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**SCANNING ELECTRON MICROSCOPIC STUDY ON
CONTRACAECUM MAGNIPAPILLATUM
(NEMATODA: ANISAKIDAE) FROM CORMORANTS IN WADI
AL-RAIYAN LAKE AREA, FAYOUM, EGYPT**

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Key Words : SEM, *Contracaecum*, Anizakidae, Cormorants, Wadi Al-Raiyan Lake, Fayoum.

ABSTRACT

Contracaecum magnipapillatum nematode were obtained from the proventriculus of the migrating piscivorous Cormorants fish feeders *Phalacrocorax carbo* locally called Anaz, hunted from the area surround Wadi Al-Raiyan lake in the western desert in Fayoum Governorate in Egypt. Infection in the proventriculus is highly effective and induces an extensive inflammatory response. This represents a new locality record for these nematodes. The morphological data serve as a basis for comparative studies of other species in the genus. Only two cases of *Phalacrocorax carbo* out of five examined cormorants were infected. The study revealed some taxonomic features, including three lips (one dorsal and two subventral) provided with three cephalic papillae and two amphids. Cuticular transverse striations occurring along entire body posterior to lips. A ventral sucker like-shaped is situated a short distance anterior to the cloacal opening of the male. The cloacal papillae include: 2 pairs of precloacals, 2 pairs of lateral and 2 pairs of ventral postcloacals. Each papilla is a dome-shaped with centrum surrounded by two circles of thickening. One pair of lateral and one pair of ventral subterminal papillae.

INTRODUCTION

Neither encysted nor free *Contracaecum* larvae can severely affect fish. Tissue reaction, inflammation, epitheloid formation and fibrous encapsulation around encysted larvae is localized. Multiple infection of the mesenteries resulting in extensive

inflammation, fibrosis and even some visceral adhesions were seen only in large fish with no apparent impact on their body condition (Mbahinzireki, 1980). As worms tend to migrate to the surface of fish once fish die, such (wormy) fish deter customers. In such cases fish have to be de-gutted and filleted in

order to be sold for consumption and the cost difference of which has to be paid by the producers. For prevention of larval nematode infection by keeping away piscivorous birds is impractical not only in fishing areas in natural habitats or man-made impoundments but even in fish ponds. In fish ponds preventive treatment of *Contracaecum* by elimination of copepods (by insecticides such as Masoten or Bromex) may be of some value if suitably timed, soon after its contamination by pelicans. (Paperna, 1996).

Contracaecum infection occurs in Israel (Paperna, 1964), Zaire, Mali, Niger (Khalil, 1971), and in lakes Victoria, George, Nakuru, Naivasha, Baringo and Magadi (Paperna, 1974; Malvestuto and Ogambo, 1978). South Africa, where it was also reported from brackish water hosts (Boomker, 1982; Van As and Basson, 1983). Infections of the pericardia in cichlid fish occur in Israel (Landsberg, 1989). Egypt, Mali, most large and small East African (Rift Valley) lakes including lakes Kivu, Edward and Albert, (Paperna, 1996), Larvae of *Contracaecum* tend to escape from their cysts, and crawl out of their host body after its death.

Regarding the definitive hosts of the members of the genus *Contracaecum* are piscivorous birds and mammals associated with fresh,

brackish and Sea water (e.g. cormorants, pelicans & seals). The parasites inhabit the stomach and consume food ingested by the host. Otherwise they attach to the stomach wall.

Scanning electron microscopy provides a means to define more objectively the surface topographical features of species than light microscopy. Al-Bassel (1990) redescribed *Contracaecum* larvae from freshwater fishes in Lake Nasser in Egypt by light microscopy.

The aim of the present work is demonstrate the transverse striations, spicules, lips, papillae and phasmid of these nematodes. The high resolution of the SEM enabled the author to give a clear description of the fine structures and added more details of the previously reported nematode. In the present work *Contracaecum magnipapillatum* is reported for the first time in Wadi Al-Raiyan lake area in the western desert at Fayoum Governorate, Egypt.

MATERIALS AND METHODS

Five from the migratory piscivorous *Phalacrocorax carbo* (Pelecaniformes: Phalacrocoracidae) locally called Anaz were hunted in spring from the beach of lake Wadi Al-Raiyan in the western desert in Fayoum Governorate in Egypt. Nematodes were removed from the proventriculus of the dissected bird

, rinsed in 0.7% saline, fixed in cold 10% buffered formalin and cleared in lactophenol for morphological study using light microscopy. Some specimens were postfixed in ethanol, dehydrated in ascending series of ethanol, then transferred through a series of intermediate fluids (3:1, 2:1 and 1:1) for 100% ethanol & amyl acetate, then to liquid CO₂ in Polaron critical point dryer. The dried specimens were then coated with gold/palladium and examined with JEOL scanning electron microscope.

RESULTS

Contraecaecum magnipapillatum: (Figs. 1-12)

Body relatively stout, with maximum width of body slightly posterior to midbody. Total body length about 5-8 times oesophageal length. Cuticular transverse striations occurring along entire body posterior to lips (Figs. 1,3). Neck striation plate-like with free anteriorly directed edges, (Figs. 1,2). Body length are 35-40 mm long for males and 45-50 mm for females. Mouth opening is guarded with three large lips, one dorsal and two subventral, the inner surface of each lip is provided with a thick smooth cuticular plate (Figs. 1, 2). Each lip has a wide base and a narrow anterior extremity with a trilobed appearance (Figs. 1,2). Three cephalic papillae and two amphids are found on the outer surfaces of the lips (Figs 1,2). The ventral sucker like-shaped is

situated a short distance anterior to the cloacal opening of the male, it covered with several small cuticular bosses (Figs. 4, 5). The cloacal opening of the male is situated on the ventral surface near posterior extremity (Figs. 4, 5). The cloacal papillae are arranged as follows: 2 pairs of precloacals, 2 pairs of lateral and 2 pairs of ventral postcloacals (Figs. 4,5), one pair of lateral and one pair of ventral subterminal papillae (Figs. 4,5). Each papilla is a dome-shaped with centrum surrounded by two circles of thickening (Figs. 6,7). A narrow lateral alae extend the whole length of the body except the two extremities (Figs. 1,3,11). The tail of the male relatively pointed, with terminal pyramid-shaped projection curved ventrally, it covered with cuticular folds, one pair of Phasmid and two pair of papillae (Figs. 5,8). The two spicules are equal 2.6-3.1 long. Cross section of the right spicule show that, they are hollow structure (Fig. 5). The wall of the right spicule forms incomplete ring (Fig. 5). The surface of the left spicule is smooth (Fig. 5). The tail of the fourth stage larvae is pointed posteriorly, with distinct terminal mucron curved dorsally (Fig. 9). The tail of the female is relatively pointed, lacking distinct mucron (Fig. 10). The anus of the female is crescent-shaped, situated near posterior extremity (Fig. 10). Cross section of the worm at the middle of the body show that, oval in shaped possessing a central tube of the intestine, longitudinal and

transverse muscle layers and lateral alae in each side (Fig.12).

DISCUSSION

Contracaecum larvae was recorded by Myers *et al.*, (1962) from Egyptian fishes. Salgado *et al.* (1978) reported *Contracaecum* sp. larvae from *Mugil cephalus* from Mexico. Other workers reported *Contracaecum* sp. larvae from freshwater fishes in different localities in the world (Boomker, 1982 ; Goude and Vanicek, 1985; Fatima, 1985 and Torres and Cubillos, 1987).

Valtonen *et al.* (1988) reported *Contracaecum osculatum* in seven species of fish and two species of seals from Bothnian Bay , Baltic Sea . Highest prevalence were salmon (*Salmo salar*) (20%), bull-trout (*Myxocephalus scorpius*) (20%) , burbot (*Lota lota*) (16%) and cod (*Gadus morhua*) (15%). Prevalence were higher in larger fish but no seasonal variations were noted in prevalence and intensity . The parasite occurred sporadically in the resident ring seal (*Phoca hispida*) but a mean of 640 worms was found in grey seal (*Halichoerus grypus*) which visits the area studied for a few weeks each spring. These animals probably maintain the infection in fish in the area . In grey seals , worms tended to occur in aggregations consisting of third-and fourth-stage larvae and adults (Anderson,1992).

McClelland and Ronald (1974) found *C. osculatum* in the stomach of harbour (*Phoca vitulina*) and grey seals from Cape Breton Island , Canada and in harp seals (*Pagophilus groenlandicus*) from the gulf of Lawrence .

Navone *et al.*,(2000) reported *Contracaecum multipapillatum* from the esophagus and stomach of the great egret *Egretta alba* from Argentina. Tolgay and Tolgay (1966) reported *Contracaecum* larvae from Anchovies (*Engraulis encrasicolus*) from the Black Sea and experimental feeding of the laboratory animals in Turkey. They summarized the life cycle as follows: the life cycle involves 3 hosts . The definitive host is a fish eating bird in inland freshwater aquatic system. Unembryonated egg are shed in water with the host's faeces. The egg embryonates and the larva moults to the second stage (L 2) stage within the egg. When the egg is eaten by a crustacean, the L 2 hatches and penetrates into the haemocoel. When the infected crustacean is eaten by a fish second intermediate host, the larva penetrates the gut wall into the body cavity where it grows and moults to the (L 3). These third-stage larvae are commonly found encysted in the body cavity or the viscera . when an infected fish is eaten by the bird definitive host the (L 3) larvae moult twice to the adult stage (Tolgay and Tolgay, 1966). Dick

(1987) was found 95 % of the larvae of *Contracaecum* spp. were found in the atrium of the heart of fathead minnows (*Pimephales promelas*). Atria containing nematodes were two to three times larger than an atria from uninfected fish, were thin walled and the muscular trabeculae were damaged. Atria with larvae were congested with blood that appeared to be organizing and extending as a thrombus into the ventricle, but there was no other evidence of a host response. Alternatively, there was an intense cellular response surrounding larvae in the body cavity of these and other host species. Apparently, this parasite which lacks host specificity has evolved an additional strategy to evade the host immune response (Dick, 1987). In Egypt, *Contracaecum* sp. larvae were recorded from freshwater fishes by several investigators (Imam *et al.*, 1979, Abu El-Ez, 1988 and Al-Bassel, 1990).

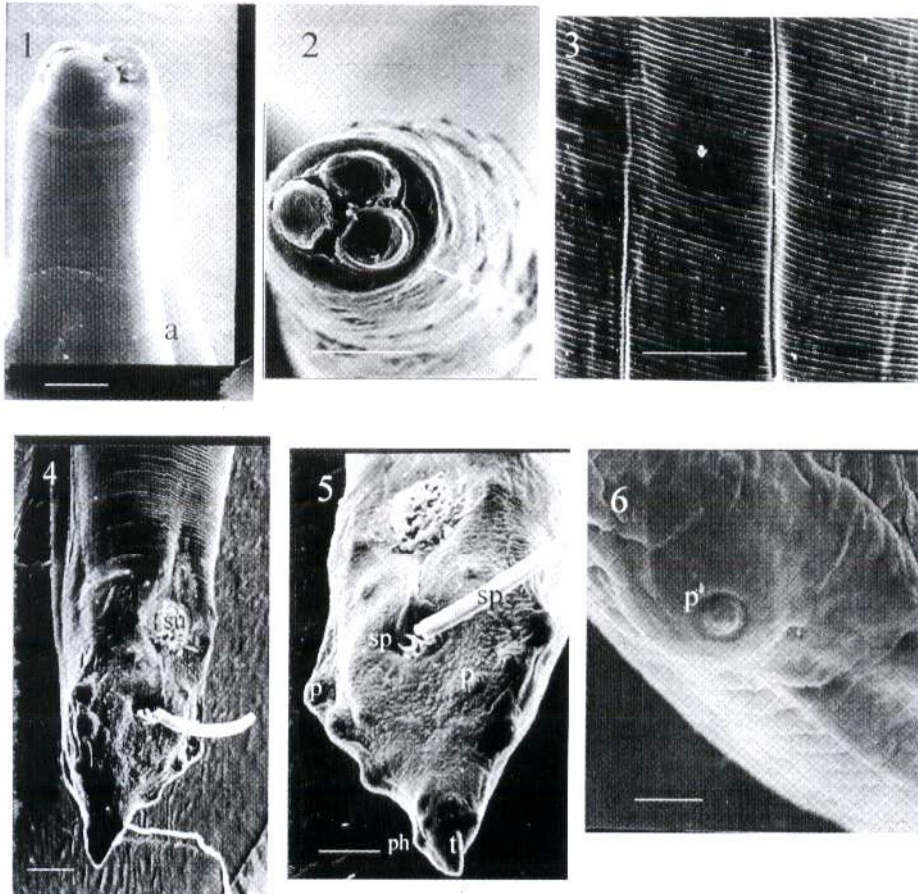
Fagerholm *et al.*, (1996) redescribed *Contracaecum magnipapillatum* from the proventriculus of *Anous minutus* from the Great Barrier Reef.

The present material is similar to Fagerholm's specimens in the main characteristics but the present work added more details about the structure of cloacal papillae and subterminal papillae. Moreover, certain minor differences in the body length and its range of distribution extends into Lake Wadi Al-Raiyan in Egypt.

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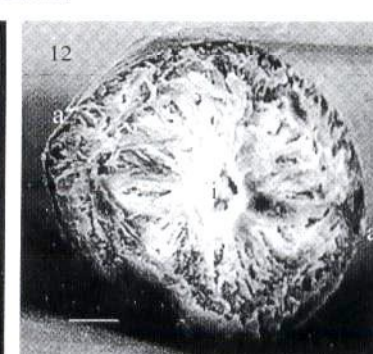
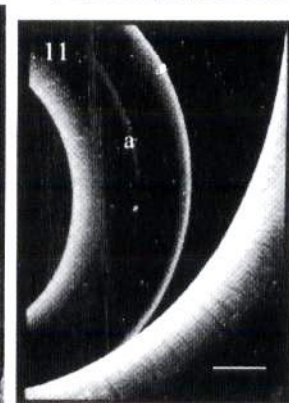
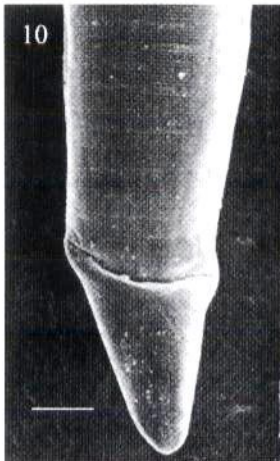
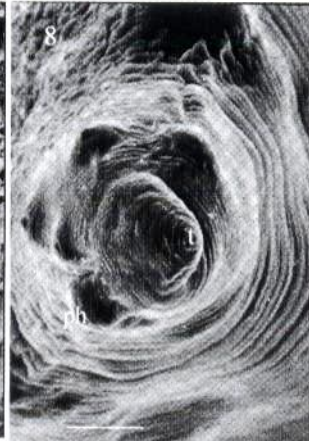
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Scanning Electron Microscopic study on *Contraecum magnipapillatum* (Nematoda: Anisakidae) from cormorants in Wadi Al-Raiyan Lake area, Fayoum, Egypt

46



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EXPLANATION OF FIGURES

- Fig. 1 : Scanning electron micrograph of the anterior region of *Contracaecum magnipapillatum* a, Alae. Scale bar = 250 μ m.
- Fig. 2 : Scanning electron micrograph of *Contracaecum magnipapillatum* en-face view showing the three lips. Scale bar = 412 μ m.
- Fig. 3 : Scanning electron micrograph of *Contracaecum magnipapillatum* showing transverse striations, c, cuticular ridge. Scale bar 15 μ m.
- Fig. 4 : Scanning electron micrograph of the posterior region of male *Contracaecum magnipapillatum* showing spicules, su, ventral sucker like-shaped. Scale bar = 200 μ m.
- Fig. 5 : Enlarged Fig.4, p, papilla; sp, spicule; ph, Phasmid. Scale bar = 105 μ m.
- Fig. 6 : Enlarged Fig.5 showing papillae, p, papilla. Scale bar = 20 μ m.
- Fig. 7 : Enlarged Fig.6 for showing the structure of papillae. p, papilla. Scale bar = 6 μ m.
- Fig. 8 : Scanning electron micrograph of the posterior extremity of male *Contracaecum magnipapillatum* showing terminal end. ph, Phasmid; t, terminal end. Scale bar = 30 μ m.
- Fig. 9 : Scanning electron micrograph of *Contracaecum magnipapillatum*, posterior extremity of fourth-stage larvae. an, anus. Scale bar = 490 μ m.
- Fig. 10 : Scanning electron micrograph of *Contracaecum magnipapillatum*, posterior end of female ventral view. Scale bar = 260 μ m.
- Fig. 11 : Scanning electron micrograph of *Contracaecum magnipapillatum*, lateral view showing alae. a, alae. Scale bar = 264 μ m.
- Fig. 12 : Scanning electron micrograph of *Contracaecum magnipapillatum*, transverse section showing intestine. i, intestine; a, alae. Scale bar = 74 μ m.

دراسة بالمجهر الالكتروني الماسح على دودة كونتراسيكم ماجنيبيايلايم
(نيماتودا: انيساكيدي) من طائر الغاق في منطقة بحيرة
وادي الريان بمحافظة الفيوم بمصر

ديهوم عبد الحميد الباسل

قسم علم الحيوان - كلية العلوم - جامعة الفيوم - الفيوم

تم في هذه الدراسة عزل الطفيلي كونتراسيكم فاريجاتم (نيماتودا : انيساكيدي) من قانصة طيور الغاق المهاجرة والمتغذية على الاسماك والتي تسمى محليا بالعنز ، و تم صيدها من منطقة بحيرة وادي الريان في الصحراء الغربية بمحافظة الفيوم بمصر . وكانت الاصابه شديدة حيث سببت للتهاب حاد في القانصه . وهذه النيماتودا تسجل لأول مرة من تلك المنطقه . وهذه المعلومات المورفولوجيه تعتبر اساس للدراسه المقارنه للانواع الاخرى لنفس الجنس . وقد وجدت الاصابه في اثنين فقط من بين خمس عينات تم فحصها . وقد تم دراسة السطح الخارجى بالميكروسكوب الالكترونى الماسح ، وقد اظهرت مزيدا من الصفات التصنيفيه مثل وجود ثلاث شفاقواحدة ظهريه واثنين بطنيان وهذه الشفاه مزودة بثلاث حلقات عنقيه واثنين من الامفيد ، وقد اظهرت الدراسة ايضا ان الجسم مغطى ببشبات جلديه مستعرضة كما اظهرت ما يشبه الممص على السطح البطنى للذكر على مقربة من الشرج (فتحة المجمع) . وكذلك وجود العديد من الحلقات الشرجية يقع زوجان منها أمام الشرج وزوجان على جانبي فتحة الشرج وزوجان خلف الشرج .

كما يوجد نوع اخر من الحلقات يعرف بالحلمات الطرفية منها زوج جانبي وزوج اخر بطنى .

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