

# *Introduction*

Tea [*Camellia sinensis* (L.) O. Kuntze] is the most important hot beverage in the world today. It occupies a very important position in Indian agriculture being one of the major foreign exchange earners of the country. The world's best quality of tea is produced in Darjeeling. It is also grown in the slopes of Nilgiris and Annamalai hills of Peninsular South India and Bhramahputra Valley of Assam located 100m. above sea level. Being a perennial, the tea plant possibly interacts with, and samples more environmental problems than do most other plants. A number of fungal pathogens cause diseases of tea which reduces the quality and quantity of tea production. Sclerotial blight caused by *Sclerotium rolfsii* Sacc. (telomorph: *Athelia rolfsii* (Curzi) Tu and Kimbrough = *Corticium rolfsii* Curzi) is one of the fungal diseases which appears in the nursery grown tea seedlings. The fungus is a soil borne rotting pathogen of very aggressive nature and causes considerable damage of young tea seedlings in the nursery which is very common in the plains but rare in the hills.

*S. rolfsii* affects the lower stems and roots of tea seedlings at or near the soil line. During infection whitish mycelial growth of the fungus can be seen at the junction of the branch with the stem close to the soil level, which is the most favoured point of attack. A dark brown lesion on a tea seedling's stem near the soil line is a very early indicator. With time, the disease progresses and a white mycelial web spreads over the soil and the basal canopy of the plant, followed by the appearance of sclerotia of mustard seed size on the infected areas. In its advanced stage infection becomes prominent in the root system and subsequently the entire shoot withers and falls and finally the plants die. Seedling death usually occurs rapidly. The fungus can remain in the soil as mycelium, living as saprophytes on diverse organic material and grows actively only in moist soil at moderate to high temperature (25-35<sup>0</sup>C). Light brown to yellow, round sclerotia may remain viable for many years. *S. rolfsii* occurs in diverse soils, has a very wide host range and world wide distribution (Punja, 1985). It infects at least 500 species in about 100 plant families. Diseases caused by *S. rolfsii* are especially rampant where temperatures are sufficiently high to permit the growth and survival of the fungus.

Plants respond to infection by pathogens in a number of ways which are triggered by the initial recognition phenomenon. Successful establishment of the

pathogens in host depends upon some kind of molecular similarity between two partners (De Vay and Adler, 1976). However, only certain key common antigens are important in host parasite compatibility (Chakraborty and Purkayastha, 1983; Chakraborty *et al.*, 1995a; 1997a; 2002d). It has been observed that with increased antigenic disparity, the response of host may prevent further activity of the parasite (Chakraborty, 1988). Disease detection by immunological means is gaining ground in case of fungal diseases (Chakraborty and Chakraborty, 2003; Gawande *et al.*, 2006). Though significant advances have been made in the development of rapid and sensitive assays for fungi in recent years, commercially available techniques are limited to a few pathogens and diseases. Such detection techniques make it possible to detect microquantities of the pathogen within a few hours of infection, which is much more advantageous than the conventional techniques involving pathogen inoculation, visible symptoms and microscopy. On the other hand detecting spores of plant pathogens in air samples can be used predictively to forecast the likelihood of important transmission events (Kennedy *et al.*, 1999; 2000). Detection and quantification of pathogen inoculum has considerable applicability in diagnosis and management of existing and emerging plant diseases within agricultural crop production systems (Kennedy and Wakeham, 2006). These have tremendous potential for plant disease control measures since detection of a pathogen at the initial stage of infection can lead to formulation of control measures before much damage has been done.

The art and science of plant disease control has moved in the direction of biological control of plant pathogens, including use of introduced antagonists. It is now widely recognized that biological control of plant pathogen is a distinct possibility for the future and can be successfully exploited in modern agriculture, especially within the framework of integrated disease management systems. Integrated control is a flexible, multi-dimensional approach to disease control utilizing a range of control components such as biological, cultural and chemical strategies needed to hold diseases below damaging economic threshold without damaging the agro-ecosystem.

The interaction between plants and their pathogens is complex and may be very specific to a given combination of the plant and the fungus. The biochemical

mechanisms responsible for containment of fungal pathogens in the resistant interactions are undoubtedly multifold. Many biochemical changes occur in plants after infection, and some of these have been associated with the expression of defenses that are activated after infection (Chakraborty, 2005). Available evidence also indicates that resistance to disease in many cases is the result of activation of more than one biochemical defense mechanisms (Bera and Purkayastha, 1999; Hammerschmidt, 1999; Chakraborty *et al.*, 2004). It has long been recognized that responses are characterized by the early accumulation of phenolic compounds at the infection site and that limited development of the pathogen occurs as a result of rapid (hypersensitive) cell death. The responses include the formation of lignin, the accumulation of cell-wall appositions such as papillae (Prats *et al.*, 2005), and the early accumulation of phenols (Chakraborty *et al.*, 1995b) within host cell walls. Since it is not practicable to consider all the probable factors associated with disease resistance of tea against *S. rolfsii*, a few promising ones have been undertaken in the present investigation.

The basic objectives of the present investigation are (a) screening of tea varieties for resistance to *S. rolfsii*; (b) estimation of host parasite proteins before and after infection; (c) raising of polyclonal antibodies against antigens of *S. rolfsii* and tea roots; (d) detection of serological cross reactivity between *S. rolfsii* and tea varieties using immunoassays and cellular location following immunofluorescence; (e) detection of *S. rolfsii* in artificially inoculated tea roots by ELISA, dot immunobinding assay and indirect immunofluorescence; (f) determination of the level of phenolics in tea roots following infection by *S. rolfsii*; (g) ascertaining the antifungal activity of phenolics associated with differential host response to infection; (h) assay of phenylalanine ammonia lyase, peroxidase and polyphenol oxidase activities in tea roots following inoculation with *S. rolfsii*; (i) *in vitro* studies of *S. rolfsii* with biocontrol agents, plant extracts and systemic fungicides and (j) developing effective integrated management strategies against sclerotial blight disease of tea.

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