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# SCANNING ELECTRON MICROSCOPY OF THE TEGUMENT OF MATURE FASCIOLA GIGANTICA WORMS (With 14 Figs.)

By  
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دراسة جلد الدودة الكبدية باستخدام الماسح الإلكتروني

عبد الرحمن ، فاطمة هيكل ، يسري السكري

باستخدام الماسح الإلكتروني وجد أن جلد الدودة الكبدية عديم الأشواك في السمات  
الظهرية والبطنية وكذلك في الأكتاف والجوانب الخارجية للجسم . وكانت الأشواك في السطح  
الظهري لسانية الشكل بينما كانت حرشفية الشكل على السطح البطني . وقد وجد أن كل شوكة  
تحتل عند القمة أشواك صغيرة ثانوية أصبعية الشكل . وكان عدد الأشواك الثانوية حوالي ١٠ - ١٥  
لكل شوكة أولية في الجزء الأمامي للجسم وتزداد هذه الأشواك إلى حوالي ٣٠ - ٣٥ عند  
وسط السطح البطني للجسم . وقد وجد أن الاختلافات المورفولوجية للجلد على السطح الظهري  
والبطني تساعد على التمييز بين الدودة الكبدية الهيباركا والجيغانكا .

## SUMMARY

Scanning electron microscopy of mature *Fasciola gigantica* worms revealed  
no spines on the tegument of the oral and ventral suckers, the shoulders  
and the lateral sides of the body.

The spines located on the dorsal surface were tongue shaped, while  
those on the ventral side were scale-like in appearance. Each spine  
was divided at its tip into small pointed secondary spines forming a  
fringe like structure. The number of these secondary spines was about  
10-15 per each primary one at the level of the anterior part of the  
body and increased to about 30-35 at the middle part of the ventral  
surface.

The differences in the morphological picture of the tegument of the  
dorsal and ventral surfaces of *Fasciola gigantica* might be used for distinguishing it from *Fasciola hepatica*.

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## INTRODUCTION

Fasciola worms are considered one of the most important worms infesting ruminants and cause great economic losses (PACHAURI, et al. 1988). With the use of Scanning Electron Microscopy (SEM), a new approach has been possible for elucidating the ultrastructure of the parasite surface and interpreting its functional morphology and significance in taxonomy.

The tegument of the mature F.hepatica and its development was described and reported by GORCHILOVA and POLYAKOVA-KRUSTEVA (1980) and BENNETT (1975 a&b) using SEM. In addition the tegument of Fasciolopsis buski has been also described by XU, et al. (1983) using the same scanning technique. To the best of our knowledge, SEM studies on the tegument of F.gigantica are lacking. Only one report on the description of the tegument of F.gigantica by light microscopy has been published in Egypt by HAIDA and SALIM (1960).

The aim of the present study is to give a detailed description of the tegument of F.gigantica infesting cattle in Egypt by the aid of SEM. Comparative study with previous reports on F.hepatica was also attempted.

## MATERIAL and METHODS

Adult flukes of different sizes were collected from the bile ducts of freshly slaughtered cattle and fixed for 2-4h in aqueous 4% glutaraldehyde 1% osmium tetroxide in a ratio of 3:1. The material was washed repeatedly in distilled water and then dehydrated through ethanol and amyl acetate. It was subsequently critically point dried using carbon dioxide. Then fixed to stubs with colloidal carbon and coated with gold palladium in a sputtering device. Specimens were examined and photographed using scanning electron microscope (SEM) operating at 20 kv.

## RESULTS

The SEM examinations revealed that the oral sucker of F.gigantica (Fig. 1) was circular in shape and its dorsal half was usually thicker than its ventral one. It exhibited variable numbers of dome-shaped contact receptors on its dorsal half (Fig. 2). The internal surface of the buccal tube carried balloon-like papillae measuring 2-2.3  $\mu$  (Fig. 3).

The ventral sucker (Fig. 4) showed also a circular outline and its anterior half was slightly thicker than the posterior one and showed more contact receptors all over



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its circumference. Many sensory papillary receptors were observed encircling the ventral sucker (Fig. 5). Some of these papillae had dome shape and others possessed single microvillus-like projections.

The tegument overlying both suckers was free from spines. However, the area of the tegument just anterior to the ventral sucker was slightly elevated, free from spines and contained the genital opening (Fig. 4).

The region of the shoulders and the lateral sides and their adjacent parts were free from spines (Fig. 6). The tegument of the rest of the worm was densely covered with backwardly directed spines which arise from the tegumental syncytium. On the dorsal surface the spines were observed at the area just posterior to the rim of the oral sucker and these spines extended to the area just anterior to the posterior extremity.

Anterior to the level of the shoulders (Fig. 7), the spines were nearly tongue-shape (12.8x10 U) and each spine divided at its tip into small pointed secondary spines in a fringe-like appearance. The number of these secondary spines was about 10-15 per each primary one. Between the primary spines, the tegumental apical plasma membrane was irregularly folded without a clear pattern of corrugation but some of them may have an invagination anterior to the base of the spines.

Posterior to the level of the shoulders, (Fig. 8) the spines were tongue shaped and increased in length (23x10 U) throughout the middle part of the dorsal surface. Each spine showed longer secondary spines in comparison to those of the anterior part of the body. Toward the posterior part of the body, the spines decreased in density and lengths (10-14 U x 7-12 U) and many appeared in a variable sizes (Fig. 9). In this area, the tegumental plasma membrane possessed transverse, circumferential folds in a regular pattern forming valleys between the similarly appeared orientated plateaux and a rectangular pattern of valleys was arranged around each spine.

The posterior extremity of the body was completely free from spines but showed irregular and deeper invaginations (valleys) and more prominent plateaux (Fig. 10). In addition to the transverse invaginations, some short longitudinal ones could also be reported.

On the ventral surface the area of the tegument just posterior to the oral sucker has in general less scale like spines (15x11 U) compared with the dorsal surface (Fig. 11). The number of the secondary spines in this region was nearly equal to those on the dorsal one. Just posterior to the ventral sucker, the spines increased in number posteriorly and became typically scale-like with a semicircular free border (29x30 U) (Fig. 12). The secondary spines were observed to occupy most of this border, they were longer than those of the scales of the anterior part of the body. The number of these secondary spines was about 30-35 per each primary one.



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Toward the posterior end of the ventral surface, the density as well as the size of the spines decreased (8x15 U) and the tegumental plasma membrane showed clear transverse folds forming valleys and plateaux (Fig. 13).

The spines at that part were found to arise from slightly elevated areas and each spine was observed to possess a transverse invagination anterior and posterior to its base. At the extremity of the ventral surface, the spines appeared very few in number and small in size (4x8 U) and have no elevated areas (Fig. 14). The free borders of these spines were recorded to carry also secondary spines. The tegumental plasma membrane showed also longitudinal folds in addition to the transverse ones, and the invaginations were much deeper than those observed on the anterior part of the body.

### DISCUSSION

From the results recorded in the present study, it could be concluded that the tegument overlying both, the oral and the ventral suckers of F.gigantica was free from spines. This result was in agreement with that reported for F.hepatica by BENNETT (1975) as well as that by GORCHILOVA and POLYAKOVA-KRUSTEVA (1980), who explained this character to be the cause of giving the fluke more freedom for light adhere to the host's tissue. The internal surface of the buccal tube carried balloon-like papillae as observed in Gastrothylax crumenifer by Veena TANDON and MAITRA (1981) who suggested that buccal lining might also be involved in food absorption. In addition, the ventral sucker was observed to have many sensory papillae beside the contact receptors, which were only present on the oral sucker, a result which agreed that reported by BENNETT (1975) for F.hepatica. According to GORCHILOVA and POLYAKOVA-KRUSTEVA (1980) these sensory formations contribute to the good orientation of the fluke and perceiving of the stimulations of the environment.

Moreover, the genital opening was found to be slightly elevated than its surrounding area. This, gave the fluke a chance to release the eggs away from its body and facilitate their passage to the bile.

Contrary to F.hepatica, the tegument of F.gigantica over the shoulders, the lateral sides and their adjacent parts were free from spines. In the author's opinion, this character might be attributed to the large size of the parasite under investigation as well as to facilitate its gliding movement inside the bile ducts especially the narrow ones. concerning the remaining parts of the tegument, the middle part of the body showed an increased number of primary and secondary spines in comparison to those found on the anterior cone. Similar results were also recorded in F.hepatica by BENNETT (1975). According to DAWES (1963), the observed multipointed spines could be considered to be the cause of hyperplasia of the bile duct epithelium for feeding purposes.



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They would cause an immediate environment of highly nutritious cellular debris which could be the tegument.

The occurrence of the plasma membrane behind the level of the shoulders with its clear plateaux and valleys as well the rectangular pattern of valley around each spine was also reported by THREADGOLD (1963), and BENNETT (1975) in case of *F. hepatica*. In the other's opinion and according to that mentioned by the previous authors, the function of these valleys is to allow the maximum independent movement of each spine to irritate the bile duct epithelium and their absence from the tegument of the anterior body in general might result in increased rigidity of the spines which were present there to anchor the fluke.

Regarding the shape of the spines, it was observed that those located on the dorsal surface of the body were tongue shaped and more elevated than those found on the ventral one. The latter appeared to have scale-like out-lines. These differences in the shape and size of spines on both surfaces could be attributed to the living behaviour of the parasite and a modified character of its effect on the habitat. In general the parasite is attached to its habitate (bile ducts) by its ventral surface. Hence its spines are wide, short and carry many secondary spines all over their free borders, giving an ideal form for maximal irritation. Meanwhile, the dorsal surface was a little bit far from the epithelial lining of the habitat, so it needed longer tongue shaped spines with secondary one only at its tip to cause irritation in the other direction.

In addition to the previously mentioned differences recorded between the spines of the dorsal and ventral surface of the body, the posterior end showed few scale-like spines on its ventral side which were absent from the dorsal one. On the contrary, BENNETT (1975) could not observe any great differences between the dorsal and ventral surfaces of *F. hepatica*.

Indeed, it could be concluded that the established differences in the morphological picture of the dorsal and ventral surfaces of the anterior and posterior part of the body in *F. gigantica*, expressed by the number, shape and size of the tegumentary spines, appearance, arrangement and depth of the outer membrane invaginations might be added as points of differentiation between *F. gigantica* and *F. hepatica*.

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### LEGENDS TO FIGURES

- Fig. 1-14:** Scanning electron micrographs of the *Fasciola gigantica* tegument.
- Fig. 1:** Oral sucker (OS) of adult *F. gigantica*. It's dorsal half (dh) is thicker than the ventral one (vh). The tegument overlying the sucker is free from the spines. Contact receptors (cr). 234 X.
- Fig. 2:** The contact receptors (cr) on the dorsal lip (dl) of the oral sucker. 468 X.
- Fig. 3:** Internal surface of the buccal tube carries balloon-like papillae (arrow) 2340 X.
- Fig. 4:** Ventral sucker (vs) and its adjacent parts. Sensory papillae (sn). Anterior cone of the body (ac). Genital opening (go). Spines back-wardly directed (sp). 70 X.
- Fig. 5:** Contact receptors (cr) and sensory papillae (sn). 234 X.
- Fig. 6:** The shoulder region (sh) which was free from spines. Tegument with spines (sp) 234 X.
- Fig. 7:** The spines (sp) at the dorsal surface of the anterior body cone. Each spine is divided at its tip into small pointed secondary spines. 2340 X.
- Fig. 8:** The tongue-shaped spines (sp) at the middle part of the dorsal surface. Notice the long secondary spines at the tips of the primary spines. 1500 X.

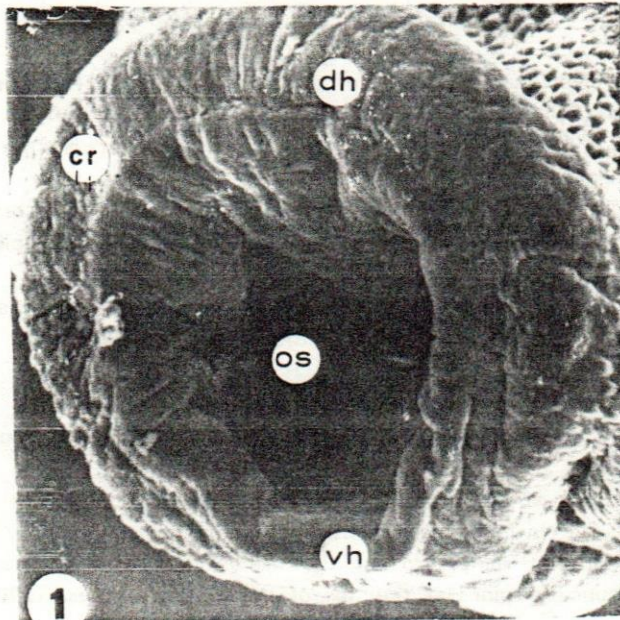


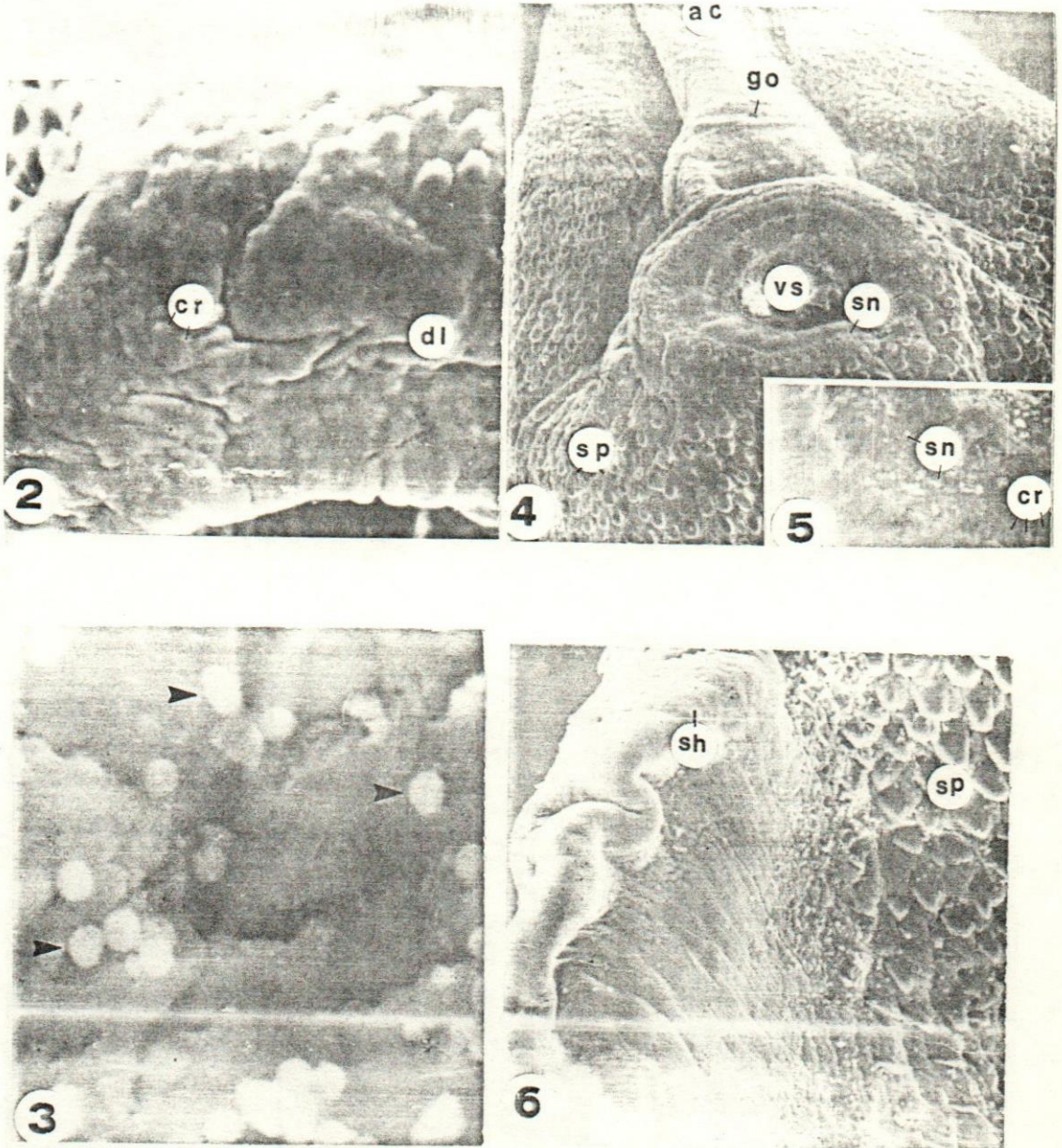
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- Fig. 9:** Tegument with less dense spines at the posterior part of the dorsal surface. The plasma membrane shows transverse invaginations (valleys) (v) with plateaux inbetween (p). Spine (sp) with valleys around it 468 X.
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