

GIARDIA MURIS AND GIARDIA DUODENALIS GROUPS: ULTRASTRUCTURAL DIFFERENCES BETWEEN THE TROPHOZOITES

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SUMMARY

Trophozoites of the *Giardia muris* group from hamsters, domestic rats and mice and of the *Giardia duodenalis* group from hamsters and domestic rats were examined by transmission electron microscopy. The basic ultrastructure of the trophozoites was similar. Differences were shown in the morphology of the ventro-lateral flange of the trophozoites of *Giardia muris* and *Giardia duodenalis* groups. Marginal plates are less developed in the species of the *Giardia duodenalis* group. In this group, the distal extremity of the lateral flange is short and thick and the marginal plate does not penetrate into the distal extremity of the flange. In the *Giardia muris* group, the ventro-lateral flange is well developed and narrow and the marginal plate penetrates the distal extremity of the flange. The osmiophilic lamella, which accompanies the dorsal surface of the marginal plate is seen only in the *Giardia muris* group.

KEY WORDS: *Giardia muris*; *Giardia duodenalis*; Trophozoite, Ultrastructure.

INTRODUCTION

Based on the median body morphology and the trophozoite's size and shape one can differentiate *Giardia muris* and *Giardia duodenalis* by light microscopy⁴. The data in the literature about such differentiation by electron microscopy are scarce. NEMANIC et al.¹² were unable to show differences between trophozoites of *Giardia muris* from mice and those of *Giardia lamblia* from man.

Taking in account these results and since the morphology of *Giardia* from hamsters and domestic rats had not been described yet, we decided to study comparatively the ultrastructure of *Giardia muris* and *Giardia duodenalis* trophozoites found in hamsters, domestic rats and mice.

MATERIAL AND METHODS

Giardia species from naturally infected animals were previously identified by light microscopy. Smears of faeces or mucosa were fixed with Schaudinn's solution and stained with Heidenhain's haematoxylin. The species of *Giardia* were identified by morphological features and trophozoite length and width measurements, at 1.000 x magnification.

Trophozoites of the *Giardia muris* group from hamsters, domestic rats (*Rattus rattus rattus*) and mice and *Giardia duodenalis* group from hamsters and domestic rats were used. Fragments of the host's mucosa were fixed in 2.5% glutaraldehyde buffered at pH 7.3 with

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0.1M phosphate buffer, post-fixed in 1% osmic tetroxide in the same buffer and embedded in Araldite. The ultrathin sections were stained with uranyl acetate and lead citrate and examined with an EM 301 Philips electron microscope.

RESULTS

The basic ultrastructure of the trophozoites of *Giardia muris* group from hamsters, domestic rats and mice and *Giardia duodenalis* group from hamsters and domestic rats is similar. The trophozoites have a microtubular adhesive disk surrounded by an adjacent cytoplasmic flange known as the ventro-lateral flange and four pairs of flagella (Fig. 1) whose basal bodies are located between the two nuclei. Osmiophilic material is seen next to the axonemes of the flagella and vacuoles are present in the dorsal region of the trophozoite. Scattered in the cytoplasm, we have noticed cytoplasmic clefts without being enveloped by membranes, with a little dense homogeneous content (Fig. 1), glycogen and free ribosomes. Two parallel rows of microtubules (funis) accompany the intracytoplasmic track of the caudal flagella. The median body is formed by microtubules wi-

thout apparent relationship to other structures. Mitochondria, endoplasmic reticulum and Golgi apparatus are not noticed.

We have not observed differences between *Giardia duodenalis* group from hamsters and domestic rats or among *Giardia muris* group from hamsters, domestic rats and mice. Differences were, however, evidenced in the morphology of the ventro-lateral flange of trophozoites of *Giardia muris* and *Giardia duodenalis* groups.

In the two groups studied the flange is arranged peripherally to the striated disk presenting a cytoplasmic lip supported by marginal plates which are associated with one of the axonemes of the antero-lateral flagella.

In the *Giardia duodenalis* group both from hamsters (Fig. 2) and from domestic rats (Fig. 3), the cytoplasmic lip that makes up the ventro-lateral flange is short and thick, and does not show electron dense material in its distal extremity. The plates that support the flange are striated, little developed and are just associated with the plasmic membrane of the ventral surface of the ventro-lateral flange. In this group, the osmiophilic lamella is not observed.



Fig. 1 — *Giardia muris* trophozoite from hamster. General aspect. Cross section showing vacuoles (V), cytoplasmic clefts (CC), striated disk (SD) and ventro-lateral flange (F). Axonemes of the flagella (A). X 15,500.

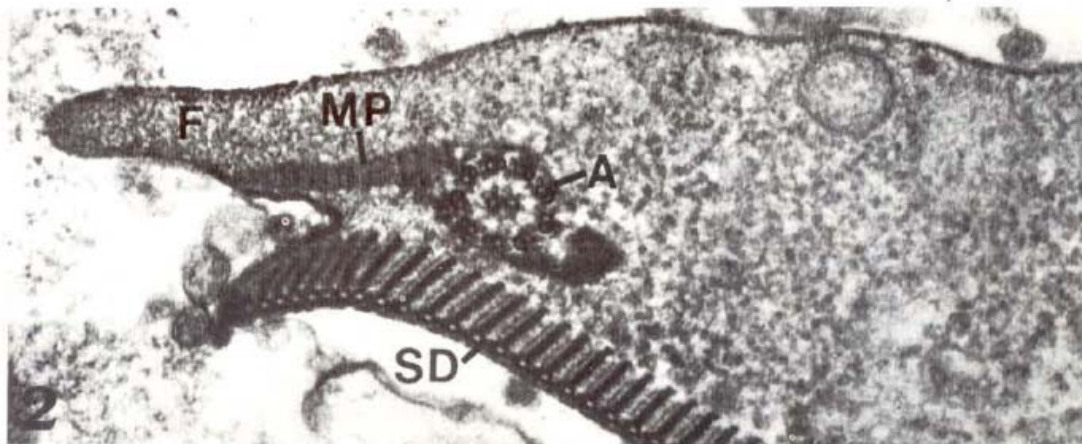


Fig. 2 — *Giardia duodenalis* trophozoite from hamster in cross-section. The ventro-lateral flange (F) is short and thick. The marginal plate (MP) is poorly developed, does not penetrate its extremity and is not accompanied by an electron-dense lamella. The axoneme (A) of the anterolateral flagella with adjacent osmiophilic material and the striated disk (SD) are shown. X 72,000.



Fig. 3 — *Giardia duodenalis* trophozoite from domestic rat in tangential section. The ventro-lateral flange (F) and its marginal plate (MP) show the same morphological aspects as *G. duodenalis* trophozoites from the hamster. X 65,000.

In *Giardia muris* group from hamsters (Fig. 4), domestic rats (Fig. 5) and mice (Fig. 6) the distal extremity of the cytoplasmic lip is narrow and developed, and it is darkened by dense cytoplasm. The marginal plates seem to occupy the whole flange extension. They extend from the dorso-lateral surface of the striated disk into the ventro-lateral flange being

associated with the flange plasmic membrane either dorsally or ventrally. These marginal plates show striations and penetrate through the flange and become lost in the dense material of their distal extremity. A dense lamella of osmiophilic material that accompanies the

dorsal surface of the plate is noticed associated with the marginal plates.

DISCUSSION

Giardia muris and *Giardia duodenalis* groups from hamsters and domestic rats, described for the first time in this paper, show a basic ultrastructural similarities to the other species already described in the literature, such as *Giardia muris* from mice^{2,5,6,7,14}; *Giardia lamblia* from man^{1,10,15}; *Giardia duodenalis* from rabbits³ and *Giardia canis* from dogs¹³.

Our results show that *Giardia* belonging to the same group (*duodenalis* or *muris* groups) do not present ultrastructural differences when located in different animals (hamsters-domestic rats and hamsters-domestic rats-mice, respectively).

However, *Giardia* belonging to different

groups (*muris* and *duodenalis*) from the same host (hamster or domestic rat) show marked differences in the ventro-lateral flange ultrastructure regarding their size, arrangement of the marginal plates and presence of osmiophilic lamella.

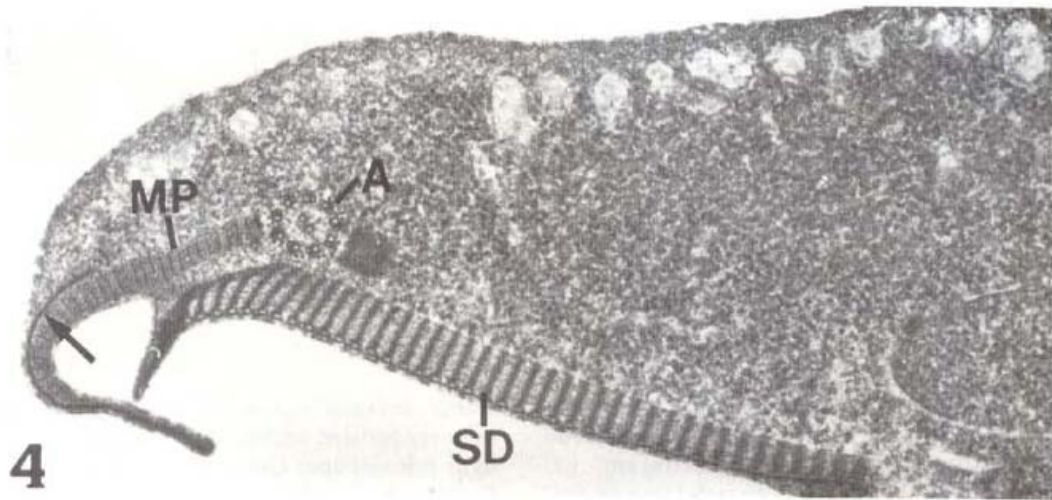


Fig. 4 — *Giardia muris* trophozoite from the hamster in cross-section. The ventro-lateral flange is long and narrow and the marginal plate (MP) penetrates through the distal extremity of the flange. A lamella of electron-dense material (arrow) accompanies the dorsal surface of the marginal plate. The striated disk (SD) and the axoneme of antero-lateral flagella (A) with adjacent osmiophilic material area shown. X 46,500.

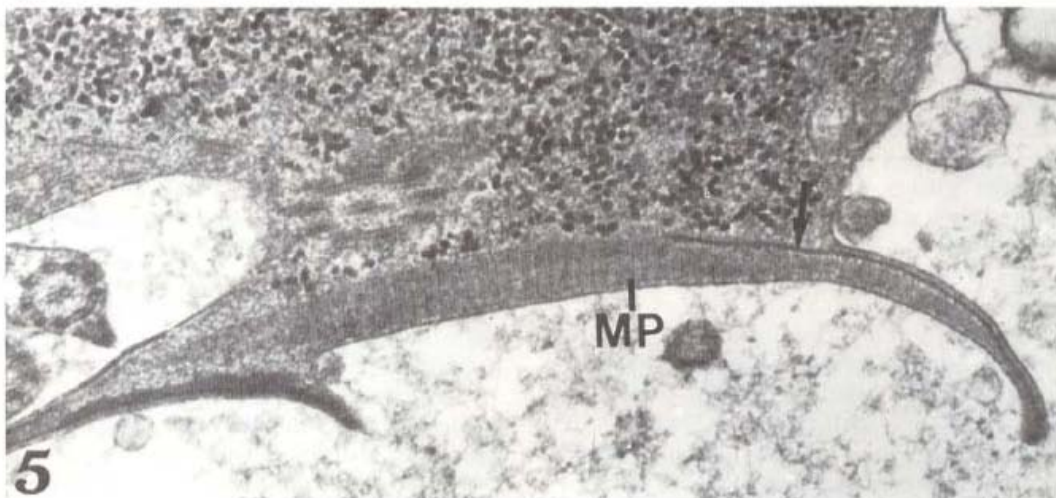


Fig. 5 — *Giardia muris* trophozoites from domestic rat in cross-section. The ultrastructure of the ventro-lateral flange (F), marginal plate (MP) and the osmiophilic lamella (arrow) is similar to those from *Giardia muris* from the hamster and mouse. X 46,200.

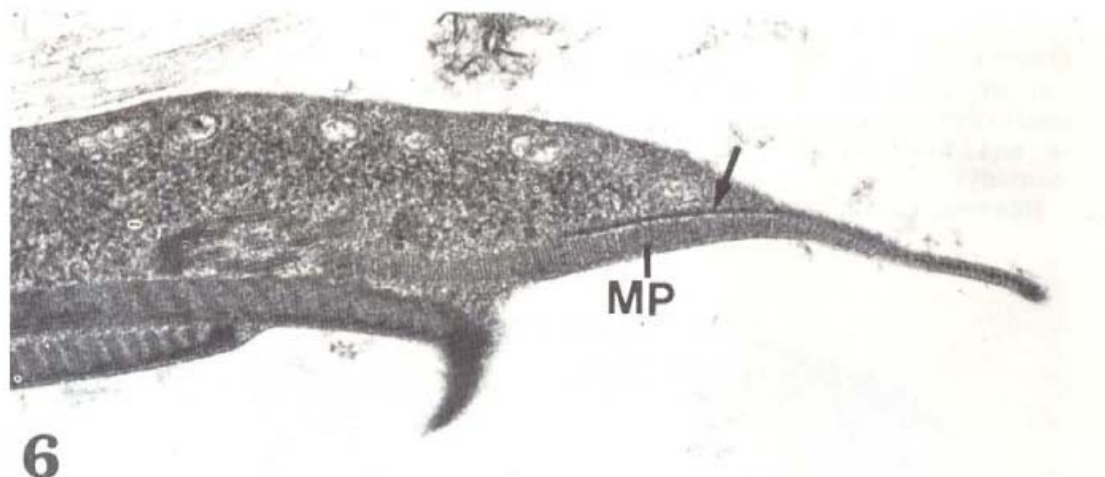


Fig. 6 — *Giardia muris* trophozoites from the mouse in cross-section, showing the ventro-lateral flange (F), marginal plate (MP) and osmiophilic lamella (arrow). X 72,000.

According to HOLBERTON⁷, it seems that the form adopted by the ventro-lateral flange may act on the attachment or detachment of the host to the substrate as the magnitude of the suction force caused by the striated disk depends on the caliber of the lateral channels between the ventro-lateral flange and the substrate. Therefore, the differences found between *muris* and *duodenalis* groups in relation to the ventro-lateral flange suggest that there is some difference in the attachment of these species to the substrate. The fact that the flange is less developed in *Giardia duodenalis* leads us to believe that it is not probable to have the formation of lateral channels in this group. This would lead the species of *Giardia duodenalis* group to attach weakly to the substrate. Such a fact could explain the lack of any mirror-image attachment of the striated disk to the intestine in cases of human biopsies¹¹.

Previous publications did not provide evidence of ultrastructural differences between *Giardia muris* and *Giardia lamblia* trophozoites¹², although KULDA & NORÝNKOVÁ⁸ in a review about *Giardia* ultrastructure, have suggested that the marginal plates in *Giardia intestinalis* are less developed than in *Giardia muris*.

It is well known that the taxonomy of the *Giardia* species is still somewhat controversial.

Three morphological different groups have been recognized among the species described by light microscope: *Giardia agilis* group which is found in amphibians, *Giardia muris* and *Giardia duodenalis* groups, which are found in mammals, each having a number of species⁹. FILICE⁴ defined these groups as species but according to KULDA & NORÝNKOVÁ⁸ they apparently represent higher tax.

The ultrastructural differences between *Giardia muris* and *Giardia duodenalis* groups and the similarities within each group of *Giardia*, shown in this paper, confirm this general tendency that point to the recognition of two morphological groups of *Giardia* in mammals. Although we do not know if these ultrastructural differences have some significance at the level of species, we can use the morphological feature of the ventro-lateral flange as an auxiliary morphological character to help a differential diagnosis among groups of *Giardia* in favorable sections examined with electron microscope, even if the host is infected by more than one type of *Giardia*.

The axenic culture of *Giardia duodenalis* group obtained in laboratory have permitted the use of other methods besides those morphological features to study differences or similarities among isolates of *Giardia* from different animal species. However, these results have not yet been used to systematize *Giardia* species⁹.

RESUMO

Giardia muris e *Giardia duodenalis*: diferenças ultraestruturais entre trofozoitos

Neste trabalho foi analisada, através de Microscopia Eletrônica de Transmissão, a morfologia de trofozoitos de *Giardia muris* obtidos de hamsters, ratos peridomiciliares e camundongos, bem como de trofozoitos de *Giardia duodenalis* obtidos de hamsters e ratos peridomiciliares. Basicamente a ultraestrutura dos trofozoitos estudados foi semelhante. Foram observadas, no entanto, diferenças na morfologia da franja ventro-lateral dos trofozoitos de *Giardia muris* quando comparados com os de *Giardia duodenalis*, diferenças estas relacionadas ao tamanho, arranjo das lâminas marginais e presença de lamela osmiofílica.

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