

DESCRIPTIONS ON THE MORPHOLOGY OF SOME NEMATODES OF THE MALAYSIAN DOMESTIC CHICKEN (*Gallus domesticus*) USING SCANNING ELECTRON MICROSCOPY

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ABSTRACT. A total of five species of the nematodes, *Acuaria spiralis*, *Gonyglonema ingluvicola*, *Ascaridia galli*, *Heterakis gallinarum* and *Oxyspirura mansoni* were recovered from the Malaysia domestic chicken, *Gallus domesticus* and studied for their morphology. Scanning electron microscopy was used to study and observe the outer surface of the nematodes. The morphological of the five species of nematodes were described and compared.

Keywords: morphology, nematodes, Malaysian domestic chicken, scanning electron microscopy

INTRODUCTION

Nematodes are invertebrate roundworms and they inhabit marine, freshwater, and terrestrial environments. Nematodes belong to the phylum Nematoda (or Nemata) which includes parasites of plants and of animals, including that of humans; they also feed on bacteria, fungi, algae and on other nematodes (Platt, 1994). Due to its large number of species, nematodes are the most important group of helminth parasites of poultry. These parasitic species

can cause damage to the host, especially in severe infections.

Nematodes are elongated, cylindrical and un-segmented roundworms. They are covered with a tough, non-cellular layer called the cuticle and they have a well-developed alimentary tract. Most species of nematodes are bisexual and they a single worm (Butcher, Hogsette and Jacobs, 1997).

In terms of size, they appear small, inconspicuous and seemingly unimportant to humans. However, some nematodes can cause diseases of great importance to humans and domestic and wild plants and animals (Gerald and Larry, 1996) whereas some are beneficial in attacking insect pests, mostly sterilizing, or otherwise debilitating their hosts (Grewall, Ehlers and Shapiro-Ilan, 2005). According to Gerald and Larry (1996), nonparasitic nematodes may accidentally find their way into a vertebrate and become short-lived, and they can become a pathogenic parasite.

Most nematode species are of bilateral symmetry (Croll, 1976). Lee (1965) stated that the male nematode has a single testis and the female possesses ovarian

tubules. Many species of nematodes are oviparous, while some are ovoviviparous or viviparous. The adult stage is reached after 4 larval stage molts. Male nematodes are small in size compared to female (Lee, 1965). Eggs are the transmission stage; some produce the infective larvae that penetrate host body. Nematodes have six stages in their life cycle; egg, four juvenile stages and adult (Croll and Matthews, 1977).

A typical nematode has a cylindrical or filariform body with the extremities usually more or less attenuated or truncated. The length varies from less than one millimeter to more than one meter, the ratio of length to breadth varying considerably. They have more elastic and tough cuticle as compared with cestodes and generally smooth, seldom spined, scaled or scattered with bosses and transversely, longitudinally or rarely obliquely striated (Yamaguti, 1963).

The body wall of nematodes comprise of the cuticle, hypodermis, and body wall musculature. The outermost covering is the cuticle, a complex structure of great functional significance to the animals. The cuticle also lines the stomodeum, proctodeum, excretory pore, and vagina. A carbohydrate-rich coat, the surface of the cuticle is found in free-living and parasitic nematodes, 5 to 20 nm in thickness (Blaxter *et al.*, 1992). The cuticle is formed by an underlying sub-cuticle layer, called the hypodermis. This layer forms four longitudinal thickenings on the inner aspect, situated dorsally, ventrally

and laterally and known as the longitudinal lines. Lateral lines contain the longitudinal canals of the excretory system. For the muscular layer, which follows next and lines the body cavity, consists of a number of cells having a basal contractile portion which is transversely striated, and a cytoplasmic portion which contains the nucleus and it is also connected to the nerve trunks in the dorsal or ventral line. This muscular layer is divided into four quadrants by longitudinal lines (Soulsby, 1968).

The present paper describes the morphology of five species of nematodes in the Malaysian domestic chicken, *Gallus domesticus* using scanning electron microscopy. Morphological differences between the five species were compared.

MATERIALS AND METHODS

Samples

The study was conducted on five species of nematode. The five species were obtained fresh from the Malaysian domestic chicken, *Gallus domesticus*, obtained from various farms in the northern parts of Peninsular Malaysia. The five species of nematodes i.e. *Acuaria spiralis*, *Gonyglonema ingluvicola*, *Ascaridia galli*, *Oxyspirura mansoni* and *Heterakis gallinarum* were identified as according to the descriptions of Lancaster (1957), Lim (1971), Mustaffa-Babjee (1980) and Rahman *et al.* (2009).

Preservation of samples

The nematodes, both male and female were preserved in 70% alcohol. Higher concentrations of alcohols were avoided which may result in structural dehydration artifacts, including shrinkage and body surface distortions sufficient to obscure features required for morphological identification and analysis, thereby compromising precise morphometrics (Naem *et al.*, 2010)

Light microscopy

The morphology of the nematodes was carried out by clearing in lacto-phenol with an addition of a few drops of 10% lactic acid and examined under the light microscope at 10×, 20×, and 40× magnifications. The morphological structures were identified as according to the descriptions of Soulsby (1968).

RESULTS AND OBSERVATIONS

From the observations using LM, *Acuaria spiralis* is characterised by having 'cordons' (cuticular ridges or grooves) at the cuticle of the anterior part of the body (Plate 1). The cordon may be non-recurrent or recurrent where it runs down the body and turn back forwards again. The lips are usually small and triangular in shape whereas the pharynx is cylindrical. Plate 2 showed the anterior and posterior of the female worm. At the posterior end is the vulva (Plate 3).

Gongylonema ingluvicola has cuticular plates at the anterior end of their body (Plate 4) and the posterior part of the male bears wide caudal alae and is supported by numerous pedunculated papillae (Plate 5). The female opens posteriorly, showing the eggs and the vulva of this species (Plate 6).

The anterior part of *Ascaridia galli* shows the presence of the cuticular plates

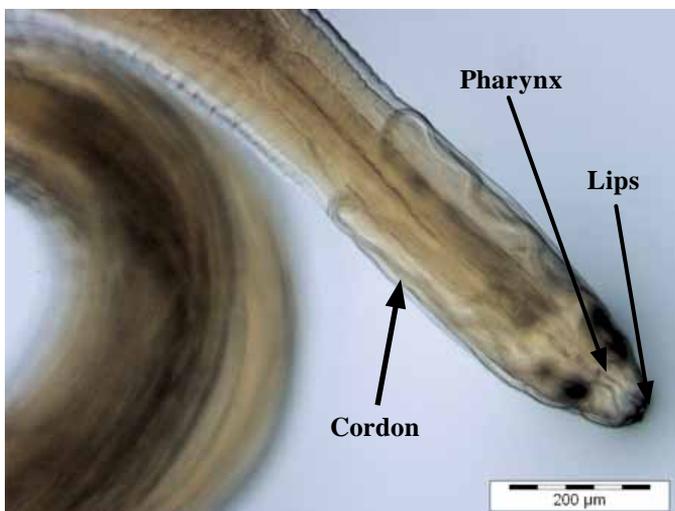


Plate 1. Lateral view of a male *Acuaria spiralis* showing the cordons at the anterior end

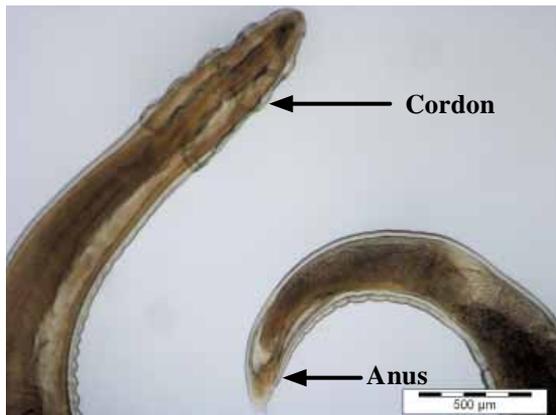


Plate 2. Anterior and posterior of female *Acuarria spiralis*.

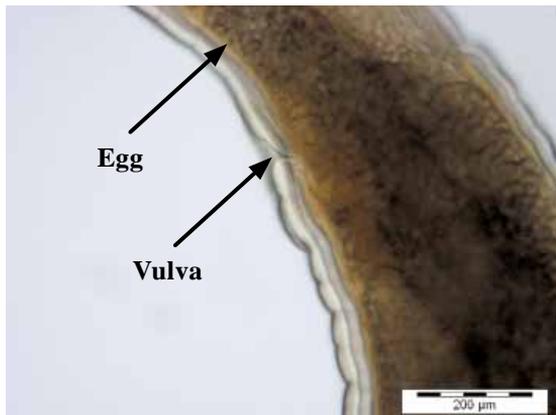


Plate 3. The vulva at the posterior part of the body of a female *Acuarria spiralis*.

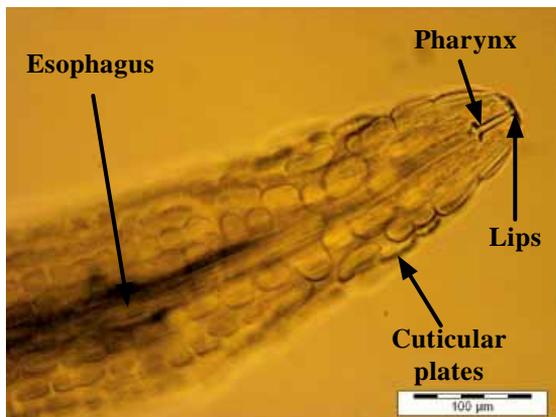


Plate 4. Anterior end, ventral view of male *Gongylonema ingluvicola* showing the cuticular plates



Plate 5. Posterior end of male *Gongylonema ingluvicola* showing the caudal alae

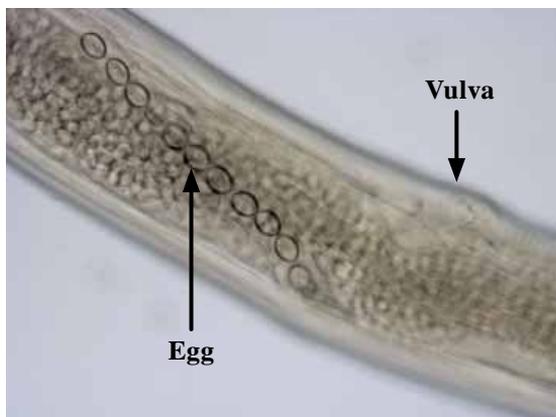


Plate 6. Vulva and eggs of *Gongylonema ingluvicola*.

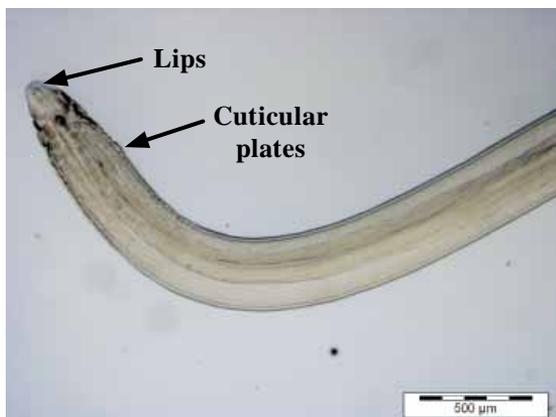


Plate 7. Anterior part of female *Ascaridia galli* showing cuticular plates

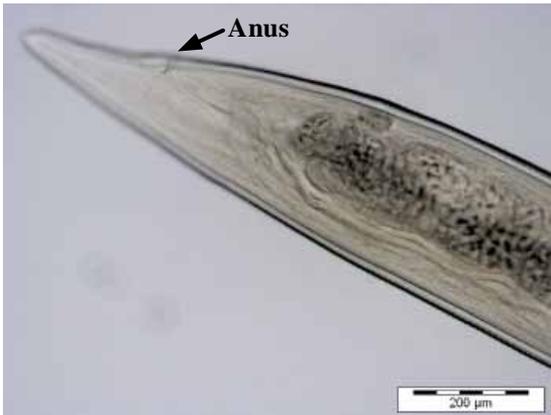


Plate 8. Posterior part of female *Ascaridia galli* showing the anus

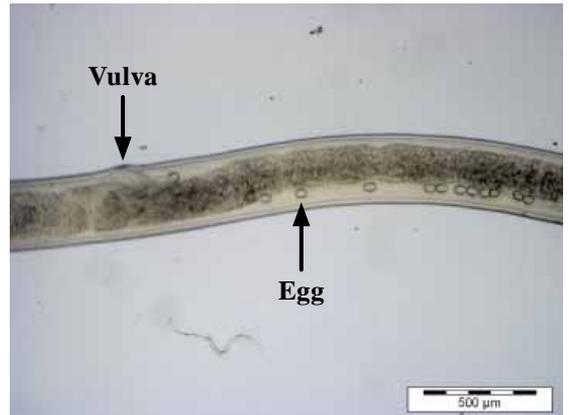


Plate 9. Vulva and eggs of female *Ascaridia galli*.

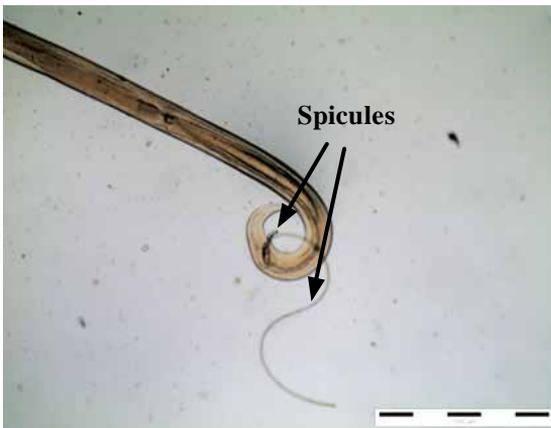


Plate 10. Posterior end of male, *Oxyspirura mansoni* showing the two spicules of different lengths

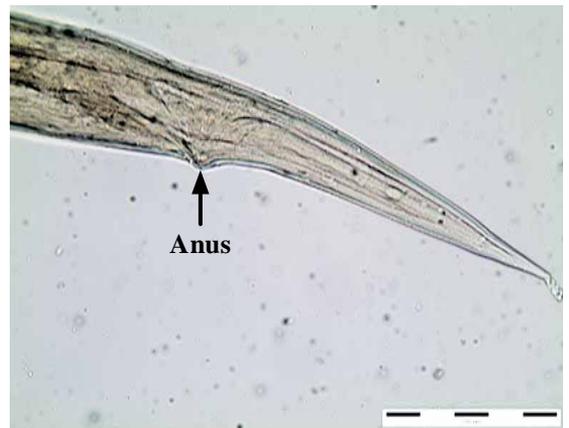


Plate 11. Posterior end of female *Oxyspirura mansoni* showing the anus



Plate 12. Anterior end of *Heterakis gallinarum*, showing the bulb-structure at its alimentary tract

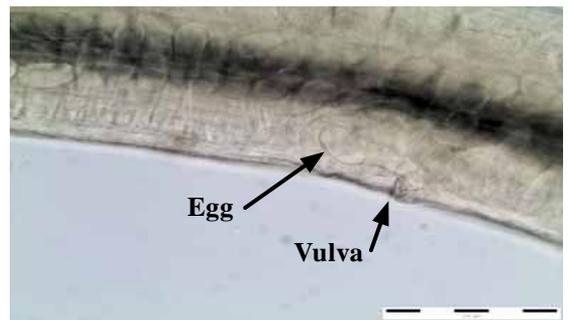


Plate 13. The vulva and eggs of *Heterakis gallinarum*.

only in the female (Plate 7). The vulva of female *Ascaridia galli* is situated a short distance anterior to the middle of the body (Plate 8) with eggs in the uterus (Plate 9).

The left spicule of male *Oxyspirura mansoni* is slender with the right long and stout at the posterior part (Plate 10). Female vulva of this species is situated in the posterior part of the body (Plate 11).

Heterakis gallinarum is characterised by the presence of a bulb-like structure at its alimentary tract (Plate 12), with the presence of numerous eggs in the uterus in the female (Plate 13).

DISCUSSION

Bodies of nematodes are generally elongate and cylindrical, tapering at both ends. A certain degree of sexual dimorphism exists, for the posterior end of males is commonly armed with special structures, for example alae and papillae and are curved ventrally. Almost all males of nematodes are smaller than females. Nematodes differ greatly in size, depending on the species. Some of nematode species are microscopic and other species measure no more than 1 mm (Cheng, 1964).

According to Cheng (1964), the cuticle of parasitic nematodes is generally smooth and the various structures such as spines, bristles, warts, punctuations, papillae, striations and ridges may be present. The arrangements and positions of such structures are of taxonomic importance. Under the layer of cuticle that

covers the nematode body is a thin layer of hypodermis.

In the present research, the anterior end of *Acuaria spiralis* has four cuticular cordons beginning at sites of lips and running to posterior end, not uniting and not forming marked curves. The cordons end before reaching the posterior end of body (Skrjabin, 1969). Both sexes also have cordons; this species is easily separated from others. The caudal end of male has lateral alae and the posterior end of female is blunt. The vulva of this species lie in the posterior third of that body (Skrjabin, 1969).

Gongylonema ingluvicola is normally found in the upper digestive tracts of birds and mammals. The cuticle of the anterior end is covered with large bosses (or cuticular plates), or irregular secutes and arranged in eight longitudinal rows (Gerald & Larry, 1996). *Gongylonema ingluvicola* has false lips and mouth capsule is present. Male left spicule is slender and have alate. Female vulva opens posteriorly and furthermore, lips of this species are small and also have a short pharynx with simple wall (Soulsby, 1968).

Gerald and Larry (1996) stated that the members of the Ascaridae are characterised by having three prominent lips, valvulated bulb absent, and pre-cloacal sucker is sometimes present. It is large, thick, and yellowish-white with oral opening at the anterior end. The mouth is surrounded by three large prominent lips and the esophagus is without a distinct posterior bulb. Male of

this species is slightly smaller, and more slender than female. The tail of male has small caudal alae and bears a number of caudal papillae, most of which are short and thick. The posterior end is obliquely truncated with pre-cloacal sucker oval or circular on ventral side of tail. Spicules are well developed, protruding out at the anal opening. Female of this species is larger, stouter and vigorous than male. The tail tapers and the vulva is situated a short distance anterior to the middle of body.

The mouth of *Oxyspirura mansoni* is without defined lips and a short buccal capsule is usually present. The male is with or without caudal alae, preanal papillae simple, numerous and arranged in a linear row. The spicules of male are usually unequal and the vulva in female may be either anterior or posterior. In male, the tail is usually spiral, caudal alae absent and the spicules unequal in length. In female, the vulva is located in front of the anus. The male tail is curved ventrally, without caudal alae. The spicules are unequal, one is slender and the other is long and stout. The vulva of this species is situated in the posterior part of the body (Soulsby, 1968).

Soulsby (1968) stated that *Heterakis gallinarum* has three well-defined lips, the esophagus with a short narrow anterior portion (pharynx) and a long posterior part ending in a bulb. The cuticle is usually with lateral flanges. Esophagus has a short narrow anterior portion (pharynx) and with a long broader posterior portion ending in a well-developed bulb containing a valvular apparatus. The worm is small

and white in colour. Mouth surrounded by three lips, a small buccal or pharynx. Esophagus ends in a well-developed bulb containing a valvular apparatus. The tail of male has large caudal alae extending some distance down the sides of the body, bearing a number of caudal papillae and a prominent pre-cloacal sucker. The spicules are well developed, unequal, protruding out at anal opening. The tail of female is also elongated, narrow and pointed. The vulva of the *Heterakis gallinarum* is situated at the middle of the body.

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