IN VITRO ANTIBODY-DEPENDENT CELL MEDIATED MECHANISMS

IN

KENYAN BADOON (PAPIO ANUBIS)

TOWARDS

SCHISTOSOMULA OF SCHISTOSOMA MANSONI

by

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ABSTRACT

The nature of cellular damage to antibody coated, skin transformed schistosomules was examined *in vitro* using two methods to assess damage i.e. ^{51}Chromium release from labelled schistosomulae or uptake of vital dye.

First a whole buffy coat preparation from the peripheral blood had to be purified into its mononuclear, neutrophil and eosinophil components. This was done by centrifugation over stepwise density gradients of ficoll-hypaque or metrizamide to yield highly enriched cell preparations. Eosinophils were also purified by removal of phagocytic cells from a cell preparation devoid of its mononuclear components. Results indicate that only neutrophils are capable of causing marked ^{51}Chromium release from labelled schistosomula; eosinophils cause much lower Chromium release while mononuclear cells have no effect. Eosinophils did however cause release of Chromium from antibody-coated chicken red blood cells indicating their functional viability after separation. It must, however, be pointed out that cells were obtained from *Schistsoma mansoni* or *Trichinella spiralis* eosinophilia induced animals after yields of adequate numbers of eosinophils from normal subjects proved futile. In any case treatment of whole buffy coat with anti-neutrophil serum abolished activity against schistosomula. Microscopy observation of vital dye uptake shows both neutrophils and eosinophils causing damage to schistosomula but only neutrophils elicit a significantly higher killing than an unpurified leucocyte preparation. Attempts to stimulate cells with ECF-A did not enhance killing. From these observations it

would appear that the neutrophil is the more potent cell in damaging schistosomula *in vitro*.

The onset of cytotoxic antibody as assessed by isotope release is at 6 weeks after a primary infection and peaks at 9 weeks dropping to background levels within a year in the case of the eosinophils, but remaining steadily high in relation to neutrophils for the duration of the two years of study. Damage by neutrophils, eosinophils or unpurified cells as assessed microscopically, shows a different pattern, with linear increase in cytotoxic antibody with duration of infection.

The second part of the investigation dealt with the possible role of immune complexes in blocking cytotoxic capacity of the effector cells via the Fc receptors.

First, as expected, neutrophils were found to bind more baboon aggregated IgG than either eosinophils or mononuclear cell preparations. Aggregated IgG would also block whole buffy coat preparation, in a dose dependent manner, from causing Chromium release from labelled organisms. Likewise exogeneously prepared immune complexes of purified adult worm antigens and immune serum would block the cytotoxicity capacity of peripheral blood leucocytes. Serum from infected animals, and known to contain immune complexes, failed to prevent damage, when pre-incubated with effector cells, to either antibody-coated schistosomules or chicken red blood cells. Peripheral blood cells luecocytes from animals exposed to *S. mansoni*

or T. spiralis continued to damage schistosomules during the period when immune complexes would be expected in circulation, and even after drug chemotherapy.

It would appear then that in an active infection immune complexes have no adverse effect in limiting the cytotoxic capacity of peripheral blood leucocytes.

These findings suggest that the baboon is a suitable animal model of the disease. Whereas both eosinophils and neutrophils from man and baboon can cause damage of schistosomules in vitro the much higher number of circulating neutrophils in both species favour the neutrophil as the effector cell of more significance in vivo. This is augmented by the higher cytotoxity of baboon neutrophils in vitro.