1) *Entamoeba histolytica* cells were grown en masse in axenic and monoxenic culture media. The harvested amebae were pooled and subsequently used for preparing antigen extracts.

2) Hyperimmune antiserum was obtained by immunizing the rabbits with axenic and monoxenic culture preparations along with an equal volume of Freund's complete adjuvant.

3) A large number of human sera samples obtained from patients of clinically proven cases of invasive amebiasis were also included in the several tests carried out for studying the antigen-antibody interactions.

4) Characterisation of *Entamoeba histolytica* antigen was carried out by means of column chromatographic separations in Sephadex G-200 gel matrix. It was shown that *Entamoeba* has a multiple antigen system comprising of several species of proteins having different molecular weights. The isolated antigen component eluting as peak 1 consisted of a high molecular weight protein and behaved as a potent antigen in various antigen-antibody reactions. The heterogeneity of the eluted antigen components was further checked by testing the purity of the isolated fractions in
polyacrylamide gel (PAGE) electrophoresis. The optimum antigen dilution subsequently used in various reactions was determined by taking into consideration the kinetics of quantitative precipitin titration reactions.

5) Antiamoebic antibodies belonging to different classes of immunoglobulins were isolated and characterised by means of ion-exchange column chromatography and antigen-antibody interactions. The isolated immunoglobulin fractions possessing antibody activity were further identified against monospecific anti-immunoglobulin sera by means of radial immunodiffusion tests and immunoelectrophoretic analyses. Most of the antiamoebic antibody activity was found mainly confined to IgG fraction. Some minor antibody activity was also detectable in IgM fractions, whereas none was detected in immunoglobulin fraction IgA. Similar results were also obtained from immunofluorescent studies of the immune serum from experimental animals and human subjects.

6) Sequentially appearing antiamoebic antibodies were also identified in primary and secondary response sera samples obtained from immunizing rabbits. The sequence of appearance of antiamoebic antibody activity, as confined to different classes of immunoglobulins, vis-a-vis time of antigenic stimulation was studied in radial
immunodiffusion tests carried out against monospecific immunoglobulin antisera. During the initial phases of antigenic stimulation, the antibody activity was predominantly confined to IgM immunoglobulin. In secondary response sera samples, the antibody activity was mostly linked with gamma globulin G. No antibody activity from immunized sera samples was detectable in IgA immunoglobulin. On the basis of the above studies, it would not be a mere speculation to assume that IgM class of antibody activity is more closely related to an infection of recent origin.

7) Characterisation of humoral immune responses in invasive amebiasis was carried out by detecting the presence of circulating antiamebic antibodies. A large number of serologic reactions were employed for identifying the specific antiamebic antibodies in whole serum and as well as in the isolated immune serum fractions. The reactivity of the isolated immunoglobulins in various serologic test procedures was further studied. The role of isolated immunoglobulins in the formation of antigen–antibody complex in various serologic tests was also investigated.

8) Characterisation of cellular immunity with special reference to the appearance of protective antibodies was carried out in experimental animals models. It was found that amebic antigens, in combination with nonspecific stimulators of
cellular immunity, are capable of eliciting a protective immune response. Such immunized animals were found to possess a high grade immunity capable of protecting the animals against the lethal challenging doses of the causative organisms. The appearance of typical pathology, or lack of it, was studied in histopathological tissue preparations.

9) The presence of cell mediated immunity in amebic infections was further investigated by detecting the appearance of a delayed type of hypersensitivity reaction in sensitized guinea pigs. A delayed type skin hypersensitivity reaction was readable in sensitized animals in about 60 - 70 hr after the administration of the challenging doses.

10) In vitro growth inhibition of amebas was obtained by growing the parasites in the presence of hyperimmune serum and the isolated fractions thereof. About 30 per cent concentration of the immune serum, or the equivalent gamma globulin, was found to inhibit the amebic growth in vitro. The cytotoxic effect of the immune serum in the presence of complement was found to materially alter the typical characteristics of ameba growth curve.