Available online www.jocpr.com

Journal of Chemical and Pharmaceutical Research, 2017, 9(1):69-74

Review Article

ISSN : 0975-7384
CODEN(USA) : JCPRC5

Graphium Salixicum: A New Species Explored from *Salix Alba*

Sunita Chandel[1*], Sabina Rana[2] and Sanjeev Thakur[3]

[1]*Department of Plant Pathology, Dr. Y.S. Parmar University of Horticulture and Forestry, India*
[2]*Department of Biotechnology, Dr. Y.S.Parmar University of Horticulture and Forestry, India*
[3]*Department of Tree Improvement and Genetic resources, Dr. Y.S.Parmar University of Horticulture and Forestry, India*

ABSTRACT

In the present investigation, a new fungal species, Graphium salixicum was isolated and identified from the plant, Salix alba. The current study is the first study ever done to report the said species. The infected samples associated with spots on bark, stem, branches of Salix alba family: Salicaceae (willow family) from Regional Horticultural Research and Training Station, Sharbo, district Kinnaur (Himachal Pradesh) forest were collected. The diseased samples were placed in moist chambers at 20ºC to induce fungal growth and sporulation. The new synnematal anamorphs assigned to the new species of Graphium were reviewed and compared with the previous species reported initially. The taxonomic characteristics of the species were resembled to the genera Graphium and the differentiating characteristics were considered to represent a new species.

Keywords: Tap water; Underground water; Atomic absorption spectroscopy

---

INTRODUCTION

The genus *Graphium* has not been monographed yet and contains numerous poorly characterized species. Among them, species from insect galleries have been relatively well studied. Phylogenetically related lineages of *Parascedosporium*, *Petriella*, *Pseudallescheria* and *Scedosporium* occur on mammals, in dung, soil and wood, often produce *Graphium* type conidial morphs and form sexual stages. *Graphium* was first described for five species, including *Graphium penicillioides* [1] later named the lectotype species for the genus, which was defined as having per current conidial development [2]. *Graphium*-like synnematous fungi, occur in the Microascales (*Graphium*), of family Microascaceae which is known to produce proliferation per currently and terminal annelledic Three *Graphium* species were recently described from *Adansonia*, namely *Graphium adansoniae* (from South Africa), and *Graphium madagascariense* and *Graphium fabiforme* from Madagascar in addition to *Graphium jumulu* is insect-vectored, and commonly associated with tree wounds [3].

During our investigations infected samples associated with spots on bark, stem, branches of *Salix alba* family: Salicaceae (willow family) from Regional Horticultural Research and Training Station, Sharbo, district Kinnaur (Himachal Pradesh) forest were collected. This tree contains phenol glycosides, salicortin and salicin is an analgesic through which salicylic acid and aspirin is prepared. The diseased samples were placed in moist chambers at 20ºC to induce fungal growth and sporulation. The new synnematal anamorphs assigned to the new species of *Graphium* after reviewing the literature and comparing their key taxonomic characters with known species from other host as well as with *Graphium carbonarium* recorded on *Salix babylonica* from China. The new taxon is described as follows:

***Graphium salixicum* Chandel, Rana and Thakur sp. nov**

Colonies, cottony, effuse hyaline to light grey, overgrowing hyphae 2-3µm wide covered with swollen , globose thin hyaline chlamydospores 4-10 µm diameter, Synnemata scattered, clearly visible under low power stereo binocular microscope. Mycelium superficial or immersed, Setae and Hyphopodia absent sporulated heavily on the host surface after 8-10 days in moist chamber at 20ºC. Synnemata 50-170 µm long, 12-20 µm wide, base some time swollen 15-28 µm diam., clustered with thick walled, brown, spherical Chlamydospores of 5-8 µm diam, developing from smooth walled creeping hyphae. Conidiophores arranged in synnematal, indeterminate, scattered over the surface, sporulation abundant on the host surface after 8-10 days by producing terminally conidial slimy head 40-70 µm diameter or conidia hanging together. Each synnema base swollen with group of chlamydospore, individual stipe of conidiophores light brown, unbranched, narrow upwards, straight or flexuous, smooth, 2-4 µm wide , bearing (1-3) conidiogenous cells arising at different level at the apex on a variable number of primary branches or may diverge penicillatly. Conidiogenous cells integrated, terminal, monoblastic discrete, subulate, or cylindrical, tapering 2-5 µm wide x 30-70 µm long, swollen or straight and pointed. Conidial development was annelledic. Conidia 0 - 3 septate, globose, or cylindrical to obovoid, thin walled , smooth, colorless or light pale brown with age 3-5 µm wide x 5-15 µm length . Teleomorph not observed.

Habitat

On bark, stem and branches of *Salix alba* trees forest, Regional Horticultural Research and Training Station, Sharbo-172 107, District Kinnaur (Himachal Pradesh), India; Collected by Sabina Rana, Dated 24-25th July, 2015, NCFT No 7836 .16 (Figures 1 to 9).

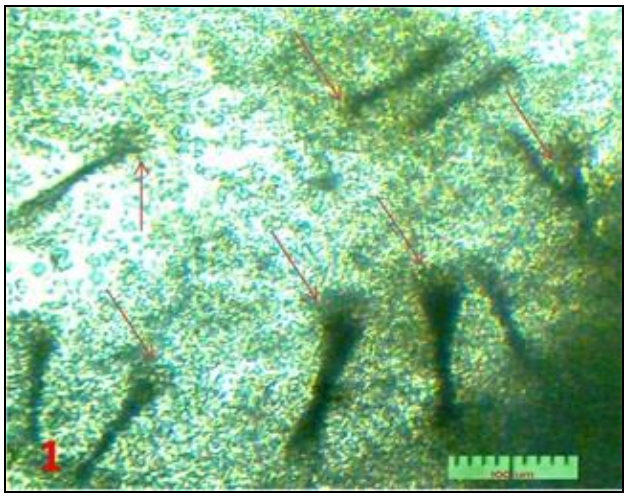

Figure 1: Synnematous conidiomata colony on host bark light micrographs

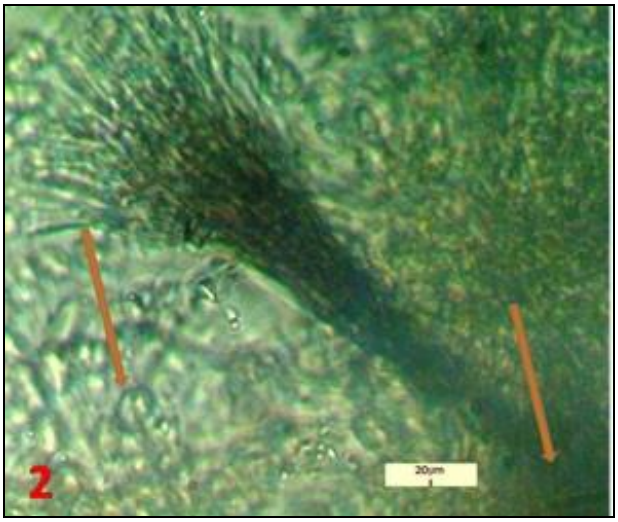

Figure 2: Synnematous conidiomata with parallel hyphae in the stipes

Figure 3: Synnematous conidiomata with Chlamydospores

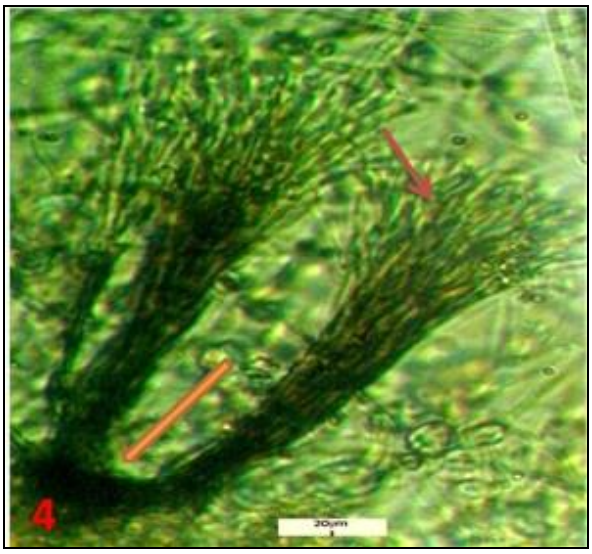

Figure 4: Synnematous conidiomata with divergent swollen base of Chlamydospores

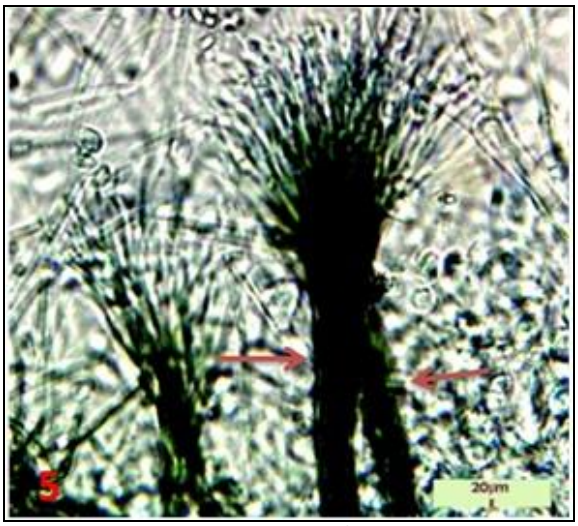

Figure 5: Divergent capitulum of conidiogenous cells

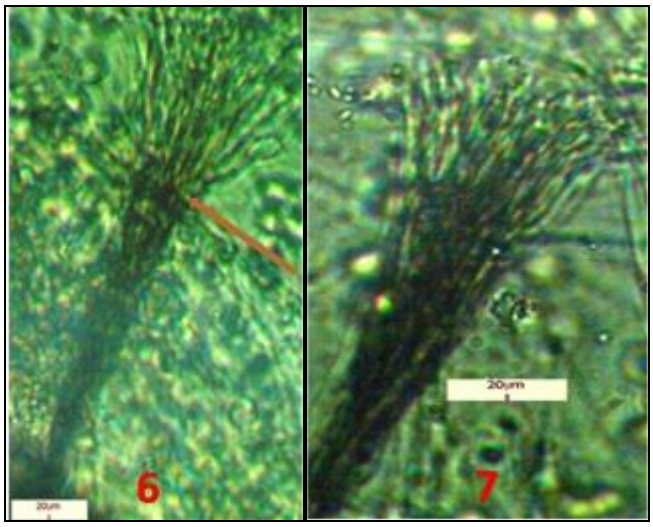

Figure 6 &7: Branching patterns of the metulae

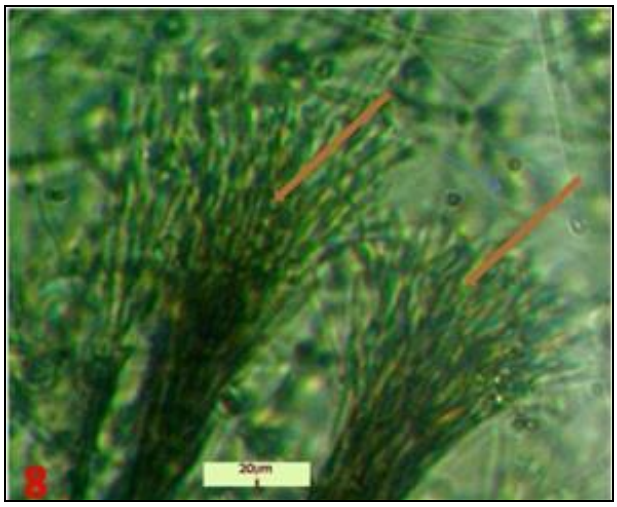

Figure 8: Conidiogenous cell showing annelledic conidium development

Figure 9: Conidia with 0-3 septation

Key to *Graphium* species

1. Synnemata up to 200 µm high, conidia straight, curved, cylindrical at apex truncate base 4-7 x 1-2 µm ……*G. penicillioides.*

2. Synnemata up to 300 -1000 µm high, conidia ellipsoidal to cuneiform, pale olivaceous brown 5-11x 2-4 µm ……..*G. puteridinis* (Corda)

3. Synnemata up to 5 mm high, conidia ellipsoidal to obovoide, olivaceous brown-black ellipsoidafal to ovoid, brown to black 1.5-3 x 1-2 µm ……..*G. calicioides* (Fr,)

4. Synnemata pale to dark brown up to 88–140 x 60–97 µm, Conidia aseptate, hyaline, cylindrical to obovoid, 4–5 × 1.5–2.5 µm, becoming brown with age Conidiophores with 2–4 branches, conidiogenous cells annellated…. *G adansoniae* [4].

5. Synnemata scattered, Stipes pale to dark brown, 89–174 µm long and 15–53 µm wide at the apex. Conidiophores with 2–4 branches, conidiogenous cells annellated. Conidia aseptate, hyaline, cylindrical to obovoid 3–5 × 1.5–2 µm, produced in a hyaline mucilaginous mass on the Synnemata, bright transparent when young, becoming brown with age …..*G. madagascariense*. Cruywagen and de Beer [4].

6. Synnemata dark brown, 319–496 x 308–674 µm, Conidia aseptate, hyaline, 3.5–4.5 × 1.5–2 µm, cylindrical to obovoid, with reniform becoming dark brown to black with age, Conidiophores with 2–4 branches conidiogenous cells annellated. …… *G. fabiforme* Cruywagen, and de Beer, (2010)...10 hium fabiforme Cruywagen & Z.W. de Beer [4].

7. Synnemata tuft-like, 100–150 x 5–15 µm diam at base with rhizoids, 20–60 µm diam at apex; stipe olivaceous-brown, apex pale olivaceous, straight to slightly curved, sub cylindrical with obtuse apex, somewhat swollen, 4–5 × 2.0 µm; conidiophores branching into up to four conidiogenous cells at apex. Conidiogenous cells olivaceous at base, pale olivaceous at apex, roughened at base, with several per current proliferations at apex, 20–35 × 1.5–2 µm. base truncate, 1–1.5 µm diam, with minute marginal frill. ….. *G. jumulu* [5].

8. Synnemata up to 1 cm long, conidiophores funicles; monoverticillate, biverticillate or irregularly branched, conidiogenous cells arising at various branching levels, discrete, 2–5 per branch, tapering distally, neck curved, 9.5 - 12 × 1.6 - 2 µm, Conidiation annelledic,. Conidia clavate, variable in size 4.5 - 8 × 1.5 - 2 µm or larger 8 -11 × 2.8- 5 µm; larger conidia are typical of young conidiophores, smaller conidia of older conidiophores, Swollen 'sprout' cells, 13 -20 × 5 - 8 µm, originate from conidia, ellipsoidal, lemon-shaped or irregular, thick-walled with a lumen …. *G. scolytodis* [6].

9. Conidiophores length 118--230 µm, conidia 4-6x1-2µm conspicuous basal frill present, Conidia in chains present, non septate, hyaline, cylindrical sub rounded end. Rhizoids absent, recorded on *Larix deciduas*….. *G. laricis* [7].

10. Synnemata dark pigmented, Rhizoids present Conidiophores mononematous *Sporothrix* type, 113–263 µm long, Conidia single celled from annelledic produced in chains and have conspicuous basal frills. Unicellular, hyaline., tunicate base , cylindrical to sub round apices 4-7.5 x 1.5-2.5 µm, recorded on *Pinus* sp. …… *G. pseudormiticum* [7].

11. Synnemata 118–230 µm long Conidia 4.4-12x2.0 µm size with conspicuous basal frill present and rhizoids absent, recorded on *Picea abies*…. *G. fimbriasporum* (Morelet) Jacobs [7].

12. Synnemata 134–225 x 31– 36 µm, and 46–58 µm wide at the apex singly, or in groups Rhizoid-like structures abundant, Conidiophores (stipe) with 2-4 branches, dark grayish brown at the base, becoming hyaline toward the apex, Conidiogenous cells 2–3 per branch point, 15–18 long and 1–3µm wide, with nodular annulations. Conidia aseptate, hyaline, curved, cylindrical with truncated bases, 4–6 ×1–3 µm, aggregate in a hyaline mucilaginous mass bright transparent white when young, becoming darker with age……..*G. carbonarium* Paciura, de Beer, Zhou & M.J. Wingf, (2010) on *Salix babylonica* [8].

13. Synnemata 50-170 x 12-20 µm size, and 15-28 µm wide swollen base composed of thick walled chlamydospore, light brown , spherical of 5-8 um diam., Conidiophores (stipe) light brown, unbranched, indeterminate, straight or flexuous, smooth walled, 2-5 µm wide, narrowed upwards, colorless terminally, bearing 1-3 conidiogenous cells. Conidiogenous cells 30-70 µm long and 2-5 µm wide, subulate, or cylindrical or tapering .Conidia 0 - 3 septate, globose, or cylindrical to obovoid, smooth walled, 3-5 µm wide x 5-15 µm long, aggregated in mucilaginous mass colorless or light pale brown with age …….*G salixicum* sp. nov recorded on *Salix alba* (India) [8].

JUSTIFICATION

Thirteen *Graphium* species cited above were reviewed for their taxonomical characters with regards to Synnemata, conidiophores (stipe), conidiogenous cells and conidia development in the light of their size, color, septation, rhizoid, chlamydospore formation to justify the new taxon Non amongst the above cited species do not found taxonomically closer in above cited characters to the new taxon of *Graphium salixicum*. Therefore, host specific species *Graphium carbonarium* recorded on *Salix babylonica* (Paciura, Z.W. de Beer, X.D. Zhou & M.J. Wingf. (2010) compared. The proposed new fungi *Graphium salixicum* recorded on ***Salix alba*** differed drastically with *Graphium carbonarium* in having simple, smaller, light brown, thin, and basal scleroidal Synnemata against branched, longer, dark brown to black , thick walled rhizoidal Synnemata, respectively in *Graphium carbonarium*. Conidiophores (stipe) unbranched, longer 30-70 µm longer and 2-5 µm wider. Conidiogenous cells metulae in *Graphium salixicum* found longer **(**30-70 x 2-5 µm) and branched as reported (15–18 x1–3µm) long and unbranched in *Graphium carbonarium***.** Conidia of *Graphium salixicum* were colorless or light pale brown, 0 - 3 septate, globose, or cylindrical to obovoid, smooth walled, base non truncated, (5-15 x 3-5 µm) size. While, Conidia of ***G. carbonarium*** were hyaline to dark brown, aseptate, cylindrical, curved with bases truncated, (4–6 ×1–3 µm) size [8].

ACKNOWLDGEMENT

The authors are highly thankful to the services rendered by Dr. Pankaj Gupta, Associate Director and Head, Dr. D. P. Bhandari Assistant Scientist (Plant Pathology) for extending the facilities in collection of diseased samples of *Salix alba* from forest plantations located at Sharbo, Kinnaur (Himachal Pradesh) and Dr. P.N.Chowdhry, Principal Mycologist, National Centre of Fungal Taxonomy (www.ncft.in), New Delhi for confirming above new taxon.

REFERENCES

[1] ACI Corda, **1837**, 1, 18.
[2] ACI Corda; SJ Hughes. *Canadian J Botany*, **1958**, 36: 770.
[3] EM Cruywagen; ZW De Beer; J Roux; MJ Wingfield, *Persoonia*, **2010**, 25, 61-71.
[4] P Barber; PW Crous. *Persoonia*, **2015**, 34, 167-266.
[5] M Kolařík; J Hulcr. *Mycol. Res.*, **2009**, 113, 44-60.
[6] IK Jacobs; T Kirisits; MJ Wingfield. *Mycologia*, **2003**, 95, 714-727.
[7] M Mouton; MJ Wingfield; PS Vanwyk; PWJ Vanwyk. *Mycol. Res.*, **1994**; 98, 1272-1276.
[8] D Paciura; ZW DeBeer; XD Zhou; MJ Wingfield. *Fungal Diversity*, **2010**, 40, 75-88.