



Tropentag, October 5-7, 2011, Bonn

“Development on the margin”

Characterisation of Novel Bacteria of the Genus *Pseudomonas* from Dieback affected *Dalbergia sissoo* in Bangladesh

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Abstract

Dieback of sissoo (*Dalbergia sissoo* Roxb.) is a disastrous disease, which destroyed millions of trees in South Asia. This novel dieback disease is characterised by a combination of symptoms, beginning with wilting of leaves. In later stage leaves become necrotic and fall down as well as smaller twigs do, leading to increasing crown transparency. Loss of branches follows and the disease ends up in the final stage of stagheadedness, where the affected trees loose almost all parts of the canopy. Black spots with gummosis appear on the basis of the trunk and are found up to a height of three to five meters with progressing disease. Various biotic and abiotic factors have been discussed as cause of the dieback disease, but the causal agent(s) could not yet be identified unequivocally. Our recent studies are focused on the molecular detection and characterisation of bacteria of the genus *Pseudomonas*, which are found to be associated with dieback affected sissoo trees from various sites of Bangladesh, including the regions around Dhaka (Mirpur), Tangail, Sirajganj, and Bogra. The bacteria were detected in roots, bark, branches and leaves of dieback affected *Dalbergia sissoo* trees. Phylogenetic analyses based on sequences of 16S rDNA and single copy gene *gacA* revealed that the isolated bacteria represent strains of a yet unassigned species of *Pseudomonas*. To proof Koch's postulates, sissoo seedlings were inoculated via various techniques including leaf infiltration and submersing the roots of very young seedlings in suspensions of representative *Pseudomonas* isolates. The symptom development was analysed during a period of culture ranging from one week up to twelve months. To analyse whether the *Pseudomonas* isolates used for inoculation could be identified later in the inoculated plants, bacteria were re-isolated from these plants and characterised by sequencing.

Keywords: Bacteriosis, dieback, shisham, sissoo