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Histological Studies of the Schistosomulum of *Andschistosoma mansoni* and *Andschistosoma margreboweie* in the Lung of Mouse

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Abstract: Histological studies on the *Schistosoma mansoni* and *Schistosoma margreboweie* development in the lung of definitive host mouse have been reported after 2 to 21 days post-infection (dpi). During the lung phase of development shape of schistosomula were relatively thin, elongated and round. Histologically, the schistosomulum was characterized by an outer cuticle layer. The layer was variable in appearance because of very thin layers of underlying smooth muscle fibers and a homogenous layer with occasional ridge areas. The thin and wide regions of the body of schistosomulum was characterized by undifferentiated and differentiated cells with oval or irregular shaped nuclei rich in chromatin and little cytoplasm. Other structures included development of gut caeca with presence of natural or black pigment. Moderate type of infiltration of cells including a large numbers of eosinophils, some macrophages and a few lymphocytes were seen around *S. margrebowiei* but no such reaction was observed around *S. mansoni* schistosomula after 8 dpi. An appearance of plasma cell was also noticed close to the surface of *S. mansoni* schistosomula.

Key words: Histology, schistosomulum, *Schistosoma mansoni*, *Schistosoma margreboweie*, lung mouse

INTRODUCTION

Schistosomula leaving the skin via blood or lymphatic vessels will ultimately pass through the right side of the heart via the venous system and will be distributed to the lungs via the pulmonary arterial system. The increase in numbers of schistosomula within pulmonary capillaries from day 2 to 7 after exposure indicate their effectiveness as a physical barrier to migration[1]. In the venous system, schistosomula are carried to the lungs within 3 to 4 days and reach their highest concentration at 4 to 6 days[2]. Following arrival in the lungs of mice, schistosomula undergo a phase of elongation, which is not accompanied by an increase in mass[3]. This process of elongation may be a necessary prelude to the passage of schistosomula along the lumina of both pulmonary and systemic capillaries[1]. This process of elongation may be necessary prelude to the passage of a schistosomulum through the narrow lumina of capillaries in the lung[4]. The changes in the tegument of *S. mansoni* during development from the cercariae to the adult stages. The presence of homogenous, discoid and membranous bodies in the tegument of 7 days old schistosomula and also reported the change in outer trilaminate tegumental membrane into a heptalaminate structure. They conclude that the membrane originated from multilaminate membrane-bound vacuoles arising from the subtegumentry cells[5] .

Several workers Smith *et al.*,[6]; Ghandour and Baibker,[7] have studies *in vivo* the development of other species of schistosomes. The developing schistosomulum of *S. mansoni* showed marked changes in body form during their migration from skin to the hepatic portal system[3]. The present study describes the histological changes in the schistosomula of *S. mansoni* and *S. marqrebowiei* development in the lung of mouse.

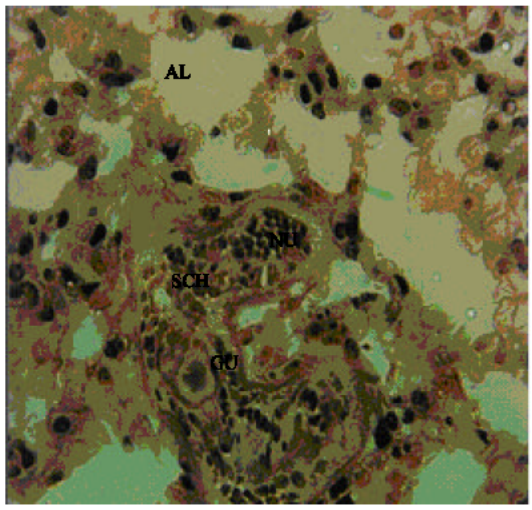
MATERIALS AND METHODS

Aged matched female mice of the Bantim and Kingman Tyler's Original (BKTO) strain, weighed approximately 20-35 g each were infected with 200 cercariae of either *S. mansoni* (Puerto Rican strain maintained in albino *Biomphalaria qlabrata* snails and random-bred. To mice following the methods of Taylor *et al.*,[8] or *S. margrebowiei* (originally obtained from Lochinrar National Park, Zambia) and maintained in *Bulinus natalensis* intermediate host snails (the original stock was obtained from the Experimental Taxonomy Unit of the British Museum of Natural History, London). Before administering the cercariae, the experimental animals were anaesthetized with Sodium pentobarbitone (Nembutal) and the abdominal hairs were clipped. The cercariae were applied to the abdominal skin by using ring. All mice were killed at 2, 3, 4, 6, 8, 10, 16 and 21 dpi and autopsies were performed immediately after the animals were killed by dislocation of neck region. The lungs from each mouse were fixed in Heidenhain's Susa fixative, washed, dehydrated ethanol, infiltrated and embedded in historesin[9]. The animals and plastic

embedded blocks of lungs of mice were provided by Dr. N.W. Runham, School of Biological Sciences, University of North Wales Bangor Gwynedd LL57 2UW United Kingdom. Selected 4 μm thick sections were stained in haematoxylin and eosin method. The sections were interpreted on Ernst Leitz Light Microscope (Model No. 786554).

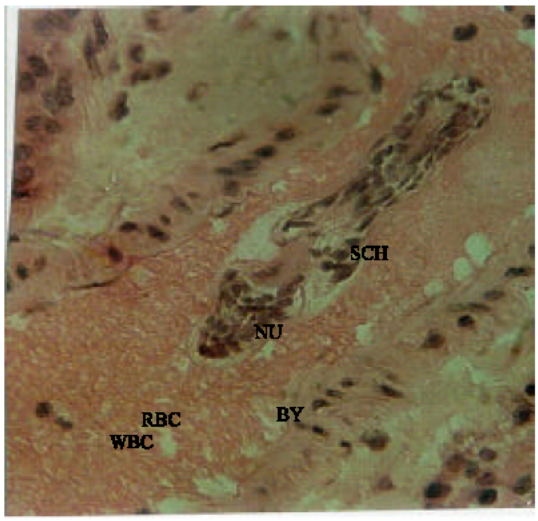
RESULTS

Histology of the lung schistosomulum: During the lung phase of migration (2-21 dpi) schistosomula were relatively thin, elongated and rounded. The schistosomulum was characterized by an outer cuticle layer. Which was variable in appearance because of very thin layers of underlying smooth muscle fibers and a homogenous layer with occasional ridge areas in both parasites. In *S. margrebowiei* larvae PAS-positive small granules were observed within the cuticle on day 16 but these were absent in *S. mansoni*. The body of the schistosomula was characterized by undifferentiated and differentiated cells with oval or irregular shaped nuclei rich in chromatin and little cytoplasm on 2 and 3 dpi (Fig. 1). Other structures included thin and wide regions of the body, musculature and development of gut caecae. The gut was characterized by a thin wall with nuclei and the presence of natural or black pigment from 4 to 10 dpi in both parasite infections (Figs 2 and 3). On day 7 irregular *S. mansoni* and elongated *S. margrebowiei* schistosomula consisted of an increased number of the nuclei and musculature of body. After 8 dpi the body of the parasite showed vacant spaces, some close to each other and a few pyknotic nuclei. Similar structures were also observed in schistosomula of *S. mansoni* on 10 dpi. Due to the small amount of the lung available for sectioning on 10 dpi no observation could be made on *S. margrebowiei*. Round *S. mansoni* and *S. margrebowie* schistosomula were characterized by variable shaped nuclei with uniformly distribute chromatin and some vacant space on day 16. A few of these types were of *S. mansoni* and *S. margreboweie* schistosomula were observed on day 21 inside the branches of the pulmonary veins and capillaries of the lungs of mouse. Due to the presence of schistosomula in the capillaries and blood vessels causing obstruction and rupturing the lumen. Moderate type of infiltration of cells including a large numbers of eosinophils, some macrophages and a few lymphocytes were seen around *S. margrebowiei* but no such reaction was observed around *S. mansoni* schistosomula after 8 dpi. An appearance of plasma cell was also noticed close to the surface of *S. mansoni* schistosomula.



25 μm

Fig. 1: Histology of the schistosomulum after 3 dpishowing prominent gut with black pigments and differentiated nuclei in the body



25 μm

Fig. 2: Histology of the schistosomulum after 4 dpi showing thin and wide region with differentiated nuclei, vacant spaces and musculature

DISCUSSION

In present study increase in number of *S. mansoni* schistosomula was recorded within the pulmonary capillaries of the lung on 6 dpi. But the results reported by Wheater and Wilson,[1] indicate that the increase in numbers of schistosomula within pulmonary capillaries

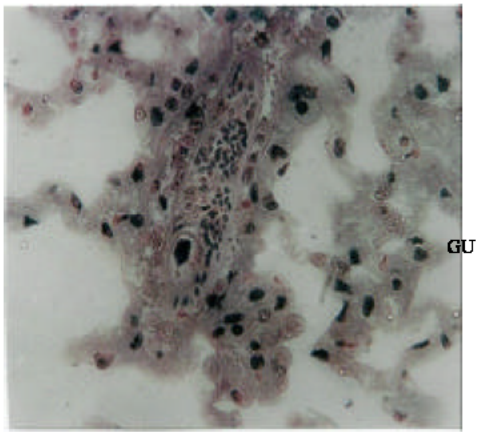


Fig. 3: Histology of the schistosomulum after 7 dpi showing dark pigments in the gut and well differentiated nuclei in the body

from day 2 to 7. Miller and Wilson,[2] have reported that in the venous system, schistosomula are carried to the lungs within 3 to 4 days and reach their highest concentration at 4 to 6 days. This study is nearly agreement with present findings. In the present study thin and wide regions of body of the schistosomulum was elongated and round in shape. Whereas, Wilson *et al.*,[3] have reported that following arrival in the lungs of mice, schistosomula undergo a phase of elongation, which is not accompanied by an increase in mass. This study is nearly agreement with results reported in the present study.

An appearance of plasma cell was also noticed close to the surface of *S. mansoni* schistosomula in present study. The results reported by Bruce *et al.*,[10] are agreement with the present study. Which showed that due to the presence of intravascular organism, free within the lumen of the blood vessel and surrounded by plasma cells and circulating blood cells. In the present study moderate type of infiltration of cells including a large numbers of eosinophils, some macrophages and a few lymphocytes were seen around *S. margrebowiei* but no such reaction was observed around *S. mansoni* schistosomula after 8 dpi. The results reported by Tizes *et al.*,[11] that though living worms produce little reaction, dead worms cause a toxic necrotic reaction with congestion of lung tissue and eosinophils, polymorphonuclear and histiocytes infiltration. Bently *et al.*, have reported that the schistosomula are relatively large organisms for intracellular migartion which caused considerable trauma and destruction of the tissues. These findings are nearly agreement with the present study. The results reported by Mastin *et al.*,[12], Von Lichtenberg *et al.*,[13], Crabtree and Wilson,[14] are different with present findings. They have shown that in normal mice, newly arrived lung schistosomula are intravascular and do not attract any inflammatory responses.

Histologically, the outer layer of the cuticle consists of underlying muscle fibers. Occasional ridges and PAS-positive granules on it. In addition to these there was a development of gut caecae characterized by a thin muscular wall with nuclei and the presence of natural or black pigments from day 4 to 10 in both species. The results reported by Wilks[15], Wilson *et al.*,[3], Wheater and Wilson,[1] Crabtree and Wilson,[16], Mastin *et al.*,[17] for general features of the body of *andS. mansoni* schistosomula and Ogbe,[18,19] and Soomro,[9] for development of the *S. margrebowiei* larvae. However, Oliver [20], Clegg[21], Wilks[15] and Bloch[22] for *S. mansoni* and Ghandour, 1978 for *S. haematobium* schistosomula are in close agreement with the finding in this study in respect to the development of gut caecae and appearance of the black pigments.

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