

First report of downy mildew of bhringaraj [Wedelia calendulacea (L.) Less.] caused by a new Plasmopara species, *Plasmopara wedelia* from West Bengal

B. N. PANJA^{*}, R. CHEJARA, M. RANJANA DEVI, S. KUNDU AND J. SAHA



J. Mycopathol, Res, 54(2) : 311 -315, 2016;
ISSN 0971-3719

© Indian Mycological Society,
Department of Botany,
University of Calcutta,
Kolkata 700 019, India

This article is protected by copyright and all other rights under the jurisdiction of the Indian Mycological Society. The copy is provided to the author(s) for internal non-commercial research and educational purposes.

SHORT COMMUNICATION

First report of Downy Mildew of Bhringaraj [*Wedelia calendulacea* (L.) Less.] caused by *Plasmopara wedelia* from West Bengal

B. N. PANJA^{*}, R. CHEJARA, M. RANJANA DEVI, S. KUNDU AND J. SAHA

Department of Plant Pathology, Bidhan Chandra Krishi Viswavidyalaya, Mohanpur 741252, West Bengal

Received : 22.04.2016

RMS Accepted : 18.05.2016

Published : 25.07.2016

Wedelia calendulacea (L.) Less., known as Bhringaraj in Sanskrit and Bengali literature, belongs to the family Asteraceae, is an important herbal medicinal plant. Since 2013-14, the herb, grown in the medicinal plant garden of the University, has been suffering severely from downy mildew disease that infects the leaves, sepals, petals and leaving all the affected plants in burnt appearance within 4 - 6 weeks. Pathogenicity of that disease on Bhringaraj has been established. However, the pathogen fails to infect locally available eight annual flowering plants, viz. aster, calendula, dahlia, chrysanthemum, gerbera, sunflower, French marigold, African marigold, belong to Asteraceae. The pathogen associated with the disease produces sporangiophores which are aseptate, hyaline, branched and measured 183.9 – 412.7 μm (av. 289.2 μm) x 4.9 to 13.5 μm (av. 8.4 μm). The distance of first primary branch of sporangiophores from base is 95.9 – 239.6 μm (av. 161.3 μm), total number of primary branches 1 – 5 (av. 3.2), size 27.5 – 158.3 μm (av. 77.1 μm) x 3.2 - 8.9 μm (av. 5.3 μm), total number of secondary branches 1 – 4 (av. 2.1), size 13.1 - 68.5 μm (av. 40.3 μm) x 2.2 – 6.2 μm (av. 4.5 μm), branching angle between the main axis and primary branches as well as between primary and secondary branches 72.9° - 120.1° (av. 92.8°) and 66.5° - 123.7° (av. 90.2°) respectively. Blunt and truncated tips [size 4.9 – 16.8 μm (av. 8.1 μm) x 2.1 – 5.8 μm (av. 3.4 μm) and angle 63.4° - 122.3° (av. 87.7°)] of three are arranged either in single cluster or 2+1 fashion whereas the same of five are arranged commonly in 3+2, sometimes 3+1+1 and rarely 2+1+2 fashions. Sporangia are hyaline, round to ovoid, papillate, 14.0 - 27.8 μm (av. 20.1 μm) x 11.8 – 21.1 μm (av. 16.1 μm) in size and 138.6 - 469.5 μm^2 (av. 264 μm^2) in surface area. Based on above descriptions, the causal agent of Bhringaraj downy mildew disease has been identified as new *Plasmopara wedelia* that differs from sunflower infecting *Plasmopara helianthi* in many respects.

Key words: Bhringaraj, downy mildew, medicinal plant, *Plasmopara*, *Wedelia calendulacea*, West Bengal

Wedelia calendulacea (L.) Less., familiar as Bhringaraj (in Sanskrit and Bengali literature), is a procumbent, perennial herbal medicinal plant, be-

long to the family Asteraceae. It is commonly found in wet places of Uttar Pradesh, Assam, Arunachal Pradesh, along the coastal areas of Bengal and Konkan, and districts of Madras Presidency (Tamil Nadu) in India. Outside India, this crop has been

*Corresponding author : birenpanja@rediffmail.com

Table 1 : Comparison between host range, morpho-metrical descriptions of sporangium and sporangiophores of *Plasmopara* from bhringaraj and *Plasmopara helianthi* reported earlier from sunflower

Parameters recorded on	<i>Plasmopara</i> from Bhringaraj	<i>Plasmopara helianthi</i> from sunflower (Novotel'nova, 1960)
No. of hosts examined under <i>Asteraceae</i>	Eight hosts examined but none is infected	Over 80 hosts are infected
Sporangiophores		
Length and breadth	183.9 – 412.7 μm (av. 289.2 μm) in length \times 4.9 to 13.5 μm (av. 8.4) in breadth	300 – 750 (av. 450) μm in length \times 7 – 14 μm in breadth
Distance of first primary branch on the main axis from the base of sporangiophores	95.9 – 239.6 μm (av. 161.3 μm)	Branching at upper half of the sporangiophore
Primary branch total no., length and breadth, angle with the main axis	1 – 5 nos. (av. 3.2), 27.5 – 158.3 μm (av. 77.1 μm) in length \times 3.2 – 8.9 μm (av. 5.3 μm) in breadth, 72.9° - 120.1° (av. 92.8°) in angle	7 – 8 in nos., right angle
Secondary branch total no., length and breadth, angle with the primary branches	1 – 4 nos. (av. 2.1), 13.1 – 68.5 μm (av. 40.3 μm) in length \times 2.2 – 6.2 μm (av. 4.5 μm) in breadth, 66.5° - 123.7° (av. 90.2°) in angle	2 – 5 nos., 40 – 86 μm long, right angle
Number of tips**, its length and breadth at base, angle	Commonly 3 – 5 but rarely 2 in no., 4.9 – 16.8 μm (av. 8.1 μm) in length \times 2.1 – 5.8 μm (av. 3.4 μm) in breadth, 63.4° - 122.3° (av. 87.7°) in angle	3 – 5 in nos., 8 – 15 μm long, right angle
Sporangia		
Shape	Round to ovoid	Ovoid to ellipsoid
Length and breadth	14.0 – 27.8 μm (av. 20.1 μm) in length \times 11.8 – 21.1 μm (av. 16.1 μm) in breadth	18 – 30 in length \times 14 – 20 μm in breadth
Area	138.6 – 469.5 μm^2 (av. 264 μm^2) in area.	–
Antheridia	Searched but not observed	Antheridia club-shaped, 30 μm in length \times 12 μm in breadth
Oogonia	Searched but not observed	Spherical, 30 – 40 μm diameter, colourless
Oospores	Searched but not observed	Spherical, 15 – 30 (23) μm diameter, yellow-brown, with a slightly wrinkled wall, 3 μm thick

** Tip of the secondary branches was either unbranched or branched. When unbranched it borne mostly three blunt tips [arranged commonly in single clustered of three tips but rarely in two clusters of three tips in 2 + 1 fashion] and rarely two blunt tips in single cluster (Fig. 1c). When branched once it had mostly five blunt tips [arranged commonly in 3 + 2, sometimes 3 + 1 + 1 and rarely 2 + 1 + 2 fashions].

reported to grow in wet areas of Burma, China, Ceylon (Srilanka), Malay Archipelago (Brunei, East Malaysia, East Timor, Indonesia, Singapore and Philippines) and Japan (Kirthikar and Basu, 2006). Leaf, stem and even whole plant parts of *W. calendulacea* have been used as a folklore medicine for various ailments and reported to possess anti-oxidant, anti-inflammatory, analgesic, anti-microbial, hepato-protective, central nervous system depressant, anti-osteoporotic, anti-convulsant, wound healing, sedative, anti-stress, anti-ulcero-

genic and anti-cancerous activity (Koul *et al*, 2012) due to the presence of flavonoids, diterpenes, triterpene, saponins and phytosteroids (Masoodi *et al*, 2011) and main active constituents coumestans i.e., wedelolactone and demethyl-wedelolactone. Several biotic and abiotic factors limit the production of this medicinal plant and reduce the biosynthesis of active principles in the plant parts.

Since 2013, the present investigator group ob-



Fig. 1a: Symptoms of downy mildew affected bhingaraj plant and leaves with different grades of severity showing whitish downy growth of fungus on lower leaf surface

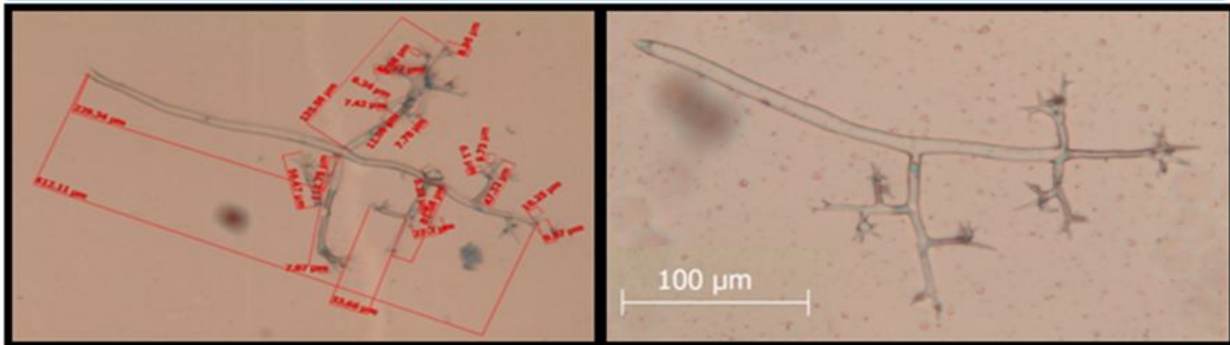


Fig. 1b: Dimension of the main axis, primary and secondary branches of sporangiophores

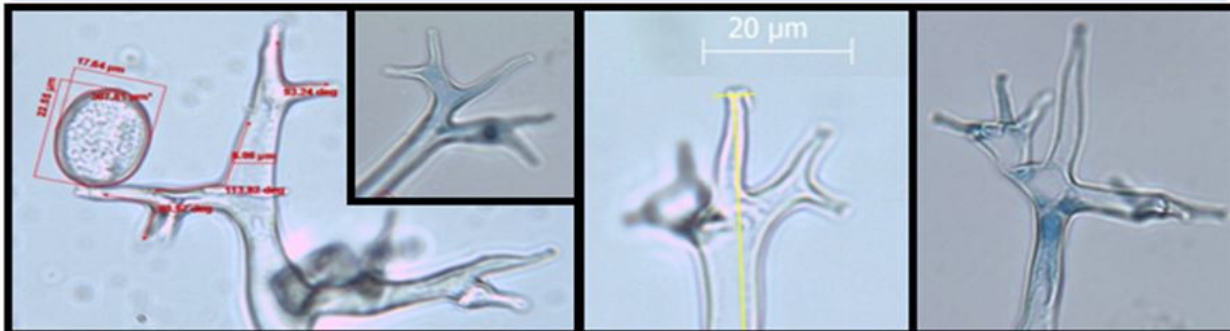


Fig. 1c: Sporangiophores showing different terminal branching pattern with blunt and truncated tips at their ends

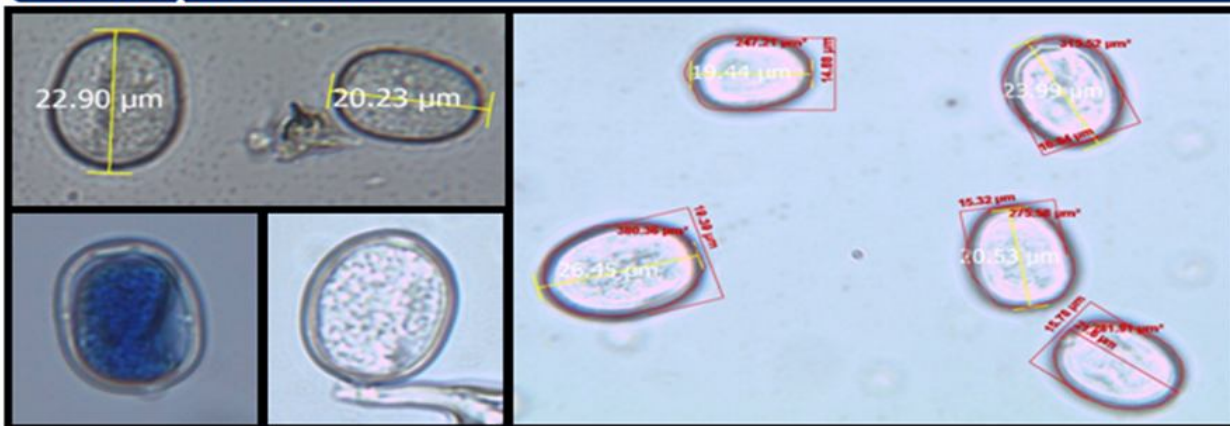


Fig 1d: Size, shape and surface areas of papillate sporangia

Fig. 1: Downy mildew disease symptoms of bhingaraj plant; dimensions, branching patterns and angles of sporangiophores, dimensions and surface areas of sporangia of *Plasmopara wedelia*

serves that *W. calendulacea* is suffering severely from a new disease with downy mildew like symptoms (Ranjana Devi, 2014). A detailed study on the disease along with its causal agent has been conducted during present investigation.

Affected leaves with different grades of disease severity were collected from medicinal plant garden of Bidhan Chandra Krishi Viswavidyalaya (BCKV) (Latitude 22°94' and Longitude 88°53') at Mohanpur, Nadia, for studying the disease symptoms, reproductive structures of pathogen, pathogenicity and host range. Symptoms were described based on field observation and by examining collected leaves. For studying asexual and sexual reproductive structures, the whitish downy growth of pathogen and the thin transverse sections of the diseased portion of leaves were taken respectively on a clean glass slide with a drop of lacto-phenol, followed by with or without staining with cotton blue, covered these carefully with cover slip and observed under phase-contrast and compound light microscope. Necessary micro-photographs were taken under different magnifications of Leica Laborlux K microscope with dedicated camera of model Leica EC3. Dimensions of various structures of the pathogen were measured using Carl Zeiss Axio Vision Rel. 4.8.2 software. For pathogenicity and host range studies, leaves showing fresh whitish downy growth of pathogen were selected, cut into small pieces, suspended in sterile distilled water to form a spore suspension of approx. 2×10^5 sporangia per ml. After proper cleaning of the apparently healthy leaf surfaces of Bhringaraj and other eight annual flowering plants, belonged to the family Asteraceae, viz. aster (*Callistephus chinensis* Linn.), calendula (*Calendula officinalis* Linn), dahlia (*Dahlia variabilis* Desf.), chrysanthemum (*Chrysanthemum indicum* Linn.), gerbera (*Gerbera jamesonii* Bolus ex Hook f.), sunflower (*Helianthus annuus* Linn.), french marigold (*Tagetes patula* Linn.), african marigold (*Tagetes erecta* Linn.) with sterile distilled water and absolute alcohol, they were artificially inoculated with spore suspension keeping suitable control, covered with transparent polythene packets for 48 hrs and observed regularly till symptom development.

Symptom appears on the under surface of lower leaves as whitish fluffy growth of the pathogen. The corresponding upper surface of leaf shows light yellowish-green mosaic areas. As the disease advances, the entire under surface of affected leaf is

covered by the white fluffy downy growth of the pathogen and the corresponding upper surface shows uniform yellow discolouration which subsequently turns dark brown giving a necrotic or burned appearance within a few days (Fig. 1a). Finally, the affected leaves dry up; gradually fall off from the twigs in acropetal succession leaving the bare twig with or without a few young leaves at the top. Affected petals and sepals too show similar pathogen growth, exhibit brown necrotic lesion and finally dry up as the disease progresses. Based on the symptoms, the disease was identified as Downy mildew. It spreads very rapidly to all other plants and burns the affected plants within 4 – 6 weeks depending upon the prevailing weather condition. It becomes visible in the month of February and continues till the end of April. After a brief disappearance during warm and rainy months, it reappears in the month of September and continues up to November.

During pathogenicity and host range studies of the pathogen, typical whitish fluffy downy growths of pathogen resembling field under naturally infested condition were developed at the under surface of lower leaves of Bhringaraj plant within 10 – 12 days after inoculation whereas no such sign and symptom was observed, even one month after inoculation, at the under surface of leaves of the above mentioned eight annual flowering plants.

Pathogen produced white fluffy downy growth comprising of sporangiophores and sporangia at the under surface of leaves (Fig. 1a). Detailed description and dimensions of sporangiophores – main axis, primary and secondary branches, its branching pattern (i.e. monopodial) (Fig. 1b), branching angle (i.e. 90° or above) (Fig. 1c), nature of tips (i.e. blunt and truncated tips) (Fig. 1c) of sporangiophores as well as size, shape and surface area of papillate sporangia (Fig. 1d) including the host range were recorded and compared with the morphological descriptions and host range of *Plasmopara helianthi* Novotel' nova [= *Plasmopara halstedii* (Farlow) Berlese & de Toni] (Novotel' nova, 1960) obtained from sunflower (Table 1). From the above comparison, it can be argued that the two *Plasmopara* species are different. It would be worthy to mention here that there was no report on the incidence of *Plasmopara helianthi* (= *P. halstedii*) on sunflower or any other member of Asteraceae from West Bengal (CABI/EPPO 2014). Hence, the causal

agent of Bhringaraj Downy mildew disease can be considered here as a new species of *Plasmopara*, *Plasmopara wedelia*. Species name of the pathogen has been chosen as *wedelia* based on the genus name of the host. The disease specimen has been deposited in the Herbarium Cryptogamae Indiae Orientalis (HCIO), Division of Plant Pathology, Indian Agricultural Research Institute (IARI), New Delhi (HCIO No. 51810), India.

ACKNOWLEDGEMENTS

The authors are grateful to the Officer In-charge, Medicinal Plant Garden, BCKV, Mohanpur, Nadia, West Bengal for providing necessary facilities in carrying out pathogenicity establishment experiment under field conditions. We want to acknowledge HCIO, IARI, New Delhi for providing necessary technical support to get accession of the submitted disease specimen.

REFERENCES

- CABI/EPPO. 2014. *Plasmopara halstedii* [Distribution map] Distribution Maps of PLant Diseases, No. October, Wallingford, UK; CABI, Map 286 (Edition 6).
- Kirthikar, K.R. and Basu, B. D. 2006. *Indian Medicinal Plants*. International Book Distributor, Dehradun. pp. 1345 – 1364
- Koul, S., Pandurangan, A. and Khosa, R. L. 2012. *Wedelia chinensis* (Asteraceae) - An overview. *Asian Pac. J. Trop. Biomed.* **2**: S1169 - S1175
- Masoodi, M. H., Ahmad, B., Wali, A. F., Zargar, B. A. and Dar, M. A. 2011. Recent developments in phytochemical and pharmacological studies of *Wedelia calendulaceae*- A review. *Indian J. Nat. Prod.* **27**: 3-7
- Novotel'nova.1960. Downy mildew on sunflowers, Sbornik Doklad. Nouchun.[Conference on Plant Protection]Tallin, Estonia, pp. 129-138.
- Ranjana Devi, M. 2014. *Studies on Diseases of Some Common Medicinal Plants in West Bengal*. An unpublished M. Sc. (Ag.) thesis submitted to the Bidhan Chandra Krishi Viswavidyalay, Mohanpur, Nadia, West Bengal, India, p.59