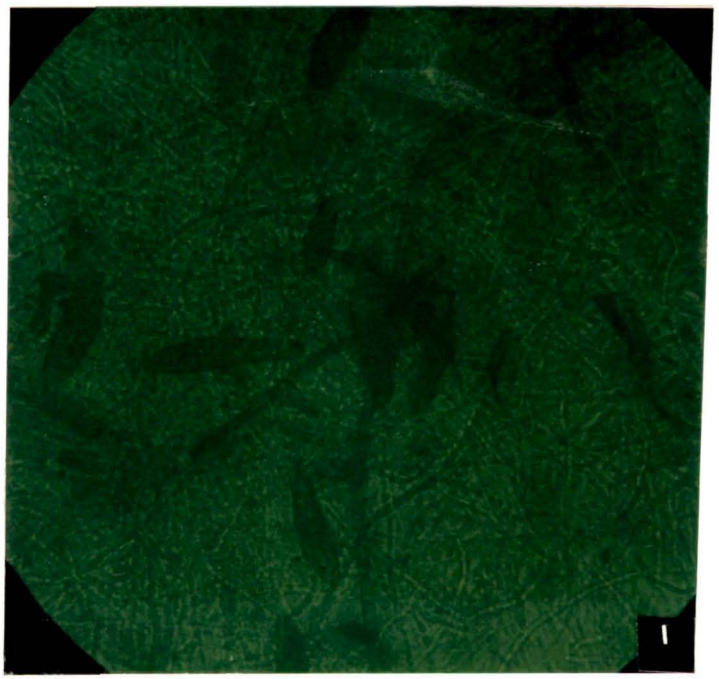
B. carbonum. Similarly antigen of untreated and nickel chloride treated tea leaves (TV-18) and B. carbonum (BC1) were cross reacted with antiserum of TV-18. In the homologous reactions TV-18 exhibited 6 precipitin arcs. Nickel chloride treated leaves of TV-18 shared only 4 of the 6 antigenic constituents of untreated leaf.

Part XI : Studies on the tissue and cellular location of major cross reactive antigens shared by Camellia sinensis and Bipolaris carbonum

Fluorescent antibody labelling with fluorescein isothiocyanate (FITC) is known to be one of the powerful techniques to determine the cell or tissue location of major cross reactive antigen shared by plant host and parasite. In the present study, immunodiffusion and immunoelectrophoretic tests as well as indirect enzyme linked immunosorbent assay clearly indicated the presence of major cross reactive antigenic substance (CRA) common to C. sinensis and B. carbonum. It was decided to determine the tissue and cellolar location of CRA in tea leaves.

Antibodies indirectly labelled with fluorescein isothiocyanate (FITC) were used to determine the location of CRA in sections of tea leaves (TV-18) and fungal cells (B.carbonum).

PLATE - XIII (figs. 1 & 2) Conidia (x60) and mycelia of Bipolaris carbonum (BC1) treated with antiserum to tea leaf (TV-18) and FITC-antibodies of goat specific for rabbit globulins. fig. 1- photographed under phase - contrast fig. 2-same field photographed under UV-fluorescent conditions for comparison of treatments.



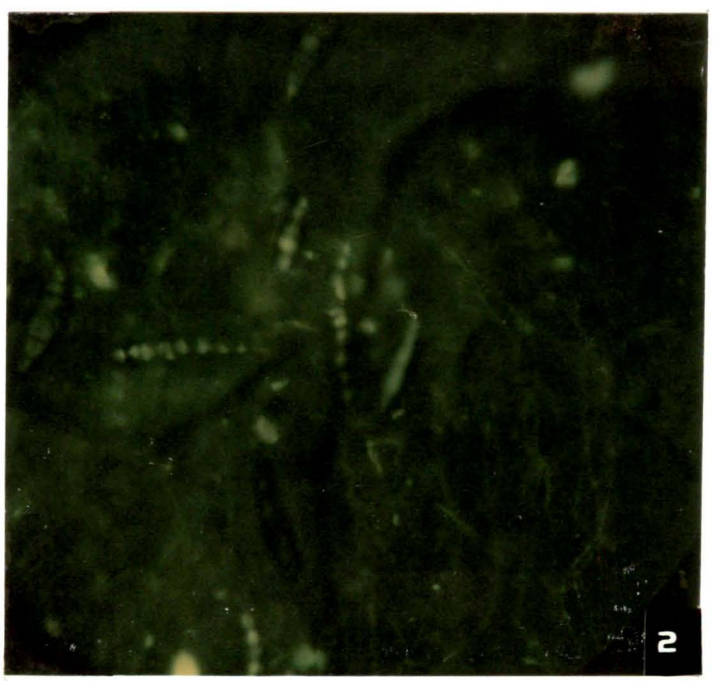
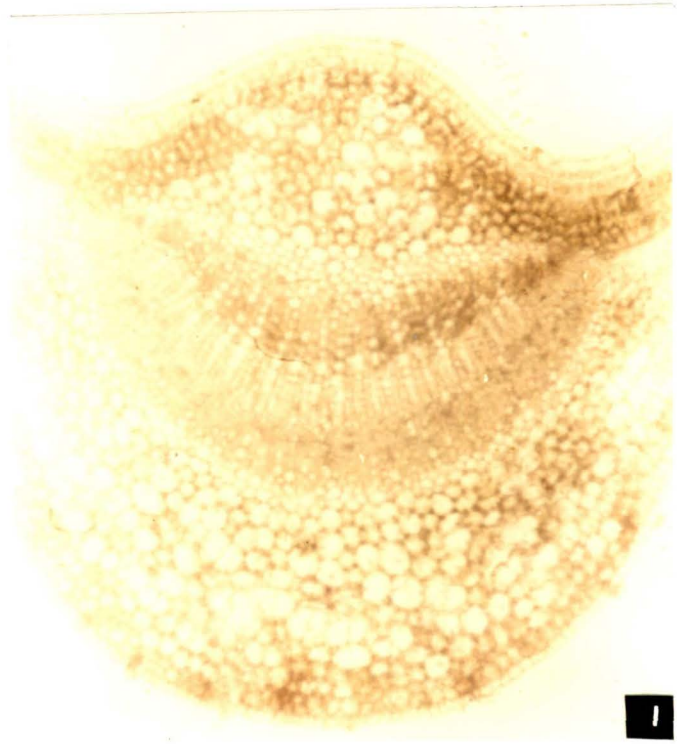


PLATE -XIII

PLATE - XIV (figs. 1 & 2). Fluorescent FITC- antibody staining of tea leaf tissues (TV-18) for cross-reactive antigen shared with Bipolaris carbonum (BC1). fig. 1 - unstained cross section of leaf ; fig. 2-autofluorescence of unstained leaf section.



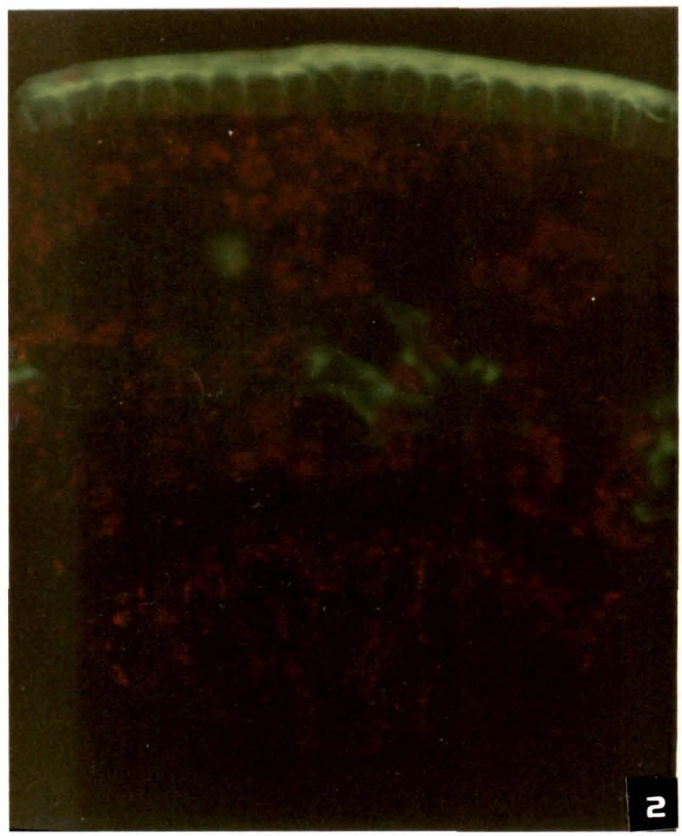
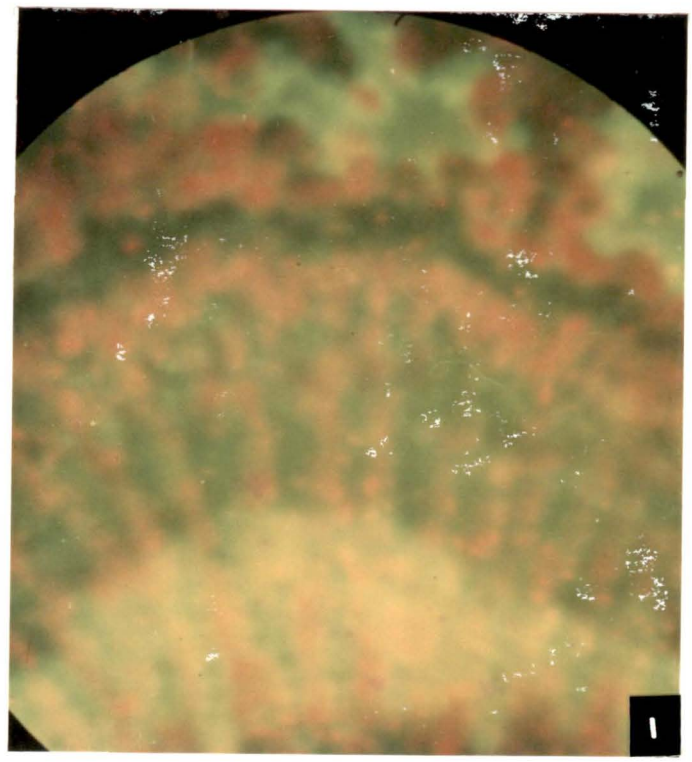


PLATE -XIV

PLATE - XV (figs. 1 & 2). Fluorescent FITC- antibody staining of tea leaf tissues (TV-18) for cross-reactive antigen shared with Bipolaris carbonum (BC1). figs. 1 & 2-leaf sections treated with antiserum to tea leaf (TV-18) and FITC-antibodies of goat specific for rabbit globulins.



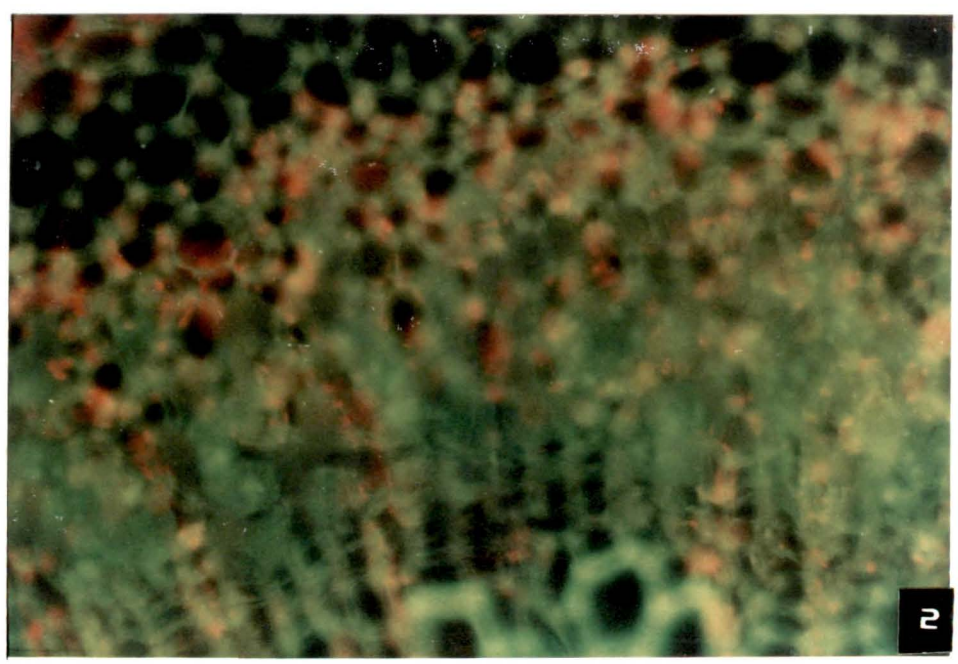
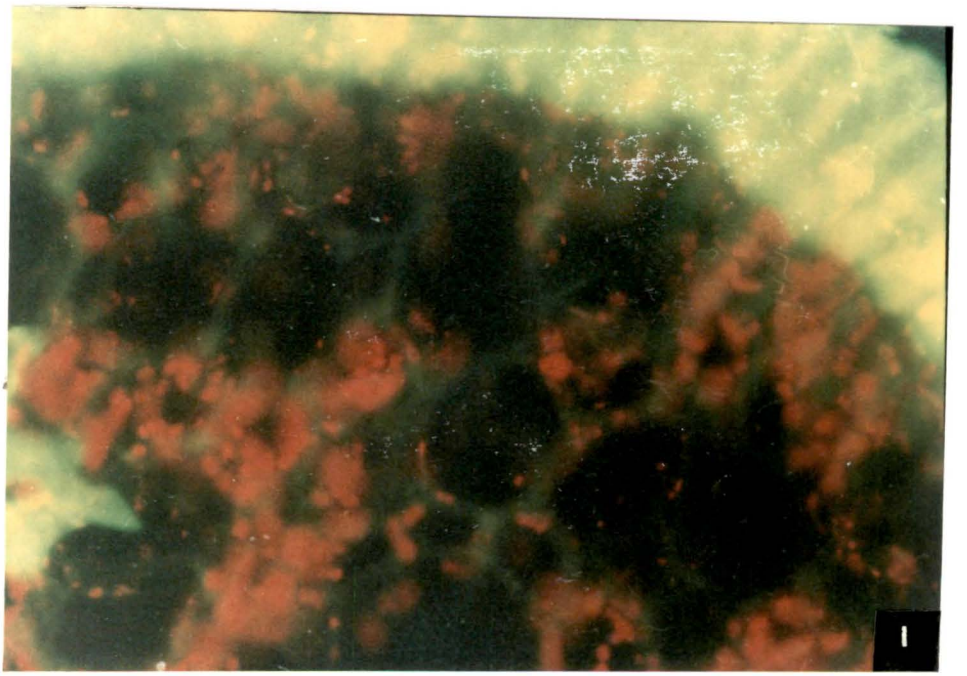


PLATE - XV

PLATE - XVI (figs. 1 & 2) Fluorescent FITC- antibody staining of tea leaf tissues (TV-18) for cross-reactive antigen shared with Bipolaris carbonum (BC1). figs. 1 & 2-leaf sections treated with antiserum to B. carbonum and FITC - antibodies of goat specific for rabbit globulins.



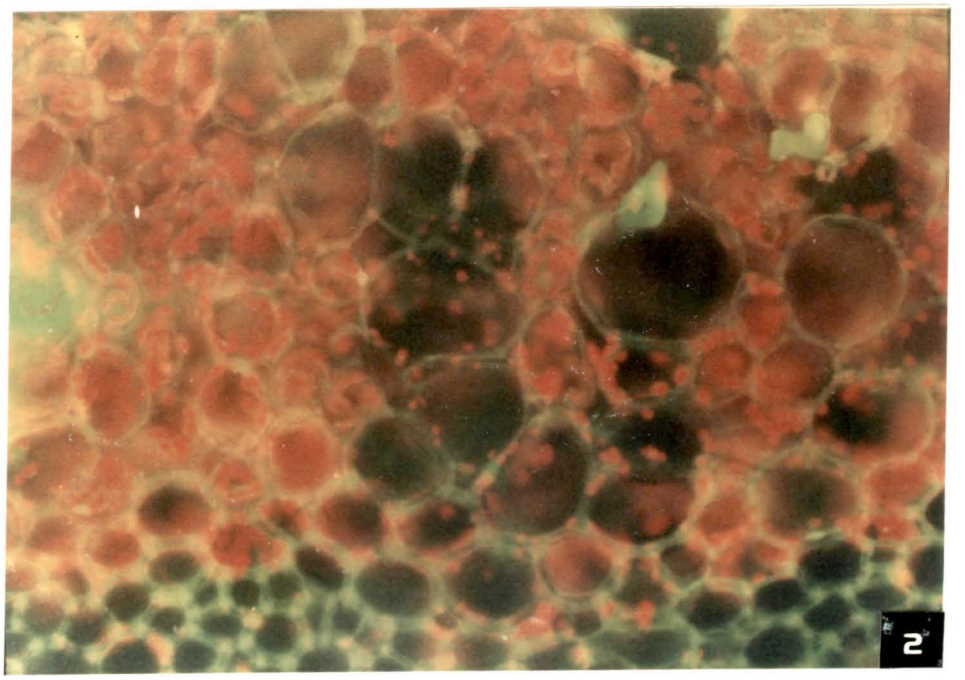


PLATE - XVI

Details of antibody staining process have been described under materials and methods. Leaf sections and mycelial preparations were photographed under both phase contrast and UV-fluorescent condition for comparison of treatment. These are presented in Plate XIII - XVI.

Conidia and mycelia of B. carbonum were not autofluorescent nor did they fluoresce when treated with normal serum followed by FITC. Treatment of mycelia and conidia of B. carbonum with homologous antiserum and FITC showed a general fluorescence that was more intense on hyphal tips and the patch like areas of the conidia. When fungal cells were reacted with antiserum of tea leaf (TV-18) and FITC, bright fluorescence in patch like areas was evident on some conidia and hyphal tips. (Plate XIII, fig. 2). Fresh cross sections of leaves (TV-18) through midrib were cut and treated with homologous and heterologous antisera, then reacted with FITC. Leaf sections exhibited a natural autofluorescence on the cuticle and sclereids present in the mesophyll tissue (Plate XIV, fig. 2). Same observations were noted when the leaf sections were treated with the normal serum and FITC. Leaf sections treated with homologous antisera (T18A) and then reacted with FITC developed bright fluorescence which was distributed throughout the leaf tissue-mainly epidermal cells, mesophyll tissue and xylem elements (Plate XV, figs. 1 & 2). Of much significance was the strong reaction of

antiserum of B. carbonum with leaf sections. CRA was concentrated mainly around epidermal cells (Plate XVI, fig. 1). Similarly, but with less fluorescence, mesophyll tissue also reacted with antiserum of B. carbonum (Plate XVI, fig. 2).