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LIGHT AND ELECTRON MICROSCOPE STUDIES OF THE ADULT OF *PLEUROGENOIDES MEDIANS* (OLSSON, 1876) (TREMATODA: LECITHODENDRIIDAE) FROM IRAQI MARSH FROGS *RANA RIDIBUNDA*

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ABSTRACT

The morphology of the *Pleurogenoides medians* from *Rana ridibunda* in Iraq was studied by light and scanning electron microscopy. The light microscopical studies confirmed the original observations with the ventral sucker being smaller than the oral sucker and the genital pore being laterally situated. Electron micrographs of adult *P. medians* revealed that the body surface had a rough appearance by hand-like spines, which are more sparsely distributed towards the posterior end with a presumed function in nutrient absorption.

INTRODUCTION

Pleurogenoides medians (Olsson, 1876), a digenian lecithodendriid trematode , parasitizes numerous aquatic vertebrate species including frogs, fresh water fishes, urodeles and anurans (Smyth and Smyth, 1980). Frogs infection with *P. medians* has been reported worldwide and this trematode is the most common encountered frog intestinal parasite in Europe and Asia (Dawes, 1968; Cox, 1971; Hristovski and Less, 1973; Brooks, 1976; Gupta and Chopra, 1985). The life cycle of *P. medians* had been studied in detail by Buttner (1951), while that of other related species in the same genus namely *P. japonicus, P. tener* and *P. orientalis* were studied by Shibue (1953), Macy (1964) and Madhavi *et al*. (1987) respectively. Certain differences concerning the intermediate host and the structure of larval stages have been visualized.

In fact to date, the helminth parasites occuring in Iraqi frogs *Rana ridibunda* are poorly known. Except of some early works on trematodes (Saoud and Roshdy, 1970; Dauood, 1974; Hamad, 1985), no comprehensive list of *Pleurogenoides* species has been published. The present study undertaken to open the way for more detailed investigations of trematodes of frogs from Iraq to help in the elucidation of the morphology of adult worms and to establish the special adaptations of individuall species within their host.

MATERIALS AND METHODS

Adults of *P. medians* were obtained from the intestine of frogs *Rana ridibunda* Sulaimanya, Iraq. The flukes were fixed in AFA solution under coverglass pressure and stained with alum carmine according to standard protocols outlined by Humason (1972).

Diagrams were drawn with the aid of a camera lucida and all measurements were in millimeters and were based on stained specimens.

Scanning electron microscopy (SEM): Adult worms were fixed for 2-3 hr in 2.5% glutaraldehyde buffered with cacodylate to pH 7.4, washed in buffer and postfixed for 1-2 hr

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in cacodylate-buffered osmium tetraoxide, dehydrated in ethanol and freeze-dried. The mounted specimens were coated with gold and examined in a Jeol 840 scanning electron microscope.

RESULTS

The morphology of the adult form of *P. medians* is best illustrated in fig. 1. The body is small, oval and 1.52-2.16 mm by 1.10-1.43 mm in size. The tegument is spinulate with spines densely distributed over the anterior surface. The oral sucker is subterminal and 0.27 mm; the ventral sucker measures 0.14 mm is smaller than the oral sucker and situated slightly above the middle of the body. The pharynx is globular and measures 0.06 mm in diameter; the oesophagus is short (0.07 mm) and bifurcates into two short ceca (0.51 mm) which are terminated at level of the ventral sucker. Testes symmetrical, situated near the cecal termination, right testis measures 0.3X0.28 mm whereas the left testis measures 0.29X0.27 mm. The genital pore is lateral and situated near the right body margin at the level of pharynx. The cirrus sac is elliptical, running obliquely from the front margin of the ventral sucker to the genital pore and encloses the seminal vesicle, the well developed prostatic complex and the cirrus. The ovary measures 0.22X0.20 mm, is submedian to left testis and situated to the left of the ventral sucker. The vitellaria follicular are extracaecal and extends from the level of pharynx to a level slightly beyond the ovary. Uterus is much coiled and the coils occupy the posterior half of the body. Eggs are small, oval, operculate, amber coloured and measures 0.018X0.010 mm.

As illustrated in figs. 2-10 throughout the worm body. The tegument is carpeted with regularly arranged flattened hand-like spines and each spine is composed of 5-8 finger-like appendages (figs. 9-10). No structures corresponding to the microvillus-like projections or knobs were seen. It appears that the spines are more densely arranged towards the anterior end and more sparsely distributed towards the posterior end, although the spines all over the body surface are of similar size. It is to be note that the spines occur in association with suckers as well as genital pore but not with excretory pore at the extreme posterior end.

DISCUSSION

The digenean trematode belonging to Pleurogenoides found in the present study resemble and corresponds morphologically to *P. medians* described by Mathias (1924), Neuhaus (1941), Vojtkova (1974) and Smyth and Smyth (1980). The present form, however, differs from those described by these authors as well as from the rest known species of *Pleurogenoides* in having: (i) an ovary which is situated to the left of the ventral sucker, (ii) a genital pore which is present near the right body margin at the level of pharynx.

The differences noted above appear to be sufficient to treat the form described here as a new variety of *P. medians* for which the name *Pleurogenoides medians* var. *sulaimanialis* is being proposed.

This is the first time that *P. medians* has been recovered in *Rana ridibunda* in Iraq.

There have not, however, been any reports on the tegumental surface of *P. medians* nor indeed of its function, although there is an indication that the morphologically similar tegument of the echinostome trematodes and nematodes have pinocytic activity and a digesting effect on the host cells (Smales and Blankespoor, 1984; Koie, 1986, 1987; Imai *et al*., 1989). Nevertheless, this is the first attempt to study the tegumental surface in *P. medians* with scanning electron microscope and various conclusions can be made.

The highly increased tegumental surface due to the densely arranged spines suggests that the tegument is likely to be active in absorption of nutrients and probably also in digestion of host cells. It is possible that the digestive system of the adult worm probably does not function

or only to a very limited degree, in the absorption of nutrients as indicated by the presence of short intestinal caeca and this may result in the increment of the tegumental surface area that could probably have a possible role in *Pleurogenoides* nutrition. Future studies should give more critical attention to this possibility .

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دراسة المثقبة البالغة ***Pleurogenoides medians*** بالمجهر الضوئي والإلكتروني المتوسم من الضفادع العراقية ***Rana ridibunda***

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الخلاصة

تم دراسة بنية الديدان البالغة *Pleurogenoides medians* التي تستوطن أمعاء الضفادع العراقية *Pleurogenoides medians* باستخدام المجهر الضوئي والمجهر الإلكتروني المتوسم وقد تبين باستخدام المجهر الضوئي بان للدودة البالغة محجماً بطنياً أصغر من المحجم الفمي وفتحة تناسلية جانبية الموقع. وأما باستخدام المجهر الإلكتروني فقد لوحظ بان الجسم مغطى بأشواك شبيهة بأصابع اليد والتي تقل في كثافتها باتجاه النهاية الخلفية ويعتقد بأنها تؤدي دوراً وظيفياً في امتصاص المواد الغذائية.

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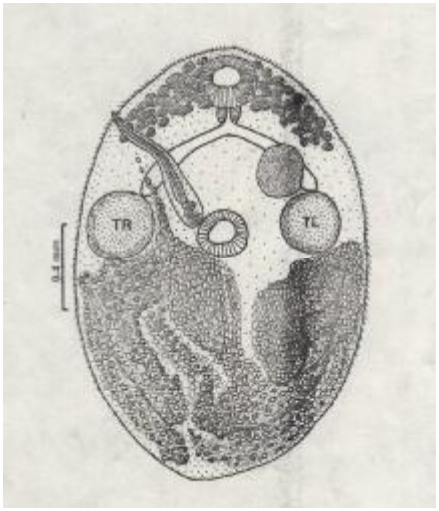

Fig. 1: Adult worm *Pleurogenoides medians* var. *sulaimanialis* drawn from testis; TL, left testis

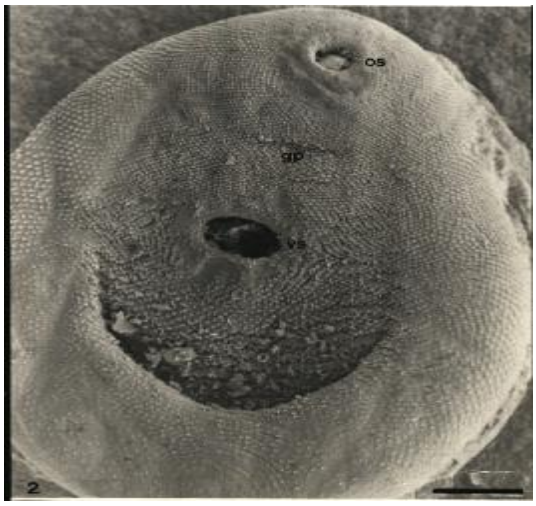

Fig. 2: Scanning electron micrograph of *Pleurogenoides medians* var. *sulaimanialis* showing spinulate surface (bar=0.5um). gp, genital pore; os, oral sucker; vs, ventral sucker.

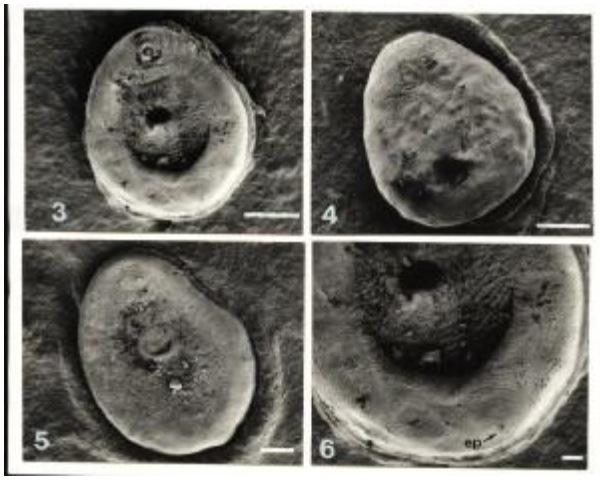

Fig. 3-6: Scanning electron micrograph of *leurogenoides medians* var. *sulaimanialis*. Fig. 3 (bar=1um) showing the ventral side; Fig. 4