

Asiatic Fungal Weevil (*Eurymycter napinatus*)-transmitted *Fusarium thapsinum* Infection in *Uraria picta* (Jacq.) DC.

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ABSTRACT

Fusarium thapsinum infection (characterized by macro- and microconidia morphology along with growth characteristics on PDA) transmitted by Asiatic fungal weevil (*Eurymycter napinatus*) is first reported for the species *Uraria picta* (Jacq.) DC. (Family: Leguminosae, Papillionoidae; an important herb in Ayurvedic medicine) from control, M_1 and M_2 populations (grown as a rainfed *kharif* crop – May to November) in random plants. Symptoms noted were drying up of apical buds, downward spiral curling and stem rot in the near vicinity at the onset of flowering. Microscopic examination showed degeneration of parenchymatous pith and anthers, browning of vascular bundles and pollen grain degeneration, agglutination and sterility with progressive infection. The infection did not impair the productivity of the affected plants as frequent lateral buds were produced (uncharacteristic to the species morphology) on the side of the infected buds.

Keywords: U. picta, weevil, F.thapsinum

Abbreviations: dES, diethyl sulphate; EMS, ethyl methane sulphonate; HA, hydroxyl amine; M_1 , first mutant generation; M_2 , second mutant generation; PDA, potato dextrose agar

Fusarium species are reported to possess a broad host range causing crown and root rots, stalk rots, head and grain blights and vascular wilt diseases (Nelson et al. 1981, Summerell et al. 2001) primarily and to lesser extent diseases such as malformation in mango (Ploetz 2001), bakene disease in rice (Summerell *et al.* 2003) amongst others. *Fusa*rium infection was identified microscopically from diseased parts (apical bud, adjoining leaflets and stem) of Uraria picta (Jacq.) DC. (Family: Leguminosae, Papillionoidae; an important herb in Ayurvedic medicine) from control as well as mutagenized population (treatments: ethyl methane sulphate (EMS, 0.02M phosphate buffer at pH 6.8 used for dilutions), diethyl sulphate (dES, aqueous) and hydroxyl amine (HA, aqueous); 0.25, 0.50 and 1.00% for 3 and 6 h; seeds were treated and sown in experimental garden along with control representing M₁ population, seeds collected from selfed inflorescences of individual plants and sown in plant to rows formed the M2 generation) grown in an experimental field (as rainfed Kharif crop: May to November) of University of Kalyani (Latitude: 22° 11′ 6″ to 23° 5′ 2″ N, longitude: 88° 2′ to 89° 5′ E) during the onset of flowering (July to mid-August: Meteorological data obtained from B.C.K.V. is pooled over 2009 and 2010; temp. 31.6 to 39.7°C max., 24.0 to 26.8°C min; relative humidity 92.2 to 99.2% max, 56.8 to 84.7% min; rainfall - 2.08 mm to 18.18 mm). Along the course of induced mutagenesis experiment to raise a desirable 'plant type' in U. picta, symptoms of infection in relation to normal (Fig. 1) was observed as premature drying of apical buds (Fig. 2) followed by downward spiral curling (Fig. 3) of adjoining leaflets and finally rotting (browning) of stem (Fig. 4) in the near vicinity. Identical symptoms were studied in control, $M_1\,(38\ plants\ infected\ out\ of\ 273\ -\ 13.92\%)$ and $M_2\,\,(54\ out\ of\ 1263\ -$ 8.04%) plant populations.

Serial transverse hand sections of the stem from 5.0 cm above and below affected region showed progressive disintegration and browning of the parenchymatous pith (**Fig. 5**). Closer to infection zone vascular tissues also showed signs of browning. Complete pollen grain deformation, agglutination and sterility (as per 1% acetocarmine staining – Marks 1954) were noted at the onset of apical bud drying; anther degeneration was closely followed with the progression of symptoms.

It is interesting to note that insects (Encylopedia of Entomology – 4^{th} Ed. Springer 2006) namely, Asiatic fungal weevil (*Eurymycter napinatus*; Family: Anthribidae- Fig. 6) and Six spotted ladybug (*Anisolemnia dilatata*; Family: Coccinellidae) were predominant visitors (collected using drop trap and fly paper) during flowering. Wells and Payne (1976) reported a total of 2392 fungal isolates from weevil damaged pecans. Parry *et al.* (1995) proposed that the mode of dispersal of Fusarium ear blight (scab) in small grain cereals due to arthropod vectors.

Fusarium infection was evidenced from field samples from the typical morphology of the microconidia under light microscope (simple swabs/rubbings on a glass slide stained with cotton blue, mounted with lactophenol). Macroconidia (size: $113 - 126 \mu m$, mean = 119.50 ± 0.62) was relatively slender, slightly falcate (Fig. 7), thin walled, septation commonly 3 - 5 rare often 6, apical cell curved and slightly tapering, basal cell relatively poorly developed not foot shaped; microconidia (size: $20.6 \ \mu m \pm 1.06$) single septate, club shaped, flattened base (Fig. 8); chlamydospores absent. Similar morphological observations of conidial structures were noted in cultured samples (inoculums: homogenized infected field samples - apical bud, adjoining leaflets, affected stem parts and homogenized mouth parts of both insect visitors) on potato dextrose agar (PDA -Atlas 2004) excepting that of Six spotted ladybug (no growth in PDA).

Upon culturing on PDA (including subcultures on PDA at an interval of 96 h) all 4 inoculums yielded colonies with abundant white mycelium on the upper surface (**Fig. 9**) and tell tale yellowish pigmentation on the lower surface of the agar (**Fig. 10**), incubated at 37°C for 48 to 72 h. Colonies showed characteristic radial growth followed by purplish pigmentation in the white mycelium (168 h onwards). Based on macro- and microconidia morphology and growth



Figure plate I (1-6) *Uraria picta.* (1) Uninfected floral bud. (2) Infected (dried) apical bud with lateral bud formation and six spotted ladybug. (3) Symptom showing downward spiral curling of leaflets. (4) Stem rot. (5) Transverse section of stem showing complete disintegration of parenchymatous pith and browning of vascular bundles (scale bar = 5 mm). (6) Asiatic fungal weevil feeding on young uninfected floral bud.

characteristics on PDA it seems that the infecting organism is *F. thapsinum* (white mycelium and yellowish pigmentation in PDA is species specific) which corroborates taxonomic evidence (Klittich *et al.* 1997) and the species epithet (Leslie and Summerell 2006). Earlier Summerell *et al.*



Figure plate II (7-10) *Fusarium thapsinum.* (7) Falcate macroconidia. (8) Napiform microconida. (9) White mycelial growth on PDA – upper surface. (10) Yellow pigmentation on PDA – lower surface. Scale bar = $100 \mu m$.

(2003) reported stem rot of Sorghum by F. thapsinum.

Being the very first report of any disease in *U. picta*, the *F. thapsinum* infection did not seem to impair the productivity of the plants as frequent lateral buds were produced (not a characteristic morphological feature of the species) by the side of the infected buds. Further, this report confirms the possible transmission of *F. thapsinum* by Asiatic fungal weevil (*Fusocarnia napinatum*).

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