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SUPEECHA KUMKATE: DEVELOPMENT OF *FASCIOLA GIGANTICA*
(COBBOLD, 1856) IN MICE AND CHARACTERIZATION OF A MONOCLONAL
ANTIBODY AGAINST ADULT WORM ANTIGEN. THESIS ADVISORS:
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The antigens of *Fasciola gigantica* were characterized by SDS-PAGE. Surface tegumental antigens contained many protein bands ranging from molecular weight 116 kD to less than 6 kD and antigens from crude extract of adult worms contained protein bands ranging from 97 kD to less than 6 kD. The excretory-secretory antigens showed the intense band at molecular weight 27kD, while metacercarial antigens were composed of proteins ranging from molecular weights 97 kD to less than 14 kD.

In this study, the hybridoma clones, namely 1C12-1 and 1C12-2, produced monoclonal antibodies, which had been found to be IgM, κ -light chain were characterized. These monoclonal antibodies were reactive by immunoelectrotransfer blot analysis with the single antigenic molecule of surface tegumental antigens at molecular weight 57 kD and molecules of metacercarial antigens at molecular weights 57 kD and 35 kD . Anatomical localization of antigenic molecules on frozen sections of metacercariae by immunoperoxidase assay using Avidin-Biotin and immunofluorescence methods indicated that the target molecules detected by monoclonal antibodies were on the surface membrane and the underlying narrow cytoplasmic zone of the tegumental layer underneath the innermost cyst wall layer of the metacercaria.

Swiss albino mice were infected with metacercariae to study the susceptibility to *Fasciola gigantica* infection. The fluke recovery rates varied between 0-10% and the average size of juvenile fluke increased from 0.14mm in width and 0.25mm in length at the seventh day to 1.42mm in width and 3.90 mm in length at the fiftieth day of infection. Antibody responses demonstrated as antibody titer against surface tegumental antigens by indirect ELISA did not correlate with the fluke recovery. However, electroimmunotransfer blot analysis revealed that antibodies were progressively produced against surface tegumental antigens, antigens from crude extract of adult worm and excretory-secretory antigens during the course of infection.

The development of internal organs and surface topography of juvenile flukes recovered from mice were studied by light microscopy and scanning electron microscopy. The juvenile flukes gradually developed as their bodies became elongated, testes and ceaca showed branching and progressive outgrowths. In contrast, ovaries and uteri remained obscure. The prominent features of surface topography of juveniles were the presence of single-pointed spines and numerous microvilli. The surface areas between spines appeared as ridges, which may develop, from the coalescence of microvilli. The division of spine tips to become serrated with two to three points was first observed on the juvenile fluke recovered on the fiftieth day of infection.