

1. INTRODUCTION

Tea (Camellia sinensis (L.) O. Kuntze) forms the backbone of the economy of North-East India. Being a perennial the tea plant possibly interacts with, and samples more environmental (both physical and biological) problems than does any other plant. Grey blight caused by Pestalotiopsis theae (Sawada) Stey. is one of the foliar diseases of tea which is prevalent throughout the whole of North-East India. This disease is extremely common on old tea leaves that are about to fall and also on leaves of all ages. On old leaves it starts from any damage - a cut, a break or a bruise - on the leaf blade. The diseased patches appear light to dark brown in colour with a greyish centre on the upper surface, roughly circular to oval, marked with concentric zonations almost from the centre to the very edge (Plate I). Black pustules of the sporodochia are produced in concentric lines on the upper surface. These are slightly larger than what is seen in brown blight caused by Colletotrichum camelliae Mass. (= Glomerella cingulata (Stoneman) Spauld and Schrenk). The diseased patches may occur at the margin or in the middle of the lamina. On young leaves the patch is usually dark brown to almost black, rather irregular in shape and not marked with concentric rings. On older leaves the patches lead to the breaking away of the lamina into irregular pieces and in severe cases the fungus can considerably reduce the maintenance foliage leading to the debility of the bush and die back. Acervuli are found on lesions on leaves, irregularly distributed on both surfaces of the leaf but most frequently epiphyllous, and are globose, punctate, subepidermal, erumpent, dehiscence by irregular rupturing of epidermis by a pore.

The biochemical mechanisms responsible for containment of fungal pathogens in the resistant interactions are undoubtedly multifold. Available evidence also indicates that resistance to disease in many cases is the result of activation of more than one biochemical defense mechanism (Ebel and Grisebach, 1988). Since it is not practicable to consider all the probable factors associated with disease resistance of tea against P. theae, a few promising ones were



Plate I : Naturally infected tea leaves showing the symptoms of grey blight disease caused by Pestalotiopsis theae.

undertaken in the present investigation. There is often a greater increase in phenolic biosynthesis in resistant host species than in susceptible hosts (Sridhar and Ou, 1974; Purushothaman, 1974; Hammerschmidt and Nicholson, 1977; Vidyasekharan, 1988; Borkar and Verma, 1991; Chakraborty and Saha, 1994a) and it is sometimes postulated that the increase in phenolic compounds is part of the resistance mechanism. Some of these compounds are toxic to pathogenic and non pathogenic fungi and have been considered to play an important role in disease resistance (Mahadevan, 1991; Nicholson and Hammerschmidt, 1992). Polyphenols are major constituents of tea leaves and their involvement in the defense mechanism either as preformed or induced chemicals seems highly probable (Chakraborty et al., 1995a).

On the other hand the resistance or immunity of plants to a pathogen may also depend on the speed and extent of protein synthesis induced in the host by the pathogen (Uritani, 1976; Agrios, 1978). Alteration in protein synthesis in the plants can lead to the development of local resistance or immune layer around infection sites (Chakraborty and Purkayastha, 1987). In several plant species it has been demonstrated that upon infection with fungi, bacteria or viruses, the development of symptoms is accompanied by the appearance of one or more new proteins, designated as pathogenesis related proteins (Gianinazzi et al., 1980; Gessler and Kuc, 1982; Ahl and Gianinazzi, 1982; Parent and Asselin, 1984; Bryngelsson et al., 1988; Christ and Mosinger, 1989; Fischer et al., 1989; Bryngelsson and Green, 1989; Ye et al., 1990). In view of these findings it was considered worth while to investigate whether protein also changes in the resistant and susceptible varieties of tea, after infection with P. theae and if so, at what level ?

Microbes have tremendous potential significance in controlling plant diseases (Blakeman and Fokkema, 1982; Cook and Baker, 1983; Baker et al., 1985; Rytter et al., 1989; Inglis and Boland, 1990; Andrews, 1992; Fokkema, 1993; Chakraborty et al., 1994c). Two basic principles are followed for biocontrol of leaf diseases - (a)

manipulation of the natural phylloplane microflora to render it more unfavourable to pathogens, or (b) introduction of a known antagonist into given biocenosis. This involves both introduction of naturally occurring as well as the foreign antagonists. The chemicals diffusing from inside the leaf on its surface provide nutrition to phylloplane microflora. Nutrients on the phylloplane such as, carbohydrates, amino acids, organic acids, sugar, alcohols, minerals, trace elements, vitamins and hormones as well as antimicrobial compounds such as phenols, terpenoids which originate endogenously are important not only because of their direct role as microbial substrate but also because of their likely indirect effects on synthesis of antibiotics and siderophores on the phylloplane. The surface of aerial plant parts of tea provide an excellent opportunity to investigate its phylloplane microflora in order to select potential microorganism antagonistic to P. theae.

The basic objectives of the present investigation are (a) to select tea varieties susceptible and resistant to P. theae, and to study the cultural characteristics of the pathogen; (b) to estimate host-parasite proteins before and after infection and to analyse their protein pattern; (c) to determine the level of phenolic compounds in the leaves of resistant and susceptible varieties before and after infection with P. theae; (d) to ascertain the antifungal activity of phenolics associated with differential host response to infection; (e) to evaluate the changes in the levels of antifungal compounds in resistant and susceptible plant; (f) to determine the disease reaction elicited by the cell wall of P. theae, their antifungal activity and chemical characterization, (g) to isolate and identify the phylloplane microorganisms of tea from Doars and Hill regions of North Bengal; (h) to screen the potential microorganism(s) antagonistic to P. theae in vitro, (i) to evaluate the effect of selected antagonist(s) for controlling the incidence and development of grey blight disease of tea, (j) to partially purify the antifungal compound, if any, from the antagonistic microorganisms; (k) and to test the isolated compound in vivo and determine its efficacy.

Before going into the details of the present work, a brief review in conformity with this study has been presented in the following pages.