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Identification and *in vitro* control of pathogen causing leaf spot disease in *Centella asiatica* in Jaffna peninsula

A. Keshiga, P. T. J. Jashothan, T. C. Jeyaseelan and E. C. Jeyaseelan*

Department of Botany, Faculty of Science, University of Jaffna, Sri Lanka *christyjeyaseelan@gmail.com

Centella asiatica is an important medicinal herb that is widely used as a leafy vegetable in several Asian countries including Sri Lanka, Production of C. asiatica has been severely affected by leaf spot disease. The aim of the present study was to identify the pathogen causing leaf spot disease in C. asiatica in Jaffna peninsula and to test the effect of natural extracts on the pathogen. Diseased leaf samples were collected from commercial fields from Valikamam, Vadamarachchi and Thenmarachchi regions and three sites were selected from each region. Symptoms were characterized and pathogen was isolated on potato dextrose agar (PDA) medium following standard procedure. Culture and morphological characters were studied for each isolate. Koch's postulates were carried out to confirm the pathogenicity of the isolates; 10 µl spore suspension (70 spores/µL) was wound inoculated to C. asiatica. This assay was done under greenhouse conditions. Furthermore, extracts of dried root powder of Curcuma longa and fresh leaves of Lawsonia inermis, Azadirachta indica and Vitex negundo were prepared using water and tested for antifungal activity. Leaf lesions were irregular or angular, pale brown to greyish white in the center, and surrounded by dark purplish halo. The lesion had pycnidia, scattered, dark brown to rusty brown, globose and immersed in host tissue. In PDA medium, young cultures appeared white, with fluffy and aerial mycelia and mature cultures appeared to be whitish grey to black with pycnidia. Hyaline in young hyphae turned brown when mature, after two weeks. Chlamydospores were observed in culture. Conidia were formed in the pycnidia. The conidia were initially unicellular, hyaline, ellipsoid to sub-ovoid, turning to dark brown, bi-celled, thick walled and ellipsoid at maturity. Based on the morphological features the fungal isolates were identified as Lasiodiplodia sp. There was no variation among the isolates collected from different regions in Jaffna peninsula. Koch's postulates confirmed the pathogenicity of Lasiodiplodia sp. C. longa water extract (1000 mg/L) significantly reduced mycelial growth and spore germination in vitro. Association of Lasiodiplodia sp. with C. asiatica has not been reported earlier in Sri Lanka. Therefore, this is the first report of leaf spot disease caused by Lasiodiplodia sp. on C. asiatica in Sri Lanka. Further studies are being carried out to identify the pathogen based on molecular techniques.

Keywords: Centella asiatica, Jaffna, Lasiodiplodia, leaf spot