



Morphological Characterization of *Monezia expansa* Rudolphi, 1810 (F: Anoplocephalidae) isolated from the intestine of the domestic sheep, *Ovis aries* (Bovidae) by light microscopy

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ABSTRACT

In the present study, A cestode parasite was recovered from the intestine of the domestic sheep, *Ovis aries* (Bovidae) from the main slaughterhouse of Cairo, Egypt during the year of 2015. The parasite was observed attached to the wall of the host intestine by unarmed scolex with suckers only. Five out of 10 gastrointestinal tracts (50%) were found to be infected. Light microscopic examination showed that the adult worm of this species was whitish in color measured 421-492 (470±0.4) cm in length and 4.2-6.99 (5.6±0.7) mm in width. The scolex was with prominent four suckers measured 0.38-0.42mm in diameter. The scolex led into a long neck followed by segments. The mature segment was broader than longer, each proglottid measured 3.23-5.29 (4.6± 0.2) mm in width and 1.0-1.64 (1.4±0.02) mm in width and showed a two set of genital organs. The ovaries and the vitelline glands formed a ring on either side, median to the longitudinal excretory canals, while the testes were distributed throughout the central field or they may be concentrated toward the sides. Each ovary was of ovoid shape, measured about 0.16-0.19 (0.14±0.01) × 0.12-0.15 (0.13±0.02) mm and was located 0.8± 0.1 mm apart from the lateral side. The testes were concentrated on both sides. At the posterior border of each proglottid, a row of interproglottidal glands was arranged around small pits. Gravid segments measured 0.022-0.076 (0.048±0.2) mm in length and 2.10-5.02 (3.05±0.02) mm in width. The present parasite was compared with the previous species of the same host which showed that the parasite isolated is *Monezia expansa*.

INTRODUCTION

Moniezia is a genus of tapeworms parasitic in mammals, comprises four known species such named *M. expansa*, *M. benedeni*, *M. autumnalis* and *M. baeri*. *M. expansa* is the most well-known species within the genus because of its high prevalence. Members of the genus are among the largest cestodes reaching up to 10 m in length. They inhabit the small intestine of the mammalian host. Their life cycle is indirect requiring intermediate hosts, which are the oribatid mites. They are characterized by the presence of interproglottid glands (Mehlhorn, 2008).

Moniezia expansa is commonly known as sheep tapeworm or double-pored ruminant tapeworm. It could be considered as the most important cestode parasite infecting sheep causing monieziasis which constitutes a problem in sheep breeding (Becker *et al.*, 1981, Polec 1990 and Maziad and El-Nemr, 2002). It is a large tapeworm inhabiting the small intestines of ruminants such as sheep, goats, and cattle (Gómez-Puerta *et al.*, 2008). There is an unusual report of human infection in Egypt (El-Shazly *et al.*, 2004). It is characterized by the presence of an unarmed scolex (i.e., hooks and rostellum are absent), two sets of reproductive systems in each proglottid, and each proglottid being short but very broad. *M. expansa* has a typical cestode body, consisting of the anterior scolex, followed by the neck and a highly extended body proper, the strobilus. It is an extremely long tapeworm and can reach an enormous length of up to 6–10 m. The scolex bears four large suckers, which are the holdfast organs to the host (Bashtar *et al.*, 2011). There are no rostellum and rostellar hooks, and the suckers are devoid of spines. This tapeworm, being monocious, contains both male and female reproductive organs in an individual. Thus, each proglottid is a complete reproductive unit. Moreover, one defining feature of the genus is that there are two sets of reproductive organs situated at lateral sides with the associated cirrus pouches and genital pores in each proglottid. The testes are numerous (Bashtar *et al.*, 2011). *M. expansa* infections are generally harmless and asymptomatic, even when the tapeworms are present in large numbers in young lambs. However heavy infection may cause intestinal obstruction, diarrhea, and weight loss (Elliott, 1986). The complete life cycle requires two hosts, ruminants as definitive hosts, and oribatid mites as intermediate hosts (Sinitsin, 1931 and Denegri *et al.*, 1998). Eggs are passed out from the intestine of the ruminant host along the gravid proglottids in the feces into the soil. The eggs are eaten by soil mites. Eggs

must reach the gut of mite hosts within 1 day of release otherwise they are desiccated. However, chances of development are very good as soil mites can be so numerous on a pasture that even if only 3% are infected (with 4–13 cysticercoids each), a grazing ruminant may ingest over 2,000 cysticercoids per kilogram of grass. Once inside the intestine of mites, the eggs hatch, and the oncospheres penetrate into the haemocoel and develops into the cysticercoid stage. This stage may take up to 4 months. When the infected mite is eaten by the grazing ruminants, mature cysticercoids are digested out of the mite and develop into mature tapeworms in the small intestine within 5–6 weeks (Denegri *et al.*, 1998). In the present study morphological characterization of *M. expansa*, a cestode parasite in the intestine of the domestic sheep, *Ovis aries* is carried out on the basis of light microscopy with a complete description of its different body parts including scolex, immature, and mature segments.

MATERIALS AND METHODS

The present study was conducted on ten gastrointestinal tracts collected from the domestic sheep, *Ovis aries* (Bovidae), from the main slaughterhouse of Cairo, Egypt, the work was approved by the institute of animal care and ethics committee; Faculty of veterinary medicine. After dissection and isolation of the gastrointestinal tracts, they were transported to the Parasitology laboratory. The various organs were separated from each other, placed individually in shallow plastic jars containing normal saline (0.85%), and were examined for helminth parasites followed by standard methods of Boomker *et al.* (1989). The contents of the abomasa, intestine, and stomach for each tract were put into separate plastic containers and each was made up of 1000ml with water. The contents were thoroughly mixed using a glass pipette and the digest of abomasa and small intestine were sieved through a sieve with 25 µm mesh size. The various aliquots of the ingesta and

the entire digests were taken into large Petri dishes and were examined under the microscope for parasitic worms. The parasites after their recovery from the hosts were washed in normal saline to free them from mucus. Relaxation is the first important step during the examination of cestodes, worms were placed in 4% formalin, 2 - 4 hours. After fixation, samples were washed in distilled water for 15 minutes to remove the excess fixative and then processed to staining which is carried out by using acetic acid alum carmine for 5-10 minutes according to (Carlton, 1967). After staining, a differentiation step must be carried out to remove the excess stain by placing the stained worms to a dilute solution of acid alcohol (0.5 ml in 1000 ml alcohol), it is better to carry out this process under a binocular dissecting microscope to detect the end point of differentiation. This is followed by dehydration in an ascending series of ethyl alcohol, 30%, 50%, 70%, 90%, 95% and absolute alcohol, leaving parasites for 2-5 minutes in each grade. The specimens were then cleared in xylene, then mounted in Canada balsam, covered with cover glass, and left to dry in an oven at 40C. Photomicrographs were taken by the use of Olympus BX53 microscope (Olympus Corporation, Tokyo, Japan). Drawings were made by camera Lucida.

RESULTS

Worms were recovered from the intestine of the examined host, where they were morphologically described and identified as *Moniezia expansa* (F: Anoplocephalidae).

Moniezia expansa Rudolphi (1810)

Figures.1-10

Description: Light microscopic examination showed that the adult worm of

this species was whitish in color measured 421-492 (470±0.4) cm in length and 4.2-6.99 (5.6±0.7) mm in width. The scolex was with prominent four suckers measured 0.38-0.42mm in diameter. The scolex led into a long neck followed by segments. The mature segment was broader than longer, each proglottid measured 3.23-5.29 (4.6± 0.2) mm in width and 1.0-1.64 (1.4±0.02) mm in width and showed a two set of genital organs. The ovaries and the vitelline glands formed a ring on either side, median to the longitudinal excretory canals, while the testes were distributed throughout the central field or they may be concentrated toward the sides. Each ovary was of ovoid shape, measured about 0.16-0.19 (0.14±0.01) × 0.12-0.15 (0.13±0.02) mm and was located 0.8± 0.1 mm apart from the lateral side. The testes were concentrated on both sides. At the posterior border of each proglottid, a row of interproglottidal glands was arranged around small pits. Gravid segments measured 0.022-0.076 (0.048±0.2) mm in length and 2.10-5.02 (3.05±0.02) mm in width. A line diagram illustrated *M. expansa* scolex and mature segments were shown in Fig. (10).

Taxonomic Summary:

Taxonomy: Animalia, Platyhelminthes, Cestoda, Cyclophyllidea, Anoplocephalidae.

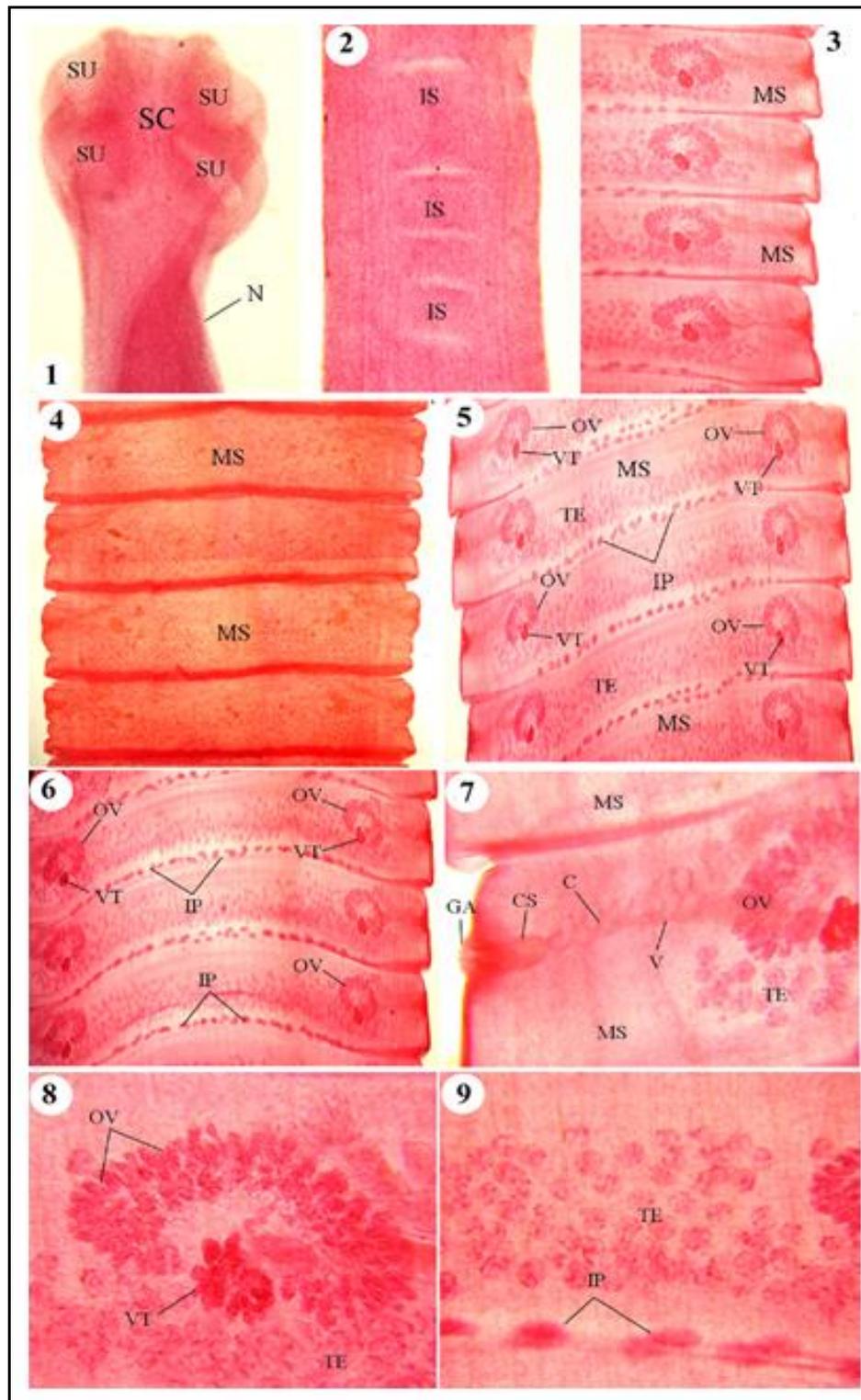
Type species: *Moniezia expansa* Rudolphi (1810).

Type host: the domestic sheep *Ovis aries* (F: Bovidae)

Type habitat and infection site: the adult worms were isolated from the intestine of the infected sheep

Type locality: Egypt.

Prevalence: 5(50%) out of the ten examined gastrointestinal tracts were found to be naturally infected by this parasite.



Figs.1-9: Photomicrographs of *Moniezia expansa* (F: Cyclophylidae) infecting intestine of the domestic sheep *Ovis aries* showing high magnifications of: (1) Scolex (SC) with four prominent suckers (SU) followed by a long neck (N), $\times 100$. (2) Immature proglottids (IS), $\times 120$. (3, 4) Mature broad proglottids (MS) with more developed genital structures, $\times 120$, $\times 200$. (5-7) Mature segments (MS) with two sets of genital organs per segment. The two horse shoe shaped ovaries (OV) surrounding a small mass of vitellaria (VT) forming a ring in the lateral sides of each segment, testes (TE) were distributed throughout the central field or they may be concentrated towards the sides. At the posterior border of each proglottid, a row of interproglottidal glands (IP) arranged as small pits. (C) cirrus, (CS) cirrus sac, (GA) genital atrium, (V) vagina, (fig. 5, 6 $\times 300$, fig. 7 $\times 500$). (8) Ovary (OV) and vitellaria (VT), $\times 800$. (9) Testes (TE) and the interproglottidal glands (IP), $\times 800$.

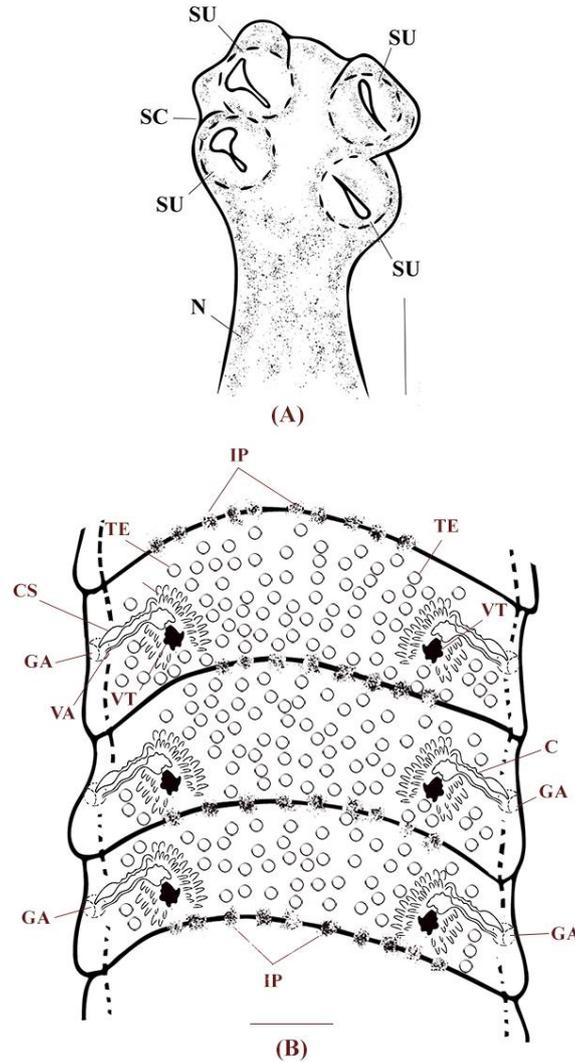


Fig. 10: A line diagram of *M. expansa*, (A) scolex, scale bar 0.1mm; (B) mature segments, scale bar 0.5mm.

DISCUSSION

Sheep and goats cover more than 30% of all domestic meat consumption and generate cash income through the export of meat and edible organs (Fletcher and Zelalem, 1991). Even though the livestock sub-sector contributes much to the national economy, its development is hampered by different constraints which include rampant animal diseases, poor nutrition, poor husbandry, poor infrastructure, shortage of trained manpower, and lack of government policies (Fletcher and Zelalem, 1991). Tapeworms have been noticed to be the most abundant helminth parasites infecting sheep through postmortem examinations. These

parasites play an important role in the large economic losses of farm animals (Maziad and El-Nemr, 2002). *Moniezia expansa* (Family: Anoplocephalidae) and other tapeworms infect sheep, goats, cattle (Khan *et al.*, 1989) and constituted a big problem in sheep raising countries (Tinar *et al.*, 1993). The present study has indicated a natural infection rate of 50% (5/10) which is in agreement with many investigators (Khan *et al.*, 1989, Kaur *et al.*, 1995, Umur and Gicik, 1995) whereas this value is higher than that reported by Hassanien (1978) and lower than those recorded by Ndarathi *et al.* (1989), Tinar *et al.* (1993) and Swarnkar *et al.* (1996) (16.21%). The present study reported that the

highest peak of infection was recorded in the winter season which is similar to some authors (Swarnkar *et al.*, 1996 and Tilahun, 1996). Other reports observed higher infection rates during spring (Hassanien, 1978 and Arvinder, 1995) and during summer (Arvinder *et al.*, 1993). These differences may be attributed to many environmental factors, vectors, parasites, and host habits in different countries (Arvinder *et al.*, 1993). The morphological studies of the present specimens confirmed their identification as *Moniezia expansa* as described by (Bambroo, 1969, Bali, 1970 and Soulsby, 1982) on the basis of various morphological characters as body length, maximum width, and size of suckers, scolex, presence of two sets of reproductive organs per mature segment, and presence of a continuous band of interproglottids. However, some intraspecific variations in the size ratio of the body and various other body organs were recorded in the present specimens. *M. expansa* was the most dominant helminth species identified during the course of the study. These results agreed with many records of previous authors (Tinar *et al.*, 1993, Arvinder *et al.*, 1993, Kaur *et al.*, 1995, Umur and Gicik). *Moniezia* is a unique member among Cyclophyllidea in possessing groups of interproglottidal glands in the parenchyma along the posterior edge of each proglottid. These gland cells are arranged in a follicular form containing a central sac. The secretions of these glands contain acetylcholin-esterase and alkaline phosphatase (Gunn and Probert, 1983).

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ARABIC SUMMARY

الوصف الشكلي لطفيل *مونيزيا إكسبانسا* (الديدان الشريطية) الذي تم عزله من أمعاء الخروف *أوفيس إبيريس* باستخدام الميكروسكوب الضوئي

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خلال الدراسة الحالية تم عزل نوع من انواع الديدان الشريطية التي تصيب الخراف والتي تم ذبحها بالمسلخ الرئيسي بالقاهرة، مصر عام 2015. تم ملاحظة وجود الطفيل متشبثا بجدار الامعاء بواسطة منطقة الرأس وعن طريق ممصات فقط. تم عزل الطفيل من خمسة خراف فقط من العشر خراف التي تم فحص القناة الهضمية لهم. فحص الطفيل باستخدام تقنية الميكروسكوب الضوئي أوضحت أن الطفيل الذي تم عزله هو طفيل بالغ أبيض اللون. تحتوي منطقة الرأس على أربعة ممصات. تلي منطقة الرأس منطقة العنق الغير مقسمة يتبعها القطع اللسانية الغير ناضجة ثم القطع اللسانية الناضجة. وتم وصف الجهاز التناسلي والاعضاء التكاثرية للقطع اللسانية الناضجة مثل المبيض، والخصي والرحم. وبعد مقارنة الطفيل بمثيله من الانواع السابقه من الديدان الشريطية تم تصنيف الطفيل على أنه طفيل ال *مونيزيا إكسبانسا*.