

Larval Eustrongylides (Nematoda: Dioctophymatidae)
Occured in Rice-Field Eels (*Fluta alba*)

黃鱔體內寄生線蟲Eustrongylides幼蟲之形態

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Abstract

In the investigation of diseased rice-field eels (*Fluta alba*) collected from a cultured farm in vicinity of Taipei city, parasitic nematodes were found in almost 90% of examined fishes. The moribund fish show a symptom of redden and swollen in outer appearance of ventral side of the body. Inflammation and hemorrhage in the digestive tract were invariably noticed in dissection. The nematodes found in the intestinal lumen were all unencysted, they can invade to the body cavity through the intestestinal wall and encystment in mesentry close to the intestine. According to a SEM observation, there are 12 cephalic circumoral papillae arranged in two circles, six in each circle. The nematode is classified as a second larval stage of *Enstrongylides sp.* Oligocheta may be the first intermediate host and some species of aves the final host.

Introduction

Rice-field eels (*Fluta alba*) culture had been tried in vain for many years. Many presumptive reasons were suggested to explain the failure of the culture, but none had been confirmed yet. Among the suggestions, infectious disease apparently is the exciting cause. In many epidemic cases, the eels show symptom of redden and slightly swollen in ventral side of the body, inflammation and hemorrhage in the digestive tract were invariably noticed in dissection. Preliminary study revealed that both bacteria and were parasite involved in causing this symptom and the subsequent mortality. Bacteriological investigation will be be discuss in another report, this paper describe the morpholigical and statistical characters of the parasite *Enstrongylides sp.* found in the intestine lumen and mesentry of the rice-field eels, and suggest a possible life cycle and the parasitic effect on fishes.

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Materials and Methods

Larval eustrongylides were collected from diseased ricefield eels (*Fluta alba*), that were obtained from a culture farm in vicinity of Taipei city. The juvenile rice-field eels used for culture in this farm were captured from rice-field of undefined districts. The body weight of the rice-field eels were in the range of 8-10 g: Parasites were removed from the eels and relaxed in 20% ethanol then fixed in AFA. Permanent specimen were prepared by cleared in glycerin-alcohol and mounted in glycerine jelly. Measurements of 25 parasites were made under 100X light microscope, except that body length were measured without magnification. Morphology without magnification. of the worms were observed by light microscope and SEM at 5 KV. to 20 KV. The specimen used in SEM study were prepared by fixed in 2.5% glutaraldehyde buffered with 0.2 M phosphate buffer at PH 7.1 for one hour and then postfixed in 1% buffered osmium tetroxides for another one hour. The fixed specimen were dehydrated in a serial concentrations of ethanol and acetone and critical-point dried in liquid CO₂. The dried specimen were then sprinked on to a stub bearing adhesive and coated with gold.

Results

The larval Eustrongylides found unencysted in the intestine lumen (Fig. 2). Number of 1 to 16 worms were found in a fish. Usually the infected fish could be diagnosed easily by the slightly swollen and reddened outer appearance of the ventral side of the body. Not unoften, the parasites could be found transpassing the intestinal wall, and invading outside into the coelom, so that part of the worm was inside the intestine and part was outside. Sexuality of larvae examined were either female or unable to identify. The larval worms are large size nematodes, total length up to seventy's mm (table 1). Body cylinder-shaped, tapering towards the anterior end (Fig. 3). Living or fixed worms are rosebeiges color in the posterior end, opaque yellow in the anterior end. Cuticle relatively transparent, without spines, transverse striations of cuticle not prominent. Oral opening slit form, lacking lips. 12 cephalic papillae arranged in two circles, around the circumoral position (Fig. 7). Each circle with 6 papillae, two lateral and four submedian. The inner circle papillae spine-like in form, having ten annules, total height is about 6 μ (Fig. 8). The outer circle papillae bottom form, with wider base and lower height than the inner circle papillae (Fig. 9). Esophageal glands just between the end of oral cavity and the beginning of esophagus (Fig. 5). Bucal cavity narrow. Nerve ring just posterior to the start of esophagus (Fig. 5). Three-lipped cardiac valve at junction of esophagus into intestine (Fig. 3). Anus medial terminal. Ovary single, slightly coiled, arising in posterior half of body, extending anterior to about middle of body. Oviduct and uterus not yet distinguishable from ovary in all examined specimens. Vagina terminate to a position closed to anus. Measurements of the larval Eustrongylides shown in Table 1: Taxonomy according to Yamaguti S. (1961), the parasite is belong to Order Dioctophymidea, Family Dioctoph-

ymidae, *Eustrongylides* Jagerskjold 1900. According to Inglis W.G.(1983), it is belong to class Enoplea, Subelass Dorylaimina, Order Dioctophymatida, Family Dioctophymidae, *Enstrongylides* Jagerskjold 1900.

Table 1. Measurements of Larval *Eustrongylides* sp.
from Rice-Field Eels (*Fluta alba*)

Morphological characters	Range	Mean
Total length	40.0-78.0mm	55.8mm
Buccal cavity length	91-217 μ m	133 μ m
Anterior body width (at nerve ring level)	126-217 μ m	160 μ m
Maximum body width (at junction of esophageal to intestine level)	420-609 μ m	505 μ m
Esophageal length	8.0-17.5mm	11.6mm
Esophageal width	245-420 μ m	333 μ m
Nerve ring to anterior end	168-329 μ m	231 μ m

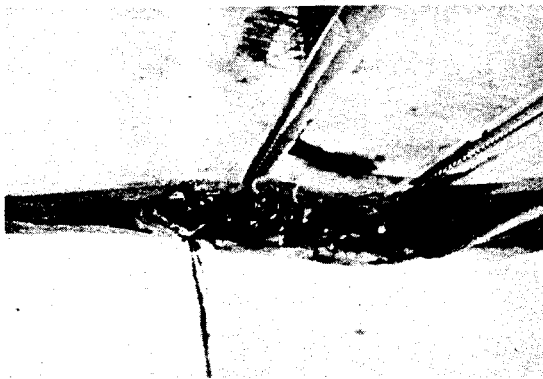


Fig. 1.

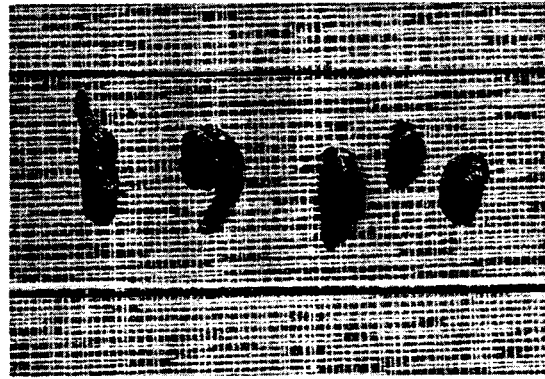


Fig. 2.



Fig. 3.

Figs. of Larval *Eustrongylides*:

- Fig. 1. Parasitic on rice-field eel(*Fluta alba*) intestine.
2. Encyst in mesenteric connective attached to intestine.
3. Junction of esophagus into intestine.

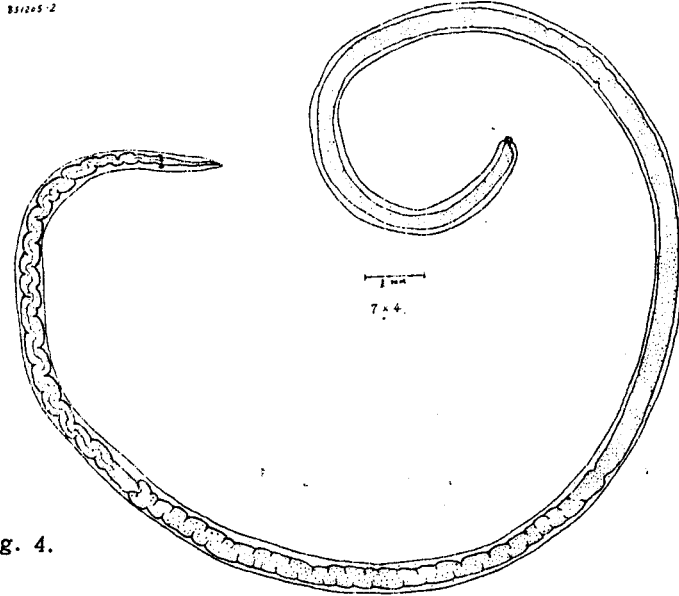


Fig. 4.

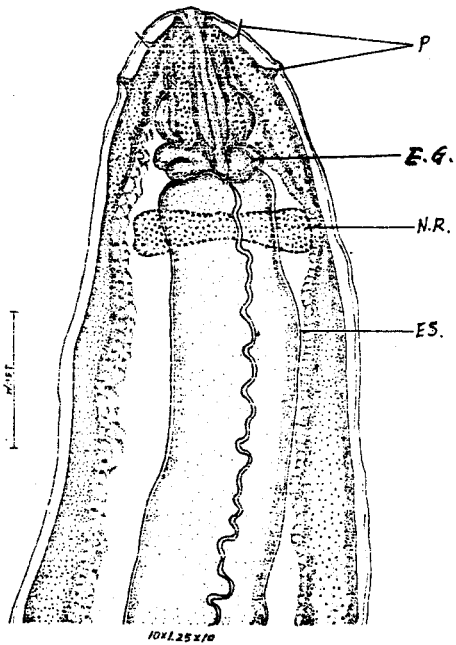


Fig. 5.

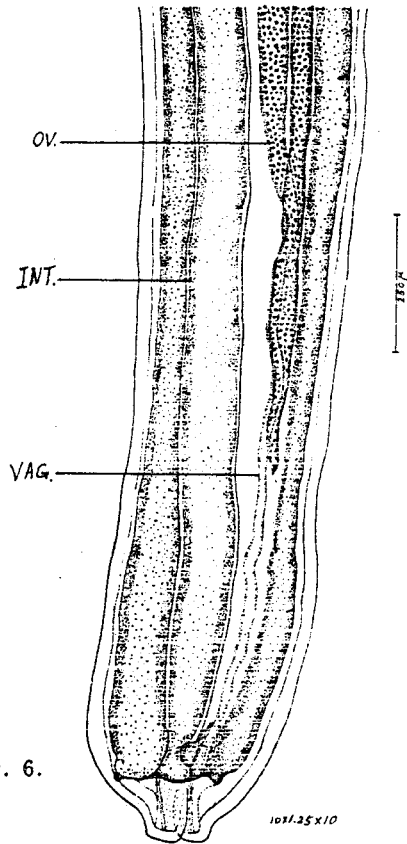


Fig. 6.

4. Diagram of entire worm.

5. Diagram of anterior end.

P.: popillae, E.G.: esophageal gland, N.R. : nerve ring, ES:esophagus

6. Diagram of posterior end.

OV: ovary, Int: intestine, Vag: Vagina

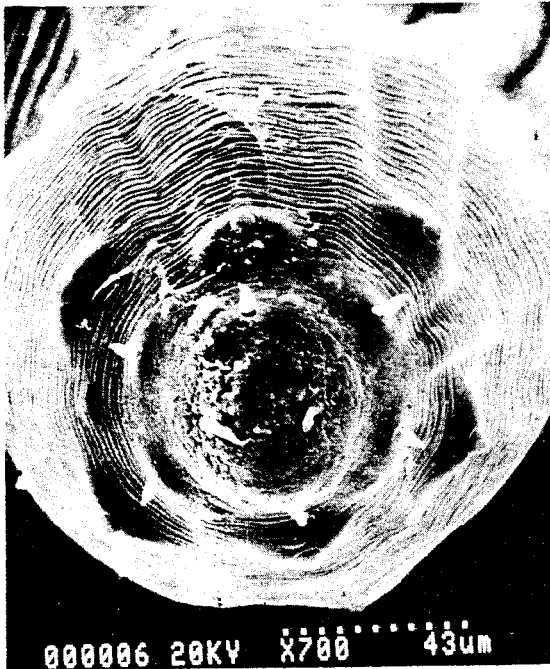


Fig. 7.

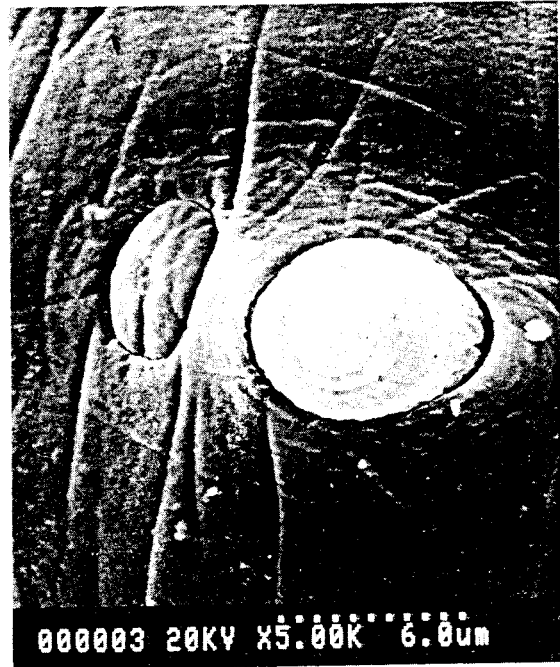


Fig. 8.



Fig.9

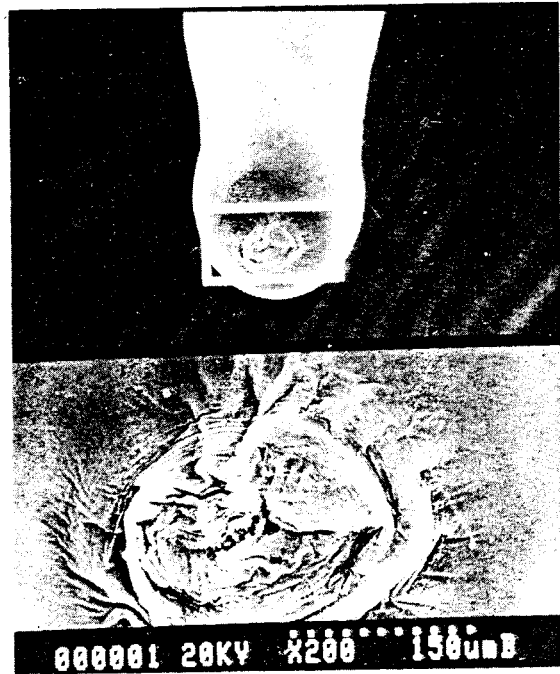


Fig. 10.

7. oral opening and cephalic circumoral papillae.
8. Inner circle cephalic circumoral papillae, spine form.
9. Outer circle cephalic circumoral papillae bottom form.
10. A. posterior end to show the anus.
B. Enlargement of the anus showing a Y shape opening.

Discussion

Fish nematode fauna is considered to be limited, in comparison with other vertebrate nematode fauna. Only 17 families of nematode occurred in fishes and only 5 are unique to fishes (Anderson, 1983). Among the two groups of fish nematode, philometrid nematode most often infect fishes, as a final host, in contrast, enoplid nematodes less occurred in fish, if it did occur fishes are only act as secondary or tertiary intermediate host, and not the final host, as in the philometrid nematodes.

In the piscine host, larval Eustrongylides may enter the host and then invade into various tissues or cavities (Burse, 1982; Cooper et al 1978; Kennedy & Lie 1976, Sprinkle-Fastzkie & Crites, 1977). It usually cause a hyperplasia of connective tissue, resulting in encystment. Most encystment occurred in mesenteric connective tissues (Cooper et al., 1978). In this case, the fish apparently infected by ingestion of the larval parasites, because unencysted worms were found in the intestinal lumen, the parasite then move through the intestinal wall and encysted in the mesentery close to intestine. Life cycle of this parasite is incompletely known, three or more hosts may be involved. The first larval stage is found to be fresh water oligochaeta in many investigations (Kennedy & Lie, 1976; Paperna, 1974; Shirajian et al 1984). Rice-field eels are benthic feeding in habit, very probable that fishes were infected by ingestion of oligochaete carrying a first stage larva of the parasite in it.

The parasites cause great damage, when it moved out from the intestinal lumen to the coelom, and as many as more than ten worms were found in a fish, a serious or even lethal effect imposed to the infected fish is obviously, in spite of conflicting result as to the pathogenicity of larva in fish had been reported (Kennedy & Lie, 1976). Pathogenic *Aeromonas hydrophila* was also isolated from the infective fish, it is thought that the parasite not only act as a primary pathogen but also induced a secondary infection of motile aeromonads. Similar phenomenon had been reported (Shirajian et al; 1984).

Although *Eustrongylides* sp. had been found in catfish, yellow perch and mullet etc. in mainland China (Hsu, 1933; Liu & Wu, 1941). but it had never been found in fishes of Taiwan. Meyers & Kuntz (1962). reported nematodes from fishes, amphibia, reptiles and birds taken on Lan-Yu, Taiwan. but none were belong to Eustrongylides. A presumptive suggestions of the hosts in life cycle of the parasites would include freshwater oligochaetes, rice-field eels and avian animal (include chicken and duck). In lacking of adult worm specimen, a speciation of the parasite become impossible in the present study.

Because of potential infection of the parasite to other animals including human being had been reported (Shirajian et al., 1984). further study of the ecology and life cycle of the parasite is not only favourable to aquaculture of rice-field eel in large scale but also important to hygiene.

中文摘要

黃鱔養殖失敗，感染性疾病的影響可能是最重要的原因。本省養殖之黃鱔常見由下頰至肛門之腹部皮膚出現輕微之淤血變紅或凸腫，解剖魚體可見消化道有炎症及淤血出血之現象，腸管內有線蟲寄生，此線蟲可穿過腸壁在緊臨腸壁外之腹腔繫膜 (Mesentery) 上形成囊泡 (cyst)，引起嚴重病變，嚴重時會導致黃鱔死亡。

此線蟲為其生活史中之第二幼蟲期，體長約 40~80mm。體前端之口周圍有12個頭突 (cephalic papillae)，排成二圈，每圈 6 個由掃描式電子顯微鏡可清晰看見其微細構造，此線蟲在分類學上應屬於胃瘤線蟲屬之未定種 (*Eustrongylides sp.*)。本線蟲之第一幼蟲期可能存在於淡水種之寡毛綱環節動物 (oligocheta)，而最終寄主可能存在於鳥、雞或鴨類中，尚待查明。

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