

Biochemical and immunological studies on soluble antigens of *Entamoeba histolytica*

Cited in:

Parasitol Kcs (1993) 79:365- 371

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Received: 6 November 1993 Accepted: 17 February 1993

Abstract.

The soluble antigens of *Entamoeba histolytica* trophozoites were analysed in detail by biochemical and immunochemical methods. The antigen was highly complex and heterogeneous as revealed by Sepharose S-300 column chromatographs, which showed four distinct fractions. The molecular mass of fractions FI, FII, FIII and FIV was 660, 170, 65 and 13 kDa, respectively. Protein was the major constituent in crude soluble antigen (CSA) and fractions FI and FII (67%, 80% and 90%, respectively). Polysaccharide was predominant in the FIII fraction (59%). Antigenic activity observed after different physico-chemical treatments revealed that CSA and FI antigens were predominantly glycoprotein in nature. However, the antigenicity of FIII antigen was greatly reduced after sodium metaperiodate treatment, whereas no alteration in reactivity was discerned after trypsin treatment. Sodium dodecyl sulphate-polyacrylamide gel electrophoresis (SDS-PAGE) analysis demonstrated nearly 28 Coomassie blue bands for CSA and 20, 16, 15 and 3 polypeptide bands for the FI, FII, FIII and FIV fractions, respectively. The molecular mass of the polypeptides of these bands ranged from 210 to 20 kDa. Antigenic activity was observed in CSA and in the first three fractions, both in counter immunoelectrophoresis (CIEP) and in enzyme-linked immunosorbent assay (ELISA). However, the highest antigenic activity was noted in fraction FI. Major immunoreactive polypeptides of CSA and FI antigens against whole trophozoite antibody were observed in the 10- to 170-kDa regions. However, major differences in the immunoreactivity of the two antigens were noted at 116 and 14 kDa for FI antigen and at 34, 30 and 20 kDa for CSA. The binding of the FI antibody to the surface of the live parasite and the loss of immunoreactive polypeptides from the FI antibody after its adsorption with live trophozoites of *E. histolytica* suggest a correspondence between FI and surface antigens of *E. histolytica*. The efficacy of active immunization with CSA and its different fractions showed that antigenicity and immunogenicity were closely associated with the high-molecular-weight proteins, as 91% protection was observed for the FI antigens, whereas the FII and FIII fractions and CSA provided only 41%, 33% and 41% protection, respectively. These data suggest that FI antigen can be used in the serodiagnosis of and immunoprophylaxis against amoebiasis.

Reference

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