filaments and that its smooth ridges must be due to distension by a higher hydrostatic pressure on one side of the filter.

Unfortunately the observed filter parameters may not be the same as those of the unfixed micro-filter. First of all the house and feeding filter will shrink to some degree as soon as the muscular activity of the animal is stopped. Secondly, the fixation and especially the acetone dehydration will cause further shinkage. Although the overall size of the house may be reduced drastically after drying, it is uncertain to what degree the micro-filter filaments shrink. Thirdly, the evaporation of a conductive layer on the specimen will increase the apparent thickness of the filaments and thereby reduce the pore size.

In spite of these factors, I find it reasonable to believe that the micro-filter traps particles down to about 0.1 μm in diameter. The size distribution of the particles seen behind the filter (figure 3) speaks in favour of this view. Compared with the size-range of bacteria and other well defined nanoplankters, this is an exceedingly low limit. It may accordingly be supposed that smaller living and dead particles are an important food source of *O. dioica*.

The observed micro-filter constitutes a continous layer throughout the feeding filter. A second layer, making up the opposite side of the ridges or channels, may also be present, but has not been disclosed so far. It seems reasonable to believe that this micro-filter is essential for the particle-trapping function of the feeding filter and that water passes across the meshwork[6]. The small rectangular meshes and the smooth surface, combined with the large open area fraction, will delay clogging of this micro-filter as particles are unlikely to enter the pores without being able to penetrate them. When the animal stops its tail movements, as it does every few sec, and continues to suck particle-enriched water from the feeding filter, the water current through the micro-filter may also be reversed and the particles lifted away from the meshwork.

A water passage exclusively along the filter ridges in socalled 'Reusengänge' or 'Reusenbahnen' and a particle-trapping mechanism based on numerous trabeculae within these tunnels[3] will render the micro-filter functionless and give a very small filtering surface and rapid clogging of the filter. Likewise, the presence of any sticky material in the feeding filter[5] would prevent particles from being sucked into the mouth of the animal and cause immediate clogging of the filter.

By continued scanning and transmission electron microscopic studies, I hope to reveal how the beautifully spaced meshwork of the micro-filter is produced by cellular secretion and to disclose its relation to the other components of the expanded feeding filter.

A similar, but much coarser silk micro-filter has recently been described in a larval philopotamidae (Trichoptera)[8].

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Presence of ATPase on the vesicular membrane of *Cysticercus cellulosae*. A high resolution cytochemical study

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Summary. ATPase was demonstrated by high resolution cytochemistry in the microtriches of *C. cellulosae*. It is thought that the enzyme is important for the parasite's acquisition of raw materials for surviving and distribution in host tissues.

Porcine cysticercosis is produced by *Cysticercus cellulosae*. In the human, cysticercosis occurs when man accidentally becomes the intermediate host of the cestode *Taenia solium*.

C. cellulosae is preferentially localized in the brain of man, whereas in the hog it is usually lodged in skeletal muscle[2]. Although the reasons for such a preferential distribution are not yet known, it is evident that the adaptation of *C. cellulosae* to different microenvironments which offer distinct metabolic recourses, must require the possession, by the parasite, of a carefully and strictly regulated transport system. Therefore the study of the presence and distribution of those enzymes associated with transport mechanisms might shed some light on the explanation for this particular body tissue distribution[3]. Many studies have provided evidence for the close association of a specific (Na, K, Mg) activated ATPase with active cation transport[4–6]. In some systems this active cation transport may also be linked to the transport of nonelectrolytes[7–10]. Therefore ATPase might be highly significant to this particular distribution of the parasite. The purpose of the present study was to localize the cytochemically demonstrable ATPase activity of *C. cellulosae*'s vesicular membrane at ultrastructural level, and relate it to the possible role in the preferential tissue distribution of this parasite.

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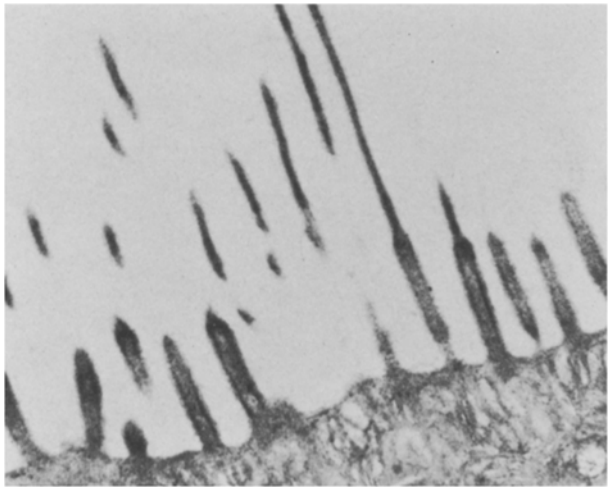

Fig. 1. Vesicular membrane of *C. cellulosae*. The tegument discloses numerous microtriches. Section stained with uranyl acetate and lead citrate. ×25,000.

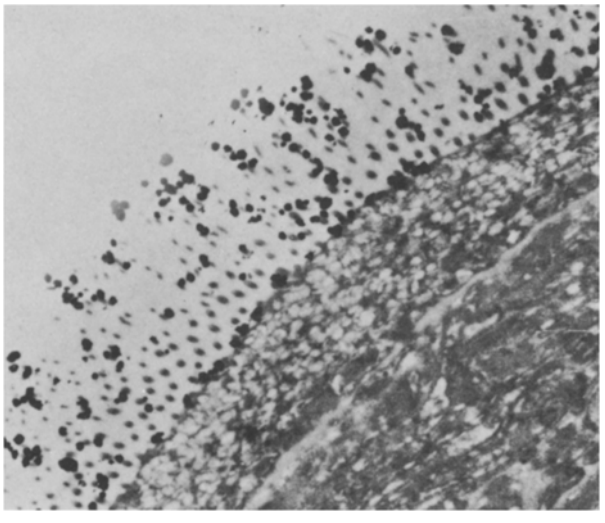

Fig. 2. The picture discloses positive ATPase reaction as evidenced by numerous electron dense deposits corresponding to lead precipitates confined to the external surface of the vesicular membrane mainly around the microtriches. Unstained section. ×11,500.

Fig. 3. Vesicular membrane of a cysticercus fixed in glutaraldehyde prior to the cytochemical reaction. No electron dense deposits are present. Unstained section. ×10,000.

Material and methods. A total of 180 cysticerci were excized from the anconeous muscle of 6 parasitized hogs slaughtered in the abattoir of Mexico City. In order to preserve the integrity of the parasites, we refrained from any drastic treatment and only 2 gently washings in 40 mM Tris-maleate buffer (pH 7.0) were carried out to clean them out from surrounding material. In order to demonstrate the Mg-Na-K-stimulated ATPase activity, the method of Wachstein and Meisell was used as modified by Marchesi and Palade[11]. The reaction was performed at 37 °C for 15 min either with unfixed or prefixed cysticerci (20–30 min in a mixture of 2.5% glutaraldehyde in 0.1 M cacodylate buffer, pH 7.4). Control experiments were carried out with the following modifications of the incubation media: a) Containing ADP, AMP, -glucose-6-phosphate or Na_2HPO_4 instead of ATP; b) containing ATP, but with added 1 mM ouabain and c) without ATP. After incubation, specimens were fixed in 3% glutaraldehyde, cacodylate buffered to pH 7.4, and washed thoroughly with the same buffer, dehydrated in ascending concentrations of ethanol and embedded in Araldite as mentioned elsewhere[12]. Sections in the silver colour range cut on a Porter-Blum MT-2 ultramicrotome were collected on copper grids and observed in a Philips EM-300 electron microscope.

Results. The cysticercus' tegument free surface, as is the case of the tegument of other tapeworms[3], is endowed with numerous slender microvilli called microtriches[13] readily seen with scanning and transmission electron microscopy[14] (figure 1). Electron-dense deposits of lead phosphate along the tegument of *C. cellulosae* were seen and interpreted as evidence that this external surface was a site of ATPase activity. Precipitates appeared as dense irregularly distributed granules confined exclusively to microtriches (figure 2). Preparations made after the glutaraldehyde-prefixation described above were generally free from any lead phosphate deposit (figure 3). Controls in which ATP was absent or replaced by Glu-6-P, as substrate, disclosed no electron dense deposits. Ouabain containing samples revealed no lead phosphate precipitates; nor were there any deposits formed when Na_2HPO_4 was added to the incubation medium instead of ATP. Considerable amount of precipitates were found at the external surface in preparations containing ADP instead of ATP; however, the localization of the reaction products was patchy along the outer limiting membrane. A similar pattern of precipitates but in minor quantities was observed when AMP was added instead of ATP.

Discussion. Following the report of Rosenthal et al.[15], many doubts have been raised concerning the validity of lead methods for the histochemical demonstration of plasma membrane-bound ATPase activity[11,16–18]. However, reliability in the W-M method for demonstration of ATPase seems to be successfully maintained

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both by changing the concentrations of lead and substrate[17,19] and by applying this method to unfixed sections[20]. Thus, at present the histochemical validity of the ATPase reaction has been recovered. Therefore our results provide evidence for the presence of ATPase activity in the exposed surface of the vesicular membrane of *C. cellulosae*. This is in keeping with data obtained by Lumsden in the outer surface of tapeworm tegument[21]. Since all postembryonic stages of these parasites lack a digestive tract, the assimilation of material from the environment must take place across the tegument. In this case, those enzyme systems which are concerned with the transport of substances, or with the modification of solutes to which parasites are otherwise impermeable, may be quite important to the survival or specific distribution of the parasite in the host tissues. The demonstration, therefore, of ATPase in *C. cellulosae* may be indicative of the parasite unzymatic dependance for its nutrition and selective tissue dwelling. If it is so, the inhibition of ATPase activity would hamper the survival of the parasite. Recent observation by Sengupta on the presence of a mobility-inhibitory factor which acts as a potent ATPase inhibitor opens the possibility of a search for this factor in various tissue fluids[22]. Theoretically the physiological neutralization or absence of this factor in skeletal muscle and brain tissue fluids would explain such preferential distribution of *C. cellulosae*. Further studies are needed to support this theory.

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Studies on protein composition and synthesis in the ovary of *Rhynchosciara americana* (Diptera, Sciaridae)[1]

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Summary. A comparative study of electrophoretic protein patterns from ovary and haemolymph was undertaken. The synthetic activity of the ovaries was also studied by measuring protein precursors incorporation. Based on these analysis, the origin of vitellogenic proteins is considered.

Vitellogenic proteins have been extensively studied in many insects with respect to their identification and isolation[2], sites of synthesis[3,4], and hormonal regulation[5]. There seems to be 2 mechanisms involved in yolk formation: the synthesis by the ovary itself and the incorporation of proteins, synthesized in other tissues, from the haemolymph.

A study was undertaken on the synthetic activity of the ovaries of *Rhynchosciara americana*, during pupal and adult stages, when yolk deposition occurs. Ovary follicles of *R. angelae* (*R. americana*)[6] have 1 oocyte and only 1 nurse cell, enveloped by a layer of follicular cells[7]. The ovary grows rapidly from the middle stage of pupa, when typical polytene chromosomes develop in nurse cells.

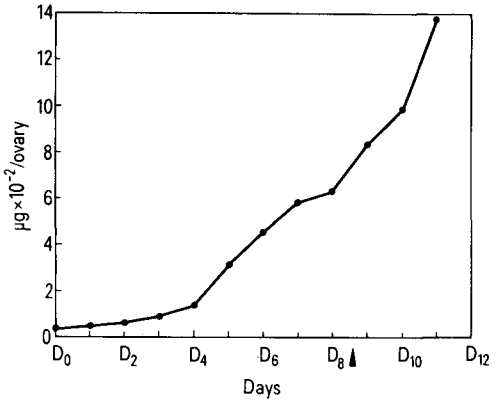

Fig. 1. Determinations of protein content in the ovaries of *R. americana* during pupal and adult stages, using crystalline bovine serum albumin as standard. Each point represents an average of 3 determinations. D_0 corresponds to the newly ecdysed pupae. Arrow indicates adult eclosion.

From this stage up to the adult, a large number of yolk granules can be seen in the oocyte cytoplasm[7]. A fertilized female lays a group of eggs which develop synchronously, producing individuals of the same sex[8].

Quantitative determinations of proteins have been made in order to verify the correlation between protein accumulation and ovarian growth. Following dissection, the ovaries were transferred to 1 ml of a NaOH 1 N solution, at 45 °C for 1 h, and a sample of 0.2 ml from this solution was used for the dosage of soluble proteins, according to Lowry et al.[9]. The spectrophotometric analysis showed an increase of protein concentration at about the 5th day of the pupal stage, which intesifies towards the end of the adult life (figure 1). The ovarian growth observed from mid pupa to adult stage[7] must, therefore, involve a remarkable accumulation of proteins, probably related to vitellogenesis.

The comparative analysis of protein patterns, revealed by polyacrylamide gel electrophoresis[10], showed a re-

1 Supported by Fundação de Amparo à Pesquisa do Estado de São Paulo (FAPESP) and Conselho Nacional de Pesquisas (CNPq, PIG-SIP/04-001). We are indebted to Dr J. M. Amabis for his continuous support and interest in the work, and to Dr L. C. G. Simões for critical reading of the manuscript.
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