale olivaceous,
	globular	septate, bluntly rounded tip, $3.0-5.0 \times 10-40 \ \mu m$	3.0-5.0 septate, 2-4× 20-85 μm
P. eugeniae-	Devdoped, dark	Dark olivaceous, 1-5 septate, 26.0-84.5× 3.5-6.4	Light to moderate olivaceous, 1-5
heynianae sp. nov.	olivaceous	μm	septate, 14.0-52.5× 3.5-6.0 µm

A perusal of the morphotaxonomic features presented in the table suggests that *Cercospora eugeniae* (which appears to be assignable to *Pseudocercospora* as per present day concept in the taxonomy of *Cercospora* complex) differs from the fungus in question in bearing exclusively hypogenous colonies, much longer conidiophores and smaller conidia as opposed to amphigenous colonies, smaller conidiophores and longer conidia in the latter. Therefore, our collection has been described and illustrated here as a new species.

Survey of Literature Kamal *et al.*, 1986 ^[25]; Ellis, 1971 ^[22], 1976 ^[23]; Deighton, 1967 ^[17], 1973 ^[18], 1974 ^[19], 1976 ^[20], 1979 ^[21]; Braun, 1987 ^[10], 1988a, b ^[11, 12], 1991 ^[13]; Braun *et al.*, 1992 ^[14]; Bagyanarayan and Braun, 1991, 1992, 1999; Bagyanarayan *et al.*, 1991 ^[1], 1992 ^[2, 3], 1994 ^[4], 1995 ^[5]; Vasudeva, 1963 ^[16]; Chupp, 1954 ^[16]; Bilgrami *et al.*, 1979 ^[7, 9], 1981 ^[8], 1991 reveals that there is no record of *Pseudocercospora eugeniae-heynianae* species of this type on the host family. Therefore, it is described and illustrated as a new species to accommodate it.



Fig 1: *Pseudocercospora eugeniae-heynianae* Sp. nov. a. Conidia b. Conidiophore c. Stroma

Conclusion

The Region of Tilkoma Forest, Gorakhpur U.P. India is rich in phytodiversity in general as well as the diversity of fungal organisms inhabiting plant leaves in particular and it provides great scope for study of foliicolous fungi. Correct identity of a fungus absolutely free from ambiguities is vital for its employment in applied disciplines in general and it is more so for plant pathology where precision of details about the biology of the pathogen is primarily conditioned by its identity. In fact, without being equipped for ascertaining the correct identity of a fungal pathogen all studies concerning its phytopathological aspects would simply be misleading. However the end is still not insight and further investigation is warranted.

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