
The tegument of the rat tapeworm Hymenolepis diminuta acts as an absorption interface facilitating chemical exchange between the cestode and its external environment. Several enzymic markers have been identified in tegumental functions of H. diminuta. One of these markers Ca"-dependent ATP'ase is sensitive to the calmodulin antagonist trifluoperazine (Hipkiss et al, 1987a).

Using immuno electron microscopy we have identified a number of calmodulin/calmodulin binding proteins distributed at various subcellular sites (Eastlake et al, 1994). Treatment of H. diminuta with trifluoperazine has potential cestocidal properties influencing both tegumental protein release, activity of calmodulin-dependent enzymes and the uptake of glucose (Branford White & Hipkiss, 1986). In this study we report that treatment in vivo of H. diminuta infected rats with trifluoperazine influences the distribution of tegument membrane associated enzymes.

Subcellular fractions were obtained using differential centrifugation. The majority of alkaline phosphatase and 5' nucleotidase were recovered in the tegument structure. Type I phosphodiesterase, a recognised tegument membrane enzyme (Gamble & Pappas, 1981) was located exclusively in this fraction. Acid and alkaline Ca"- ATP'ases were enriched in the tegument and no activity was located in the cytosol. Based on this evidence, the above enzymes were taken as being reliable markers for the tegument.

Zonal centrifugation of the prepared tegument fraction from control showed that alkaline phosphatase and type I phosphodiesterase as distinct unimodal peaks at a modal density with a major peak at 1.16g cm'3; a minor peak at 1.12g cm'3 and some activity remained in the sample layer. Bi-modal pattern was exhibited by 5' nucleotidase with major and minor peaks at 1.17g cm'3 and 1.12g cm'3 respectively. Differences in the density distribution of alkaline Ca" ATP'ase gave a dimodal distribution of 1.12 and 1.18g cm'3 whereas the acid form was unimodal at 1.18g cm'3. The sharpest peaks of activity are coincident with the densities of plasma membrane markers.

Tegument isolated from H. diminuta recovered from rats treated with trifluoperazine (134mg/kg body weight per day) was also subjected to isopycnic zonal centrifugation. Alkaline phosphatase and type I phosphodiesterase showed unimodal distribution with median densities 1.19 and 1.21g cm'3 respectively. Both these densities had increased compared to samples from untreated cestodes, 5' nucleotidase exhibited bi-modal distributions with activity peaks of 1.14 and 1.22g cm'3. Both forms of Ca"- ATP'ase showed skewed density distribution with over 60% of the enzymes remaining in the starting layer.

Glucose uptake by the cestode was monitored using a glucose oxidase system (Hipkiss & Branford White, 1986). Even at this high drug concentration, glucose transport was 30% active compared to untreated H. diminuta suggesting that some respiration properties are retained by the cestodes.

From the density distribution it would seem that trifluoperazine causes selective loss of protein from the tegument especially in domains enriched with ATP'ase and 5' nucleotidase as there was a shift to the lighter density region in the sucrose gradient. We have previously shown that incubation of H. diminuta in culture results in destruction of the tegumental protein. Gross changes in ultrastructural appearance and cellular organisation also occur (Hipkiss et al, 1987b). These observations concurred with earlier work where high levels of soluble protein and lactate dehydrogenase were released into the media following incubation with the calmodulin antagonist. Under the conditions used here no morphological changes were evident although some subtle changes to tegument enzymes and the cestodes ability to utilise glucose was observed. Using calmodulin as a probe we are currently investigating the links between trifluoperazine and the glucose transport system.

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