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PARASITOLOGICAL ASPECTS OF *LANISTES CARINATUS* (Olivier, 1804, Cyclostoma) SNAILS AND THEIR ROLE IN TRANSMISSION OF PARASITIC DISEASES TO ANIMAL AND MAN

(With 7 Tables, 9 Figures and One Plate)

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الجوانب الطفيليه لقواقع ؛لانستس كاريناتس؛ ودورها فى نقل الامراض الطفيليه للانسان والحيوان

نصر معوض الباهي

فى سبع مناطق من محافظة كفر الشيخ وعلى مدار عام كامل تم دراسة الانواع المحتلفه للاطوار اليرقيه الموجوده فى قواقع؛ لانستس كاريناتس؛ وكذلك الطفيليات الموجوده فى القناه الهضميه للقواقع مع حساب نسب الاصابه على مدار شهور العام وكذلك فصول السنه الاربع. ويعد قوقع لانستس كاريناتس؛ احد قواقع المياه العذبه ذو الحجم الكبير واللون الداكن وفتحته تتجه نحو اليسار. وهو قوقع منتشر فى مصر فى اماكن عديده حيث المياه الساكنه فى المجارى المائيه الضحله. وعندما تمت عدوى البط البكينى بالسركاريا المتحوصله المتكونه فى قواقع تم تربيتها فى المعمل وعدوتها بالسركاريا النشيطه المنبعثه من القواقع المجمعه من الحقل امكن الحصول على الديدان الناضجه وغير الناضجه لطفيل ؛ اكينوباريفيم ريكريفاتم؛ وكذلك عند عدوى الفئران البيضاء باطوار النيماتود التى تم تجميعها من القواقع الحقليه امكن الحصول على الديدان الناضجه وغير الناضجه لطفيل ؛ أنجيوسترنجيلس كانتوننسيس؛. وكذلك تم عزل طفيليات ؛ اليوسوما؛ و ؛نيديدا؛ والتى امكن استخدامهما كطريقه للمقاومه البيولوجيه لقواقع المياه العذبه مثل قواقع البولينس والبيومفلاريا وقوقع ليمينيا. وايضا تم عزل طفيل ؛ هارباكتيكويدا ؛و؛ تريتاتوريم.

SUMMARY

The role of *Lanistes carinatus* in parasitic diseases transmissible between animal and man were studied. Their parasitic fauna of the mantel and gut were also defined. Redia & encysted metacercaria of *Echinoparyphium recervatum* and larvae of *Angiostrogylus cantonensis* were obtained from the

snail tissue. Two species of *Oligochaeta* (*Aleosoma* and *Naidida)* and one *Copepoda sp.* (*Harpacticoida*) were found in the mantel cavity and gut. The examined *L. carinatus* snails were collected from 7 localities at Kafr El-Sheikh Governorate, Egypt, during one year from June/96 to May/97. The identity of the larvae and cercariae recovered in *L. carinatus* were confirmed by bioassay in rats, ducks and fish. Albino rats were experimentally infected with the isolated *A. cantonensis* larvae where premature stages and mature worms were recovered 35&75 d.p.i from the brain and bronchial arteries respectively. Damage of the cerebral blood vessels, the cerebral hemisphere and medulla oblongata tissue with hemorrhages were observed in histological sections of the experimentally infected rats. Immature and mature *E. recurvatum* worms were recovered after 15 & 35 days from ducks infected by being fed on snail tissue containing encysted metacercariae *Aleosoma sp.* and *Naidida sp.* showing a promising value as a biological predators against *Bulinus truncatus, Biomhpalaria alexandrina* and *Lymnaea caillaudi* snails.

Key words: *Snails-Parasitic diseases-Anmals and Man.*

INTRODUCTION

Lanistes carinatus snails are large , operculated, left handed snails distributed all over Egypt. They have special ability to resist different adverse conditions.

For long time this snail was considered as non-pathogenic, but Rysavy et. al. (1975) described two types of cercariae (Xyphidio type 1 & type 2) shed from *L. carinatus* in Warak El-Arab, Giza, Egypt. In 1979 Yousif & Ibrahim recorded only one Xiphidio cercaria but morphologically different from the above. While Nada (1983) in Sharkia Governorate mentioned that no cercaria was shed from this snail sp.

Recently in 1993 Diab mentioned that one Xiphidio cercaria was shed from snails of Behyra Governorate, Egypt.

Other than the previous trematode cercariae Ibrahim & Yousif (1986), extracted nematode larvae from *L. carinatus* tissue and identify them as *Angiostrogylus cantonensis* larvae.

Angiostrogylus cantonensis is a neurotropic parasite of the pulmonary arteries of rat (Alicata 1988).The infective larvae invade the central nervous system carried in the blood after penetration of the intestinal wall and in the neural parenchyma growing to young adults.Worms invaded the cerebral vein and travel to the heart and pulmonary arteries where they mature (Rosen

et al. 1967). Oviposition of single-celled egg carried in blood to the lungs and digestive system embryonated within one-week (Bhaibulaya 1975).

It is worthy to mention that all of the previous identification depend mainly on the morphological characters of the obtained larvae. There was no biological confirmation related to produce the adult stages of these developmental larvae in vertebrate hosts.

The present study throws more light on:

- The rate of infection of this snail speces with different larval stages in Kafr El-Sheikh Governorate, Egypt.
- More accurate identification of different parasitic stages collected from the infected snails was through developing them to mature worms in suitable experimental hosts.
- Other parasites detected in the examined snails were tested as means of biological Control against some medically important fresh water snails.

MATERIAL and METHODS

1-Study sites:

Through separate work in one of Schistosoma research project in Kafr El-Sheikh Governorate, *L.carinatus* snail samples were collected from the banks of shallow irrigation canals, tertiary canals and different fresh water collections in seven localities representing different snail habitat in Kafr El-Sheikh Governorate Egypt. These localities include Mesier, Shinno, Abioka, El-Ryad, Sedi-Salem, Keline and El-Hamoul.

2-Snails:-

In co-operation with Schistosoma Snail Competing Office (Ministry of Health in Kafr El-Sheikh Governorate), nine hundred seventy *L.carinatus* snail of different size were collected from different water ways in the previously mentioned study sites during a year (June 1996 - May 1997), through one month visit for each study site. The snails were transferred directly to the laboratory. Identification was according to Brown (1994). The snails were reared in the lab, the laid egg masses were transferred to a new aquarium and kept to hatch to produce the new juvenile snails according to El-Bahy (1993).

3- Examination of snails:-

The field collected snails were kept for 24hr. in the laboratory in dechlorinated tap water. The snail examined by exposure to light and crushing technique according to Abdel-Ghani (1955) and El-Bahy (1993). In

the first method, individual snail was exposed in a suitable beaker to artificial light for ½ hr. daily. Different types of shed cercariae were collected, enough samples were fixed for identification while the rest were left for recording further development of these cercariae.

The snails were kept in the laboratory for breeding. At the end of the first week 50% from the collected snails were crushed. All of its internal organs and tissue were examined microscopically to collect the developmental stages of the parasites and the encysted metacercariae that may be found in their tissues (digestive glands, visceral hump, mantle and foot). Rate of infection by different parasites was recorded and the percentage of infection was calculated monthly and seasonally. Snail tissue containing encysted metacercariae were collected , counted and kept under water surface in refrigerator to test their development in experimental hosts.

Permanent mounted specimens were prepared according to Prichared & Krus(1982). These samples were identified according to Dimitrov (1990).

4-Preparation of Encysted metacercariae (EMC):

At the end of exposure time, infected snails were replaced back to the aquarium. Water containing large no. of shed cercariae were divided into three parts. In the first part pieces of Cellophane sheets were dropped . In the second part three medium size lab. bred *L. carinatus* snails were dropped. In the third part one *Clarias lazera* fish (80-100gm. body weight) was held with special precautions to facilitate penetration of the cercariae into its body. Finger-ling fish were brought and reared in the lab. till reach the recommended weight and before infection some of them were killed and carefully examined to ensure the absence of any EMC. The exposed snails and fish were kept under observation for one week. Snails tissues were macerated and examined. All fish were killed, the subcutaneous tissue and muscles were carefully examined microscopically.The present EMC in snails and fish were fixed, mounted and measured.

5-Calculation of EMC dose:

After maceration of the infected snails (Lab. infected and naturally infected). Foot was divided into small pieces, compressed between two glass slides, all EMC present were counted and prepared in identified doses to be used in further infection of the tested host after storing for one week in refrigerator.

6- Infection of ducks:

Twenty apparently healthy Bikini ducks, one day old were brought and reared in the lab. After one week, feces were examined. They were divided into three groups each of 6. The first group was infected with metacercariae

encysted in *L. carinatus* snails under lab. conditions.The second group was infected with encysted metacercariae collected from naturally infected *L. carinatus* snails. The third group was left as non-infected control group.Each duck (in 1st & 2nd group) was infected with 20 EMC by feeding on infected snail tissue according to Mouahid & Mone (1988). Half of each group were severed at 15 days post infection (dpi) and the other half at 35 dpi. The intestinal content and intestinal wall were carefully examined. The recovered worms were counted, worm burden was calculated. Mounted specimens were measured and identified according to Kiseliene & Grabda Kazubska (1990).

7- Collection of the nematode larvae from infected snails:

The macerated snail tissue containing nematode larvae were warped in a piece of gauze and left in hanged funnel containing warm saline resembling Baerman apparatus. The larvae were collected after 30 minutes settlement from the few drops of saline at the end of the funnel. The collected larvae were examined and identified according to Galves-Ovied and Oviedo (1986). They were separated into L1, L2 and L3. Total no. of larvae per snail were calculated.

8- Infection of Albino rats:

Ten Albino rats (100-150 gm.) were divided into 2 groups. Six rats were infected each with 10 L3 per os using rubber tube and 4 were left as a non infected control group. At 35 dpi half of each group was killed to recover the immature worms. At 70 dpi the other half of the infected and control groups were killed to obtain the mature worms according to Noda et al (1987). Brain, Lung, Heart and intestine of killed rats were examined for worms, eggs and larvae. Samples from brain of infected rats, were fixed in formaline 10%, processed for histological sections micron thickness, stained with H&E, then examined, according to Loker (1978).

9- Study the other fauna present in field collected snails:

There was no. of prey and predators present in the buccal cavity and mantle of field collected *L. carinatus* snails. They were identified according to Pennak (1853), Fauchald (1977) and Jamieson (1978). The most common ones (*Aleosoma sp.* and *Naidida sp.*) were tested as a mean of biological control against *Lymnaea cailliaudi*, *Bulinus trancatus* and *Biomphalaria alexandrina*. Ninty lab. bred, medium size *L.cailliaudi, B.truncatus* and *B. alexandrina*.30 each were prepared in nine separate aquaria, each of 10 snails.Ten *Aleosoma sp.*/snail were transferred to 3 aquaria (one from each snail sp.) Another 10 *Naidida sp.*/snail were transferred to another separate 3 aquaria (one from each snail sp.).The last 3 aquaria were left as a control

without predators. The aquaria were observed daily, water change and counting the no. of dead snails were done at 3, 5, 7 and 10 days.

RESULTS

Snail morphology and ecology:

Lanistus carinatus are large sized, fresh water snail, thick shell, their width more greater than high, with short spire and blunt apex.They are left handed and operculated. The shell consisted of four whorls separated by oblique, deep sutures. The shell was demarcated by distinct spiral lines. The body whorl was concave in the upper surface. The aperture was rounded, the umbilicus deep and perforated all over the shell except the upper lamina.The color was brownish-yellow to blackish-brown according to the size, the smaller the lighter, and the local burettes of the aquarium, the least dirty the lighter color, this as shown in Fig. (1). According to the width of snails they were grouped into three categories, small size group, (not more than 1.0 cm.), medium size group (1.0 - 2.9 cm.) and large size group (more than 3.0 cm.). Their measures ranges were recorded in Table (1). The most common infected snails were of large size group, rare of medium size and non of small size.

L. carinatus snails living in colonies is muddy, calm, shallow pits at the end of tertiary irrigation canal or beside the main canals where the water distract their edges. These areas are suitable for the life of mice and also favorable for swimming and feeding of ducks & geese.

Incidence and rate of infection of *L. carinatus* snails:

The study was carried out on 970 *L.carinatus* snails collected from 7 location (Mesier, Shino, Abioka, Ryad, S. Salm, Kelien and Hamoul) in Kafr El-Sheickh governorate, Egypt, along a year from June 1996 to May 1997.

Results in Tables (2 & 4) revealed that rate of infection of *L. carinatus* with rediae, sporocysts, encysted metacercariae and nematode larvae were high during spring season to reach 13, 13.3, 8 & 7.2% respectively. This was followed by Autumn and Winter seasons while Summer season showed the lowest rate of infection (1.5, 1.6, 0.2 and 0.8% respectively).

Table (3) and Plate (1) showing the infection rates of *L. carinatus* with rediae were 10.8% in Mesier followed by Shinno (9.1%), Keline (6.6%), Abioka (5.6%) Sedi-Salm (5.4%) El-Ryad (4.8%) and El-Hamoul (3.8%)

with the peaks during March, May, April, March, February, October & November in the above locality respectively.

The highest rate of infection with sporocysts were in Mesier (10.7%) followed by Shino (7.6%) while the lowest were in Keline (4.5%) & El-Hamoul (4.3%).The maximum infection rates occurred during April, March, in Mesier & Shinno respectively (Table 3& Plate1).

Infection of *L.carinatus* with encysted metacercariae were recognized in Mesier (7.5%) followed by Shinno (6.6%) with the maximum rate during March and April respectively (Table 3 & Plate1).

The rate of infection with nematode larvae were high in snails collected from Mesier (5.7%) followed by Keline (3.8%) with the maximum rate during January & February (Table 3).

Monthly infection rates of *L.carinatus* collected from each locality (Plate1) clarify that Mesier is the most common location with high infection rates, while El-Hamoul showed the lowest infection rate.

Morphology and identification of trematode larvae:

On exposing the field collected *L. carinatus* snails to artificial light two types of cercariae were shed. The first type of cercaria was (Fig. 2E) leptocercus, the tail slightly shorter than the body.The body measures 95-123 × 50-56 µm, the tail measures, 92-100 × 16-17 µm,. Oral sucker diameter 28-32 µm, Acetabulum diameter 25-32 µm, Stylet length 16-18 µm.The acetabulum lies posterior to the middle of the body. The intestinal caeca are relatively short and narrow. Two pairs of penetration glands infront of the acetabulum. A main excretory duct extending posteriorly on both sides to open in the excretory vesicle at the posterior extremity.

Cercariae did not encyst in vitro (on cellophane sheet). On dropping 3 medium size lab. bred *L.carinatus* snail in the water containing these cercariae marked decrease in the number of cercariae in the surrounding water was observed. Examination of these snails after this, resulting in encysted metacercariae present in the foot connective tissue and few number in the digestive glands. This observation was not recorded in the control group (not exposed to the cercariae).

The encysted metacercariae were oval, ovoid or rounded in shape (Fig. 2F), measured 90-120 × 60-65 µm. They were double walled & containing full mature embryo with dark blackish ball and vaculated portion.

Snails that shed this type of cercariae, when dissected after this, two forms of rediae were detected one form containing cercariae and the other not containing germinal balls (Fig. 2A,B&D). Both of them were elongated

cylindrical bodies measured 0.6-0.8X0.05-0.07mm. provided with oral opening, muscular pharynx and simple gut. Birth pore occurred slightly behind the head colar.There was 2 lappets at the lateral aspects of the posterior extremity.

Another type of leptocercus cercaria was observed after exposure of another field collected L.carinatus snails to artificial light. These cercariae have the following characteristic morphology. The body measured 85-110X65-78 µm. The tail was 71-82X23-26 µm. Oral sucker diameter 34-42 Um. Acetabulum diameter 24-27 µm. Stylet length 17-19 µm.The mouth is subterminal and the pharynx behind the oral sucker. Short narrow oesophagus bifurcate near the anterior border of the acetabulum. The blind intestinal ceaca end in front of the excretory vesicle. There are 3 pairs of penetration glands on either sides of acetabulum.The exact no.of flame cells could not be accurately determined. Two lateral excretory ducts directed backward to open in excretory vesicle (Fig. 3C).

On exposing of the shed cercariae to *Clarias lazera* (Cat fish), marked decrease in the number of cercariae present in the surrounding water was observed. When the exposed fish were killed in the second day, there was a large number of encysted metacercariae found in the sub-cuticle tissue and the muscles. The control non exposed group of fish were free from any encysted cercariae. The encysted metacercariae recovered from the infected fish were oval or rounded in shape occurred singly in the muscles or in numbers of closely contact cysts in the penetration site. They measured 55-60X50-55 µm, with double wall filled with mature embryo contain no vesicles (Fig. 3D).

It is worthy to mention that snails which shed this type of cercariae harboring two stages of sporocystes when dissected.The first stage was transparent mass with ill-defined shape, containing clusters of embryonic cells (Fig. 3A).The other stages were less defined in shape, thick walled, and containing fully developed leptocercus cercaria. When the infected snails were crushed, the sporocyst ruptured and cercariae were emerged in an active jerky movement with its strong tail (Fig. 3B).

Experimental infection of Ducks:

Results in Table (5) revealed that EMC of the the first described cercariae type developed in all experimentally infected ducks.Severing of 6 ducks at 15dpi revealed *Echinoparyphium recurvatum* immature worms (Fig. 4B).The immature worms were detected in both infected with either EMC prepared in the lab.from lab. bred snails or from field collected snails.

On severing of the remaining ducks at 35 dpi The mature *E. recurvatum* worms were recovered from all infected ducks.The condition was not recorded in the control non-infected groups slaughtered at the same time.

The recovered worms (Fig.4A,B&C) have the characteristic features *of E. recurvatum*. The recovered 15 days old worm were 3.2-4.0mm. long X 0.3-0.4mm. width. There was no testes, no ovary, with few lateral vitelline glands .

The recovered 35 days old worms were 4.9-5.1mm. long X 0.7-0.8mm. width.Ovary and testes still smaller size with thin coiled uterine tube and small common genital pore.Uterus contained no eggs. The lateral vitelline glands were well developed.The head collar of immature and mature worms have 45 spines of which 4 are corner spines on either side. The anterior end is curved ventrally and armed with spines anterior to the ventral sucker. The ventral sucker is 0.30-0.37mm. wide and situated at the first quarter of the body.

Identification of nematode larvae:

On the other side the collected nematode larvae from *L.carinatus* snails were identified as *Angiostrongylus cantonensis* larvae. The first larval stage measured 0.263-0.301mm. The second larval stage measured 0.342-0.356 mm. The first and 2nd larval stages provided with rahbditiform esophagus and has long straight tapered tail ending in sharp point (Fig. 5A).

The 3rd larval stage was 0.427-0.465mm. in length and 0.021-0.027mm. wide with filariform oesophagus. The tail tapered caudal, pointed end and straight. The excretory pore was on the anterior fifth of the body (Fig.5B).

Experimental infection of Albino rats:

Early mature *A. cantonensis* worms were recovered from the brain of infected rats 35 dpi. (Fig.7A).The recovered immature worms were filliform in shape. Males measure 15-18 mm. Long (Fig.5C) and female 17-22 mm. long. Bursa small in size with distinguished rays. Females with medium sized tail, no serration, no rings, vulva in the posterior third of the body.

Adult mature males were 17-20 mm. long (Fig.5D).The bursa was atypical, small size, and all rays distinguished. The ventral rays were fused for most of their length and the dorsal rays were stout with short terminal branches (Fig. 6A&B).

Adult females were 18-23 mm long. The vulva situated in the posterior third of the body or nearly subterminal in location (Fig. 6C&D).

Eggs were oval or spheroid in shape measuring 70-80 µm X 40-45 µm. They were operculated, double walled, containing single-celled embryo (Fig. 5E).

Histopathological studies of the brain tissue:

Histopathological studies of the brain of experimentally infected albino rats showed damage of the cerebral blood vessels owing to migration of young adult from the brain to the lung. Severe hemorrhage was the commonest picture accompanied the destructive behavior of worm movement.

There was severe damage of the cerebral hemisphere and medulla oblongata which appeared in the form of furrows in migration sties of the immature stages (Fig. 7b & Fig. 8A&B).

Other parasitic fauna of *L. carinatus* snails:

Examination of the buccal cavity mantel, buccal cavity and gut of *L. carinatus* snails.Two spp. of *Oligochaeta* (*Aeolosoma* and *Naidida)* and one *Copepoda sp.* (*Harpacticoida*) were found. *Naidida sp.* was elongated, the cuticle provided with a number of constrictions, tapered anteriorly and the posterior end was provided with a number of finger-like projections (Fig. 9A).They multiply by sectioning division into two separate anterior and posterior part. Each part developed at the same time to form a complete individual. Rapid multiplication giving rise to a large number of colonies of *Naidida sp. Aleosoma sp.* unilocular ciliate, Olegocheata member. The body is cylindrical, with tapering anterior and posterior ends. Oral opening in the anterior end followed by simple gut without anal opening. The cuticle was rough and partially covered with cilia (Fig. 9B). *Tritatorium sp.* the individual was formed of body and tail; the tail furcated at its end (Fig. 9D). Multiplication by cyst formation. The cyst was oval with two knobs and contained clusters of embryonic cells (Fig. 9C). *Harpacticoida sp.* is a copepode. The head was provided with two oval cuticular lamellae and anterior mouth parts and posterior segmented tail (Fig. 9E&F).

Studying the predatory effect of some *Olegocheates*:

The effect of *Aleosoma sp.* and *Naidida sp.* as biological predators were studied. Results displayed in table (6) showed that *Naidida sp.* are more powerful than *Aleosma sp.* in distruction of snails where the mean destruction rates were 50.34% for *Naidida sp.* while it was 40.34% for *Aleosoma sp.*

The destruction effect of *Naidida sp.* on different snail spp. appeared as 70% on *L. cailliaudi*, followed by 60% on *B. truncatus*, while the lowest effect was on *B. alexandrina* (30%). By the same way *Aleosoma spp.* effect

was high on B. truncatus (60%) followed by *L. cailliaudi* (50%) and *B. alexandrina* (20%).

DISCUSSION

Snail morphology and size were described after classifying them into three categories; small, medium and large sized shell. Similar to that described by Nada (1983) who classified snails depending on the shell diameters. Ecology, distribution and climatic conditions of *L.carinatus* snails were not previously described in Egypt.The same conditions as shown in this study were mentioned by Liang-Houkuen et al. (1983) in Guangzhou, China. The highest infection rates among snails populations with different parasitic larvae were during Summer season while the lowest was during the Winter season. Moderate infection rates appeared during Autumn & Spring seasons. Part of these observations came in agreement with the work of Ibrahim & Yousif (1983) in Assiut, Beni Swif and Behyra governorates, Eygpt when they studied the infection rates of *L.carinatus* with *A. cantonensis* larvae. Noda et al. 1987 mentioned that in Yoron, Nansei Islands, Japan, prevalence of *A. cantonensis* in snails was low in August & increased gradually to April, the intensity was maximum in June. Among the examined localities snails collected from Mesier showing the highest infection rate followed by Shinno while El-Hamoul showed the lowest rate of infection. This may be due to the nature of the soil and water salinity as well as methods of irrigation and the types of cultivated crops.

Concerning cercariae shed from *L. carinatus* snails, the first type was defined in this study as *E. recurvatum* cercaria after bioassay in ducks and recovery of the adult worms. This type of cercariae slightly resembled those described by Yousif and Ibrahim (1979) from *L. carinatus* in Giza, Eygpt, but there was a slight difference in measurements. Rysavy et al.(1975) described two types of xiphidio cercariae from *L. carinatus* in Warak-El-Arab,Giza, Egypt. These cercariae differed in body dimensions, number, shape and position of penetration glands and the shape of excretory vesicle.

According to the morphological features of the second type of cercariae, which encysted in fish muscles, there was no authors who previously worked in Egypt *on L. carinatus* snails described any cercaria resembling those described in this study. Diab (1993) described slightly morphologically similar cercaria sp. shed from *Cleopatra sp* snails collected from Behyra governorate, Egypt, but they differ greatly in the body/tail measurement, size of acetabulum and penetration glands. The morphology of

the encysted metacercariae and their encysted sites in the foot tissue and the digestive glands resembled those mentioned by Harper (1929) who mentioned that cercaria were encysted in the digestive glands of the same snails and may migrate to other snails.

The present work proved that *L. carinatus* act as 1st. and 2nd. intermediate host for *E. recurvatum* where the rediae callected from naturally infected snails were recorded and described. The cercariae shed were identified and used in infection of lab. bred snails that recovered EMC. EMC produced immature and mature *E. recurvatum* worms in experimentally infected ducks. These results differ greatly from the work of Nada (1983) who mentioned that there was no cercariae shed from *L. bolteni* and there was no developmental stages in this snail tissue. He found only, few EMC in the snail foot and thought that *L. bolteni* acts as 2nd intermediate host for *E. vecurvatum*.

In the present study the author defined *L. carinatus* snails as intermediate host for *A. cantonensis* after experimental infection of Albino rats with the nematode larvae collected from naturally infected snails. The immature *A. cantonensis* worms recovered from the brain while the mature male and femal worms were recovered from the respiratory tract of experimental infected rats. The larvae were morphologically identical to those described by Uchikawa et.al. (1983) in Japan, who extracted 1st 2nd & 3rd larval stages from Biomphalaria glabrata. Campbell & little (1988) descried the morphology of L1, in fecal pellets of experimentally infected rats and use these larvae in infection of *L.carinatus* snails by consuming these larvae and then collected L_2 & L_3 larvae. In Egypt. Ibrahim & Yousif (1983) mentioned bat *L.carinatus* snails act as intermediate host of *A. cantonensis* on the basis of the morphology of the larvae collected from naturally infected snails, without evidence of experimental infection of rats or bioassay of these larvae.

In the present work extraction of the rediae and sporocysts from *L. carinatus* snails with their description and accurate identification of their progeny are mentioned for the first time.

Bioassay was the method adopted in the present work for accurate identity of larvae collected from *L. carinatus* snails. The recovered worms from the intestine of experimentally infected ducks with encysted metacercariae were morphologically typical to that described by Soulsby (1969) and Wang-Xiym (1985) who recorded worms in small intestine of domestic ducks with the anterior end curved ventraly and armed with forty five hooks and four corner spines. Kiseliene&Grabda-Kazubska (1990)

described *E. recurvatum* sp. With 45 spines in the basis of rediae and cercariae from naturally infected *Planorbis planorbis*. The collected number of flukes were small in comparison with the number of encysted metacercariae. This may be due to the method of infection, where losses may occur during ingestion ,or the reduced susceptibility of the duck species, or due to the age of the encysted metcercariae.

After experimental infection of Albino rats with *A. cantonensis* larvae, the premature adults of *A. cantonensis* recovered from the brain of experimentally infected albino rats and the adult worms recovered later on from the respiratory tract. This insure the identification of the collected larvae.This method was recommended by Higa et.al. (1986).Campbell & Little (1988) who infected rats by ingestion of raw intermediate and paratenic hosts. Also, Limaye et.al. (1990) mentioned infection of white rat by contaminated water and/or food.Yousif & Ibrahim (1978) recorded *A. cantonensis* worms in *Rattus rattus* in Egypt.

Infection in human was recorded in different countries in the world. Uchikawa et.al. (1986), in Japane recorded infection of man with *A. cantonensis* after eating of raw or incompletely cooked Ampullaridae snails. Anderson et.al. (1986), mentioned that *A. cantonensis* causes esinophilic meningitis in human. But there was no records in the ministry of public health in Egypt about the infection of human in these examined localities.

Immature worms were recovered from the brain and the mature adults from the respiratory system, this resembles the work of Platt & Harris (1985) in experimentally infected mice. The morphology of immature and mature worms resembled those described by He-Jingzhi et.al. (1983) and Andreson (1992).

The morphology of immature eggs collected by smears from lungs of experimentally infected rats were the same as that described by Higa et.al.(1986) who mentioned that eggs were oblong or spherical in shape and unembryonated.

Concerning other fauna of *L.carinatus* snails; This study identify *Naidida sp., Aleosoma sp., Tritatorium sp.* with its multiplication cyst and *Herpacticoida sp.* as naturally inhabiting *L.carinatus* snails with neglected effect on the snail life.These parasites were identified according to Pennak (1953), Fauchald (1977) and Jamieson (1978).

Naidida sp. and *Aleosoma sp.* were tested, as biological predators against the most dangerous intermediate host snails; *L. cailliaudi, B. truncatus and B.alexandrina.* Wald & Rayport (1977), described some *Oligocheates* as biological predators against planorbid snails.

From the present results we can consider both *Oligocheate spp.* as promising biologic predators especially if they have no paratactic behavior on other aquatic animals, birds or man.

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Table (1); Morphology and measurment of the collected *L. carinatus* snails collected from Kafr El-Sheikh governorate, Egypt.

Item	Small size	Medium size	Large size
Width	less than 1.0	1.10 - 2.9	more than 3.0
Hight	less than 0.60	0.70 - 1.2	more than 1.3
Spire	less than 0.30	0.31 - 0.50	more than 0.6
Aperature	less than 0.35	0.40 - 1.0	more than 1.1
Body whorl hight	less than 0.40	0.50 - 1.0	more than 1.1

Tabl (2); Total infection rates of *L. carinatus* snails during different seasons in different localities in Kafr El-Sheikh governorate from June1996 up to May1997.

Season	% of infection			
	Rediae	Sporocyst	E. mtacercariae	Nematode larv.
Summer	1.5%	1.6%	0.2%	0.8%
Autumn	6.5%	5.1%	4.0%	2.1%
Winter	5.3%	5.2%	5.0%	2.0%
Spring	13.0%	13.3%	8.0%	7.2%

Table (3): Total infection rates with different larval stages in the examined snails in different localities in Kafr El-Sheikh governorate from June1996 up to May1997.

Location	Rediae	Sporocysts	Encys. Metac.	Nemat.larv.
Mesier	10.8 %	10.7 %	7.5	5.7
Shino	9.1	7.6	6.6	2.7
Abioca	5.6	5.9	4.8	1.6
El-Ryad	4.8	5.8	1.8	3.0
S. Salm	5.4	5.8	2.6	1.7
Kelien	6.6	4.5	4.8	3.8
Hamoul	3.8	4.3	2.0	2.5

Table (4): Seasonal rates of infection of *L. craniatus* snails with parasites in different localities in Kafr El-Sheikh governorate from June1996 up to May1997.

Season	Localities	% of infection			
		Rediae	Sporocysts	Encys. Metac.	Nemat. larv.
Summer (Jun, Jul.& Aug. 1996)	Mesier	1.6	2.8	0.4	0.8
	Shino	2.0	2.0	0.0	1.2
	Abioka	3.2	2.5	0.0	0.6
	El-Ryad	0.0	0.0	0.0	0.7
	S.Salm	1.6	1.6	0.0	0.4
	Kelien	2.0	1.6	0.8	1.2
	Hamoul	0.0	0.7	0.0	0.7
Automn (Sep.,Oct.& Nov. 1996)	Mesier	9.7	7.1	5.8	3.2
	Shino	9.6	8.8	9.6	3.2
	Abioka	8.4	6.6	5.6	3.4
	El-Ryad	4.4	3.2	0.6	1.2
	S.Salm	4.7	3.5	1.7	0.5
	Kelien	5.8	3.2	3.4	2.5
	Hamoul	3.1	3.6	1.0	0.5
Winter (Dec.1996, Jan.& Feb. 1997)	Mesier	7.9	7.9	5.5	4.0
	Shino	8.0	7.0	5.5	2.7
	Abioka	2.5	3.4	6.0	0.0
	El-Ryad	5.6	7.8	4.4	2.2
	S.Salm	3.8	4.8	4.8	1.9
	Kelien	7.2	2.7	3.6	2.7
	Hamoul	2.0	3.0	5.0	0.0
Spring (Mar.,Apr.& May 1997)	Mesier	23.9	24.8	18.3	14.7
	Shino	16.9	11.3	11.3	3.8
	Abioka	8.2	10.9	7.6	2.1
	El-Ryad	9.2	12.3	2.0	8.2
	S.Salm	11.4	13.4	3.8	3.8
	Kelien	11.5	10.6	11.5	8.7
	Hamoul	9.9	9.9	2.1	8.8

Table (5): Experimental infection of one week old lab. breed bekeni ducks with encysted metacercaria.

Duck groups (6 each)	Source of encysted metacercariae and infection dose	Number of the recovered worms	
		15 dpi	35 dpi
First group (G1)	Field collected *L. carinatus* snails 10/Duck	7 worm /3 duck	6 worm /3 duck
Secound group (G2)	lab. infected *L. carinatus* snails 10/duck	8 worm /3 duck	5 worm /3 duck
non-infected control group (G3)	non-infected	0 worm/3 duck	0 worm /3 duck

Table (6): Experimentally infected Albino rats.

Albino rats		No. of recovered worms		worm / rat	eggs no.
serial no.	death day	Brain	Respiratory s.		
1	35 d.p.i.	7	0	7	0
2		6	0	6	0
3		3	0	3	0
4	75 d.p.i.	0	8	8	3
5		0	6	6	1
6		0	5	5	2
Non-infected control gp.					
7 & 8	35 dpi.	0 & 0	0 & 0	0 & 0	0 & 0
9 & 10	75 dpi.	0 & 0	0 & 0	0 & 0	0 & 0
Total		13	19	6.4	6

Table (7): Effect of some *Olegocheats* as biological predators.

Days past exposure	Snail spp.	Nidida spp.	Aleosoma spp.	Control gps.
		dead no./total (%)	dead no./total (%)	Dead no./total (%)
5 d.p.i.	*L. cailliaudi*	2/10	1/10	0/10
	B. truncatus	2/10	1/10	1/10
	B. alexandrina	1/10	0/10	0/10
7 d.p.i.	*L. cailliaudi*	2/8	1/9	0/9
	B. truncatus	3/8	2/9	0/9
	B. alexandrina	1/9	1/10	0/10
10 d.p.i.	*L. cailliaudi*	3/6	3/8	0/9
	B. truncatus	1/6	3/7	0/9
	B. alexandrina	1/8	1/9	0/10
Total no. of dead snail	*L. cailliaudi*	7/10 (70%)	5/10 (50%)	1/10 (10%)
	B. truncatus	6/10 (60%)	6/10 (60%)	1/10 (10%)
	B. alexandrina	3/10 (30%)	2/10 (20%)	0/10 00 %
Mean effect		50.5%	40.34%	

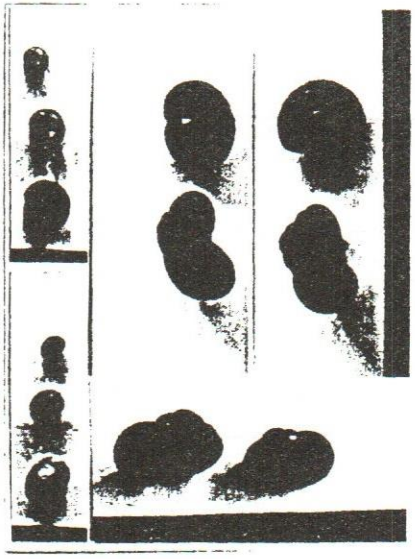

Fig. (1):
Lanistus carinatus snails different shell size; Large, medium and small, collected from Kafr El-Sheikh governorate.

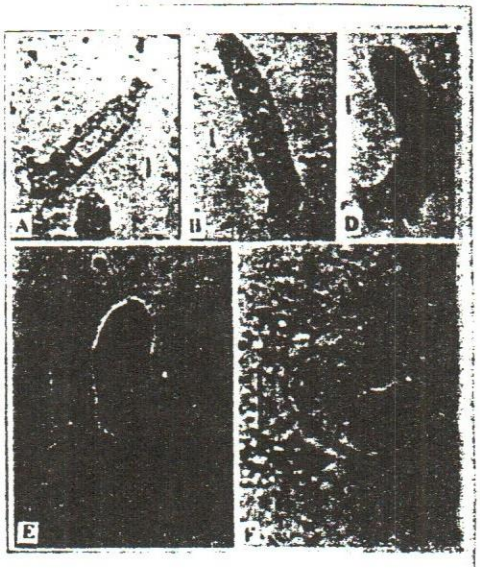

Fig. (2):
Developmental stages of *E.recurvatum* extracted from *L.carinatus* snails
A-Ventral view of well developed Redia
B-Lateral view of immature Redia containing embryonic cells.
D-Lateral view of mature Redia containing cercariae
E-Leptocercus cercaria.
F- Encysted metacercaria.
(Par = 0.05mm.)

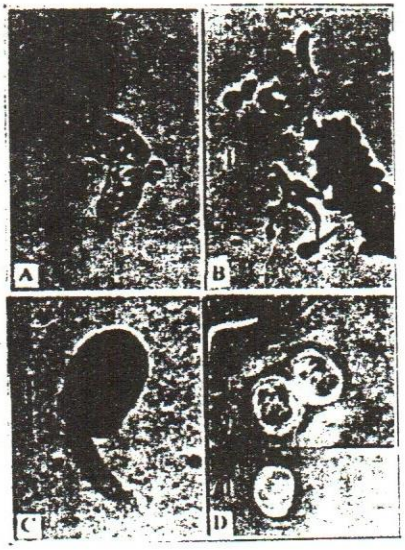

Fig. (3):
Developmental stages extracted from *L. carinatus* snails shed another cercariae species.
A- sporocyst with indifined wall and containing clusters of embryonic cells.
B- sporocyst with cereriae emerging from it.
C-Leptocercus cercariae.
D-Encysted metacercariae collected from experimen tally infected Catfish.
(Par = 0.05mm.)

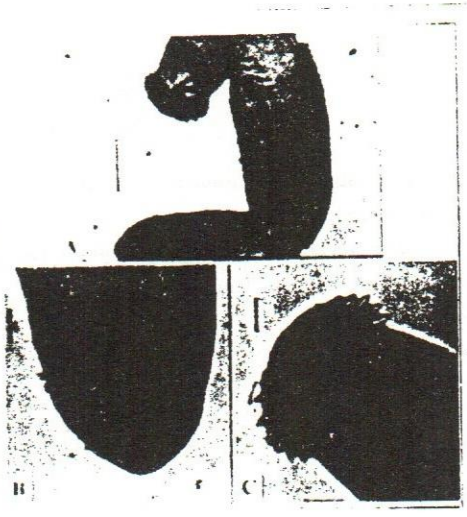

Fig. (4):
***E. recurvatum* worms recovered from experimen taly infected Ducks.**
A-Mature worm recovered 35 dpi
B-Worm 15 dpi., posterior half of the body (undevel oped Vitelline glands and gonads.
C-Head collar with 45 spines & 4 corner spines.
(Par = 1mm.)

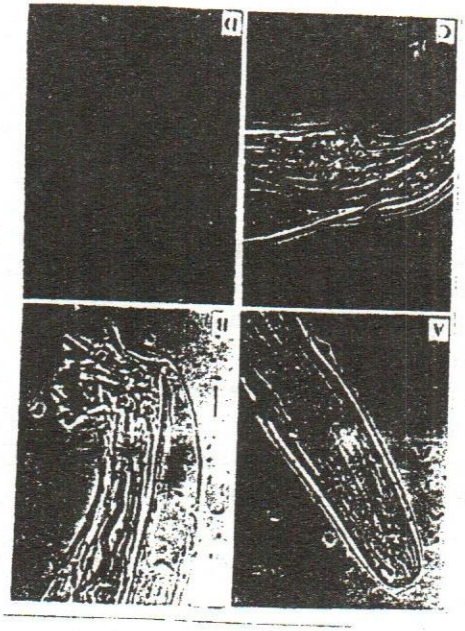

Fig.(5):

A-*A. cantonensis* first & second larval stages.

B-*A. cantonensis* Third larval stage extracted from L. carinatus snails.

C-Immature *A. cantonensis* worm recovered from the brain of experimently infected Albino rat 35 dpi.

D-Mature *A. cantonensis* worm recovered from the bronchial arteris of experimently infect Albino rat 70 dpi.

E-Egg of A. cantonensis obtained from respiratory tract of experimently infected Albino rat.

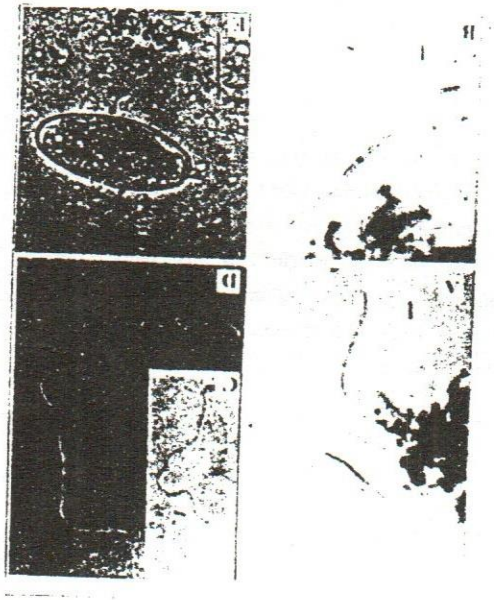

Fig. (6):

Adult male and female *A. cantonensis* worm.

A-Anterior end.

B-Posterior end of male showing copulatory bursa.

C-& D- Posterior end of female showing the vulva.

(Par = 1 mm.)

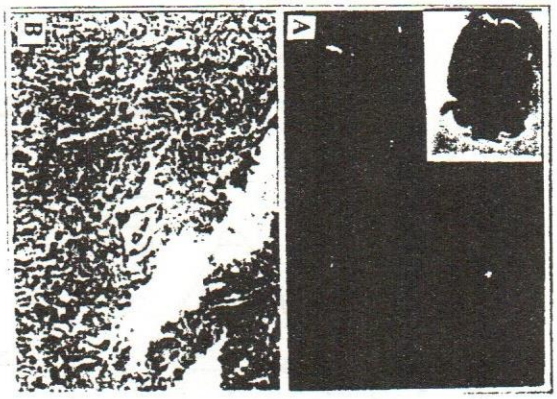

Fig. (7):
A-Macroscopic appearance of the brain of experimentally infected rat showing the immature *A. cantonensis* worms in between the cerebral hemisphere and medulla oblengata.
B-Histological section in the medulla oblongata showing distructed blood capillary (arrow).
(X40 H&E)

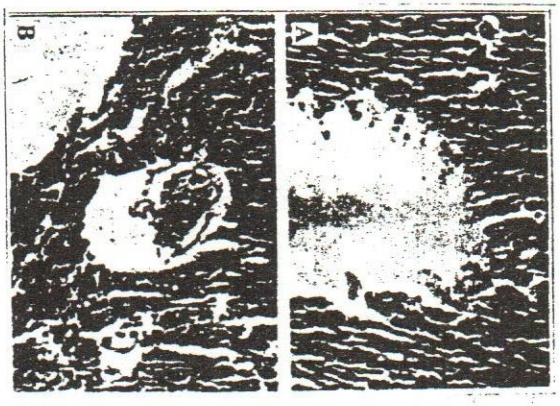

Fig. (8):
A-Histological section in the cerebral hemisphere showing damaged tissue and hemorrhage. (arrow)
(X40 H&E)
B-Histological section in cerebral hemisphere showing ruptured blood capillary and damaged tissue. (arrow) (X40 H&E).

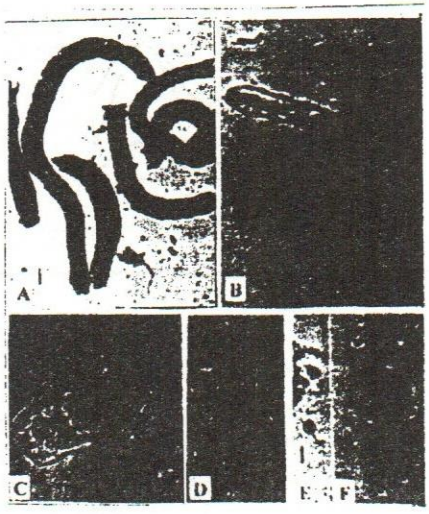

Fig. (9):
A-*Naidida sp.*
B-*Aleosoma sp.*
C-*Tritatorium sp.* cyst.
D-*Tritatorium sp.*
E & F- *Harpacticoida sp.*
(Par = 0.05mm.)

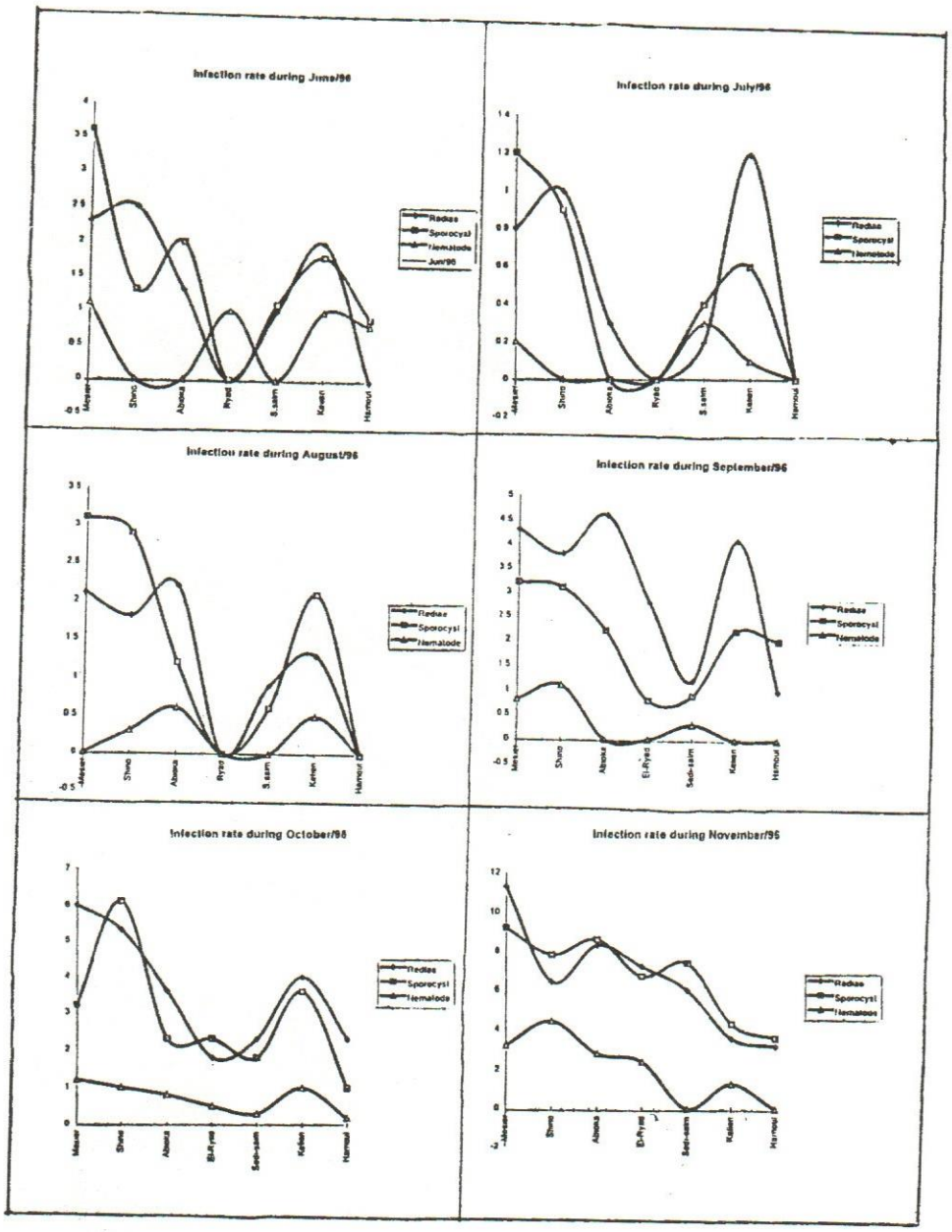

Plate (1):
Monthly infection rate among *L. carinatus* snails with Rediae, Sporocysts and *A. cantonensis* larvae.

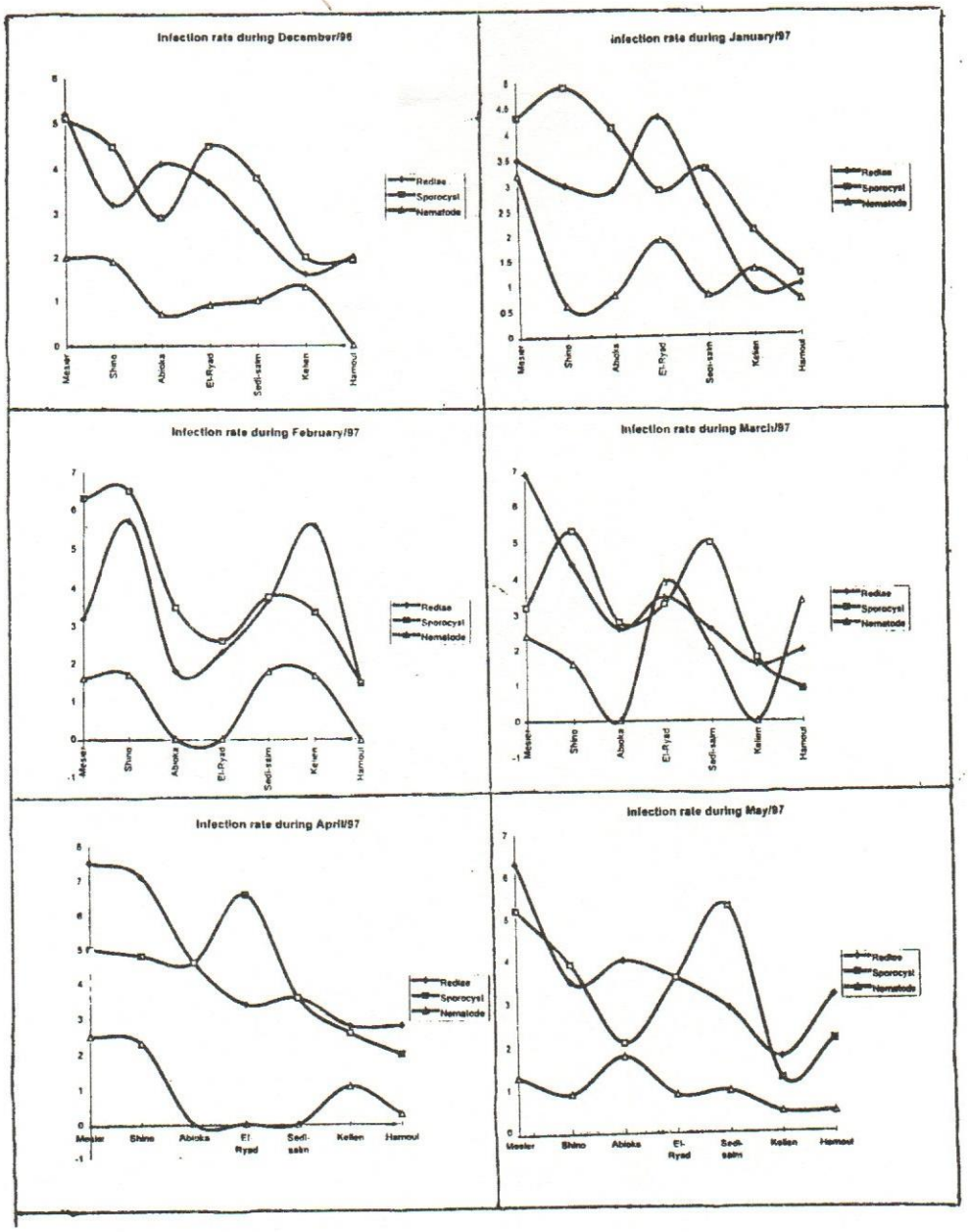

Plate (1) continue
Monthly infection rate among *L. carinatus* snails with Rediae, Sporocysts and *A. cantonensis* larvae.