

STUDIES OF SCHISTOSOMA SPINDALE, MONTGOMERY, 1906

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Abstract

Schistosoma spindale (Montgomery, 1906) is a trematode inhabiting the portal and mesenteric veins of *Bubalus bubalis*. It was discovered by Montgomery (1906) from cattle in Mukteswar, India [1]. *Indopanorbis exustus* is the intermediate host of *Schistosoma spindale*. It is the site where the development of egg to the cercariae takes place. The cercaria is a feeble swimmer and in water its movements are mostly up and down. The cercariae can infect the host by oral route or by piercing the skin. It causes hepato-intestinal Schistosomiasis. The pathology caused by the parasite is due to injury caused by the migrating cercaria, injury by the adult parasite in the blood vessels and by the injury caused by ova. Animals with heavy infection suffer from anaemia, debility, emaciation and diarrhea, ending fatally. Two separate processes of invasion in the life history can be recognized. The first invasion involves the entry of the parasite into the host's body, while the second invasion occurs when the parasite invades the particular tissue of the host where it lives. The adaptability of the *S. spindale* in the host is aided by its unique morphological and physiological characteristics. The present study had been carried out on the ultrastructure of *S. spindale*. The adults show sexual dimorphism. The male *S. spindale* is shorter, coiled and stouter. The female *S. spindale* is long and slender. The male bears the female in the ventral canal, the gynaecophoric canal. There is bisexual pairing of adult worms. They live in the bloodstream of warm-blooded hosts. A spindale-shaped ova which is the characteristic of *S. spindale* is found.

Key words: *Schistosoma spindale*, Gynaecophoric canal, sanguivorous worm, Schistosomiasis, SEM, Scanning Electron Microscopy

I. INTRODUCTION

Schistosomes are best known blood flukes. They cause Schistosomiasis or bilharziasis in man and animals. Schistosomiasis can cause illness, debility and death in cattle and buffaloes. Several forms of this disease show enlarged liver and spleen, calcified bladder, deformity of the ureter or mal-functioning of the kidneys. The pathology of experimental *S. spindale* infection in buffaloes was studied by Fairley and Jasudasan [2]. The presence of a groove present in the body inspired the name Schistosome or 'split body'. Their peculiar characteristics make them different from other trematodes. The adult worms feed on the cellular and plasma fractions of the blood. As an adaptation of microhabitat, the worms are thread-like in shape. The worms show considerable sexual dimorphism. Cylindrical females are held in an extensive ventral groove, the gynaecophoric canal of the shorter and massive males. Armstrong (1965) suggested that the male produces pheromones that control the maintenance and maturation of females [3]. Schistosomes live in a host location in which the immunological defenses might be expected to be rapidly effective but these parasites possess their own adaptations which enable them partially to counter these defenses. Adult schistosomes are thus, able to live and reproduce for many years in a host which is having resistance to new infections. The present work is an attempt to know the ultrastructure of *S. spindale*.

II MATERIALS AND METHODS

Collection of parasites: The intestine of buffalo infected with *S. spindale* were collected from the local slaughter house in Hyderabad. Mesenteric portal veins were observed and localization of the parasites in blood vessel of the intestines was done. The parasites were collected by dissecting and then with the help of needle and brush.

Histology: The histology of the parasite's anatomical structure was done by paraffin embedding process of the parasite to make blocks which were sectioned and stained using Grenacher's Borax Carmine and Ehrlich's Haematoxylin-Eosin. [4]

Light Microscopy: The morphology was studied by flattening between two slides and fixed in 4% formalin for 24 hours. These worms were used for morphological studies after staining them using stains like Borax Carmine, Haematoxylin Eosin stain and Periodic Acid Schiff Method [5]

Scanning Electron Microscopy (SEM): Male and female *S. spindale* were obtained from the mesenteric veins of naturally infected *B. bubalis* in Hyderabad, India. The parasites were fixed in 2.5% Glutaraldehyde in 0.05M phosphate buffer (pH 7.2) for 24 hr at 4°C and post fixed 2% aqueous Osmium tetroxide in the same buffer for alcohol and processed for 2 hr. After the post fixation, samples were dehydrated in a series of graded alcohol and processed for critical point drying with Electron Microscopy

Science CPD Unit. The dried samples were mounted over the stubs. Finally, applied a thin layer of platinum metal over the sample using an automated sputter coater (JEOL JFC-1600) for 5 min. Then samples were observed and scanned in SEM (JOEL-JSM 5600) at various magnifications. SEM studies were carried as per the principles and techniques described by John and Lonne (6).

III. RESULTS

- 1. Male:** An adult *S. spindale* is seen coiled with smooth tegument under light microscope(Fig.1,2,3,6) and rough texture of tegument when seen under SEM(Fig.4,5). The male is seen coiled(Fig. 2,3).
- 2. Female:** The female is seen as a long thread like shape. (Fig. 1). The female is seen in the gynaecophoric canal of the male.(Fig.2,4,5).
- 3. Anterior end:** The anterior end of *S.spindale* consists of Oral sucker and Ventral sucker (Fig. 3,4,5,6)
- 4. Gynaecophoric canal:** The gynaecophoric canal shows male *S.spindale* with female in its gynaecophoric canal(Fig.2,4,5).
- 5. Ovary:** The ovary is very prominent in the female *S. Spindale* (Fig.10,11,12,13).
- 6. Ova:** Spindale shaped ova is very clearly seen(Fig.10,11,12)differentiating the *S.Spindale* from other species. The characteristic spindale shaped egg of *S. spindale* having terminal spine is seen under light microscope(Fig 15, 16).
- 7. Testes:** 4 to 5 testes are seen in the anterior end.(Fig. 6,7,8,9).
- 8. Caeca:** Caeca are very clearly observed in the female *S. spindale* (Fig. 1,2,3,11)

IV DISCUSSION

In the present study, the male and the female *S. spindale* shows smooth tegument when seen under light microscope. But the tegument when seen under higher magnification through SEM shows a rough texture. The tegumental surface was found to be uniform from anterior to posterior end and it is non-tuberculated in *S. spindale*, as per the studies done by Gupta and Aggrawal(7). The present study correlates with the studies made by Gupta and Aggrawal (2002) and by Kruatrachue (1983)[7,8]. Kruatrachue(1983) found the body surface of male *S.spindale* was fairly uniform with transverse ridges and papillae of various types evenly distributed over the body surface. The anterior end of the *S.spindale* shows subterminal oral sucker and a pedunculated ventral sucker. Kommu Sudhakar et al. 2018 have worked on the suckers and found ventral sucker to be larger than oral sucker. They found the Ventral sucker to be pedunculated round thick-rimmed and to be situated below the oral sucker. They also noticed numerous papillae and spines on the sucker.[9]. The suckers serve to maintain their position in the blood vessels against the circulatory current. The oral sucker surrounds the mouth of the adult and the ventral sucker is located posterior to the bifurcation of the gut. There is oesophagus which is followed by paired intestinal caeca which come together posteriorly, forming a single caecum that extends the remaining length of the body. According to Bhalerao(1932) intestinal caeca were seven to eight times as long as common caecum in Schistosomes[10]. The ovary is conspicuous. The present study shows striking resemblance to the studies made by Bogtish et al.,2005[11]. Bogtish et al. found single ovary located in the anterior portion of the body of the female. They also found uterus to be long or short, depending on the position of the ovary relative to the female genital pore. 4 to 5 testes were conspicuously seen in the male *S. spindale*. There is a difference in the number of testes recorded in the studies done by different people. Montgomery(1906) recorded 6-7 testes, Bhalerao(1932) 3-6, Chauhan et al.(1973) 3-7 and Mishra(1991) 3-5[1,10,12,13]. In the present study, 4-5 testes were found. Spindale shaped ova is very clearly seen in the female differentiating *S.Spindale* from other species of Schistosoma. The ovum of *S.spindale* is elongated spindle and zygomorphic. The characteristic spindale shaped egg of *S. spindale* shows the bulged region at the centre with horns on either sides. One of the horns is tapered into a spine at one end and at other end of the horn is blunt. Miracidium is present in the centre of the ovum which is released in the faecal matter. The results of the present study also correlates with the studies made by Hossain et al., 2015 and Kommu Sudhakar et al, 2018 who have also reported spindle shaped eggs with a terminal spine in the uterus[14,9] in *S.spindale*.

Comparative information of the different Schistosomes, source of information, their host, site of infection, egg excretion, snail vector, geographic location and Shape of Egg

Parasite species	Source of information	Definitive Host	Site of Infection	Egg excretion	Sail Vector	Geographic location	Shape of the Egg
<i>S. haematobium</i>	Rollinson D et al. 2001[15] CDC[16][17]	Humans, primates	Veins of urinogenital system	Urine	<i>Bulinus globosus</i> , <i>B. umbilicatus</i> , <i>B. truncatus</i> , <i>B.nasutus</i>	Africa, Parts of Middle east and Mediterranean region	oval with terminal spine
<i>S. mansoni</i>	Teixeira et al. 2007 [18] Anderson et al 2015[19]. CDC[16][17]	Humans Rodents	Intestinal mesenteric veins	Faeces	<i>Biomphalaria</i>	Africa, Middle East, America	oval with lateral spine
<i>S. japonicam</i>	Wang and Mao,1989[20] CDC[16][17]	Humans Ruminants, carnivores	Intestinal mesenteric veins	Faeces	<i>Oncomelania</i>	South East Asia	Rounded with lateral spine

<i>S. Intercalatum</i>	Almeda J et al., 1996[21], Rollinson D et al., 2001[15] CDC[16][17]	Humans Rodents, Cattle	Intestinal mesenteric veins	Faeces	<i>Bulinus, Physopsis</i>	Africa	Elongate (central bulge) and terminal spine
<i>S. spindale</i>	This study	Cattle, Buffalo	Intestinal mesenteric veins	Faeces	<i>Indoplanorbis exustus</i>	India, Africa	Spindle shaped having two horns and a spine on one horn

V CONCLUSION

Intestinal Schistosomiasis due to *S. spindale* is an economically important blood fluke infection widespread in India and other developing countries which is manifested in India and other countries. It is manifested as a chronic diarrhoeic disease if a large number of worm pairs inhabit the mesentery as per the studies done by Agarwal and Southgate, 2000[22]. According to Degheidy and Shalaby, 2010 an understanding of micro-morphological features plays an important role in the development of vaccines[23]. El-Shabasy et al., 2015 had revealed that alterations in the ultrastructure of schistosome worms were useful for evaluation of antischistosomal drugs for which there is necessity of understanding of normal ultrastructural morphology [24]. Thus the present study aims to have a better and indepth insight about the ultrastructure of *S. spindale*, for future research, for an effective targeted drug designing.

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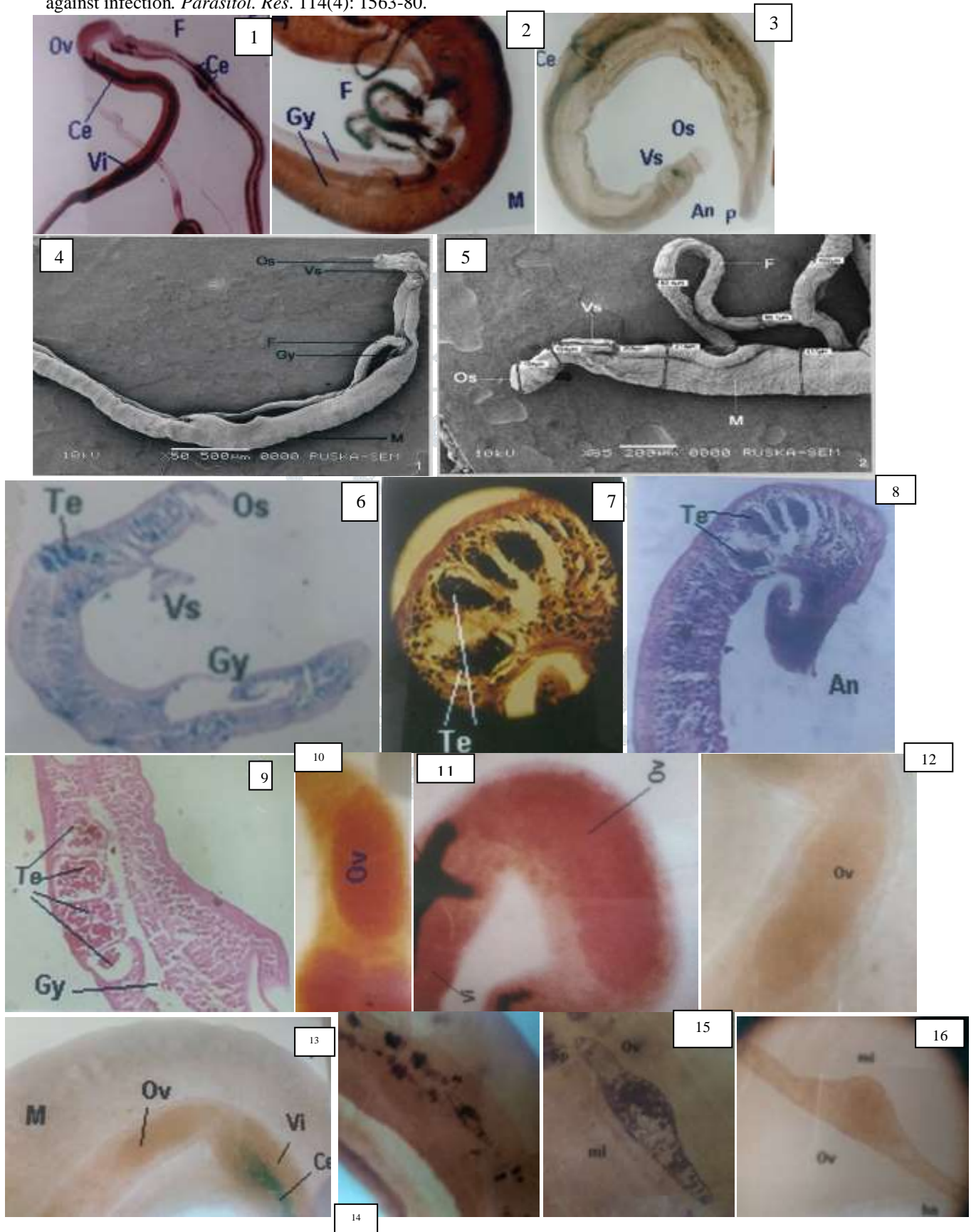
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Studies on *Schistosoma spindale* ,Montgomery,1906:

Fig. 1. B.C. stained Female *S. spindale* Fig. 2. B.C. stained Female in the gynaecophoric canal of the male. Fig. 3. .B.C. stained Male *S.spindale*. Fig. 4.. SEM of Adult Male *S. spindale* having female in its Gynaecophoric canal. Fig.5. SEM of the anterior end of *S. spindale* .Fig.. 6..Haematoxylin Eosin Stained section of male *S.spindale* showing oral sucker ,ventral sucker and gynaecophoric canal. Fig.7. Testes of male *S spindale*. stained by B.C. Fig 8. Section of the anterior end of male *S. spindale*.stained by Haematoxylin Eosin . Fig. 9. Section showing testes of male *S. spindale* stained by PAS method.Fig. 10. Fig. 11. Fig.12. Fig.13 . Different Photographs of Female *S. spindale* showing Ovary stained by B. C. Fig. 14. Ovum of *S. spindale* seen in the female present in the gynaecophoric canal of the male. Stained by B.C.. Fig.15 .Ovum in the female *S.spindale* by B.C.. Fig. 16. Ovum of *S.spindale* as seen in wet mount, Ovum found in the faecal matter of *Bubalus bulalis* (F-Female, M-Male, An-Anterior end, ,P-Posterior end, Gy-Gynaecophoric canal, Ov- Ovum, Ce-Caecum,Os-Oral Sucker, Ts-Testes, Vs-Ventral Sucker, Vi-Vitellaria, Mi-Miracidia, B.C,-Borax Carmine, PAS- Periodic Acid Schiff).

