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Biological studies on parasitic crustaceans infesting different fishes and their histopathological impacts

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دراسات بيولوجية على قشريات طفيلية تصيب أنواع مختلفة من الأسماك وتأثيراتها الهستوباثولوجية

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INTRODUCTION

Introduction

Invertebrates are animals without a backbone. They are an extremely diverse group, both in form and function. They inhabit, virtually, every type of environment found on the earth. They dominate the earth's biota, both in terms of numbers of species and biomass. Arthropods are animals covered with a jointed exoskeleton and form a major component of the invertebrate group. They include organisms such as spiders, insects and crustaceans. **Platnick (1992)** stated that biodiversity is essentially equivalent to speaking about arthropods and other animal species and plant groups are just a gloss on the arthropod theme.

Worldwide, more than 42,000 species of crustacea are known. They are divided between eight subclasses and include terrestrial, marine and freshwater forms. About 500 species of freshwater crustaceans are listed for Australia only. Due to the difficulty of identifying some crustaceans, it is unknown how many species occur in South Australia for example **(Hawking and Smith, 1997)**. Freshwater crustaceans such as: Daphniidae, Moinidae, Macrothricidae, Chydoridae, Sididae, Bosminidae, Sayciidae and Ilyocryptidae or microcrustaceans including Ostracoda (seed shrimps), Copepoda (copepods), Cladocera (water fleas) and Conchostraca (clam shrimps) are often small and many species from these groups tend to be planktonic **(Gooderham and Tsyrlin, 2002)**. In contrast, the crustaceans groups including Anostraca (fairy shrimps), Notostraca (shield or tadpole shrimps), Amphipoda (scuds or side-swimmers), Isopoda (water slaters or sow bugs) and Decapoda (freshwater crabs, crayfish, prawns and shrimps) are called macrocrustaceans. These animals tend to be larger and benthic rather than planktonic.

Generally, crustaceans are highly variable in body form but some general rules apply. All crustaceans have two pairs of antennae, one pair of mandibles and two pairs of maxillae on their heads. Characteristically, they also have a pair of appendages, laterally, on each body segment although sometimes these are reduced or absent from various parts of the body depending on the species. The appendages can be modified in shape to perform special tasks such as swimming, walking, feeding, respiration or copulation.

Crustacean bodies are composed of several segments generally between 16 to 60 segments. The first six segments form the head and the remainders make up the thorax and abdomen. Most crustaceans have a carapace and all have two pairs of antennae but their structures are greatly varied. The larger freshwater forms are very similar to their marine cousins and are easily recognised. The tiny microcrustaceans are harder to recognise and it may be difficult to see their features without a microscope **(Gooderham and Tsyrlin, 2002)**.

Crustaceans can be found in just about all kinds of waters-fast-flowing, still, fresh and saline. They can be found living in the water column, on the bottom of a water body, or among aquatic plants. Some are tolerant to pollution, while other species are intolerant and prefer clean water. Smaller crustaceans can be found in just about every water body. Some of the larger ones are less common or are found only in particular areas of the state. Adult crayfish have been known to roam the banks of creeks for a period of time and to move from one water body to another if conditions become unfavorable (**Martin and Davis, 2001**).

Small crustaceans are generally filter feeders, using specialized appendages to create a current of water that passes over the ‘filter’. The collected particles; including bacteria, algae and small planktons; are removed by special combing or brushing hairs and transported to the mouth. Larger forms feed by grabbing prey with their large front claws; they locate their prey by smell and take insects, mollusks and fish. Some larger crustaceans, including yabbies, eat plants. Whereas, a number of crustaceans are parasites, often during their larval stages.

Most crustaceans have separate sexes. Some reproduce parthenogenetically. Eggs may be attached to certain appendages or contained within a brood pouch or in an egg sac. The most common form of larva is the ‘nauplius’, which is free-swimming and has only three pairs of appendages the first, second antennae and the mandibles. All crustaceans moult and additional appendages are formed as the animal grows. Many of the smaller crustaceans have resting eggs that are resistant to desiccation but these are not found in amphipods, isopods and decapods. The life span of a crustacean can be between two weeks and 15 years, depending on the species **(Gooderham and Tsyrlin, 2002)**.

No group of plants or animals on Earth exhibits the range of morphological diversity as seen among the extant Crustacea (**Martin and Davis, 2001**). This structural disparity is best demonstrated by the Copepoda, which by virtue of their immense vertical distribution from the abyss to the high Himalayas, spanning three quarters of the possible vertical range on Earth are also arguably the most abundant metazoans (**Hardy, 1970; Huys and Boxshall, 1991**).

Occurrence of copepods:

The name copepod is derived from the Greek words *cope* meaning 'oar' and *podos* meaning 'foot" and literally means 'oar-footed'. This name refers to their broad, paddle-like swimming legs and forms the basis of their common name in other languages, such as the German 'Ruderfusskrebs', the Dutch 'Roeipootkreeft' and the Norwegian 'Hoppekrebs'. Copepoda is the largest subclass of small crustaceans and more over 8500 species have been recorded. Most species are marine, but there are many freshwater species that live in moss, soil-water films and leaf litter. Also, there are many copepods that are parasitic (over 1000) on various marine and freshwater animals, especially fish. Most copepods are ranging in length from less than 1mm to more than 5mm, although there are larger (17mm) free-living species. Some copepods are ectoparasitic on fish and attach to the gill filaments, fins or the integument. Other copepods are commensals or endoparasites of polychaete worms, bivalves, echinoderms and tunicates. Many copepod species are also parasites of Cnidarians. All degrees of modifications from the free living copepod form are exhibited by these parasites.

Ancestral forms are usually ectoparasites and resemble free-living species. On the other hand, some ectoparasites and endoparasites are so highly modified and bizarre that they no longer have any resemblance to the free-living species. In most parasitic copepods, the adults are adapted for parasitism and the free-swimming larval stages are usually similar to those of the free-living copepods. Contact with the host occurs at various times during the life cycle of the copepod and modifications appear with each molt (**Ruppert & Barnes, 1994)**.

Morphology of copepods:

To appreciate the extent of morphological changes undergo by parasitic copepods for adaptation to their mode of life, one should be acquainted with the morphology of the free-living copepod which could be concerned as the typical structure. The body of a free-living copepod is commonly tapered from anterior to posterior and is somewhat cylindrical. The trunk is composed of a thorax and abdomen. The head is either rounded or pointed. Compound eyes are absent, but the median naupliar eye is a typical and conspicuous feature of most copepods. Also, appendages are the uniramous first antennae which are generally long and held outstretched at right angles to the long axis of the body. The head is fused with the first thoracic segments and sometimes with the second thoracic segment as well. The first pair of thoracic appendages has become modified to form maxillipeds, used for feeding **(Ruppert and Barnes, 1994)**. The cephalothorax is followed by four free thoracic segments, each bearing a pair of swimming legs. These segments vary in their proportion lengths from species to species, but as a rule diminish from the second to the fifth leg bearing segment, the last of them being usually much reduced. This segment is followed by the genital complex and made up of two segments.

The presence of vestigial pair of legs on the genital complex is an indication of the thoracic origin of the genital complex as no appendages are carried on the abdominal segments **(Kabata, 1979)**. The abdomen is composed of five segments, which are commonly narrower than those of the thorax. There are no abdominal appendages, except for the anal segment bearing two caudal rami (Fig. 1).

Adaptation to the parasitic mode of life, with fish as hosts, has led to substantial changes in the morphology of the copepods. Those changes took the form of addition of parasitic developmental stages to the free-swimming copepod life cycles and are directed towards the attainment of two main objectives:

(1) Maintenance of secure hold to the host.

(2) Accommodation of increased reproductive activity.

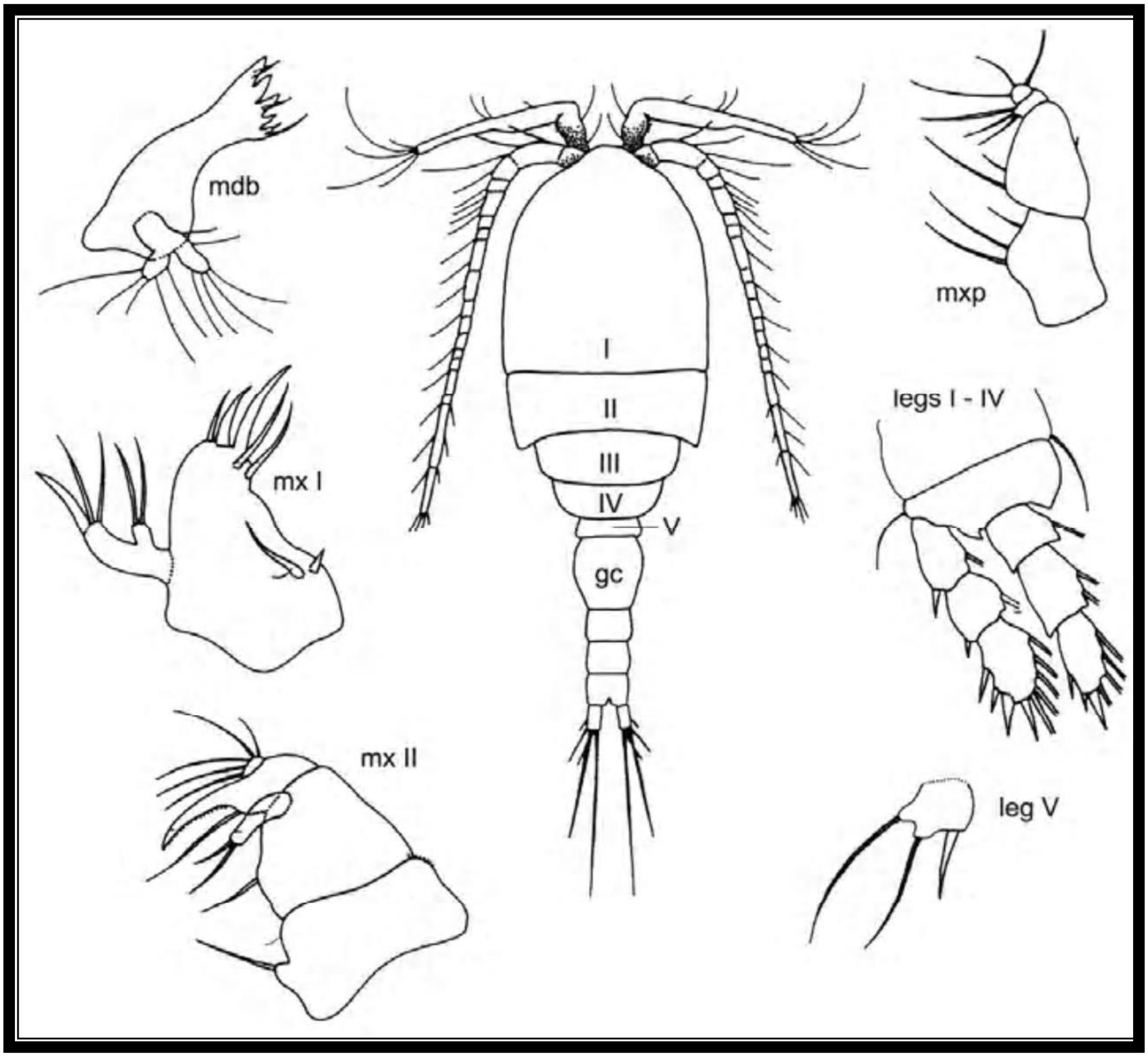


Figure 1: Schematic drawing showing the classic structure of free-living cyclopoid copepod; mandible (mdb), 1st maxilla (mx I), 2nd maxilla (mx II), maxilliped (mxp), 1st, 2nd, 3rd and 4th thoracic legs (legs I – IV), 5th thoracic leg (leg V), thoracic segments (I – IV) and genital complex (gc) **(**redrawn from **Ruppert and Barnes, 1994)**.

The first resulted in the development of various prehensile mechanisms, in some instances; extensive metamorphosis has been adopted as the mechanism transforming free-swimming dispersal stages into sessile parasitic adults **(Kabata, 1992)**.

More examples of modifications undergo by members of this taxon in order to adapt to the parasitic mode of life include: reduction in locomotory appendages; development of adaptations for adhesion both by development of new structures and modification of appendages; increase in size and change in body proportions caused by much greater growth of genital regions; fusion of body somites and loss of external evidence of segmentation; reduction of sense organs; and a reduction in numbers of instars that are free-living, both through the passing of more stages before hatching and through larval instars becoming parasitic **(Roberts and Janovy, 2005)**.

Out of ten copepod orders; Calanoida, Platycopioida, Misophrioida, Harpacticoida, Mormonilloida, Gellyelloida, Monstrilloida, Cyclopoida, Poecilostomatoida and Siphonostomatoida, only the latter three orders are parasitic on fishes. About 75% of parasitic species belong to Siphonostomatoida, 20% to Poecilostomatoida and only some of 5% to Cyclopoida **(Ho, 1990)**. According to **Kabata (1970)** classification of the three parasitic orders of Copepoda is partly based on morphology of their mouth parts (Fig. 2).

Members of the suborder Cyclopoida have the buccal orifice not projecting much above the surface of the body. It is usually a partly uncovered hole that anteriorly overhung by the flap of the upper lip. The cyclopoid mandible consists of two parts; the basal part is short and stocky and the distal part is falciform, long

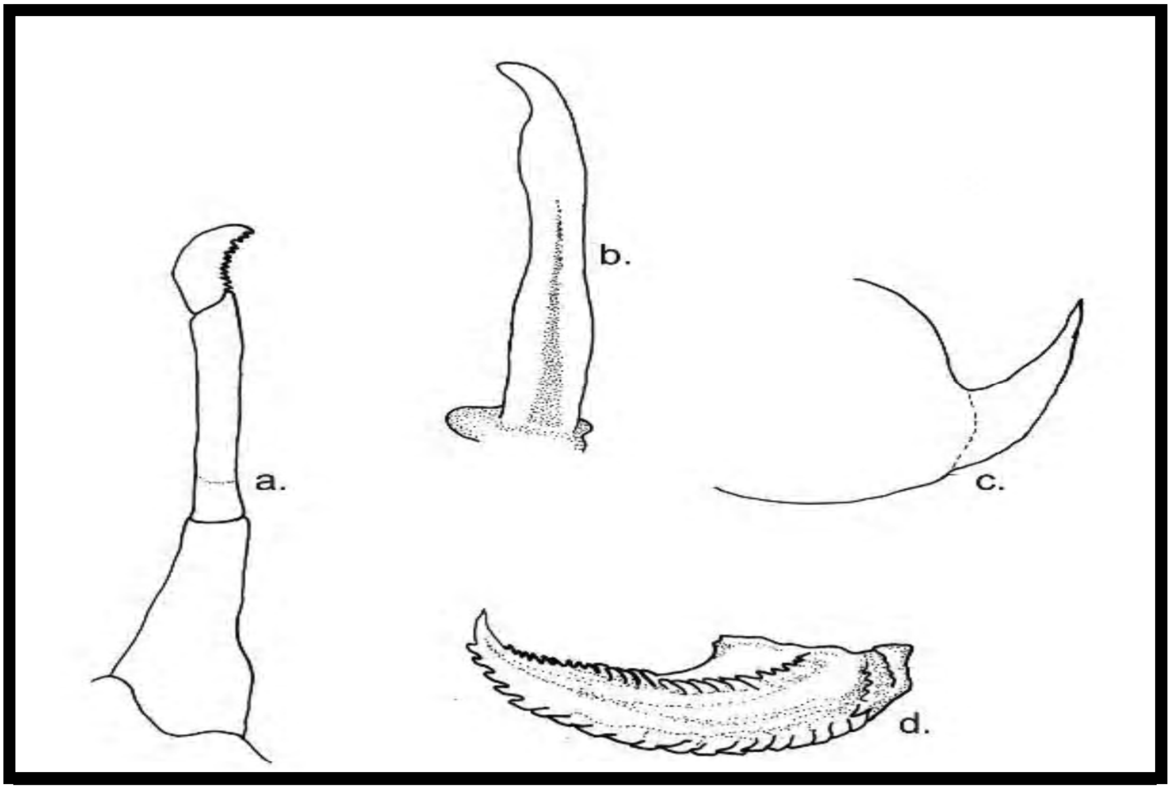


Figure 2: Schematic drawing showing the morphological features of the mandible in three parasitic orders; Siphonostomatoida (a&b), Cyclopoida (c) and Poecilostomatoida (d) members of subclass Copepoda, **(**redrawn from **Kabata, 1988**).

and pointed with rows of small denticles running along the entire or almost entire lengths of both its margins. This is apparently the ancestral condition and is possessed by several copepod suborders **(Kabata, 1970)**. Unlike the other two suborders, Cyclopoida has no authenticated records of parasitic species on marine fishes. A large proportion of cyclopoid species parasiting on freshwater fishes belongs to the family Lernaeidae **(Kabata, 1992)**.

Association between the morphological changes of the parasitic copepod and the type of host-parasite relationship determine the extent of the deleterious impact exerted by copepods on their fish hosts. A wide range of effects have been observed from relatively innocuous to lethal. Generally speaking, the damage caused to the fish by copepods is directly proportional to the degree of closeness of this relationship. Mobile ectoparasites tend to be less harmful than sessile, mesoparasitic species penetrating host tissue and in some cases invading vital internal organs. The final effects depend, however, on more than one factor and are influenced by the intensity of infection, the site of infection and often by environmental parameters such as temperature, oxygen content or salinity level of water **(Kabata, 1992)**.

Classification of copepods:

- Phylum Arthropoda
  - Subphylum Crustacea
    - Class Maxillopoda
      - Subclass Copepoda
        - 1-Order Cyclopoida
          - 2-Order Poecilostomatoida
            - 3-Order Siphonostomatoida

1-Order: Cyclopoida

The order Cyclopoida is a large group of copepods, most species of which are free-living. Free-living cyclopoids occupy important niches as primary consumers in many aquatic habitats, particularly freshwater. Several families of cyclopoids are parasites on invertebrates and the Lernaeidae is a highly specialized group of fish parasites **(Ho and Kim, 1997)**.

Family: Lernaeidae

Distribution and occurrence of species of the family Lernaeidae:

Lernaeidae is a major family of cyclopoid copepods parasitizing on fresh-water fishes. Some of them are among the most harmful parasites of cultured fishes; consequently, they have become the target for extensive studies **(Ho and Kim, 1997)**. **Kabata (1979)** stated that the family Lernaeidae contains genera displaying a broad range of morphological differences (Fig. 3).

At one extreme are species of *Lamproglena* **von Nordmann, 1832**. Its morphology bears many traces of its free-living ancestry. At the other are members of *Lernaea* L., the notorious 'anchor worm' that has been metamorphosed to the point of losing, at first sight, all indication of its resemblance with Crustacea **(Kabata, 1983)**. The genus *Lamproglena* is the most primitive member of the family Lernaeidae. Following attainment of maturity, the females increases in size only moderately and though their external segmentation is at least partially lost and does not show any significant tagmosis. All segments present, following the moult of the last copepodid stage, are still more or less recognisable and their proportions have been largely retained. The genital segment

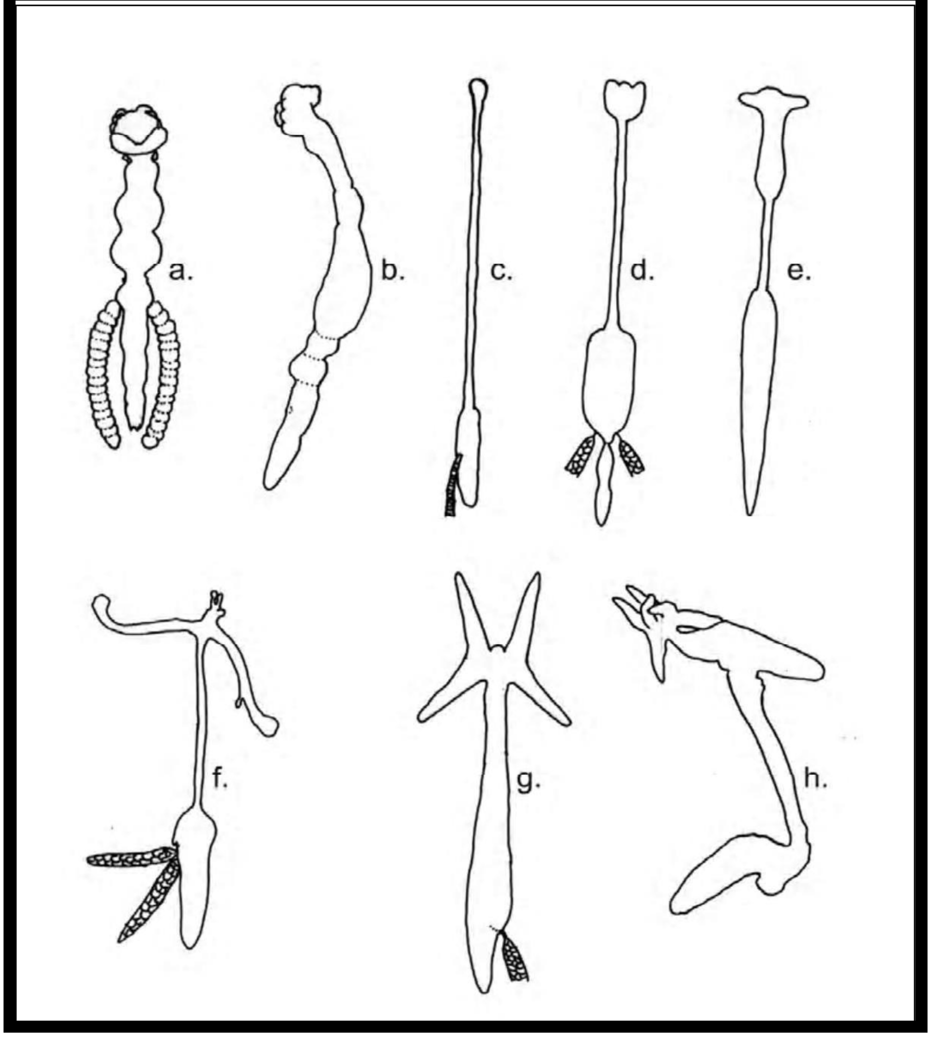


Figure 3: Schematic drawing showing the morphological differences of the members of the family Lernaeidae: a, *Lamproglena*; b, *Lamproglenoides*; c, *Afrolernaea*; d, *Lernaeogariffa*; e, *Areotrachelus*; f, *Taurecheros*; g, *Lernaea* and h, *Opistholernaea* **(**redrawn from **Kabata, 1979).**

is usually separated from the segments anterior to it by a small fifth leg bearing segment that forms a waist-like constriction **(Kabata, 1979)**.

The mesoparasitic lernaeid genera have adult females profoundly modified as the result of metamorphosis. The essence of morphological changes imposed upon them by their mode of life consists in development of a holdfast intended to secure the position of the head within the host, elongation of the body necessary for the penetration of the host and retention of contact with the external environment as well as expansion of the genital region to accommodate increased reproductive efforts. In consequence of these changes, the original segmentation and appearance are lost and the body of the female becomes roughly divisible into three regions: the cephalothorax (with more or less prominent holdfast); elongated neck (cylindrical or gradually expanding posteriorly) and the posterior part of variable structure.

The Lernaeidae is predominating in the Old World. Although some genera have american representatives **(Brian, 1924; Wilson, 1924)** and are known from America only the majority of the lernaeid species occur in Eurasia or Africa. These vast continental masses continue to yield records of new genera and species of Lernaeidae **(Kabata, 1983)**. About 110 species of lernaeid copepods are known from 332 species of freshwater fishes belonging to 161 genera in 41 families. They are mostly known from females which are highly modified and parasitic on freshwater fishes. Some of them are mesoparasites anchoring to their hosts with the transformed cephalothorax (bearing holdfast), but others are ectoparasites attaching to hosts by means of the enlarged and powerful maxillae. From the works of the life cycle of *Lamproglena chinensis* **Kuang (1962)** and *Lernaea cyprinacea* **Grabda (1963)**, it is understood that the adult male lernaeids are not as transformed as their opposite sex and they die soon after mating **(Kuang, 1962; Grabda, 1963)**. *Lamproglena* and *Lernaea* are the two largest genera of the

Lernaeidae and they together account for more than two thirds of the known species of this family. Species of *Lamproglena* are not as widely distributed as those of *Lernaea*; they are absent from the new world and south Pacific including Australia and New Zealand **(Ho, 1998)**.

Biogeography of the family Lernaeidae:

Asia and Africa have the largest number of lernaeids (about 89%) and species of Lamprogleninae are confined to Africa and Asia. This peculiar pattern of lernaeid distribution seems to have resulted from an explosive cladogenesis on the mobile ‘Indian Raft’. This piece of Gondwana-originated subcontinent carries nine of 14 lernaeid genera (*Afrolernaea*, *Catlaphilla*, *Indolernaea*, *Indopeniculus*, *Lamproglena*, *Lamproglenoides*, *Lernaea*, *Pillainus* and *Pseudolamproglena*) including all seven genera of the *Lamprogleninae*. Since species of *Afrolernaea*, *Lamproglena*, *Lamproglenoides* and *Lernaea* are known from both Africa and India, it is logical to assume that these four genera were originated within Gondwana before the separation of India from Africa. Then, the remaining five must have evolved through an explosive cladogenesis of this Indian Raft. This historic event explains why the occurrence of *Indopeniculus* is seen in Thailand and *Pseudolamproglena* is seen in China, but not in Africa. It also explains why the modern Lamproglenids are restricted to Africa and Asia with more than half of them occurring in Asia **(Fryer, 1968; Ho, 1998)**. *Lamproglena monodi* infesting the gill filaments of Tilapia in Egypt **(Ibraheem and Izawa, 2000)**. *Lernanthropus kroyeri* infesting the gill filaments of the sea bass fish, *Dicentrarchus labrax* caught from the Mediterranean Sea, the Coast of Ras El-Bar near Damietta City **(Abu Samak, 2004)**.

Systematics of the family Lernaeidae:

Lernaeidae are parasites on freshwater teleosts and because of their morphological plasticity, their systematics are only too often beset with uncertainties, spurious taxa and confused synonyms. They are characterised by pronounced sexual dimorphism because their males do not undergo metamorphosis and retain a similarity to their free-living cyclopod relatives as well as to the unmetamorphosised females **(Kabata, 1992)**. The familial diagnosis based on metamorphosised female and adult male is as follows:

Female: Body divided into cephalothorax, long sub-cylindrical thoracic neck and more or less distinct trunk like posterior section. Cephalothorax with holdfasts of various types. Neck with or without secondary holdfast. Posterior section either containing indistinguishable fused genital segment and abdomen or indistinctly divided in their two components. Antenna absent or present, other appendages cyclopoid. One to four pairs of biramous legs, fifth and sixth legs also present or absent.

Male: Cephalothorax without holdfast and is well delimited. Four free leg-bearing segments are diminished in size from first to fourth. Genital segment is small and sub-spherical in shape. Abdomen is of fewer than four segments. Appendages as in unmetamorphosised female, but antennae always present **(Kabata, 1992)** (Fig. 4).

Extensive morphological modifications in all genera of this family, except *Lamproglena* and *Lamproglenoides* make the morphology of fully adult females difficult to understand and also tend to obscure the phylogenetic affinities of the

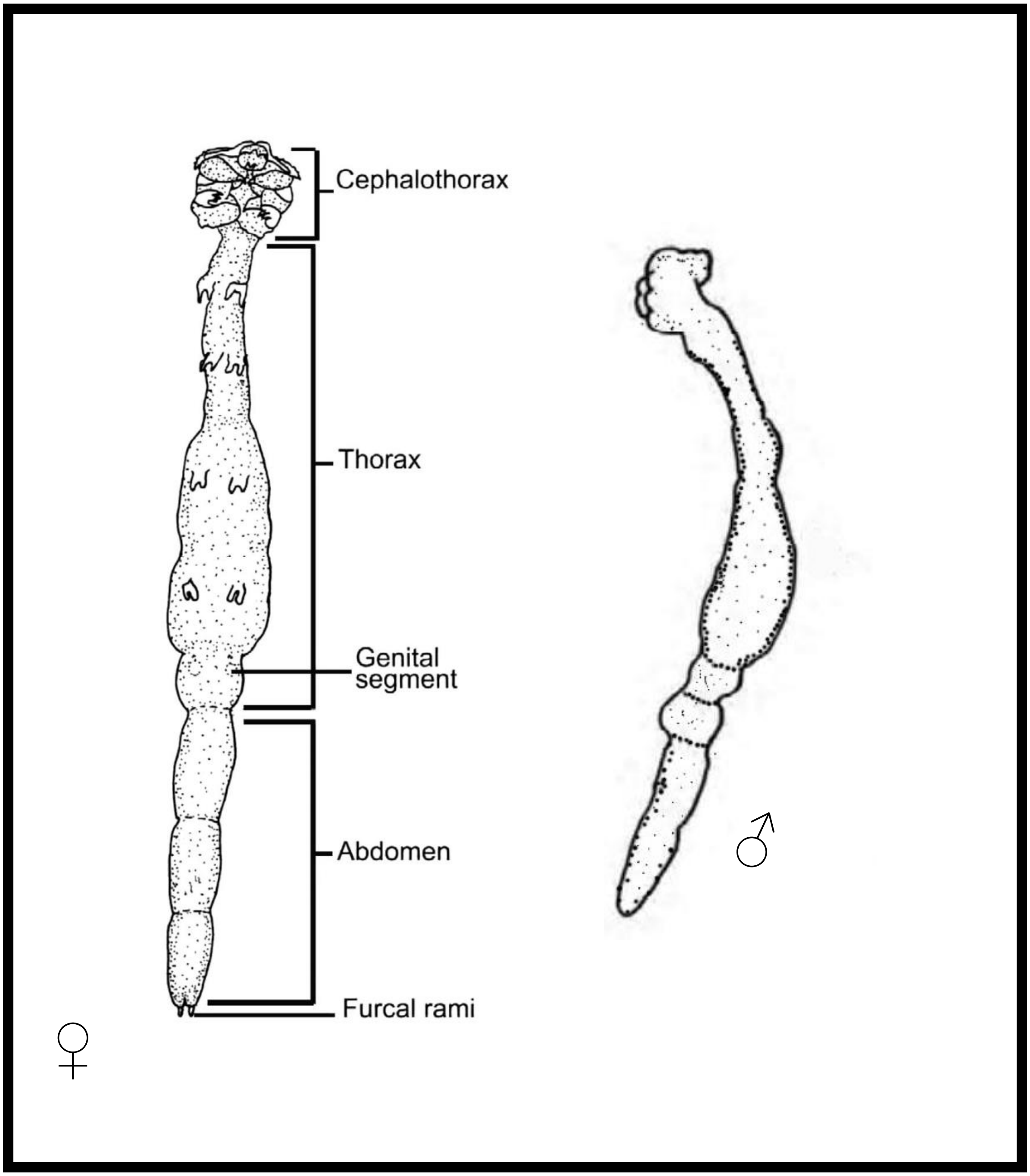


Figure 4: Schematic drawing showing the morphological criteria of female *Lamproglena clariae* and male *Lamproglenoides* spp. (redrawn from **Marx and Avenant-Oldewage, 1996**).

family. The true nature of the lernaeid copepods can be revealed only through knowledge of their developmental stages and currently either unknown or known very imperfectly **(Fryer, 1964)**.

Life cycle of the family Lernaeidae:

The first three stages of the life cycle are nauplii which are slightly different from each other by the progressive increase in length. The third nauplius at the latest phase contains the first copepodid stage within its cuticle. There are five copepodid stages resembling those of other cyclopoid families. At the copepodid fifth stage, the sex of the parasite is distinguishable. This stage is succeeded by the young adult male and female stages which sometimes referred to as cyclopoids (**Grabda, 1963)**. The larva (cyclopoid) uses the second antennae and maxillipeds for prehension. After settling on the gills it continues to use these appendages (Fig. 5). Males use this method as they are not permanently parasitic, but the female when searching for the host burrow or push the cephalothorax into the host's tissue until it is completely buried and only the genital segment as well as the abdomen are left on the outside **(Thatcher, 1998)**. The females become fixed parasites and consequently there is a loss or reduction in locomotory appendages and there is great diversity in the means of prehension. The male does not finally become a fixed parasite, but swims about freely until his death. It is only after the female has become a fixed parasite, the male has perished that the former increase enormously and becomes transformed **(Paperna, 1996)**. The body of the female is always a little longer than that of the male, and just before as well as for some time after fertilization the genital segment increases greatly in length, but remains of the same width. Thus, giving the larva destined to become a female an

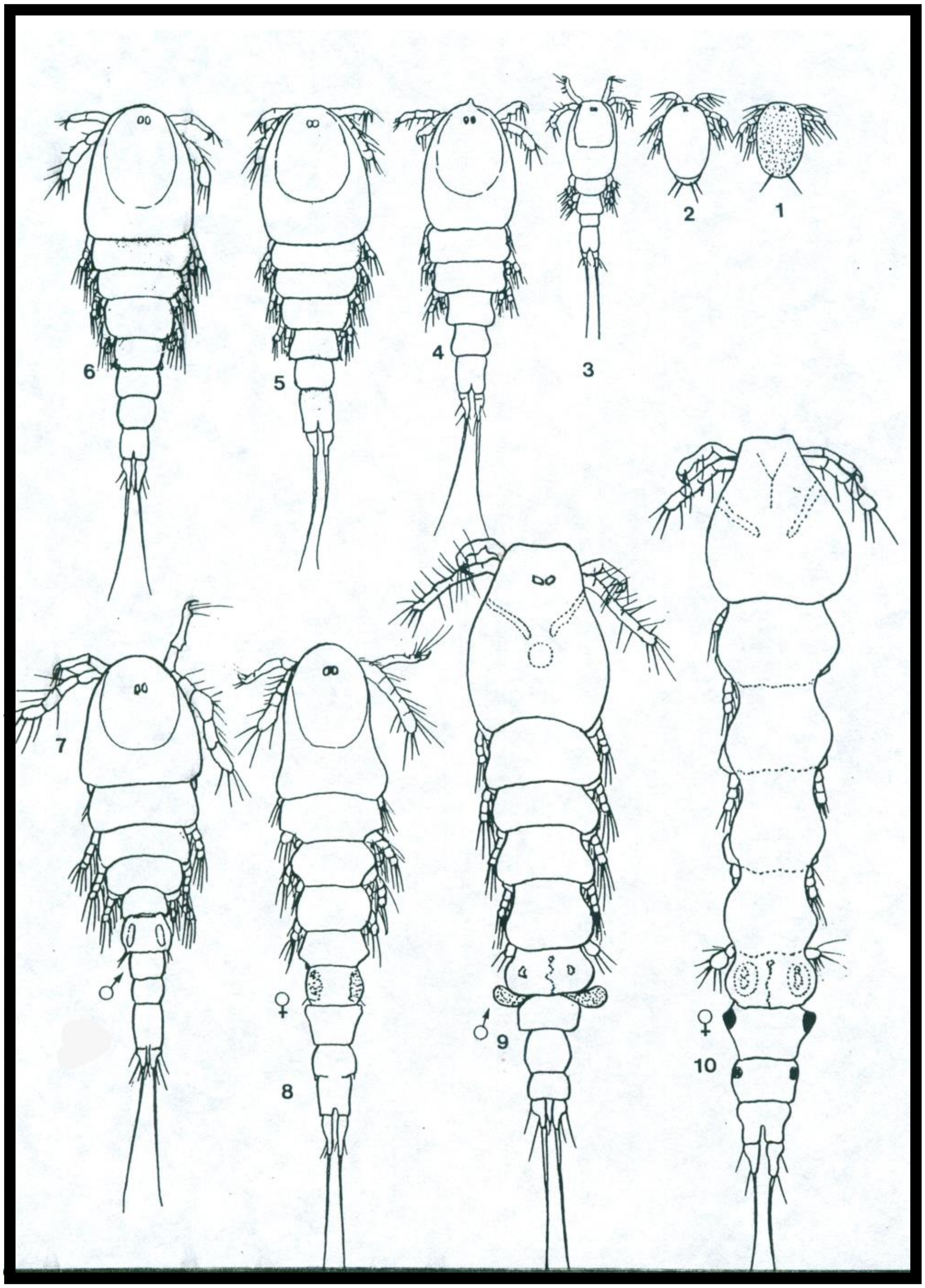


Figure 5: Schematic drawing showing life cycle stages of *Lernaea cyprinacea*; Nauplius (1), Metanauplius (2), Copepodite 1 to 4 (3-6), Copepodite 5 male (7), Copepodite 5 female (8), Adult male (9) and Adult female (10) **(**redrawn from **Paperna, 1980)**.

exceptionally elongated form. The only dimorphism is one of length and not of structure, a difference which is common to all copepods, but is here carried a little farther than usual **(Kabata, 1971)**. The environment of adult female lernaeid is the fish tissue and they can modify this habitat by means of their attachment and feeding strategies **(Thatcher, 1998)**.

Pathology caused by members of the family Lernaeidae:

According to **Kabata (1981)**, initial mechanical disruption of the gill epithelium is a result of penetration by the second antennae which are characteristically used for primary attachment in parasitic copepods, however, there are exceptions to this statement as other copepods such as members of the genus *Lamproglena* that uses second maxillae **(Sproston *et al.*, 1950)** and maxillipedes **(Marx and Oldewage, 1996)** for attachment, whilst second antennae are sensory in function **(Sproston *et al.*, 1950)**. Lernaeids feed directly on blood and can cause primary anaemia in infested fish. They produce hyperplasia and metaplasia in gill filaments which can reduce respiratory capacity in fish **(Thatcher, 1998)**. Infection with lernaeids may be very detrimental to the fish. The point of attachment of the adult female to the host integument (skin, gills, buccal or branchial mucosa) shows a typical inflammatory reaction. A fibrotic capsule is eventually formed around the embedded head and neck. Alternatively, the area around the attachment site may ulcerate with resulting focal necrosis and in skin infection also cause loss of scales. In lernaeids there is more selectivity to site of attachment, relatively fewer numbers of parasitic females may cause severe damage to their host. If aggregated into a limited attachment zone gills or fins, the entire attachment zone will ulcerate and undergo necrosis. The resulting lesion may become secondarily infected by bacteria, fungi or epizoic protozoans **(Paperna, 1996)**.

2-Order: Poecilostomatoida

This order illustrates a progression from little specialized parasites (*Ergasilidae*) to some highly modified and bizarre forms (*Philichthyiae* and *Sarcotacidae*). Poecilostomes have been especially successful as symbionts of other invertebrates, particulary with cnidarian hosts. Of 1475 species of copepods known from invertebrates, 416 belong to this order and 373 species of poecilostomes are associated with cnidarians **(Roberts, 1970)**.

Family: Ergasilidae

Distribution and occurrence of species of the family Ergasilidae:

Most species in the family Ergasilidae belong to the genus *Ergasilus* of which 65 species are parasitic on freshwater fish and 33 species on marine teleosts **(Kabata, 1979)**. *Ergasilus sieboldi* infects over sixty species of fish throughout much of the USSR (except the Pacific Province). The main hosts are *Tinca tinca*, *Esox lucius*, *Acerina cernua*, *Lucioperca lucioperca*, and *Silurus glanis* **(Dogiel *et al*., 1961; Zmerzlaya, 1972)**. Several thousand parasite individuals may occur on the gills of one fish. Cyprinids and salmonids, other than *Coregonus* spp., are less severely infected **(Abrosov and Bauer, 1959; Dogiel *et al*., 1961)**. In Germany 39 of 79 species of fish are host to *E. sieboldi*, the most important being *Esox lucius* and *Abramis brama* (**Dogiel *et al*., 1961**). *E. sieboldi* is also common in British freshwater fishes **(Abdelhalim *et al*., 1991)**. *Ergasilus labracis* infests a wide range of marine species, including salmonids **(Hogans, 1989)**. Ergasilids on amazonian fishes are very host-specific according to **Thatcher and Boeger (1983)**. *Ergasilus lizae* is a species which is restricted largely to the Mugilidae, though it has been found on eels, tilapia, carp as well as mullet in aquaculture ponds in Israel **(Paperna and Overstreet, 1981; Kabata, 1992)** (Fig. 6).

Biogeography of the family Ergasilidae:

Ergasilid copepods occur worldwide on fish. Ergasilids are among the most common copepods, parasitic of fishes. They have been a "thorn in the flesh for many valuable fisheries in the Old World" for a long time **(Kabata, 1970)** and often frequent, the gills of a variety of fishes in North America **(Rogers and Hawke, 1978)**. *Ergasilus sieboldi* is seen in Europe **(Dogiel *et al*., 1961; Grabda, 1991)**. *Ergasilus lizae* is important, occurs worldwide and is known in the Mediterranean Sea and Middle East **(Paperna, 1975; 1991)**. *Ergasilus caeruleus* and *Ergasilus versicolor* are known throughout USA and Canada. The genus *Paraergasilus* has been reported from Asia, East Africa and the Gulf of Mexico. *Pseudergasilus zacconis* is seen in Japan on *Plecoglossus altivelis* **(Nakajima *et al*., 1974)**.

Systematics of the family Ergasilidae:

Roberts (1970) suggested that ergasilid copepods are specific to North American species. **Kabata (1979)** recommended that identification of ergasilid copepods for European species, and **Do (1982)** for Japanese species. Other species have been described from Australia **(Kabata, 1992)** and India **(Ho *et al*., 1992)**. Ergasilid copepods are less modified than most fish-parasitic copepods and resemble free-living copepods in segmentation. Only fertilized females are parasitic; males and developmental stages live in the water column.

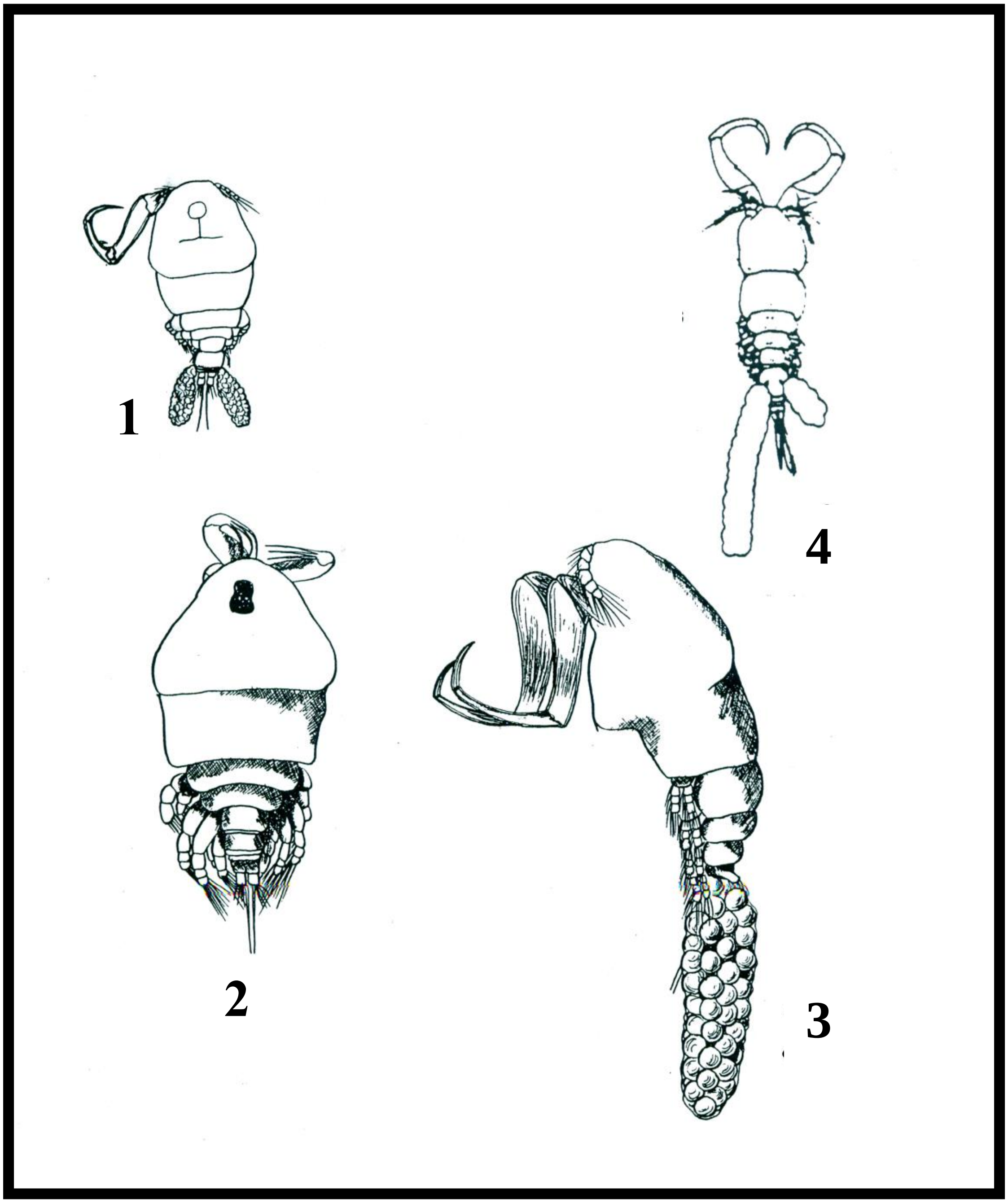


Figure 6: Schematic drawing showing the morphological differences of the members of the family Ergasilidae; *Ergasilus kandti* (1), *Paraergasilus minutus* (2), *Ergasilus latus* (3) and *Ergasilus* spp. (4) **Paperna (1980)**.

Female: The mature female of *Ergasilus* spp. has a cephalothorax formed from the cephalosome and the first leg-bearing segment. Four leg-bearing segments follow, the last intimately associated with the swollen genital segment which gives rise to paired multiseriate egg sacs, each with more than 100 eggs **(Kabata, 1979; Grabda, 1991)**. Antennule 6-segmented, tapering distally. The large second antennae are used for attachment; they either pierce or encircle a gill filament. The second antennae cuticle is swollen in genera such as *Dermoergasilus*. In *Ergasilus* and *Sinergasilus* spp. the second antennae end in a large curved spine; in *Diergasilus* and *Thersitina* spp. the second antennae end in two spines; in *Paraergasilus* three spines. In *Ergasilus briani*, *Ergasilus lizae* and *Ergasilus australiensis* the second antennae end in a stout pointed, clasper-like smooth claw lake in tooth **(von Nordmann, 1832)**. Mouth-parts are on the mid-ventral surface of the cephalothorax, and consist of a two-segmented mandible with a falcate terminal segment, a short first maxilla with two distal setae and a falcate second maxilla with dentiform setae **(Kabata, 1979)**. The stomach extends anterior and posterior to the mouth-parts, merging posteriorly with the straight intestine that leads to the anus at the end of the abdomen between the paired uropods.

Male: Body slender, cyclopiform. Cephalothorax formed from the cephalosome and the first leg-bearing segment. Antennule segmentation and setation as in female. Antennae always present. Urosome 6-segmented comprising short fifth pedigerous somite, genital and 4 free abdominal somites **(Araujo and Boxshall, 2001) (**Fig. 7**)**.

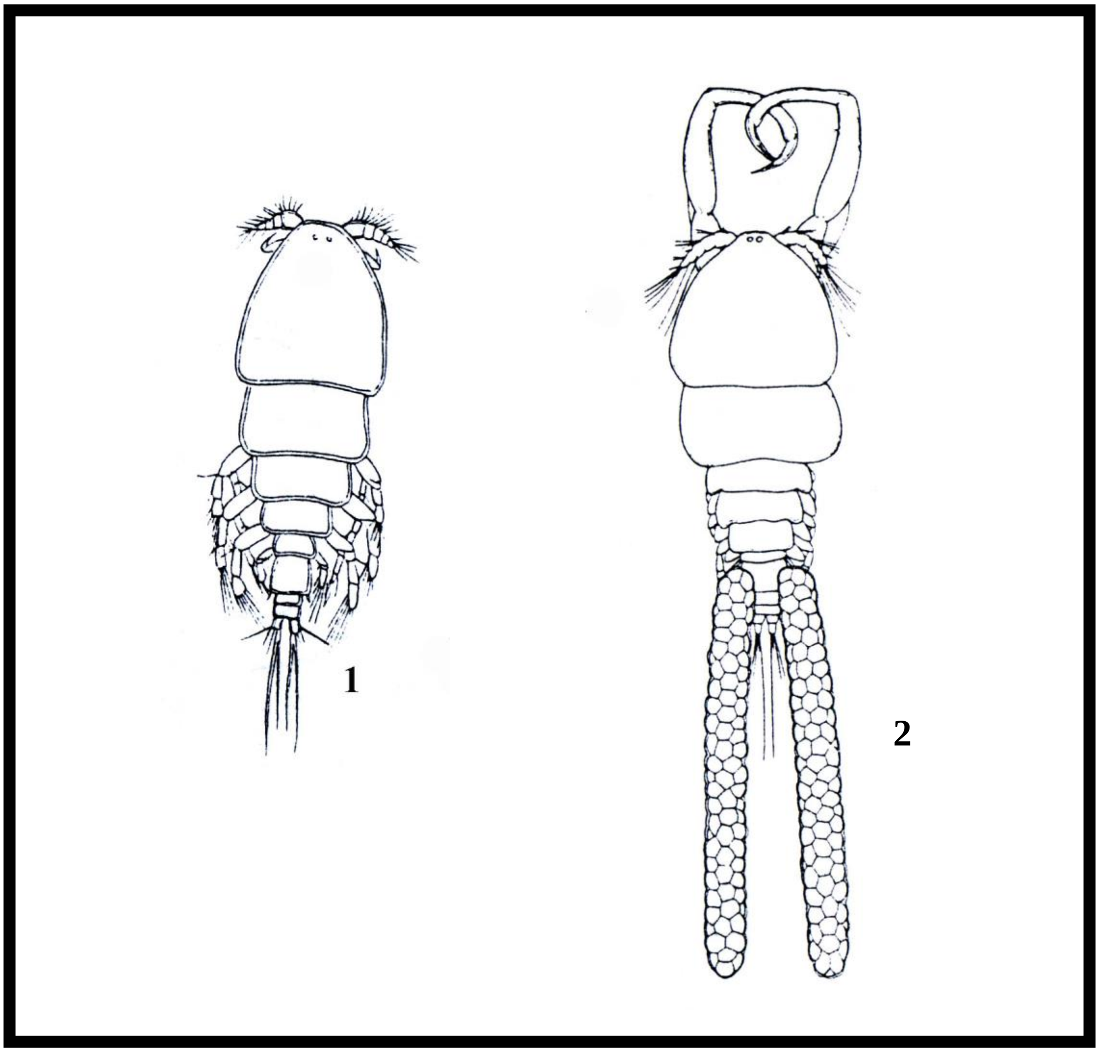


Figure 7: Schematic drawing showing the morphological differences between male *Ergasiloides brevimanus*, **(1) (**redrawn from **Sars, 1909)** and female *Ergasilus borneoensis*, **(2) (**redrawn from **Yamaguti, 1959)**.

Life cycle of the family Ergasilidae:

Eggs, laid in egg sacs, hatch as free-swimming nauplii with oval bodies and six legs. In the plankton, molting occurs between multiple naupliar stages, and several copepodid development stages. Males mate with free-swimming females, and then die. Females find a fish host, attach to the gills, mature and begin producing eggs. Sexual differentiation is evident in the fourth copepodid stage **(Zmerzlaya, 1972; Abdelhalim *et al*., 1991)**. It takes 22 days for the eggs of *E. sieboldi* to develop into free-living males and females **(Zmerzlaya, 1972)**. After fertilization, the female attaches to a fish. **Zmerzlaya (1972)** observed that overwintered females of *E. sieboldi* produced eggs in the spring which meant that sperm was stored for several months. At least two generations per year, including one that over winters, were suggested by **Tuuha *et al*. (1992)** for *E. sieboldi*, *E. briani* and *Neoergasilus japonicus* in Finnish lakes. Non-ovigerous females of *E. labracis* transferred from a winter temperature of 7.2ºC to the laboratory and the temperature increased slowly to 20ºC produced egg sacs **(Paperna and Zwerner, 1976)** (Fig. 8).

Pathology caused by members of the family Ergasilidae:

Fish such as *Tinca tinca* that are heavily infested with *Ergasilus sieboldi* die, especially in summer when water temperatures are highest. There is extensive gill damage and severe haemorrhage with inflammation and exsanguinations associated with the attachment and feeding of the parasite. Blood vessels in the gill filaments are blocked and this leads to atrophy of gill tips **(Dogiel *et al*., 1961)**. Similar histopathology is associated with *Pseudergasilus zacconis* **(Nakajima *et al.*, 1974)**. Infection of *Abramis brama* with *E. sieboldi* results in the disturbance of blood cell maturation with lymphopaenia and reactive

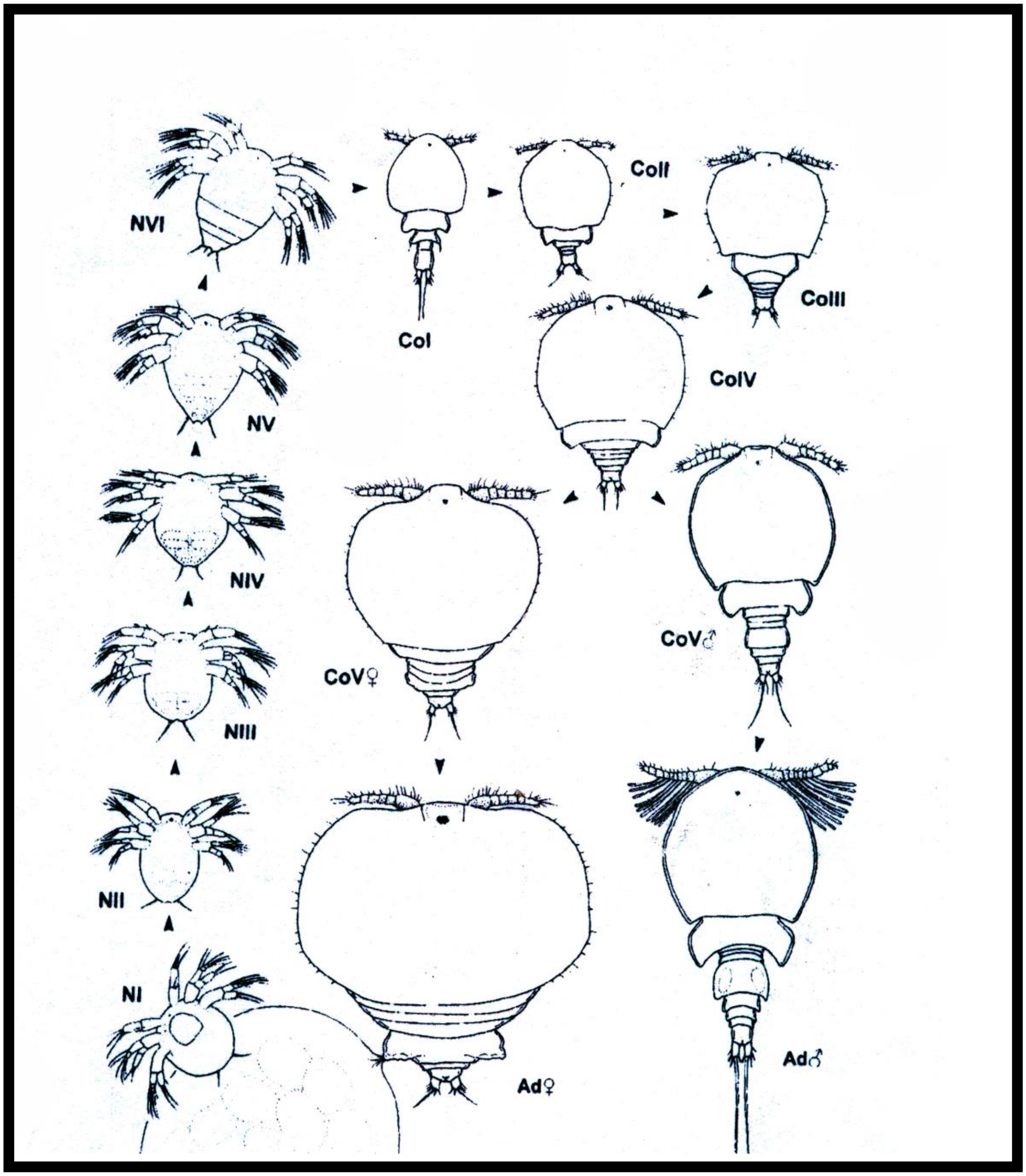


Figure 8: Schematic drawing showing life cycle stages of family Ergasilidae comprising six nauplius stages and five copepodid stages plus the adults of both sexes; Nauplius stages 1 to 6 (NI – NVI), Copepodid stages 1 to 5 (CoI – CoV) and adult male and female (Ad) (redrawn from **Boxshall and Halsey, 2004)**.

granulocytosis in heterophil and basophil leucocytes **(Einszporn-Orecka, 1973a, b)**. *Mugil cephalus* infected with several hundred *E. lizae* were emaciated **(Paperna, 1975)**. *Ergasilus australiensis* causes epithelial hyperplasia in the basal half of the gill filaments and pseudobranchs of *Acanthopagrus australis* **Roubal (1986)**. *Dermoergasilus acanthopagri* which attaches to the tip of the gill filaments by second antennae that encircle the filament, causes localized swelling and occlusion of the filament arterial vessels **(Roubal, 1987; 1989)**. Increase in number of mucous cells, fusion of lamellae and filaments due to epithelial proliferation, lymphocyte infiltration and exsanguination was reported by **Rogers (1969)** for *Ergasilus cyprinaceus*. A similar effect was described by **Thatcher and Boeger (1983)** for *E. colomesus*. **Paperna and Zwerner (1981)** reported an increase in number of mucous cells, epithelial hyperplasia and infiltration of macrophages, lymphocytes and eosinophils in gill filaments of *Morone saxatilis* infested by *E. labracis*.

Schäperclaus (1992) reported that ergasilosis resulted in large-scale mortality of fish and threats to entire standing crops which have been experienced predominantly among tench, pike and bream. Ergasilids take their nourishment from the epithelial and mucous cells of the gills. The digestion of the gill epithelium takes place externally by digestive enzymes and then ingested for the internal digestion. The gill tissues involved reacts by an augmented formation of mucous cells and the coalescence of the gill lamellae due to the proliferation of epithelial cells. **Molnar and Szekely (1997**) reported that *E. sieboldi* infested the pikeperch *Stizostedion lucioperca* and Volga pikeperch *Stizostedion volgense* in Lake Balaton. *Ergasilus* established themselves in the folds on the external surface of the operculum and on the base of the pectoral fins and only a small proportion cling to the gill filaments. Although *Ergasilus* is usually infested the gill filaments but the lesions caused by the presence of copepods on the operculum are restricted to the epithelium even if the infestation was intensive.

3-Order: Siphonostomatoida

The members of this large group are mostly parasites of fishes. Only 31 species have been recorded from cnidarians, but **Humes (1985)** believes that this small number is probably due to neglect in research and so is not real. Although even the most primitive siphonostomes show some adaptations to parasitism, like the poecilostomes, an array from generalized to extremely modified and bizarre can be demonstrated. The majority of siphonostomes are parasites of marine fishes, and since aquaculture of marine fishes has not yet been widely practiced, the actual or potential economic importance of the various siphonostomes is unknown (**Izawa, 1969)**. In western Japan, where culture of yellowtail (*Seriola*) is practiced intensively in small bays, *Caligus spinosus* has inflicted considerable damage (**Izawa, 1969)**.

Family: Caligidae

Distribution and occurrence of species of the family Caligidae:

Lepeophtheirus salmonis has a circumpolar distribution in the northern hemisphere where it is largely restricted to the Salmonidae, especially the genera *Salmo*, *Salvelinus* and *Oncorhynchus* **(Kabata, 1979; Egidius, 1985)**. **Berland (1993)** found the prevalence and intensity of *L. salmonis* and *C. elongatus* on wild salmon in west Norway to be similar in 1973 and 1988 but significantly increased in 1992. During a survey of *L. salmonis* on salmon in the northern Pacific, **Nagasawa *et al*. (1993)** found 78% and 15% of all individuals in this species on pink salmon, *Oncorhynchus gorbuscha* and Chinook salmon, *O. tschawytscha* respectively, with fewer on steelhead trout (*O. mykiss*), coho (*O. kisutch*) and chum (*O. keta*) salmon. *Caligus elongatus* is a cosmopolitan species and has been found on over 80 species of fish in 17 orders and 43 families (including salmonids, pleuronectids, clupeids and gadids). *Caligus elongatus* is rare on wild salmon but is more common on wild sea trout, *Salmo trutta* (**Wootten *et al*., 1982)** and has been recorded on cultured brook trout, *Salvelinus fontinalis* and rainbow trout, *O. mykiss*, in eastern Canada **(Hogans and Trudeau, 1989)** (Fig. 9).

Biogeography of the family Caligidae:

Within the Mediterranean Sea there is an adaptive heterogeneity in *L. europaensis*, which occur at higher densities and with greater sizes on flounder than on brill; eggs and free-living stages tolerate a greater salinity range if obtained from flounder **(Meeüs *et al*., 1993a, b)**. *Caligus elongatus* is the most common species of parasitic copepod in British waters **(Parker, 1969; Boxshall, 1974; Kabata, 1979)**. *Lepeophtheirus salmonis* is seen in northwest and northeast Ireland and in the west 94% of *L. salmonis* is known from farmed salmon **(Tully and Nolan, 2002)**.

Systematics of the family Caligidae:

The caligiform families within the order Siphonostomatoida are characterised by dorso-ventrally flattened bodies divided into an anterior cephalothorax and a post-cephalothoracic genital trunk. These families can be viewed in sequence of increasing modification of body form particularly in the number of leg-bearing somites incorporated into the cephalothorax and fused beneath the dorsal cephalothoracic shield **(Boxshall and Defaye, 1993)**.

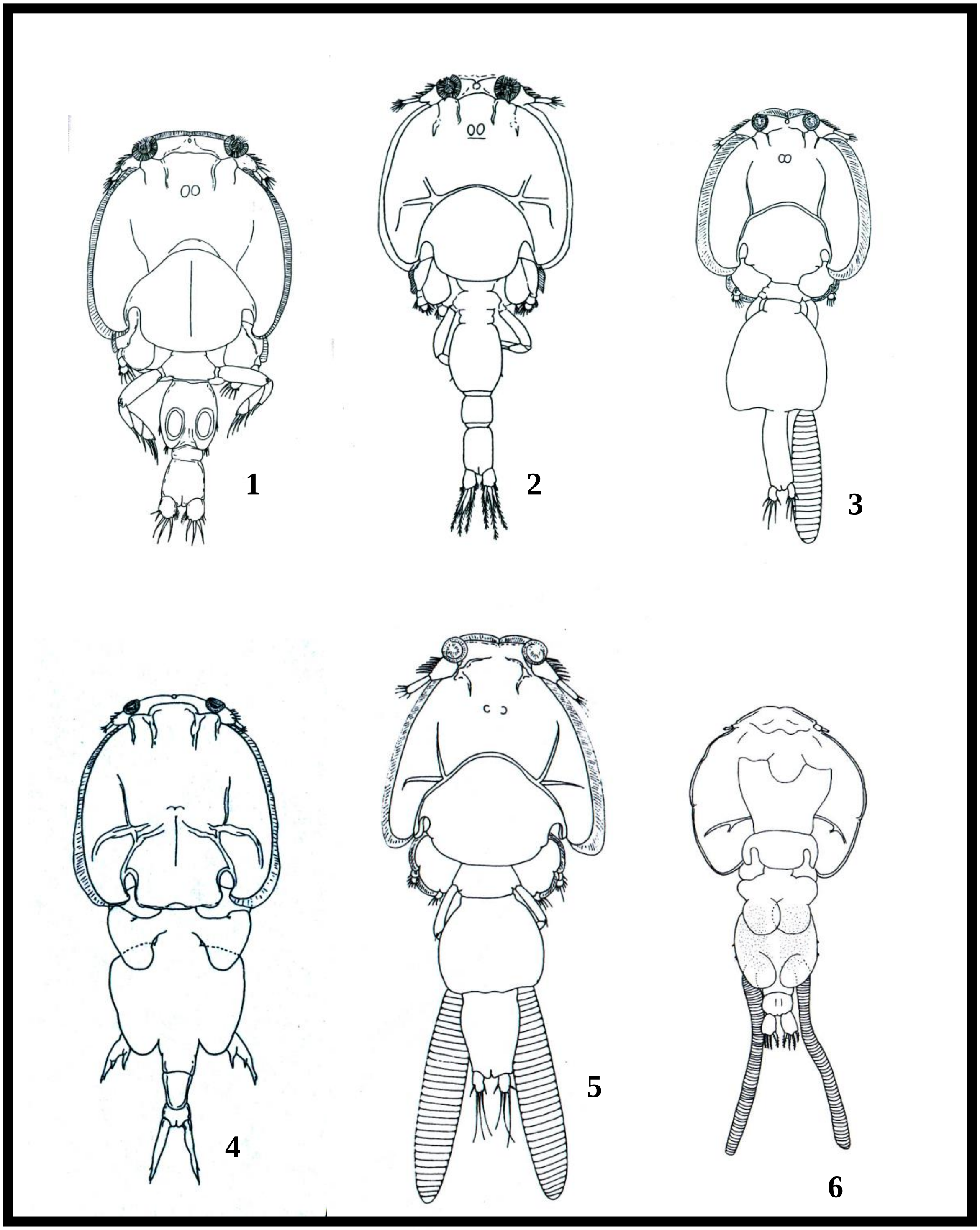


Figure 9: Schematic drawing showing the morphological differences of the members of the family Caligidae; *Caligus diaphanus* (**1**), *Caligus omissus* (**2**), *Caligus biseriodentatus* **(3**), *Tuxophorus cybii* (**4**), *Caligus savala* **(5**) and *Elytrophora brachyptera* (**6**) **(**redrawn from **Cressey, 1980)**.

Caligidae is the most speciose family of parasitic copepods on fish, comprising over 465 species and includes the sea lice which can cause severe economic losses to fin-fish aquaculture, particularly of salmonids **(Boxshall and Defaye, 1993; Pike and Wadsworth, 1999)**. Caligids and related caligiform families typically attach to the host using a combination of claws and suction. The entire cephalothorax is rimmed with marginal membrane and modified limbs and forms an effective seal against the host surface. On the ventral surface and within the cephalothoracic sucker, there are the clawed antennae and maxillipeds which serve primarily for attachment by hooking into the skin of the fish. In some caligids, attachment is further enhanced by additional paired suckers, the lunules located ventrally on the frontal plates as well as by the various spines and processes which enhance friction between parasite and host. Adult caligids usually show sexual dimorphism. Males are smaller than females, with slimmer posterior body region.

Female: The female is usually larger than the male. Cephalothoracic shield subdivided into plates. Shield with marginal membrane, small sensory setules, and single sensory crypt on each of lateral sides. First antenna armed with larger setae. Second antenna is subchelate. Basal segment is bearing powerful sclerotized round-tipped process projecting posteriorly. Sternal furca with trapezoid base and two elongated digitiform processes. Caudal ramus is dominated by three large, centrally located pinnate setae flanked by one small medial and one small lateral setae. One additional small seta observed laterally on ventral side **(Piasecki and Mackinnon, 1995)**. Two egg sacs each contain about 30 eggs **(Mackinnon, 1992)** (Fig. 10).

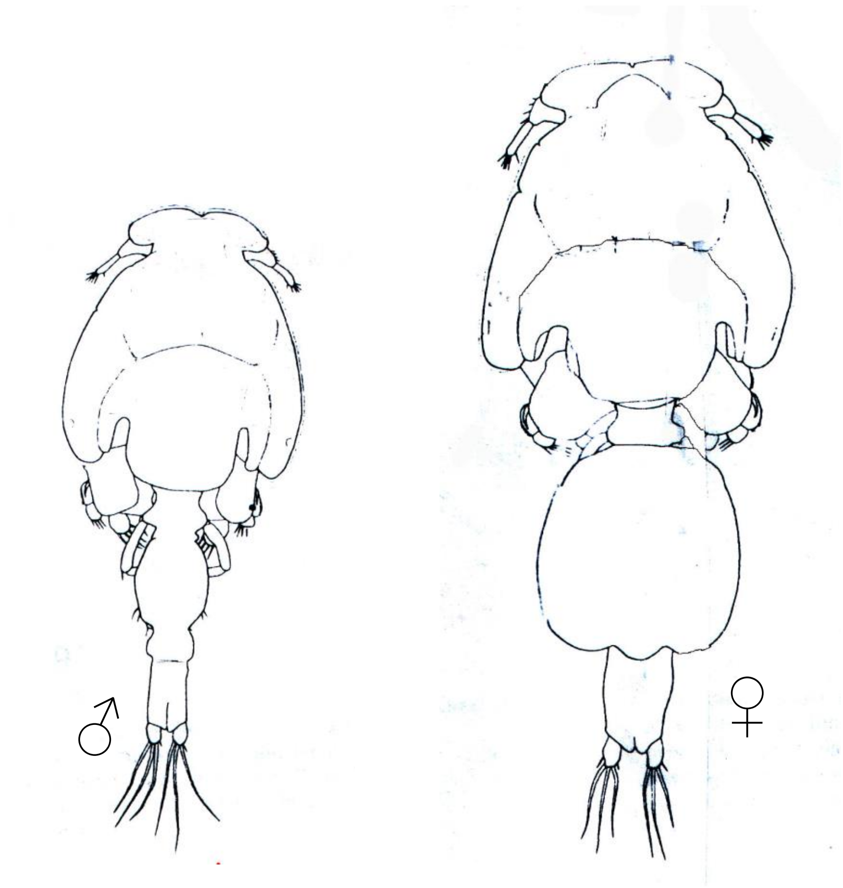


Figure 10: Schematic drawing showing the morphological shapes of male and female *Caligus elongatus* **(**redrawn from **von Nordmann, 1832**).

Male: The body of male is elongated, small than female, with slimmer posterior body region. Cephalothoracic shield and first antenna as in female, but second antenna with basal segment more robust than of female, more strongly sclerotized and enlarged posterolaterally, and equipped with two corrugated pads on ventral surface. One pad large and elongate, other minute and circular. Sternal furca is smaller than in female and with slimmer processes. Larger lateral processes near base. Abdomen two indistinct segmented in male. Genital complex of male is suboval, consisting of two elongated diagonal cuticular flaps shielding genital openings. Caudal ramus is as in female **(Piasecki and Mackinnon, 1995**).

Life cycle of the family Caligidae:

The life cycle of *Caligus* spp. consists of 8 stages separated by molts: nauplius I, nauplius II, copepodid, chalimus I, chalimus II, chalimus III, chalimus IV and adult (Fig. 11). The first three stages are free-swimming. The copepodid is infective and all subsequent stages live on fish **(Piasecki and Mackinnon, 1995**). Shape of newly hatched larva (Nauplius I) reflecting shape of egg (i.e., a very short cylinder). Nauplius II resembling previous stage. Body of copepodid is elongated, with efficient hydrodynamic shape. Larvae of chalimus I are longer and wider than copepodids. Segmentation of chalimus II is less obscure than in previous stage. On the other hand, segmentation of chalimus III is more pronounced than in previous stage. Chalimus IV is dorsoventral flatting pronounced. Cephalothorax is wider and dorsal shield subdivided into plates. Frontal region is triangular with obtuse angle. Sternal furca is present. Young adults attached by frontal filament. Older ones break free but some retain proximal portion of filament for the rest of their lives.

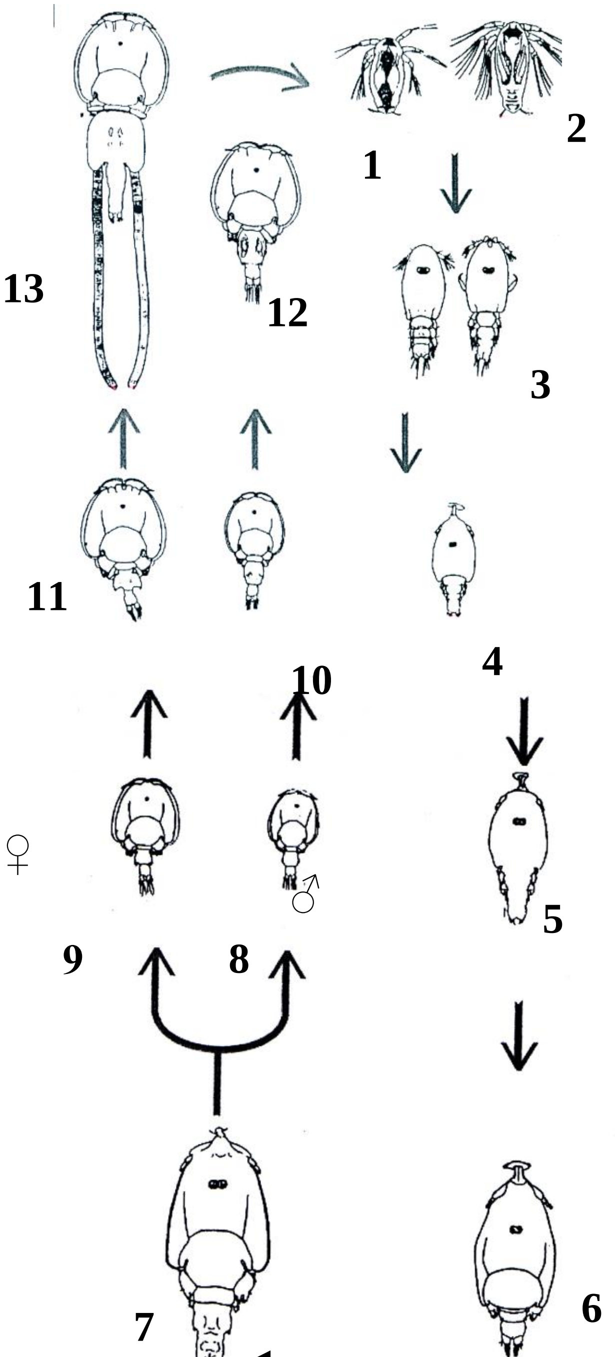


Figure 11: Schematic drawing showing life cycle stages of *Caligus* sp. comprising two nauplius stages I and II (1and 2), copepodid (3), four chalimus stages I, II, III and IV (4, 5, 6 and 7), preadults stages I and II male (8 and 10) and female (9 and 11) and adults male and female (12 and 13) (redrawn from **Gregor**, **2003)**.

Sexual dimorphism is apparent. Males are smaller than females with slimmer posterior body region **(Lin and Ho, 1993; Piasecki and Mackinnon, 1995**).

Pathology caused by members of the family Caligidae:

Caligidae, often referred to as "sea lice" and is responsible for most disease outbreaks. Sea lice (Copepoda: Caligidae) is crustacean ectoparasite associated to wild and cultured salmonid species that feed on the mucus, skin and blood of the host(**Pike and Wadsworth, 2000; González and Carvajal, 2003**). Sea lice is an ectoparasite which occur on many fish worldwide and are regarded as having the most commercially damaging effect on cultured salmon in the world with major economic losses to the fish farming community resulting each year **(Bristow and Berland, 1991).** Sea lice affects salmon by reducing fish growth, causing loss of scales which leaves the fish open to secondary infections **(Wooten *et al.*, 1982)** and by damaging the fish which reduces its marketability. In the eastern Mediterranean, the copepods *Caligus pageti* and *Pseudocaligus apodus* caused skin sores, emaciation and mortality in brackish and sea water ponds always found on the gills of mullets. The two species of sea lice found on cultured salmonids in Ireland are *Caligus elongates* **(von Nordmann, 1832)** a species of parasite that infests over 80 different types of marine fish, and *Lepeophtheirus salmonis* **(Krøyer, 1863)** which infests only salmon and other salmonids.

L. salmonis is regarded as the more serious copepodean parasite of the two species and has been found to occur most frequently on farmed salmon **(Jackson *et al.*, 1997; Jackson *et al*, 2005)**. Most of the damage caused by these parasites is thought to be mechanical, which is carried out during the course of attachment and feeding **(Kabata, 1974; Brandal *et al.*, 1976; Jones *et al.*, 1990**). Inflammation and hyperplasia (enlargement caused by an abnormal increase in the number of cells in an organ or tissue) have been recorded in Atlantic salmon in response to infections with *L. salmonis* **(Jones *et al*., 1990; Nolan *et al*., 2000)**. Increases in stress hormones caused by sea lice infestations are thought to increase the susceptibility of fish to infectious diseases (**Nolan *et al*., 2000)**.

The infection with sea lice can affect the growth, fecundity, and survival of their hosts because their feeding may cause skin lesions leading to osmotic problems and consequently the occurrence of secondary infections and if untreated, they can reach a level that is highly detrimental the fish (**Pike and Wadsworth, 1999; Nolan *et al*., 2000; Bjørn *et al*., 2001; Bjørn and Finstad, 2002; Tully and Nolan, 2002; Heuch *et al*., 2005**). Both wild and farmed salmonids can act as hosts to sea lice, and the possible interaction and cross-infection of the parasite between farmed and wild fish is causing much concern (**Tully, 1992; Birkeland and Jakobsen, 1997; Tully and Nolan, 2002; Marshall, 2003; Morton *et al*., 2004**).

Origin and evolution of the parasitic copepods:

The evolution of any character under natural selection is mediated through its impact on life-history. Thus, a central challenge of biology is to understand the effect of each trait that an organism possesses whether molecular, developmental, physiological, morphological, or behavioral. On survival and reproduction in the environment in which it lives, one route to understanding the linkages between traits and environments is through a comparison of the life histories of organisms sharing a common ancestry but living in different habitats. The Copepoda are a particularly good group for such an exploration they are clearly monophyletic and inhabit a variety of environments including epibenthic and interstitial habitats, the open ocean pelagic zone, estuaries, inland waters, ground water, moist terrestrial soils and symbiotic associations with a wide variety of animal taxa **(Huys and Boxshall, 1991)**.

Each major habitat in which copepods are found has experienced repeated independent invasions **(Huys and Boxshall, 1991)**. Ancestral copepods are assumed to have been marine epibenthic animals. The Platycopioida and the Misophrioida have remained in this habitat, as have many of the Harpacticoida and Cyclopoida. The marine pelagic zone has been colonized by five orders the Calanoida, the Mormonilloida and a few Harpacticoida, Cyclopoida and Poecilostomatoida via at least 10 independent invasions. According to **Huys and Boxshall (1991)** inland waters have been independently invaded 19 times by six copepod orders (the Calanoida, Cyclopoida, Harpacticoida, Gelyelloida, Poecilostomatoida, and Siphonostomatoida). A number of groups the Poecilostomatoida, the Siphonostomatoida, the Monstrilloida as well as a few species of Cyclopoida and Harpacticoida have taken on associations with a variety of marine and freshwater animals, often as parasites. These symbiotic relationships have apparently been independently established many times.

Despite the fact that copepods have successfully taken up life in a wide variety of habitats, their basic life history remains relatively conserved Adults that mature to produce multiple clutches of eggs during a single year. Most are iteroparous annuals. Postembryonic development always includes a fairly familiar set of instar types nauplii and copepodids including adults. The only exceptions are the elimination of nauplii and the introduction of the chalimus by some parasitic taxa. **McMahon, (2002)** proposed that much of evolution can be attributed to heterochrony; that ‘‘the vast majority of differences among closely related organisms are due to simple changes that are manifested relatively late in ontogeny.’’ For the Copepoda, **Huys and Boxshall (1991)** discuss several morphological features, such as the separation of the genital and first abdominal somites, the sixth legs and the geniculate antennules, whose interspecific variation can be interpreted as heterochronic in nature. Certainly, the loss or addition of instars and the numbers of these stages relative to the major nauplius to copepodid metamorphosis are evolutionary changes that must have as an underlying basis alternations in the timing of developmental events.

Not only is more research required on phenotypic and genetic variation within species, but such studies need to be coupled with carefully chosen comparisons between taxa with different levels of relatedness living in different types of environments. For this purpose, phylogenetic hypotheses must be constructed that attract some broad level of consensus and these must be based on characters that are independent of the adaptively significant life history traits we hope to understand. Molecular data are ideal for this purpose. In addition, much more information is needed on the basic ecology of taxa living in the unusual habitats in which copepod species sometimes occur e.g., terrestrial leaf litter, ground water, deep sea, various host organisms. Combining studies of interspecific variation and phylogeny with a thorough knowledge of the selection pressures exerted by different environments will be a powerful route to understanding the origin and maintenance of the diversity of copepod life histories.

Control of parasitic copepods:

Parasitic copepods are common on cultured and wild marine finfish, and there is a vast literature describing their taxonomy and host ranges. Many of these species have long been recognized to have the potential to affect the growth, fecundity and survival of wild fish hosts **(White, 1940; Kabata, 1958; Hewitt, 1971; Neilson *et al.*, 1987; Johnson *et al.*, 1996)**. With the development of semiintensive and intensive brackish water and marine aquaculture, the importance of parasitic copepods as disease causing agents has become more evident.

Parasitic copepods feed on host mucous, tissues and blood. Moreover, their attachment and feeding activities are responsible for any primary disease that develops. The relationship between number of parasitic copepods and severity of the disease is dependent on:

1) The size and age of the fish.

2) The general state of health of the fish.

3) The species of copepod and the developmental stages present (**Pike and Wadsworth, 1999)**. Losses associated with disease are the result of direct mortality, mortality due to secondary infections, reduced growth, loss of carcass value and costs of treatment **(Lin *et al.* 1994; Pike and Wadsworth, 1999; Ho, 2000)**.

Elimination of the parasite usually requires treatment over several weeks to break the life cycle at the larval stage because embedded females are difficult to kill **(Kabata, 1985)**. Control of the disease is based largely on use of chemical treatment as formalin, malachite green, chloramine T and toltrazuril **(Cross and Hursey, 1973; Farley and Heckmann, 1980; Schmahl *et al*. 1989; Mathews, 1994)** but the cumulative effects for such treatments on edible fish can be questionable (**Bernoth, 1991**). Consequently, elimination of free-living stages as tomites or theronts by repeated changes of water and sediment in cultures can decrease population density of the parasite without any drastic effects on fish health.

An important aspect of effective control of fish diseases caused by parasites is reliable diagnostic (**des Clers, 1993**) preferably in early phases of a disease, enabling the application of adequate prophylactic measures and treatment or prevention of serious outbreaks. Several compounds have been successfully used in a wide spectrum to control and treat these parasites. Formalin is a one of the oldest treatment agent of fish ectoparasites where it has a markedly deleterious effect on a wide range of parasites (**Allison, 1957; Gopalakrishnan, 1964; Schmahl, 1991; Flores-Crespo *et al*. 1995; Tonguthai, 1997)**. It is parasiticidal effect depends upon its fixative action, while it has the disadvantage of the high toxicity for the fish (**Schmahl, 1991**).

Hoffman and Williams (1999) reported that Dimilin, a growth regulator is eradicated *L. cyprinacea* from golden shiner (*Notemigonus crysoleucas*). To avoid environmental and other side effects of chemical treatments, several biological methods have been proposed. **Kabata (1985)** reported that the copepod *Mesocyclops* preyed on free-swimming larval *Lernaea* and suggested that planktonic predators could be used in biological control.

Hogans (1989) reported the successful treatment of *Ergasilus labracis* on Atlantic salmon parr with Neguvon. Fresh water and hyposaline sea water were ineffective against *E. lizae* **(Conroy and Conroy, 1986)**.

Chemotherapeutics used against sea lice include formaldehyde, organophosphate insecticides (malathion, trichlorphon, dichlorvos, azamethiphos), ivermectin, pyrethrum, carbaryl, diflubenzuron, hydrogrn peroxide and aquatic extracts of onions and garlic **(Roth *et al*. 1993)**. The treatment reduced the number of lice and prevented damage to the host though there was a narrow safety margin. **Ho *et al*. (1992)** concluded that ivermectin was not well suited to oral treatment because high concentrations reached the central nervous system and the drug was excreted slowly.

It is noteworthy mentioning that, most of the previous chemicals and drugs applied in controlling and treatment of the parasites of cultured fishes are either highly toxic for fishes (e.g. formalin, organophosphates and trichlorphon) or must be used under specific conditions. Moreover, the use of chemotherapeutic agents in fish aquacultures has certain disadvantages. They may be toxic to the natural food fish (non-target organisms), may produce harmful accumulation in the fish flesh and environment and their over use (multiuse) may lead to the development of drug resistance (**Spence *et al*. 1982; Tonguthai, 1997; Abu Samak and Khidr, 1998; 2000**). Therefore, it was found necessary to search for more effective and suitable ways for controlling fish parasitic-diseases especially in fish farms by using either a biological control or natural products (e.g. plant extracts). In this respect, these ways may be more safe to the target (i.e. fish) and non-target organisms (i.e., all biota in the fish farms) and also to the environment (**Didier *et al*., 1988; Roy and Tandon, 1996; Abu Samak and Khidr, 1998; 2000; Khidr *et al*., 2003)**.

The present work evaluated the efficacy of two natural extracts Curcumin, (*Curcuma longa*) and Myrrh, (*Commiphora molmol*) and synthetic drug (Mebendazole) treatment against infections with the crustacean parasites (*Lamproglena minuta*) infesting *Sarotherodon galilaeus* and (*Ergasilus celestis*) infesting *Anguilla anguilla*. To date there appears to be no information on the efficacy of curcumin and Myrrh against crustacean parasites. The use of medicinal plants for the treatment of many diseases is associated to folk medicine from different parts of the world. Natural products from some plants continue to be used in pharmaceutical preparations either as pure compounds or as extracts **(Leon and Araujo, 2001)**. In the current study, the plant extracts were applied on the parasitic copepods of fish hosts. The first natural extract curcumin, *Curcuma longa* is a medicinal plant having a short stem with large leaves and bears ovate or oblong rhizomes, which are often branched and brownish-yellow in colour **(Ishita *et al*., 2004)**. The ethanol extract of the rhizomes of *Curcuma longa* has anti-*Entamoeba histolytica* activity **(Ishita *et al*., 2004)**. curcumin has anti-*Leishmania* activity in *vitro* **(Koide *et al*., 2002)**. Several synthetic derivatives of curcumin have anti-*L. amazonensis* effect **(Ishita *et al.,* 2004)**. **Rasmussen *et al.* (2000)** reported the efficacy of an ethanolic extract from *Curcuma longa* against *Plasmodium falciparum* and *L. major,* which was able to inhibit, in *vitro,* growth of these parasites. **Unnikrishnan and Rao (1995)** studied the antioxidative properties of curcumin and its three derivatives (demethoxy curcumin, bisdemethoxy curcumin and diacethyl curcumin). **Mazumber *et al.* (1995)** demonstrated that curcumin has an antiviral activity, being a HIV-1 integrase inhibitor and suggested that curcumin analogs may be developed as anti-Aids drugs. **(Leon and Araujo, 2001)** suggest that curcumin may be useful in the therapy of cancer and bone inflammation diseases that increase bone resorption. Curcumin decreases the severity of pathological changes and thus protects from damage caused by myocardial infarction **(Nirmala and Puvanakrishnan, 1996)**. Curcumin and its derivatives and many other extracts from the rhizomes recommended previously as anti-inflammatory, antioxidant, anticarcinogenic, anticoagulant, antifertility, antidiabetic, antibacterial, antifungal, antiprotozoan, antiviral, antifibrotic and antivenom agent **(Ishita *et al*., 2004)**. The second natural extract Myrrh, *Commiphora molmol* is an oleo-gum resin obtained from the stem of the plant *Commiphora molmol* **(Massoud *et al*., 2001)**. Myrrh has been shown to be a safe and effective anti-schistosomal drug at a dose of 10 mg/kg/day for six days **(Massoud *et al*., 2001)**. Myrrh proved to be safe and very effective in treatment of *Schistosoma haematobium* and *Schistosoma mansoni* parasites **(Abo-Madyan *et al.,* 2004)**. **Botros, *et al.* (2004)** determined the efficacy of Myrrh in the treatment of *Schistosoma haematobium* and *Schistosoma mansoni* parasites. Myrrh may prove efficacious for the treatment of Fasciolaiasis disease caued by *Fasciola* (liver fluke) that infects sheeps, goats and cattle, for which humans act as an accidental host **(Massoud *et al.,* 2001)**. Also, Myrrh has molluscicidal effect on infected *Bulinus truncates* and *Biomphalaria alexandrina* snails **(Massoud and Habib, 2003)**. There are many medicinal uses for Myrrh, for example, to treat wounds, intestinal parasites, diarrhea, persistent cough and chest ailments **(Ghazanfar, 1994)**. It is also commonly used to treat diabetes **(Al-Rowais, 2002)**. Finally, the third agent is a synthetic drug, Mebendazole. It has not been investigated for any parasite *in vitro*, but, *vivo*, its effects have been studied against helminthes other than amphistomes. The time-related topographical changes in mature cysticerci of *T. taeniaeformis* induced after treatment of infected mice with mebendazole were reported by **Vaughan (1975)**. Treatments with mebendazole have been utilized to eradicate *Pseudodactylogrys bini* and *P. anguillae* (**Szekely and Molnar, 1987; Buchmann and Bjerregaard, 1990; Buchmann *et al*., 1992; 1993).** Lower efficacy of mebendazole was observed by treatment infection with *Gyrodactylus sp*. in rainbow trout, *Oncorhynchus mykiss*, and (**Tojo *et al*., 1992**). **Maurício *et al.* (2001)** evaluated the efficacy of mebendazole treatment against infections with *Anacanthorus penilabiatus,* gill parasites of *Piaractus mesopotamicus*. **Buchmann (1994)** reported slight aberrations of the tegument; partial destruction of the microtubule and disintegration of the terminal cell in mebendazole treated monogenean. Mebendazole is a known broad spectrum anthelmintic used widely to treat nematode and cestode infestations of domestic animals **(Martins *et al*., 2001)**.

AIM OF THE WORK

Aim of the work

The Mediterranean coast of Egypt is about 1100 km. long. It extends from El-Sallum in the West to El-Arish in the East. Most of the fishing activities are concentrated in the eastern zone, where the continental shelf (up to the depth of 200 m) is very wide and flat **(Ibrahim and Soliman, 1996)**. Egypt with its dense and fast growing population, concentrated along the River and Delta region, considers that fish and fishery products play an important role in the country's food security and domestic economy.

The Mediterranean Sea fishes especially in coastal waters of A.R. Egypt are considered as highly valued fish food. Also, Egyptian cichlids are the most popular fish kinds dewelling the Nile River because of their qualities, grow quickly and abundant throughout the year. Therefore, this work is planed to study parasitic crustaceans infesting some economic marine fishes caught from the Mediterranean Sea, coast of Lake Manzala near Damietta City, Damietta Province and coast of the Nile River near El-Zarka City. Moreover, it is designed to investigate:

1- The survey and depict parasitic crustaceans infesting some economic marine and fresh fishes serving for the Egyptian database.

2- The morphological and anatomical features of these collected parasitic crustaceans.

3- The effect of environmental and biological on these parasites.

4- The geographical range of the collected parasitic crustaceans.

5- The histological characters of the examined fishes in order to morphologically clarify the fine structures of the host gills.

6- The histopathological effect caused by parasitic crustaceans.

7- The effect of plant extract of *Curcuma longa* and *Commiphora molmol* on the parasitic crustaceans. In this respect, Curcumin and its derivatives and many other extracts from the rhizomes recommended previously as anti-inflammatory, antioxidant, anticarcinogenic, anticoagulant, antifertility, antidiabetic, antibacterial, antifungal, antiprotozoan, antiviral, antifibrotic and antivenom agent **(Ishita *et al.*, 2004)**. Moreover, there are many medicinal uses for Myrrh, for example, to treat wounds, intestinal parasites, diarrhea, persistent cough and chest ailments **(Ghazanfar, 1994)**.

8- The effect of synthetic drug anthelmin on the parasitic crustaceans which has been used as an alternative against human nematodes and cestodes infections and has been widely used in veterinary medicine, as it is active against flukes and cestodes **(Yanong and Watson, 2005).**

MATERIALS AND METHODS

Materials and Methods

1- Study areas and host fishes collection

A total of **900** fishes of different species were monthly collected during the present study. At least 50 fishes /month were collected from April 2005 until March 2006. The fishes were collected from three sites. The study areas are located in Damietta Province, northern part of Egypt, Lake Manzala (Fig. 12). Site 1 lies close to Shata village on the Mediterranean Sea, site 2 lies close to El-Deba village and site 3 lies close to the Nile River at El-Zarka City. The fish was collected by trawling nets. The collected fishes were brought to the laboratory alive in tanks provided with ventilated either sea or fresh water. Fishes were examined directly after arrival to the laboratory.

2- Determination of host length, weight and sex

The fish standard length was measured from the tip of the snout to the posterior end of the body. Fish were grouped into several lengths (from 10 to 50 cm). The fish weight (in grams) was recorded. Fishes were grouped into several weights (from 20 to more than 900 g). Host sex was determined by dissecting the fish to expose the gonads if present. The fishes were classified as male (group I) and female (group II).

3- Identification, systematic position and nomenclature of the examined host fishes:

Twelve species are belonging to nine families were identified throughout the present investigation depending on **Wheeler (1985)** and **Ibrahim and Soliman (1996)**.

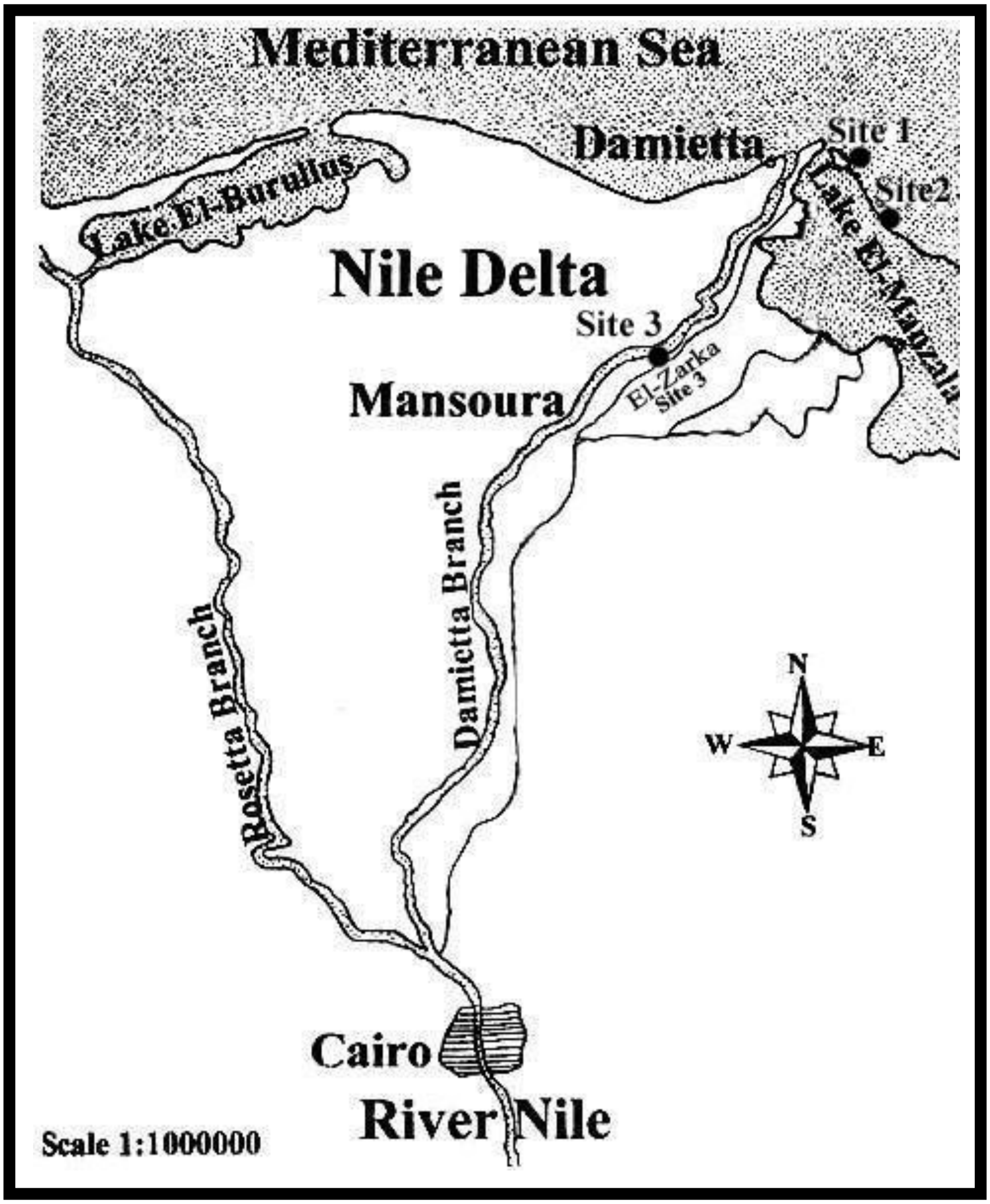


Figure 12: Map showing the study areas and fish collection sites; site 1 at Shata village, site 2 at El-Deba village and site 3 at El-Zarka City.

The systematic position and nomenclature of the examined fishes in the present study are as follows:

Family : Serranidae.

e.g. : *Dicentrarchus punctatus* **(Bloch, 1792).**

English name: Spotted seabass

Arabic name: نقط (Site 1 and 2) (Fig. 13i, A).

e.g. : *Dicentrarchus labrax* **(L., 1758).**

English name: European seabass

Arabic name: قاروص (Site 1 and 2) (Fig. 13i, B).

Family : Mugilidae.

e.g. : *Mugil cephalus* **(L., 1758).**

English name: Flathead Grey Mullet

Arabic name: بوري حر (Site 1 and 2) (Fig. 13i, C).

e.g. : *Valamugil seheli* **(Forsskal, 1775).**

Arabic name: سهيلي (Site 1 and 2) (Fig. 13ii, D).

e.g. : *Liza ramada* **(Risso, 1826).**

English name: Thinlip Mullet

Arabic name: طوبار (Sit 1 and 2) (Fig. 13ii, E).

Family : Sparidae.

e.g. : *Diplodus sargus* **(L., 1758).**

English name: White Sea Bream

Arabic name: شرغوش (Site 1) (Fig. 13ii, F).

Family : Anguillidae.

e.g. : *Anguilla anguilla* **(L., 1758).**

English name: Common eel

Arabic name: حنش (Site 1 and 2) (Fig. 13iii, G).

Family : Soleidae.

e.g. : *Solea solea aegyptaica* **(Chabanaud, 1927).**

English name: Egyptian Sole

Arabic name: سمكة موسى (Site 1 and 2) (Fig. 13iii, H).

Family : Sciaenidae.

e.g. : *Argyrosomus regius* **(Asso, 1801).**

English name: Meagre

Arabic name: لوت (Site 1 and 2) (Fig. 13iii, I).

Family : Lutjanidae.

e.g. : *Lutjanus lineolatus* **(Allen, 1985).**

English name: Striped snapper

Arabic name: شكرم (Site 1) (Fig. 13iv, J).

Family : Mullidae.

e.g. : *Upeneus asymmetricus* **(Lachner, 1954).**

English name: Golden Striped Goatfish

Arabic name: بربوني مخطط (Site 1 and 2) (Fig. 13iv, K).

Family : Cichlidae.

e.g. : *Sarotherodon galilaeus* **(Boulenger, 1899).**

English name: Tilapia galilaea

Arabic name: بلطي جاليلي (Site 3) (Fig. 13iv, L).

4- Collection of parasitic copepods:

The hosts' skin, fins, gills, buccal and branchial cavities, and eyes were examined for searching for parasitic copepods. The skin and fins were examined by scrapeing them with scalpel blade. Gills were removed, placed into glass Petri dishes filled with filterated sea water or freshwater and examined under dissecting microscope.

5 -Identification, systematic position and nomenclature of the parasitic copepods:

Specimens were fixed in 10% formaldehyde. They were washed under running tap water over-night to remove formalin then dehydrated in ascending series of alcohol. They were transferred into lactic acid (85%) for 1 to 2 days, cleared in xylene and mounted in lactic acid.

The parasitic copepods were identified using **von Nordmann (1832); Markewitsch (1933), Capart (1943), van Beneden (1851), Kroyer (1863), Roberts (1970), Kabata (1979), Roubal (1981), Rahemo (1982)**.

Identification, systematic position and nomenclature of the parasites under investigation are as follows:

Figure 13i: Photomicrographs showing the fish species examined during the present work.

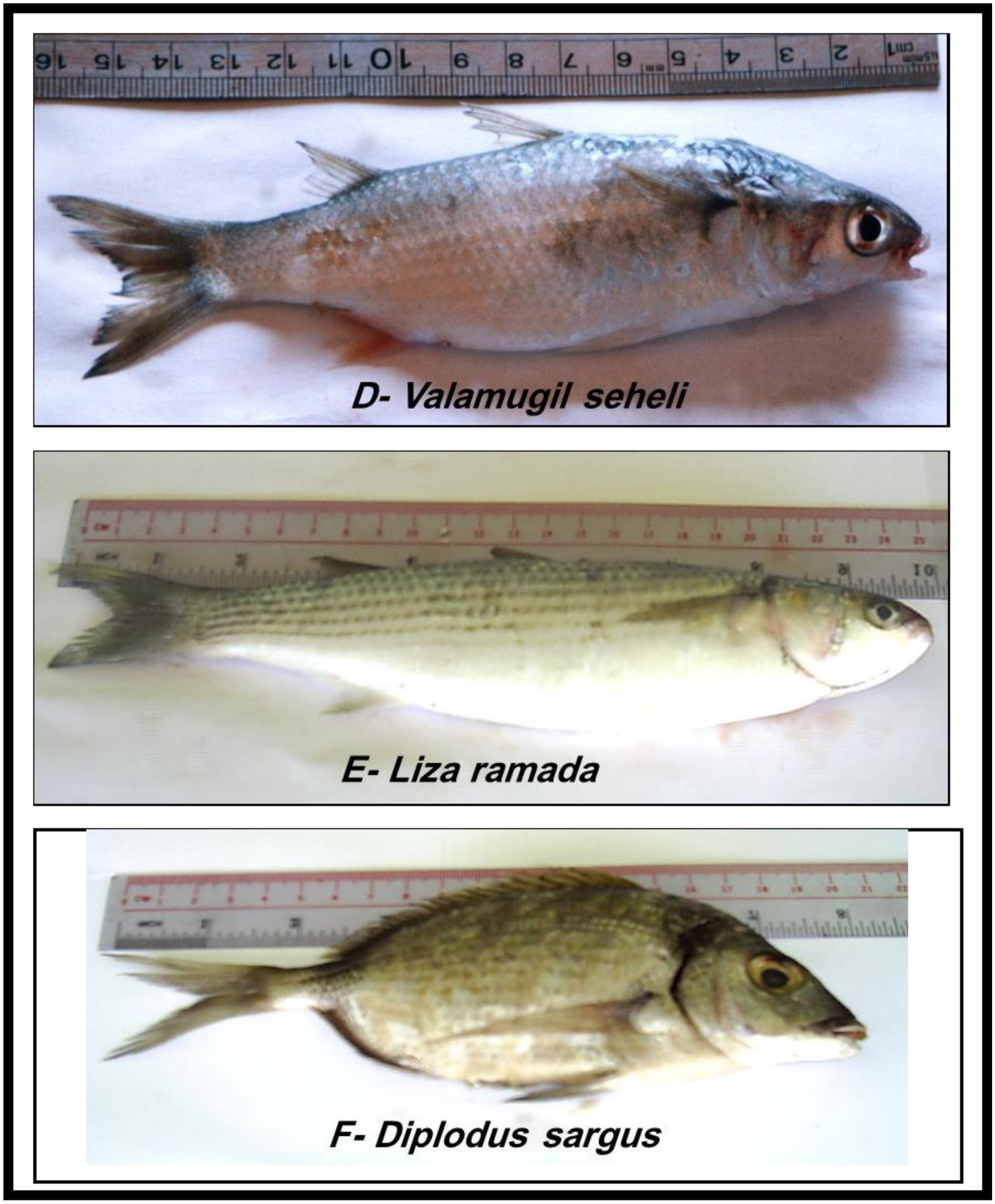


Figure 13ii: Photomicrographs showing the fish species examined during the present work.

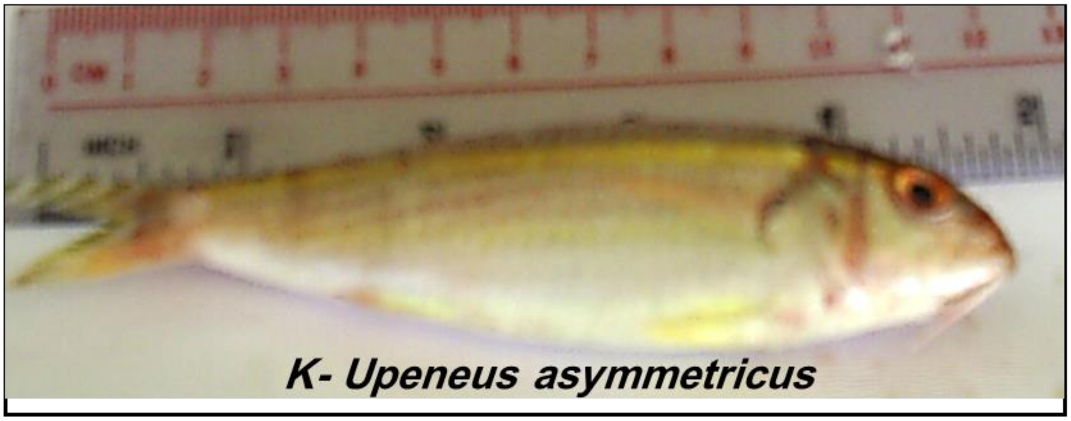
G- *Anguilla anguilla*

H- *Solea solea aegyptaica*

I- *Argyrosomus regius*

Figure 13iii: Photomicrographs showing the fish species examined during the present work.

J- *Lutjanus lineolatus*



K- *Upeneus asymmetricus*

L- *Sarotherodon galilaeus*

Figure 13iv: Photomicrographs showing the fish species examined during the present work.

Phylum: Arthropoda

Subphylum: Crustacea

Class: Maxillopoda

Subclass: Copepoda

1- Order: Cyclopoida

Family: Lernaeidae **Sproston *et al.*, 1950**

Genus: *Lernanthropus* **de Blainville, 1822**

Species: *Lernanthropus kroyeri* **van Beneden, 1851**

Genus: *Lamproglena* **von Nordmann, 1832**

Species: *Lamproglena minuta* **Capart, 1943**

2- Order: Poecilostomatoidea

Family: Ergasilidae **von Nordmann, 1832**

Genus: *Ergasilus* **von Nordmann, 1832**

Species: *Ergasilus briani* **Markewitsch, 1933**

Species: *Ergasilus lizae* **Kroyer, 1863**

Species: *Ergasilus australiensis* **Roubal, 1981**

Species: *Ergasilus mosulensis* **Rahemo, 1982**

Species: *Ergasilus celestis* **Roberts, 1970**

3- Order: Siphonostomatoida

Family: Caligidae

Genus: *Caligus* **Müller, 1785**

Species: *Caligus elongatus* **von Nordmann, 1832**

6- Morphological and anatomical studies of the parasitic copepods:

a- Living preparation:

Many specimens of parasitic copepods from the studied area were examined alive. Each parasite was transferred with a drop of freshly filtered either sea or fresh water onto a clean slide and covered with a cover slip. The specimens were flattened by withdrawn some water from the space under cover slip with a filter paper until the internal organs were visible.

b- Whole mount preparation:

1- Relaxation

Each parasitic copepod was transferred with a drop of fresh water onto a clean slide and covered with a coverslip then one or two drops of formaldehyde were added to fix the parasitic copepod on extension. The specimens were well flattened by pressure resulting from withdrawing some water and formaldehyde from the space between the coverslip and the slide with a filter paper. They were examined using oil immersion equipment.

2- Fixation and preservation

The collected copepods were fixed in 10% formaldehyde for whole mount preparations. The fixative solution is:

10% formalin

Formaldehyde	10ml
Distilled water	90 ml

3- Staining methods

Carmine stain is the most commonly used for the whole mount preparations. The clarity of a good whole mount results from the limitation of the carmine to the nuclei with the cytoplasmic areas completely or relatively free of the dye. When a whole mount fails to show sharp details, this usually indicates that the stain is too diffuse (over-stain), dehydrated in ascending series of alcohol (80%-100%), cleared in xylene and mounted in DPX. The carmine staining fluid is prepared by the following methods.

Grenacher's Borax Carmine & Aceto-carmine (Cited from Weesner, 1968).

Add 4 gm of Borax and 2 gm of Carmine to 100 cc. of distilled water and mix this mixture very well, allow to stand for several days with stirring daily. The solution may be boiled gently and mixed with 100 cc. of 70% alcohol, allow to stand for several days and filter. Add a crystal of thymol to avoid growing of bacteria.

4- Drawing and Measurements

Parasitic copepods were measured using a measuring ocular micrometer lens calibrated against a stage micrometer slide. Illustrations of the parasites were drawn free-hand with the aid of an eye-piece graticule. All measurements are in micrometers Figs. 14 and 15.

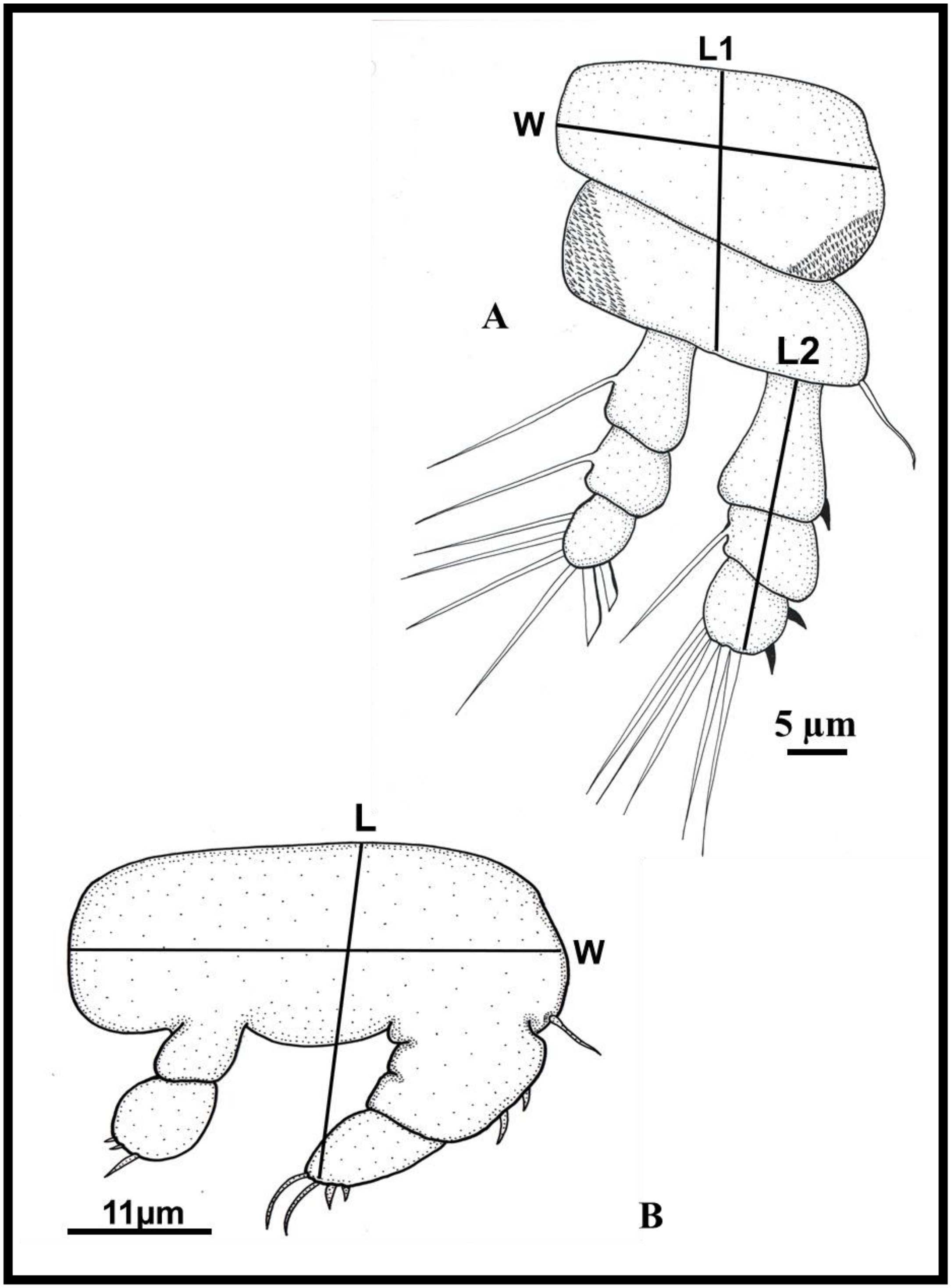


Figure 14: Schematic drawing of the thoracic legs of Genus *Ergasilus* (A) and Genus *Lamproglena* (B) showing the dimension of measurements. L, total length, L1, first length, L2, second length and W, total width.

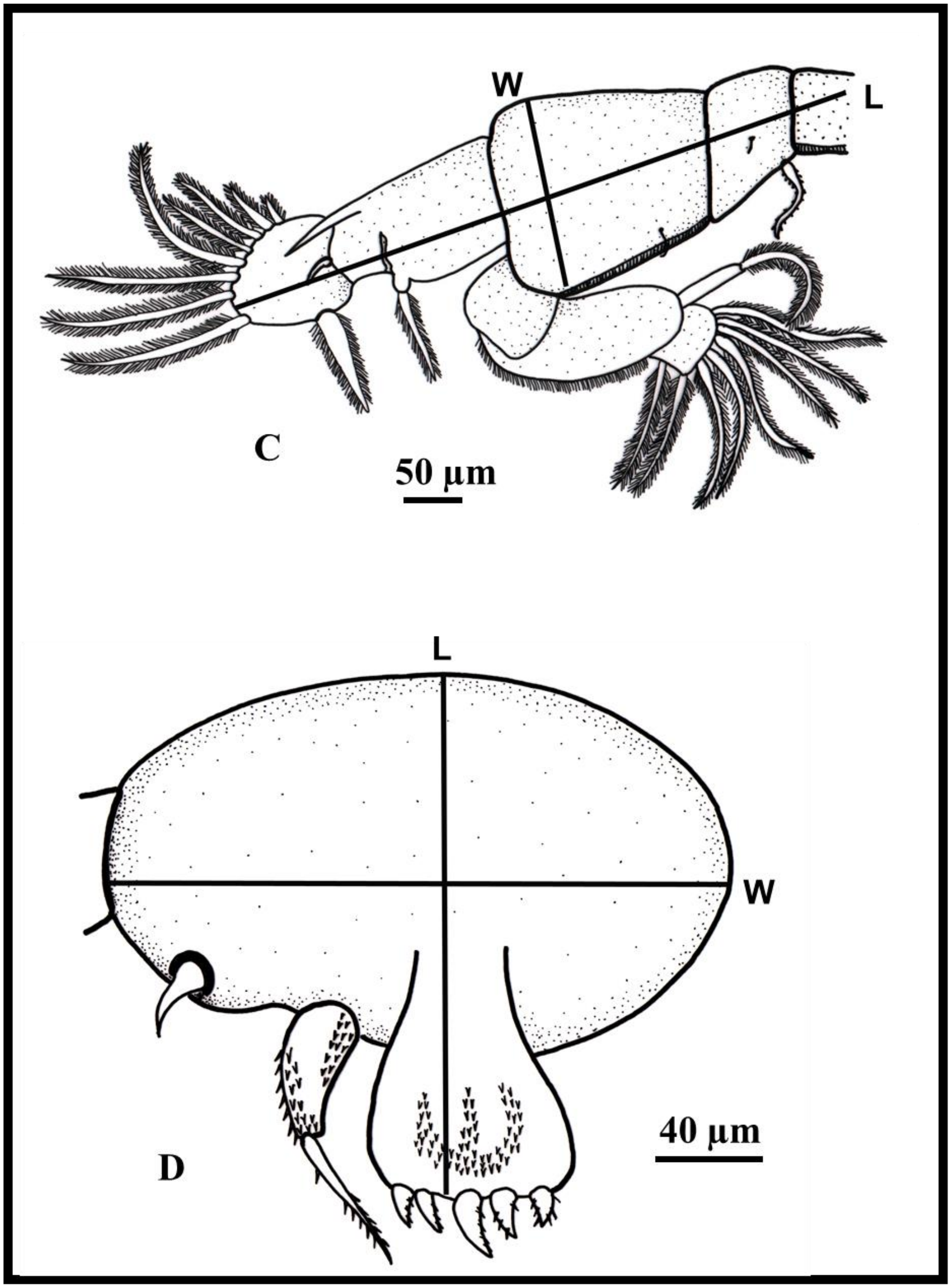


Figure 15: Schematic drawing of the thoracic legs of Genus *Caligus* (C) and Genus *Lernanthropus* (D) showing the dimension of measurements. L, total length and W, total width.

7- Ecological studies:

Water temperature, hydrogen ion concentration (pH), salinity and oxygen content were measured monthly at each site from April 2005 to March 2006.

1- Water temperature and pH

Water temperature was recorded with a simple thermometer. Water temperature is expressed in degree centigrade (°C). Hydrogen ion concentration (pH) of each monthly collected water sample was estimated electrometrically in the same day of sampling with an Orion electrode standardised to pH 7.0 with a pH buffer kit.

2- Water salinity

The water samples were collected in labelled, tightly-stoppered bottles and stored until examined in the laboratory. Water salinity was measured with an Aqua salinometer. The salinometer was standardised by using distilled water as zero salinity and commercially obtained salinity kits.

3- Water oxygen content

Oxygen content of the water sample was measured according to Winkler method as described by **Strickland and Parsons (1972)**.

The calculations were performed as follows:

Oxygen content (mg O_2/L) = $V_t\ N\ E \times 1000/\ V_s\ (V_b - V_w) \times V_b^{-1}$

Where:

V_t = volume of thiosulphate

N = normality of thiosulphate

E = equivalent weight

V_s = volume of sample

V_b = volume of bottle

V_w = volume of Winkler's reagent

8- Histological and Histopathological studies:

Specimens of infested and uninfested gills were also fixed in 10% formaldehyde for histopathological examinations and the routine haematoxylin and eosin staining technique. The specimens were washed under running tap water over-night to remove the excess of fixative solution. They were dehydrated in ascending series of alcohol, processed through xylene-alcohol and then cleared in two changes of xylene, 30 minutes each. They were transferred into a mixture of xylene and melted paraffin wax for 1 hour and then into two changes of pure paraffin wax, 30 minutes for each. The specimens were embedded in pure paraffin wax. Serial sections were cut at a thickness of 5 microns using rotary microtome. Sections were stained in haematoxylin and eosin according to **Drury and Wallington (1967)**.

In order to study the histological feature of non-infested gills and the histopathological changes of infested gill arches and primary gill lamellae caused by the infection were as the following techniques:

Wax embedding preparations.

Specimens of infested and uninfested gills were fixed in 10% formaldehyde.

Fixed specimens were washed under running tap water overnight to remove the excess of fixing solution. They were dehydrated in ascending series of alcohol, processed through xylene-alcohol and then cleared in two changes of pure xylene, 10 minutes each change. They were transferred into a mixture of xylene and melted paraffin for 10 minutes and then into two pure paraffin changes. The specimens were embedded in pure paraffin to form tissue blocks. Serial sections were cut at a thickness of 5 microns by using rotary microtome. Sections of uninfected and infected gill arch were stained accordingly the following stain:

a- Haematoxylin and eosin

Sections were stained in haematoxylin and eosin according to **Drury and Wallington (1967)**. Finally, the stained sections were cleared in xylene and mounted in canada balsam.

9- Control of parasitic copepods:

The parasitic copepods were subjected to three different types of treatment substances. The first substance is a natural plant extract, available in common market and cheap in price. The curcumin, *Curcuma longa* whish was identified according to **Maxwell, (2000)**. The rhizome is the portion of the plant used medicinally; it is usually boiled, cleaned, and dried, yielding a yellow powder. The extract of *Curcuma longa* was weighted and preserved in tightly closed glass vials.

The second substance is also a natural plant extract, available in common market and cheap in price. The Myrrh, *Commiphora molmol* which was identified according to **Ford, (1992)**. The rhizome is the portion of the plant used medicinally; it is usually boiled, cleaned, and dried, yielding a fine powder. The extract of Myrrh was weighted and preserved in tightly closed glass vials.

The third substance is a cheap and available synthetic drug obtained from Pharco pharmaceuticals, Egypt. Anthelmin contains 30% mebendazole.

The three substances were diluted to 10, 100, 500 and 1000ppm in distilled water and were added to the parasites in vitro.

For each substance, the mortality time of the parasitic copepods at different concentrations was detected.

10- The statistical analysis

Calculations of the parasitic copepods' prevalence and mean intensity were performed using the **SPSS** (version **11.5**), where the prevalence was the percentage of fishes infested and mean intensity the number of parasites per infested fish. Also, calculations of the simple linear regression of the increase in drug concentration and the time of parasitic copepods viability were performed using the **SPSS** (version **11.5**).

Individuals of the parasites at the level of studied species were counted monthly to calculate the prevalence and mean intensity infection in each fish species.

The relationship between the host body length, host body weight, host sex and the infestation level of the parasitic copepods was determined.

The statistical analysis of the prevalence and the mean intensity the parasitic copepods' were conducted using **SPSS** statistical facilities. In order to look for differences in the monthly fluctuations in the mean intensity and prevalence, **ANOVA**’s based on a General Linear Model followed by a Post Hoc

Test and Paired Sample Tests were conducted. In the case of significance differences, the Multiple Range Comparisons (Least Significance Difference, LSD) was applied in order to detect the distinct variances among the variable.

Also, significant differences in prevalence and the mean intensity among the seasons were again obtained using **ANOVA**’s based on a General Linear Model. The mean intensity of the parasitic copepods in relation to host length and weight classes and body sex were tested by applying simple linear regression analyses on the raw data and the significances of the regressions were obtained using **ANOVA**’S.

Variations in the mean intensity in relation to water temperature, pH, salinity and oxygen content at all sites were analysed statistically using **ANOVA**’s based on a General Linear Model. The significance of differences between prevalence and mean intensity values was calculated using one way analysis **ANOVA**’S or a Student-*t* Test for equality of means.

The degrees of freedom were obtained for most statistical tests and the critical P value was set at P=0.05* (significant), P=0.005** (highly significant) and P=0.0005*** (very highly significant). The arcsine square root of the prevalence and log transformed values of the mean intensity and abundance were used to normalise the data where appropriate.

RESULTS
AND
DISCUSSIONS

CHAPTER I

Morphological and Anatomical studies

Section I

Lernanthropus kroyeri

Genus: *Lernanthropus* de Blainville, 1822

Lernanthropus kroyeri van Beneden,1851

(1) Redescription:

The redescription of the copepodian parasite *Lernanthropus kroyeri* is illustrated in figures 16, 17, 18 & 19 and its measurements are shown in table (1).

This redescription is based on the study of twenty-eight living adult parasite individuals and ten mounted specimens of *Lernanthropus kroyeri* collected from the gill filaments of the spotted sea bass fish, *Dicentrarchus punctatus*.

(a) Adult female

Body of female parasite is elongated with an average total length, 1875 (1500-2190) µm excluding egg sacs (Fig. 16A & B).

Cephalthorax is narrower anteriorly, posterior margin slightly concave, posterolateral corners rounded, anterolateral extended ventrally as prominent, rounded lobes and curved ventrally on each side. It measures 632 (500-700) µm x 732 (600-950) µm.

Cephalon and first thoracic segment (Fig. 16) fused to form cephalothorax, slightly wider than long. The remaining thoracic segments fused forming genital complex. Genital complex is wider than long, 805 (630-900) µm x 1015 (950-1080) µm. There is deep constriction between cephalothorax and pregenital trunk. The latter is with prominent, rounded anterolateral corners and slightly convex lateral margins. Inside the genital complex there are two oval dorsal ovaries and large ventral cement glands. Two uniseriate, elongate egg sacs being 1655 (930-2010) µm x 207 (165-240) µm. Each sac emerged from a genital orifice containing

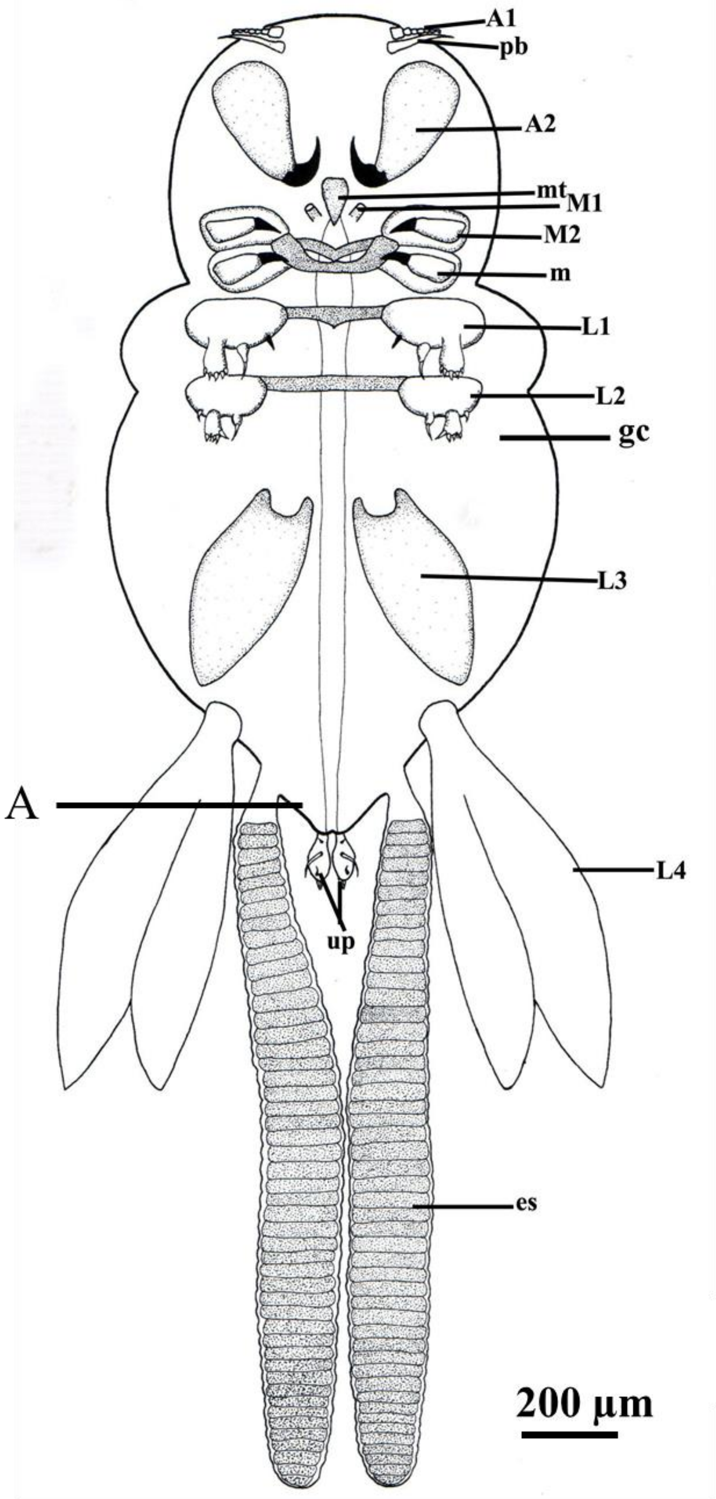


Figure 16A: Schematic drawing of ventral view of the female copepodian parasite, *Lernanthropus kroyeri* **van Beneden, 1851.**

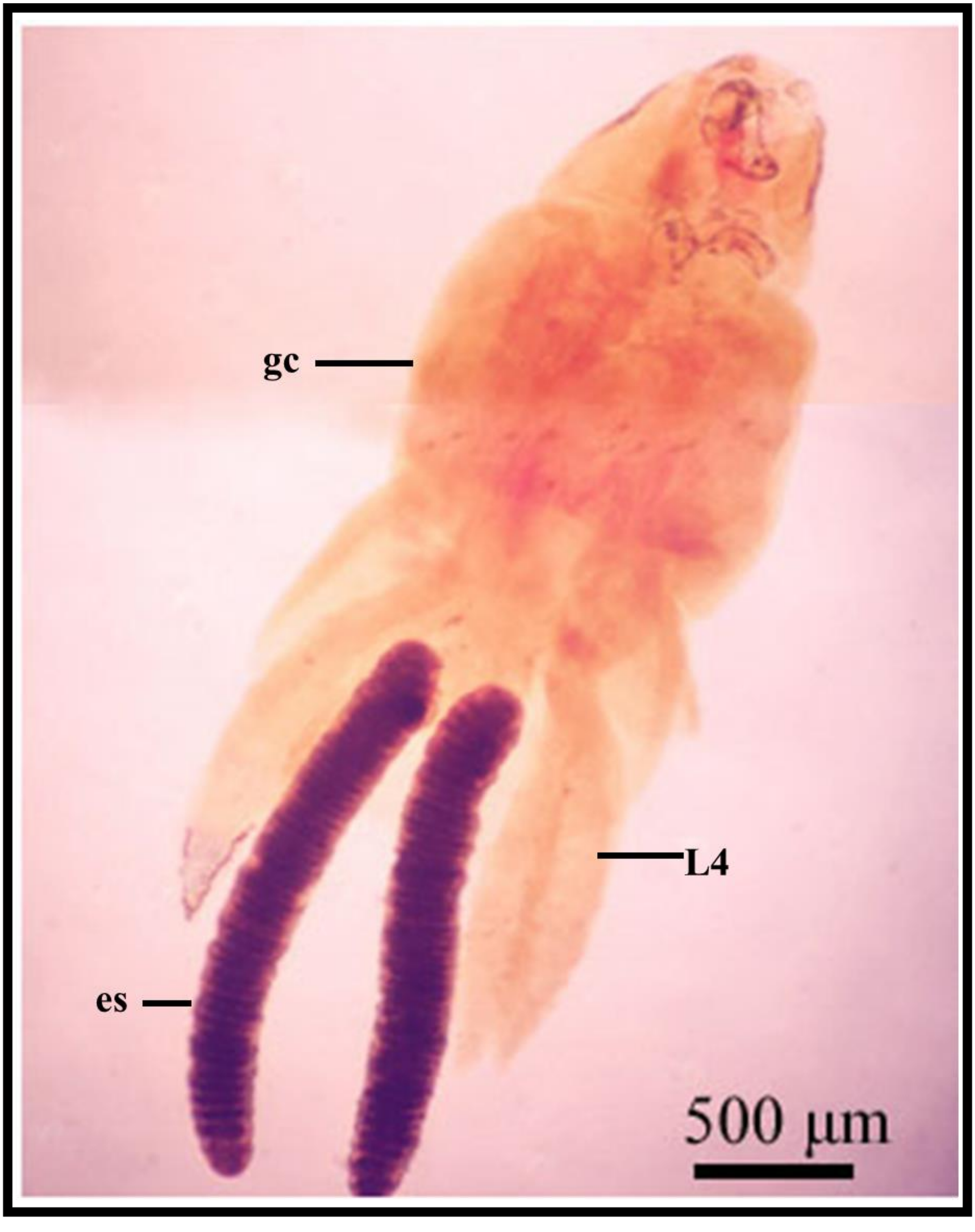


Figure 16B: Photomicrograph of the female copepodian parasite *Lernanthropus kroyeri* **van Beneden, 1851** stained with alum carmine.

54 (44-61) disc-shaped eggs (Fig. 16A & B). Abdomen is short and distinguished at beginning of dorsal plate extension (Fig 16A & B).

Antennule (first antenna) (Fig. 17A) seven-segmented; first segment with one seta on anterior margin, second segment with three setae (two on anterior margin and one on posterior side), third segment with one seta on anterior margin, fourth segment with three setae on anterior margin, fifth segment with one seta on anterior margin, sixth segment with two setae on anterior margin, terminal segment with 8 setae (three apical setae on anterior margin and five subapical setae on posterior side. Parabasal flagellum with broader base and is pointed distal part.

Antenna (second antenna) (Fig. 17B) is two-segmented and sturdy, subchelate; corpus large, tapering distally with single small myxal process. Subchela is curving inwards with a spiniform process on inner surface close to the base.

Maxillule (first maxilla) (Fig. 17C) is biramous. Exopod is short, rounded in distal end with two terminal small spines and subterminal horny spine and setule cover. Endopod subcylindrical with three apical strong spiniform processes, setule cover.

Maxilla (second maxilla) (Fig. 17D) is uniramous, brachiform and two-segmented: proximal segment (= lacertus) large elongate unarmed; second segment (= brachium) slender with two subterminal spines (one on anterior margin and other on posterior margin), rows of minute spines and terminal spiny claw armed with two sharp denticles rows, each comprising 13 teeth.

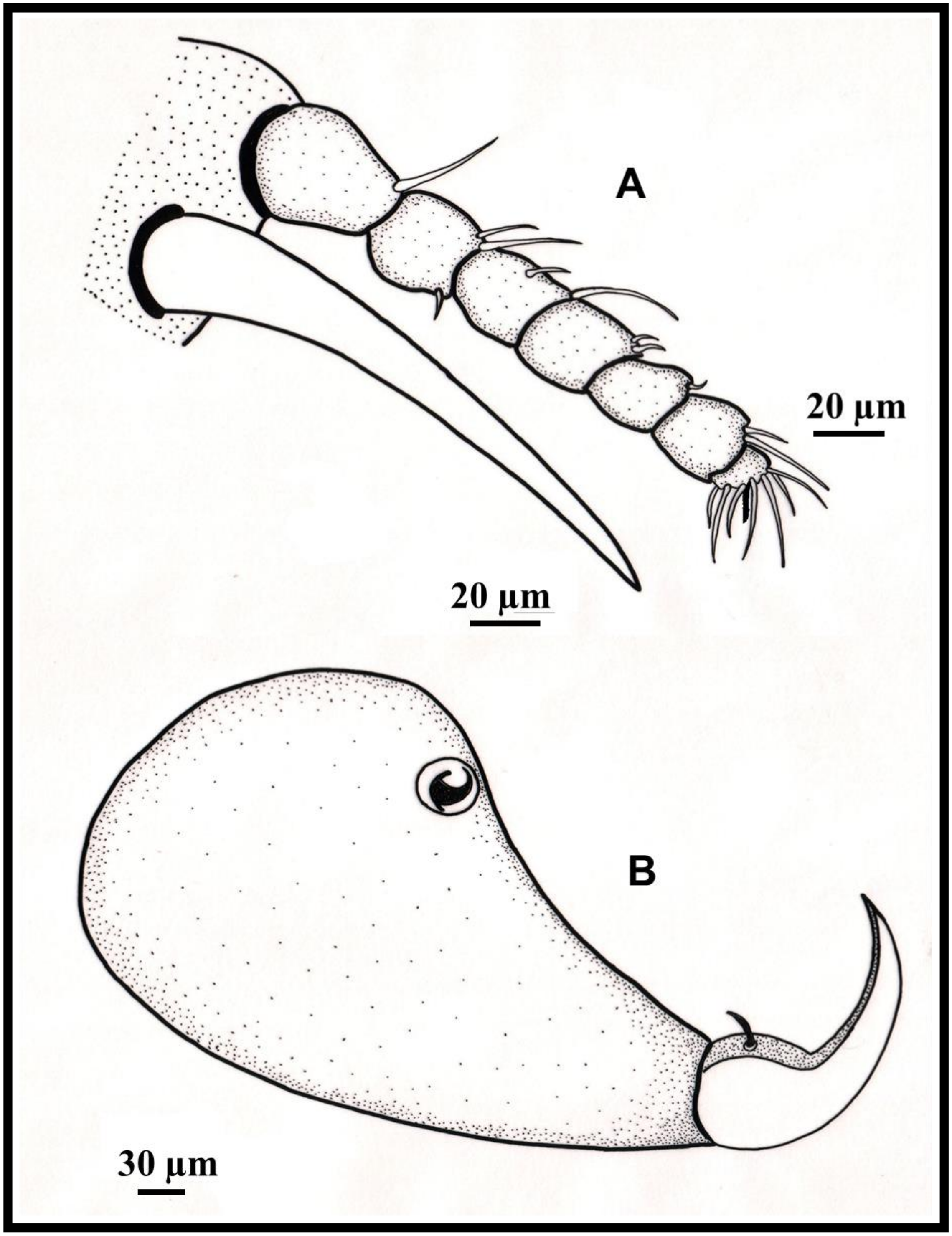


Figure 17A&B: Schematic drawing of the antennule and parabasal flagellum, (A) and antenna (B) of the female copepodian parasite, *Lernanthropus kroyeri* **van Beneden, 1851.**

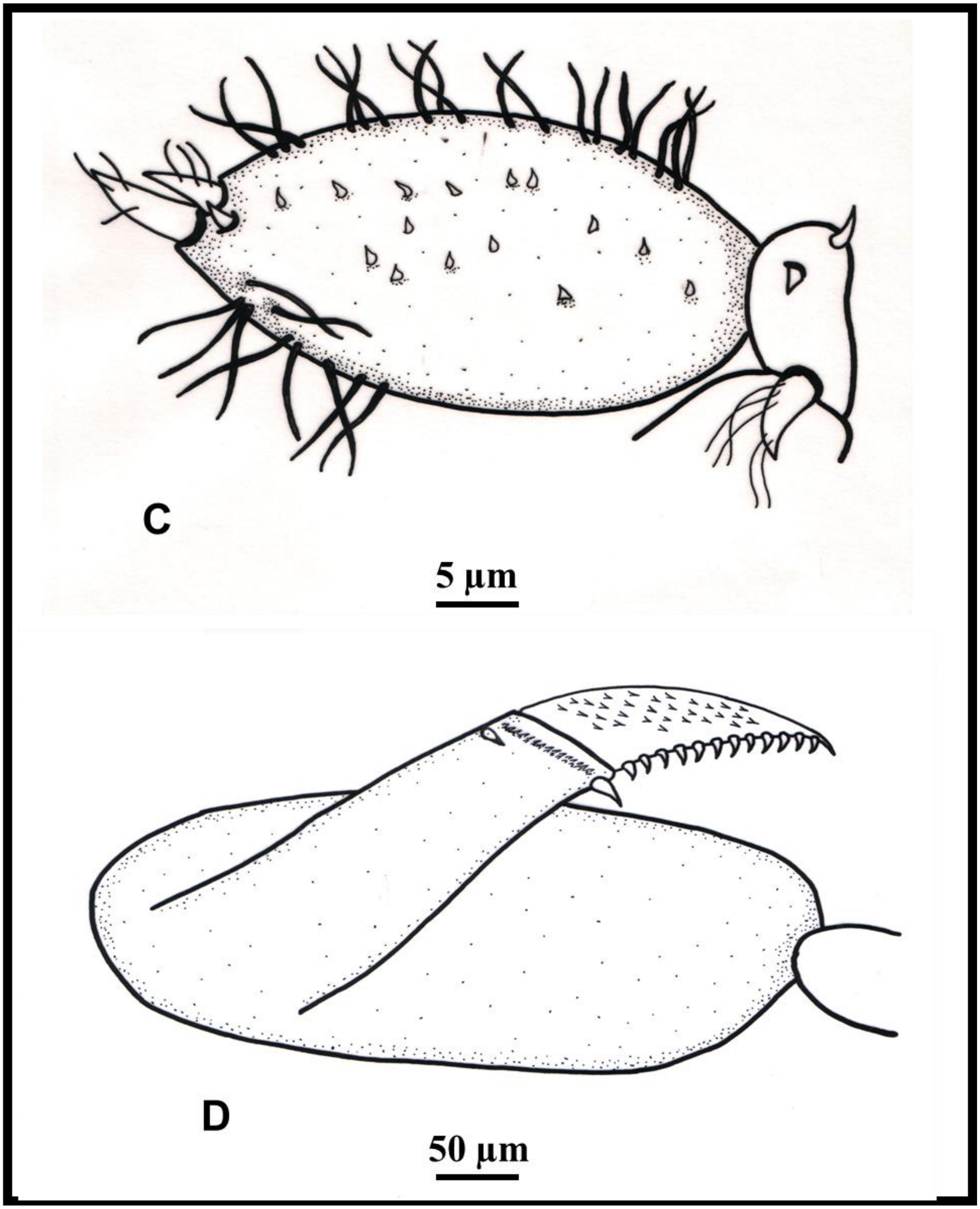


Figure 17C&D: Schematic drawing of the maxillule, (C) and maxilla (D) of the female copepodian parasite, *Lernanthropus kroyeri* **van Beneden, 1851.**

Maxilliped (Fig. 17E) subchelate, corpus stout unarmed, subchela armed with single subterminal seta on inner margin and claw apicaly directed with longitudinal ridges.

Mouth tube (Fig. 17F) is conical with tip directed posteriorly, situated between maxillae, the labrum shorter than labium, with some integumental processes and with tube-like buccal stylet. Labium is tapering towards tip with denticulate margins. Mandibles tip with flattened shaft bearing 8 recurved teeth. Uropod (Fig. 17G) is unsegmented, fusiform with 5 setules (two terminal and three subterminal) and being 122 (100-130) μm x 53.75 (40-65) μm

First thoracic leg (Fig. 17H) is biramous: exopod broad and distally armed with 5 terminal spines (pilose in female); endopod smaller, tapering distally, margins denticulate.

Second thoracic leg (Fig. 17I) is biramous: exopod with five distal spines (naked except 1st, 4th and 5th spines) and denticles covering medial region of frontal surface. Endopod denticulate armed with short apical seta (pilose in female). Sympod armed with pilose seta.

Third thoracic leg (Fig. 16A) long is unarmed, protruding posteroventrally from medial region of genital complex, parallel to each other. Fourth thoracic leg is bilobed and unarmed, protruding ventrolaterally from distal region of genital complex.

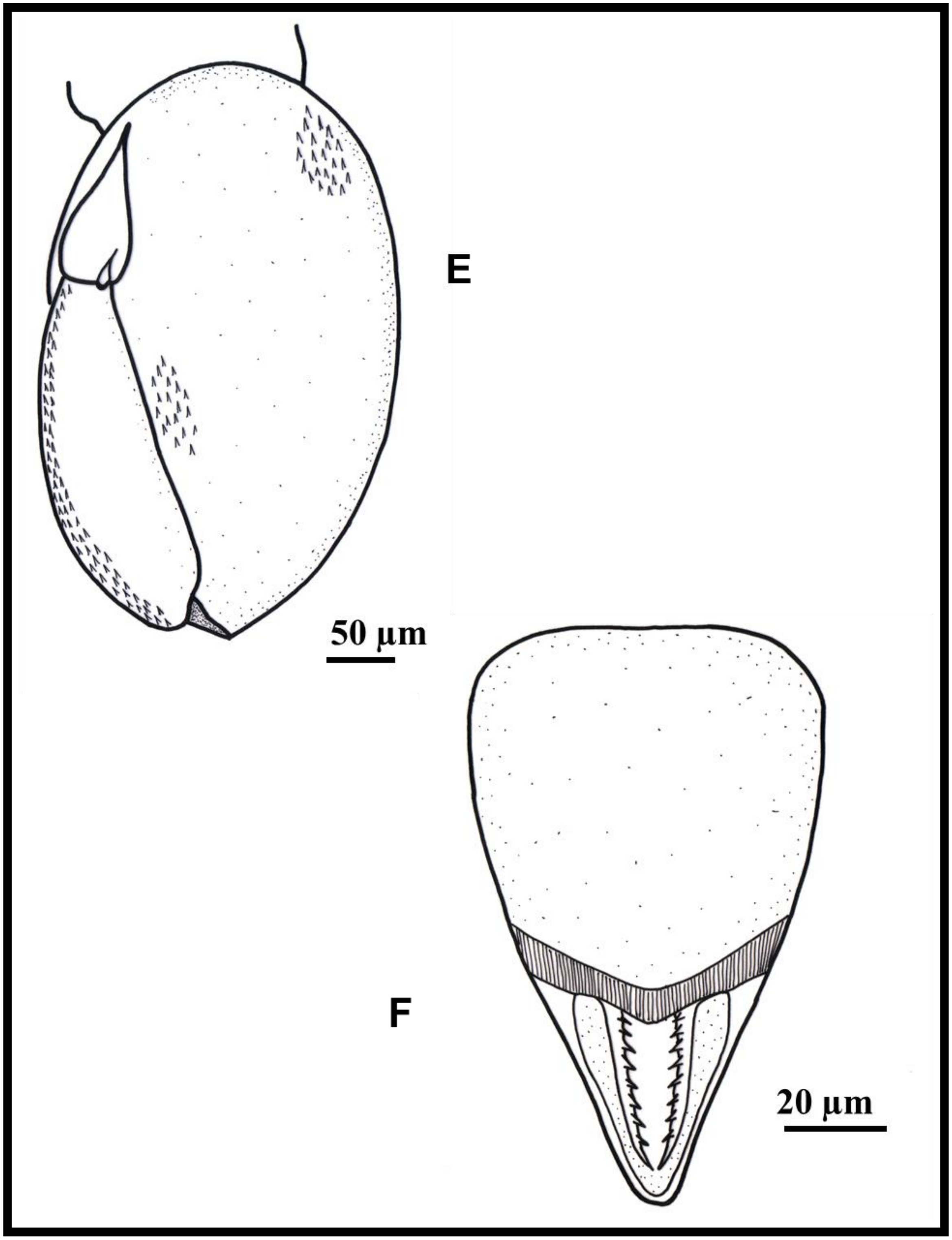


Figure 17E&F: Schematic drawing of the maxilliped, (E) and mouth tube (F) of the female copepodian parasite, *Lernanthropus kroyeri* **van Beneden, 1851.**

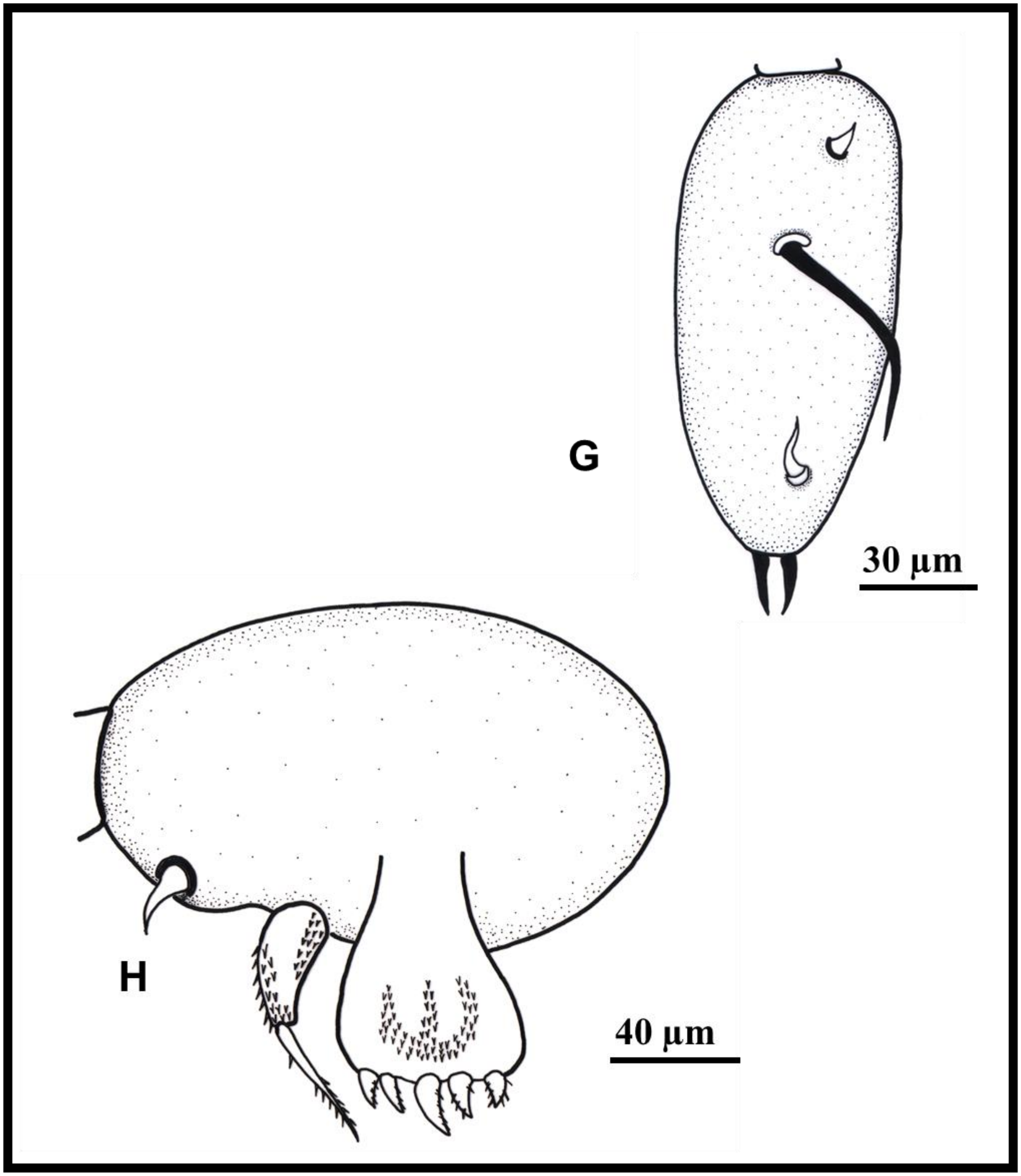


Figure 17G&H: Schematic drawing of the uropod, (G) and first thoracic leg (H) of the female copepodian parasite**,** *Lernanthropus kroyeri* **van Beneden,1851***.*

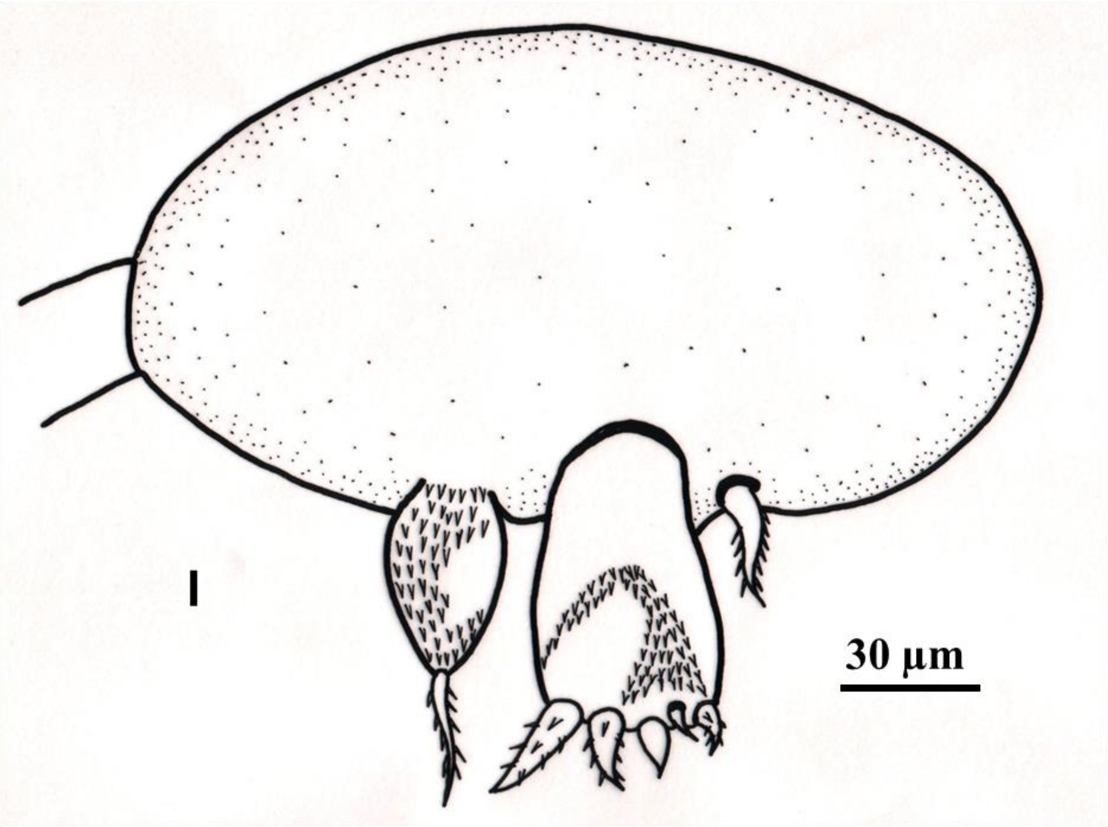


Figure 17I: Schematic drawing of the second thoracic leg, (I) of the female copepodian parasite**,** *Lernanthropus kroyeri* **van Beneden, 1851.**

(b) Adult male

Body of male parasite is elongated (Fig. 18) with average total length of 1548 (1400-2610) µm. Cephalon and first thoracic segment (Fig. 18) fused to form cephalothorax, slightly wider than long. The remaining thoracic segments fused forming genital complex. Genital complex is slightly identical in length and width, being 644 (455-1300) µm x 631(480-1100) µm.

Abdomen (Fig. 18) is short could not be clearly delimited in male. Inside the abdomen of some males, two spermatophores in posterior vasa deferentia were seen.

First antenna (Fig. 19A) seven-segmented; first segment with one seta on posterior side, second segment with three setae (two on anterior margin and one on posterior side), third segment with one seta on posterior margin, fourth segment with one seta on anterior and two on posterior margins in male, fifth segment with one seta on posterior margin, sixth segment with two setae on anterior margin; terminal segment with 8 setae (two apical, two subapical setae on anterior margin and one apical, three subapical setae on posterior side).

Second antenna (Fig. 19B) is sturdy and two-segmented, subchelate; corpus large, tapering distally with two processes on inner surface; subchela curving inwards with a spiniform process on inner surface close to the base.

First maxilla (Fig. 19C) is biramous: exopod short, rounded in distal end with terminal strong spiniform process. Endopod subcylindrical with three apical strong spiniform processes, spine cover in male.

Second maxilla (Fig. 19D) is uniramous, brachiform, two-segmented: proximal segment (= lacertus) large elongate unarmed; second segment (= brachium) slender with two subterminal spines (one on anterior margin and other

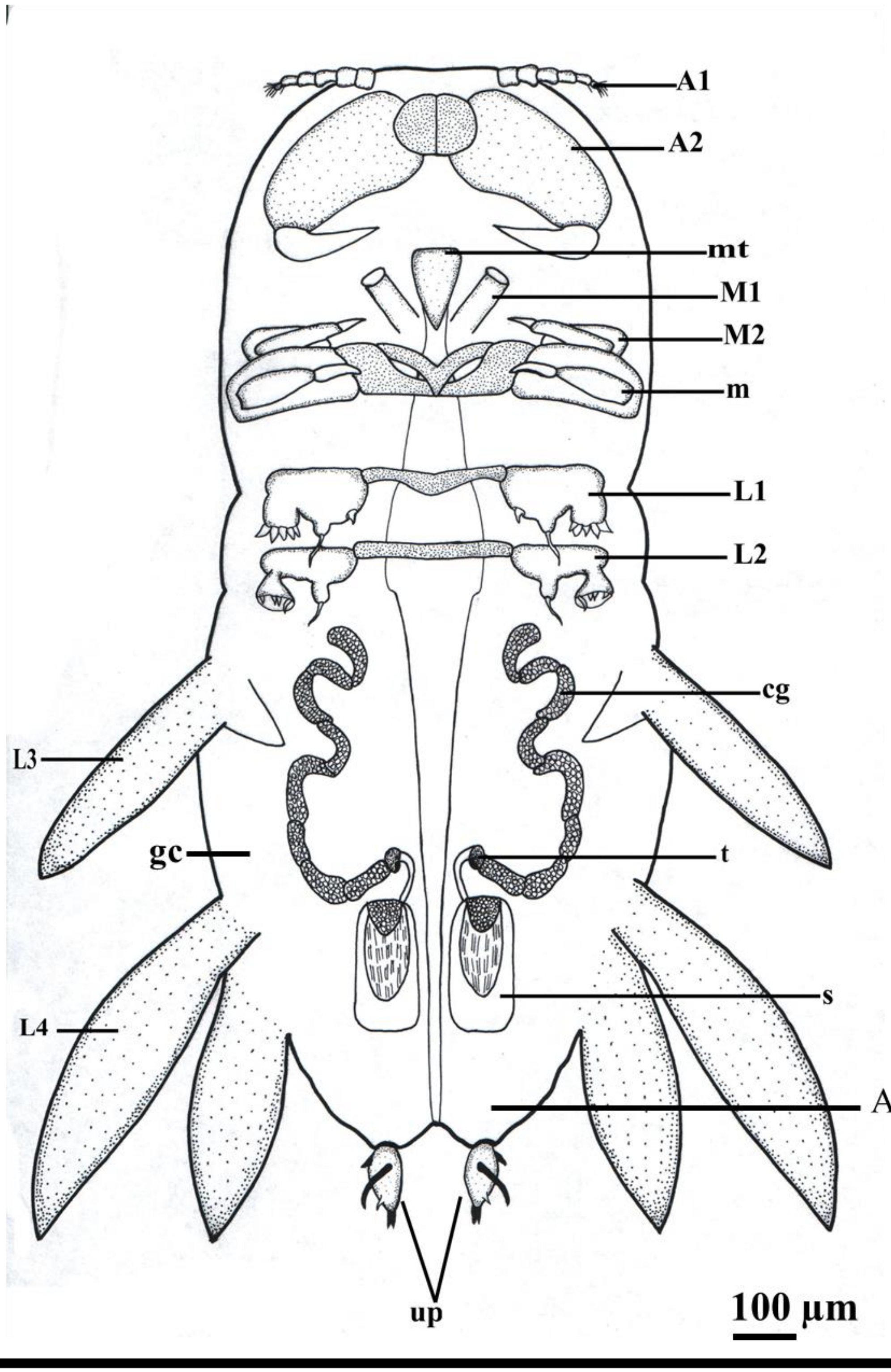


Figure 18A: Schematic drawing of ventral view of the male copepodian parasite, ***Lernanthropus kroyeri*** **van Beneden, 1851.**

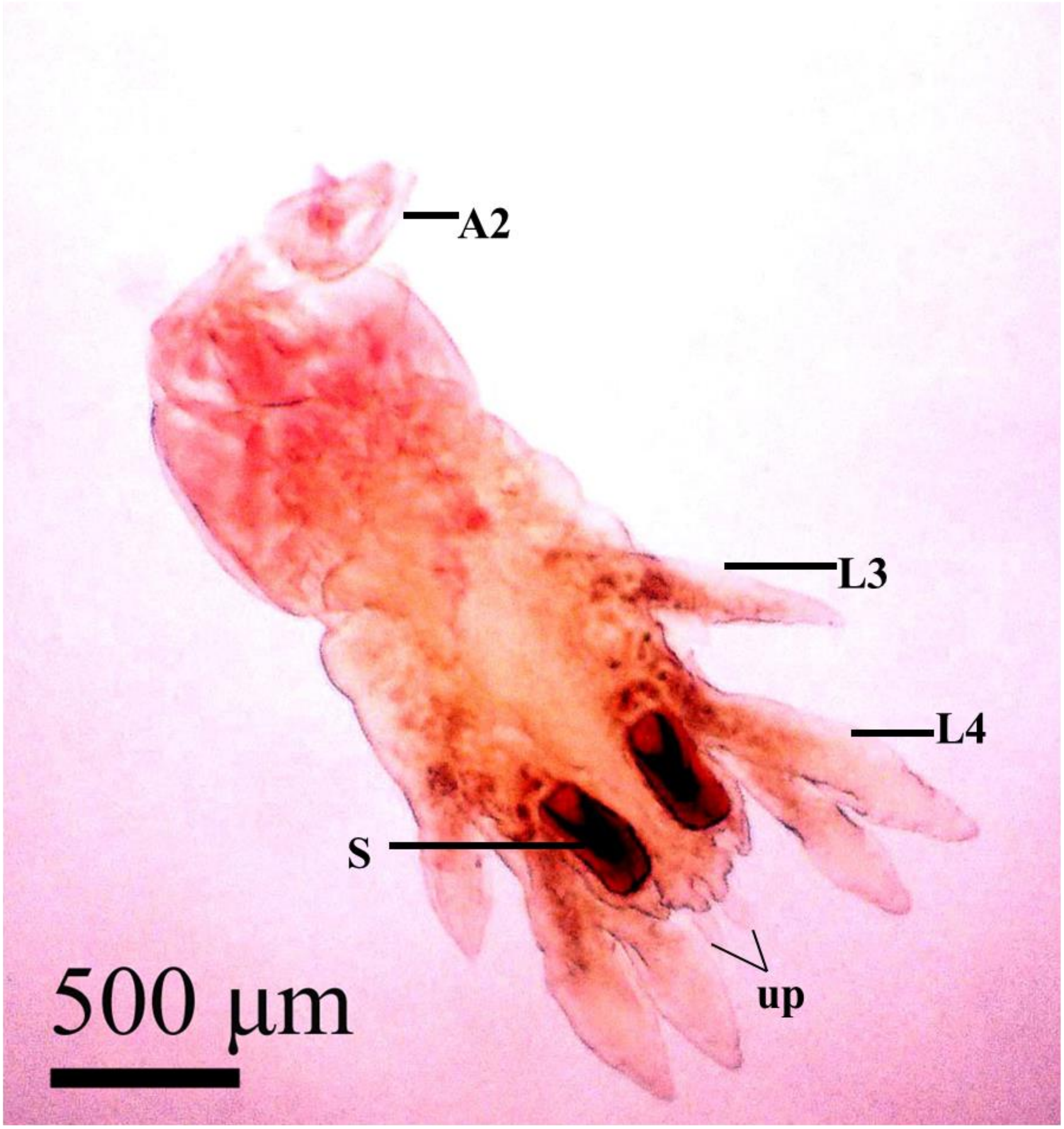


Figure 18B: Photomicrograph of the male copepodian parasite *Lernanthropus kroyeri* **van Beneden, 1851** stained with alum carmine.

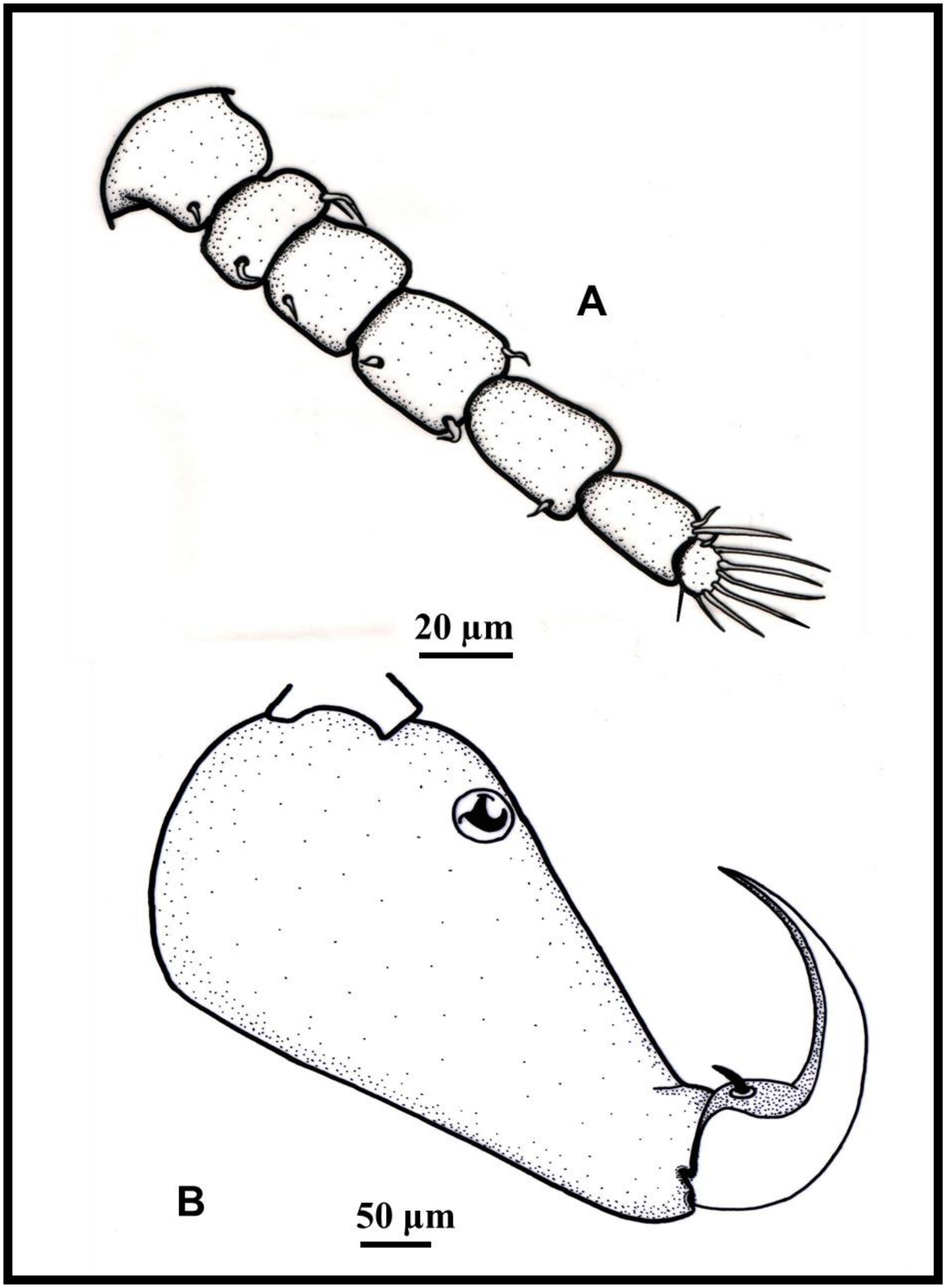


Figure 19A&B: Schematic drawing of the antennule, (A) and antenna (B) of the male copepodian parasite**, *Lernanthropus kroyeri* van Beneden, 1851.**

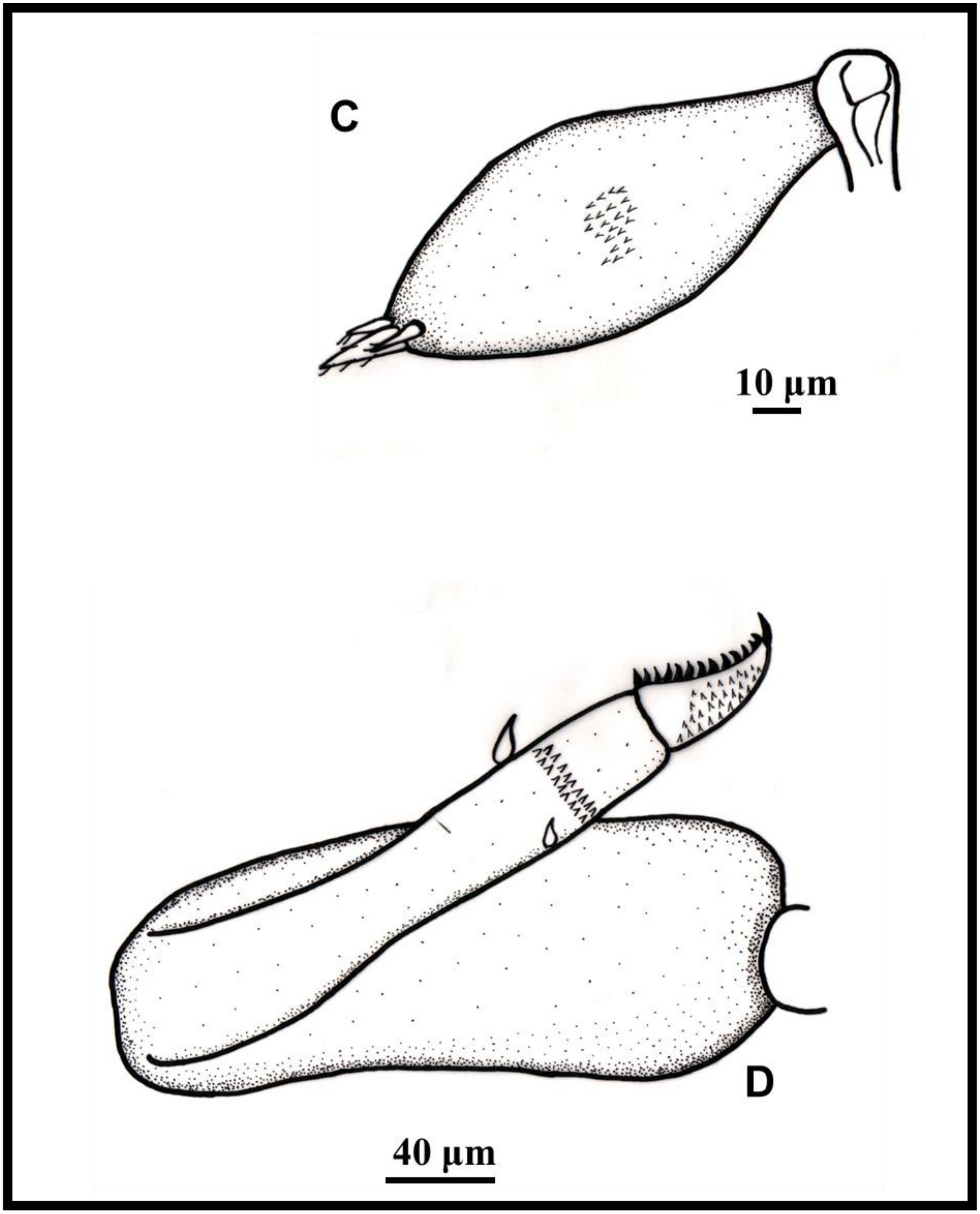


Figure 19C&D: Schematic drawing of the maxillule, (C) and maxilla (D) of the male copepodian parasite**, *Lernanthropus kroyeri* van Beneden, 1851.**

on posterior margin), rows of minute spines and terminal spiny claw armed with two sharp denticles rows, each comprising 14 teeth, ended with apical long spine in male only .

Maxilliped (Fig. 19E) is subchelate, corpus stout unarmed, subchela consisting of marginally denticulate shaft in male only, armed with single subterminal seta on inner margin and claw apicaly directed with longitudinal ridges.

Uropod (Fig. 19F) is unsegmented, fusiform with 5 setules (two terminal and three subterminal) and denticles covering most of distal half in male only being 80 (75-90) µm x 43 (40-55) µm.

First thoracic leg (Fig 19G) is biramous: exopod broad and distally armed with five terminal spines (naked except 3rd and 4th spines); endopod small, tapering distally, margins denticulate, with apical pilose seta longer than exopod. Sympod is denticulated, armed with spiniform process near base and medial to endopod.

Second thoracic leg (Fig. 19H) is biramous: exopod with five distal spines (naked- shape) and denticles covering distal base. Endopod denticulate armed with short apical seta (naked in male). Sympod armed with naked seta, near base of exopod.

Mouth tube (Fig. 19I) is conical with tip directed posteriorly, situated between maxillae, the labrum shorter than labium, with some integumental processes and with tube-like buccal stylet.

Labium is tapering towards tip with denticulate margins. Mandibles tip with flattened shaft bearing 8 recurved teeth.

Third thoracic leg (Fig. 18) is long, unarmed, protruding posteroventrally from medial region of genital complex, bilobed with long lateral lobe.

Fourth thoracic leg (Fig. 18) is bilobed and unarmed, protruding ventrolaterally from distal region of genital complex.

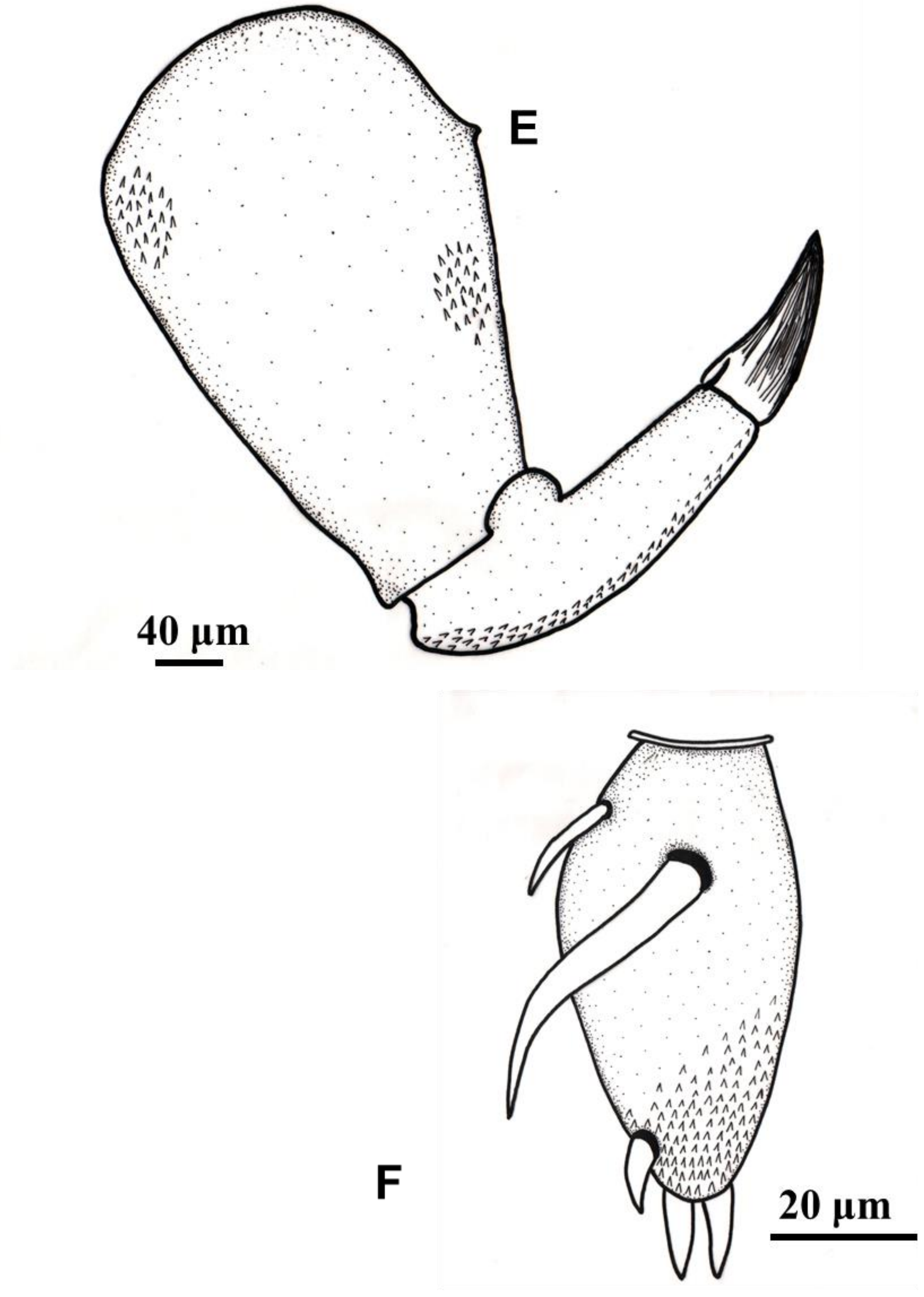


Figure 19E&F: Schematic drawing of the maxilliped, (E) and uropod (F) of the male copepodian parasite**,** ***Lernanthropus kroyeri*** **van Beneden, 1851.**

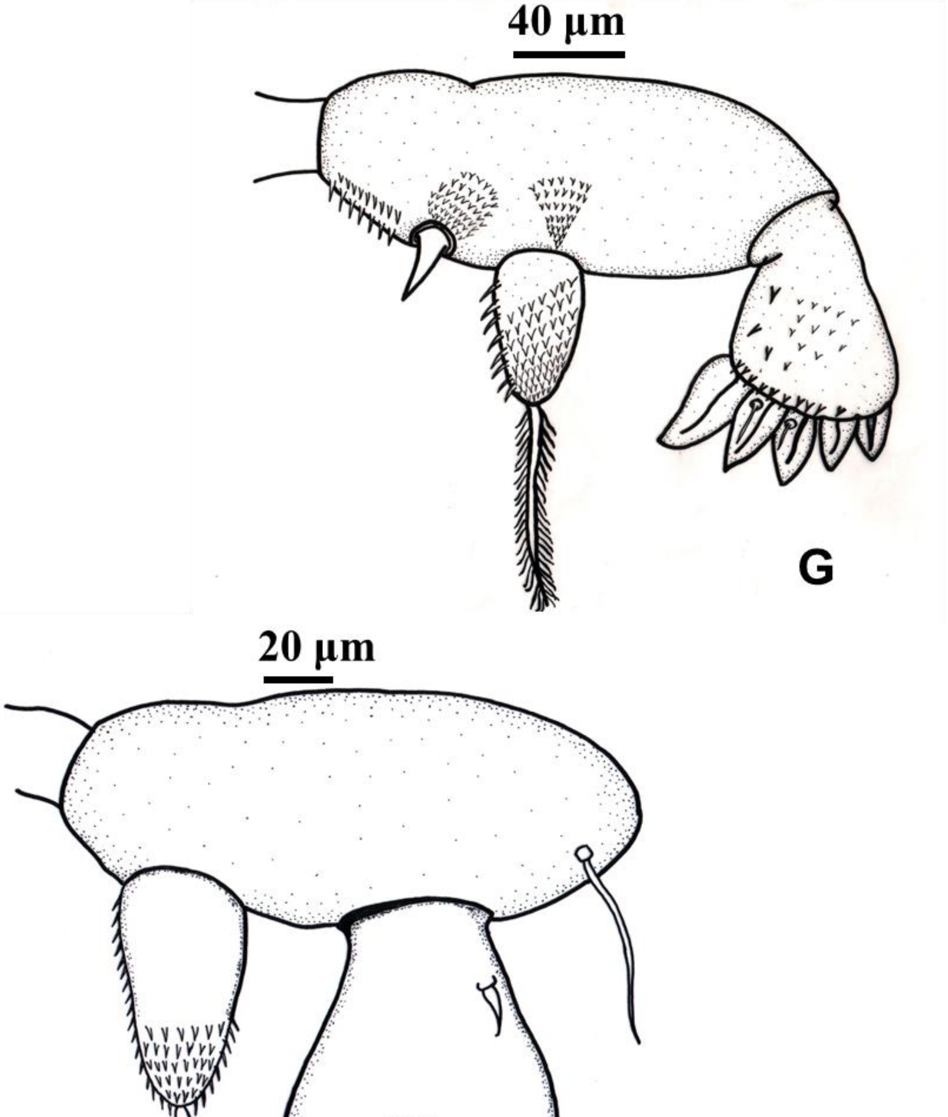


Figure 19G&H: Schematic drawing of the first thoracic leg, (G) and the second thoracic leg (H) of the male copepodian parasite**,** ***Lernanthropus kroyeri*** **van Beneden, 1851.**

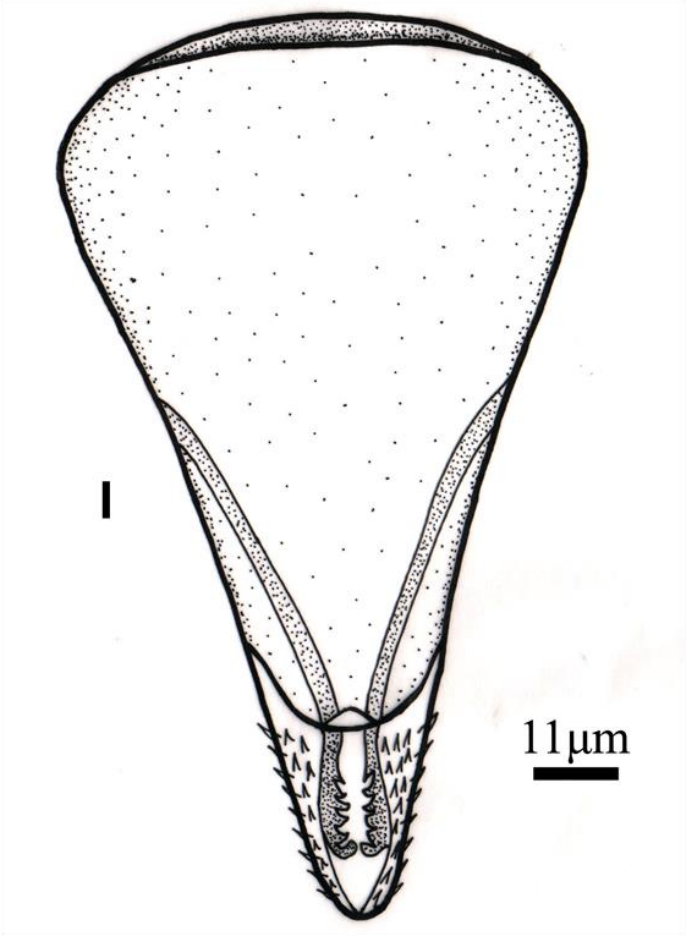


Figure 19I: Schematic drawing of the mouth tube, (I) of the male copepodian parasite, ***Lernanthropus kroyeri*** **van Beneden, 1851.**

(2) Discussion:

The anatomical structure and dimensions demonstrated in the present description are evidently to suggest that the present species described here in belongs to genus *Lernanthropus* **de Blainville, 1822** according to the following generic criteria which based by **Yamaguti and Yamasu (1960), Hewitt (1968**) and **Kabata (1971**). These generic morphological criteria are:

Cephalothorax of the female is fused with first leg-bearing segment, dorsally with well-developed shield curved ventrally on each side. Fourth leg-bearing segment have large or small dorsal or ventral plate. Abdomen is small, indistinctly one or two segmented. Egg sacs are straight or irregularly coiled under plate of fourth leg-bearing segment and uniseriate. First antenna is uniramous, usually indistinctly segmented, with or without parabasal flagellum. Second antenna is subchelate. Mouth and mouth parts are siphonostome. Maxilliped is subchelate. First leg is small, biramous and rami one-segmented. Second leg is similar to first. Third is modified into variously shaped, plate-like structure and with or without foliaceous outgrowths of sympod and exopod. Fourth leg is unsegmented, bilobed, large or small and with or without filiform tips. Fifth leg is uniramous, small or absent. Uropods are present. Cephalothorax of the male is with dorsal shield flat. There is no plate on fourth leg-bearing segment. Abdomen is one-segmented. First antenna is uniramous and usually indistinctly segmented. Second antenna is subchelate. Mouth and mouth parts are siphonostome. Maxilliped is subchelate. First leg is small, biramous and rami one-segmented. Second leg is similar to first. Third and fourth legs are uniramous or bilobed, lobes are flat or vermiform and comparatively long. Fifth leg is absent. Uropods are present.

The present individuals of the parasitic copepod *Lernanthropus kroyeri* **van Beneden, 1851** infesting gills of spotted sea bass fish, *Dicentrarchus punctatus* were collected previously, from the sea bass fish, *Dicentrarchus labrax* in Egyptian Mediterranean coast by **Abu Samak (2004**) Thus, the present study of *Lernanthropus kroyeri* represents the first record from the spotted sea bass fish, *Dicentrarchus punctatus* in Egyptian Mediterranean coast.

Lernanthropus kroyeri has been recorded from many localities along the coast of Europe, the Irish Sea and the coast of Norfolk on the sea bass fish, *Dicentrarchus labrax* **(Wilson, 1922)**. An unusual record *Lernanthropus kroyeri* was identified as being parasitic on *Lutianus griseus* from the Gulg of Mexico **(Bere, 1936)**. A single unconfirmed study, *Lernanthropus kroyeri* was listed among the parasitic fauna of the sea bass fish, *Dicentrarchus labrax* caught from Bardawil Lagoon **(Paperna and Lahav, 1975)**. *Lernanthropus kroyeri* has been recorded from many localities along the coast of Europe, from the Adriatic to the southern North Sea appears to be *Dicentrarchus labrax* **(Kabata, 1979)**. Most of the investigations about *Lernanthropus kroyeri* of the sea bass fish, *Dicentrarchus labrax* in Turkey are focused on the Aegean Coast (**Tokşen, 1999**). From the eastern Mediterranean coast, *Lernanthropus kroyeri* **van Beneden, 1851** was recorded for the first time on gills of the sea bass fish, *Dicentrarchus labrax* **(Abu Samak, 2004)**.

The present description of the parasitic copepod *Lernanthropus kroyeri* **van Beneden, 1851** infesting gills of spotted sea bass fish, *Dicentrarchus punctatus* is similar to the description of **Abu Samak (2004)**. The present specimen's dimensions are smaller than the specimens redescribed by **Kabata (1979)** and **Abu Samak (2004**) except 2nd maxilla, maxilliped and 4th leg in both sexes are larger as shown in table (1). **Kabata and Gusev (1966)** found that some species might differ considerably in size, and they suggested that the size differs in the same parasite, depending on the geographic area. The armature setation number on the segments of female and male first antenns in the present study differs from that of *Lernanthropus gisleri* which is similar to *L. kroyeri* in the study by **Kabata (1979)** and resembles that of the young female and male *L. kroyeri* in the study by **Cabral *et al*. (1984)**. The postantennal processes have been recently discovered only in 5 species belonging to *Lernanthropus* by **Olivier and Niekerk (1995a and 1995b)** and **Olivier *et al*. (1997)**.

The availability of the material and the use of SEM of both sexes of *L. kroyeri* by **Abu Samak (2004)** have led to supplement the new information of *L. kroyeri* as follows: Presence of myxal process on the second antenna; of postantennal processes posterior to the second antenna in both sexes; of armature on both labium and labrum; of two buccal stylets; of 8 teeth on tip of the mandibles in both sexes and of single subterminal seta on inner margin of maxilliped subchela in both sexes; difference in type and density of cuticular processes covering the dorsal surface of cephalothorax between two sexes; in position of the first antenna setation between two sexes; in armature on postoral processes, first maxillae and claw of the second maxillae in both sexes; in number of myxal process on the second antenna in both sexes; in type of exopodal setation and denticles pattern covering first and second thoracic legs between two sexes; besides the male endopod of first thoracic leg with apical pilose seta longer than the exopodal length.

Abbreviations of the copepodian parasite (*Lernanthropus kroyeri*) figures

A	Abdomen
A1	Antennule (1st antenna)
A2	Antenna (2nd antenna)
cg	Cement glands
es	Egg sac
gc	Genital complex
L1	1st thoracic leg
L2	2nd thoracic leg
L3	3rd thoracic leg
L4	4th thoracic leg
m	Maxilliped
M1	1st maxilla
M2	2nd maxilla
mt	Mouth tube
Pb	Parabasal flagellum
s	Spermatophore
t	testis
up	Uropod

Table (1): Comparison between the present species of *Lernanthropus kroyeri* and the previously description of this species collected from Egypt.

Author(s)	**Abu Samak (2004)**		**The present specimens**	
Host species	*Dicentrarchus labrax*		*Dicentrarchus punctatus*	
Locality	Egypt		Egypt	
Sex	Female	Male	Female	Male
Maximum lengthxMaximum width	3763x789	2094x610	1875x1377	1548x646
First antenna	179x38	214x45	112x24	161x30
Parabasal flagellum	173x25	151x31	150x25	-
Second antenna	275x225	315x178	384x255	364x210
Mouth tube	144x80	144x75	111x68	110x66
First maxilla	133x40	132x41	37.5x17.5	87.5x32.5
Second maxilla	180x67	205x68	369x170	260x95
Maxilliped	190x97	233x135	349x202	350x195
First thoracic leg	176x208	137x143	183x112	161x72.5
Second thoracic leg	182x162	160x143	181x94	123x56.2
Third thoracic leg	694x369	523x213	490x240	370x102
Fourth thoracic leg	1638x300	745x268	1847x466	1069x296

CHAPTER I

Section II

Lamproglena minuta

Genus: *Lamproglena* von Nordmann, 1832

Lamproglena minuta Capart, 1943

(1) Redescription:

The redescription of the copepodian parasite, *Lamproglena minuta* is illustrated in figures 19, 20 & 21 and its measurements are shown in table (2).

The following redescription is based on the study of ninteen living adult and twenty mounted specimens of *Lamproglena minuta*, collected from the gill filaments of the *Tilapia galilaea*; *Sarotherodon galilaeus.*

(a) Adult female

The body of the adult female *Lamproglena minuta* is elongated, consists of three distinct parts: cephalothorax, trunk and abdomen. It measures 2460 (1800-2820) µm in total length excluding egg sacs. The maximum body width is 617.5 (430-750) µm (Fig. 20).

Cephalothorax (Fig. 20) is incorporating first leg-bearing segment, of about equal length and width, slightly wider in its posterior half, with marked indentations at about midlength of lateral margins, separated from trunk by deep, neck-like constrictions. It measures in female 550 (490-610) µm in length and 520 (450-600) µm in width.

The trunk (Fig. 20) is oval, longer than wide and subdivided into three regions, first consisting of fused second and third leg-bearing segments marked off posteriorly by definite indentations on lateral margins, second consisting of

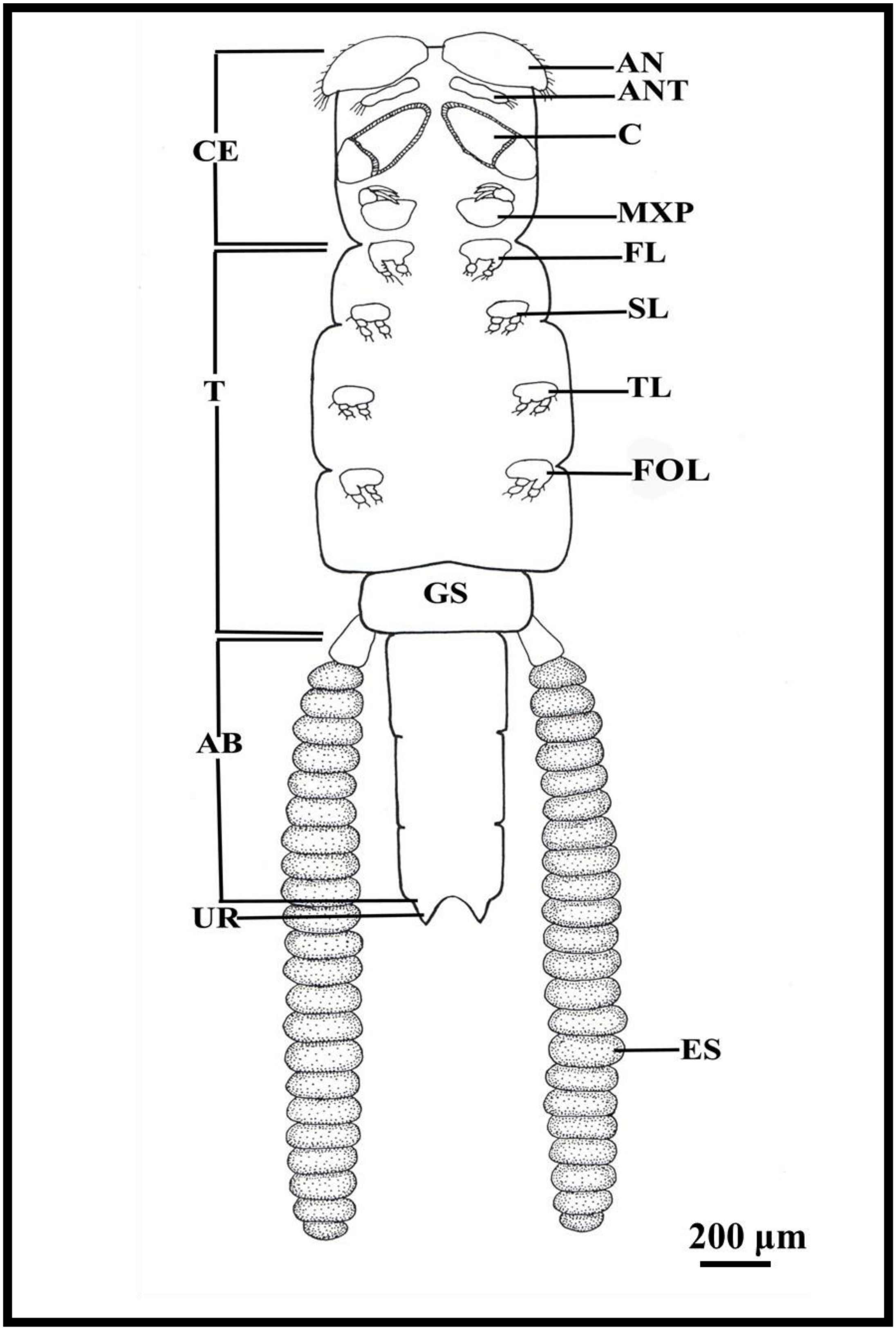


Figure 20A: Schematic drawing of ventral view of the female copepodian parasite**,** *Lamproglena minuta* **Capart, 1943.**

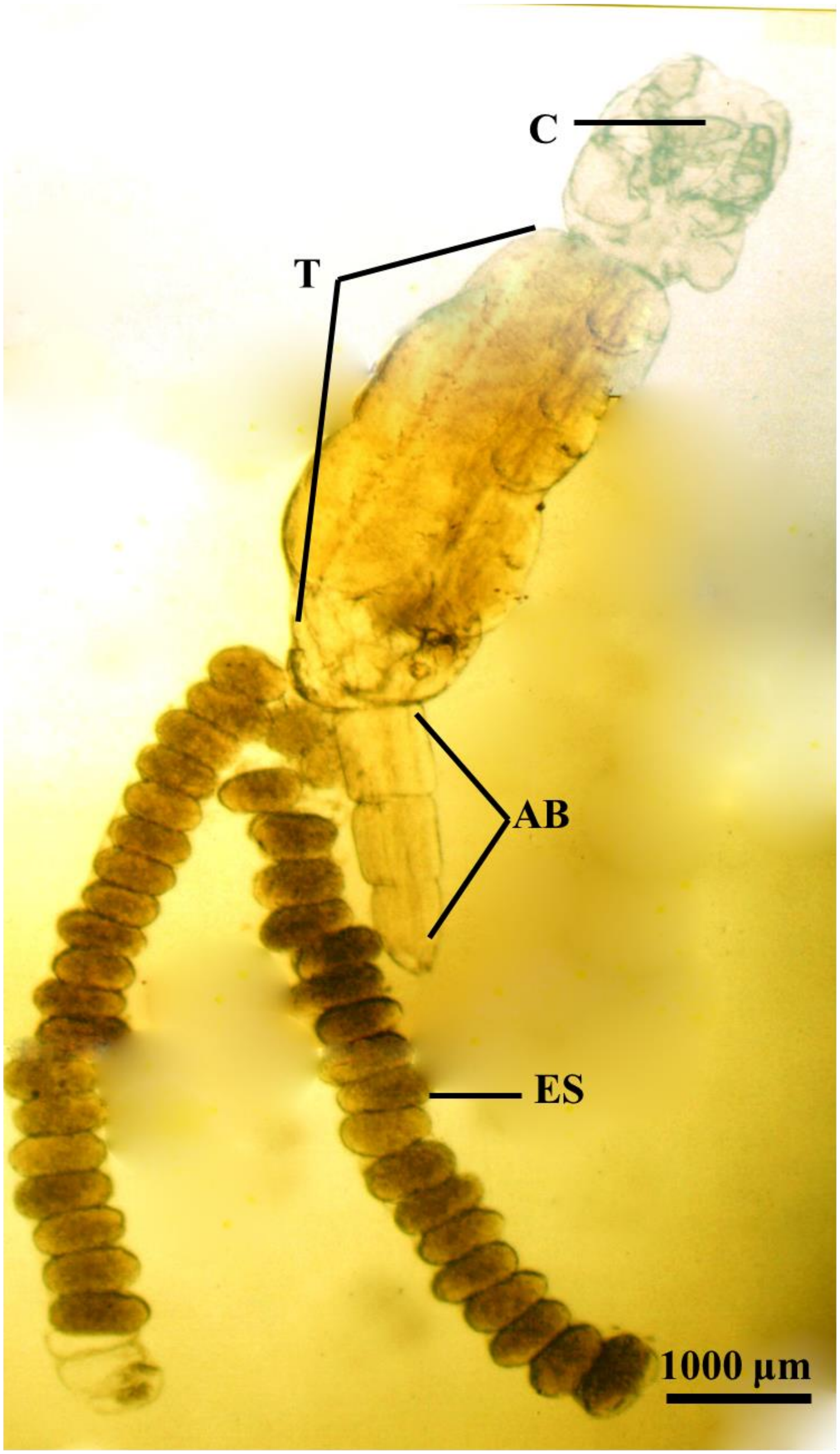


Figure 20B: Photomicrograph of the female copepodian parasite, *Lamproglena minuta* **Capart, 1943** with lactic acid.

fourth leg-bearing segment, and third (genital complex) consisting of fused fifth leg-bearing segment and genital segment. It measures in female 935 (810-1150) µm in length and 482.5 (320-570) µm in width.

Genital complex (Fig. 20) is narrower than preceding trunk regions. It measures 440 (320-500) µm in width.

Abdomen (Fig. 20) is distinctly three-segmented, sub-cylindrical, tapering somewhat posteriorly. It measures 885 (690-1230) µm in length and 275 (200-320) µm in width.

First antenna (Fig. 21A) is short, unsegmented, non-prehensile, armed with several very short setae and measures 227.5 (200-310) µm in length and 130 (99-135) µm in width.

Second antenna (Fig. 21B) is relatively poorly developed, four indistinct segments, armed with many setae. It measures 112.5 (100-120) µm in length and 40 (35-50) µm in width.

Maxilliped (Fig. 21C) is carrying at its apex three subequal curving; claw-like spines, at least one of them with papilliform outgrowth near base.

Uropod (Fig. 21D) is short, with one seta, two small papilla on or near distal margin and one seta on lateral margin.

First four pairs of legs biramous. First pair (Fig. 21E) with small protuberance on medial margin of sympod, near base; endopod apparently unsegmented, with fringe of setules on lateral margin; exopod indistinctly two-segmented, with several setae on lateral margin and single stout seta on apex.

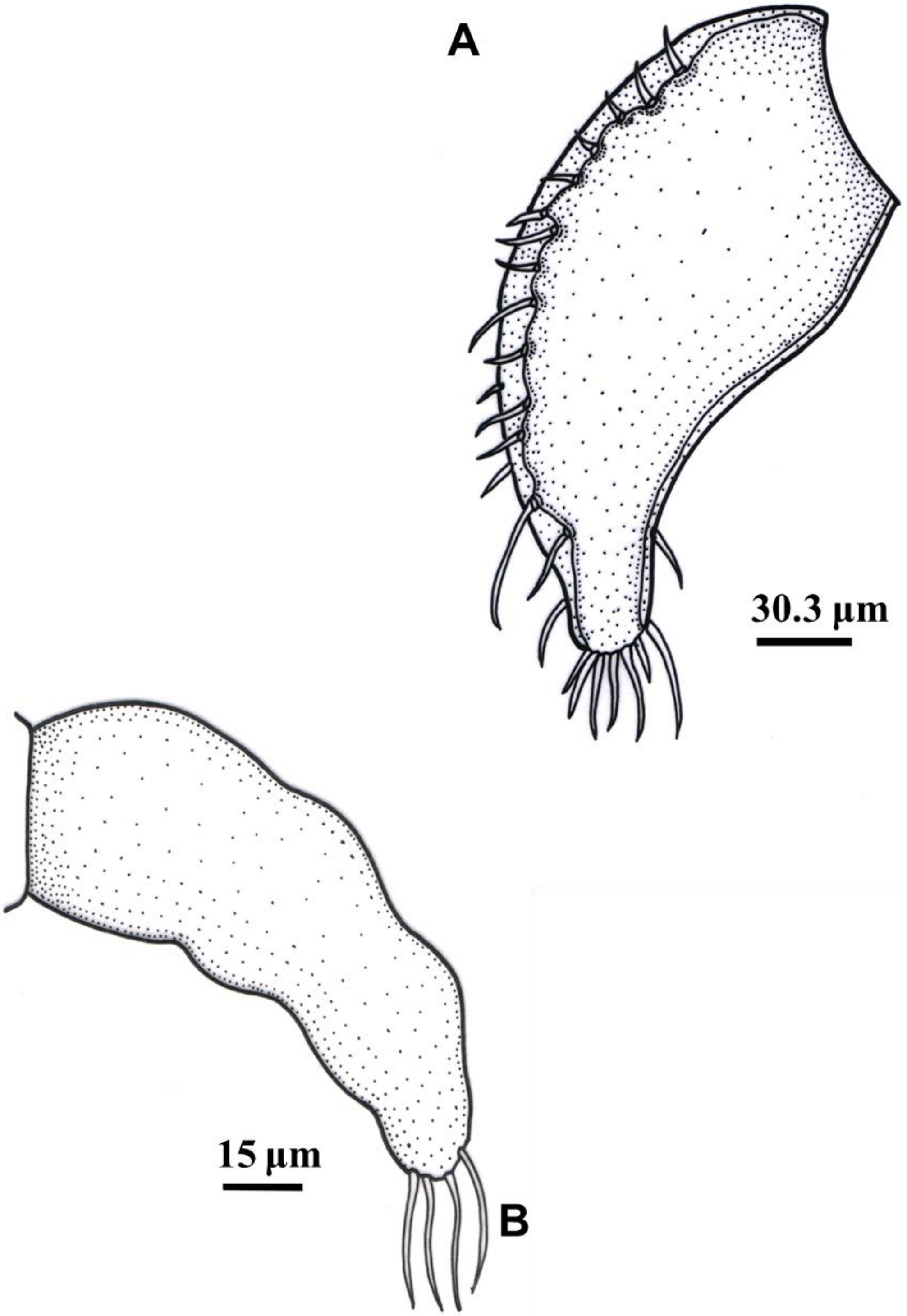


Figure 21A&B: Schematic drawing of the antennule, (A) and antenna (B) of the female copepodian parasite, *Lamproglena minuta* **Capart, 1943.**

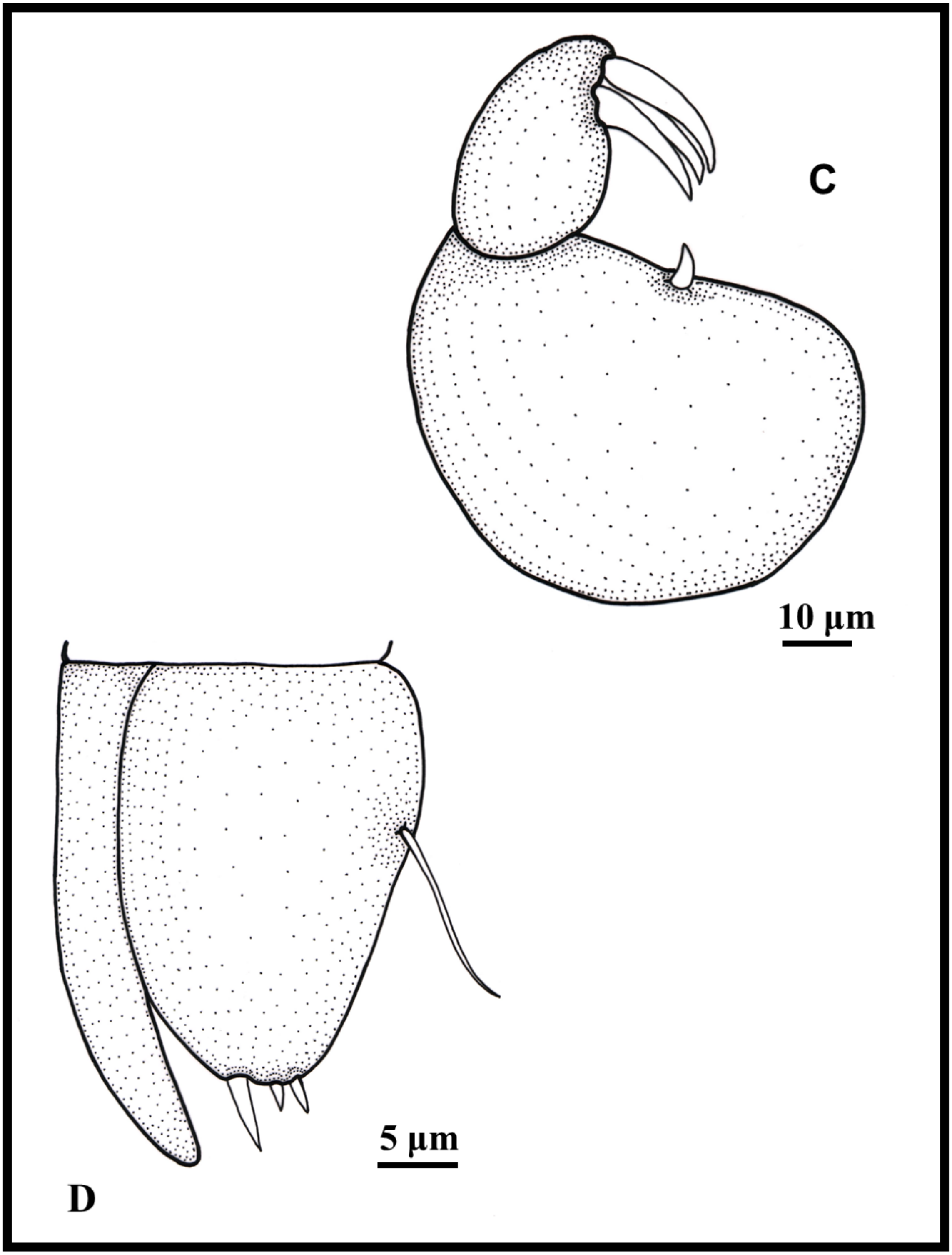


Figure 21C&D: Schematic drawing of the maxilliped, (C) and uropod (D) of the female copepodian parasite, *Lamproglena minuta* **Capart, 1943.**

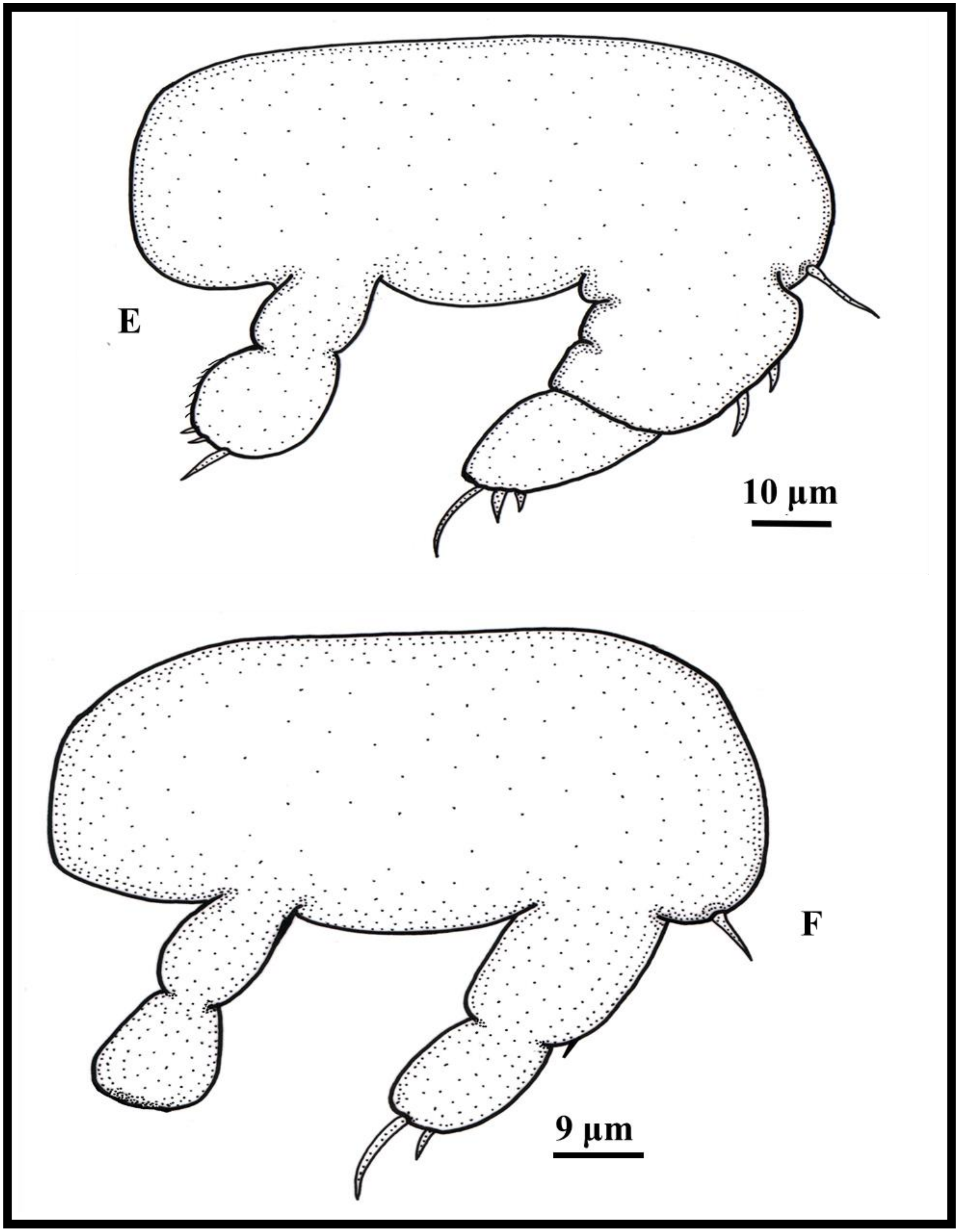


Figure 21E&F: Schematic drawing of the first leg, (E) and second leg (F) of the female crustacean parasite, *Lamproglena minuta* **Capart, 1943.**

Second leg (Fig. 21F) is bearing indistinctly two segmented rami; endopod unarmed, exopod with single seta on lateral margin of basal segment and three setae at apex.

Third and fourth pairs (Fig. 21G) are similar, rami similar but with only two setae at apex of exopod.

Egg sacs (Fig. 20) are uniseriate; each measures 1696 (1610-1830) µm in length and 203 (190-220) µm in width. Each egg sac contains 23-30 eggs.

(b) Adult male

Body of male parasite (Fig. 22) is elongated, smaller than female with average total length of 2232 (2211-2250) µm and 530 (490-610) µm in width, consists of three distinct parts: cephalothorax, trunk and abdomen.

Cephalothorax (Fig. 22) is incorporating first leg-bearing segment, of about equal length and width, slightly wider in its posterior half, with marked indentations at about midlength of lateral margins, separated from trunk by deep, neck-like constrictions. It measures in male 507(510-550) µm in length and 430 (370-510) µm in width.

The trunk (Fig. 22) is oval, longer than wide, subdivided into three regions, first consisting of fused second and third leg-bearing segments marked off posteriorly by definite indentations on lateral margins, second consisting of fourth leg-bearing segment, and third (genital complex) consisting of fused fifth leg-bearing segment and genital segment. It measures in male 867 (750-1030) µm in length and 550 (460-600) µm in width.

Genital complex (Fig. 22) is narrower than preceding trunk regions. It measures 360 (300-410) µm in width.

Abdomen (Fig. 22) is distinctly three-segmented, sub-cylindrical, tapering somewhat posteriorly. It measures in male 786 (600-930) µm in length and 218 (190-240) µm in width.

Appendages of male is similar to female appendages in structure but different from female in dimension that the appendages of male are smaller where first antenna measures 200 x 90 µm, second antenna 99 x 35.5 µm, maxilliped 130 x109 µm, first thoracic leg 80 x 95.5 µm, second and third thoracic legs 66 x 65.7 and fourth thoracic leg 54.5 x 45 µm.

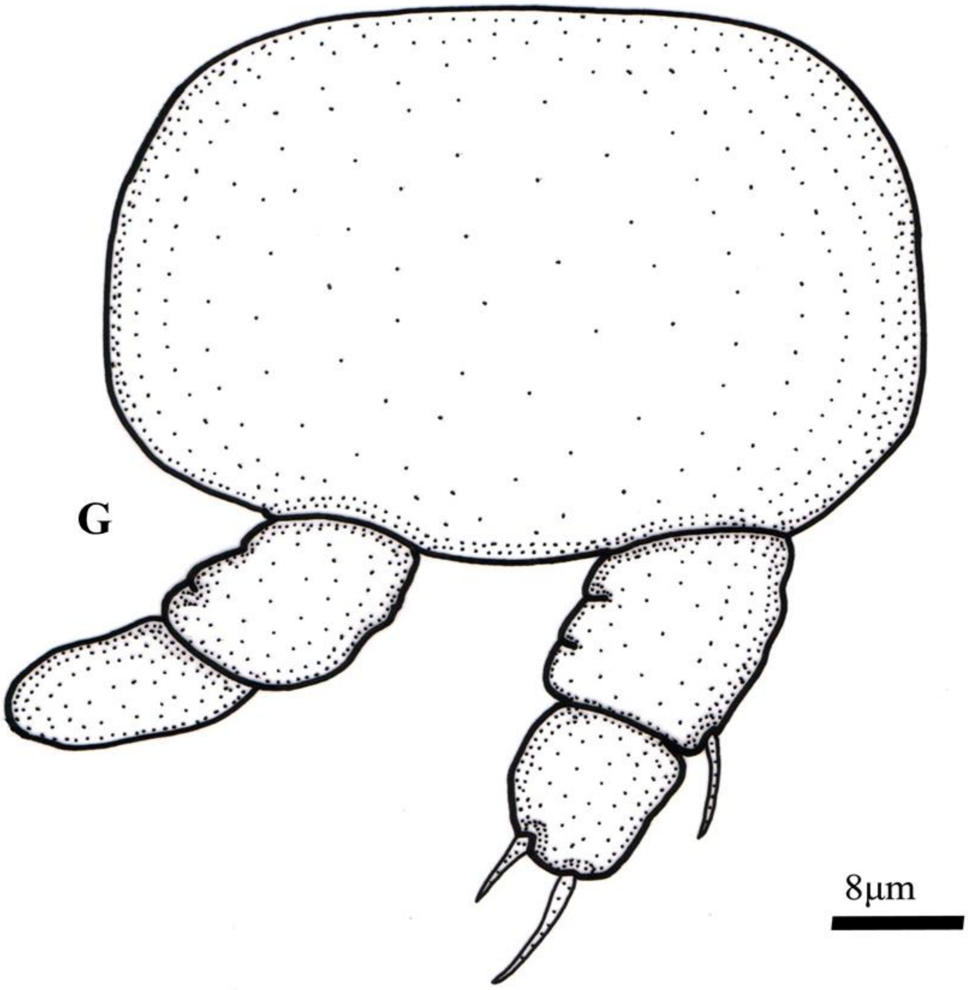


Figure 21G: Schematic drawing of the third and fourth legs, (G) of the female copepodian parasite, *Lamproglena minuta* **Capart, 1943.**

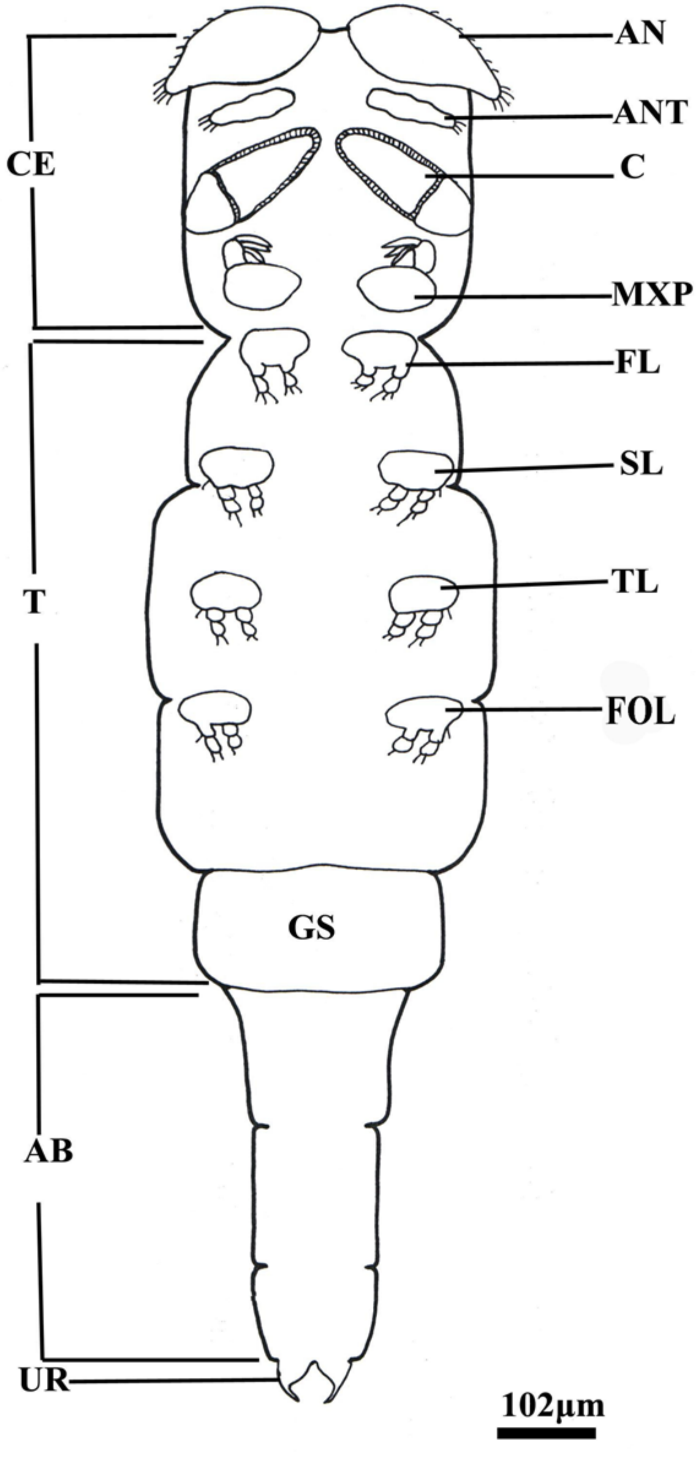


Figure 22A: Schematic drawing of ventral view of the male copepodian parasite, *Lamproglena minuta* **Capart, 1943.**

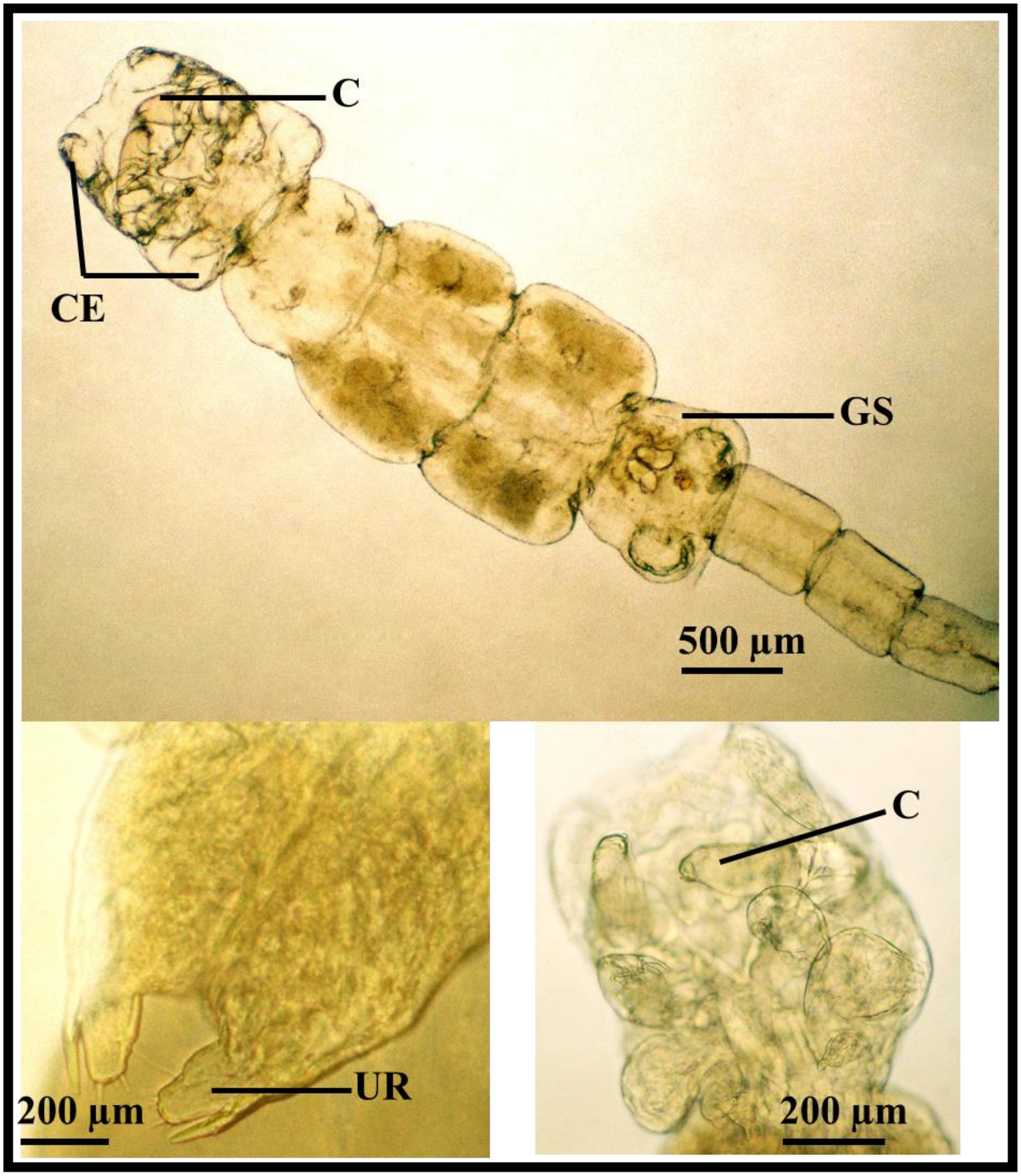


Figure 22B: Photomicrograph of the male copepodian parasite, *Lamproglena minuta* **Capart, 1943** with lactic acid.

(2)Discussion:

The anatomical structure and dimensions demonstrated in the present description are evidently to suggest that the present species described in the present study belongs to genus *Lamproglena* **von Nordmann**, **1832** according to the following generic criteria which based by **Kabata (1979).** These generic morphological criteria are: the body of the adult female *Lamproglena* consists of three distinct parts: cephalothorax, trunk and abdomen. The cephalothorax is marked from the trunk by a deep constriction; it might or might not include the first pair of swimming legs. The trunk is oval and externally unsegmented, or contains traces of segmentation (grooves and constrictions). The abdomen is sub-cylindrical; it is three- segmented, though in some species no external segmentation is observable. The first antennae are uniramous, usually unsegmented, non-prehensile; the second antennae are relatively poorly developed. The mouth parts are of the cyclopoid type. The Maxillipeds are strong and prehensile, ending usually in three claw-like spines. In most species there are four pairs of biramous swimming legs, and fifth leg being vestigial and uniramous. In some species only one pair of swimming legs is retained. *Lamproglena* differs from other lernaeid genera in uniseriate egg sacs with relatively few eggs.

The present individuals of the parasitic copepod *Lamproglena minuta* **Capart, 1943** infesting gills of the *Tilapia galilaea*; *Sarotherodon galilaeus* was recorded for the first time from Egyptian freshwaters.

Previously *Lamproglena minuta* has been recorded from many localities from South-East Asian fishes, *Anabas testudineus*, *Ophiocephalus striatus*, *Barilius barmandi*, *Puntius gonionotus* and *Puntius binotatus* **(Kabata, 1985)**. *Lamproglena minuta* has been recorded on the gill filaments of *Barilius barmandi* in Chieng Bay, Thailand **(Yamaguti, 1954)**. The present redescription is closely related to *Lamproglena minuta* **Capart, 1943** which was firstly recorded and described by **Capart (1943**) from the gill filaments of *Puntius binotatus* in Buknon, Thailand and *Tylognatbus gracilis* in Muang Pan, Thailand.

There are close similarities between the present specimens and the specimens redescribed by **Kabata (1985)** in all morphological criteria, specially shape and armature of antennule and antenna, shape of cephalothorax, armature of swimming legs, shape of trunk and abdomen segments and caudal rami. Meanwhile, the present specimens differ from the Asian specimens described by **Kabata (1985)** where the present specimens are larger in which total length of Asian specimens was 1.3 mm; cephalothorax length 0.3 mm, width 0.3 mm; trunk length 0.5 mm, width 0.35 mm; abdomen length 0.5 mm, width 0.14 mm in female see Table (2).

Abbreviations of the copepodian parasite (*Lamproglena minuta*) figures

AN	1st antenna
ANT	2nd antenna
C	Claw
MXP	Maxiliped
FL	1st leg
SL	2nd leg
TL	3rd leg
FOL	4th leg
GS	Genital segment
ES	Egg sac
CE	Cephalothorax
T	Trunk
AB	Abdomen
UR	Uropod

Table (2): Morphometric measurements (maximum length x maximum width in µm) of the copepodian parasite, *Lamproglena minuta* Capart, 1943 collected during the present study.

Sex	Female	Male
Maximum length x Maximum width	2460x617.5	2232 x 530
Cephalothorax	550 x 520	507 x 430
Trunk	935 x 482.5	867 x 550
Abdomen	885 x 275	786 x 218
Uropod	90 x 66.8	72 x 50
Egg sac	1696 x 203	-
First antenna	227.5 x 130	200 x 90
Second antenna	112.5 x 40	99 x 35.5
Maxilliped	150 x 117.5	130 x 109
First thoracic leg	82.5 x 100	80 x 95.5
Second & Third thoracic leg	67.5 x 72.5	66 x 65.7
Fourth thoracic leg	60 x 50	54.5 x 45

CHAPTER I

Section III

Ergasilus briani

Genus: *Ergasilus* von Nordmann, 1832

Ergasilus briani Markewitsch, 1933

(1) Redescription:

The body structure of the copepodian parasite *Ergasilus briani* is showed in figures 23 & 24 and its measurements are shown in table (3).

The following redescription is based on the study of eleven living adult and ten mounted specimens of female *Ergasilus briani*, where the male is free living and only the adult female is a parasite, collected from the gill filaments of the flat-head grey mullet, *Mugil cephalus.*

The body of the female parasite is slender, elongated, violin-like, markedly expanded anteriorly and gradually constricted towards the posterior end (Fig. 23). The body length of the female *E. briani*, excluding caudal setae and egg sacs, is 768 (700 – 880) μm and 301 (215-350) μm in width.

The body consists of two main parts, prosome and urosome (Fig. 23). The prosome consists of cephalosome and mesosome. The first somite of the mesosome is fully incorporated into the cephalosome forming cephalothorax which is equal approximately in length to the remaining part of the body. There is no boundary between them is indicated only by a shallow, indistinct constriction, just posterior to the anterior two thirds, and running across both lateral sides.

The cephalothorax (Fig. 23) is oblong, uninflated and bullet-shaped. Its anterior end is slightly tapering with slightly projecting antennary region forming a short rostrum and the posterior margin is transversely truncated. The cephalothorax decreases in width posteriorly. It measures 280 (255-325) μm in

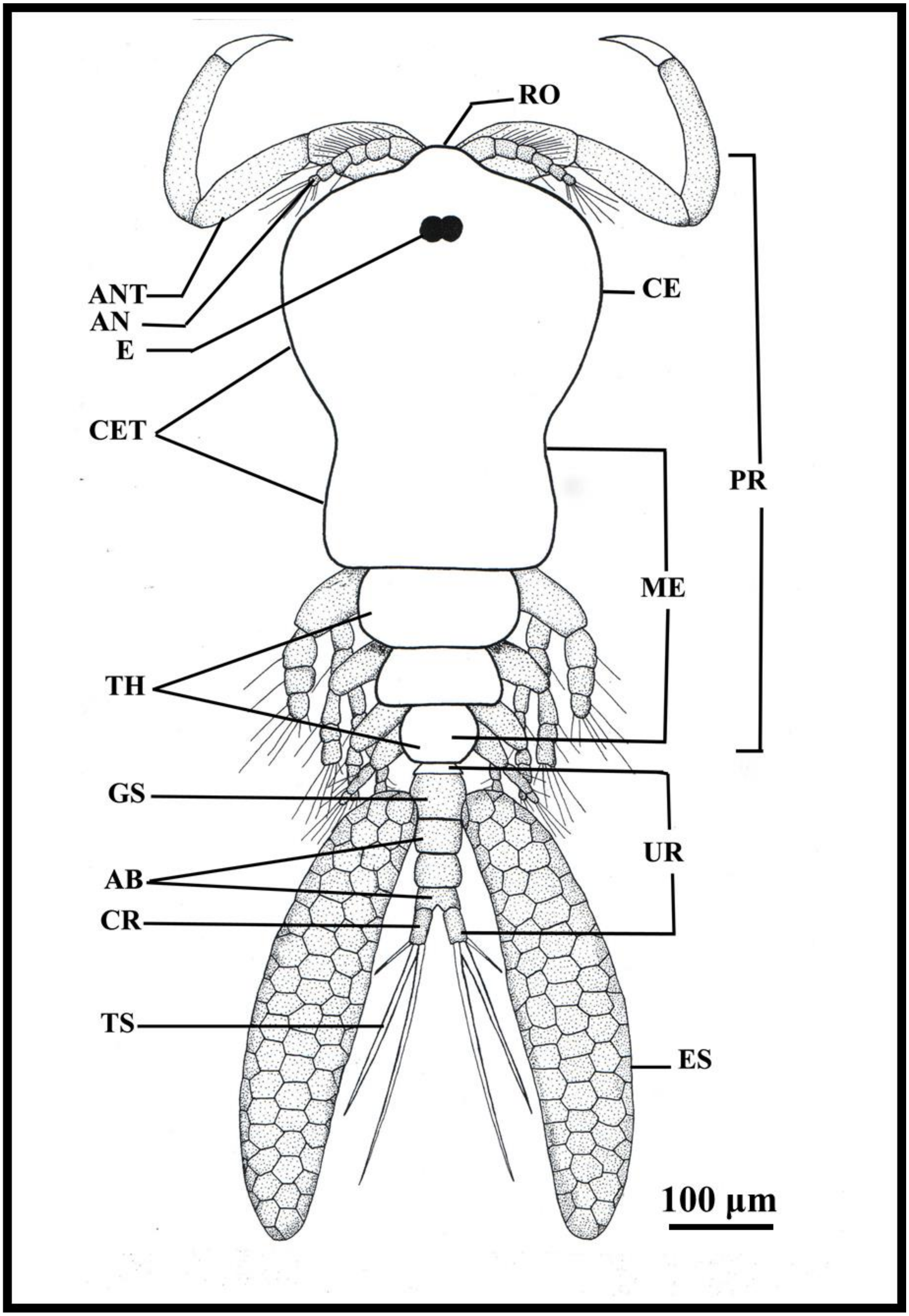


Figure 23A: Schematic drawing of the female copepodian parasite, *Ergasilus briani* **Markewitsch, 1933.**

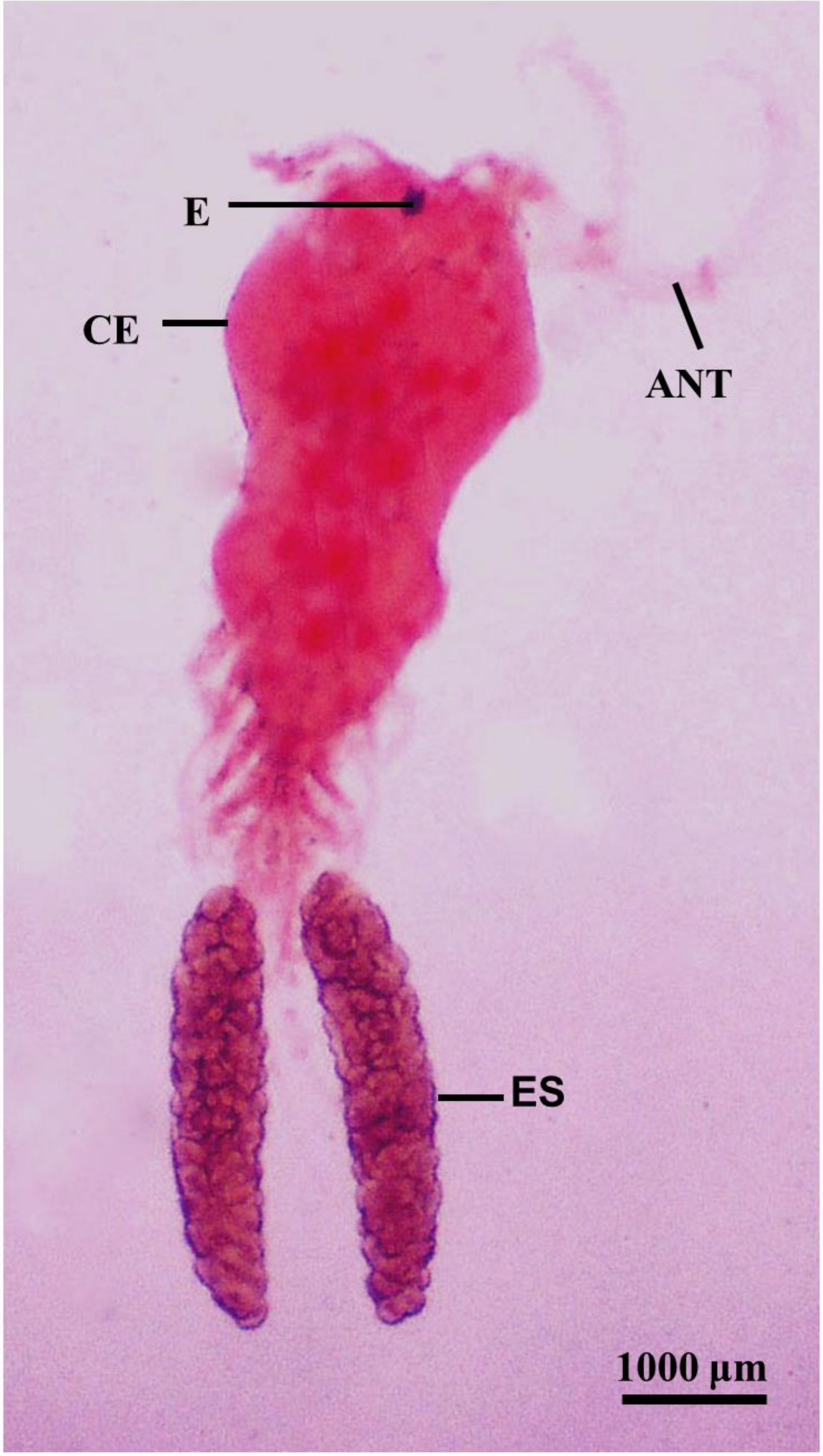


Figure 23B: Photomicrograph of the female copepodian parasite, *Ergasilus briani* **Markewitsch, 1933** stained with alum carmine**.**

length and 301 (215-350) µm in a maximum width that attained at its anterior broader region. A median eye spot lies dorsally on the cephalothorax (Fig. 23), just posterior to the rostrum.

The mesosome (Fig. 23) consists of three free somites comprising second, third and fourth thoracic somites. The metasomal somites are broader than long, progressively narrowed from cephalothorax and decrease in size towards the posterior end of the body.

The two ovisacs (Fig. 23) are long but their length is less than half of the total body length. Each ovisac extends posteriorly with slightly tapering and rounded distal end. Each one measures 440 (365-485) µm in length and 104 (80-105) µm in width. They are filled with large number of eggs. Eggs are large, spherical and visible through the thin membrane of the egg sac.

The abdomen (Fig. 23) consists of three somites. These somites are wider than long, with almost similar width and slightly diminish posteriorly. The first abdominal somite is larger than the following two. The third abdominal somite is notched posteriorly almost up to the half of its length. The last somite is almost equal to or slightly smaller than the second one. Each forked part of the third abdominal somite carries a single caudal ramus. The length of each ramus is almost three times longer than its width, almost equal to the last two abdominal segments. Each caudal ramus is armed distally with four terminal setae. The innermost one is the largest, the outermost seta is shorter than the innermost one and the middle two are ventrally located. They are small and almost unnoticeable from the dorsal side.

Cephalthorax bears one pair of antennules, one pair of antennae and eye spots. The pair of short cylindrical, setiferous, somewhat constricted distally and segmental antennules are located on the protuberant rostral area. Each antennule (Fig. 24A) measures 115 (87.5-137.5) µm in length and 26.6 (22.5-32.5) µm in maximum width and consists of six segments. The size of the segments is diminished distally towards the terminal end except the second segment which is slightly the biggest. All antennualry segments are provided with numerous simple setae, principally on antero-ventral surface. The number of setae on each segment starting from the proximal segment to the distal one is as follows: 1S: 135: 65: 45, 1a: 2S, 1a: 6S, 1a, Where “S” means naked setae and “a” means aesthetic. The number of these antennulary setae is known as “setal formula”.

A pair of stout prehensile, subchelate and segmented antennae (Fig. 24B) is situated on the slightly protruding rostral area. They are long and slender. Each antenna measures 490 (543-588) µm in length and 141 (122.5-150) µm in maximum width and consists of four segments. The first or basal segment is the coxobasis. It is short, slightly cubical in shape and bears a single tooth-like spine arising near its inner distal margin. The second segment is the first endopodal segment which is the longest one. It is widest at its proximal end and narrows towards its distal end. It bears a small tooth-like spine at its inner margin, almost at the beginning of the distal third. The third antennary segment is the second endopodal segment and is called the subchela. It is narrow and arched with the sides almost parallel and clearly bent inwards. It bears three small teeth-like spines, one almost at midway on its inner margin, one distal most on its outer margin near the end of the segment and the third one medially situated near the base of the fourth segment. The fourth or distal antennary segment is the fourth endopodal segment. It is in the form of a stout pointed, clasper-like smooth claw lacking tooth. It is the shortest segment (Fig. 24B).

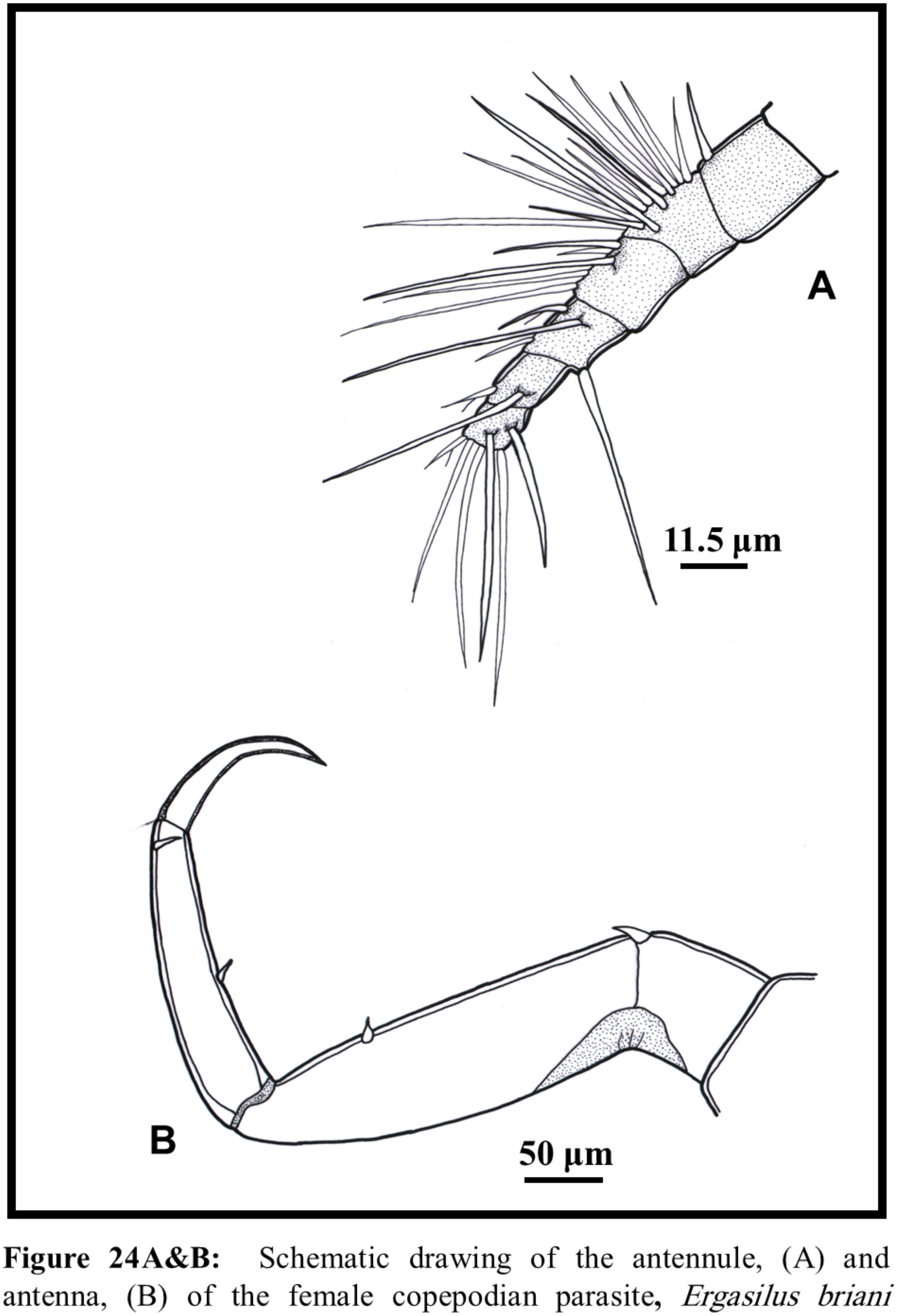


Figure 24A&B: Schematic drawing of the antennule, (A) and antenna, (B) of the female copepodian parasite**,** *Ergasilus briani* **Markewitsch, 1933.**

Five pairs of thoracic swimming legs originate from the lateral side of the first five thoracic somites. The first pair of thoracic swimming legs originates ventral-laterally from the posterior part of the cephalothorax and the last pair projects from the fifth thoracic somite. The first four pairs are biramous and exist in a completely form. Each leg consists of a proximal coxapod and a distal basipod which bears the two free rami; an exopod and an endopod. Whereas the fifth thoracic pair is greatly reduced.

The first pair of thoracic legs (Fig. 24C) consists of a coxapod and basipod that arnamented on their anterior surface with rows of spinules. The basipod is ornamented also with a single antero-lateral sensilum. Each of exopod and endopod consists of three segments that diminish distally in size. The first exopodal segmental is serrated where there is one row of spinules project from both outer and distal margins. It is also provided with a single postero-lateral rasp-like spine projecting from the outer margin and providing with one row of spinules on its outer margin. The second exopodal segmental is serrated laterally with one row of spinules arranged on its outer margin. It is provided with a single postero-lateral serrated rasp-like originating from the outer margin. A single medially situated naked seta also additionally originating from the inner margin. The terminal exopodal on its outer margin. It is provided with two laterally situated serrated rasp-like spines projecting from the outer margin. Also, it is ornamented posteriorly with five naked setae in which the outer one is pectinate. The endopodal segments are serrated where they are provided with one row of spinules arranged on the outer margins. The first endopodal segment is serrated also on the distal margin and provided with a single medially situated unarmed seta projecting from the inner segmental margin. The second endopodal segment is provided with a single postero-lateral naked seta projects from the inner segmental margin. The terminal endopodal segment is provided with two postero-lateral serrated rasp-like spine arranged on its outer margin in addition to four naked setae on both distal and inner segmental margins.

The second and third thoracic legs (Fig. 24D) are closely similar. The coxapod and basipod of each leg are ornamented on their anterior surfaces with rows of spinules arranged in scattered groups. The basipod of each leg is ornamented with a single antero-lateral sensilum projecting from the outer margin of the basipod. The exopod and endopod of both legs consist of three segments, diminishing distally in size. The first exopodal segment is provided with a single posterolateral rasp-like spine originates from its outer margin. The second exopodal segment is provided with a single medially situated unarmed seta located on the inner segmental margin. The terminal exopodal segment is provided with a single posterolateral rasp-like spine projects from the outer margin and moreover six posteriorly directed unarmed setae in which the outer is pectinate arranged on both distal and inner margins of the terminal exopodal segment. The endopodal segments are serrated where their outer margins are provided with a single row of spinules. The first endopodal segment is serrated also at distal margin and provided with a single medially situated unarmed seta located on the inner margin of the first endopodal segment. The second endopodal segment is provided with a single posterolateral unarmed seta projecting from the inner margin. The terminal endopodal segment is provided with a single posteriroly directed rasp-like projects from the distal segmental margin. Furthermore, it is provided with four posteriroly directed unarmed setae arranged on both distal and inner segmental margins.

The fourth thoracic leg (Fig. 24E) consists of a coxapod that ornamented on its anterior surface with rows of spinules arranged in scattered groups and

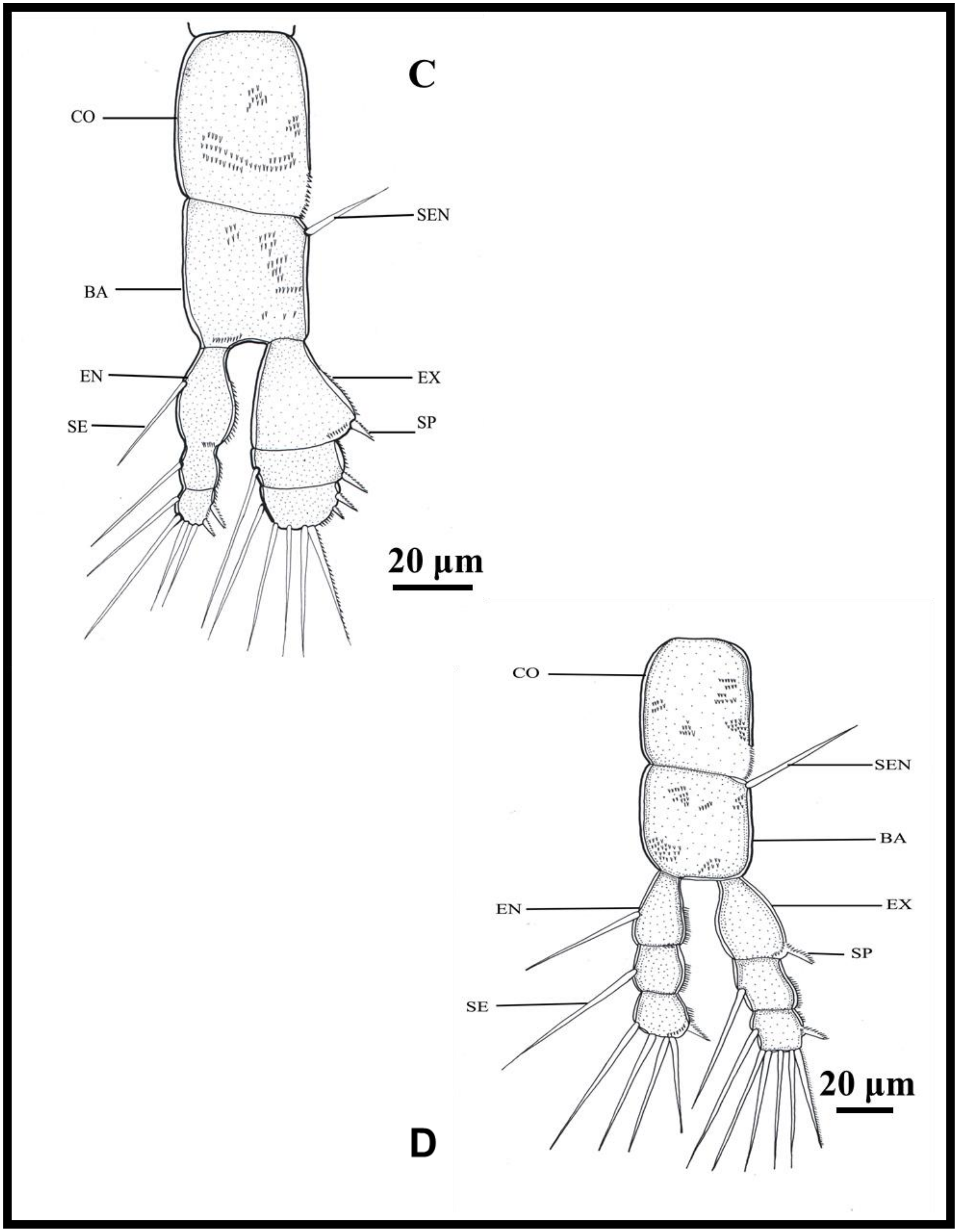


Figure 24C&D: Schematic drawing of the 1st thoracic leg, (C) and 2nd & 3rd thoracic legs (D) of the female copepodian parasite**,** *Ergasilus briani* **Markewitsch, 1933.**

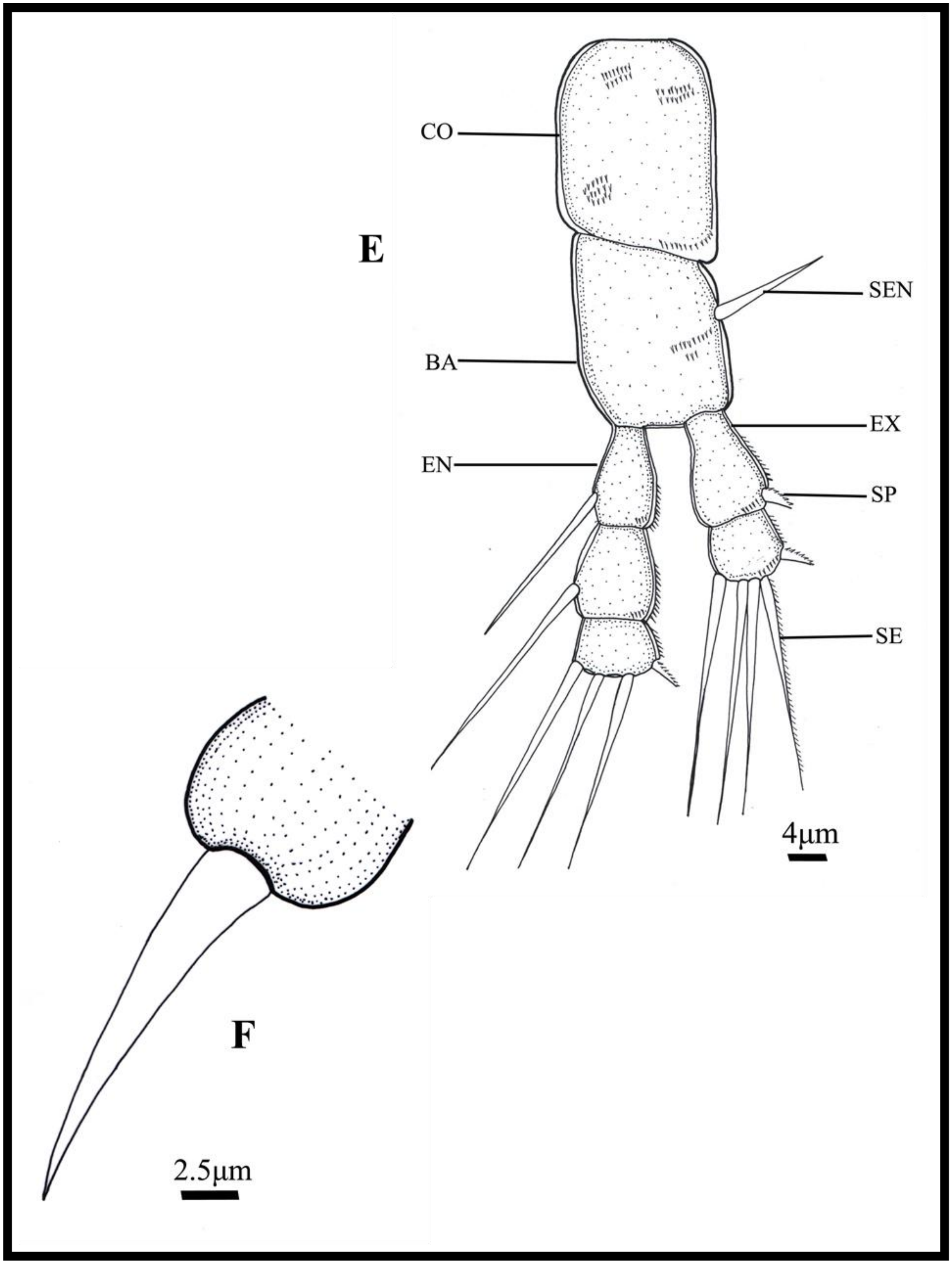


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Figure 24E&F: Schematic drawing of the 4th thoracic leg, (E) and 5th thoracic leg, (F) of the female copepodian parasite, *Ergasilus briani* **Markewitsch, 1933.**

basipod which ornamented

also on its anterior surface with a single row of spinules. Also the basipod is provided with a single antero-lateral sensilum projecting from the outer basipodal margin. The exopod consists of two segments while the endopod consists of three segments. Both exopodal and endopodal segments distally diminish in size and ornamented with a single row of spinules arranged on their outer and distal margin. The first exopodal segment is provided with a single posterolateral rasp-like; serrate spine which exists on the outer segmental margin. The terminal exopodal segment is provided laterally with a posterolateral rasp-like, serrate spine projecting from the outer segmental margin in addition to four posteriorly directed unarmed setae arranged on its distal margin; the outer one is pectinate. The first endopodal segment is ornamented laterally and inwardly with medially situated unarmed seta. The second endopodal segment is provided with a single posterolateral unarmed seta which exists on the inner segmental margin. The terminal endopodal segment is provided laterally with a single serrate, rasp-like spine located on the outer segmental margin besides three posteriorly directed unarmed setae arranged on the distal segmental margin.

Finally, the extremely reduced fifth thoracic swimming leg (Fig. 24F) is represented by papillary process ornamented with a single posterolaterally directed unarmed sensilum located on the distal end. The armature formula for spines (Roman numerals) and setae (Arabic numerals) of the thoracic legs of the adult female *Ergasilus briani* **Markewitsch, 1933** is shown in Table (4).

The urosome (Fig. 24G) consists of the two last thoracic somites (fifth and sixth), the abdominal somites and the caudal rami. The fifth thoracic somite is extremely reduced, very small and short but distinctly apart as much from the fourth thoracic segment as from the genital segment that follows it. It measures 173 (150-207.5) µm in length and 55 (35-65) µm in width. The sixth thoracic somite is the genital somite.

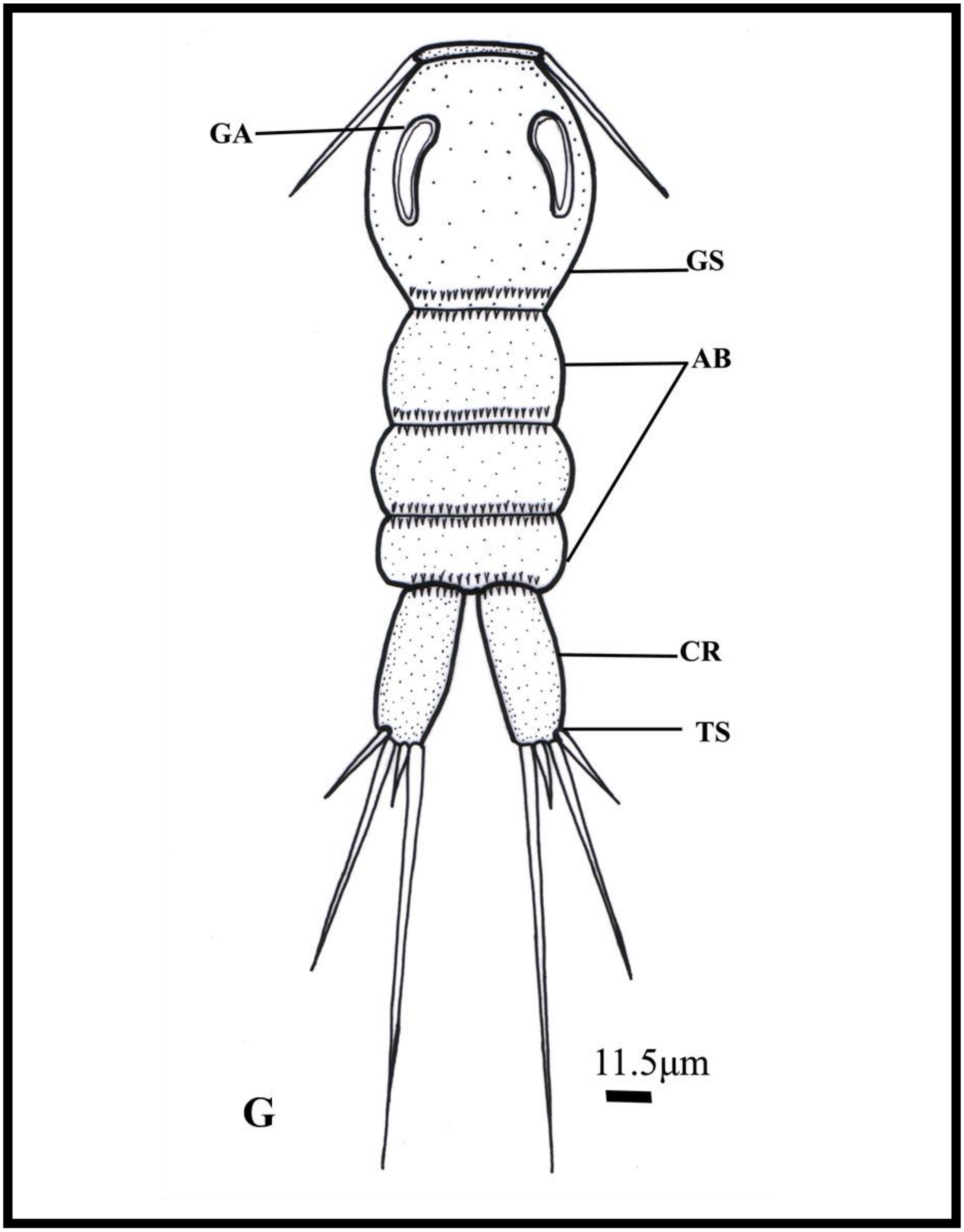


Figure 24G: Schematic drawing of the urosome, (G) of the female copepodian parasite**,** *Ergasilus briani* **Markewitsch, 1933.**

Table (3): A comparison between the present species of female parasitic copepod *Ergasilus briani* and the previously described species (in µm).

Author(s)	Markewitsch (1956)	Alston *et al.* (1996)	The present specimens
Host	*Cyprinid fishes*	*Tinka tinka*	*Mugil cephalus*
Locality	USSR	England	Egypt
Body measurements	700x1000	885.3x500	768 (700 – 880)x301 (215-350)
Cephalothorax	335.42	419.95	280 (255-325)x301 (215-350)
Urosome	-	-	173 (150-207.5)x55 (35-65)
First antenna	102.08	133.53	115 (87.5-137.5)x26.6(22.5-32.5)
Second antenna	233.33	397.25	490 (543-588)x141 (122.5-150)
First thoracic leg (L1)	-	-	93(75-112.5)
First thoracic leg (L2)	-	-	53(50-55)
First thoracic leg (W)	-	-	35(32.5-37.5)
Second&third thoracic leg (L1)	-	-	84.5(50-110)
Second&third thoracic leg (L2)	-	-	63(46.25-81.25)
Second&third thoracic leg (W)	-	-	27(17.5-36.25)
Fourth thoracic leg (L1)	-	-	60(25-77.5)
Fourth thoracic leg (L2)	-	-	53(45-62.5)
Fourth thoracic leg (W)	-	-	24(15-32.5)
Fifth thoracic leg	-	-	25x10
Egg sac	364.58-379.17x72.92	-	440 (365-485)x104 (80-105)

Thoracic legs	Coxapod	Basipod	Exopod			Endopod		
			1	2	3	1	2	3
First leg	0-0	I-0	I-0	I-1	II-5	0-1	0-1	II-4
Second leg	0-0	I-0	I -0	0-1	I-6	0-1	0-1	I-4
Third leg	0-0	I-0	I-0	0-1	I-6	0-1	0-1	I-4
Fourth leg	0-0	I-0	I-0	I-4	-	0-1	0-1	I-3

Roman numerals = Spines Arabic numerals = Setae

Table (4): The armature fomula for spines (Roman numerals) and setae (Arabic numerals) on the thoracic legs of adult female *Ergasilus briani* **Markewitsh, 1933.**

CHAPTER I

Section IV

Ergasilus lizae

Genus: *Ergasilus* von Nordmann, 1832

Ergasilus lizae krøyer, 1863

(1) Redescription:

The body of the copepodian parasite *Ergasilus lizae* is illustrated in figures 25 & 26 and its measurements are shown in table (5).

The following redescription is based on the study of eleven living adult and twenty mounted specimens of female *Ergasilus lizae*, where the male is free living and only the adult female is a parasite, collected from the gill filaments of the, *Valamugil seheli.*

The body of female parasite is panduriform, slightly elongated, markedly expanded anteriorly and progressively narrowed towards the posterior end (Fig. 25). The body length of female *Ergasilus lizae*, excluding caudal setae and egg sacs, is 925 (900-950) µm. The maximum body width that attained at cephalothorax is 368.59 (357.98 – 384.59) µm.

The body (Fig. 25) consists of two main parts; prosome and urosome. The prosome consists of cephalosome and mesosome. The first somite of the mesosome is fully incorporated into the cephalosome forming cephalothorax. There is no suture between the cephalosome and the first mesosomal somite but the boundary between them indicated only by a dorsal transverse welt with shallow notches just anterior to the midlength of the cephalothorax on the lateral margins.

The cephalothorax (Fig. 25) is oblong, enlarged, somewhat distended and guitar-shaped. Its anterior half is broader with slightly protruding antennary region forming a short rostrum and its posterior half is slightly narrower with two posteriorly rounded lateral margins. The cephalothorax decreases in width posterior. It measures 352 (350-355) µm in length and 395 (365-450) µm in a maximum width that attained at its anterior broader region.

The metasome (Fig. 25) consists of three free somites comprising second, third and fourth thoracic somites. The metasomal somites are broader than long, progressively narrowed from the cephalothorax and gradually diminish in width towards the posterior end of the body. The second thoracic somite is the largest one.

A pair of elongated, cigar-shaped, multiseriated egg sacs (Fig. 25) are originated one from each ventro-lateral side of the genital somite. The egg sacs are long and constituting more than half the total body length. Each egg sac extends posteriorly with slightly tapering and rounded distal end. Each one measures 787.5 (725-850) µm in length and 225 (220-230) µm in width. They are filled with a large number of eggs. Eggs are large, spherical and visible through the thin membrane of the eggs.

The abdomen (Fig. 25) consists of three somites. These somites are much wider than long, with sub equal sizes and slightly narrow posteriorly. The first abdominal somite is slightly larger than the following two. The third abdominal somite is deeply incised centrally at its posterior end and almost up to the half of its length. This last somite is noticeably shorter than the previous two. Each forked part of the third abdominal somite carries single caudal ramus. The two caudal rami are slightly longer than broad. Each ramus is armed distally with four

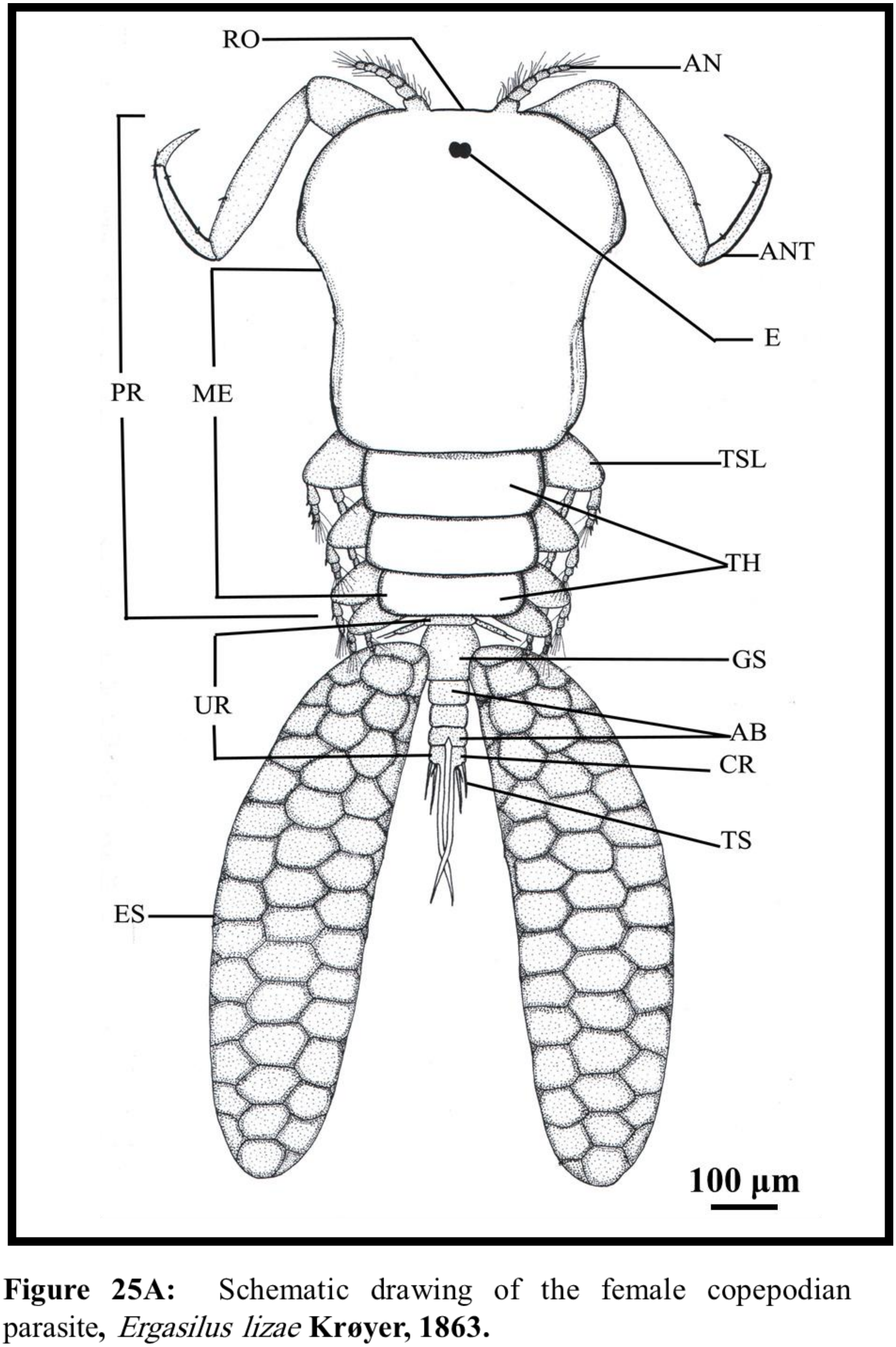


Figure 25A: Schematic drawing of the female copepodian parasite**,** *Ergasilus lizae* **Krøyer, 1863.**

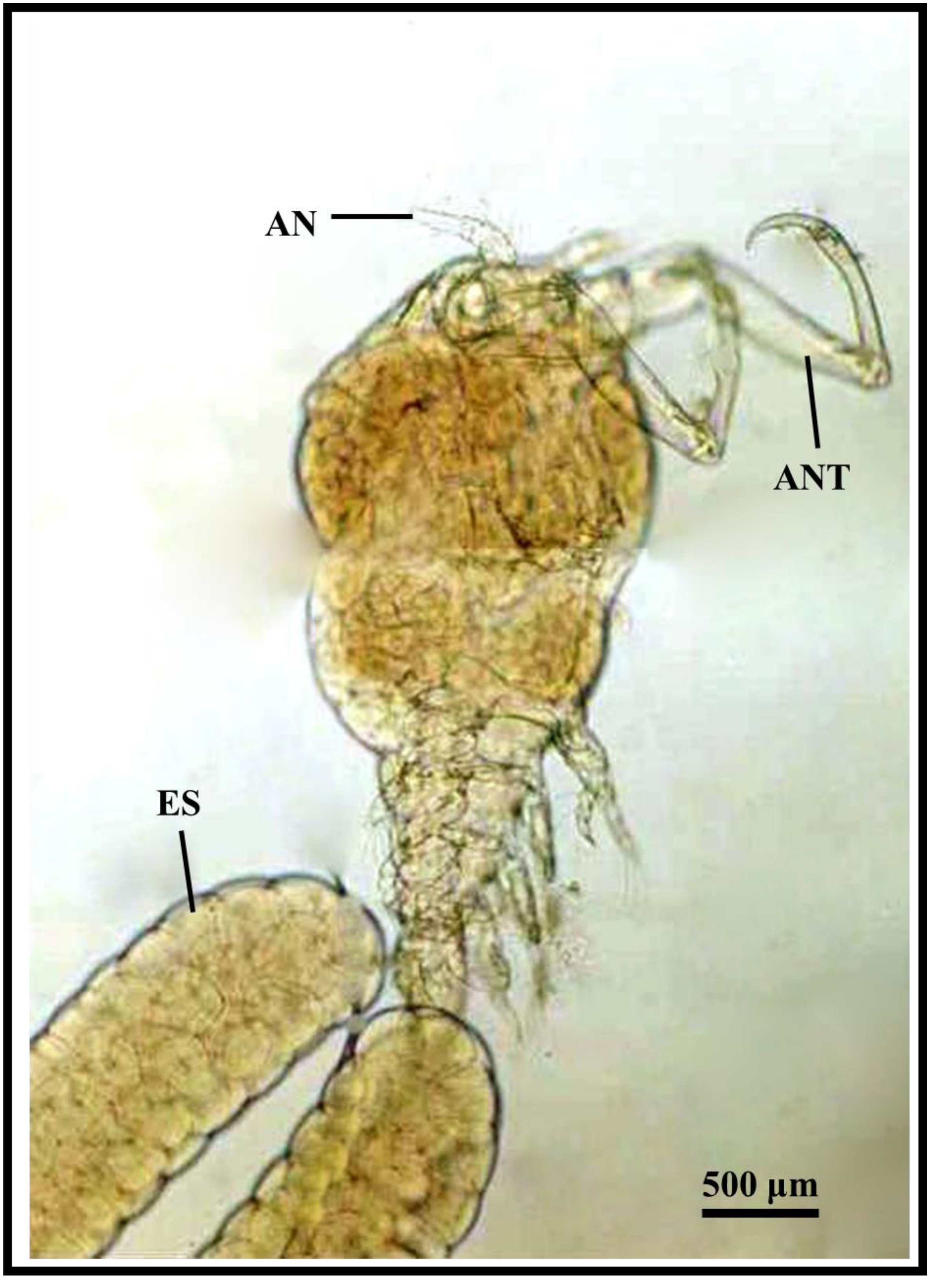


Figure 25B: Photomicrograph of the female copepodian parasite**,** *Ergasilus lizae* **Krøyer, 1863** with lactic acid.

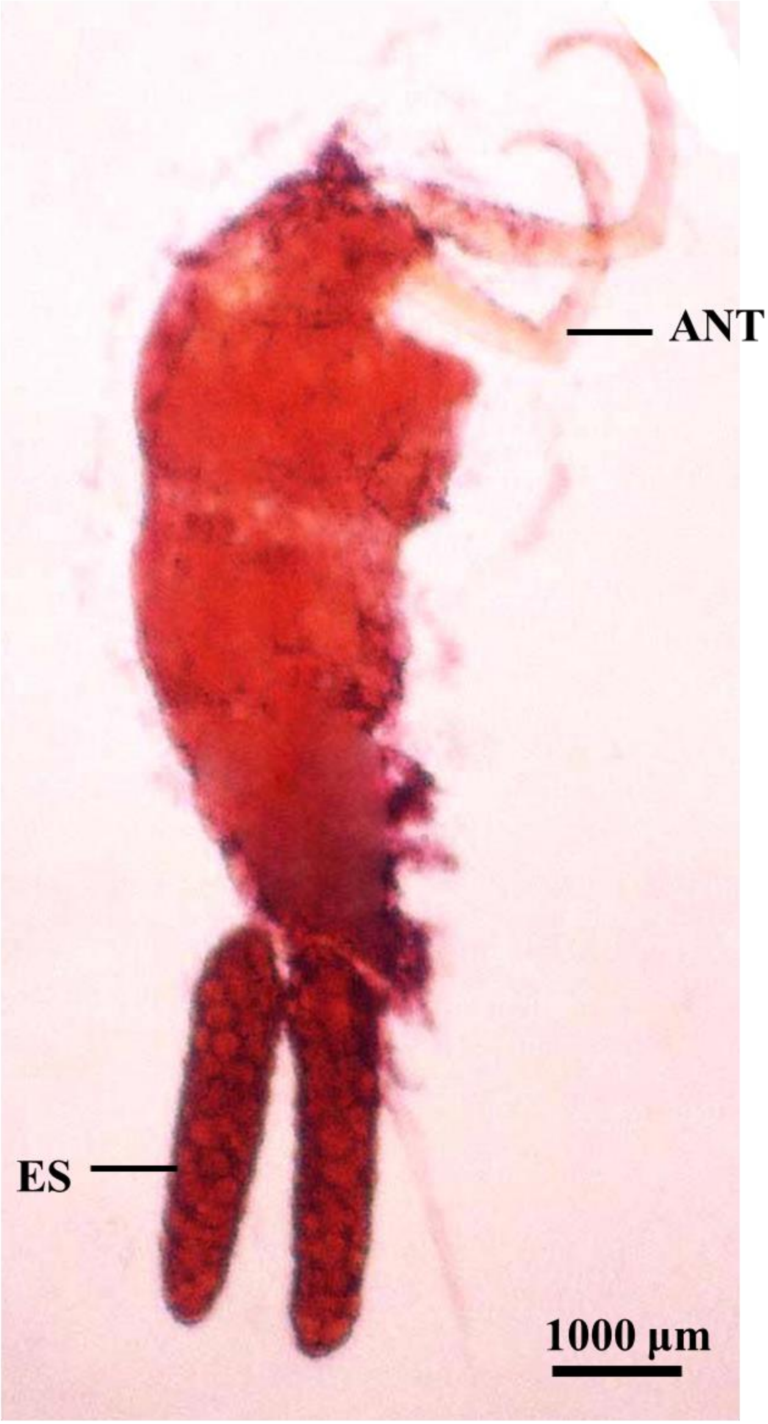


Figure 25C: Photomicrograph of the female copepodian parasite**,** *Ergasilus lizae* **Krøyer, 1863** stained with aceto-carmine.

terminal setae. The inner one is the largest and the outer seta is smaller than the inner one. The middle two are ventrally located. They are small and almost unnoticeable from the dorsal side.

The cephalothorax bears one pair of antennules, one pair of antennae and eye spots. A pair of short cylindrical, setiferous, tapering distally and segmented antennules (Fig. 26A) is located on the protuberant rostral area. Each antennule measures 112.5 (100-120.7) µm in length and 37.5 (30-39.5) µm in width and consists of six segments. These segments diminish distally in size towards the terminal end except the second segment which is slightly the largest.

All antennule segments are provided with numerous simple unarmed setae, principally on anterior-ventral surface. The number of setae one each segment starting from the proximal segment to the distal on is as follows; 3: 11: 4: 4: 3: 8. The number of these antennutary setae is known as “Setal formula” or “armature formula”.

A pair of strong, prehensile, subchelate and segmented antennae is situated on the prominent cephalic protrusion. They are long, slender and gently curved. Each antenna (Fig. 26B) measures 525 (450-590) µm in length and 190 (150-200) µm in width and consists of four segments; coxobasis and three endopodal segments. The first or basal segment (coxobasis) is stout and short, slightly subtrinagular in shape, little longer than broad, broader at its proximal end and naked from any spines or setae. The second segment is the first endopodal segment. It is the longest one, gently curved inwardly and slightly widened at its proximal end. It bears a single short tooth-like spine at its inner margin, almost at the end of its second third. The third segment is the second endopodal segment. It is narrow, elongate and arched with sides almost parallel, slightly curved inwardly

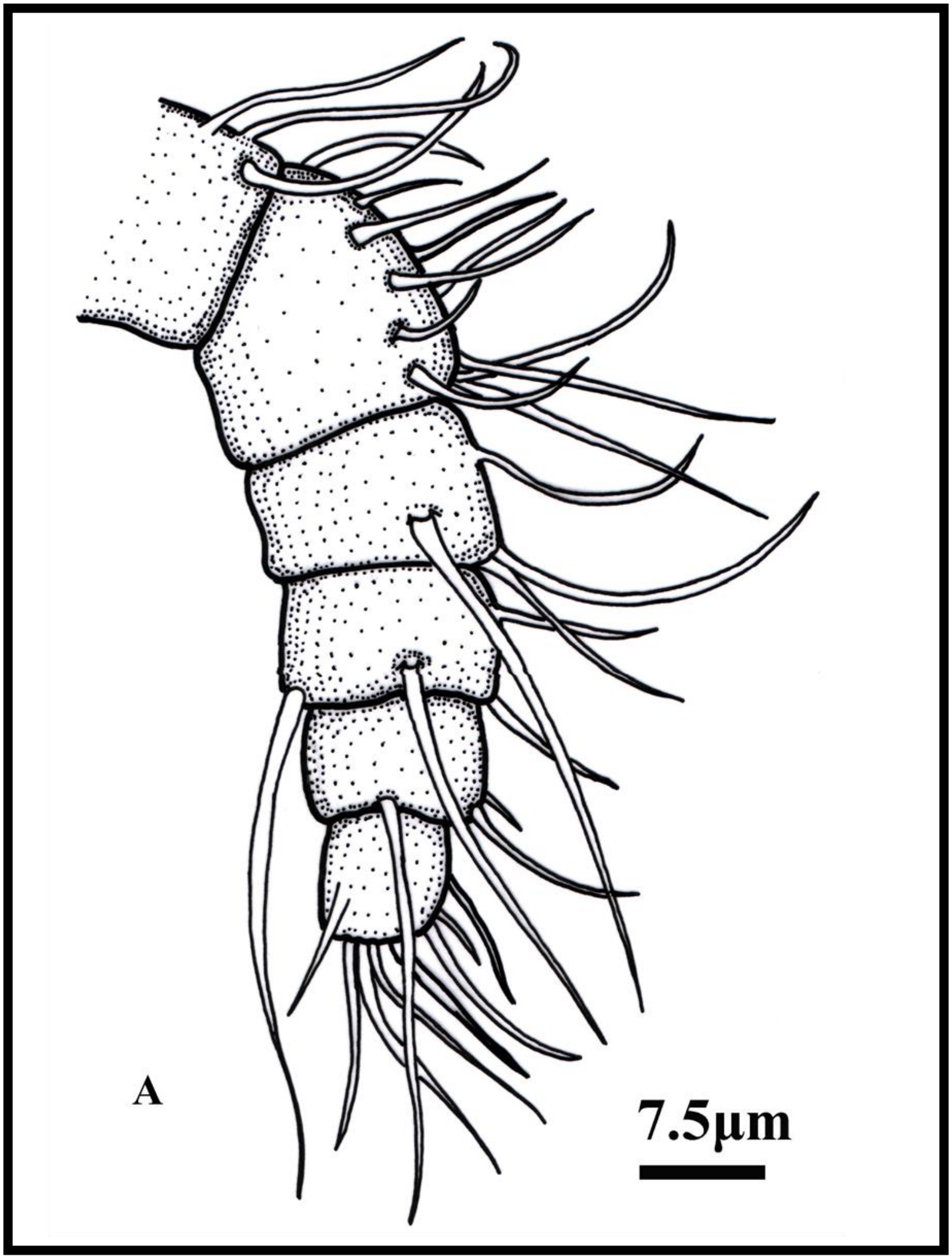


Figure 26A: Schematic drawing of the antennule (A) of the female copepodian parasite**,** *Ergasilus lizae* **Krøyer, 1863.**

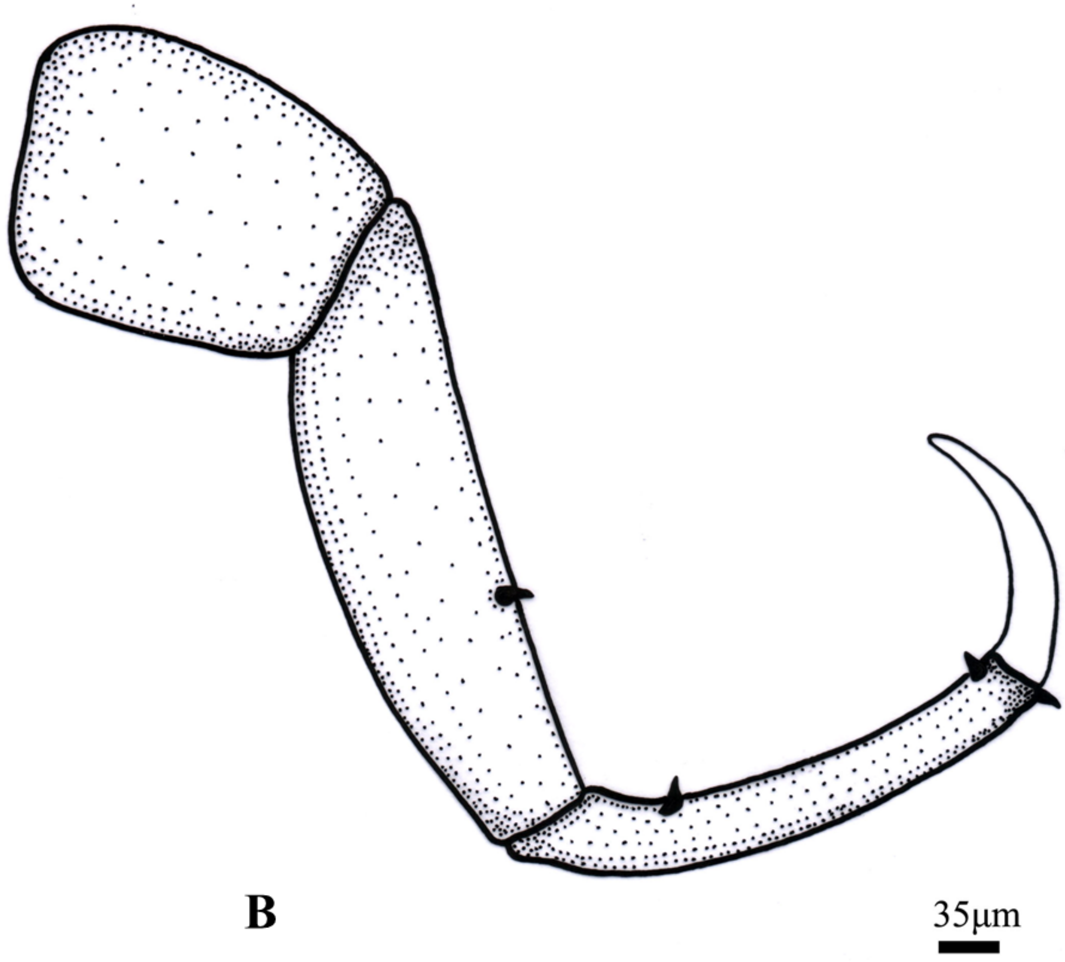


Figure 26B: Schematic drawing of the antenna (B) of the female copepodian parasite**,** *Ergasilus lizae* **Krøyer, 1863.**

and its proximal end slightly widened. It bears a single short tooth-like spine at both proximal and distal end of its inner margin and another short spine at distal end of its outer margin. The fourth or distal segment is the third endopodal segment. It is the form of an elongated, powerful, curved inwardly and sharply pointed hooked claw with smooth margins lacking teeth. It is the shortest one, comprising about half as long as the third segment.

Five pairs of thoracic swimming legs originate from the lateral sides of the first five thoracic somites. The first pair of thoracic legs originates ventral-laterally from the posterior part of the cephalothorax and the last thoracic pair projects from the fifth thoracic somite. The first four pairs are biramous and exist in a completely form. Each pair consists of a proximal coxapod and a distal basipod which bears the two free rami; an exopod and an endopod. Whereas the fifth thoracic pair is uniramous and greatly reduced.

The first pair of swimming legs (Fig. 26C) consists of a coxopod and is ornamented postero-laterally with rows of coars spinules also a basipod ornamented on its postero-medial surface with rows of coarse spinules. The basipod also is provided on its outer margin with a postero-lateral sensilum. Each of exopod and endopod consists of three segments that distally diminish size. The first exopodal segment is provided with a single postero-lateral teeth-like spine projecting from its outer margin and is arnamented with a row of postero-laterally directed strong satules arranged on its inner margin. The second exopodal segment is provided with a single medially situated unarmed seta which projects from the inner margin. The terminal exopodal segment is provided with one antero-lateral and one postero-lateral teeth-like spines besides five naked setae projecting from the posterior margin. The first endopodal segment is provided with a single

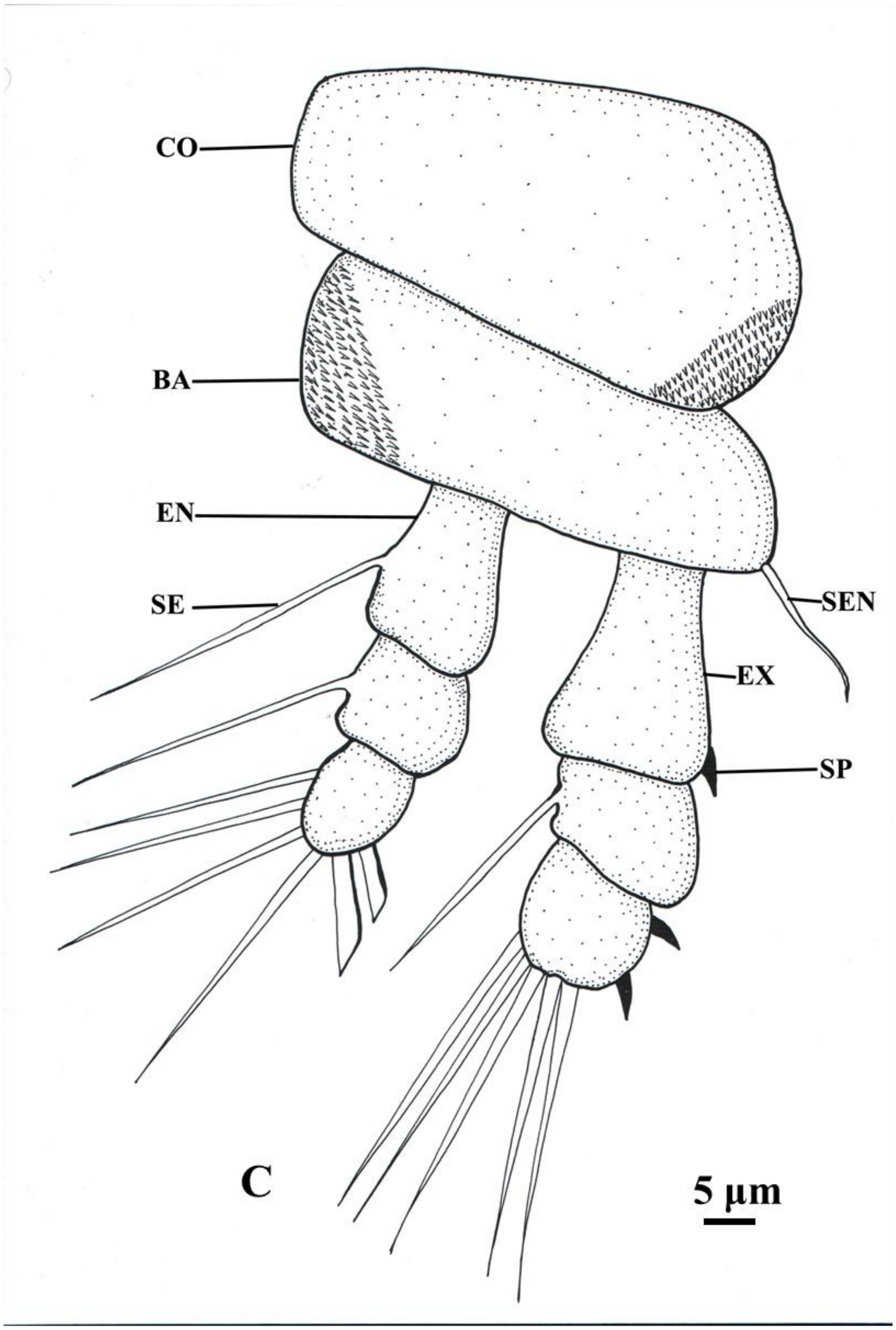


Figure 26C: Schematic drawing of the 1st thoracic leg, (C) of the female copepodian parasite**,** *Ergasilus lizae* **Krøyer, 1863.**

postero-lateral unarmed setae projecting from the inner margin. The second endopodal segment is ornamented with a single medially situated naked seta which projects from the inner margin. The terminal endopodal segment is or namented with two postero-lateral rasp-like spines, as they are serrated at their anterior margins and is ornamented also with four naked setae arranged on its distal margin.

The second and third pairs of thoracic legs (Fig. 26D) are closely similar. The coxopod of each leg is ornamented on its postero-lateral surface with rows of coars spinules. The basipod of each leg is ornamented on its postero-medial surface with rows of coars spinules in addition to a single medially situated sensilum which exists on the outer basipod margin. The first exopodal segment is provided with a single postero-lateral tooth-like spine projects from its outer margin and is ornamented with a single row of postero-laterally directed strong setules arranged on its inner margin. The second exopodal segment is provided with a single medially situated naked seta projects from its inner margin. The terminal exopodal segment is ornamented with six unarmed setae arranged on its posterior margin. The first endopodal segment is ornamented with a single postero-lateral naked seta exists on its inner margin. The second endopodal segment is provided with two postero-lateral naked setae arranged on its inner margin. The terminal endopodal segment is provided with five posteriorly direted unarmed setae arranged on the distal end.

The fourth thoracic leg (Fig. 26E) consists of an ornamented coxapod with rows of coars spinules on its postero-lateral surface and a basipod which is ornamented on its postero-medial surface with rows of coars spinules . Also the basipod is provided with a single medially located sensilum exists on the outer basipodal margin. The exopod consists of two segments whereas the endopod

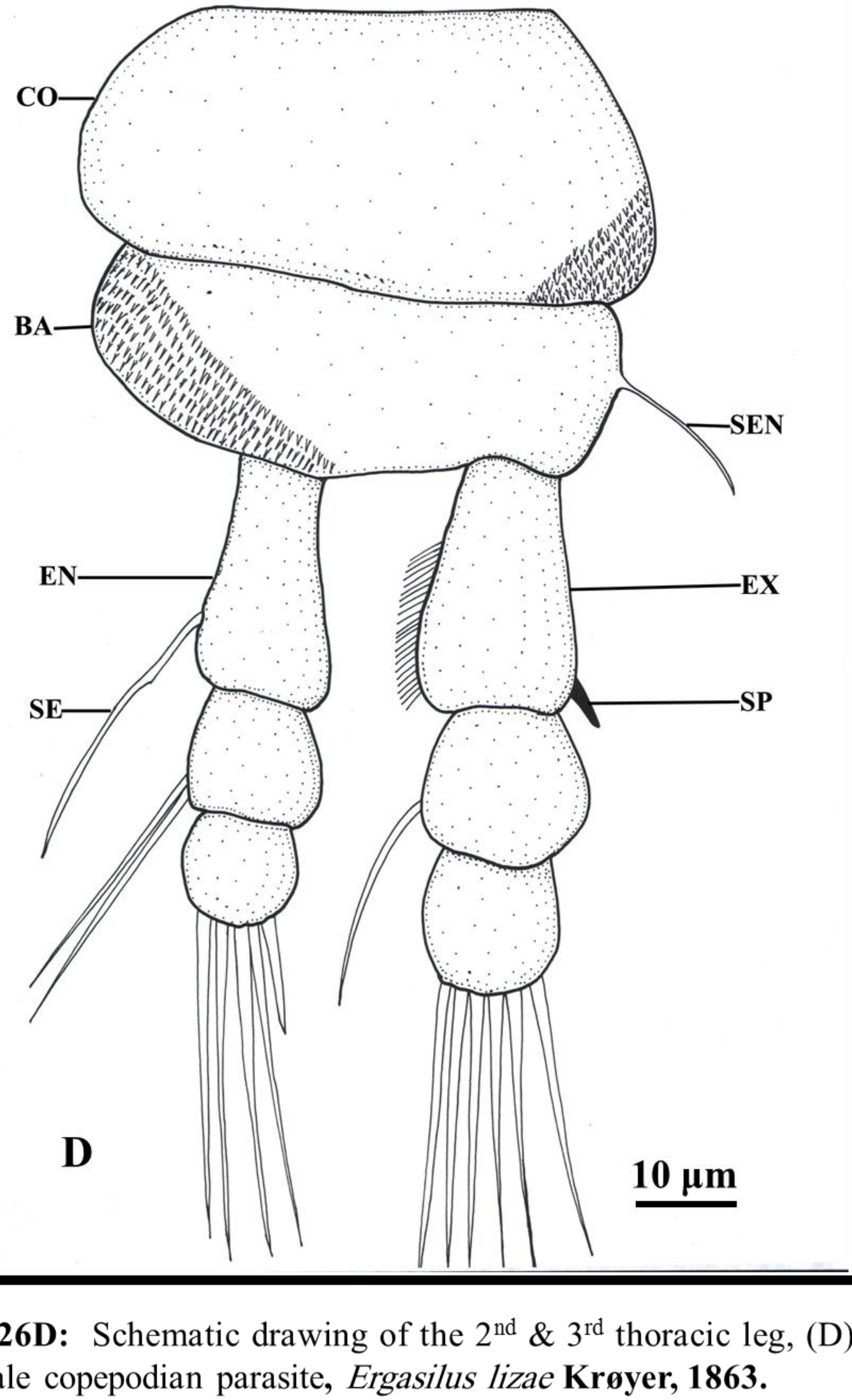


Figure 26D: Schematic drawing of the 2nd & 3rd thoracic leg, (D) of the female copepodian parasite**,** *Ergasilus lizae* **Krøyer, 1863.**

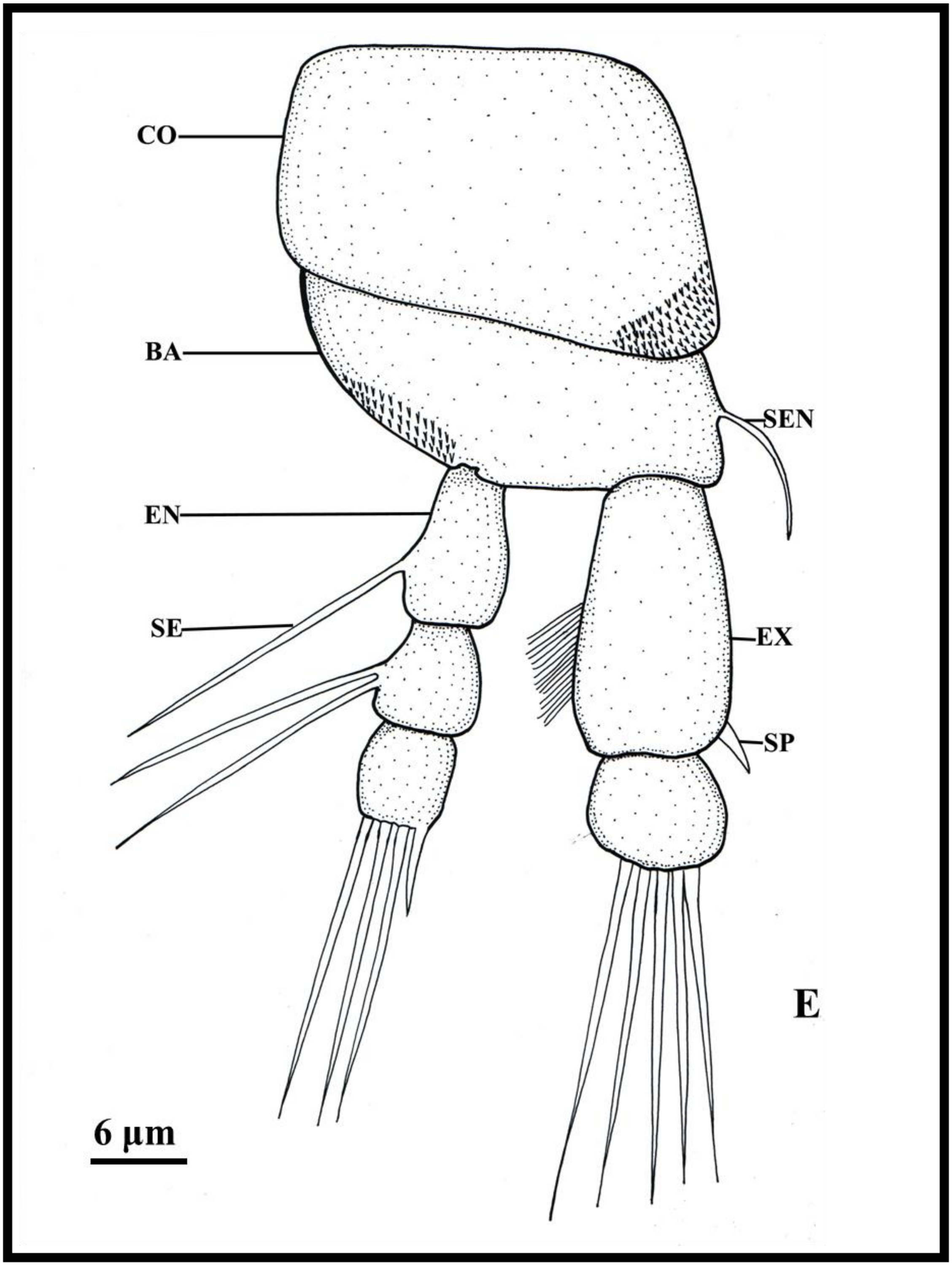


Figure 26E: Schematic drawing of the 4th thoracic leg, (E) of the female copepodian parasite, *Ergasilus lizae* **Krøyer, 1863.**

consists of three segments. Both exopodal and endopodal segments distally diminish in size. The first exopodal segment is provided with a single postero-lateral tooth-like spine which projects from its outer margin in addition to a single row of postero-laterally directed strong setules arranged on its inner margin. The second or terminal exopodal segment is provided with five naked setae projecting from the posterior margin. The first endopodal segment is provided with a single medially situated naked seta projecting from the inner margin. The second endopodal segment is provided with two laterally situated naked setae arranged on its inner margin. The terminal endopodal segment is provided with a single posteriorly directed spiniform seta and three unarmed arranged on its distal end.

Finally the fifth thoracic leg (Fig. 26F) is extremely reduced and represented by a short coxopod and on elongated basipod. The coxopod is provided with a single postero-lateral unarmed seta originating from the outer margin. The basipod appears as an oblong articulating digitate segment ornamented with two long unarmed setae located at the distal end. The armature formula for spines (Roman numerals) and setae (Arabic numerals) of the thoracic legs of the adult female *Ergasilus lizae* **kroyer, 1863** is shown in Table (6).

The urosome (Fig. 26G) consists of the two last thoracic somites, the three abdominal somites and the caudal rami. The fifth thoracic somite is extremely reduced, very short and narrow but distinctly apart as much from the fourth thoracic segment as from the sixth thoracic segment that follows it. It measures 190 (185-199) µm in length and 75 (65-77) µm in width. The sixth thoracic somite is the genital somite. It is a large somite, slightly broader than long and, wide in the middle.

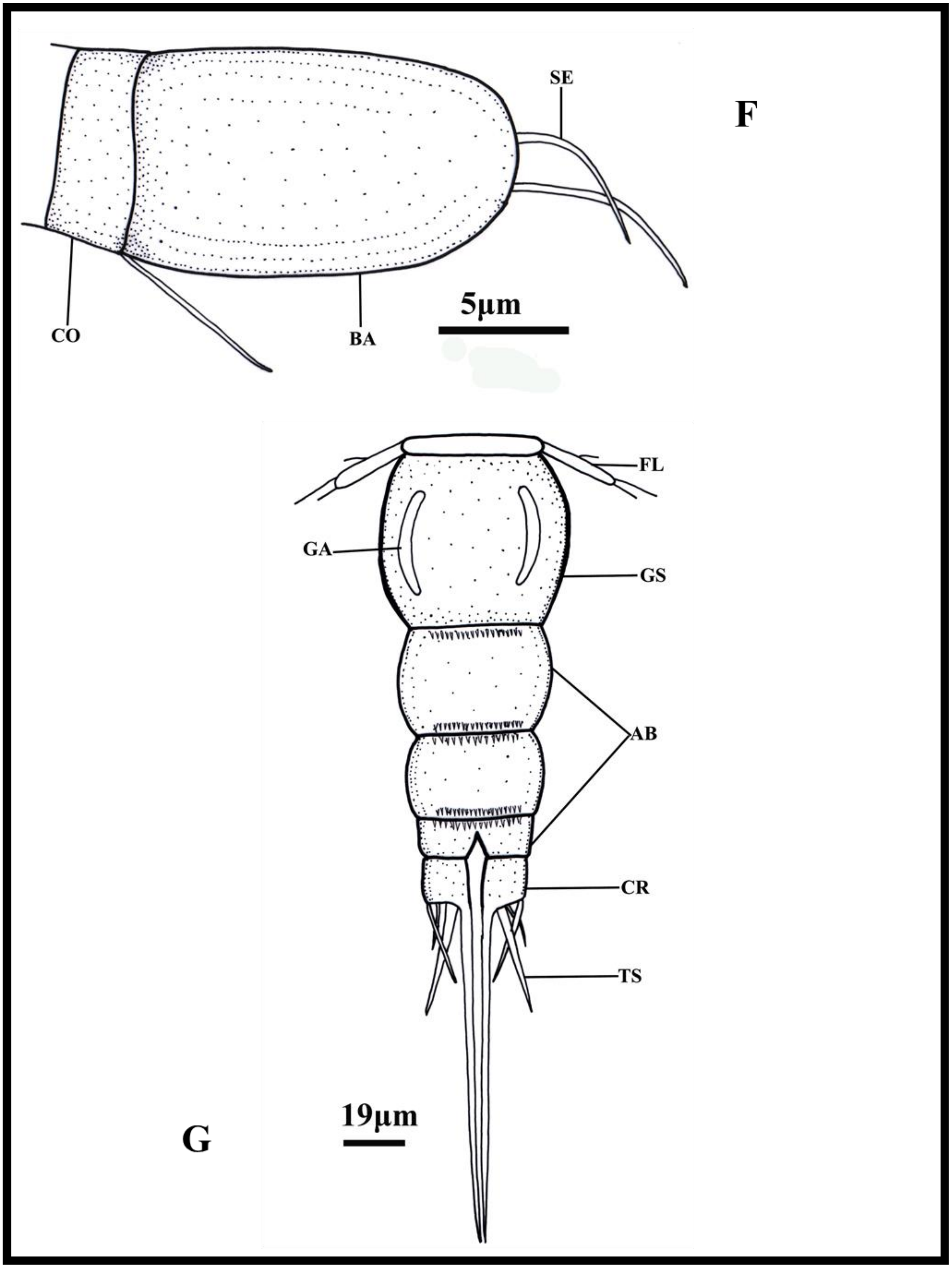


Figure 26F&G: Schematic drawing of the 5th thoracic leg, (F) and urosome, (G) of the female copepodian parasite**,** *Ergasilus lizae* **Krøyer, 1863.**

Table (5): A comparison between the present species of female parasitic copepod *Ergasilus lizae* and the previously described species (in µm).

Author(s)	**Byrnes (1986)**	**Kabata (1992)**	**The present specimens**
Host	*Acanthogyrus bucheri, A. australis, A. berda*	*Mugil cephalus, Trachystoma petardi*	*Valamugil seheli*
Locality	Australia	Australia	Egypt
Body measurements	-	-	925 (900-950)x368.59 (357.98 – 384.59)
Cephalothorax	29x540_650	274_320x280	352 (350-355)x395 (365-450)
Urosome	-	-	190 (185-199)x75 (65-77)
First antenna	135x24.75	140_144.83x30_44.83	112.5 (100-120.7)x37.5 (30-39.5)
Second antenna	697.5x62.35_90	16.2_22.41x40_61.67	525 (450-590)x190 (150-200)
First thoracic leg (L1)	-	-	75
First thoracic leg (L2)	-	-	65
First thoracic leg (W)	-	-	77.5
Second&third thoracic leg (L1)	-	-	58.75
Second&third thoracic leg (L2)	-	-	66.25
Second&third thoracic leg (W)	-	-	72.5
Fourth thoracic leg (L1)	-	-	57.5
Fourth thoracic leg (L2)	-	-	50
Fourth thoracic leg (W)	-	-	55
Fifth thoracic leg	-	-	50x25
Egg sac	-	-	787.5 (725-850)x225 (220-230)

Thoracic legs	Coxapod	Basipod	Exopod			Endopod		
			1	2	3	1	2	3
First leg	0-0	I-0	I-0	I-1	II-5	0-1	0-1	II-4
Second leg	0-0	I-0	I-0	0-1	I-6	0-1	0-2	0-5
Third leg	0-0	I-0	I-0	0-1	I-6	0-1	0-2	0-5
Fourth leg	0-0	I-0	I-0	I-5	-	0-1	0-2	0-4

Roman numerals = Spines Arabic numerals = Setae

Table (6): The armature formula for spines (Roman numerals) and setae (Aranbic numerals) of the thoracic appendages of the adult female *Ergasilus lizae* **Kroyer, 1863**.

CHAPTER I

Section V

Ergasilus australiensis

Genus: *Ergasilus* von Nordmann, 1832

Ergasilus australiensis Roubal, 1981

(1) Redescription:

The body of the copepodian parasite *Ergasilus australiensis* is illustrated in figures 27 & 28 and its measurements are shown in table (7).

The following redescription is based on the study of fifty four living adult and twenty one mounted specimens of female *Ergasilus australiensis*, where the male is free living and only the adult female is a parasite, collected from the gill filaments of the common two banded sea bream, *Diplodus sargus*.

The body (Fig. 27) of female parasite is panduriform, slightly elongated, markedly expanded anteriorly and progressively narrowed towards the posterior end. The body length of female *Ergasilus australiensis*, excluding caudal setae and egg sacs, is 940 (910-970) µm. The maximum body width is 380 (370-390) µm.

The body (Fig. 27) consists of two main parts; prosome and urosome. The prosome consists of cephalosome and mesosome. The first somite of the mesosome is fully incorporated into the cephalosome forming cephalothorax. There is no suture between the cephalosome and the first mesosomal somite but the boundary between them indicated only by a dorsal transverse welt with shallow notches just anterior to the midlength of the cephalothorax on the lateral margins.

The cephalothorax (Fig. 27) is oblong, enlarged, its anterior two-thirds wider than its posterior one-third and thick at about midlength with conically protruding oral region and barely perceptible projecting antennary region. It measures 342 (340-355) µm in length and 317.5 (300-335) µm in a maximum width that attained at its anterior broader region.

The metasome (Fig. 27) consists of three free somites comprising second, third and fourth thoracic somites. The metasomal somites are broader than long, progressively narrowed from the cephalothorax and gradually diminish in width towards the posterior end of the body. The second thoracic somite is the largest one.

A pair of elongated, cigar-shaped, multiseriated egg sacs (Fig. 27) are originated one from each ventro-lateral side of the genital somite. The egg sacs are long and constituting more than half of the total body length. Each egg sac extends posteriorly with slightly tapering and rounded distal end. Each one measures 850 (750-900) µm in length and 197 (193-205) µm in width. They are filled with a large number of eggs. Eggs are large, spherical and visible through the thin membrane of the eggs.

The abdomen (Fig. 27) consists of three somites. These somites are much wider than long, with sub equal sizes and slightly narrow posteriorly. The first abdominal somite is slightly larger than the following two. The third abdominal somite is deeply incised centrally at its posterior end and almost up to the half of its length. This last somite is noticeably shorter than the previous two. Each forked part of the third abdominal somite carries single caudal ramus. The two caudal rami are slightly longer than broad. Each ramus subquadrate, armed

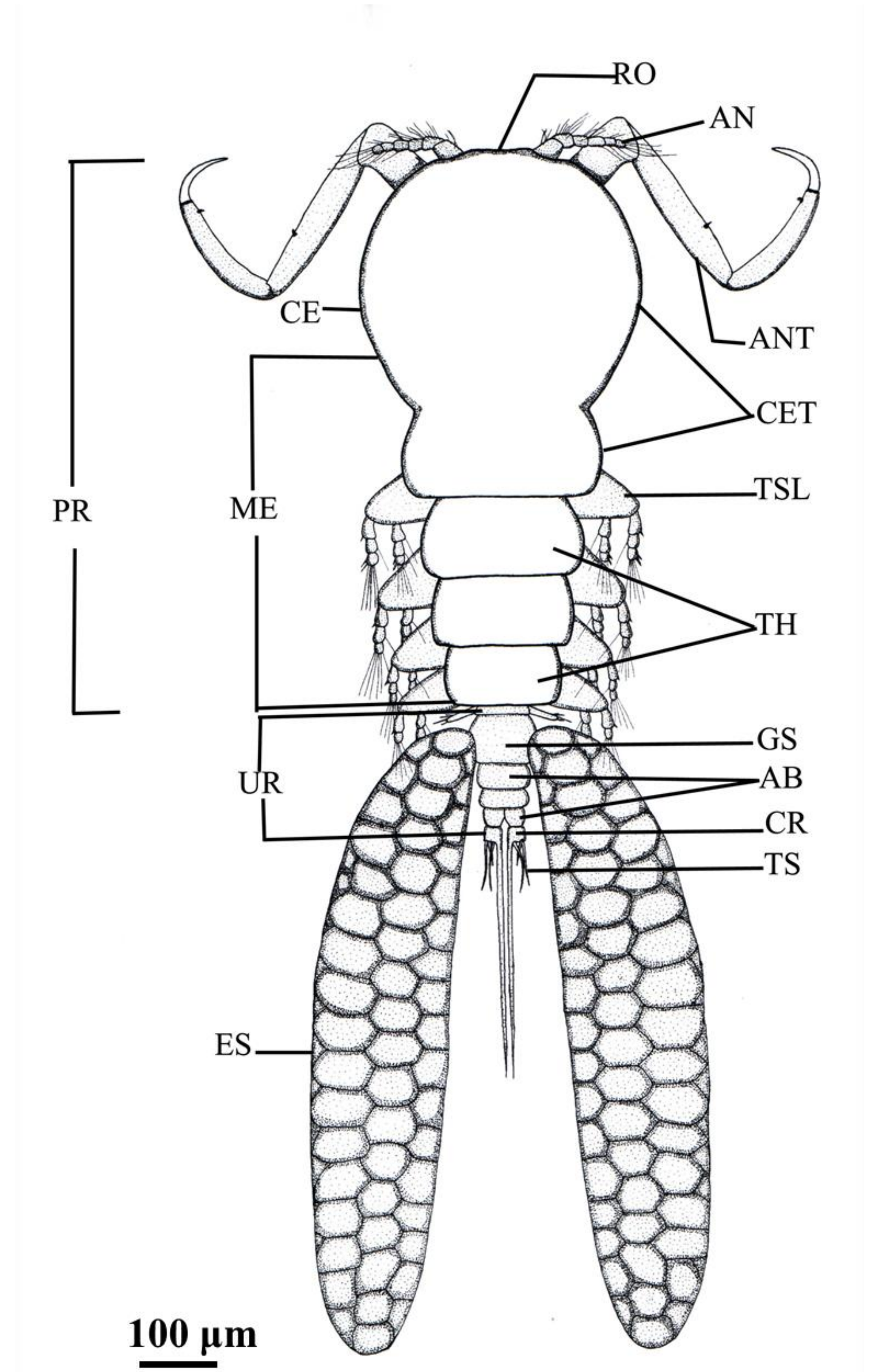


Figure 27A: Schematic drawing of the female copepodian parasite, *Ergasilus australiensis* **Roubal, 1981.**

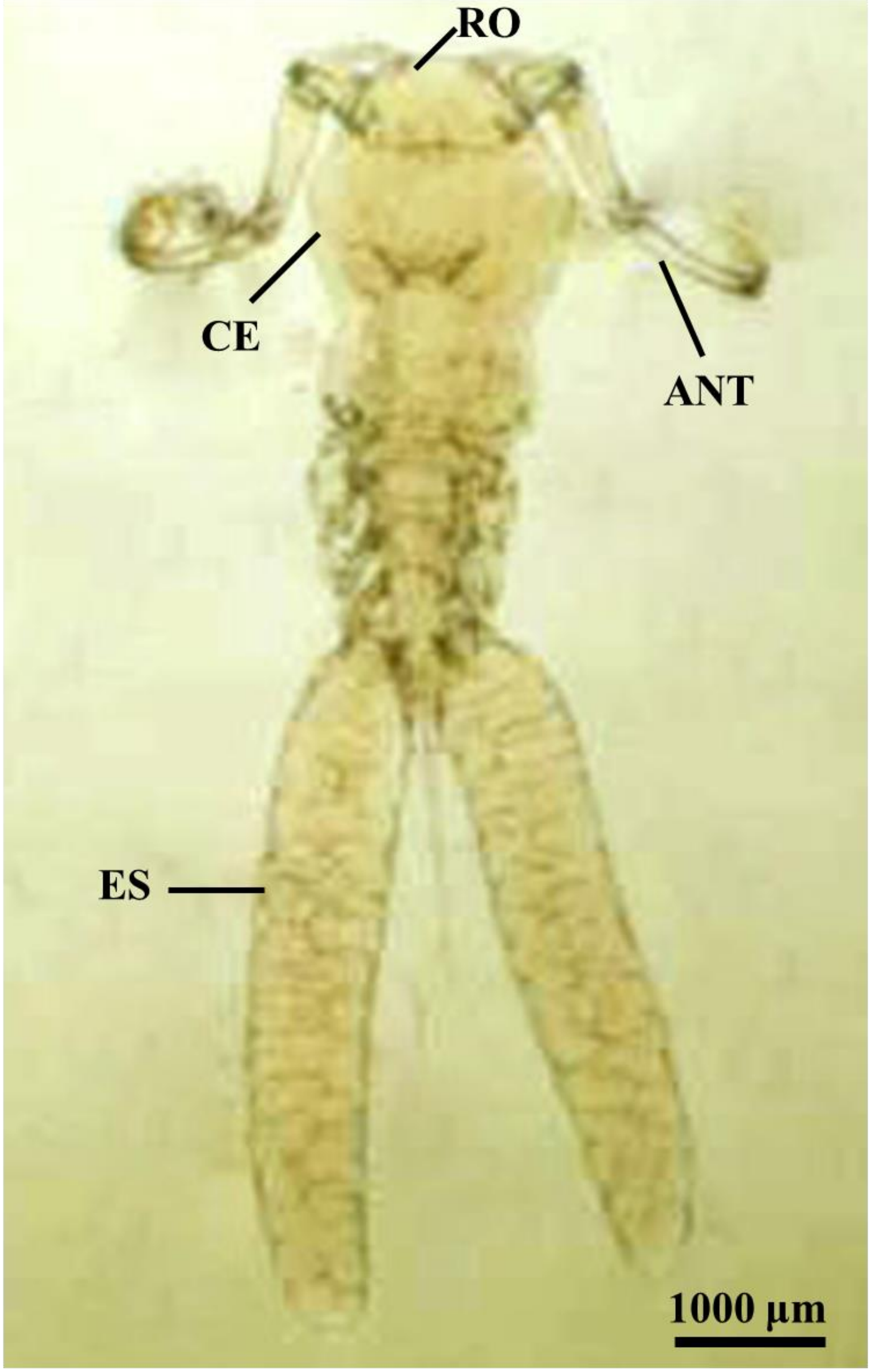


Figure 27B: Photomicrograph of the female copepodian parasite**,** *Ergasilus australiensis* **Roubal, 1981** with lactic acid.

distally with four terminal setae. The inner one is the largest and the outer two are smaller than the inner one. The middle seta is ventrally located.

The cephalothorax bears one pair of antennules and one pair of antennae. A pair of short cylindrical, setiferous, tapering distally and segmented antennules are located on the protuberant rostral area. Each antennule (Fig. 28A) measures 102.5 (100-110) µm in length and 30 (33-40) µm in width and consists of six segments. All antennule segments are provided with numerous simple unarmed setae, principally on anterior-ventral surface. The number of setae on each segment starting from the proximal segment to the distal one is as follows; 3: 12: 3: 4: 3: 8. The number of these antennutary setae is known as “Setal formula” or “armature formula”.

A pair of strong, prehensile, subchelate and segmented antennae (Fig. 28B) is situated on the prominent cephalic protrusion. They are long, slender and gently curved. Each antenna measures 467 (555-580) µm in length and 145 (135-150) µm in width and consists of four segments; coxobasis and three endopodal segments. The first or basal segment (coxobasis) is stout and short, slightly subtrinagular in shape, little longer than broad, broader at its proximal end and naked from any spines or setae. The second segment is the first endopodal segment. It is the longest one, gently curved inwardly and slightly widened at its proximal end. It bears a single short tooth-like spine at its inner margin, almost at the end of its second third. The third segment is the second endopodal segment. It is narrow, elongate and arched with sides almost parallel, slightly curved inwardly and its proximal end slightly widened. It bears a single short tooth-like spine at distal end of its inner margin. The fourth or distal segment is the third endopodal segment. It is the form of an elongated, powerful, curved inwardly and sharply

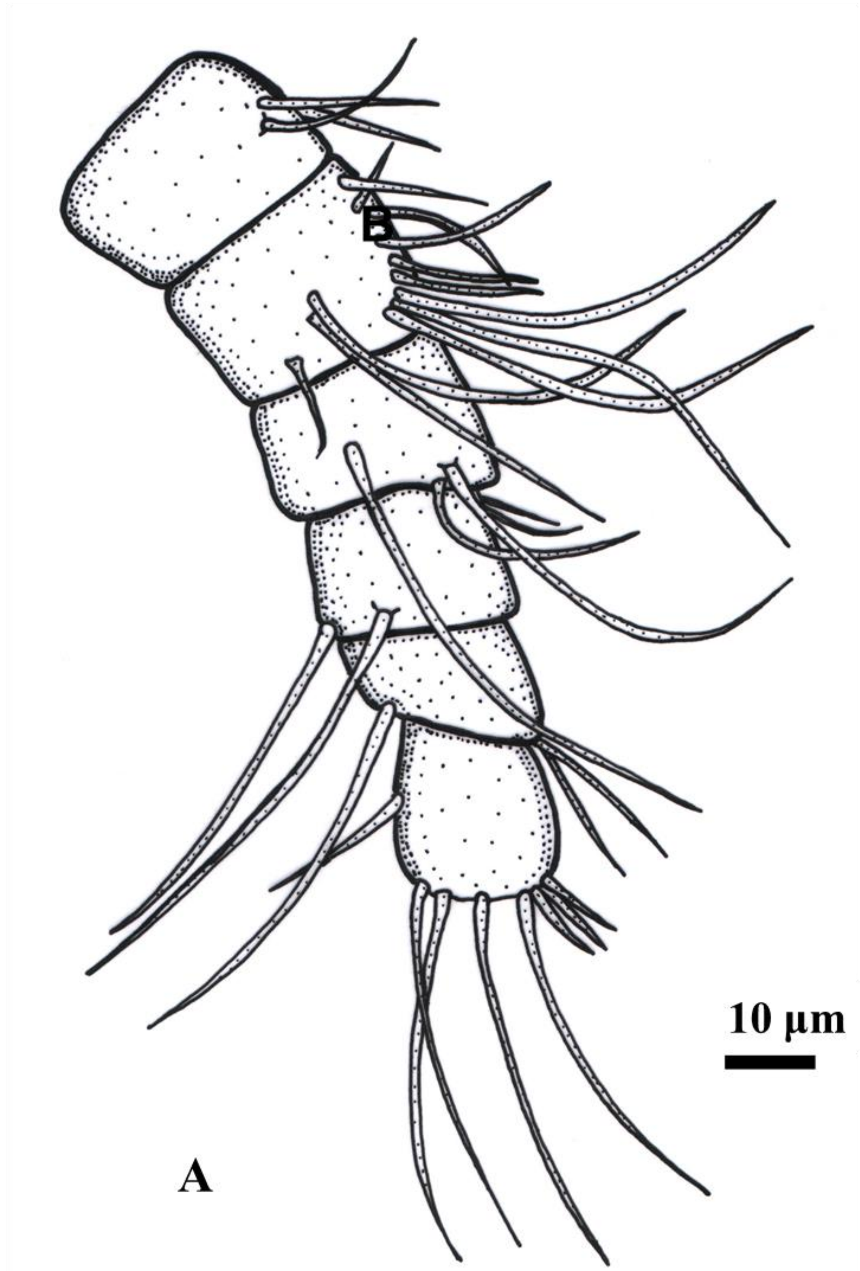


Figure 28A: Schematic drawing of the antennule, (A) of the female copepodian parasite**,** *Ergasilus australiensis* **Roubal, 1981.**

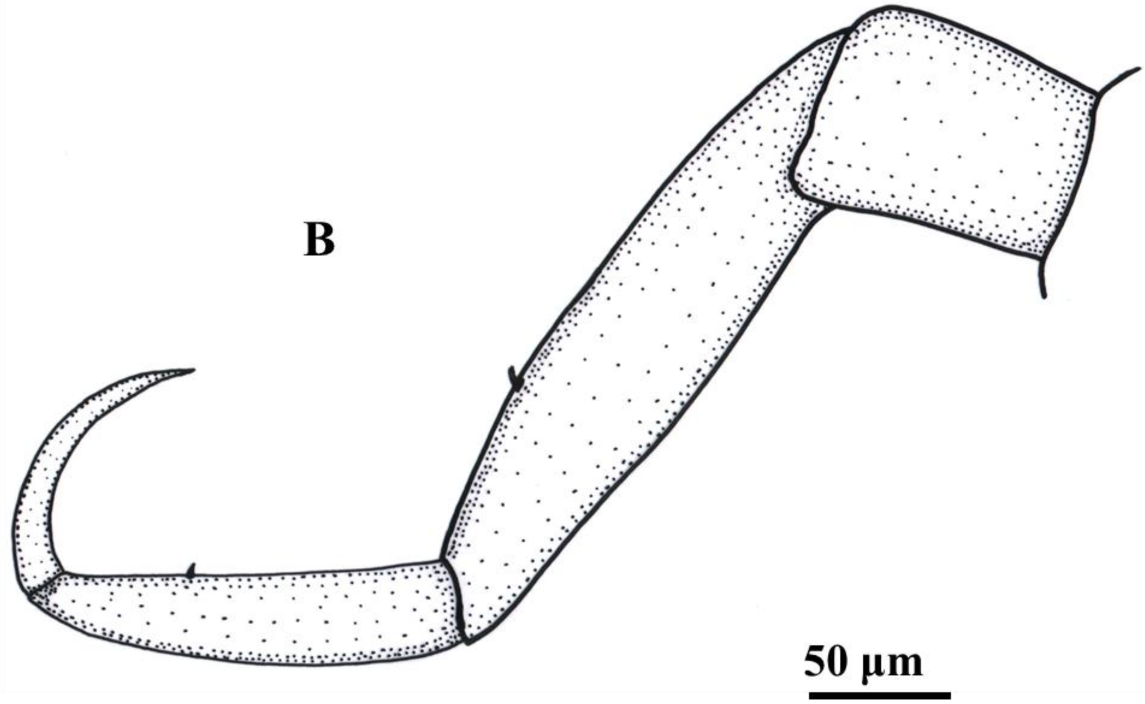


Figure 28B: Schematic drawing of the antenna, (B) of the female copepodian parasite**,** *Ergasilus australiensis* **Roubal, 1981.**

pointed hooked claw with smooth margins lacking teeth. It is the shortest one, comprising about half as long as the third segment.

Five pairs of thoracic swimming legs originate from the lateral sides of the first five thoracic somites. The first pair of thoracic legs originates ventral-laterally from the posterior part of the cephalothorax and the last thoracic pair projects from the fifth thoracic somite. The first four pairs are biramous and exist in a completely form . Each pair consists of a proximal coxapod and a distal basipod which bears the two free rami; and exopod and an endopod. Whereas the fifth thoracic pair is uniramous and greatly reduced.

The first pair of swimming legs (Fig. 28C) consists of a coxopod and is ornamented postero-laterally with rows of coars spinules also a basipod is ornamented on its postero-medial surface with rows of coarse spinules. The basipod also is provided on its outer margin with a postero-lateral sensilum. Each of exopod and endopod consists of three segments that distally diminish in size. The first exopodal segmental is provided with a single postero-lateral teeth-like spine projecting from its outer margin and is ornamented with a row of postero-laterally directed strong setules arranged on its inner margin. The second exopodal segment is provided with a single medially situated unarmed seta which projects from the inner margin. The terminal exopodal segment is provided with one antero-lateral and one postero-lateral teeth-like spines besides five naked setae projecting from the posterior margin. The first endopodal segment is provided with a single postero-lateral unarmed setae projecting from the inner margin. The second endopodal segmental is ornamented with a single medially situated naked seta which projects from the inner margin. The terminal endopodal segment is ornamented with two postero-lateral rasp-like spines, as they are serrated at their

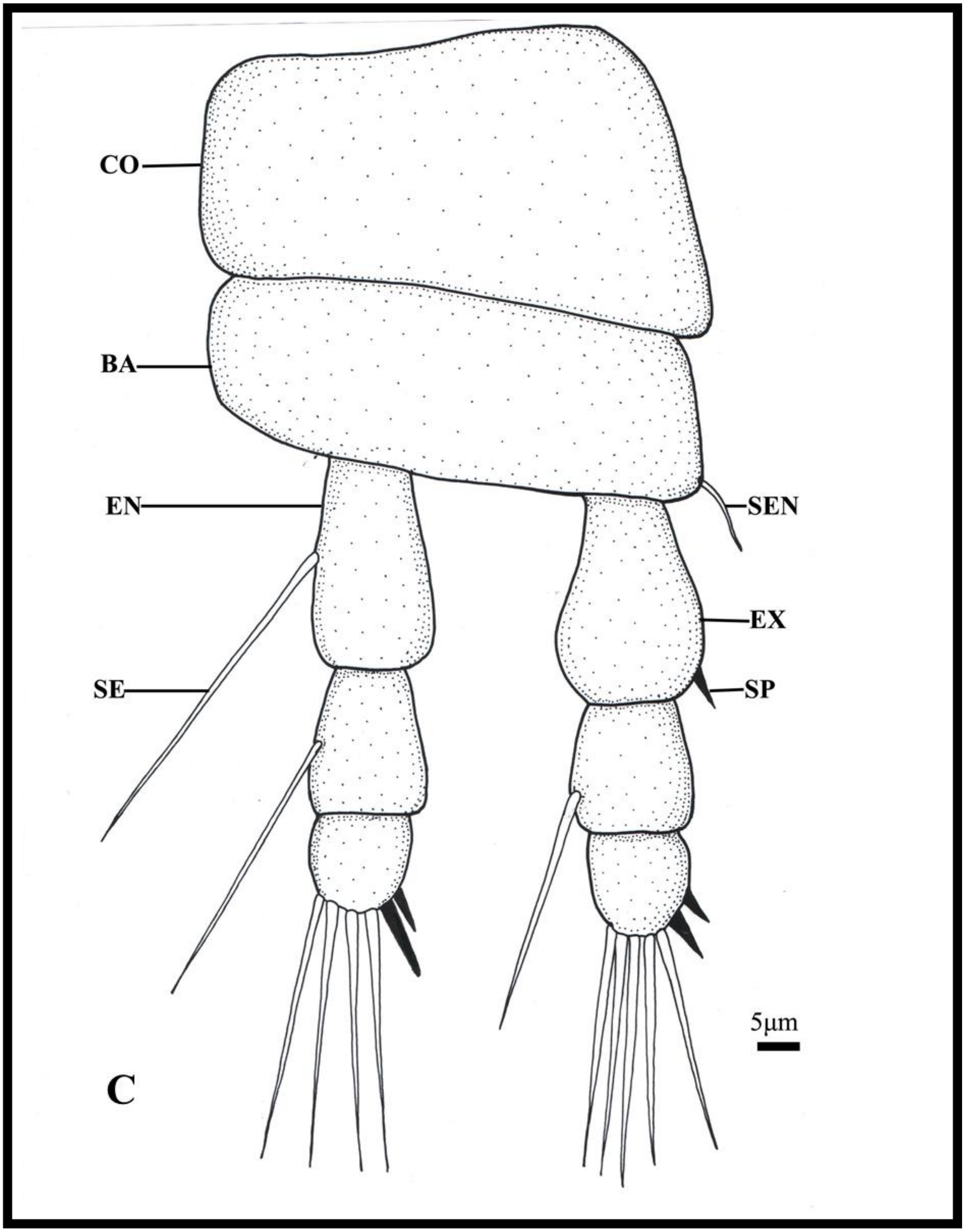


Figure 28C: Schematic drawing of the 1st thoracic leg, (C) of the female copepodian parasite, *Ergasilus australiensis* **Roubal, 1981.**

anterior margins and is ornamented also with four naked setae arranged on its distal margin.

The second and third pairs of thoracic legs (Fig. 28D) are closely similar. The coxopod of each leg is ornamented on its postero-lateral surface with rows of coars spinules. The basipod of each legs ornamented on its postero-medial surface with rows of coars spinules in addition to a single medially situated sensilum which exists on the outer basipod margin. The first exopodal segment is provided with a single postero-lateral tooth-like spine projects from its outer margin and is ornamented with a single row of postero-laterally directed strong setules arranged on its inner margin. The second exopodal segment is provided with a single medially situated naked seta projects from its inner margin. The terminal exopodal segment is ornamented with six unarmed setae arranged on its posterior margin. The first endopodal segment is ornamented with a single postero-lateral naked seta that exists on its inner margin. The second endopodal segment is provided with two postero-lateral naked setae arranged on its inner margin. The terminal endopodal segment is provided with five posteriorly direted unarmed setae arranged on the distal end.

The fourth thoracic leg (Fig. 28E) consists of ornamented coxapod with rows of coars spinules on its postero-lateral surface and basipod which is ornamented on its postero-medial surface with rows of coars spinules. Also the basipod is provided with a single medially located sensilum exists on the outer basipodal margin. The exopod consists of two segments whereas the endopod consists of three segments. Both exopodal and endopodal segments is distally diminish in size. The first exopodal segment is provided with a single postero-lateral tooth-like spine which projects from its outer margin in addition to a single

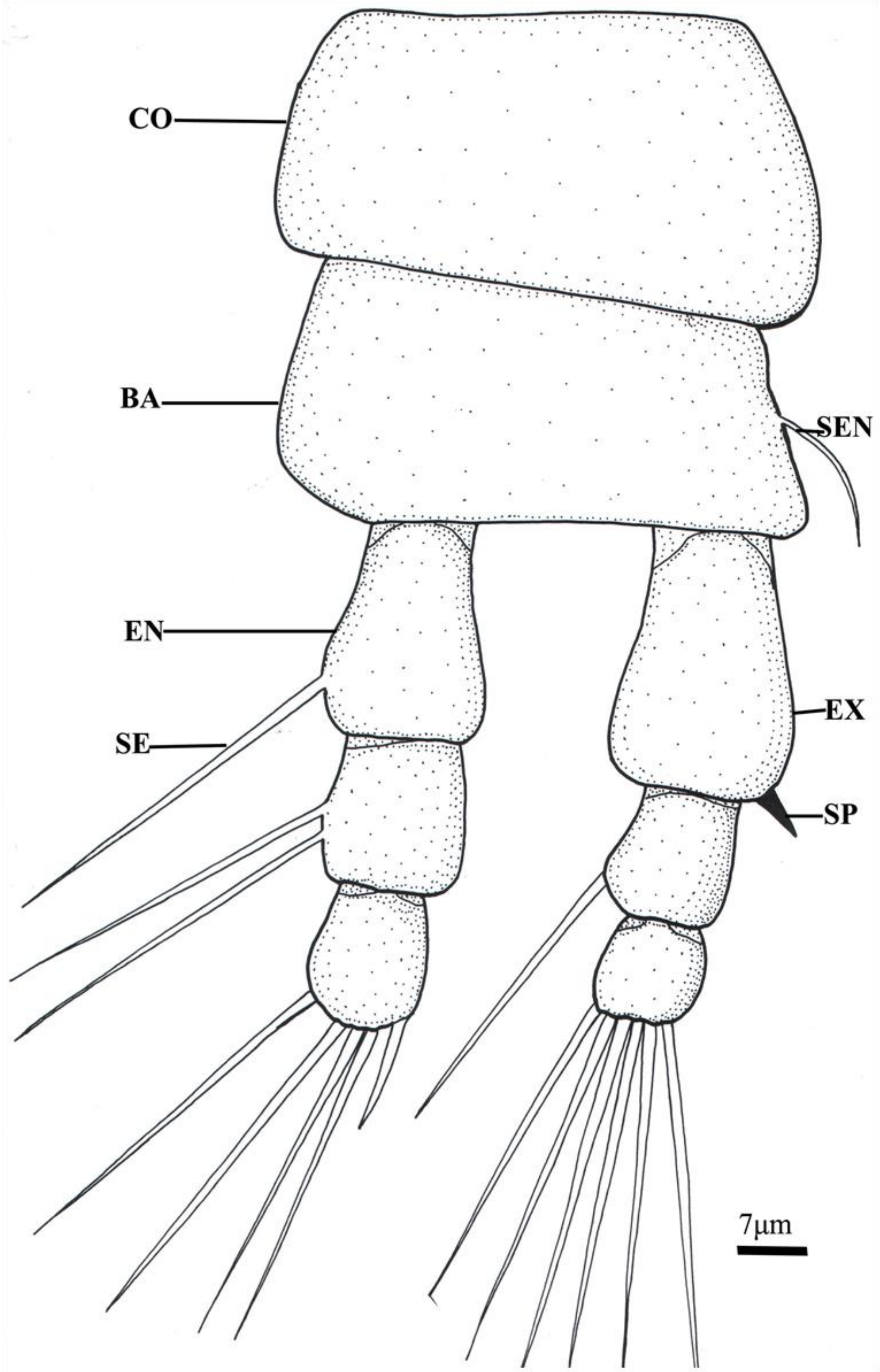


Figure 28D: Schematic drawing of the 2nd &3rd thoracic leg, (D) of the female copepodian parasite, *Ergasilus australiensis* **Roubal, 1981.**

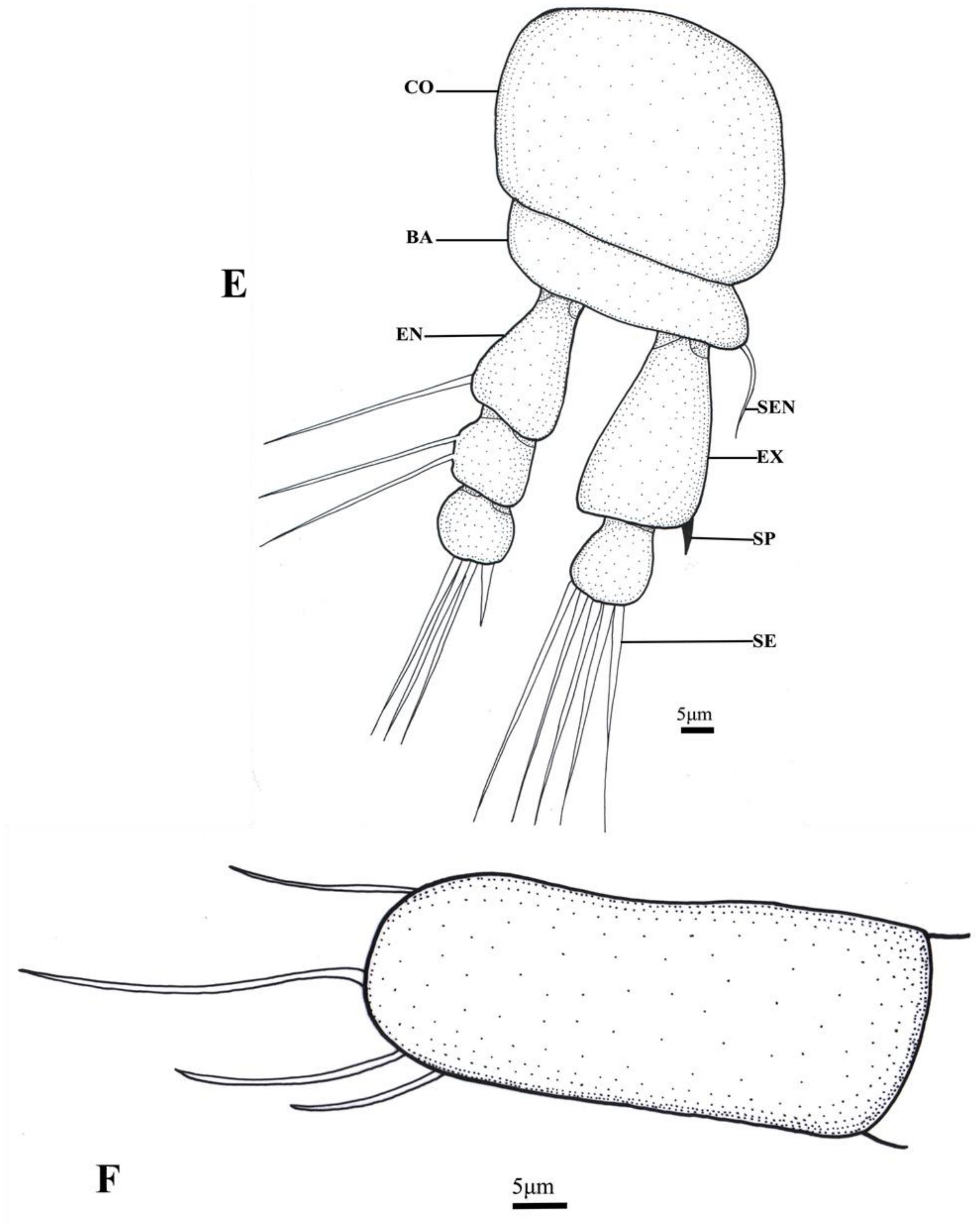


Figure 28E&F: Schematic drawing of the 4th thoracic leg, (E) and 5th thoracic leg, (F) of the female copepodian parasite**,** *Ergasilus australiensis* **Roubal, 1981.**

row of postero-laterally directed strong setules arranged on its inner margin. The second or terminal exopodal segment is provided with five naked setae projecting from the posterior margin. The first endopodal segment is provided with a single medially situated naked seta projecting from the inner margin. The second endopodal segment is provided with two laterally situated naked setae arranged on its inner margin. The terminal endopodal segment is provided with a single posteriorly directed spiniform seta and three unarmed setae arranged on its distal end.

Finally the fifth thoracic leg (Fig. 28F) is extremely reduced and represented by a short coxopod and elongated basipod. The coxopod without any setae. The basipod appears as an oblong articulating digitate segment ornamented with four long unarmed setae located at the distal end. The armature formula for spines (Roman numerals) and setae (Arabic numerals) of the thoracic legs of the adult female ***Ergasilus australiensis* Roubal, 1981** is shown in Table (8).

The Urosome (Fig. 28G) consists of the two last thoracic somites, the three abdominal somites and the caudal rami. The fifth thoracic somite is extremely reduced, very short and narrow but distinctly apart as much from the fourth thoracic segment as from the sixth thoracic segment that follows it. It measures 193 (190-195) µm in length and 92 (90-97) µm in width. The sixth thoracic somite is the genital somite. It is a large somite, slightly broader than long, wide in the middle.

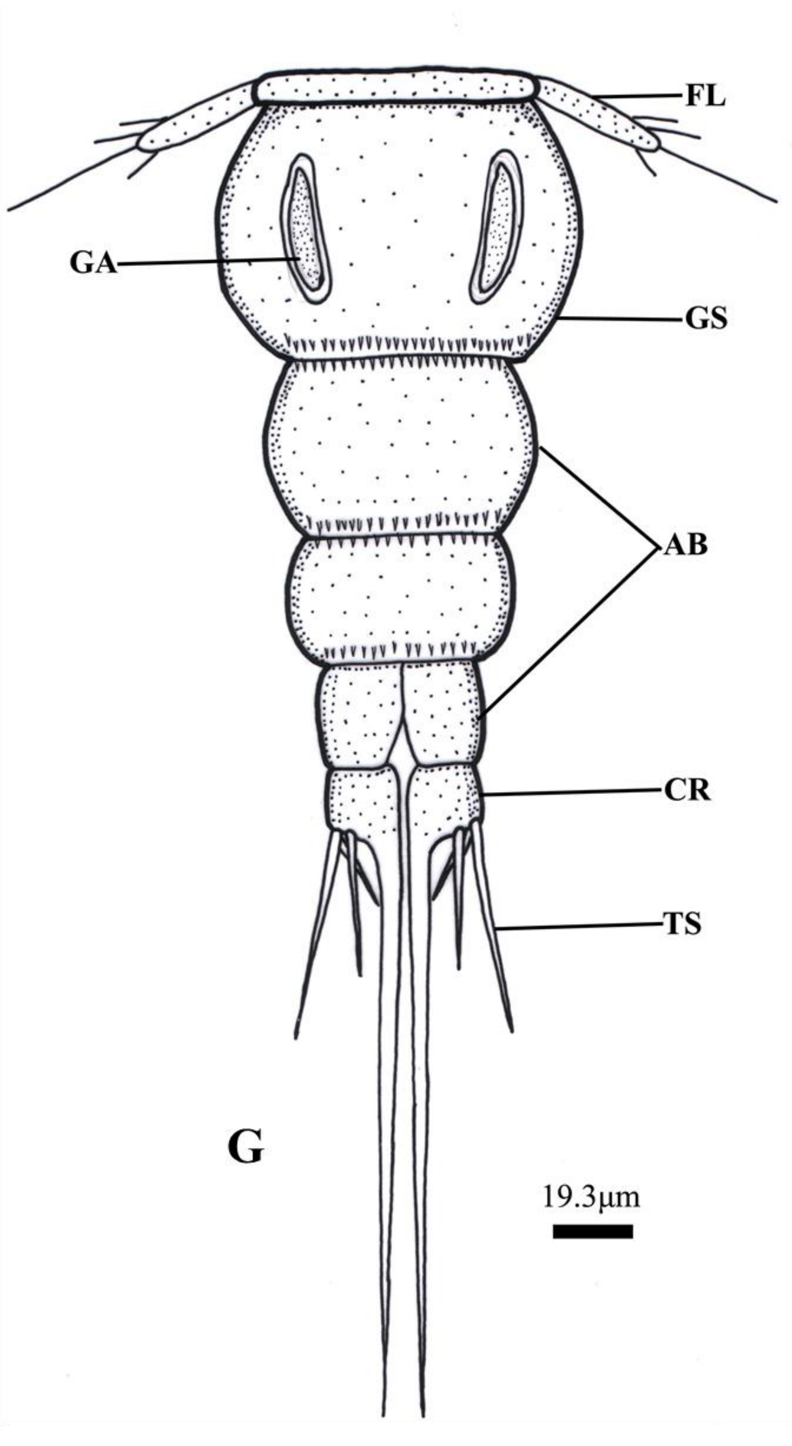


Figure 28G: Schematic drawing of the urosome, (G) of the female copepodian parasite, *Ergasilus australiensis* **Roubal, 1981.**

Table (7): A comparison between the present species of female parasitic copepod *Ergasilus australiensis* and previously described measurements of species (in µm).

Author(s)	**Kabata (1992)**	**The present specimens**
Host	*Toxotes chatareus & Acanthopagrus australis*	*Diplodus sargus*
Locality	North Queensland & New South Wales	Egypt
Body measurements	909 x 290	940 x 380
Cephalothorax	-	342 x 317
Urosome	-	193 x 92
First antenna	126 x 38	102 x 30
Second antenna	490 x 90	467 x 145
First thoracic leg (L1)	122	75
First thoracic leg (L2)	-	70
First thoracic leg (W)	74	80
Second&third thoracic leg (L1)	120	70
Second&third thoracic leg (L2)	-	75
Second&third thoracic leg (W)	80	70
Fourth thoracic leg (L1)	108	75
Fourth thoracic leg (L2)	-	65
Fourth thoracic leg (W)	60	65
Fifth thoracic leg	-	50 x 20
Egg sac	-	850 x 197

Thoracic legs	Coxapod	Basipod	Exopod			Endopod		
			1	2	3	1	2	3
First leg	0-0	I-0	I-0	0-1	II-5	0-1	0-1	II-4
Second leg	0-0	I-0	I-0	0-1	0-6	0-1	0-2	0-5
Third leg	0-0	I-0	I-0	0-1	0-6	0-1	0-2	0-5
Fourth leg	0-0	I-0	I-0	0-5	-	0-1	0-2	0-4

Roman numerals = Spines Arabic numerals = Setae

Table (8): The armature formula for spines (Roman numerals) and setae (Aranbic numerals) of the thoracic appendages of the adult female *Ergasilus australiensis* **Roubal, 1981.**

CHAPTER I

Section VI

Ergasilus mosulensis

Genus: *Ergasilus* von Nordmann, 1832

Ergasilus mosulensis Rahemo, 1982

(1) Redescription:

The body of the copepodian parasite *Ergasilus mosulensis* is illustrated in figures 29 & 30 and its measurements are shown in table (9).

The following redescription is based on the study of fifteen living adult and ten mounted specimens of female *Ergasilus mosulensis*, where the male is free living and only the adult female is a parasite, collected from the gill filaments of the thin grey mullet, *Liza ramada*.

The body (Fig. 29) of the female parasite is slender, elongated, violin-shaped, markedly expanded anteriorly and tapering gradually towards the posterior end. The body length of the female *Ergasilus mosulensis*, excluding caudal setae and egg sacs, is 905 (830-990) µm. The maximum body width that attained at cephalothorax measures 445 (430-450) µm.

The body (Fig. 29) consists of two main parts, prosome and urosome. The prosome consists of cephalosome and mesosome. The first somite of the mesosome is fully incorporated into the cephalosome forming cephalothorax. There is no suture between the cephalosome and the first mesosomal somite but the boundary between them is indicated only by a deep indentical constriction below the middle of the cephalothorax.

The cephalothorax (Fig. 29) is oblong, guitar-shaped with an anterior lobe, the cephalosome and a posterior lobe, the first mesosomal somite. Its anterior end is bluntly broad with slightly protruding antennary region and its posterior end is bluntly with two rounded posteriorly lateral margins. It measures 375 (370-400) µm in length and 407.5 (400-420) µm in a maximum width that attained at its anterior broader region.

The mesosome (Fig. 29) consists of three free somites comprising second, third and fourth thoracic somites. The mesosomal somites are broader than long, progressively narrowed from the cephalothorax and decreased in size towards the posterior end of the body. The second thoracic somite is the largest one.

A pair of elongated cigar-shaped, multiseriated egg sacs (Fig. 29) are originate one from each dorso-lateral side of the genital somite. They are long but their length is slighty shorter than the total body length. Each egg sac extends posteriorly with slightly tapering and rounded distal end. Each one measures 655 (550-760) µm in length and 155 (135-180) µm in width. They are filled with large number of eggs about 87-90. Eggs are large, spherical and visible through the thin membrane of the egg sac.

The abdomen (Fig. 29) consists of three somites. Their somites are wider than long, with almost similar width and narrow slightly posteriorly. The first and last abdominal somites are approximately similar in length. The first somite is slightly larger than the second one. The third somite is partially bifurcated posteriorly almost up to the half of its length. This last somite is longer than the previous two. A transverse row of spines is arranged on the posterior margin of the ventral surface of each abdominal somite. Each forked part of the third abdominal somite carries a single caudal ramus (uropod). The two caudal rami are slightly longer than wide. Each ramus measures 173 (155-197.5) µm in length and 102 (100-110) µm in width. Each one is ornamented ventrally on its posterior margin with a single row of spines. Additionally, it is armed distally with four well developed terminal setae. The inner one is the longest. The outer one is

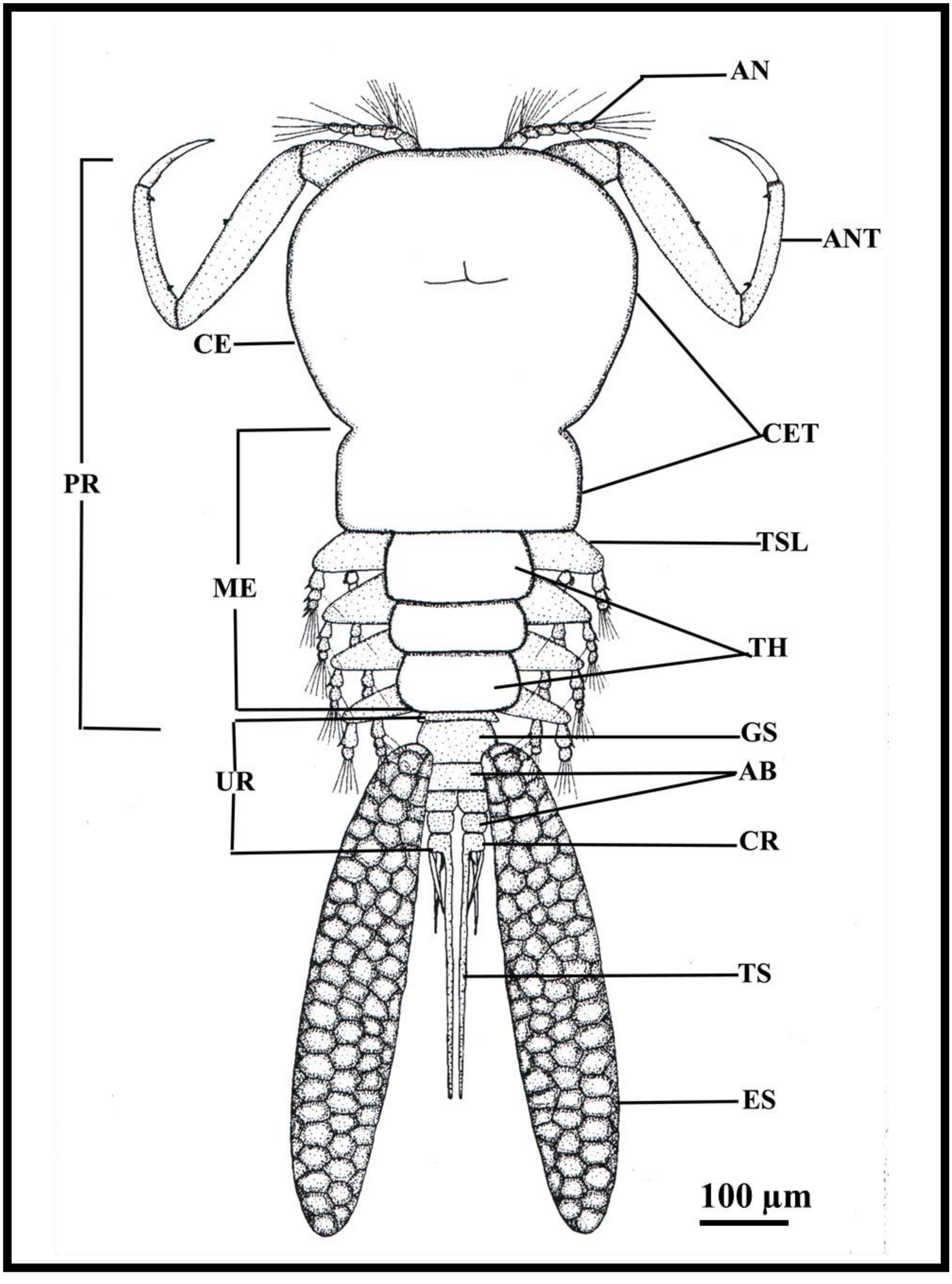


Figure 29A: Schematic drawing of the female copepodian parasite**,** *Ergasilus mosulensis* **Rahemo, 1982.**

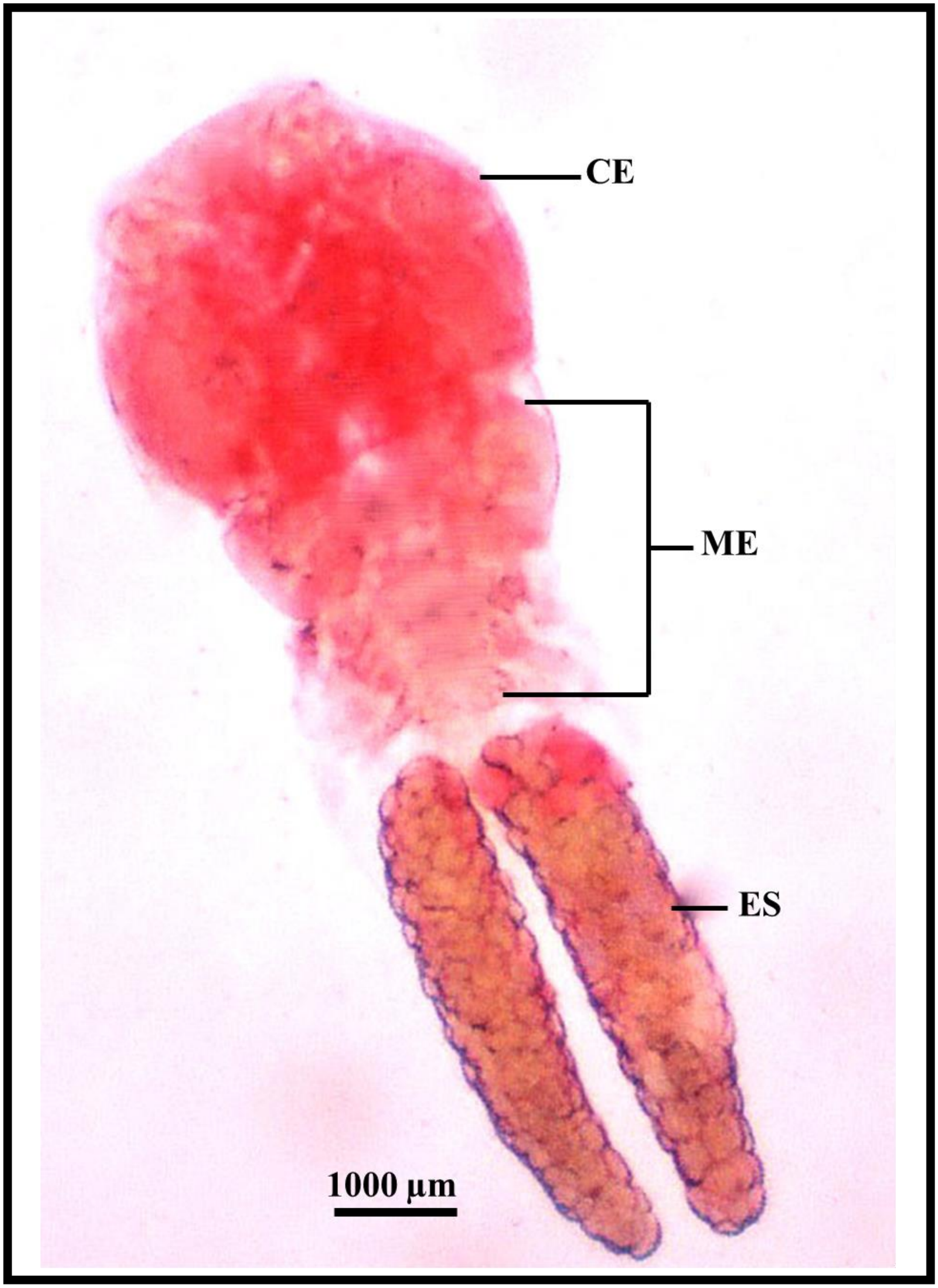


Figure 29B: Photomicrograph of the female copepodian parasite, *Ergasilus mosulensis* **Rahemo, 1982** stained with alum carmine.

shorter, and the middle two are ventrally located. They are small and almost unnoticeable from the dorsal side.

The cephalothorax bears one pair of antennules and one pair of antennae. The pair of cylindrical, setiferous, tapering distally and segmented antennules are located on the protuberant cephalic region. Each antennule (Fig. 30A) measures 112.5 (100-115) µm in length and 25 (20-30) µm in width and consists of six segments. All antennulary segments are provided with numerous naked simple setae, principally on antero-ventral surface. The number of setae on each segment starting from the proximal to the distal one is as follows: 3: 12: 6: 4: 2: 7. The number of these antennulary setae is known as "setal formula".

A pair of stout prehensile, subchelate and segmented antennae (Fig. 30B) are situated on the prominent cephalic protrusion. They are long and slender. Each antenna measures 624 (530-625) µm in length and 160 (125-165) µm and consists of four segments; coxobasis and three endopodal segments. The first or basal segment is the coxobasis. It is short, slightly cubical in shape and is naked from any spines or setae. The second segment is the first endopodal segment. It is the largest one, wides along almost its entire length except at its distal end. It bears a short delicate tooth-like spine at its inner margin of the second half. The third segment is the second enodopodal segment and forms subchela. It is narrow and arched with the sides almost parallel and bears two small teeth-like spines at its inner margin; one near its proximal end and the other distal near the end of the segment. The fourth or distal segment is the third endopodal segment. It is in the form of a stout pointed, clasper-like, fairly straight claw lacking teeth. It is the shortest segment.

Five pairs of thoracic swimming legs originate from the lateral side of the first five thoracic somites. The first pair of thoracic legs originates ventro-laterally

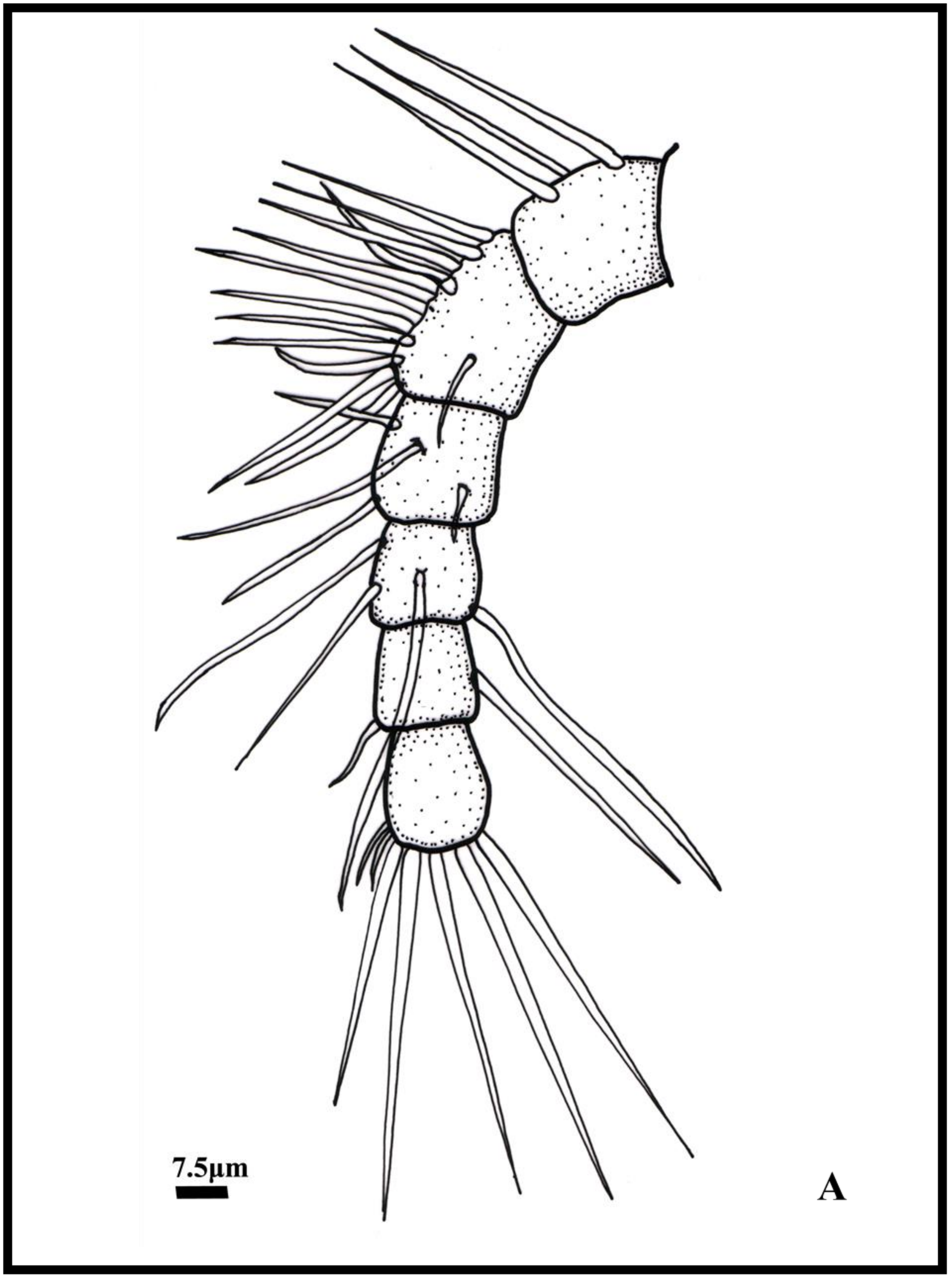


Figure 30A: Schematic drawing of the antennule, (A) of the female copepodian parasite**,** *Ergasilus mosulensis* **Rahemo, 1982.**

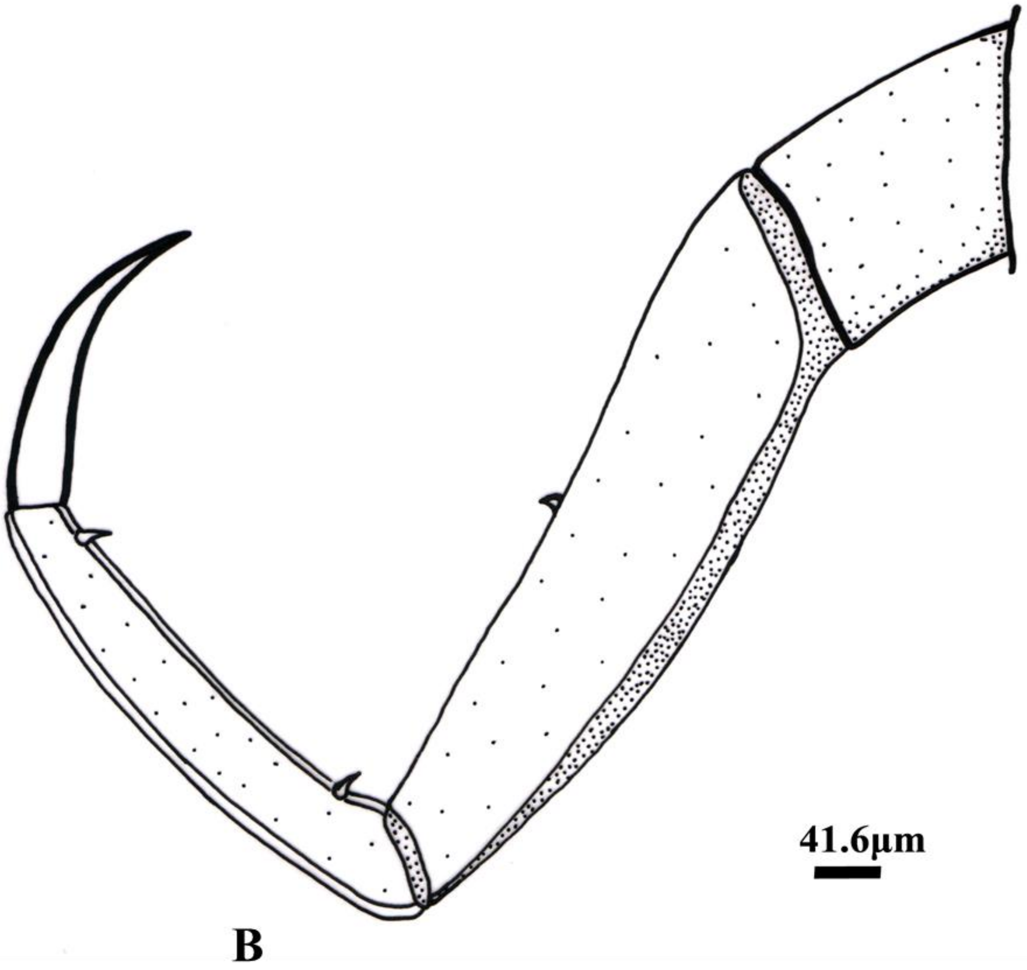


Figure 30B: Schematic drawing of the antenna, (B) of the female copepodian parasite**,** *Ergasilus mosulensis* **Rahemo, 1982.**

from the posterior part of cephalothorax and the last one projects from the fifth thoracic somite. The first four pairs are biramus and completely exist with different parts; a proximal coxapod and a distal basipod which bears both free an exopod and an endopod whilst the fifth pair is extremely reduced.

The first pair of swimming legs (Fig. 30C) consists of a setiferous or serrated coxapod where there is one row of spines arranged on its outer margin and a slightly naked basipod except from a single postero-lateral sensilum originates from the outer margin. Each of the exopod and endopod consists of three segments that diminish distally in size. The three exopodal segments are pectinate laterally; armed with a single row of spinules on the outer margin. The first exopodal segment is provided with a single postero-lateral tooth-like spine projects from its outer margin. Also it is ornamented laterally with a row of strong setules arranged on its inner margin. The second exopodal segment is armed with a single medially situated plumose seta which projects from the inner margin. The terminal exopodal segment is provided with two dorso-lateral teeth-like spines in addition to five plumose setae in which the outer is pectinate. The three endopodal segments are pectinate laterally. Both first and second endopodal segments are pectinate laterally and are provided with single postero-lateral plumose seta which projects from the inner endopodal margin. The terminal endopodal segment is provided with two slightly curved teeth-like spines and four plumose setae.

The second and third pairs of swimming legs (Fig. 30D) are closely similar. The coxapod of each leg is naked from ornamentation. The basipod of both legs is armed with a single antero-lateral sensilum which exists on the outer margin of the basipod. The exopod and endopod of both legs consist of three segments diminishing distally in size. The first exopodal segment is armed with a single postero-lateral tooth-like spine which exists on its outer margin and is

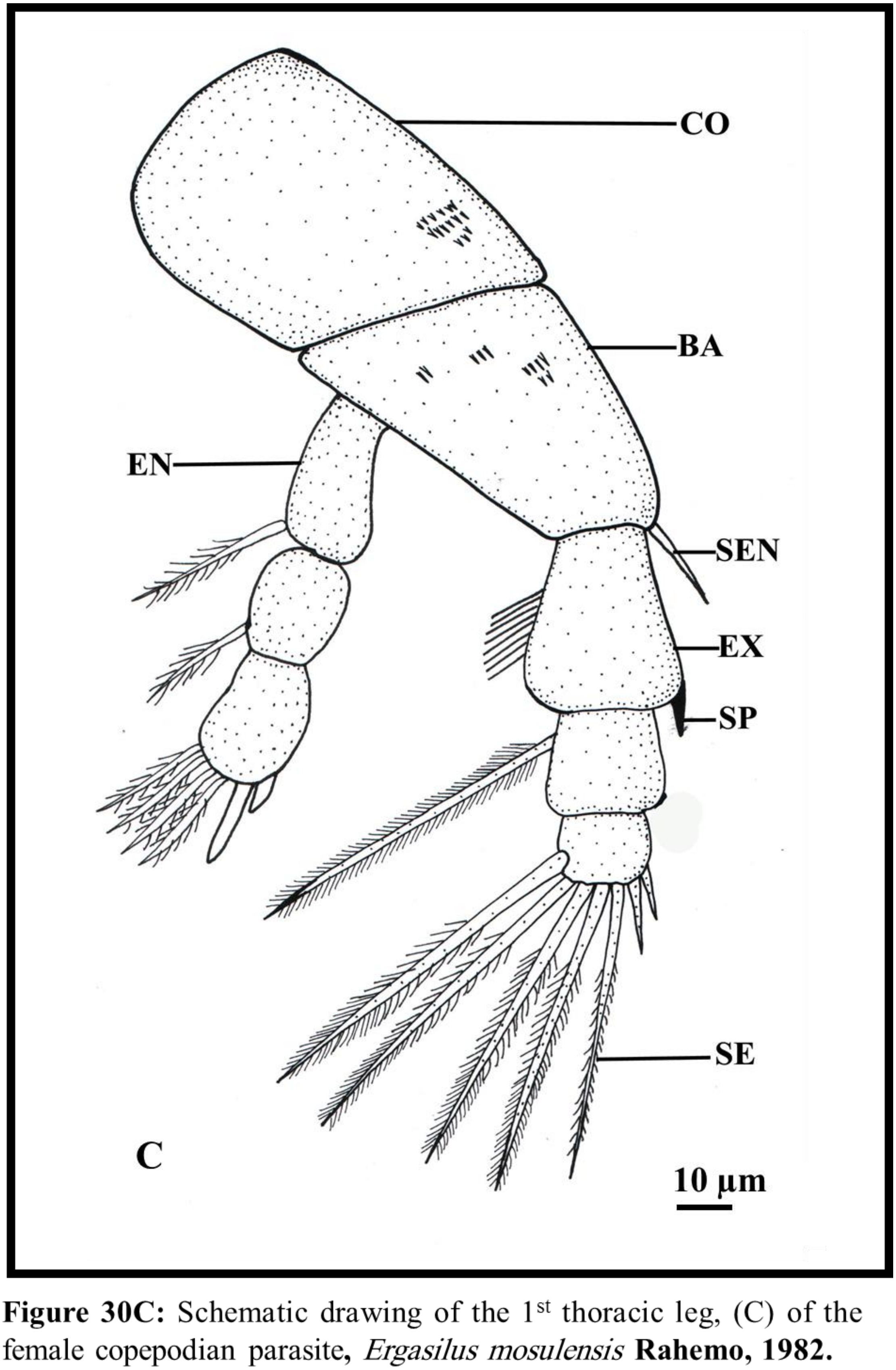


Figure 30C: Schematic drawing of the 1st thoracic leg, (C) of the female copepodian parasite**,** *Ergasilus mosulensis* **Rahemo, 1982.**

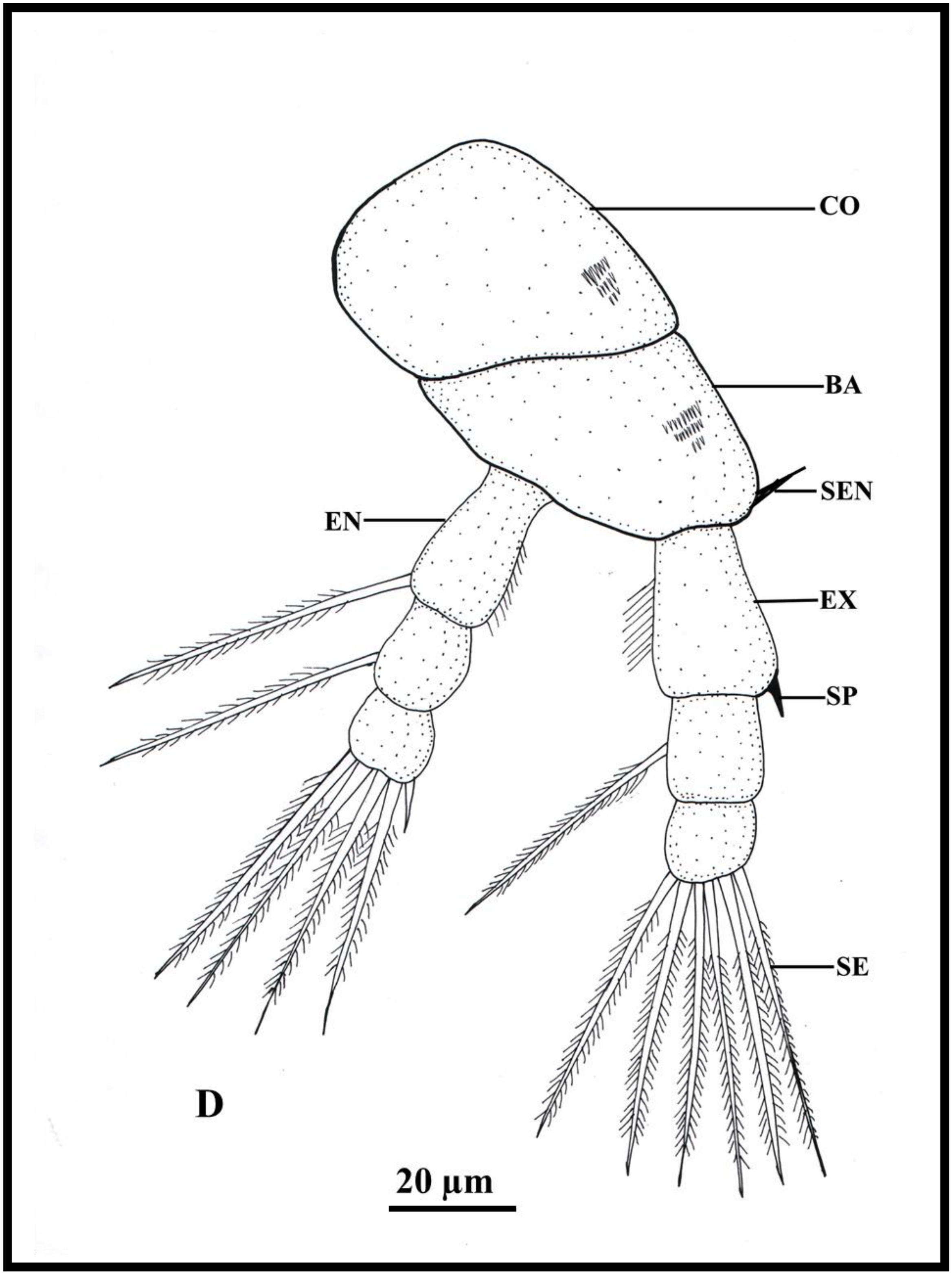


Figure 30D: Schematic drawing of the 2nd & 3rd thoracic leg, (D) of the female copepodian parasite**,** *Ergasilus mosulensis* **Rahemo, 1982.**

ornamented with a single row of strong setules which arranged on its inner margin. The second exopodal segment is provided with a single medially situated plumose seta projecting from its inner margin. The terminal exopodal segment is provided with six distally located plumose seta, in which, where the outer is pectinate. The first endopodal segment is ornamented with a row of postero-laterally strong setules arranged on its outer margin and with a single postero-lateral plumose seta which projects from the inner margin. The second endopodal segment is provided with a single postero-lateral plumose seta projects from the inner margin. The terminal endopodal segment is provided with a single tooth-like spine besides four plumose setae arranged on its distal margin.

The fourth pair of thoracic legs (Fig. 30E) consists of a coxapod naked from ornamentation and an ornamented basipod with a single postero-laterally situated sensilum projects from its outer margin. The exopod consists of two segments whereas the endopod consists of three segments. Both exopodal and endopodal segments get narrower distally. The first exopodal segment is provided with a single postero-lateral tooth-like spine and is ornamented with a row of postero-laterally directed strong setules arranged on its inner margin. The terminal exopodal segment is ornamented with five plumose setae with outer pectinate one. Both first and second endopodal segments are ornamented with a row of postero-laterally directed strong setules, located on their outer margins in addition to a single distally situated row of spines. The first endopodal segment is also provided with a single postero-lateral plumose seta. Whereas the second is ornamented with two postero-lateral plumose setae originating from their inner margins. The terminal endopodal segment is provided with a single tooth-like spine and three plumose setae posteriorly directed.

Finally, the fifth thoracic pair of legs (Fig. 30F) is extremely reduced, uniramous and represented by a short coxapod and an elongated basipod. The coxapod is ornamented with a single postero-lateral naked seta originates from its outer margin. The basipod appears as a relatively large articulating digitate segment ornamented with three apical naked setae, the middle one is the longest. The armature formula for spines (Roman numerals) and setae (Arabic numerals) of the thoracic legs of the adult female *Ergasilus mosulensis* **Rahemo, 1982** is shown in Table (10).

The urosome (Fig. 30G) consists of the two last thoracic somites (fifth and sixth), the abdominal somites and the caudal rami (uropods). The fifth thoracic somite is extremely reduced but clearly delineated from the fourth thoracic segment and the genital segment that follows it. The sixth thoracic somite is the genital somite. It is a barrel-shaped large somite, its width slightly exceeding length, wide in middle and provided with paired slit-like genital apertures longitudinally oriented on the dorso-lateral surface. Furthermore, it is ornamented with a single row of spines on its posterior margin of the ventral surface.

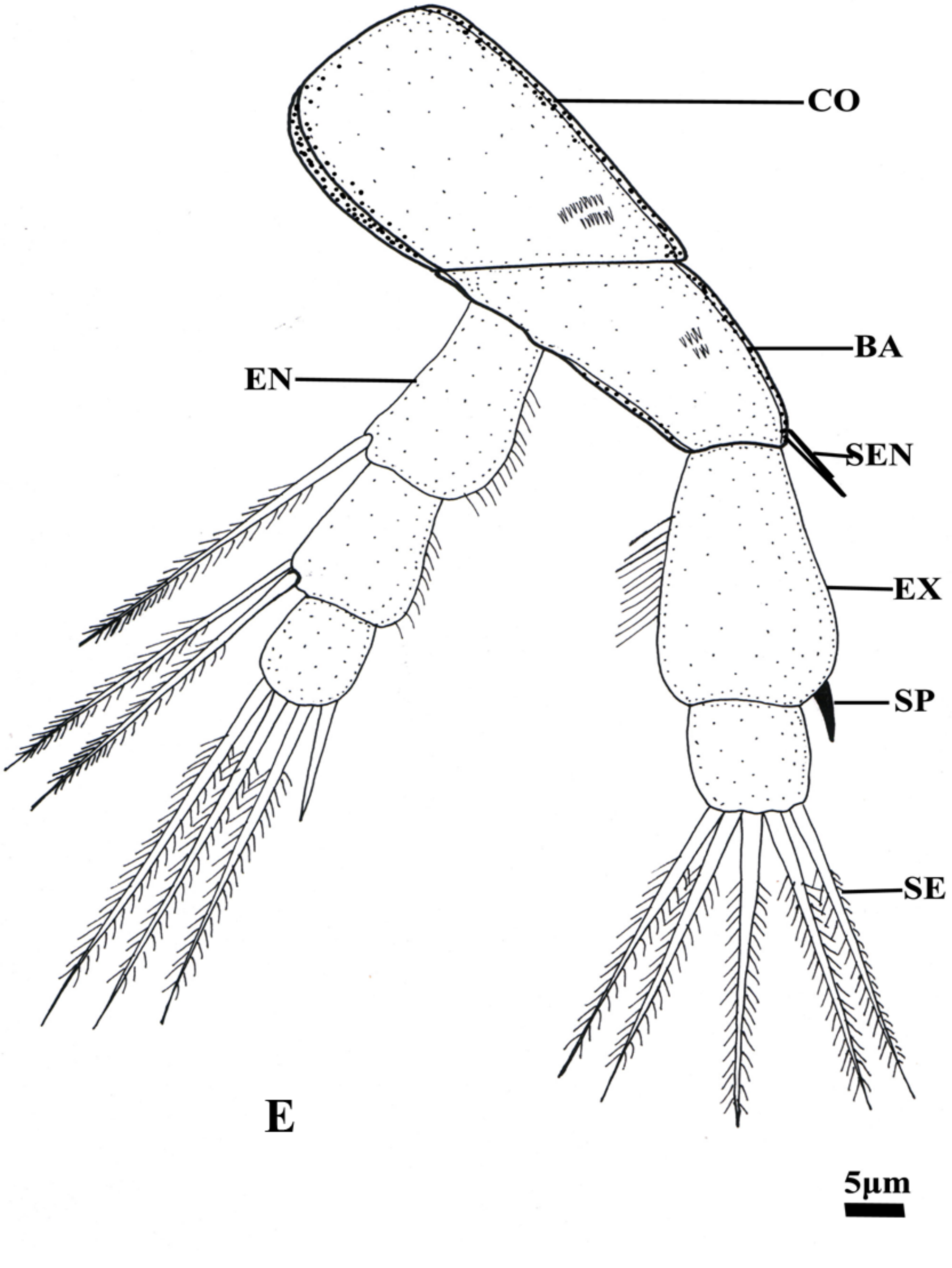


Figure 30E: Schematic drawing of the 4th thoracic leg, (E) of the female copepodian parasite**,** *Ergasilus mosulensis* **Rahemo, 1982.**

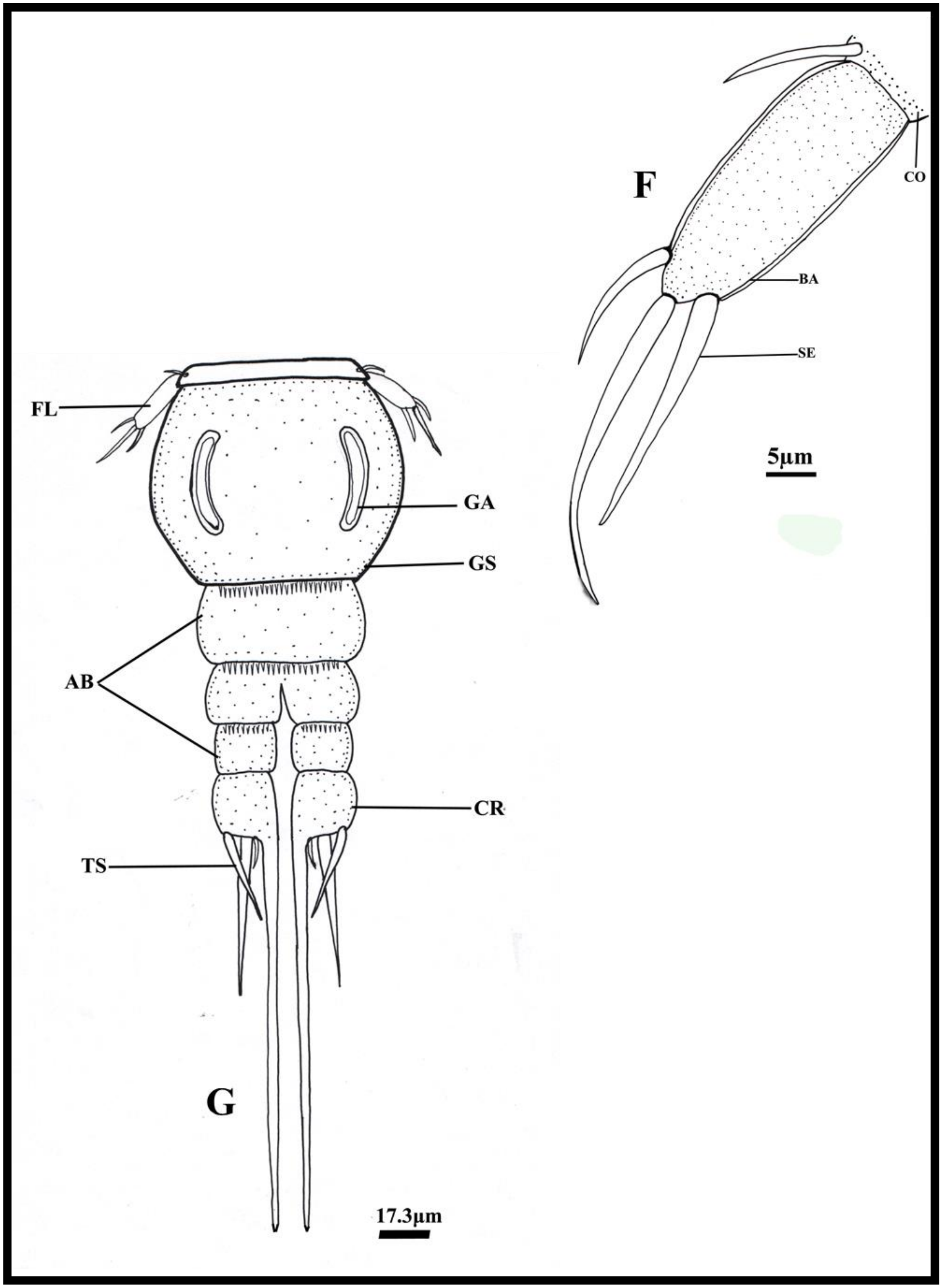


Figure 30F&G: Schematic drawing of the 5th thoracic leg, (F) and urosome, (G) of the female copepodian parasite**,** *Ergasilus mosulensis* **Rahemo, 1982.**

Table (9): A comparison between the present species of female parasitic copepod *Ergasilus mosulensis* and the more related previous descriptions of this species (in µm).

Author(s)	Ho *et al.*,	The present specimens
Host	*Liza abu*	*Liza ramada*
Locality	Iraq	Egypt
Body measurements	940x439	905 x 445
Cephalothorax	510 x 330	375 x 407.5
Urosome	-	173 x 102
First antenna	120.69 x 39.66	112.5 x 25
Second antenna	508.77 x 74.56	624 x 160
First thoracic leg (L1)	-	87.5
First thoracic leg (L2)	-	52.5
First thoracic leg (W)	-	37.5
Second&third thoracic leg (L1)	-	96.25
Second&third thoracic leg (L2)	-	70
Second&third thoracic leg (W)	-	55
Fourth thoracic leg (L1)	-	75
Fourth thoracic leg (L2)	-	50
Fourth thoracic leg (W)	-	27.5
Fifth thoracic leg	-	50 x 25
Egg sac	700_720 x 120_160	655 x 155

Thoracic legs	Coxapod	Basipod	Exopod			Endopod		
			1	2	3	1	2	3
First leg	0-0	I-0	I -0	0-1	II-1,4	0-1	0-1	II-4
Second leg	0-0	I -0	I -0	0-1	0-1,5	0-1	0-1	I-4
Third leg	0-0	I -0	I-0	0-1	0-1,5	0-1	0-1	I-4
Fourth leg	0-0	I -0	I-0	1-5	-	0-1	0-2	I-3

Roman numerals = Spines Arabic numerals = Setae

Table (10): The armature formula for spines (Roman numerals) and setae (Arabic numerals) of the thoracic swimming legs of the adult female *Ergasilus mosulensis* **Rahemo, 1982.**

CHAPTER I

Section VII

Ergasilus celestis

Genus: *Ergasilus* von Nordmann, 1832

Ergasilus celestis Roberts, 1970

(1) Redescription:

The body of the copepodian parasite *Ergasilus celestis* is illustrated in figures 31 & 32 and its measurements are shown in table (11).

The following redescription is based on the study of seventy nine living adult and twenty five mounted specimens of female *Ergasilus celestis*, where the male is free living and only the adult female is a parasite, collected from the gill filaments of the eel fish, *Anguilla anguilla.*

The body (Fig. 31) of the female parasite is slender, elongated, markedly expanded anteriorly and gradually constricted towards the posterior end. The body length of the female *Ergasilus celestis,* excluding caudal stetae and egg sacs is 1025 (900-1050) µm and the width is 405 (370-430) µm.

The body (Fig. 31) consists of two main parts, prosome and urosome. The prosome consists of cephalosome and mesosome. The first somite of the mesosome is fully incorporated into the cephalosome forming cephalothorax which is equal approximately in length to the remaining part of the body. There is no boundary between them is indicated only by a shallow, indistinct constriction, just posterior to the anterior two thirds, and running across both lateral sides.

The cephalothorax (Fig. 31) is elongated. Its anterior end is slightly tapering with slightly projecting antennary region forming a short rostrum and the posterior margin is transversely truncated. The length of cephalothorax is much exceeds its transversely truncated. The length of cephalothorax is much exceeds is width where its length is about half as long as the body length. The cephalothorax decreases in width posteriorly. It measures 550 (490-555) µm in length and 340 (325-350) µm in a maximum width that attained at its anterior broader region.

The mesosome (Fig. 31) consists of three free somites comprising second, third and fourth thoracic somites. The metasomal somites are broader than long, progressively narrowed from cephalothorax and decrease in size towards the posterior end of the body.

The two ovisacs (Fig. 31) are long. Each ovisac extends posteriorly with slightly tapering and rounded distal end. Each one measures 1050 (1000-1055) µm in length and 145 (133-160) µm in width. They are filled with large number of eggs about 70-80. Eggs are large, spherical and visible through the thin membrane of the egg sac.

The abdomen (Fig. 31) consists of three somites. These somites are wider than long, with almost similar width and slightly diminish posteriorly. The first abdominal somite is larger than the following two. The third abdominal somite is notched posteriorly almost up to the half of its length. The last somite is almost equal to or slightly smaller than the second one. On the ventral surface of each abdominal somite, there are two rows of spinules arranged on each posterior margin. Each forked part of the third abdominal somite caries a single caudal ramus. The length of each ramus is longer than its width, almost equal to the last two abdominal segments. Each caudal ramus is armed distally with three terminal setae. The innermost one the largest, the outermost seta is shorter than the innermost one and the middle one is ventrally located.

Cephalthorax bears one pair of antennules and one pair of antennae. The pair of short cylindrical, setiferous, somewhat constricted distally and segmental antennules (Fig. 32A) are located on the protuberant rostral area. Each antennule measures 105 (90-110) µm in length and consists of six segments. The size of the

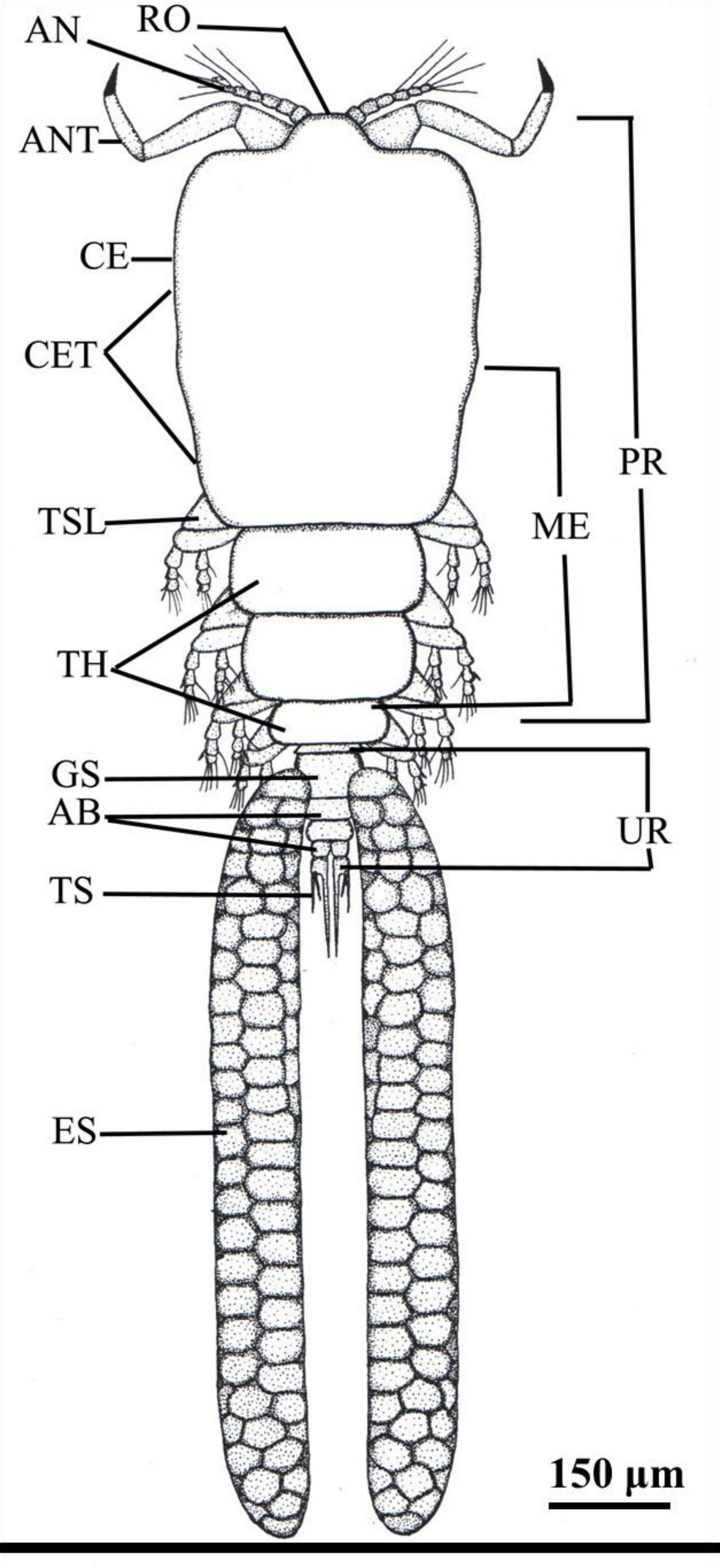


Figure 31A: Schematic drawing of the female copepodian parasite, *Ergasilus celestis* **Roberts, 1970.**

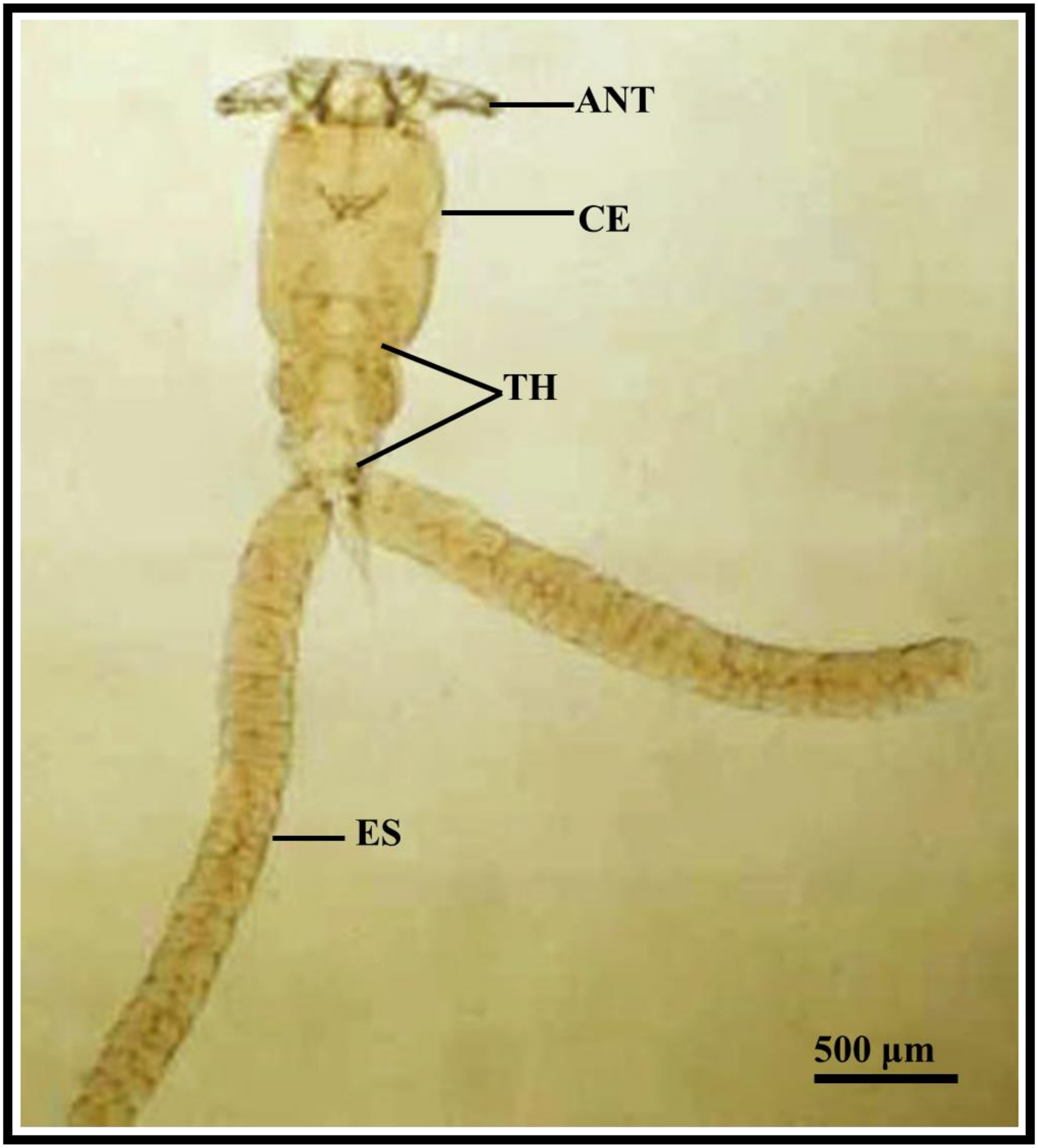


Figure 31B: Photomicrograph of the female copepodian parasite, *Ergasilus celestis* **Roberts, 1970** with lactic acid.

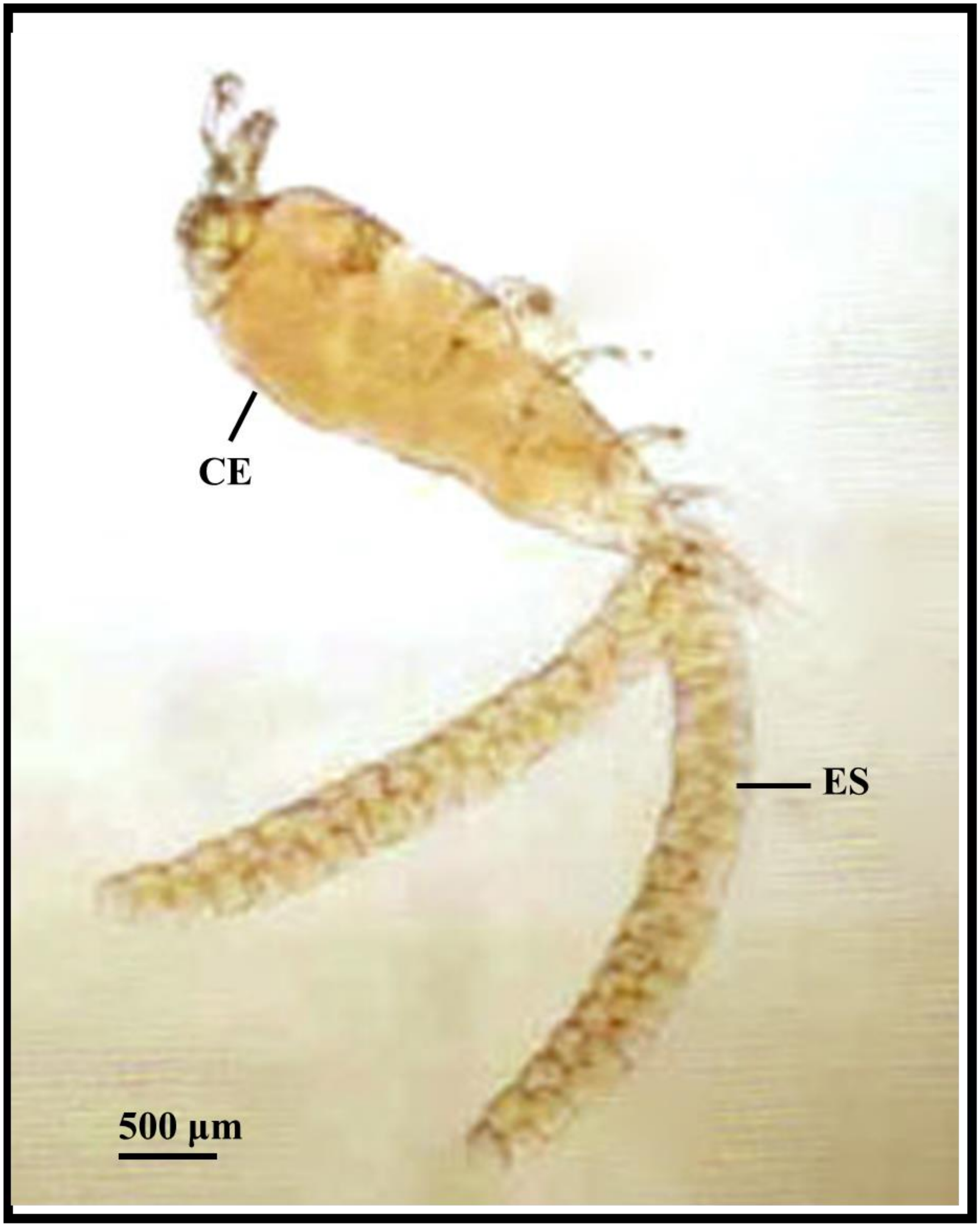


Figure 31C: Photomicrograph of the female copepodian parasite**,** *Ergasilus celestis* **Roberts, 1970** with lactic acid.

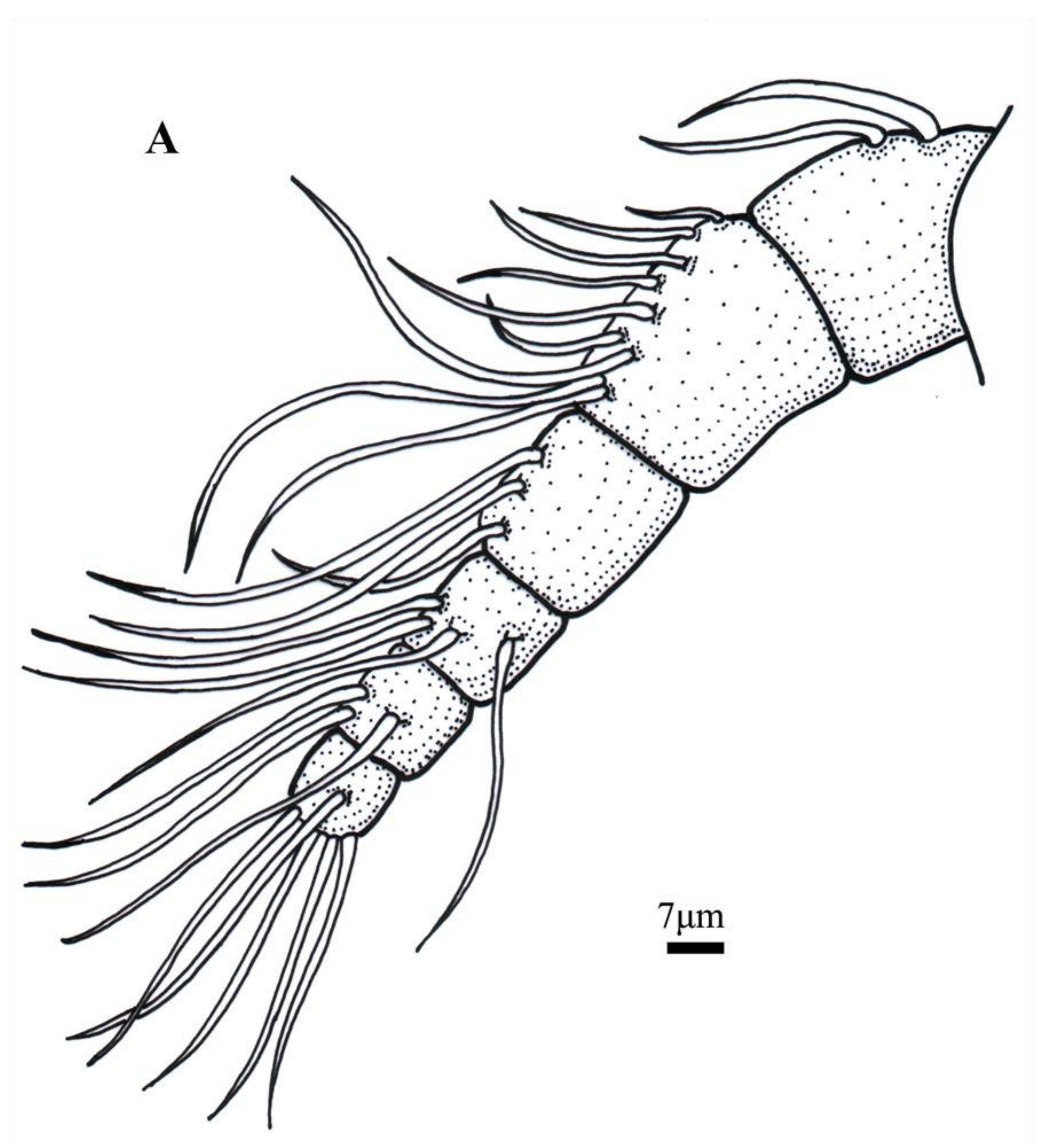


Figure 32A: Schematic drawing of the antennule, (A) of the female copepodian parasite**,** *Ergasilus celestis* **Roberts, 1970.**

segments diminishes distally towards the terminal end except the second segment which is slightly the biggest. All antennualry segments are provided with numerous simple setae, principally on antero-ventral surface. Armature formula is 2-9-3-4-3-5.

A pair of stout prehensile, subchelate and segmented antennae (Fig. 32B) is situated on the slightly protruding rostral area. They are long and slender. Each antenna measures 355 (330-359) µm in length and 100 (90-120) µm in a maximum width and consists of four segments. The first or basal segment is the coxobasis. It is short, slightly cubical in shape. The second segment is the first endopodal segment which is the longest one. It is widest at its proximal end and narrows towards its distal end. It bears a small tooth-like spine at its inner margin, almost at the medial region. The third antennary segment is the second endopodal segment and is called the subchela. It is narrow and arched with the sides almost parallel and clearly bent inwards. The fourth or distal antennary segment is the fourth endopodal segment. It is in the form of a stout pointed, clasper-like smooth claw lacking teeth. It is the shortest segment.

Five pairs of thoracic swimming legs originate from the lateral side of the first five thoracic somites. The first pair of thoracic swimming legs originates ventro-laterally from the posterior part of the cephalothorax and the last pair projects from the fifth thoracic somite. The first four pairs are biramous and exist in a completely form. Each leg consists of a proximal coxapod and a distal basipod which bears the two free rami; an exopod and an endopod. Whereas the fifth thoracic pair is greatly reduced.

The first pair of thoracic legs (Fig. 32C) consists of a coxapod and basipod. The basipod is ornamented also with a single antero-lateral sensilum. Each of exopod and endopod consists of three segments that diminish distally in size.

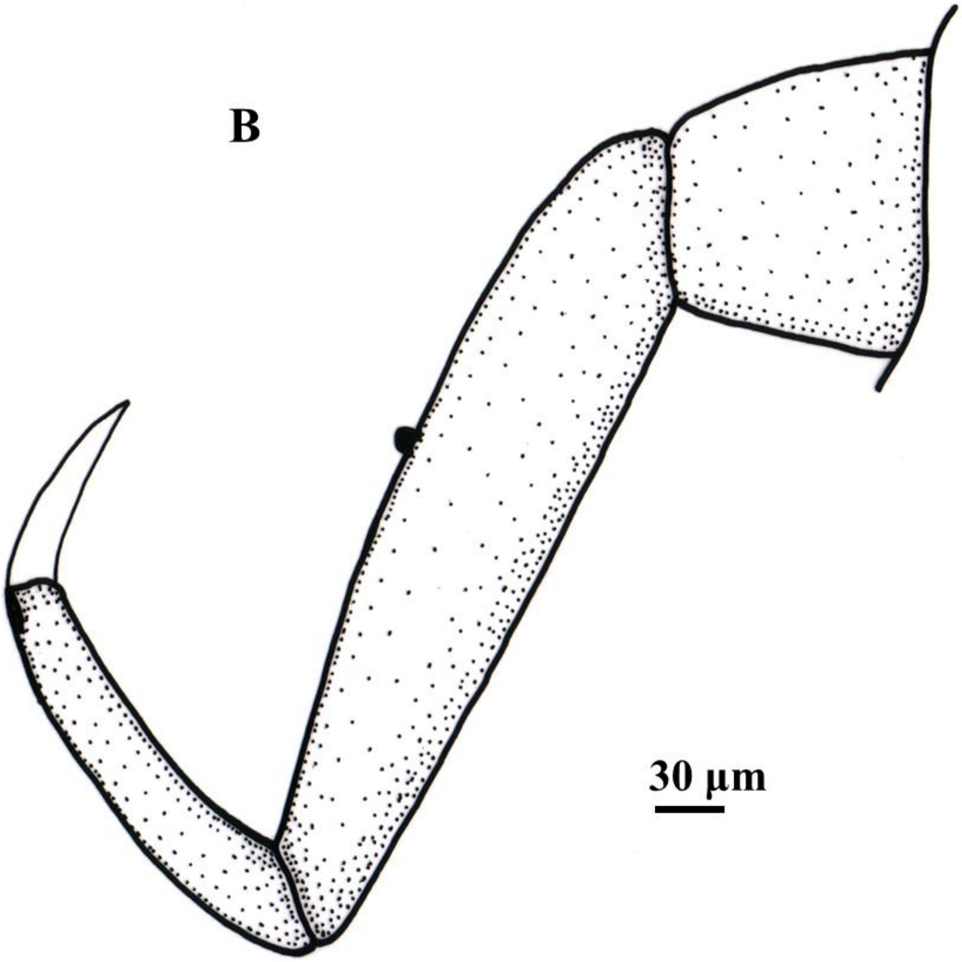


Figure 32B: Schematic drawing of the antenna, (B) of the female copepodian parasite**,** *Ergasilus celestis* **Roberts, 1970.**

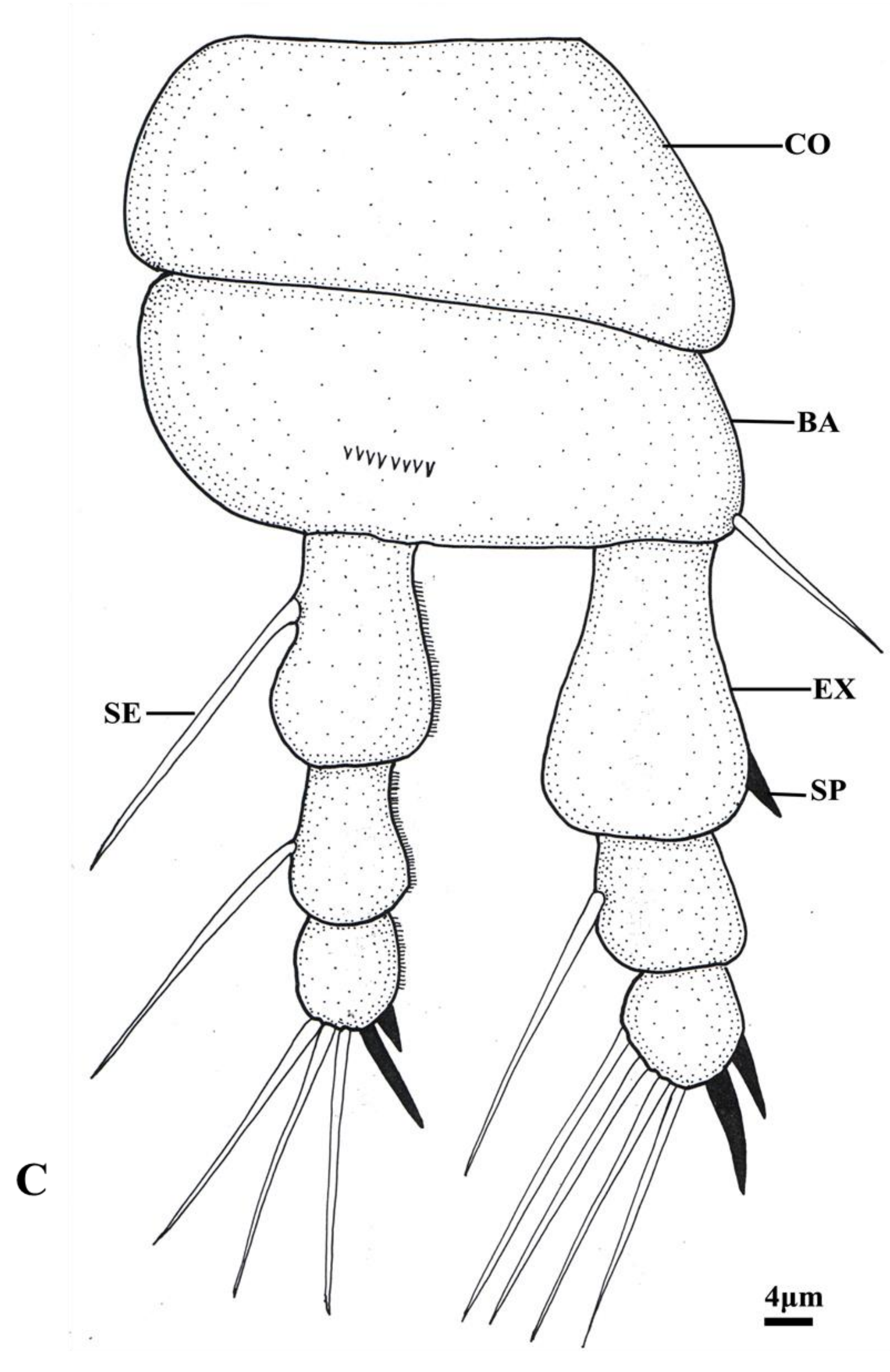


Figure 32C: Schematic drawing of the 1st thoracic leg, (C) of the female copepodian parasite**,** *Ergasilus celestis* **Roberts, 1970.**

The first exopodal segmental is provided with a single postero-lateral rasp-like spine projecting from the outer margin. The second exopodal segment is provided with a single medially situated naked seta. It is provided with two laterally situated serrated rasp-like spines projecting from the outer margin. Also, it is ornamented posteriorly with four naked setae. The endopodal segments are serrate where they are provided with one row of spinules arranged on the outer margins. The first endopodal segment is serrate on the distal margin and provided with a single medially situated unarmed seta projecting from the inner segmental margin. The second endopodal segment is provided with a single postero-lateral naked seta projects from the inner segmental margin. The terminal endopodal segment is provided with two postero-lateral serrated rasp-like spine arranged on its outer margin in addition to three naked setae on both distal and inner segmental margins.

The second and third thoracic legs (Fig. 32D) are closely similar. The basipod of each leg are ornamented with rows of spinules. The exopod and endopod of both legs consists of three segments, diminishing distally in size. The first exopodal segment is provided with a single postero-lateral rasp-like spine originates from the outer margin. The second exopodal segment is provided with a single medially situated unarmed seta located on the inner segmental margin. The terminal exopodal segment is provided with six posteriorly directed unarmed setae. The endopodal segments are serrated where their outer margins are provided with a single row of spinules. The first endopodal segment is serrated also at distal margin and provided with a single medially situated unarmed seta located on the inner margin of the first endopodal segment. The second endopodal segment is provided with a single postero-lateral unarmed seta projecting from the inner margin. The terminal endopodal segment is provided with a single

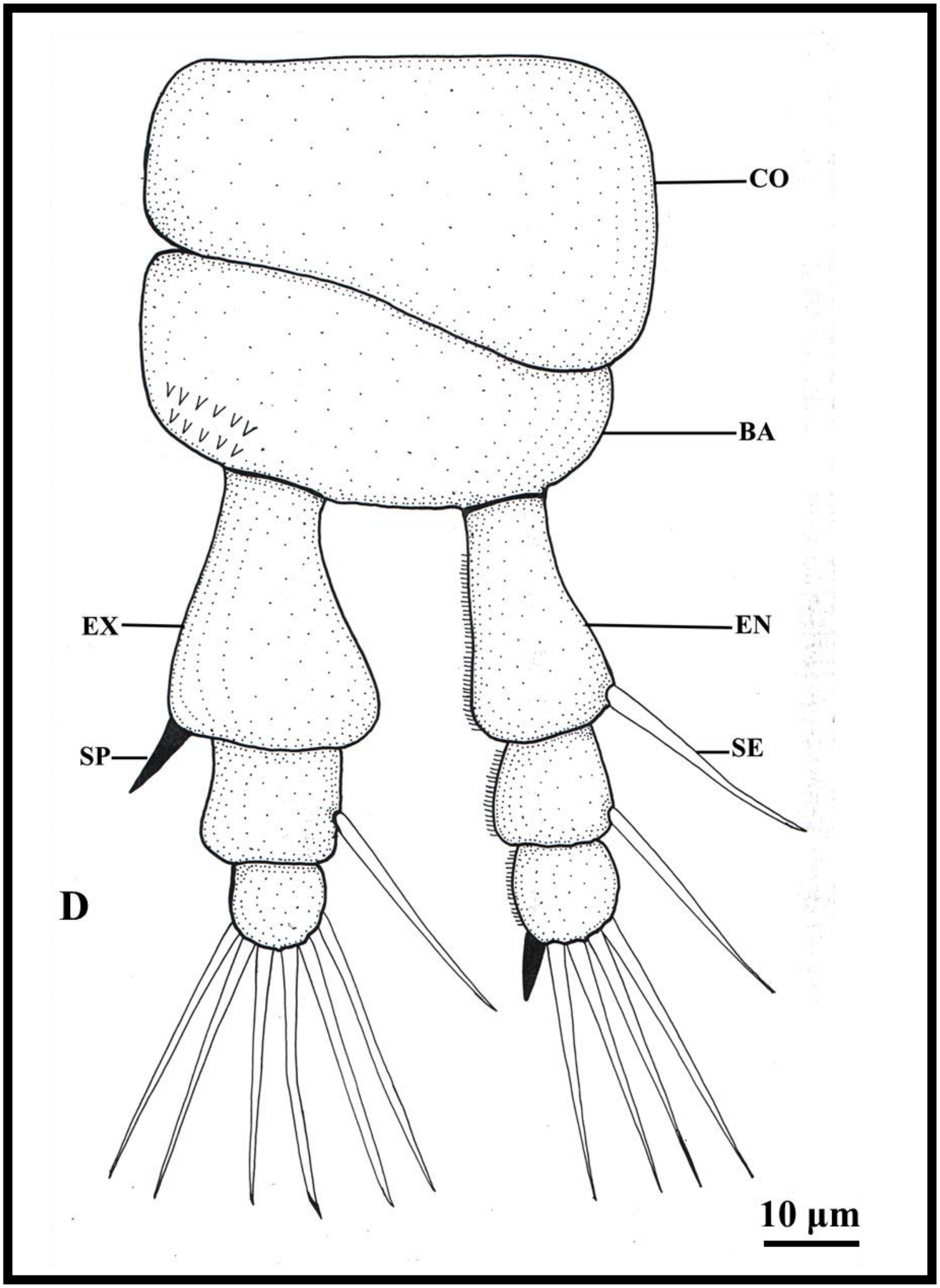


Figure 32D: Schematic drawing of the 2nd & 3rd thoracic leg, (D) of the female copepodian parasite, *Ergasilus celestis* **Roberts, 1970.**

posteriroly directed rasp-like projects from the distal segmental margin. Furthermore, it is provided with four posteriroly directed unarmed setae arranged on both distal and inner segmental margins.

The fourth thoracic leg (Fig. 32E) consists of a basipod which ornamented on its anterior surface with a double row of spinules. Also the basipod is provided with two antero-lateral sensilia projecting from the outer basipodal margin. The exopod consists of two segments while the endopod consists of three segments. Both exopodal and endopodal segments distally diminish in size. The first exopodal segment is provided with a single postero-lateral rasp-like serrate spine which exists on the outer segmental margin. The terminal exopodal segment is provided with five posteriorly directed unarmed setae arranged on its distal margin. The first endopodal segment is ornamented laterally and inwardly with medially situated unarmed seta. The second endopodal segment is provided with a single postero-lateral unarmed seta which exists on the inner segmental margin. The terminal endopodal segment is provided laterally with a single serrate, rasp-like spine located on the outer segmental margin besides three posteriorly directed unarmed setae arranged on the distal segmental margin. Armature of rami is shown in tables (12).

The urosome (Fig. 32F) consists of the two last thoracic somites (fifth and sixth), the abdominal somites and the caudal rami. The fifth thoracic somite is extremely reduced, very small and short but distinctly apart as much from the fourth thoracic segment as from the genital segment that follows it. It measures 155 (145-160) µm in length and 80 (75-90) µm in width. The sixth thoracic somite is the genital somite.

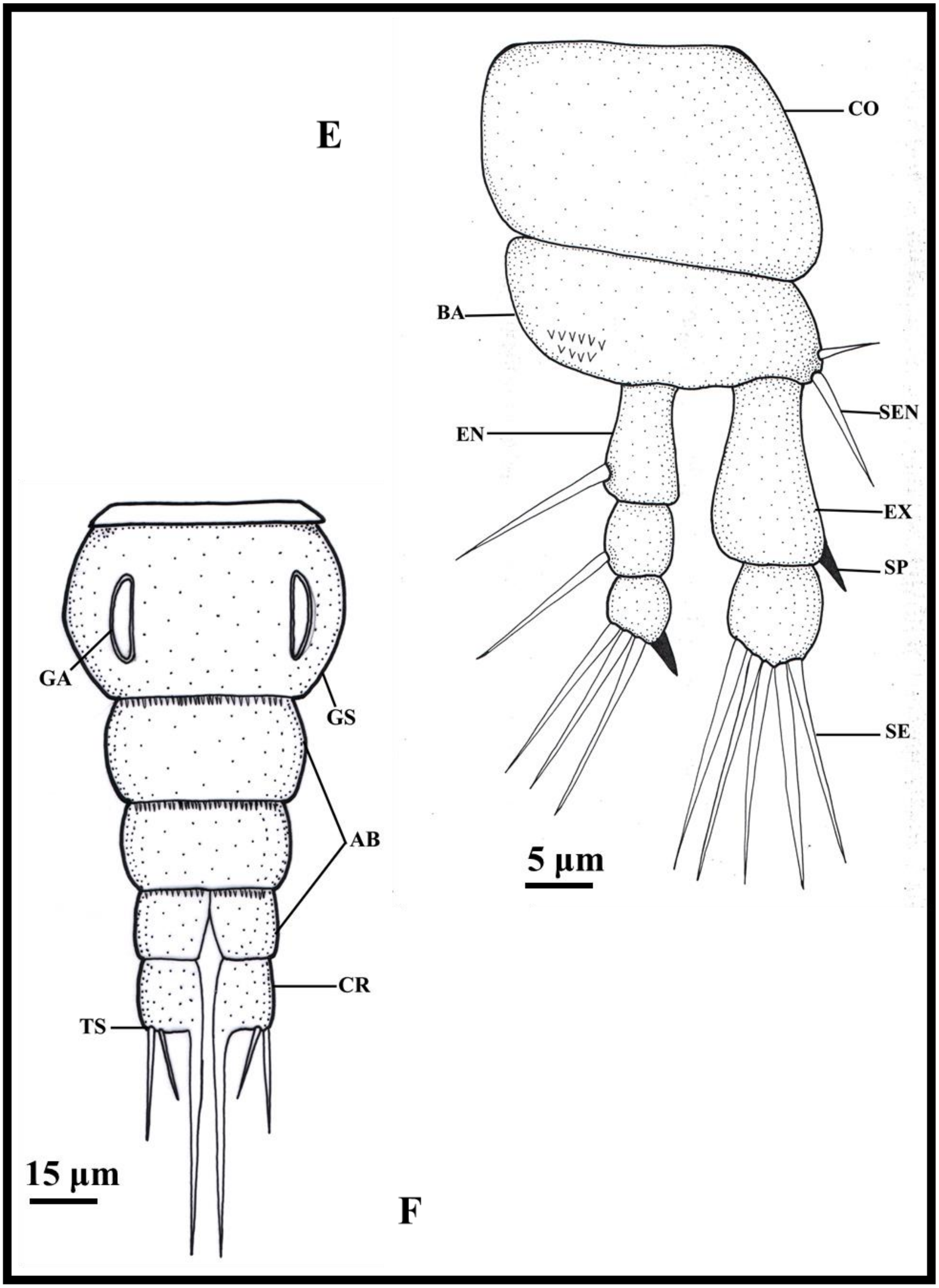


Figure 32E&F: Schematic drawing of the 4th thoracic leg,, (E) and urosome, (F) of the female copepodian parasite**,** *Ergasilus celestis* **Roberts, 1970.**

Abbreviations of the ergasilides parasite species

AB	Abdomen
ANT	Antenna (2nd antenna)
AN	Antennule (1st antenna)
BA	Basipod
CR	Caudal ramus
CE	Cephalosome
CET	Cephalothorax
CO	Coxapod
ES	Egg sac
EN	Endopod
EX	Exopod
E	Eye spot
GA	Genital aperture
GS	Genital segment
ME	Mesosome
PR	Prosome
RO	Rostrum
SEN	Sensilum
SE	Seta
SP	Spine
TS	Terminal seta
TH	Thorax
UR	Urosome
TSL	Thoracic swimming leg
FL	Fifth leg

Table (11): Morphometeric measurements of the copepodian parasite, *Ergasilus celestis* **Roberts, 1970.**

Items	Maximum length	Maximum width
Body	1025	405
Cephalothorax	550	340
Urosome	155	80
First antenna	105	27.5
Second antenna	355	100
First thoracic leg (L1)	60	-
First thoracic leg (L2)	65	-
First thoracic leg (W)	-	70
Second&third thoracic leg (L1)	50	-
Second&third thoracic leg (L2)	65	-
Second&third thoracic leg (W)	-	70
Fourth thoracic leg (L1)	50	-
Fourth thoracic leg (L2)	40	-
Fourth thoracic leg (W)	-	45
Fifth thoracic leg	-	-
Egg sac	1050	145

Thoracic legs	Coxapod	Basipod	Exopod			Endopod		
			1	2	3	1	2	3
First leg	0-0	I-0	I-0	0-1	II-4	0-1	0-1	II-3
Second leg	0-0	0-0	I-0	0-1	0-6	0-1	0-1	I-4
Third leg	0-0	0-0	I-0	0-1	0-6	0-1	0-1	I-4
Fourth leg	0-0	II-0	I-0	0-5	-	0-1	0-1	I-3

Roman numerals = Spines Arabic numerals = Setae

Table (12): The armature fomula for spines (Roman numerals) and setae (Arabic numerals) on the thoracic swimming legs of the adult female *Ergasilus celestis* **Roberts, 1970.**

(2) Discussion:

The anatomical structure and dimensions demonstrated in the present description are evidently to suggest that the present species described here in belong to genus *Ergasilus* **von Nordmann**, **1832** according to the following generic criteria which based by **Markewitsch (1956), Gusev (1962), Yamaguti (1963), Hoffman (1967, 1977), Kabata (1988), Mitchum (1995)** and **Hoffman and Williams Jr. (1999**). These generic morphological criteria are: body is cyclops-like, expanded anteriorly, tapering to posterior end. Head fused with, sometimes separated from, first thoracic segment and more or less fused with second thoracic segment forming cephalothorax which may be highly inflated dorsally, line of fusion usually visible, either throughout length or only at sides of body. An eye spot may be evident on the cephalothorax dorsally, medially and toward the anterior end. Fourth free thoracic segments (fifth thoracic segment) are small, occasionally completely imperceptible. Sixth thoracic segment is bearing oviductal openings. Abdomen is three segmented in female. Caudal rami short, well developed, each provided with four setae. Egg sac long, often cigar-shaped or rather pump, eggs small and numerous. First antenna (antennule), five or six segmented, setiferous, second antenna subchelate, prehensile, four or five segmented, the terminal segment in the form of a prominent, stout, calsper-like claw. First pair of swimming legs born on cephalothorax; first four pairs of swimming legs are biramous. Each ramus consists of three segments, except for exopodite of the fourth pair which contains two or occasionally one segment. Fifth pair uniramus, one or two segmented or reduced to a papilla and one, or two setae, rarely obsolete. Female is parasitic on gills and (rarely) in the nasal fossa of freshwater and marine teleosts, male free swimming.

Gurney (1913), Karamchandani (1952), Markewitch (1956), Roberts (1963), Yamaguti (1963), Rogers (1969), Lucky (1977), Kabata (1979), Ben

Hassine (1983), Thatcher and Boeger (1984), Kabata (1985), Byrnes (1986), Oldewage and Van As (1988a,b), Venkateshappa *et al*. (1988), Ho *et al*. (1992), Kabata (1992), Abdelhalim *et al*. (1993), Alston *et al*. (1993) and Alston *et al*. (1996) differentiated between the species of the genus *Ergasilus* **von Nordmann, 1932** on the basis of shape of cephalothorax, presence or absence of dorsal sculpture, ratio of antenna length to length of the cephalothoax, ratio of length of the cephalothorax to the total body length, size ratio of length to width of the cephalothorax, head either fused with or separated from first thoracic segment and more or less fused with second thoracic segment and the way of fusion either partial or complete fusion, number of segments of the antennule and their armature formula, size of the antenna, distal ends joined or unjoined, ratio of length of the last antennary segment to the length of the claw, number and location of spines and sensilla occurring on the segments of the antenna, size ratio of genital segment to fifth thoracic segment, size ratio between abdominal segments, bifurcation of the abdominal segments, ratio of length to width of caudal rami, setation of caudal rami, presene or absene of setae on the dorsal and ventral surfaces of the body segments, setation on the ventral surface of the abdominal segments, ratio of egg sas (ovisacs) to total body length, size of eggs, armature of swimming legs and structure of the fifth swimming legs.

According to the previous differential criteria, the first described specimens in the present study are closely related to *Ergasilus briani* **Markewitsch, 1933,** the second described specimens in the present study are closely related to *Ergasilus lizae* **Kroyer, 1863,** the third described specimens in the present study are closely related to *Ergasilus australiensis* **Roubal, 1981,** the fourth described specimens in the present study are closely related to *Ergasilus mosulensis* **Rahemo, 1982** and the fifth described specimens in the present study are closely related to *Ergasilus celestis* **Roberts, 1970**.

The present individuals of the first parasitic copepod *Ergasilus briani* **Markewitsch, 1933** infesting gills of the flat-head grey mullet; *Mugil cephalus* was recorded for the first time in Egyptian Mediterranean coast.

Ergasilus briani **Markewitsch, 1933** was previously collected from the gills of *Osmerus eperlanus, Abramis brama* and *Rutilus rutilus,* from Russia, Siberia, Poland, Germany, Czechoslovakia and China. **Markewitsch (1956)** described it from the gills of *R. rutilus, Leuciscus idus, Crusian carp, Tinca tinca* and *Carassius carassius* in Poland, Finland and Germany. **Freyer and Andrews (1983)** collected it from the bream, *A. brama* in Britain. **Alston *et al.* (1996)** collected it from the gills of the tench, *T. tinca* in Britain. **Bagge and Valtonen (1996)** collected it from the gills of the roach, *R. rutilus* in Finland. There are close similarities between the present specimens and the specimens redescribed by **Markewitsch (1956)** and **Alston *et al.* (1996)** in all morphological criteria, specially body dimensions, shape and armature of antennule and antenna, shape and dimensions of cephalothorax, armature of swimming legs and caudal rami. However, they differed in length of antenna where they are slightly longer in the present study as in Table (3).

The present individuals of the second parasitic copepod *Ergasilus Lizae* **Kroyer, 1863** infesting *Valamugil seheli* was recorded for the first time in Egyptian Mediterranean coast.

Ergasilus Lizae **Kroyer, 1863** was firstly recorded and described by **Kroyer (1863)** from the gills of *Mugil lizae* which was collected near New Orleans. Numerous workers such as **Wilson (1911), Kelly and Allison (1962), Roberts (1969, 1970), Johnson and Rogers (1973), Raibout and Ben Hassine (1977), Kabata (1979) and Kabata (1992)** recorded this species on mugilid fish hosts from many parts of the world**.** In this respect **Wilson (1911),** recorded it from the gills of *Mugil lizae*, *Mugil cephalus*, *Floridichthys carpia*, *Fundulus* *similis* and *F. heterolitus grandis* from Atlantic ocean near Louisiana in USA; **Bere (1936)** recorded it from the gills of *Mugil cephalus* from the gulf of Mexico; **Pearse (1952)** recorded it from the gills of *Mugil cephalus* from the Texas coast; **Kelly and Allison (1962)** recorded it from the gills of *Lepomis macrochirus*, *L. microlophus* and *Microepterus salmoides* from freshwater in England; **Raibaut and Ben Hassine (1977)** collected it from the gills of *Mugil cephalus, L. ramada* and *L. saliens* from the Mediterranean Sea; **Paperna (1977)** collected it from the gills of *M. cephalus* in the Mediterranean Coast of Israel; **Aouij and Zasoulai (1994)** collected the same species from the gills of *M. cephalus* from Tunisia; **Knoff *et al*. (1994)** collected it from the gills of *M. platanus* in the Brazilian coast and **Hoffman and Williams (1999)** collected it from the gills of *Fundulus heteroclitus* from USA.

In the present study, a careful comparison of the specimens collected with those described by previous authors revealed that, they are related to the specimens redescribed by **Kabata (1992)** in the body dimensions, armature and measurements of antennule and antenna, shape of cephalothorax, length of free thoracic segments, width of abdominal segments, caudal rami setation, armature of swimming legs (Table 5). Meanwhile, the present specimens differ from those described by **Kabata (1992)** in width of free thoracic segments and length of genital segment.

The present individuals of the third parasitic copepod *Ergasilus australiensis* **Roubal, 1981** infesting the common two banded sea bream; *Diplodus sargus* was recorded for the first time in Egyptian Mediterranean coast.

Ergasilus australiensis **Roubal, 1981** was originally described by **Roubal (1981)** from the gills of *Acanthopagrus australis* in northern New South Wales. It was subsequently found by **Byrnes (1986)** on the same host, as well as on *Acanthopagrus butcheri* and *Acanthopagrus berda* in numerous Australian localities. *Ergasilus australiensis* has been recorded from North Queensland and New South Wales on *Toxotes chatareus* and *Acanthopagrus australis* **Kabata (1992)**. There are close similarities between the present specimens and the specimens redescribed by **Kabata (1992)** in all morphological criteria, specially body dimensions, shape and armature of antennule and antenna, shape of cephalothorax, armature of swimming legs and caudal rami. However, they differed in length of antenna where they are slightly shorter in the present study and wider than it. They also differed in thoracic legs dimensions that being smaller in the present study as in Table (7).

The present individuals of the fourth parasitic copepod *Ergasilus mosulensis* **Rahemo, 1982** infesting the thin grey mullet, *Liza ramada* was recorded for the first time in Egyptian Mediterranean coast.

Ergasilus mosulensi was firstly collected from the gills of some freshwater mullets, *Liza abu* and *Mugil lizae* in Iraq by **Rahemo (1982)**. It has been recorded from the gills of *L. abu* in Iraq by **Mhaisen *et al*. (1988)** and **Ho *et al*. (1996)** collected and described this species from the gills of *L. abu* in Iraq.

The present material of *Ergasilus mosulensis* **Rahemo, 1982** resembles that redescribed by **Ho *et al.* (1996)** in all morphological criteria, specially antennule and antenna, cephalothorax, free thoracic segments, abdominal segments, egg sac and eggs. However, they differed in length of antenna where they are slightly longer in the present study and wider than it. They also differed in cephalothorax dimensions that being shorter in length and wider in width in the present study as in Table (9).

The present individuals of the last parasitic copepod *Ergasilus celestis* **Roberts, 1970** infesting the eel fish; *Anguilla anguilla* was recorded for the first time in Egyptian Mediterranean coast.

Roberts (1970) was firstly collected *Ergasilus celestis* from the gills of *Anguilla rostrata* from North America. It was subsequently found by **Rogers and Hawke (1978)** on the burbot *Lota lota* in Ontario, Canada. In Atlantic Canada, *Ergasilus celestis* has been recorded on gills of *Anguilla rostrata* **Kabata (1988)**. It also has been identified among wild populations of eels, *A. rostrata*, in Atlantic Canada **Kabata (1992)**.

There are close similarities between the present specimens and the specimens described by **Roberts (1970)** in all morphological criteria, specially shape and armature of antennule and antenna, shape of cephalothorax, armature of swimming legs and caudal rami. Despite slight differences between the present specimens and the closely related one in the body dimensions where the present specimens are longer in which total length of the specimens described by **Roberts (1970)** was 0.3 mm.

CHAPTER I

Section VIII

Caligus elongatus

Genus: *Caligus* Müller, 1785

Caligus elongatus von Nordmann, 1832

(1) Redescription:

The body of the copepodian parasite *Caligus elongatus* is illustrated in figures 33 & 34 and its measurements are shown in table (13).

The following redescription is based on the study of twenty seven living adult and eleven mounted specimens of male *Caligus elongatus*, collected from the gill filaments of the sea bass, *Dicentrarchus labrax.*

The body (Fig. 33) is elongated, small, with slimmer posterior body region. It measures in male 2880 (2580-3180) μm in length excluding caudal setae and 1605 (1410-1900) μm in width.

Cephalothoracic shield (Fig. 33) subdivided into plates. Shield with marginal membrane, small sensory setules, and single sensory crypt on each of lateral sides.

First antenna (Fig. 34A) is armed with larger setae. Proximal segment is large and strong, sclerotized at junction with cephalothorax, bearing 29 pinnate setae. Distal segment is armed with 14 setae. It measures 715 (650-780) μm in length and 180 (160-210) μm in width.

Second antenna (Fig. 34B) with basal segment more robust, more strongly sclerotized and enlarged posterolaterally, and equipped with two corrugated pads on ventral surface. One pad large and elongate, other minute and circular. Basal segments are extending to base of postantennary process. Corpus is strong,

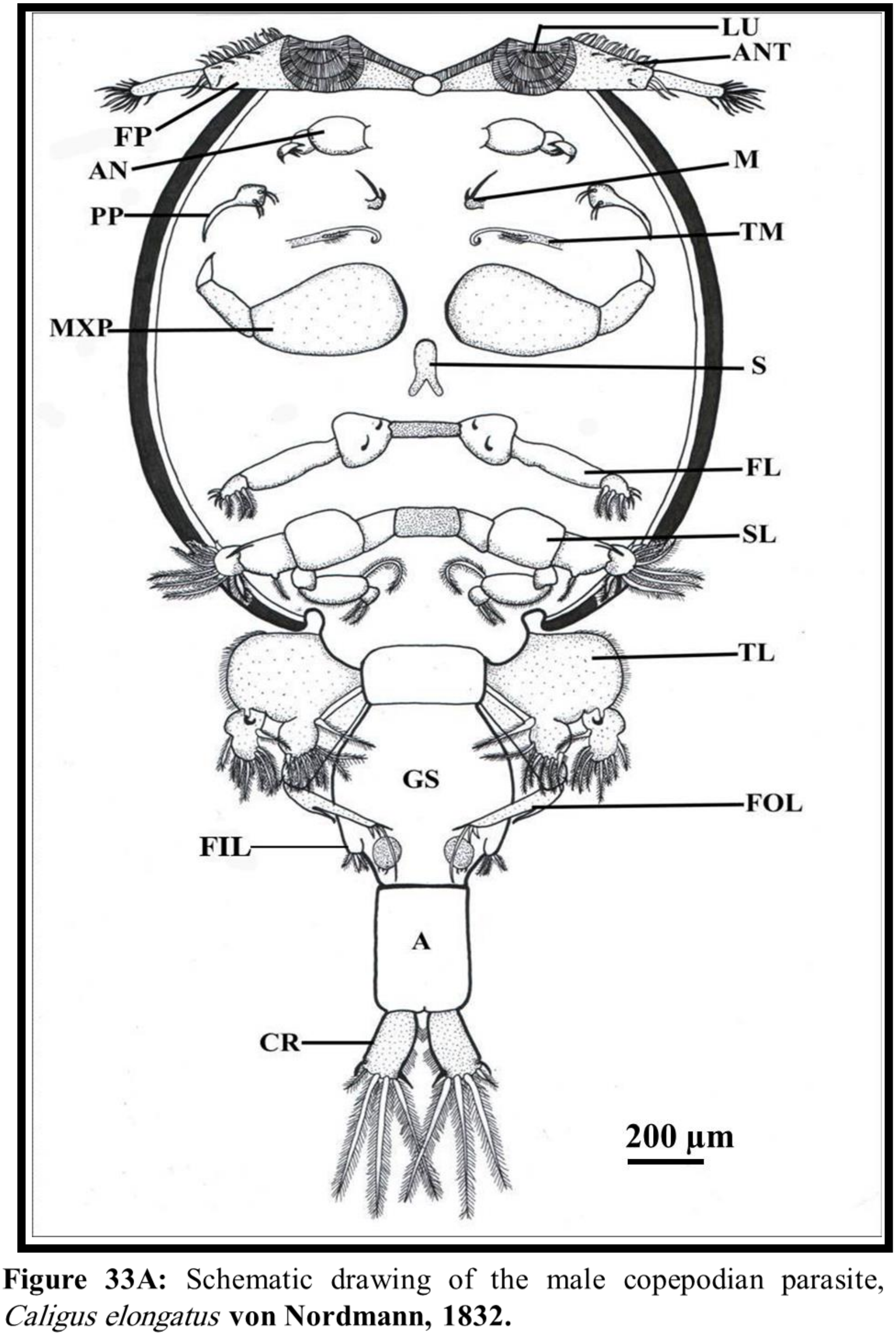


Figure 33A: Schematic drawing of the male copepodian parasite, *Caligus elongatus* **von Nordmann, 1832.**

Figure 33B: Photomicrograph of the male copepodian parasite, *Caligus elongatus* **von Nordmann, 1832** stained with alum carmine.

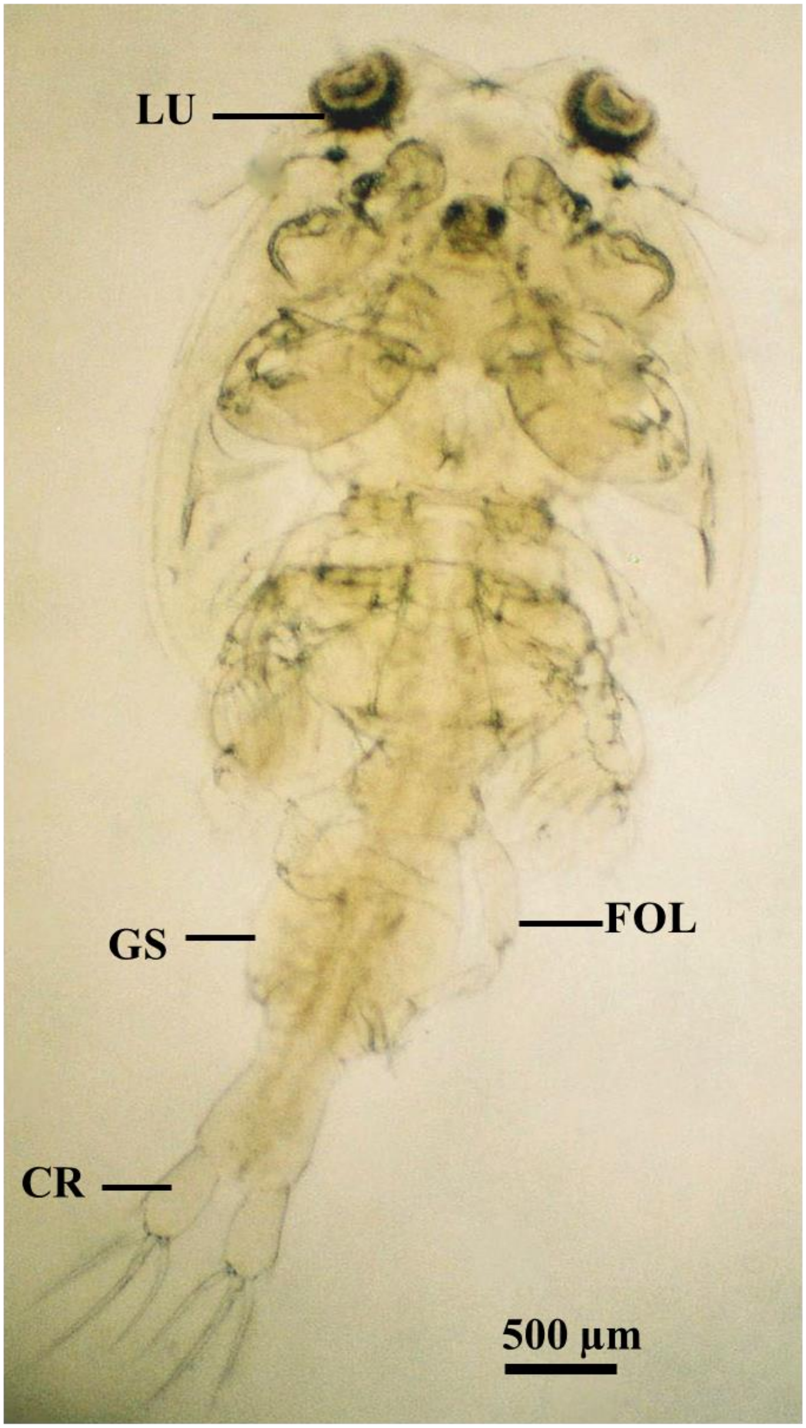


Figure 33C: Photomicrograph of the male copepodian parasite, *Caligus elongatus* **von Nordmann, 1832** with lactic acid.

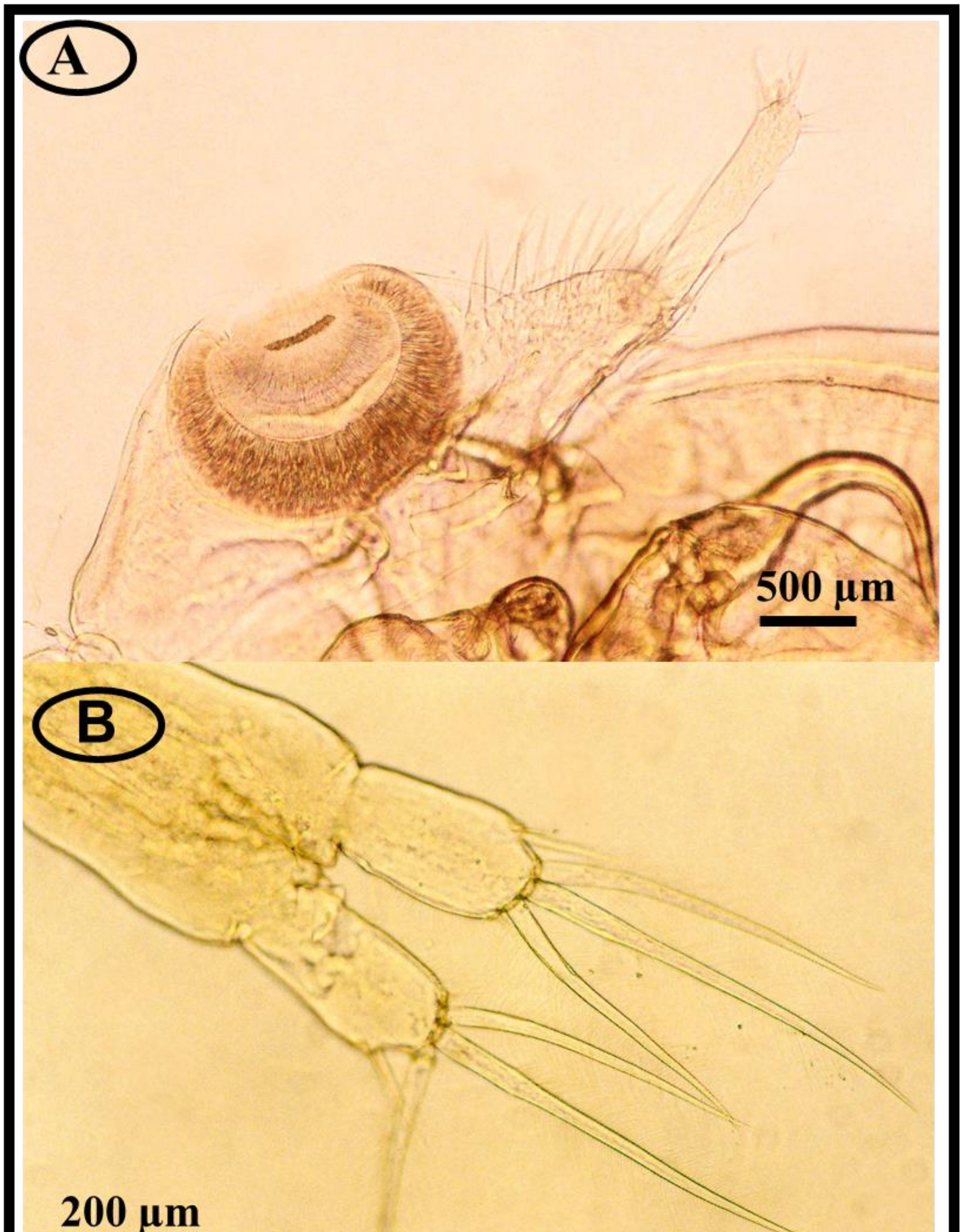


Figure 33D: Photomicrograph of the male copepodian parasite, *Caligus elongatus* **von Nordmann, 1832** showing the lunule and antennule, (A) and uropod, (B) with lactic acid.

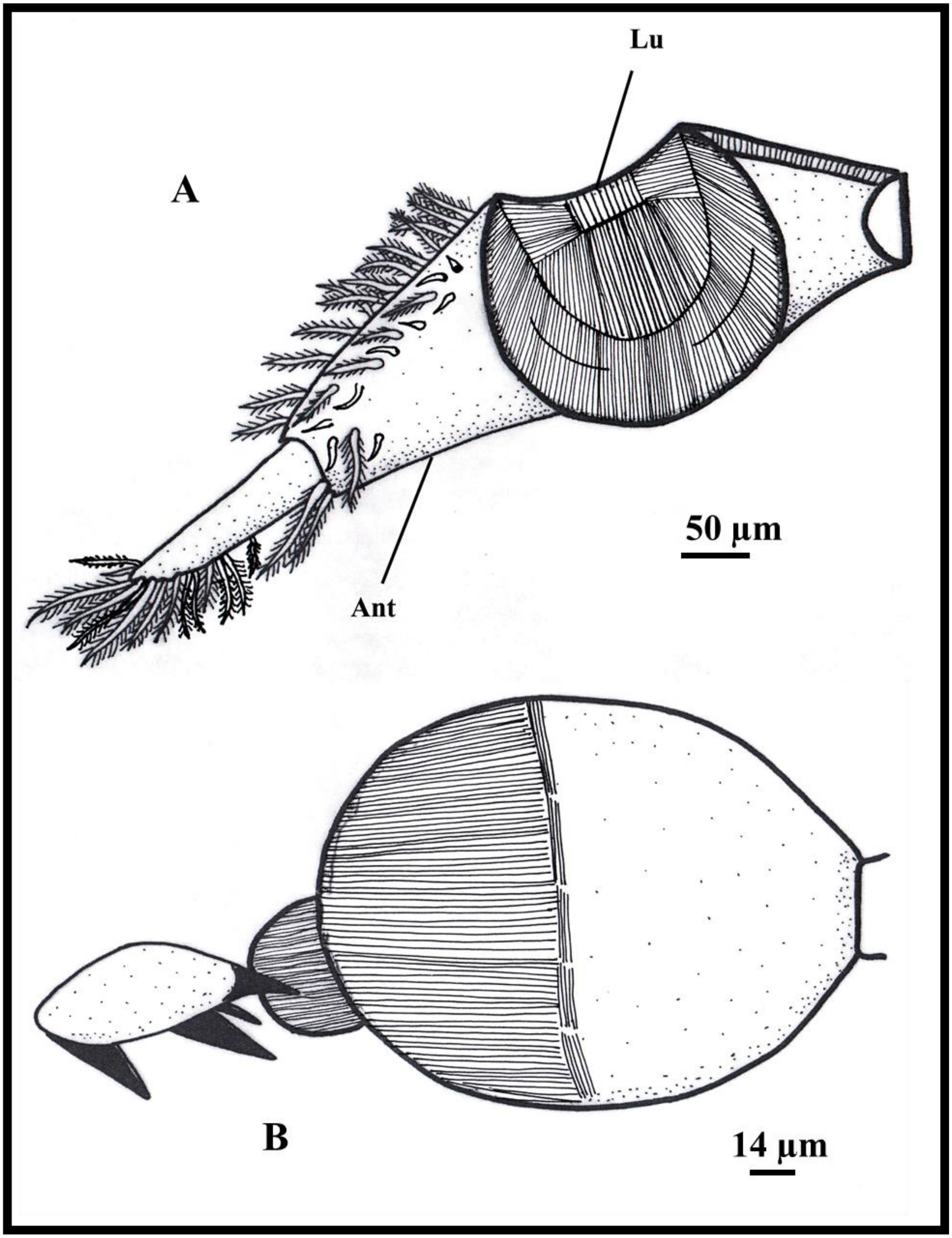


Figure 34A&B: Schematic drawing of lunule and antennule, (A) and antenna, (B) of the male copepodian parasite, *Caligus elongatus* **von Nordmann, 1832.**

sub-cylindrical, and elongate. Subchela is small, stocky, and occupies tip of corpus. Subchela is aligned perpendicularly to long axis of corpus. Myxal elevation is displaced distally. Most of ventral surface of corpus is occupied by large corrugated pad. Similar smaller pad is extending on dorsal side of corpus. Myxal elevation also is corrugated. Subchela is not clearly subdivided. Small single seta appears on each side of shaft. Small stocky claw is shielded on both sides by cuticular flaps. Claw and flaps is partly covered by elongated cuticular cap. It measures 210 (195-227) µm in length and 105 (85-130) µm in width.

First maxilla (Fig. 34C) is conical process smaller, and subdivided into base and claw. Basal segment with lateral corrugated pad. Small seta is arising laterally near claw base. It measures 32.5 (25-35) µm in length and 25 (22-30) µm in width.

Second maxilla (Fig. 34D) is strongly and elongate. Brachium almost is twice as long as lacertus and much thinner. Calamus is much longer than canna. Flabellum a well-defined process covers with setules. Subdivision is longer and visible on brachium. It measures 250 (225-280) µm in length and 27.5 (25-35) µm in width.

Maxilliped (Fig. 34E) is with broader corpus; Subchela exsists parallel to ventral surface. It measures 700 (585-810) µm in length and 405 (320-490) µm in width.

Sternal furca (Fig. 34F) is small and with slimmer processes and large lateral processes near base.

Postantennary process (Fig. 34G) consists of strong, heavily sclerotized claw directed posteriorly. Claw is equipped with two bifid sensory setules. It measures 205 (185-225) µm in length and 65 (55-80) µm in width.

First thoracic leg (Fig. 34H) with sclerotized subrectangular sympod with pinnate seta on ventromedial margin. Small pyriform endopod is armed with one

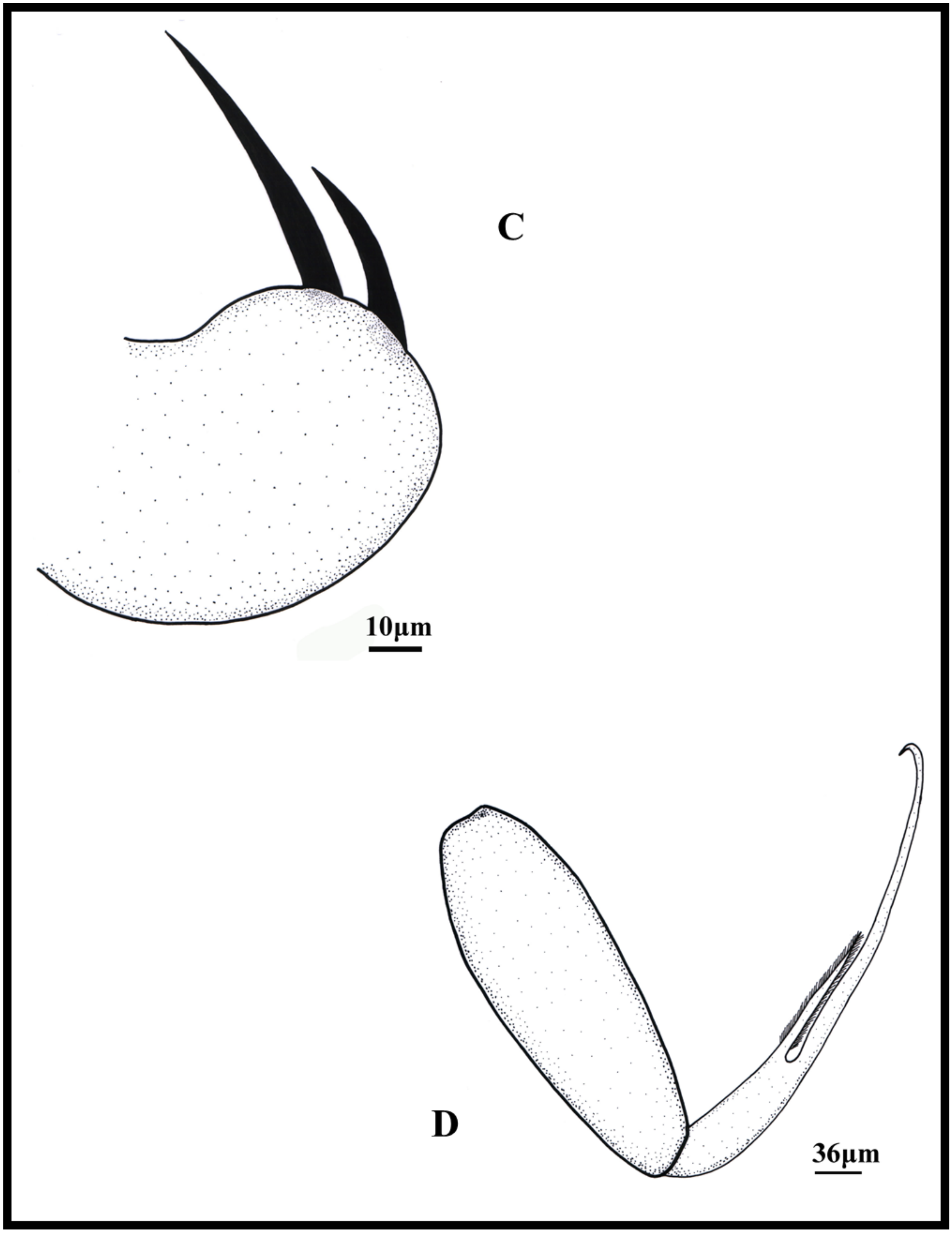


Figure 34C&D: Schematic drawing of maxillule, (C) and maxilla, (D) of the male copepodian parasite, *Caligus elongatus* **von Nordmann, 1832.**

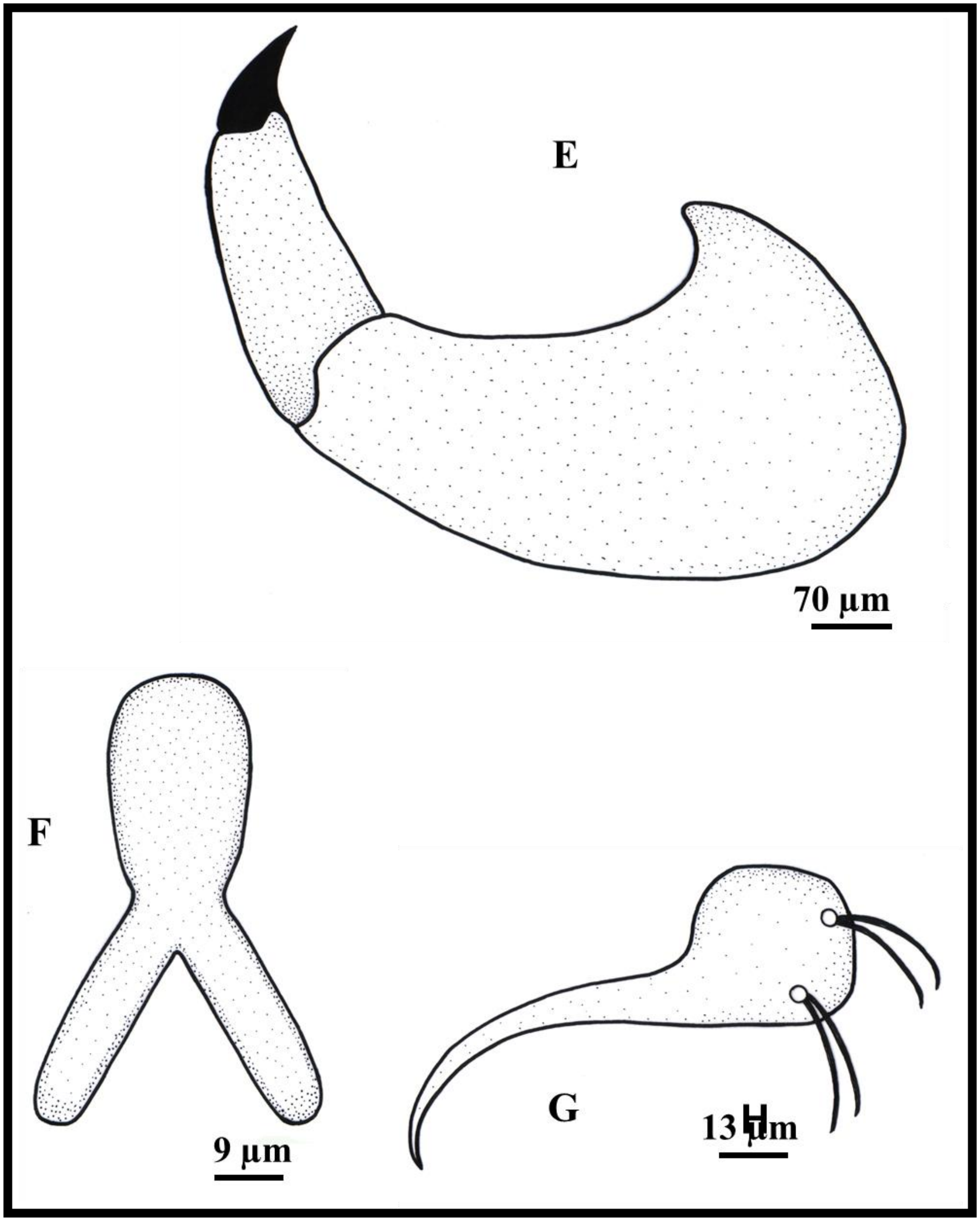
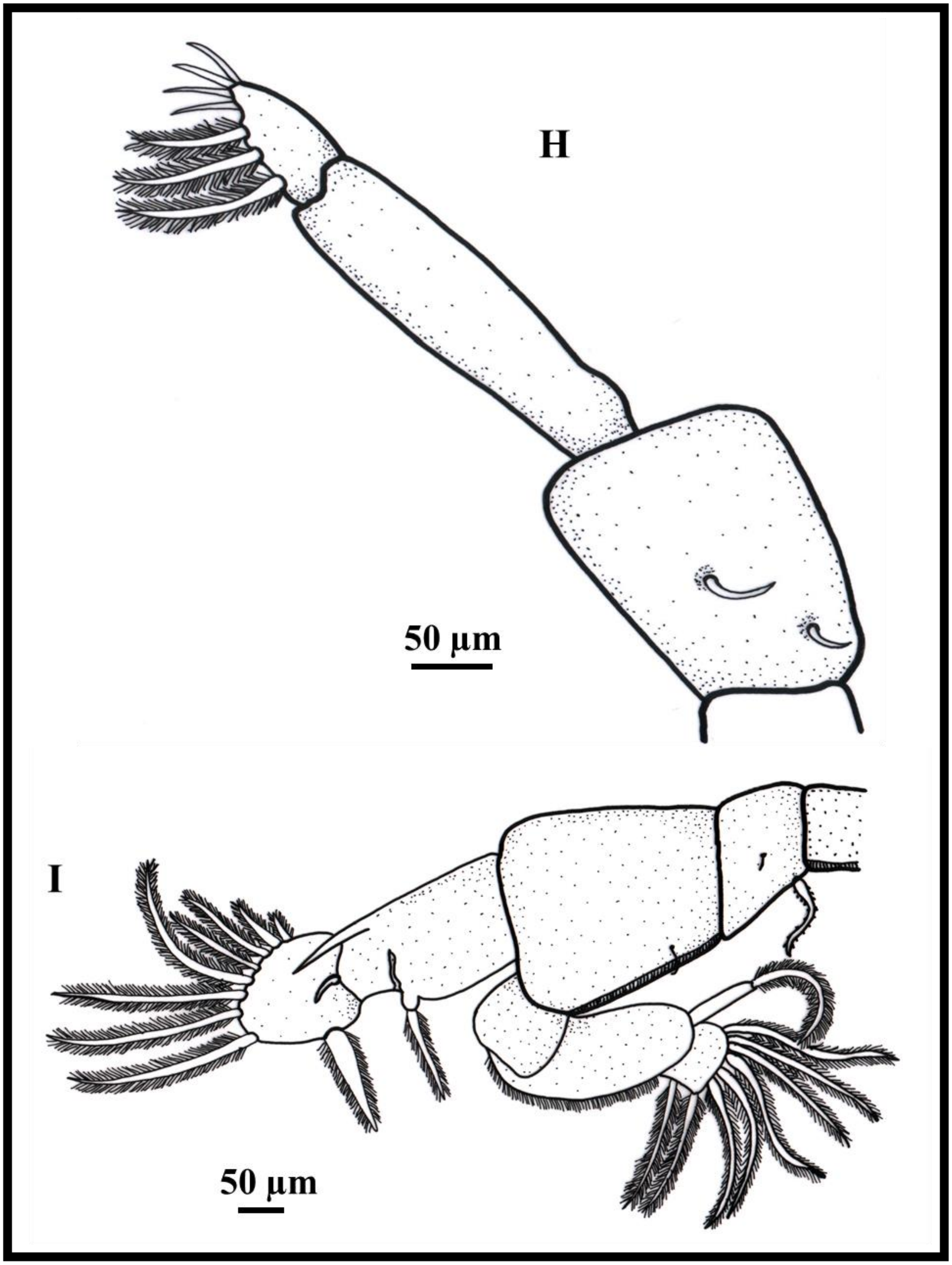


Figure 34E&F&G: Schematic drawing of maxilliped, (E) sternal furca, (F) and postantennal process, (G) of the male copepodian parasite, *Caligus elongatus* **von Nordmann, 1832.**



(Figure 34H&I): Schematic drawing of 1st leg, (H) and 2nd leg, (I) of the male copepodian parasite, *Caligus elongatus* **von Nordmann, 1832.**

small apical seta. Elongate two segmented exopod are linked to sympod with strongly sclerotized joint. Proximal segment of exopod is almost twice as long as sympod and 3 times longer than distal segment. Proximal segment is equipped with striated membrane along ventro-medial margin. Distal segment subrectangular, bears three setae with pinnae on medial margin. Distal margin is armed with three short and one long setae. Interpodal bar is narrow.

Second thoracic leg (Fig. 34I) with strong and wide interpodal bar bears striated membrane on the distal margin. Sympod with well-separated medial segment, possesses large pinnate seta and sensory setule. Rest of sympod is subrectangular with striated membrane along medial margin and single sensory setule. Lateral margin is armed with small pinnate seta. Each ramus is three-segmented and similar to others in size. Proximal segment of exopod as long as next two segments and combined with long sclerotized seta on lateral margin. Seta with two lateral membranes. Medial margin of proximal segment is armed with large pinnate seta. Second segment is similar to first one, with additional short striated membranes on medial margin. Distal segment with lateral margin is armed with one small conical seta and one seta with lateral membranes. Distomedial margin with six large pinnate setae and short striated membrane. Endopod segments are almost equal in size. The second segment is slightly larger than other two. Proximal one is armed with one large pinnate seta on medial margin and short striated membrane in distolateral corner. Second segment is equipped with two pinnate setae and striated membrane on medial margin. Lateral margin with striated membrane. Short sub triangular distal segment is armed with six large pinnate setae and short striated membrane.

Third leg (Fig. 34J) is fused with powerful interpodal bar, forming an " apron " (**Kabta 1979**). Sympod is armed with one small pinnate seta on lateral margin and large pinnate seta and two sensory setules on ventromedial margin.

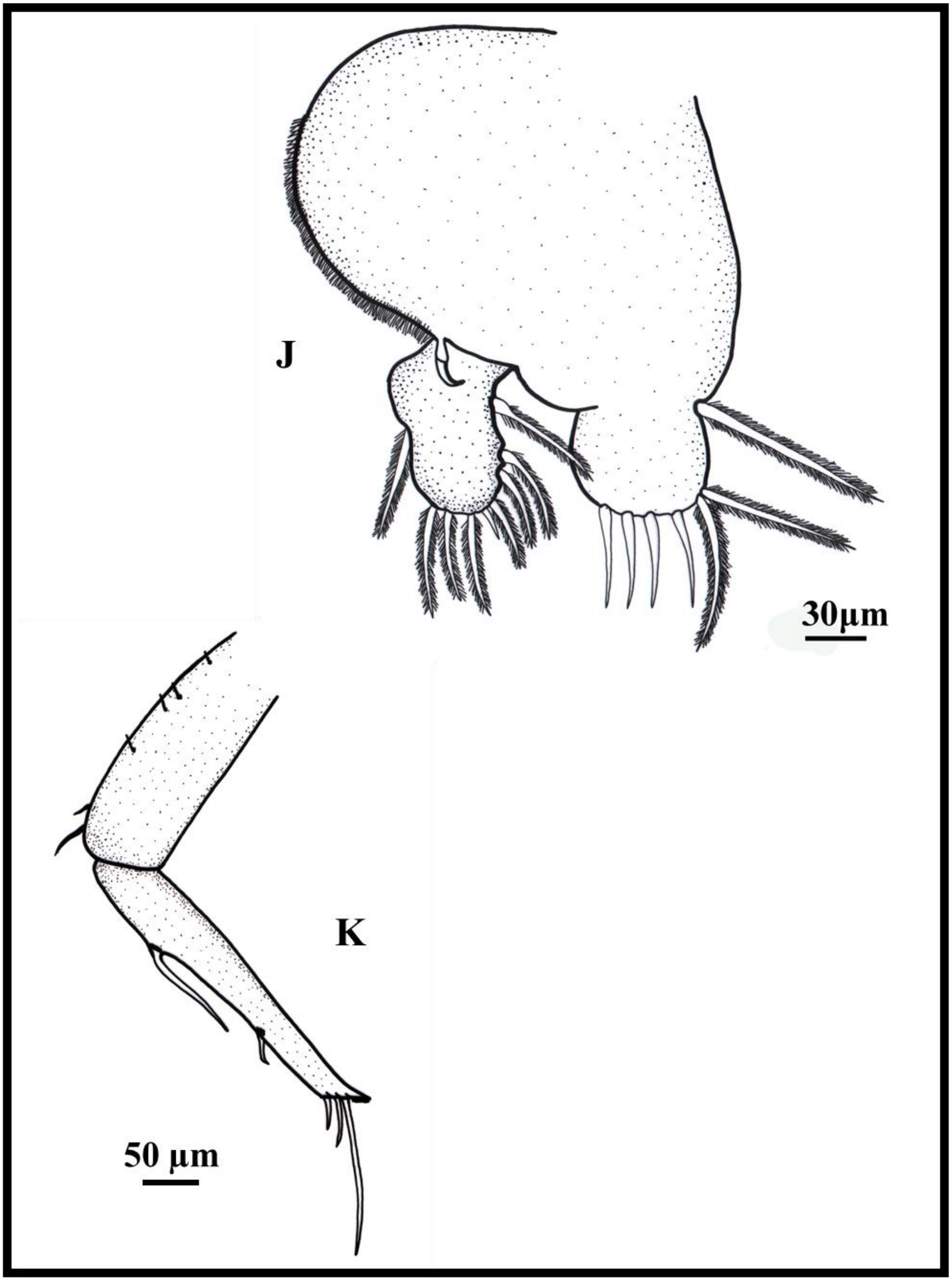
Both margins have striated membranes. Rami are widely separated and the space between them is sealed with elongated cuticular flap with striated margin. Exopod is three-segmented, with very short proximal segment that is armed with prominent sclerotized seta (claw). Second segment is twice as long, bears large pinnate seta on medial margin and small seta also membrane on lateral margin. Subcircular distal segment is equipped with membrane on lateral side and three short and four large pinnate setae on distomedial margin. Endopod is two-segmented, with small proximal segment bears large pinnate seta on medial side. Subcircular distal segment has a membrane on lateral side and six large pinnate setae on distal margin.

Fourth leg (Fig. 34K) is elongated and more sclerotized. Large setae of distal segment are equipped with setules along one side and membrane in the opposite. Membranes on bases of setae are well defined.

Fifth leg consists of three pinnate setae (two grouped together). Sixth leg is vestigial (Fig. 33).

Genital complex (Fig. 33) is suboval, consists of two elongated diagonal cuticular flaps shielding genital openings. Space between flaps is triangular. Posterior ends of flaps bears vestigial sixth legs. Each one has two small pinnate setae. Lateral sides of genital complex are with sensory setules. These structures are protecting the copulatory pores.

Abdomen is two indistinct segmented. Caudal ramus is dominated by three large, centrally located pinnate setae flanked by one small medial and one small lateral setae. One additional small seta is observed laterally on the ventral side (Fig. 33).



(Figure 34J&K): Schematic drawing of 3rd leg, (J) and 4th leg, (K) of the male copepodian parasite, *Caligus elongatus* **von Nordmann, 1832.**

(2) Discussion:

The anatomical structure in the present redescription of the parasitic copepod *Caligus elongatus* **von Nordmann, 1832** infesting gills of sea bass fish, *Dicentrarchus labrax*, in Egypt may be belongs to the genus *Caligus* according to the following criteria based by **Yamaguti (1963), Parker (1969) and Piasecki and MacKinnon (1995).** These generic morphological criteria are: the body is cyclopoid in shape and more or less distinctly segmented and depressed. The body shows four externally recognizable regions: the cephalothorax (carapace), genital segment, abdomen and caudal rami. The cephalothorax (carapace) is large and subcircular, shield-like in shape. Subcircular lunule lies at the anterior-lateral end of each frontal plate**.** Two closely placed eyes present at the center of cephalic area of cephalothorax. The genital segment is well developed, slightly cordiform in shape. Abdomen is oblong in shape.

First antenna is armed with larger setae. Second antenna is more strongly sclerotized and enlarged posterolaterally. First maxilla is conical process smaller and subdivided into base and claw. Second maxilla is strongly and elongated. Maxilliped has broader corpus. Postantennary process is consisting of strong, heavily sclerotized claw directed posteriorly. Fourth thoracic legs are consisting spines and setae. Fifth thoracic leg in the form of a setiferous plate near posterolateral corner of genital segment in both sexes.

The present individuals of the parasitic copepod *Caligus elongatus* **von Nordmann, 1832** infesting gills of sea bass fish, *Dicentrarchus labra*x was recorded for the first time in Egyptian Mediterranean coast.

Caligus elongatus **von Nordmann, 1832** was firstly recorded and described by **von Nordmann (1832)** from the gills of sea bass fish, *Dicentrarchus labra*x which was collected along the coast of Europe, the Irish Sea, Scottish and

Norwegian. *Caligus elongatus* **von Nordmann, 1832** has been recorded from Scottish, Irish and Norwegian on the sea bass and sea bream by **Paperna (1980) and Rodgers and Furones (1998).** It also collected from the gills of Atlantic salmon, *Salmo salar* and rainbow trout, *Oncorhynchus mykiss* along the coast of Europe by **Wootten *et al*. (1982), Pike (1989), Grimnes and Jakobsen (1996) and Bjørn and Finstad (1997)**.

The male *Caligus elongatus* **von Nordmann, 1832** differ from the female in structure of some appendages; second antenna, first maxilla, maxilliped, sternal furca and genital structures (**Piasecki and Mackinnon, 1995)**. Second antenna of male has basal segment more robust than in female. First maxilla of male is conical process smaller than in female.

Maxilliped of male has broader corpus than in female. Sternal furca of male is smaller than in female. Genital complex of male is suboval, consisting of two elongated diagonal cuticular flaps shielding genital openings while in female are two cuticular elevations of uneven surfaces.

The present description of the parasitic copepod *Caligus elongatus* **von Nordmann, 1832** infesting gills of sea bass fish, *Dicentrarchus labra*x is based on the study of males but females can not studied for their disappearance during present collection.

There are close similarities between the present specimens and the specimens described by **Piasecki and Mackinnon (1995)** in all morphological criteria, specially shape and armature of antennule and antenna, shape of cephalothorax, armature and shape of swimming legs, shape of sternal furca and genital complex, shape of abdomen and caudal rami.

Despite slight differences between the present specimens and the closely related one in the body dimensions where the present specimens are smaller except maxilliped, lunule and antennule are larger in length and width Table (13).

Only in recent decades and owing to the development of salmon aquaculture, the parasitic copepod *Caligus elongatus* is commonly referred to as a sea louse and becomes as a one of the most economically important parasites of fishes (**Boxshall and Defaye 1993**).

Abbreviations of the copepodian parasite (*Caligus elongatus*) figures

A	Abdomen
AN	Antenna(2nd antenna)
ANT	Antenule (1st antenna)
CR	Caudal rami
FIL	Fifth leg
FL	First thoracic leg
FOL	Fourth leg
FP	Frontal plat
GS	Genital segment
LU	Lunule
M	Maxillule(1st maxilla)
MXP	Maxilliped
PP	Post-antennal process
S	Sternal furca
SL	Second thoracic leg
TL	Third thoracic leg
TM	Tip of maxilla

Table (13): A comparison between the present species of male parasitic copepod *Caligus elongatus* and the previous description of this species (in µm).

Author(s)	Piasecki and Mackinnon (1995)	The present specimens
Host	*Lepeophtheirus salmonis*	*Dicentrarchus labrax*
Locality	Canada	Egypt
Maximum length x Maximum width	4250 x 1700	2880 x 1605
Genital complex	-	542.5 x 487.5
Abdomen	-	362.5 x 257.5
Uropod	-	202.5 x 112.5
Lunule and Antennule	625 x 177.5	715 x 180
Antenna	375 x 150	210 x 105
Maxillule	-	32.5 x 25
Maxilla	562.5 x 450	250 x 27.5
Maxilliped	586 x 391	700 x 405
Sternal furca	250 x 100	137.5 x 50
Postantennal process	187.5 x 75	205 x 65
First leg	1025 x 287.5	540 x 150
Second leg	1000 x 250	545 x 185
Third leg	700 x 600	450 x 400
Fourth leg	500 x 110	355 x 80

CHAPTER II

Ecological studies

(I)General prevalence and mean intensity of infestation with the parasitic copepods.

Twelve examined fishes belonging to nine families, eleven genera and twelve species (*Dicentrarchus punctatus*, *Sarotherodon galilaeus*, *Mugil cephalus*, *Valamugil seheli*, *Diplodus sargus*, *Liza ramada*, *Anguilla anguilla*, *Dicentrarchus labrax*, *Solea solea aegyptaica*, *Argyrosomus regius*, *Lutjanus lineolatus* and *Upeneus asymmetricus*) were collected during the present investigation from April 2005 until March 2006. Eight of them (*Dicentrarchus punctatus, Sarotherodon galilaeus, Mugil cephalus, Valamugil seheli, Diplodus sargus, Liza ramada, Anguilla anguilla* and *Dicentrarchus labrax*) were infested with parasitic copepods while the remain fishes (*Solea solea aegyptaica*, *Argyrosomus regius, Lutjanus lineolatus* and *Upeneus asymmetricus*) were non-infested.

The prevalence and the mean intensity of the infestation with the eight parasitic copepods are given throughout the year with total prevalence (61%) and total mean intensity (2.48 parasite/infested fish) (Table 14). During the present study, total prevalence of *Lernanthropus kroyeri* was 64% and its mean intensity was 2.80 parasite/infested fish, total prevalence of *Lamproglena minuta* was 73% and its mean intensity was 0.68 parasite/infested fish, total prevalence of *Ergasilus briani* was 43% and its mean intensity was 0.73 parasite/infested fish, total prevalence of *Ergasilus lizae* was 52% and its mean intensity was 8.76 parasite/infested fish, total prevalence of *Ergasilus australiensis* was 72% and its mean intensity was 4.17 parasite/infested fish, total prevalence of *Ergasilus mosulensis* was 41% and its mean intensity was 0.79 parasite/infested fish, total prevalence of *Ergasilus celestis* was 64% and its mean intensity was 4.20

Table (14): General prevalence and mean intensity of infestation with eight parasitic copepods during the present investigation.

Parasitic copepods	Prevalence%	Mean intensity (parasite/ infested fish)	Site 1		Site 2		Site 3	
			Prev.%	Mean intensity	Prev.%	Mean intensity	Prev.%	Mean intensity
Lernanthropus kroyeri	**64%**	**2.8**	**44%**	**3**	**11%**	**1.27**	**0**	**0**
Lamproglena minuta	**73%**	**0.68**	**0**	**0**	**0**	**0**	**73%**	**0.68**
Ergasilus briani	**43%**	**0.73**	**17%**	**2**	**3%**	**1**	**0**	**0**
Ergasilus lizae	**52%**	**8.76**	**23%**	**1.2**	**8%**	**4**	**0**	**0**
Ergasilus australiensis	**72%**	**4.17**	**55%**	**1.9**	**5%**	**1.67**	**0**	**0**
Ergasilus mosulensis	**41%**	**0.79**	**21%**	**4**	**15%**	**3**	**0**	**0**
Ergasilus celestis	**64%**	**4.2**	**33%**	**2.3**	**12%**	**9**	**0**	**0**
Caligus elongatus	**58%**	**2.16**	**37%**	**2**	**3%**	**0.21**	**0**	**0**
Total	**61%**	**2.48**	**56%**	**3.62**	**55%**	**3.71**	**73%**	**0.68**

parasite/infested fish and total prevalence of *Caligus elongatus* was 58% and its mean intensity was 2.16 parasite/infested fish (Table 14).

Most of the parasitic copepods were collected from two sites (1, Lake El-Manzala close to Shata village and 2, Lake El-Manzala close to Deba village) and the eighth parasite *Lamproglena minuta* was collected only from the third site (3, the Nile River at El-Zarka city). Total prevalence of infestation with the parasitic copepods in site 1 was 56% and with mean intensity 3.62 parasite/infested fish, and in site 2 was 55% and with mean intensity 3.71 parasite/infested fish. Meanwhile, site 3 based on the presence of one parasite, *Lamproglena minuta* with prevalence 73% and mean intensity 0.68 parasite/infested fish.

At the level of parasite species, the fishes collected from the site 1 had the highest prevalence and mean intensity except the infestation level with *Ergasilus lizae* and *Ergasilus celestis* was high in site 2 than in site 1 (Table 14).

(II) Relationship between the seasonal changes and the infestation with parasitic copepods

The seasonal changes in the prevalence and the mean intensity of the parasitic copepods infestation at Lake El-Manzala, close to Shata village (site 1), close to El-Deba village (site 2) and close to the Nile River at El-Zarka City (site 3) are shown in Tables 15, 16, 17, 18, 19 and 20 and in Figures 35, 36, 37, 38, 39 and 40. Where eleven examined fishes (*Dicentrarchus punctatus*, *Mugil cephalus*, *Valamugil seheli*, *Diplodus sargus*, *Liza ramada*, *Anguilla anguilla*, *Dicentrarchus labrax*, *Solea solea aegyptaica*, *Argyrosomus regius*, *Lutjanus lineolatus* and *Upeneus asymmetricus*) were collected from the marine waters of Lake El-Manzala at site 1 and 2. While, the twelve examined fish (*Sarotherodon galilaeus*) was collected from the freshwaters of the Nile River at site 3.

A- Prevalence and Mean intensity in site 1

At site 1, table 15 and figure 35 show that the prevalence of *Lernanthropus kroyeri* was high with a maximum value of 7% during summer and autumn then decline in winter. The prevalence of *Ergasilus briani* reached its maximum value of 8% during summer then decline in other seasons. The prevalence of *Ergasilus lizae* reached its maximum value of 44% during spring then decline in other seasons. The prevalence of *Ergasilus australiensis* was high with a maximum value of 8% during spring and summer then decline in winter. The prevalence of *Ergasilus mosulensis* reached its maximum value of 9% during autumn then decline in other seasons. The prevalence of *Ergasilus celestis* reached its maximum value of 58% through spring then decline in other seasons.

Table (15): Seasonal changes of prevalence (%) of parasitic copepods at Lake El-Manzala, close to Shata village (site 1).

Parasitic copepods	Season	Prevalence(%)	DF	t-value	P
Lernanthropus kroyeri	Spring	6%	0.3	1.95	0.001**
	Summer	7%			
	Autumn	7%			
	Winter	0			
Lamproglena minuta	Spring	0	0.0	-0.69	0.543
	Summer	0			
	Autumn	0			
	Winter	0			
Ergasilus briani	Spring	0	0.0	1.35	0.05*
	Summer	8%			
	Autumn	0			
	Winter	0			
Ergasilus lizae	Spring	44%	4.0	7.31	0.000***
	Summer	8%			
	Autumn	0			
	Winter	0			
Ergasilus australiensis	Spring	8%	3.0	2.11	0.01**
	Summer	8%			
	Autumn	1%			
	Winter	0			
Ergasilus mosulensis	Spring	1%	4.0	2.15	0.01**
	Summer	1%			
	Autumn	9%			
	Winter	0			
Ergasilus celestis	Spring	58%	4.0	7.51	0.000***
	Summer	1%			
	Autumn	1%			
	Winter	0			
Caligus elongatus	Spring	0	2.0	3.12	0.01**
	Summer	7%			
	Autumn	1%			
	Winter	0			

P, probability; DF,Degree of freedom;*, significant; **, highly significant; ***, very highly significant.

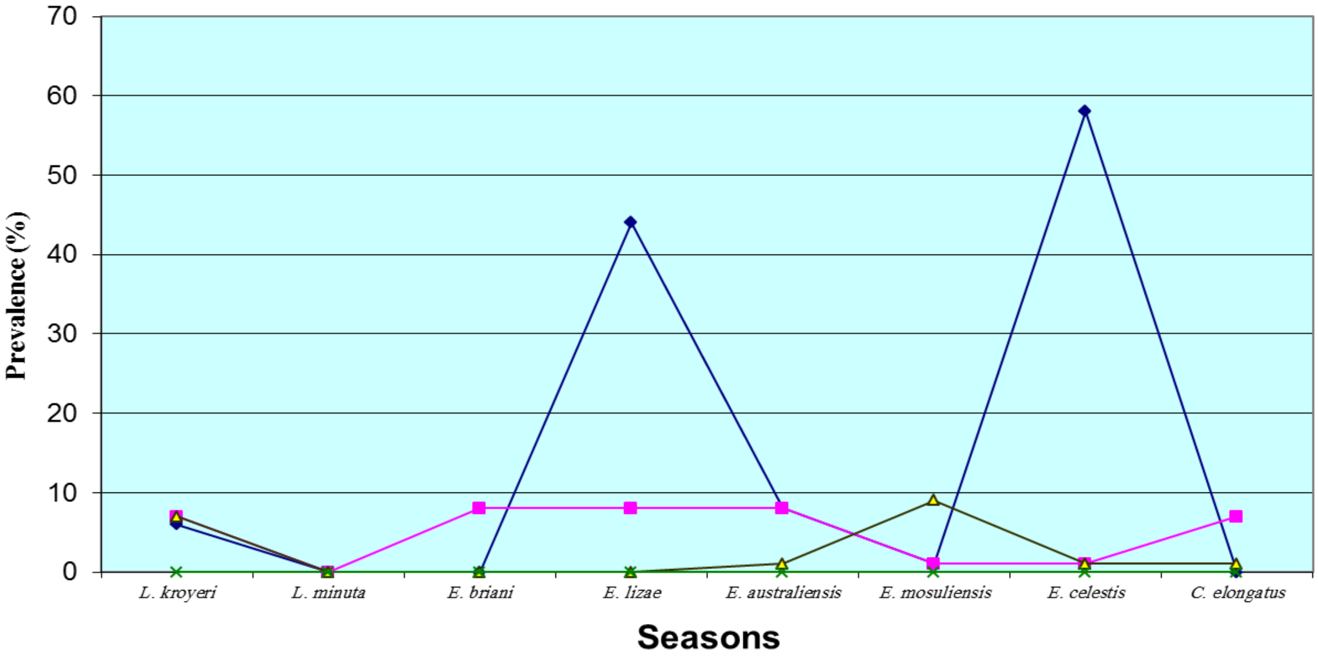


Figure (35): Seasonal changes in the prevalence of parasitic copepods at Lake El-Manzala, close to Shata village (site1).

Table(16): Seasonal changes of the mean intensity of parasitic copepods at Lake El-Manzala, close to Shata village (site 1).

Parasitic copepods	Season	Mean	±SD	*t*-value	P
Lernanthropus kroyeri	Spring	0.070	0.258	-0.05	0.971
	Summer	1.250	1.571	-1.26	0.018*
	Autumn	1.760	3.474	-1.76	0.002**
	Winter	0.000	0.00	0.000	1.000
Lamproglena minuta	Spring	0.000	0.00	-0.69	0.543
	Summer	0.000	0.00	-0.71	0.169
	Autumn	0.000	0.00	-0.29	0.524
	Winter	0.000	0.00	-0.33	0.317
Ergasilus briani	Spring	0.000	0.000	0.00	1.000
	Summer	2.000	2.90	-2	0.050*
	Autumn	0.000	0.00	0	1.000
	Winter	0.000	0.00	-0.2	0.690
Ergasilus lizae	Spring	20.380	17.90	-18.11	0.000***
	Summer	3.310	5.38	-2.41	0.000***
	Autumn	0.000	0.00	-1.75	0.003**
	Winter	0.000	0.00	-1.14	0.013*
Ergasilus australiensis	Spring	4.250	5.88	-4.25	0.01*
	Summer	4.170	7.86	-3.33	0.000***
	Autumn	0.500	1.23	-0.5	0.557
	Winter	0.000	0.00	0	1.000
Ergasilus mosulensis	Spring	0.180	0.60	-0.21	0.890
	Summer	0.800	1.30	-0.9	0.279
	Autumn	3.000	1.63	-2	0.02*
	Winter	0.000	0.00	-1.2	0.02*
Ergasilus celestis	Spring	37.300	10.92	-4.92	0.000***
	Summer	0.600	0.89	-3.22	0.000***
	Autumn	0.500	0.71	-0.25	0.806
	Winter	0.000	0.00	0	1.000
Caligus elongatus	Spring	0.000	0.00	0	1.000
	Summer	3.750	4.79	-2.5	0.018*
	Autumn	0.330	0.58	-5.5	0.000***
	Winter	0.000	0.00	0	1.000

P, probability; ±SD, standard deviation;*, significant; **, highly significant; ***, very highly significant.

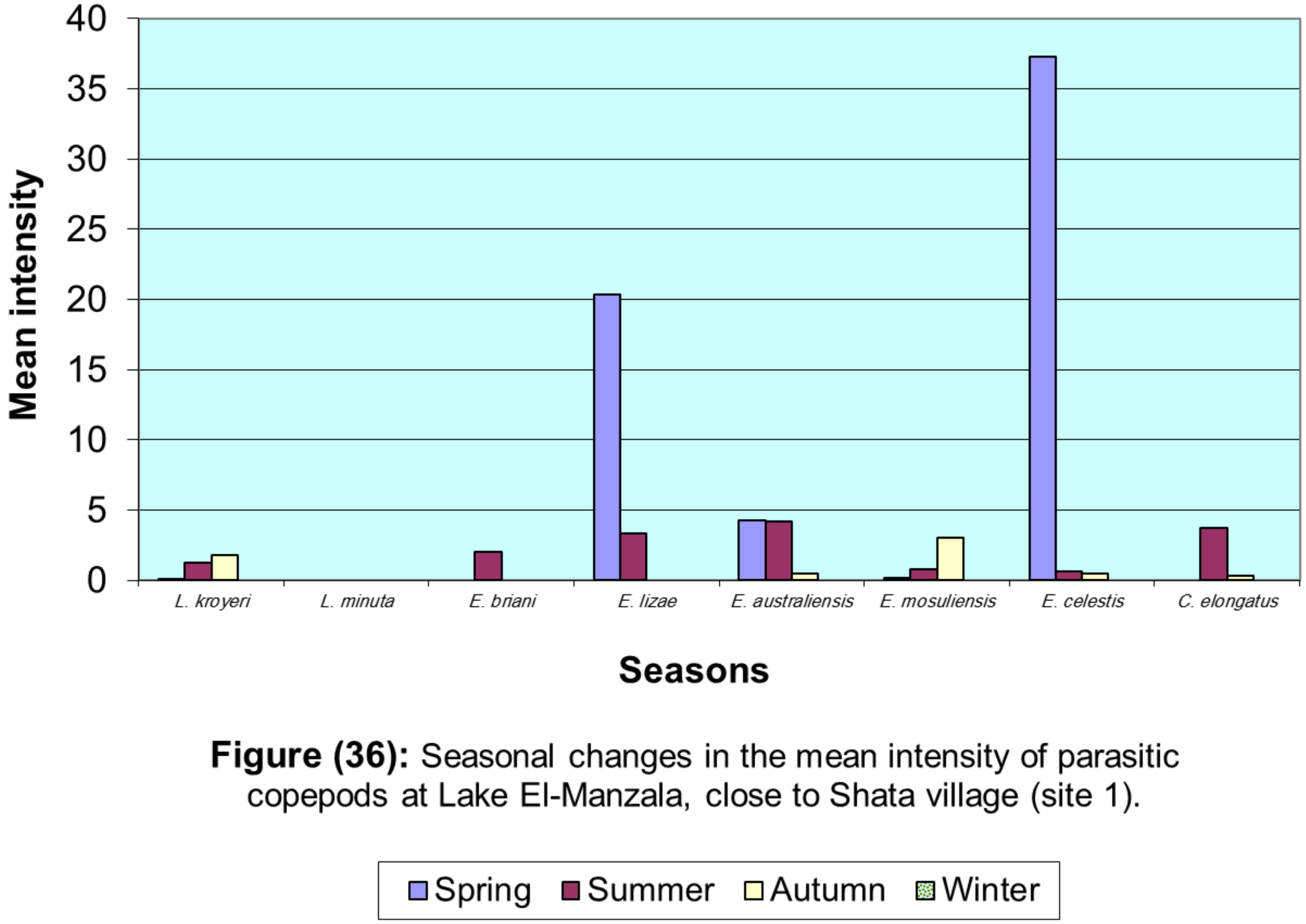


Figure (36): Seasonal changes in the mean intensity of parasitic copepods at Lake El-Manzala, close to Shata village (site 1).

Table(17): Seasonal changes of prevalence (%) of parasitic copepods at Lake El-Manzala, close to El-Deba village (site 2).

Parasitic copepods	Season	Prevalence(%)	DF	t-value	P
Lernanthropus kroyeri	Spring	0	0.5	1.33	0.02**
	Summer	1%			
	Autumn	0			
	Winter	0			
Lamproglena minuta	Spring	0	0.0	-0.49	0.423
	Summer	0			
	Autumn	0			
	Winter	0			
Ergasilus briani	Spring	0	0.0	1.33	0.05*
	Summer	0			
	Autumn	0			
	Winter	3%			
Ergasilus lizae	Spring	0	4.0	6.11	0.000***
	Summer	8%			
	Autumn	0			
	Winter	0			
Ergasilus australiensis	Spring	0	3.0	3.11	0.05*
	Summer	5%			
	Autumn	0			
	Winter	0			
Ergasilus mosulensis	Spring	1%	4.0	2.16	0.03**
	Summer	6%			
	Autumn	0			
	Winter	8%			
Ergasilus celestis	Spring	8%	3.0	3.77	0.000***
	Summer	23%			
	Autumn	0			
	Winter	0			
Caligus elongatus	Spring	0	2.0	5.11	0.01**
	Summer	0			
	Autumn	3%			
	Winter	0			

P, probability; DF,Degree of freedom;*, significant; **, highly significant; ***, very highly significant.

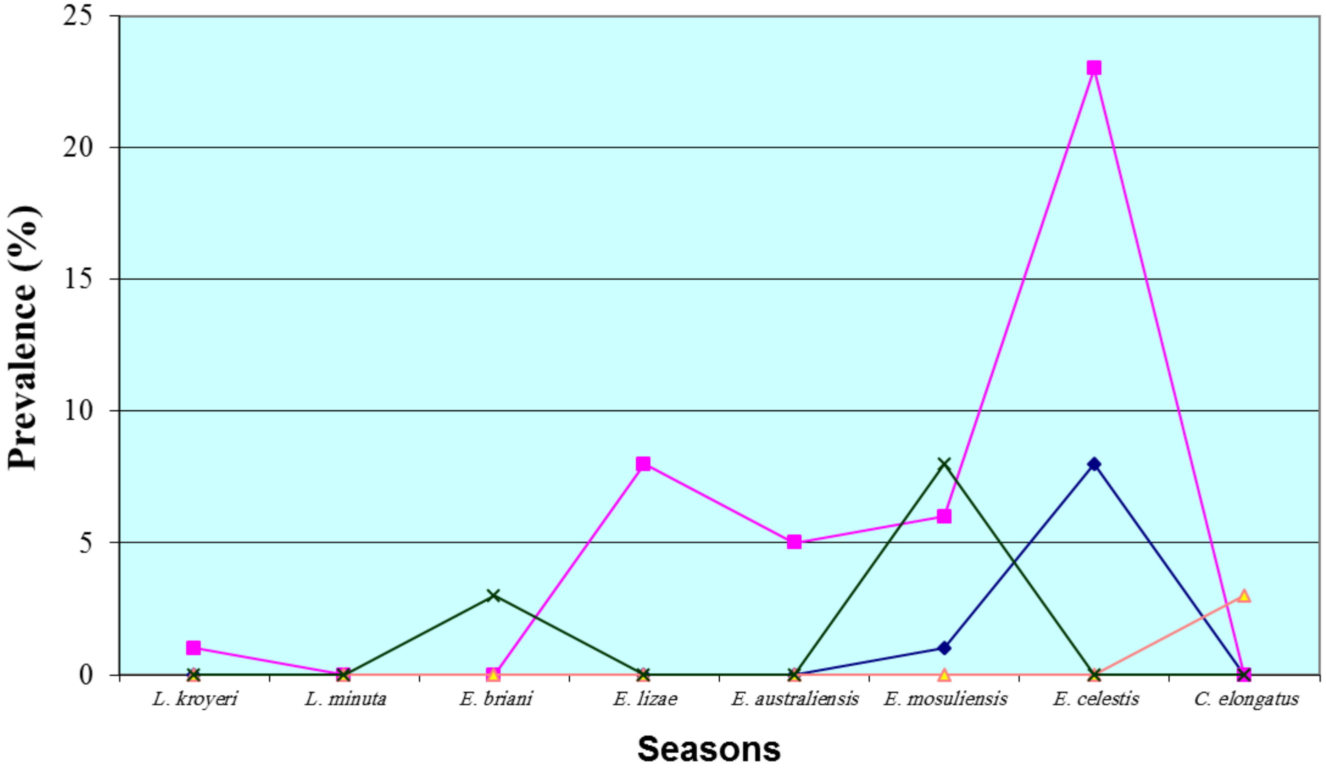


Figure (37): Seasonal changes in the prevalence of parasitic copepods at Lake El-Manzala, close to El-Deba village (site 2).

Table(18): Seasonal changes of the mean intensity of parasitic copepods at Lake El-Manzala, close to El-Deba village (site 2).

Parasitic copepods	Season	Mean	±SD	t-value	P
Lernanthropus kroyeri	Spring	0.00	0.00	0.64	0.62
	Summer	1.27	1.22	-0.55	0.37
	Autumn	0.00	0.00	-1.48	0.10
	Winter	0.00	0.00	0.33	0.53
Lamproglena minuta	Spring	0.00	0.00	0.00	1.00
	Summer	0.00	0.00	0.00	1.00
	Autumn	0.00	0.00	0.00	1.00
	Winter	0.00	0.00	0.00	1.00
Ergasilus briani	Spring	0.00	0.00	0.69	0.62
	Summer	0.00	0.00	-1.29	0.24
	Autumn	0.00	0.00	0.29	0.75
	Winter	1.00	0.00	0.13	0.78
Ergasilus lizae	Spring	0.00	0.00	-17.42	0.000***
	Summer	0.00	0.00	-1.70	0.012*
	Autumn	2.55	3.24	-1.46	0.012*
	Winter	1.60	2.07	-0.81	0.050*
Ergasilus australiensis	Spring	0.00	0.000	-3.56	0.024*
	Summer	1.67	1.528	-2.63	0.005**
	Autumn	0.00	0.000	-0.21	0.803
	Winter	0.00	0.000	0.33	0.639
Ergasilus mosulensis	Spring	0.33	0.577	0.47	0.747
	Summer	1.00	1.414	-0.19	0.826
	Autumn	0.00	0.000	-1.71	0.045*
	Winter	1.50	2.380	-0.87	0.075
Ergasilus celestis	Spring	2.83	0.980	-4.23	0.007**
	Summer	6.50	5.447	-2.52	0.007**
	Autumn	0.00	0.000	0.04	0.969
	Winter	0.00	0.000	0.33	0.574
Caligus elongatus	Spring	0.00	0.000	0.69	0.741
	Summer	0.00	0.000	-1.79	0.101
	Autumn	0.21	0.000	-5.21	0.000***
	Winter	0.00	0.000	0.33	0.487

P, probability; ±SD, standard deviation;*, significant; **, highly significant; ***, very highly significant.

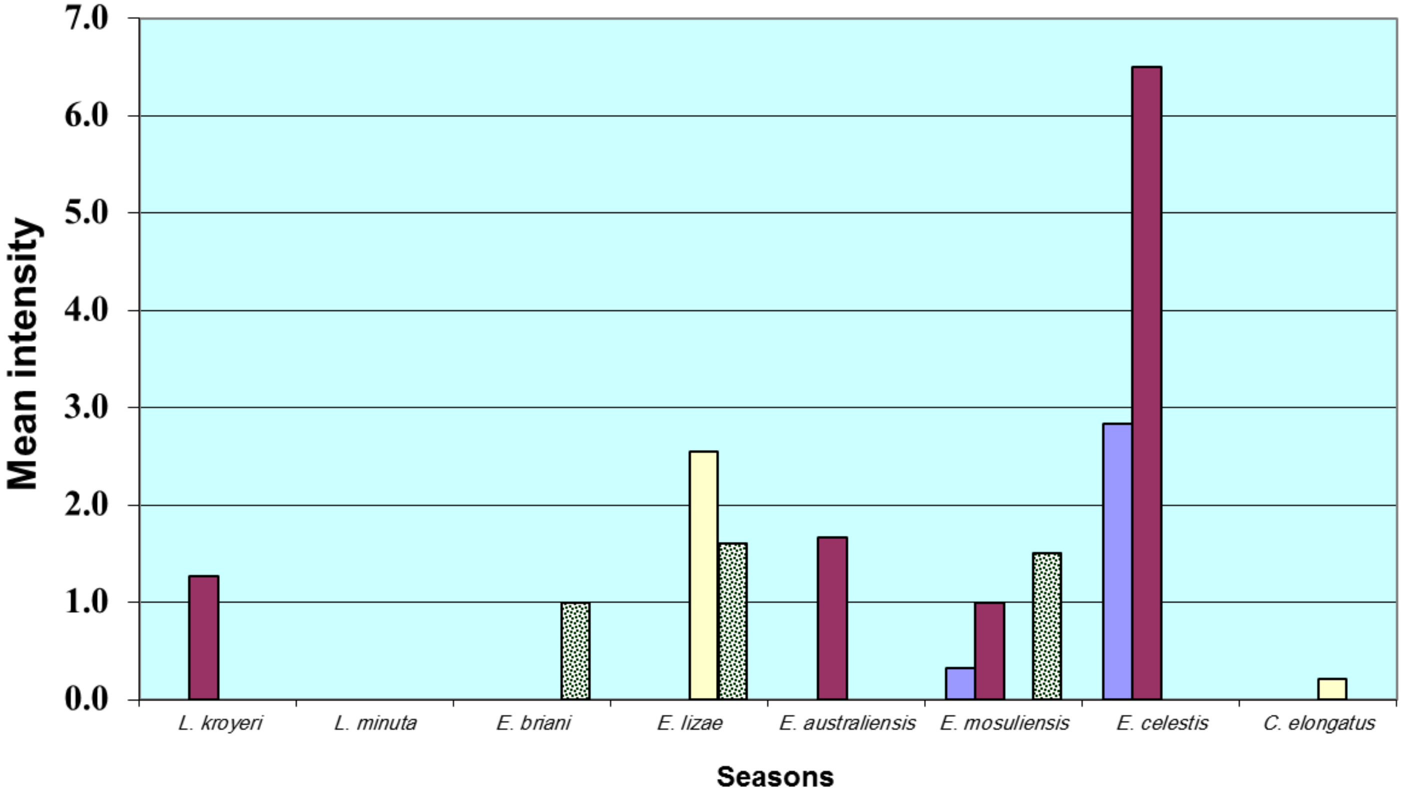


Figure (38): Seasonal changes in the mean intensity of parasitic copepods at Lake El-Manzala, close to El-Deba village (site 2).

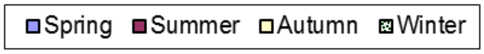


Table (19): Seasonal changes of prevalence (%) of parasitic copepods close to the Nile River at El-Zarka City (site 3).

Parasite copepod	Season	Prevalence(%)	DF	*t*-value	P
Lamproglena minuta	Spring	69%	5.0	7.29	0.637
	Summer	71%			
	Autumn	60%			
	Winter	63%			

P, probability; DF, Degree of freedom.

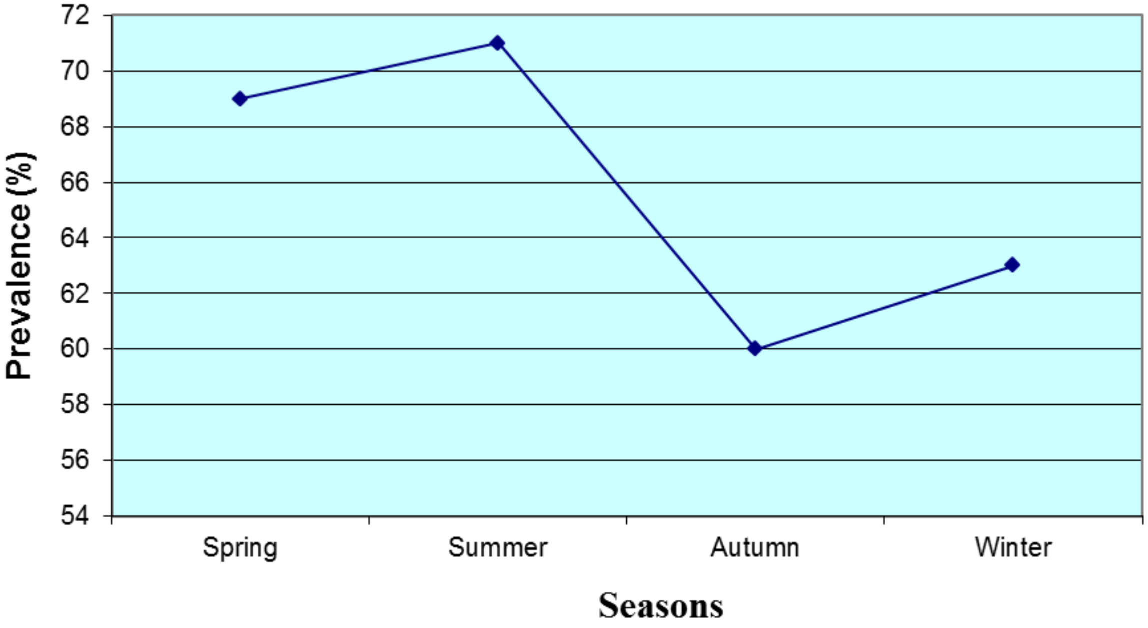


Figure (39): Seasonal changes in the prevalence of copepodian parasite, *Lamproglena minuta* infesting *Sarotherodon galilaeus* close to the Nile River at El-Zarka city (site 3).

Table(20): Seasonal changes of the mean intensity of parasitic copepods close to the Nile River at El-Zarka City (site 3).

Parasite copepod	**Season**	**Mean intensity**	**±SD**	***t*-value**	**P**
Lamproglena minuta	Spring	70.10	1.112	-0.69	0.741
	Summer	75.29	1.115	1.79	0.101
	Autumn	39.11	1.626	5.21	0.000***
	Winter	40.33	0.658	-0.33	0.487

P, probability; ±SD, standard deviation; ***, very highly significant.

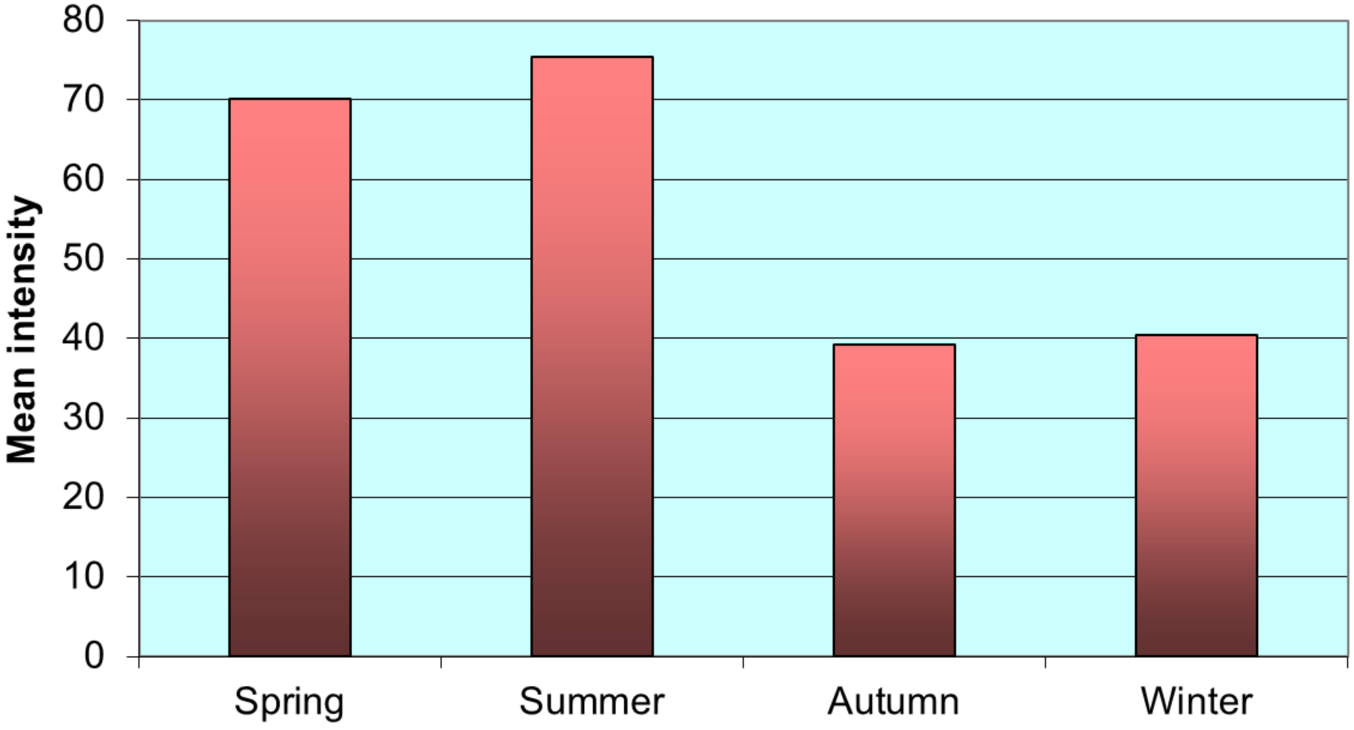


Figure (40): Seasonal changes in the mean intensity of copepodian parasite, *Lamproglena minuta* **i**nfesting *Sarotherodon galilaeus* close to the Nile River at El-Zarka city (site 3).

The prevalence of *Caligus elongates* was high with a maximum value of 7% during summer then decline in other seasons. On the other hand, *Lamproglena minuta* was disappeared during four seasons at this site.

Statistical analysis (Table 15) using *t*-student test has revealed that the differences in the prevalence of *Ergasilus lizae* and *Ergasilus celestis* among the different seasons was very highly significant (*t*-value=7.31, P=0.000) of *Ergasilus lizae* and (*t*-value=7.51, P=0.000) of *Ergasilus celestis*. Moreover, the differences in the prevalence of other copepods was highly significant ($P<0.05$).

At site 1, table 16 and figure 36 show that the mean intensity of *Lernanthropus kroyeri* and *Caligus elongatus* was low during Spring, increased to a maximum value of 3.750 parasite/infested fish of *Caligus elongatus* during Summer and Autumn. Also, it was declined dramatically to reach its minimum value during Winter of the two species. The mean intensity of *Ergasilus australiensis*, *Ergasilus lizae, Ergasilus mosulensis* and *Ergasilus celestis* increased from Spring to Autumn reaching a maximum value of 37.300 parasite/infested fish of *Ergasilus celestis* and decreased to reach a minimum value of the remain three species during Winter.

The mean intensity of *E. briani* was low during spring and increased to 2.0 parasite/infested fish in summer then decline during Autumn and Winter. On the other hand, *Lamproglena minuta* was disappeared during the four seasons at the site 1.

Statistical analysis (Table 16), using ANOVA’s and LSD tests indicated presence of significant differences in the seasonal mean intensity values among fish hosts. These differences were very highly significant for *Ergasilus lizae*, through Spring and Summer, *Ergasilus australiensis* and *Ergasilus celestis* during Summer and *Caligus elongatus* during Autumn ($P<0.05$). On the other hand, the differences in the mean intensity values were highly significant for *Ergasilus celestis* through Spring, for *Lernanthropus kroyeri* and *Ergasilus lizae* during Autumn and nearly significant for *Lernanthropus kroyeri*, *E. briani* and *Caligus elongatus* through Summer, for *Ergasilus lizae* and *Ergasilus mosulensis* during Winter and for *Ergasilus australiensis* during Spring. However, no significant changes were observed in these parasites among the different seasons ($P>0.05$).

B- Prevalence and Mean intensity in site 2

At site 2, table 17 and figure 37 show that the prevalence of *Lernanthropus kroyeri*, *Ergasilus lizae*, *Ergasilus australiensis*, *Ergasilus mosulensis* and *Ergasilus celestis* was high with a maximum value of 23% of *Ergasilus celestis* during summer then decline in other seasons.

The prevalence of *Ergasilus briani* reached its maximum value of 3% during winter then decline in other seasons. Also, the prevalence of *Caligus elongates* was high with a maximum value of 3% during autumn then decline in other seasons. On the other hand, *Lamproglena minuta* was disappeared during four seasons at the site 2.

Statistical analysis (Table 17) using *t*-student test has revealed that the differences in the prevalence of *Ergasilus lizae* and *Ergasilus celestis* among the different seasons was very highly significant (*t*-value=6.11, P=0.000) of *Ergasilus lizae* and (*t*-value=3.77, P=0.000) of *Ergasilus celestis*. Moreover, the differences in the prevalence of other copepods was highly significant ($P<0.05$).

At site 2, table 18 and figure 38 show that the mean intensity of *Lernanthropus kroyeri* and *Ergasilus australiensis* was low during Spring, increased during Summer. Also, it was declined dramatically to reach its minimum value during Autumn and Winter. The mean intensity of *Ergasilus mosulensis* and *Ergasilus celestis* increased during Spring and Summer and decreased to reach a minimum value during Autumn and Winter. The mean intensity of *E. briani* was low during Spring, increased to 1.0 parasite/infested fish in Winter then decline during Summer and Autumn. The mean intensity of *Ergasilus lizae* and *Caligus elongatus* were low during spring and Summer then increased during Autumn and Winter (Table 18). On the other hand, *Lamproglena minuta* was disappeared during four seasons at the site 2.

Statistical analysis (Table 18), using ANOVA's and LSD tests indicated presence of significant differences in the seasonal mean intensity values among fish hosts. These differences were very highly significant for *Ergasilus lizae*, through Spring, *Caligus elongatus* during Autumn (P<0.05). Also, the differences in the mean intensity values were highly significant for *Ergasilus australiensis* through Summer, *Ergasilus celestis* during during Spring and Summer and nearly significant for *Ergasilus lizae* through Summer, Autumn and Winter, *Ergasilus australiensis* through Spring and *Ergasilus mosulensis* during Autumn. However, no significant changes were observed in these parasites among the other different seasons (P>0.05).

C- Prevalence and Mean intensity in site 3

At site 3, table 19 and figure 39 show that the prevalence of *Lamproglena minuta* was high with a maximum value of 71% during Summer and a minimum value of 60% through Autumn.

Statistical analysis (Table 19) using *t*-student test has revealed that the differences in the prevalence of *Lamproglena minuta* among the different seasons was not statistically significant (*t*-value=7.29, P=0.637).

At site 3, table 20 and Figure 40 show that the mean intensity of *Lamproglena minuta* was low during Autumn to reach its minimum value (39 parasite/infested fish) and increased to a maximum value of (75.29 parasite/infested fish) during Summer.

Statistical analysis (Table 20), using ANOVA's and LSD tests show that the mean intensity of *Lamproglena minuta* showed a very highly significant difference during Autumn (*t*-value=5.21, P=0.000), but the differences were not statistically significant in the other seasons (P>0.05).

There are seasonal variation in the prevalence and mean intensity of the most parasitic copepods among the sites of collection. The prevalence and the mean intensity of infestation with most of the parasitic copepods increased in Spring and some of them increased in Autumn and others increased in Summer at site 1. Meanwhile, most of them were prevalent and intense in Summer, some of them were in Autumn and other in Winter at site 3. Clearly, most parasites if preferred, in their distribution, one season at site 1 they can prefer the next season at site 2, except the parasitic copepods; *Lernanthropus kroyeri* and *Ergasilus briani*. Otherwise, *Ergasilus lizae* had the same behaviour in its prevalence like the most parasites but had other behaviour in its intensity. On the other hand, existence of the eighth parasite, *Lamproglena minuta* increased in Summer only at site 3 during the present study.

(III) Correlation analysis of the relationship between water parameters and mean intensity of parasitic copepods

According to the output of Person Correlation Coefficients, a common response was detected for the parasitic copepods at site 1 where their mean intensity was negatively correlated with water parameters; Temperature, pH, Salinity and Oxygen content Table (21). At site 1, the relationship was very highly significant for water temperature (*t*-value = -55.501, P = 0.000). Also, it was very highly significant for pH (*t*-value = -16.410, P = 0.000), very highly significant for salinity (*t*-value = -36.617, P = 0.000) and for Oxygen content (*t*-value = -10.964, P = 0.000).

Person Correlation Coefficient has revealed that the negative correlations between water parameters; Temperature, pH, Salinity and Oxygen content and the mean intensity of parasitic copepods were very highly significant at site 1($P<0.05$; Table 21).

According to the output of Person Correlation Coefficients, a common response was detected for the parasitic copepods at site 2 where their mean intensity was negatively correlated with water parameters; Temperature, pH, Salinity and Oxygen content (Table 21). At site 2, the relationship was very highly significant for water temperature (*t*-value = -94.745, P = 0.000). Also, it was very highly significant for pH (*t*-value = -37.487, P = 0.000), very highly significant for salinity (*t*-value = -72.184, P = 0.000) and for Oxygen content (*t*-value = -20.110, P = 0.000).

Person Correlation Coefficient has revealed that the negative correlations between water parameters; Temperature, pH, Salinity and Oxygen content and the

Table (21): Mean intensity of parasitic copepods at Lake El-Manzala, close to Shata village (site 1) and close to El-Deba village (site 2), the Nile River at El-Zarka city (site 3) in relation to ecological parameters.

Parasitic copepods Sites	Water parameters		Mean intensity	±SE	*t*-value	P
Site 1	Temperature	25.19	3.62	0.422	-55.501	0.000***
	pH	7.67		0.360	-16.41	0.000***
	Salinity	24.24		5.585	-36.617	0.000***
	Oxygen content	6.21		0.405	-10.964	0.000***
Site 2	Temperature	26.15	3.71	0.268	-94.745	0.000***
	pH	7.41		0.177	-37.487	0.000***
	Salinity	25.71		8.080	-72.184	0.000***
	Oxygen content	8.389		0.378	-20.11	0.000***
Site 1	Temperature	25.19	3.62	0.422	-55.501	0.000***
	pH	7.67		0.360	-16.41	0.000***
	Salinity	24.24		5.585	-36.617	0.000***
	Oxygen content	6.21		0.405	-10.964	0.000***
Site 3	Temperature	25.46	0.68	0.261	-95.429	0.000***
	pH	7.18		0.104	-63.44	0.000***
	Salinity	12.47		4.274	-23.86	0.000***
	Oxygen content	4.354		0.204	-18.649	0.000***
Site 2	Temperature	26.15	3.71	0.268	-94.745	0.000***
	pH	7.41		0.177	-37.487	0.000***
	Salinity	25.71		8.080	-72.184	0.000***
	Oxygen content	8.389		0.378	-20.11	0.000***
Site 3	Temperature	25.46	0.68	0.261	-95.429	0.000***
	pH	7.18		0.104	-63.44	0.000***
	Salinity	12.47		4.274	-23.86	0.000***
	Oxygen content	4.354		0.204	-18.649	0.000***

P, Probability; ±SE, standard error of mean; ***, very highly significant.

mean intensity of parasitic copepods were very highly significant at site 2 ($P<0.05$; Table 21).

According to the output of Person Correlation Coefficients, a common response was detected for the parasitic copepods at site 3 where their mean intensity was negatively correlated with water parameters; Temperature, pH, Salinity and Oxygen content (Table 21). At site 3, the relationship was very highly significant for water temperature (*t*-value = -95.429, P = 0.000). Also, it was very highly significant for pH (*t*-value = -63.764, P = 0.000), very highly significant for salinity (*t*-value = -23.860, P = 0.000) and for Oxygen content (*t*-value = -18.649, P = 0.000).

Person Correlation Coefficient has revealed that the negative correlations between water parameters; Temperature, pH, Salinity and Oxygen content and the mean intensity of parasitic copepods were very highly significant at site 3 ($P<0.05$; Table 21).

(IV) The relationship between the host body length, host body weight, host sex and the infestation level of the parasitic copepods

1. Host body length

All the parasitic copepods during the present investigation had highly mean intensity (infestation level) on the length class I than the other length classes (Table 22).

According to the output of linear regression, different response patterns were detected among the parasitic copepods *Lernanthropus kroyeri*, *Lamproglena minuta*, *Ergasilus briani*, *Ergasilus lizae*, *Ergasilus australiensis*, *Ergasilus mosulensis*, *Ergasilus celestis* and *Caligus elongatus* under investigation in relation to body length of *Dicentrarchus punctatus*, *Sarotherodon galilaeus*, *Mugil cephalus*, *Valamugil seheli*, *Diplodus sargus*, *Liza ramada*, *Anguilla anguilla* and *Dicentrarchus labrax*, respectively. The two response patterns indicated that the infestation level of *Ergasilus briani* and the body length of *Mugil cephalus* and *Ergasilus australiensis* and the body length of *Diplodus sargus* are negatively correlated. The other response pattern indicated that the infestation level of the remaining copepods and the body length of their hosts are positively correlated (Table 22 and Fig. 41).

Statistical analysis (using linear regression, Table 22) has revealed the presence of highly significant correlations between the mean intensity (infestation level) of *Ergasilus lizae* and body length of *Valamugil seheli* (*t*-value = 3.292, P = 0.002), nearly significant on *Caligus elongatus* and the body length of *Dicentrarchus labrax* (*t*-value = 2.323, P = 0.034) and very highly significant in *Lernanthropus kroyeri* (*t*-value = 5.963, P = 0.000). However, non-significant correlations were detected for other parasites among fishes in different length classes (P>0.05; Table 22).

Table(22): Regression analysis of the mean intensity of parasitic copepods in relation to host body length.

Parasitic copepods	**Length classes**	**Mean intensity**	**Beta**	***t*-value**	**p**
Lernanthropus kroyeri	I	7.299(+)	1.174	5.963	0.000***
	II	0.665			
	III	0.059(-)			
Lamproglena minuta	I	0.157(+)	0.083	0.409	0.684
	II	0.041			
	III	0.001(-)			
Ergasilus briani	I	0.168(+)	-0.016	-0.054	0.958
	II	0.004(-)			
	III	0.005			
E.lizae	I	47.736(+)	0.683	3.292	0.002**
	II	4.327			
	III	0.367(-)			
E.australiensis	I	1.494(+)	-0.029	-0.099	0.922
	II	0.072			
	III	0.041(-)			
E.mosulensis	I	0.986(+)	0.279	0.509	0.614
	II	0.093			
	III	0.003(-)			
E.celestis	I	8.466(+)	0.489	1.081	0.289
	II	0.4			
	III	0.037(-)			
Caligus elongatus	I	20.278(+)	1.069	2.323	0.034*
	II	1.206			
	III	0.04(-)			

Beta, Regression coefficient; P, probability;*,**, significant; ***, very highly significant; (+) designated for the maximum value and (-) designated for the minimum value.

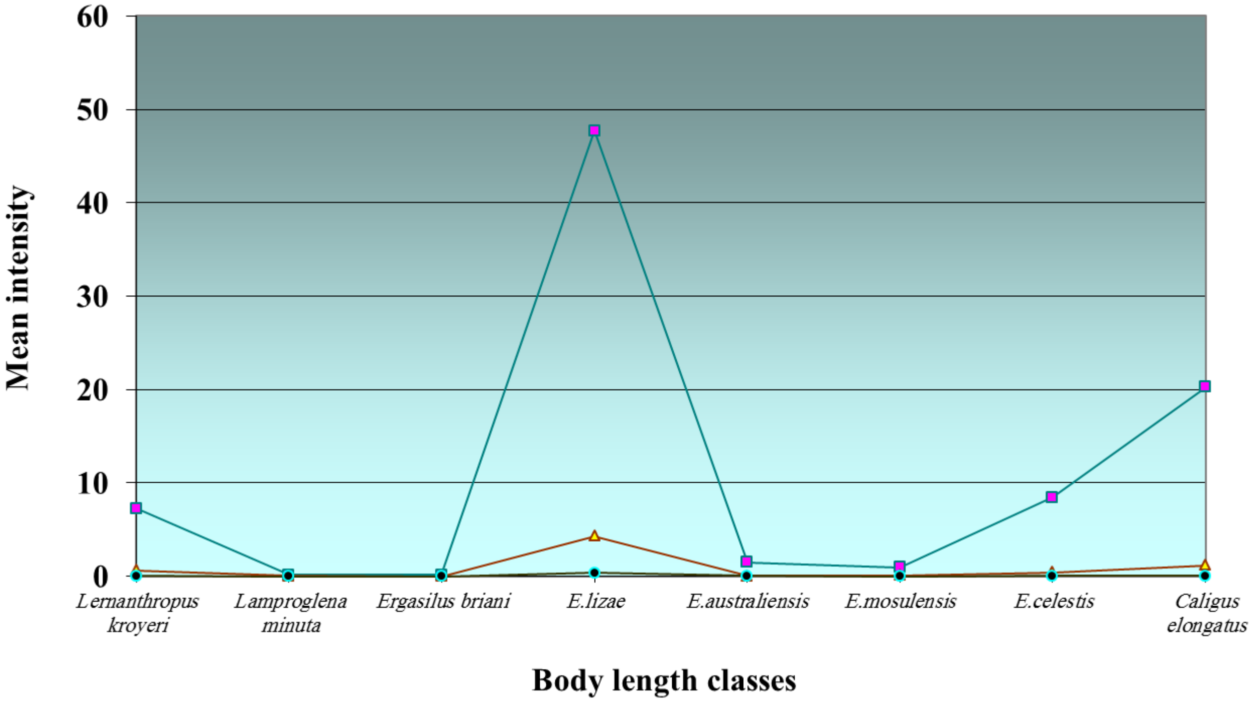


Figure (41): Mean intensity of parasitic copepods on length classes of their fish hosts.

2. Host body weight

All the parasitic copepods during the present investigation had highly mean intensity (infestation level) on the weight class I than the other weight classes (Table 23).

According to linear regression the infestation level of the copepodian parasites, *Lernanthropus kroyeri*, *Lamproglena minuta*, *Ergasilus briani*, *Ergasilus lizae*, *Ergasilus australiensis*, *Ergasilus mosulensis*, *Ergasilus celestis* and *Caligus elongatus*, in relation to the body weight of *Dicentrarchus punctatus*, *Sarotherodon galilaeus*, *Mugil cephalus*, *Valamugil seheli*, *Diplodus sargus*, *Liza ramada*, *Anguilla anguilla* and *Dicentrarchus labrax*. ANOVA indicated the presence of significant differences in the mean intensity of the investigated parasites in relation to the body weight of two hosts. On the other hand, the differences in the mean intensity values with respect to the body weight of remaining hosts were not statistically significant (P>0.05; Table 23 and Fig. 42).

Statistical analysis (using linear regression, Table. 23) has revealed the presence of a highly significant negative correlation between the mean intensity (infestation level) of *Ergasilus lizae* and body weight of *Valamugil seheli* (*t*-value = -2.529, P = 0.015). A very highly significant negative correlation was indicated between the infestation level of *Lernanthropus kroyeri* and the host body weight *Dicentrarchus punctatus* (*t*-value = -5.197, P = 0.000) and non-significant in other fishes.

Table(23): Regression analysis of the mean intensity of parasitic copepods in relation to host body weight.

Parasitic copepods	**Length classes**	**Mean intensity**	**Beta**	***t*-value**	**p**
Lernanthropus kroyeri	I	1.428(+)	-1.023	-5.197	0.000***
	II	0.112			
	III	0.011(-)			
Lamproglena minuta	I	1.124(+)	0.042	0.207	0.836
	II	0.101			
	III	0.007(-)			
Ergasilus briani	I	1.167(+)	0.283	0.964	0.343
	II	0.068			
	III	0.005(-)			
E.lizae	I	16.344(+)	-0.525	-2.529	0.015**
	II	1.315			
	III	0.145(-)			
E.australiensis	I	9.269(+)	0.219	0.758	0.456
	II	0.726			
	III	0.054(-)			
E.mosulensis	I	2.65(+)	-0.099	-0.18	0.858
	II	0.182			
	III	0.014(-)			
E.celestis	I	10.242(+)	-0.368	-0.814	0.423
	II	0.37			
	III	0.045(-)			
Caligus elongatus	I	8.579(+)	-0.614	-1.335	0.201
	II	0.519			
	III	0.03(-)			

Beta, Regression coefficient; P, probability;**, highly significant; ***, very highly significant; (+) designated for the maximum value and (-) designated for the minimum value.

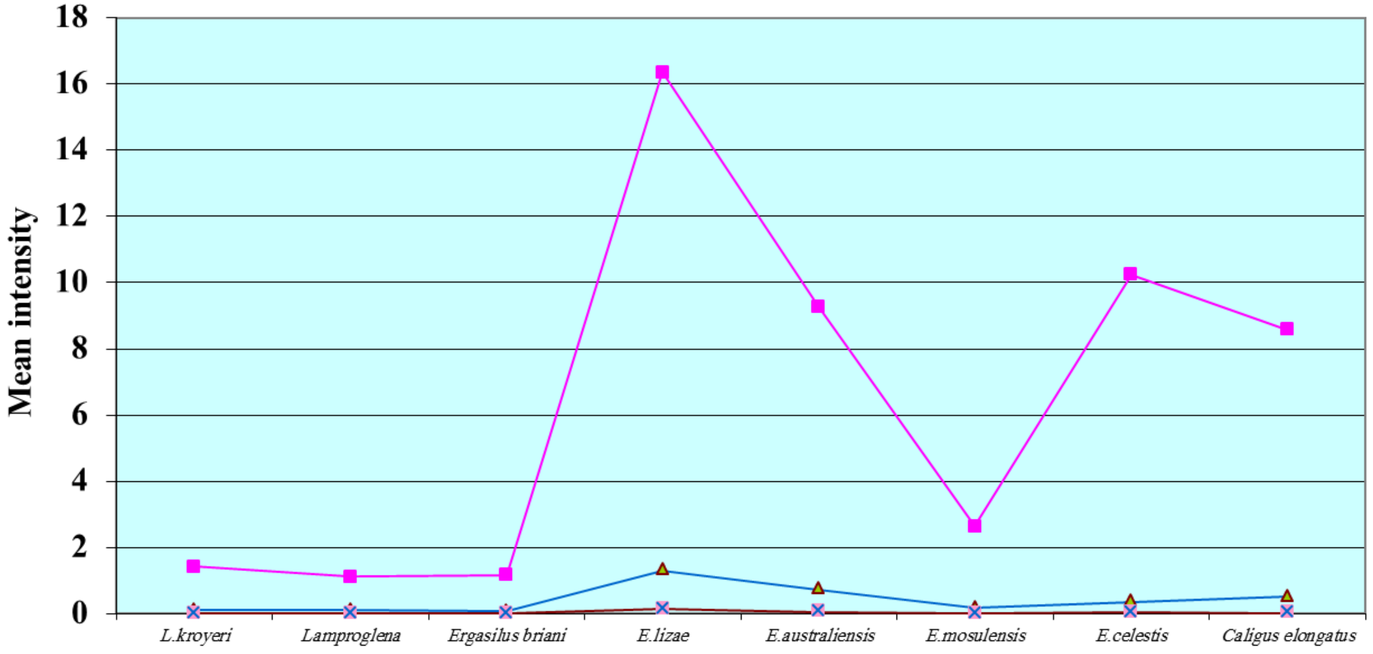
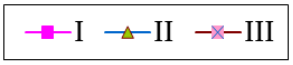


Figure (42): Mean intensity of parasitic copepods on weight classes of their fish hosts.



3. Host sex

Table 24 has revealed that half number of the parasitic copepods prefered male host fish and the other half prefered female host fish.

According to the linear regression (correlations), there is one response pattern was detected among the parasitic copepods *Lernanthropus kroyeri*, *Lamproglena minuta*, *Ergasilus briani*, *Ergasilus lizae***,** *Ergasilus australiensis*, *Ergasilus mosulensis*, *Ergasilus celestis* and *Caligus elongatus* under investigation in relation to body sex of *Dicentrarchus punctatus, Sarotherodon galilaeus, Mugil cephalus, Valamugil seheli, Diplodus sargus, Liza ramada*, *Anguilla anguilla* and *Dicentrarchus labrax* respectively. The response pattern indicated that the infestation level of all copepods and the body sex of their hosts are positively correlated (Table 24and Fig. 43).

Statistical analysis, using *student-t* test, has proofed that mean intensity (infestation level) of *Ergasilus mosulensis* between both sexes of *Liza ramada* was nearly significant (*t*-value = 7.897, P = 0.011) and very highly significant for the rest of the parasite species (P<0.05; Table 24).

Table (24): Statistical analysis of the mean intensity of parasitic copepods infesting females and males of fishes.

Parasitic copepods	**Host name**	**Host sex**	**Mean intensity**	***t*-value**	**±SE**	**P**
Lernanthropus kroyeri	*Dicentrarchus punctatus*	Female	3.26	13.296	0.245	0.000***
		Male	5.94	7.499	0.792	
Lamproglena minuta	*Sarotherodon galilaeus*	Female	4.19	6.981	0.600	0.000***
		Male	2.5	10.683	0.234	
Ergasilus briani	*Mugil cephalus*	Female	0	0	0.000	0.000***
		Male	5.15	8.066	0.639	
Ergasilus lizae	*Valamugil seheli*	Female	20.97	13.422	1.562	0.000***
		Male	26.83	27.157	0.988	
Ergasilus australiensis	*Diplodus sargus*	Female	20	0.000	0.000	0.000***
		Male	10.21	16.643	0.614	
Ergasilus mosulensis	*Liza ramada*	Female	2.2	4.491	0.490	0.011*
		Male	3.55	11.303	0.314	
Ergasilus celestis	*Anguilla anguilla*	Female	17.38	15.576	1.116	0.000***
		Male	11.62	11.487	1.012	
Caligus elongatus	*Dicentrarchus labrax*	Female	21	0.000	0.000	0.000***
		Male	7.88	10.549	0.747	

P, Probability; ±SE, standard error of mean; *, significant; *, very highly significant.**

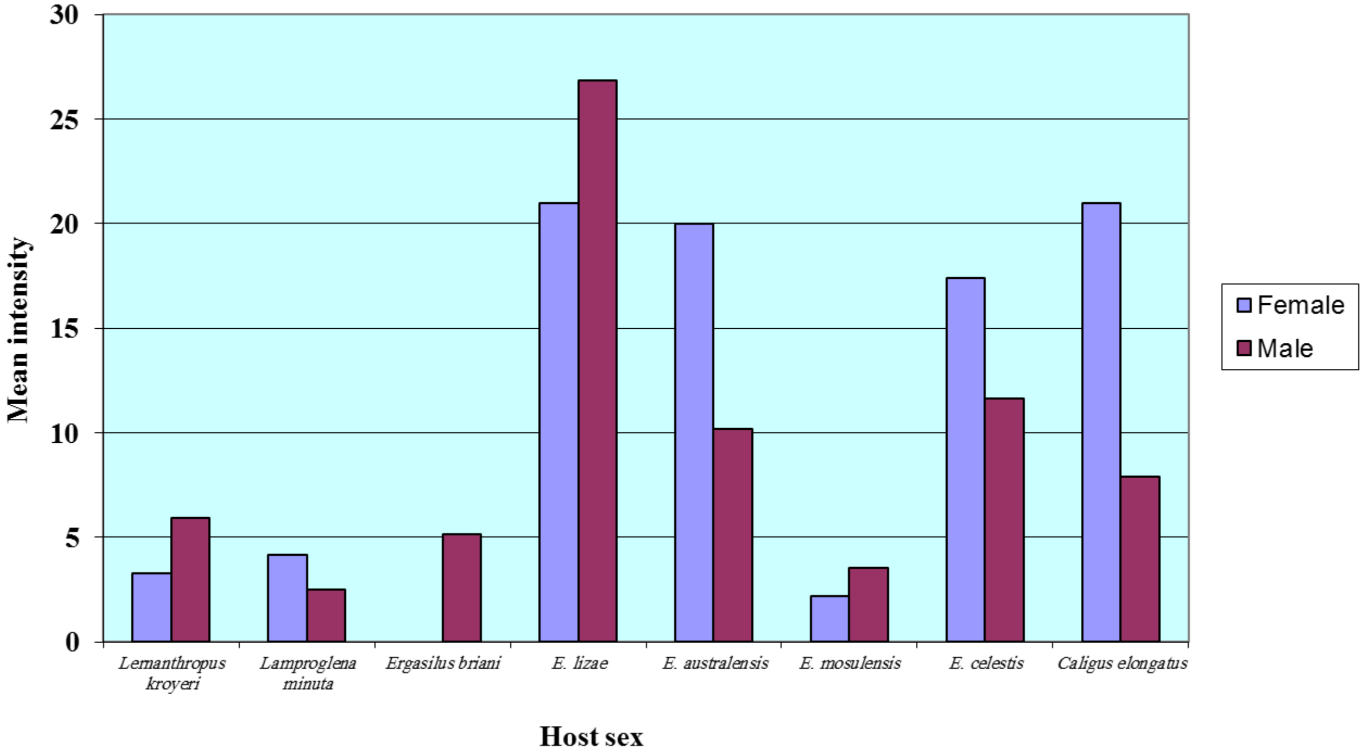


Figure (43): Mean intensity of parasitic copepods in relation to sex of fish hosts.

(V) Discussion:

1. Seasonal changes of the prevalence and mean intensity of parasitic copepods

In the present study, it was found that the parasitic copepods were dominated and have revealed the highest prevalence and mean intensity at site 1 and 2 except *Lamproglena minuta* that was disappeared from these sites during four seasons because this parasite infested the freshwater fish only that collected from site3. Therefore, *Lamproglena minuta* has attained the highest prevalence and mean intensity at site 3 only. **Ramadan *et al.* (1995)** found that the prevalence and the mean intensity with some ectoparasites (*Tricodina* spp., *Dactylogyrus* spp., *Lernaea* spp. and *Ergasilus* spp.) were higher on *Tilapia zillii* than on *Oreochromis niloticus*. The colonization of these parasites is mainly possible via the incoming water providing that the maintenance and hygiene of the farm is good and thus derived from the parasite pool of the lake or river in which the farm has its water supply **(Valtonen and Koskivaara, 1994)**. *Ergasilus sieboldi* and *Triaenophorus nodulosus* had higher prevalence and intensity in the oligotrophic lake than in Lake Vatia. Meanwhile, *Argulus foliaceous* had higher intensity in the Lake Vatia than in the oligotrophic lake **(Halmetoja *et al.*, 1993)**. **Tuuha *et al.* (1992)** compared the prevalence and intensity of *Ergasilus briani* (16.9%, 0.5), *Neoergasilus japonicus* (15.6%, 0.4) and *paraergasilus longidigitus* (2.1%, 0.002) on *Perca fluviatilis* from four lakes in central Finland.

In the present study, the infestation with the parasitic copepods did not maximized during the same season but there are seasonal variation in the prevalence and mean intensity of the most parasitic copepods among the sites of collection. The prevalence and the mean intensity of infestation with most of the parasitic copepods increased in Spring and some of them increased in Autumn and others increased in Summer at site 1. Meanwhile, most of them were prevalent and intense in Summer, some of them were in Autumn and other in Winter at site 2. Clearly, most parasites if preferred, in their distribution, one season at site 1 they can prefer the next season at site 2, except the parasitic copepods; *Lernanthropus kroyeri* and *Ergasilus briani.* Otherwise, *Ergasilus lizae* had the same behaviour in its prevalence like most parasites but had other behaviour in its intensity. On the other hand, existence of the eighth parasite, *Lamproglena minuta* increased in Summer only at site 3 during the present study.

In this respect, **(Rawson, 1977)** reported that the intensity of *Ergasilus lizae* and *Ergasilus versicolor* on *Mugil cephalus* was high in summer but declined in autumn. The intensity of *Caligus elongatus* on cultured Atlantic salmon in the lower Bay of Fundy, Canada was highest in late summer and autumn but lowest in winter **(Hogans and Trudeau, 1989)**. **Ramasamy and Ramalingam (1989)** found that the prevalence, abundance and frequency distribution of *Bicotyle vellavoli* on *Pampus chinensis* were highest than those on *P. argenteus*. They supposed that the differences in the host susceptibility to infection or the ecological and behavioral differences which render *P. argenteus* less liable to infection may be a logical interpretation. *Lepeophtheirus pectoralis* has its greatest prevalence (>95%) and intensity (5 parasite/fish) on flat fish during summer **(Boxshall, 1974)**. **Tully (1989)** found that the total intensity of the parasitic copepod, *Lepeophtheirus pectoralis*, did not rise cumulatively because mature lice disappeared from fish before maturation of the next generation was complete. Adult females from winter generations are significantly larger, produce larger egg sacs and more but smaller eggs than in summer **(Ritchie *et al*., 1993)**.

Similar findings were reported by **Voth (1972)** who found that the prevalence of parasitic copepod, *Lepeophtheirus hospitalis*, on *Platichthys* *stellatus* was increased in summer, decreased with the winter rain period and increased again in spring with the migration of other infected *Platichthys stellatus*. These phenomena occur in monogeneans which are also ectoparasites. **Ruangpan and Kabata (1984)** recorded highest intensity *Caligus epidemicus* on the telson and legs of the penaeid prawn, *Penaeus monodon*. **Byrnes and Rohde (1992)** found prevalence of *Caligus epidemicus* on four species of marine bream, *Acanthopagrus australis, A. berds, A. butcheri* and *A. latus*. **Batra (1984)** reported that the monogenean gill parasites, *Cichlidogyrus* spp., were the most numerous on *Tilapia rendalli*, *O. macrochir* and *O. andersonii*. **Landry and Kelso (1999)** recorded that the ectoparasites of *Micropterus salmoides* have attained higher prevalence and mean intensity than those of the endoparasites (trematoda, acanthocephala and cestoda).

Seasonal variation and prevalence and mean intensity of parasitic copepods that reported in the present investigation in the three sites may be considered as low in a selective breeding context. Genetic variation, potential biological co-factors and the role of the immune system in the defence against infection with parasitic copepods require further attention if fish hosts are to be selectively bred to reduce their susceptibility to infection with parasitic copepods. This variation of infection in fish hosts has been subjected to studies by many authors **Glover *et al.* (2005)** and **Glover *et al.* (2004a, b).** They reported that *Lepeophtheirus salmonis* and *Caligus elongatus* giving variation in abundance and mean intensity among different seasons and they proved that two caligid copepods may be considered as low in a selective breeding context. This variation also may be attributed to the water temperature, where the high temperature favours a short incubation period, rapid growth and high egg production and on the other hand, the temperature may not be the only controlling factor in the population dynamics of these parasites. Many suggestions have been proposed on seasonal infection of fishes **Hoglund (1990)** and **Abu Samak (1995)**. **Hoglund (1990)** attributed the seasonal variation of infection in fishes to the changes of temperature as the most factor affecting the dynamics of fish infection. On the other hand, **Abu Samak (1995)** did not agree- to some extent – with **Hoglund (1990)** in considering, the temperature as the only most important factor affecting the dynamics of fish infection. She found that the temperature is not the most relevant factor affecting in fish infection where low and high prevalence of infection occurred in the same season.

2. The parasitic copepods and environmental parameters

In the present study, statistical analysis has indicated that the relationship between the infestation level of all parasitic copepods at all localities was negatively correlated with the measured water parameters (Temperature, pH, Salinity and Oxygen content). This correlation was very highly significant for all parasitic copepods.

Temperature was reported, by many authors, to be an important factor that determines the population growth of parasitic copepods. **Hogans (1989)** found that the generation time of *Ergasilus luciopercarum* increased sharply with decreasing temperature (6ºC). Females of *Ergasilus sieboldi* and *Ergasilus labracis* infections occur during winter at 5-8 ºC **(Abdelhalim *et al.*, 1991)**.

The life cycle of *Ergasilus sieboldi* and *Neoergasilus japonicus* consist of six nauplii and five copepodid stages and one adult stage. Sexual differentiation is evident in the fourth copepodid stage **(Zmerzlaya, 1972; Abdelhalim *et al.*, 1991)**. It takes 22 days for the eggs of *Ergasilus sieboldi* to develop into free-living males and females at 7.7-10.1ºC **(Zmerzlaya, 1972)**. The generation time of *Caligus elongatus* and *Caligus pageti* is 5 weeks at water temperatures ranging from 5-7 ºC in the Bay of Fundy **(Roubal, 1990)**.

In the present investigation, the pH-values were found to be in the acidity scale at all sites. However, the water body at site 1 and 2 are obviously more acidity than that at site 3. It is thought that the high pH-values of hydrogen at site 1 and 2 could exert an additional stress on fish hosts as well as their parasitic copepods. Moreover, the mean intensity of all parasitic copepods was found to be negatively correlated with the pH-values. This phenomenon occurs also in monogeneans which are also ectoparasites. In this respect, **Singhal *et al.* (1986)** recorded a positive relationship between the infestation level of *Gyrodactylus* sp. and the pH-value. **Said (2002)** suggested that the monogenean parasites, *D. extensus,* can tolerate relatively low pH-values and they are acclimatized in a considerable manner to tolerate this condition.

Experimental studies on the effects of alkaline solutions have indicated that damage to fish generally begins at pH 9.0 (**Whitaker, 1982; Hartwell *et al*., 1986**) and pH-value up to 10.0 was reported to cause fish mortalities in freshwater environment (**Jordon and Lloyd, 1964**). Therefore, the parasitic copepods may prefer the acidic environment than the alkaline one. Or some individuals can tolerate with the acidic environment and others can tolerate with the alkaline one.

Statistical analysis in the present study has indicted that the relationship between the water salinity and the mean intensity of the copepodian parasites was negatively correlated at all sites. It is obviously noticed that these parasitic copepods are less tolerant (sensitive) to salinity changes.

The results of the present study seemed to be negative impacts of chlorides on the survival and population growth of ectoparasites of fishes that previously reported by some authors (for example, **Chan and Wu, 1984; Buchmann *et al*., 1987; Crespo *et al*., 1995)**. **Chan and Wu (1984) and Buchmann *et al.* (1987)** found that the 1ong-term treatment with sodium chloride reduced the intensity of infection of the European eel, *Anguilla anguilla*, by the monogeneans, *Pseudodactylogyrus anguillae* and *P. bini*. **Crespo *et al.* (1995)** evaluated the chemotherapeutic drugs against cichlidogyriasis in tilapia fish, *Oreochromis niloticus*, in Mexico. The authors found that sodium chloride was highly efficient in the control of *Cichlidogyrus* infestation.

In contrast to the present study, the sensitivity of parasitic copepods to salinity was previously recorded. *Ergasilus labracis* infected more than 90% of striped bass, *Morone saxatilis*, from the lower Chesapeake Bay in salinities from 0.1 to 32 ppt. *Ergasilus lizae* that has been found on eels, tilapia, carp as well as mullet in aquaculture ponds in Israel, occurred in salinities from 0.2 to 21 ppt **(Paperna, 1975, 1977 and 1991)**. *Lepeophtheirus salmonis* is absent from sites with lowered salinity. Farms in Norway are in sites with a consistently higher salinity than in Scotland and have more severe *L. salmonis* infections **(Pike, 1989)**. An epizootic of *Caligus epidemicus* within the lower Mitchell River, Victoria, Australia caused the death of bream, *Acanthopagrus butcheri,* and mullet, *Mugil cephalus*, *Aldrichetta forsteri*, *Liza argentea* and *Myxus elongatus*; the epizootic was associated with high salinity up to 28 ppt **(Hewitt, 1971)**. An epizootic of *Caligus pageti* on a fish farm in Egypt was also associated with a high salinity up to 45 ppt **(Hewitt, 1971)**.

Also, several studies demonstrated that salinity of water can influence the occurrence of adult monogeneans in different water bodies for example, **El-Naggar and Khidr (1988)** reported the absence of the monogenean gill parasite *Cichlidogyrus nematocirrus* from the gills of cichlid hosts inhabiting the River Nile at Mansoura, whereas, the same species was well represented in respective hosts in the River Nile at Damietta. They suggested that the parasite may have a preference for higher salinity at Damietta. **Kennedy, (1975)** reported that the estuarine habitat reflects the lowest level of infection of the monogenean *P.*

anguillae on the European eel, *A. anguilla*. **Paperna (1979)** studied the pattern of distribution of cichlid monogeneans in inland water in Africa and found that the infestation level in fish ponds as well as in smaller water habitats was much higher than that in lake environments. **El-Naggar (1994)** studied the occurrence of five monogenean species on the gills of the catfish *Clarias lazera* at Mansoura, Canal and Manzala Lake and reported that the relatively higher salinity values at Manzala Lake may inhibit or reduce egg production, egg hatching and growth rates of the infective stages (oncomiracidia) and/or may cause weakness or mortality of the adult monogeneans. Under these conditions, the activities of the parasites are diminished and their population growth is declined.

Statistical analysis in the present study has indicted that the relationship between the oxygen content of water and the mean intensity was negatively correlated with all parasitic copepods at all sites. It is obvious that these copepods are less tolerant (sensitive) to oxygen of the water.

These phenomena occur also in monogeneans which are ectoparasites. **Said (2002)** found *D. anchoratus* is less tolerant (sensitive) to change in the oxygen content of the water. This coincides with the observations made by **Izyumova (1958)** who showed that a fall in the amount of dissolved oxygen significantly reduced the population of *D. extensus* in Russia**.** Few laboratory investigations have been conducted to study the relationship between the activities of monogenean parasites and oxygen content in the ambient water **(Kearn, 1962; Arme and Fox, 1974; Houlihan and Macdonald, 1979).** Movements, clamp activities and egg production of adult parasites seem to be relying on adequate level of oxidative phosphorylation, one of the metabolic mechanisms by which the parasite can make use of the digested food and release the energy required for different cellular activities **(Kearn, 1962; Arme and Fox, 1974; Houlihan and Macdonald, 1979)**. **Kearn (1962) and Houlihan and Macdonald (1979)** found that the skin monogenean, *Entobdella soleae*, can maintain egg production and oxygen uptake in the face of falling oxygen partial pressure by increasing its ventillatory movements, by increasing its surface area and by reducing its thickness.

Low environmental oxygen levels are also detrimental to the host and this stress together with the burden imposed by parasites may lead to host death. **Overstreet (1993)*** identified low oxygen content of water as the most important stress factor contributing to massive fish mortality caused by the protozoan *Amyloodinium ocellatum*. **Schaperclaus (1954)*** suggested that fish could tolerate infections of high mean intensity when oxygen was freely available, but the survival chances of the fishes were reduced by the decreased availability of oxygen. **Kotlán and Kobulej (1972)* and Molnár (1994)** pointed out that adverse effects of oxygen depletion on the host are amplified when the fish are heavily infected with *D. vastator*. **Paperna (1964)** reported a substantial reduction in the respiratory surface of the gills as a result of infection with *D. vastator* and **Uspenskaya (1961)** noted that such fish take up half as much oxygen.

3. Parasitic copepods and host ageing

All fish species in the present study show strong preference to smaller fishes rather than larger fishes. This preference is significant in *Lernanthropus kroyeri*, *Ergasilus lizae* and *Caligus elongatus* but not significant in other species. *Lernanthropus kroyeri*, *Lamproglena minuta* and *Ergasilus briani* are particularly common on small fishes class I (standard length, from 3 to 17cm and body weight, from 0 to 300gm), while *Ergasilus lizae, Ergasilus australiensis*, *Ergasilus mosulensis*, *Ergasilus celestis* and *Caligus elongatus* are particularly common on small fishes (class I) and also on longer fishes (class II and III standard lengths from 17.1 to 50cm and body weight, from 300.1 to 900gm). Moreover, *Lernanthropus kroyeri* on *Dicentrarchus punctatus*; *Ergasilus lizae* on *Sarotherodon galilaeus* and *Caligus elongatus* on *Dicentrarchus labrax* have attained their maximum mean intensity values on fishes of length class I and their lowest values have been recorded on fishes of length class II and III.

These phenomena occur in monogeneans which are also ectoparasites. **Said (2002)** found a negative relationship between infection with monogenean parasites, *Dactylogyrus anchoratus* and the size of the host, *Cyprinus carpio*. He suggested that the development of immunity with advancing age is the reason for the corresponding decline of *D. anchoratus*. Also, he claimed that an immune response to *D. anchoratus* must be highly specific. His suggestion was supported by **Noisy and Maillard (1980)** whom noticed a negative relationship between infection *Microcotyle chrysophrii* and size of the host *Sparus aurata*. They attributed such a negative relationship to the fact that immunity of the host develops with increasing age as well as with the growing mean intensity of the parasites. Also, a significant positive correlation between fish size of Atlantic salmon (*Salmo salar*) have been reported to the increase in intensity and abundance of sea lice, *Caligus elongatus* and *Lepeophtheirus salmonis* **(Glover *et al.*, 2001, 2003, 2004 a, b; Tucker *et al*., 2002)**. They suggested that the fish size (body length and weight) influences sea lice infection level. This is in accordance with the findings of **Roubal (1987)** during his study on the effect of *Ergasilus* sp. on the host *Acanthopagrus australis*. He found that the relative pathogenicity is reduced when gill filament length is converted to surface area. This reduction is greater in smaller fishes because they have a smaller surface area per unit length of filament than larger fishes. This suggestion has received support from **Rizvi (1969), Anderson (1974), Ramasamy *et al.* (1985), Silan and Maillard (1989, 1990)** whom proposed that the larger fishes display a largest gill surface area and this will increase in the biotic capacity and thus resources for this parasite. **Stromberg and Crites (1975)** showed that the prevalence and intensity of

Camallanus oxycephalus on *Morone chrysops* increased with host size. **Hagras *et al.* (1995)** reported that the intensity of *Quadriacanthus* spp. and *Paraquadriacanthus nasalis* infesting *Clarias lazera* gradually increases and reaches a higher level on Medium-sized fishes and declines on larger fishes. They suggested that the host *C. lazera* acquires immunity against these parasites as it gets older. **Anderson (1974)** reported a close positive correlation between the host age and thus size of *Abramis brama* and number of *Diplozoon paradoxum*. He proposed that such a positive relationship most probably due to high density of hosts and the enclosed nature of the habitat which increasing the chance of encountering an infective larval parasites. **Srivastava and James (1967)** attributed such a positive relationship to the long term exposure of older fishes to parasites or their more gregarious or sluggish habits which facilitate the transfer of parasites from one individual to another. **Buchmann (1989)** found a significant positive correlation between total body length of the host *Anguilla anguilla* and the load of *Pseudodactylogyrus anguillae*. He suggested that the larger numbers of *P. anguillae* on the gills of *A. anguilla* may be related to the increased volume of water passing over the gills of larger eels, which will increase the probability of contact with more larvae and hence result in a higher infection level. There are other examples of a positive correlation between parasite numbers and host size. **Euzet and Marc (1963)** found that only larger specimens of *Labrus bergylta* were infected with the *Microcotyle donavini*. **Joy (1988)** found a positive correlation between numbers of *Microcotyle spinicirrus* and increasing length of *Aplodinotus grunniens*. Large hosts carried significantly more parasites than smaller ones.

Similar results were observed by **Cope (1959)** for *Ergasilus turgidus* from *Gasterosteus aculeatus*, by **Tedla and Fernando (1970)** for *Ergasilus confusus* from *Perca fluviatilis,* by **Zmerzlaya (1972)** for *Ergasilus sieboldi* from several fish species, by **Bortone *et al*. (1978)** for *Bomolochus concinnus* and *Ergasilus manicatus* from *Menidia beryllina* and *M. peninsule,* by **Ramasamy *et al*. (1985)**

for *Vallisia indica* and *Allodiscocotyla chorinemi* from *Scomberoides commersonianus*, by **Gonzalez - Lanza *et al.* (1991)** for *Diplectanum aequans* and *D. laubieri* from *Dicentrarchus labrax*, by **Ramadan *et al.* (1995)** for *Lerneae*, *Ergasilus* and *Dactylogyrus* from *Tilapia* spp., by **Jaworski and Holm (1992)** for *Caligus elongatus* and *Lepeophtheirus salmonis* from *Salmo salar*, by **Hudson *et al*. (1994*)*** for *Ergasilus nerkae* from *Pungitius pungitius*, *Gasterosteus aculeatus* and *Prosopium cylindraceum*, by **Moravec and Scholz (1994**) for *Neoechinorhynchus rutili* from *Barbus barbus*, by **Aragort *et al.* (1997)** for *Cichlidogyrus* spp. from *Oreochromis mossambicus*, by **Aloo (1999**) for *Contraceacum sp*. larva from *Micropterus salmoides*, by **Gonzalez and Acuna (2000)** for *Anisakis sp*.from *Sebastes capensis*, by **Muzzal and Bowen (2000*)*** for *Echinorhynchus salmonis* from *Salvelinus namaycush*, by **Valles-Rios *et al.* (2000*)*** for *Ergasilus versicolor* and *Contracaecum multipapillatum* from *Mugil cephalus*, by **Said (2002*)*** for *Dactylogyurs extensus* from *Cyprinus carpio* and by **Manera and Dezfuli (2003)** for *Lernanthropus kroyeri* from *Dicentrarchus labrax*.

4. Parasitic copepods and host sex

In the present study, individuals of *Lamproglena minuta*, *Ergasilus australiensis*, *Ergasilus celestis* and *Caligus elongatus* were found to prefer female than male fishes, whereas individuals of *Lernanthropus kroyeri*, *Ergasilus briani*, *Ergasilus lizae* and *Ergasilus mosulensis* were found to prefer male than female fishes. Such phenomenon of sex preference was explained by **(Glover *et al.*, 2001, 2003, 2004a, b; Tucker *et al.*, 2002)** whom showed that there is an effect of fish sex on the abundance of *Caligus elongatus* and *Lepeophtheirus salmonis.* They observed significant differences in abundance of *Caligus elongatus* and *Lepeophtheirus salmonis* to females than males of the Atlantic salmon (*Salmo salar*). **Grimnes and Jakobsen (1996)** found that the abundance of parasitic copepod *Lepeophtheirus salmonis* on females *Salmo salar* was significantly higher than on males. The monogeneans *Gyrodactylus stephanus*, *G. prolongis*, *Urocleidus angularis* and *Fundulotrema prolongis* from the killifish *Fundulus heteroclitus* were found to be more common on females than on males **(Barkman and James, 1979)**. In the case of schistosomes, the exposure of male and female mice of a variety of strains to equal numbers of cercariae resulted in a more severe clinical course and higher mortalities in females than in males (**Mendonça *et al*., 2000**). Similar findings were reported by **Ramadan *et al.* (1995*)*** for *Dactylogyrus* spp., *Ergasilus* spp. and *Acanthosentis* spp. from *Oreochromis niloticus*, *Sarotherodon galilaeus* and *Tilapia zillii*, by **Alloo (1999)** for *Contracaecum* sp. larvae from *Micropterus salmoides* and by **Said (2002)** for *Dactylogyrus extensus* from *Cyprinus carpio*.

Moreover, in the present work, there is a tendency of the parasitic copepods *Lernanthropus kroyeri*, *Ergasilus briani*, *Ergasilus lizae* and *Ergasilus mosulensis* to prefer male than female fishes. Such phenomenon of sex preference was explained by **Marques and Hogans (1996)** reported that abundance of *Ergasilus centrarchidarum* on males *Microgadus tomcod* was significantly higher than on females. **Lees and Bass (1960)** and **Thomas (1964)** found that at least one monogenean parasite, *Polystoma integerrimum*, exhibits a significant preference for one sex of host. The preference of the monogenean parasites for male hosts was previously recorded (**Paling, 1965; Tinsley, 1989; Appleby, 1996)**. **Paling (1965)** found that males of *Salmo trutta* are more heavily infected with the monogenean parasite *Discocotyle sagittata* than females. **Tinsley (1989)** found a similar phenomenon in a North American desert toad, *Scaphiopus couchii*, infected by the monogenean *Pseudodiplorchis americanus*. Also, **Appleby (1996)** found that the abundance of the monogenean *Gyrodactylus* sp. on male sand gobies, *Pomatoschistus minutus*, was significantly higher than on females. Other authors found that male hosts are heavily parasitized than female hosts; as reported by **Shotter (1973)** in *Diclidophora merlangi* from *Odontogadus merlangus,* by **Pickering and Christie (1980)** in *Gyrodactylus* spp. from *Salmo trutta,* by **Saoud and Hassan (1983)** in cestode *Anthobothrium* spp. from *Dasyatis uarnak* and by **Zharikova (1984)** in *Dactylogyrus* spp. from *Abramis brama*.

In summary total prevalence of *Lernanthropus kroyeri* was 64% and its mean intensity was 2.80 parasite/infested fish, total prevalence of *Lamproglena minuta* was 73% and its mean intensity was 0.68 parasite/infested fish, total prevalence of *Ergasilus briani* was 43% and its mean intensity was 0.73 parasite/infested fish, total prevalence of *Ergasilus lizae* was 52% and its mean intensity was 8.76 parasite/infested fish, total prevalence of *Ergasilus australiensis* was 72% and its mean intensity was 4.17 parasite/infested fish, total prevalence of *Ergasilus mosulensis* was 41% and its mean intensity was 0.79 parasite/infested fish, total prevalence of *Ergasilus celestis* was 64% and its mean intensity was 4.20 parasite/infested fish and total prevalence of *Caligus elongatus* was 58% and its mean intensity was 2.16 parasite/infested fish. The population growth of all parasitic copepods at the three sites in the present study increased with decreasing temperature, the pH-values were found to be in the acidity scale at all sites and the mean intensity of all parasitic copepods was found to be negatively correlated with the pH-values, the survival and population growth of all parasitic copepods at all sites also, was less tolerant (sensitive) to salinity changes and all parasitic copepods at all sites are less sensitive to oxygen of the water. The mean intensity of all parasitic copepods at all sites in the present study show strong preference to smaller fishes rather than larger fishes. Also, half number of all parasitic copepods prefered male host fish and the other half prefered female host fish.

CHAPTER III

Histological
And
Histopathological Studies

Histopathological changes of the gill lamellae

1. Results:

a. Normal histology of the gills:

The collected fish hosts; *Dicentrarchus punctatus*, *Sarotherodon galilaeus*, *Mugil cephalus*, *Valamugil seheli*, *Diplodus sargus*, *Liza ramada*, *Anguilla anguilla* and *Dicentrarchus labrax* have four gills on each side of their head. Each gill consists of a rigid gill arch from which project two rows of primary gill lamellae, the inner and outer hemibranchs. Each of the two surfaces of the primary gill lamella bears a single row of plate-like secondary gill lamellae which are thin-walled and represent the site of gaseous exchange between the contained blood and the water flowing through the gill. There are mucous cells on some parts of the secondary gill lamella and its basal epithelium. The primary gill lamella is supported by a median cartilaginous ray which runs along its longitudinal axis (Figs. 44, 47, 50, 52, 55, 57, 60 and 62).

Light microscope examinations of sections stained with haematoxylin and eosin showed that the primary gill lamellae are covered with a stratified epithelium which is continuous over the margins and tips but interrupted where the secondary gill lamellae project. The stratified epithelium is mainly composed of small and large cuboidal cells, each possessing a central nucleus (Figs. 44, 47, 50, 52, 55, 57, 60 and 62).

b. Parasitic infestations and histopathological changes of the gill lamellae:

1. *Lernanthropus kroyeri*:

At the light microscope level, lesions were observed with female and male *Lernanthropus kroyeri* with *Dicentrarchus punctatus* gill filaments. Typically these growths were most pronounced about the anterior of the copepod's cephalothorax and especially just anterior to the frontal plates. The subchelate maxillipeds were also used to grasp the host (Fig. 44).

A short distance from attached female and male *Lernanthropus kroyeri* the surface of the interbranchial septum appeared smooth and normal (Fig. 44 and 45). Therefore, attached female and male *Lernanthropus kroyeri* were either not associated with any gross epithelial lesions or attached within shallow ulcerations. Gut contents of both female and male *Lernanthropus kroyeri* contained host cells and cellular debris, including red blood cells, inflammatory cells and epithelial cells. The papillomatous growths associated with the attachment of female *Lernanthropus kroyeri* were characterized by marked epithelial hyperplasia with disorganization of epithelial layers, degeneration, necrosis and massive loss of filament tissues (Fig. 44).

The attachment of male *Lernanthropus kroyeri* was associated with microscopic lesions marked by a sub-acute inflammatory response. The epidermis at the margins of the ulcers has moderately hyperplasia, degeneration, necrosis and massive loss of filament tissues (Fig. 45).

2. *Lamproglena minuta*:

The attachment of female *Lamproglena minuta* was associated with microscopic lesions marked by a sub-acute inflammatory response. *Lamproglena minuta* causes epithelial hyperplasia with disorganization of epithelial layers, degeneration, necrosis and massive loss of filament tissues in gill filaments of the tilapia fish *Sarotherodon galilaeus* (Fig. 48). In figure (49), lesions associated with female *Lamproglena minuta* were compatible with necrosis of secondary gill lamellae and massive loss of filament tissues. Adult female of *Lamproglena minuta* firmly attach themselves, feed and produce embryo sacs (Fig. 49).

3. *Ergasilus briani*:

The attachment of female *Ergasilus briani* was associated with microscopic lesions marked by a sub-acute inflammatory response. Lesions associated with female *Ergasilus briani* appeared with necrosis of secondary gill lamellae and massive loss of filament tissues. Hyperplasia was not observed in association with female *Ergasilus briani* (Fig. 51). At the light microscope level, the crustacean parasite (*Ergasilus briani*) exerts a compression against the gill tissue at the site of attachment to the gill lamellae of *Mugil cephalus*.

4. *Ergasilus lizae*:

Figure (53) revealed that females *Ergasilus lizae* were associated with microscopic lesions marked by a sub-acute inflammatory response. The epidermis at the margins of the ulcers was moderately hyperplastic and infiltrated with few heterophilic granulocytes. Lesions associated with females *Ergasilus lizae* were compatible with epithelial hyperplasia, necrosis and massive loss of filament tissues in gill filaments of *Valamugil seheli* (Fig. 53).

At the light microscope level, female *Ergasilus lizae* was found to induce minor histopathological changes at the site of attachment of the gill lamellae. These changes include degeneration and hyperplasia (Fig. 54).

5. *Ergasilus australiensis*:

The attachment of female *Ergasilus australiensis* was associated with microscopic lesions marked by a sub-acute inflammatory response. *Ergasilus australiensis* causes epithelial hyperplasia, necrosis and infiltration of macrophages, lymphocytes and eosinophils in gill filaments of the sea bream *Diplodus sargus* (Fig. 56). At the light microscope level, the crustacean parasite (*Ergasilus australiensis*) exerts a compression against the gill tissue at the site of attachment to the gill lamellae of *Diplodus sargus*.

6. *Ergasilus mosulensis*:

A lesion associated with female *Ergasilus mosulensis* was compatible with necrosis of secondary gill lamellae and massive loss of filament tissues (Fig. 58).

In figure (59), female *Ergasilus mosulensis* was found to induce histopathological changes. There is extensive gill damage and severe haemorrhage with inflammation and exsanguinations associated with the attachment and feeding of the parasite. Blood vessels in the gill filaments are blocked and this leads to atrophy of gill tips. Lesions appeared as hyperplasia, degeneration, necrosis and massive loss of filament tissues (Fig. 59).

7. *Ergasilus celestis*:

At the light microscope level, female *Ergasilus celestis* was found to induce histopathological changes at the site of attachment of the gill lamellae of *Anguilla anguilla*. Occasionally, only residues of the attachment string were observed deeply embedded into the submucosa causing proliferation of gill epithelium that chronic and up to four mucous cell layers found on the surface of epithelium (Fig. 61). An additional type of lesion was observed of *Ergasilus celestis* consisting of eosinophilic inclusions in an enlarged epithelium, especially at the base of secondary lamellae and on the tips of the primary lamellae (Fig. 61).

Through the piercing action by means of its cephalothoracic appendages especially, that have terminal claw (second antennae). These deep penetrations of this appendage into the gill tissue produce major histopathological changes include chronic proliferation of primary gill epithelium, epitheliocystis-likeinclusions and massive loss of filament tissues (Fig. 61).

8. *Caligus elongatus*:

At the light microscope level, male *Caligus elongatus* was found to induce minor histopathological changes at the site of attachment of the gills of *Dicentrarchus labrax*. These changes include degeneration, hyperplasia and massive loss of filament tissues (Fig. 63). Through the piercing action by means of its cephalothoracic appendages especially, that have terminal hook (maxillipeds). These deep penetrations of this appendage into the gill tissue produce major histopathological changes include hyperplasia, degeneration and massive loss of filament tissues (Fig. 63).

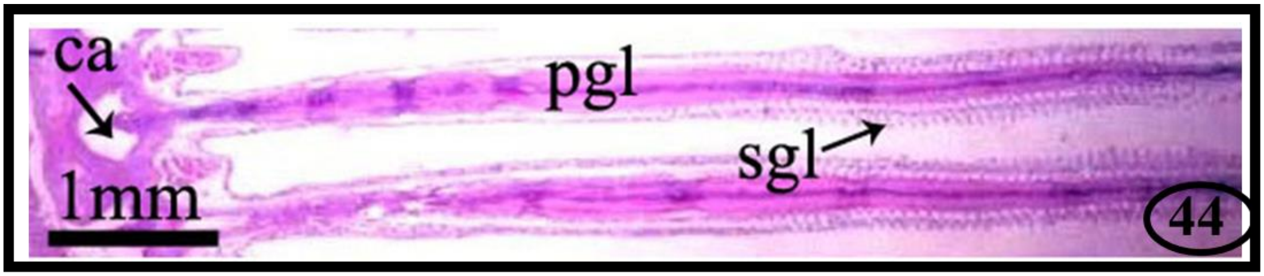


Figure 44: Photomicrograph of a paraffin wax section, stained with haematoxylin and eosin, through two primary gill lamellae of non-infested spotted seabass, *Dicentrarchus punctatus* showing the normal histology. ca, cartilage, pgl, primary gill lamella and sgl, secondary gill lamella.

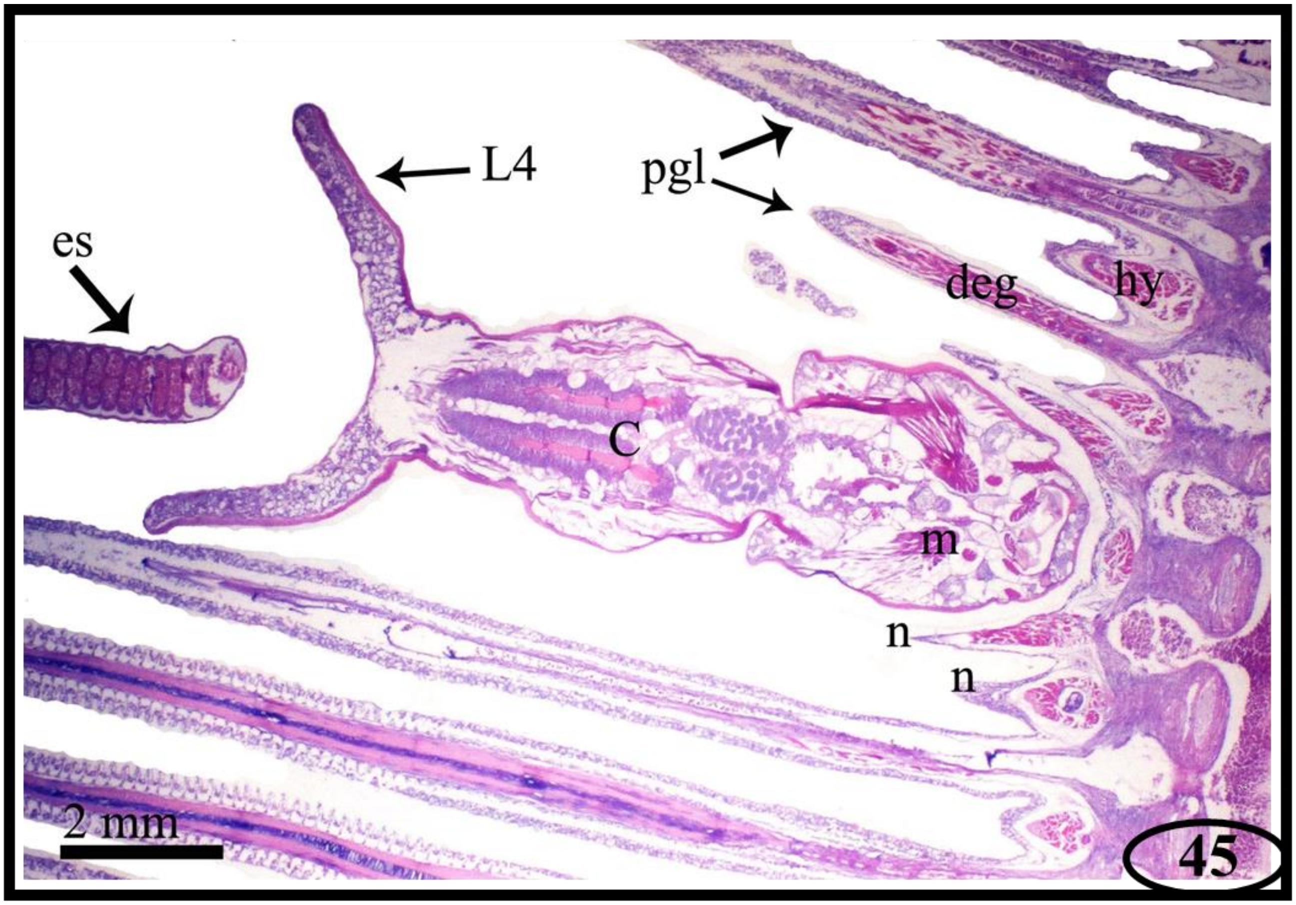


Figure 45: Photomicrograph of a paraffin wax section, stained with haematoxylin and eosin of a gill arch of spotted seabass, *Dicentrarchus punctatus* infested with female copepod *Lernanthropus kroyeri* (c) causing degeneration (deg), hyperplasia (hy), necrosis (n) of the primary gill lamellae (pgl)by using the maxilliped (m), egg sac (es) and 4th leg (L4).

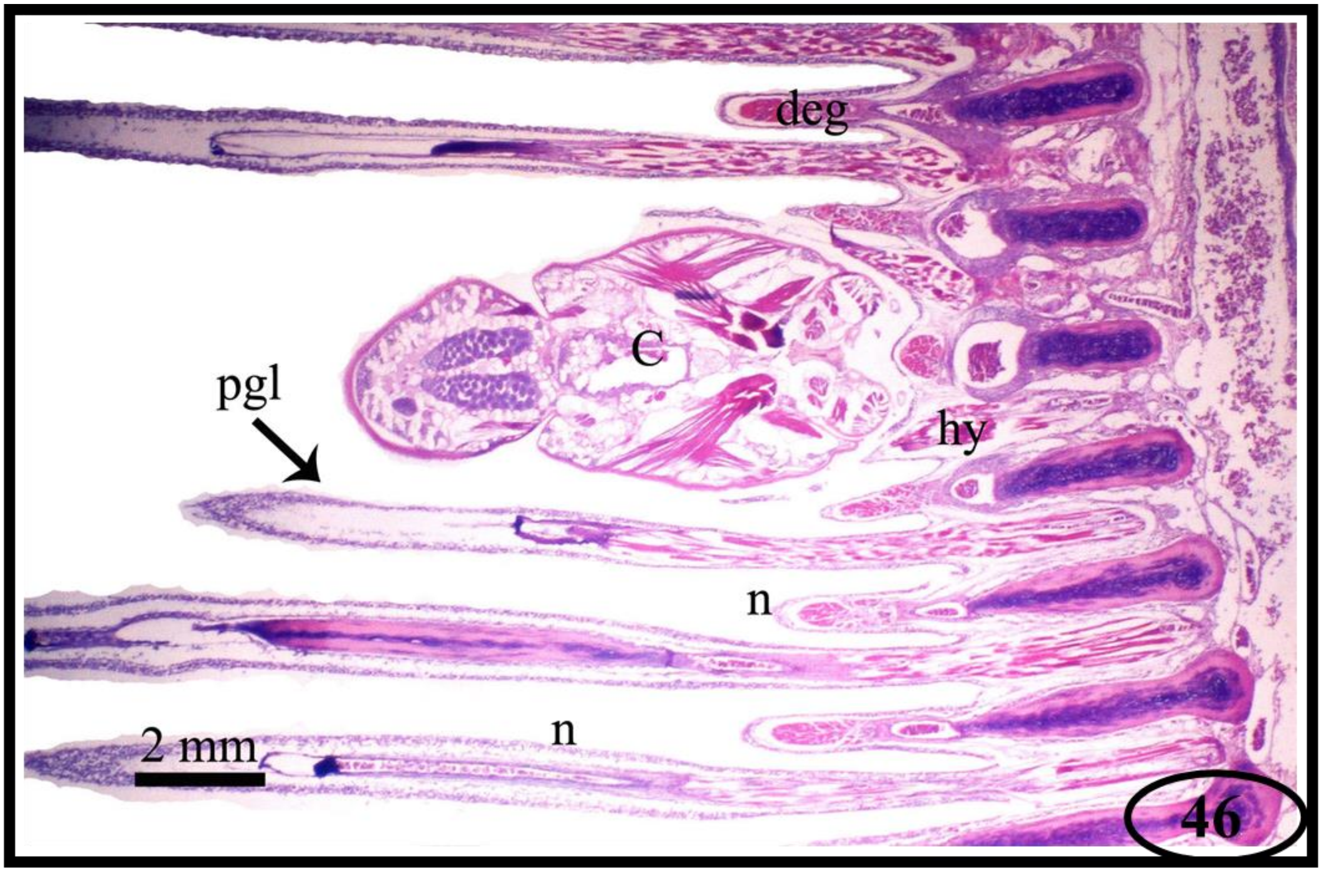


Figure 46: Photomicrograph of a paraffin wax section, stained with haematoxylin and eosin of a gill arch of *Dicentrarchus punctatus* infested with male copepod *Lernanthropus kroyeri* (c) causing degeneration (deg), hyperplasia (hy) and necrosis (n) of the primary gill lamella (pgl).

Figure 47: Photomicrograph of a paraffin wax section, stained with haematoxylin and eosin through a gill arch of non-infested *Sarotherodon galilaeus* showing the normal histology. ca, cartilage, pgl, primary gill lamella and sgl, secondary gill lamellae.

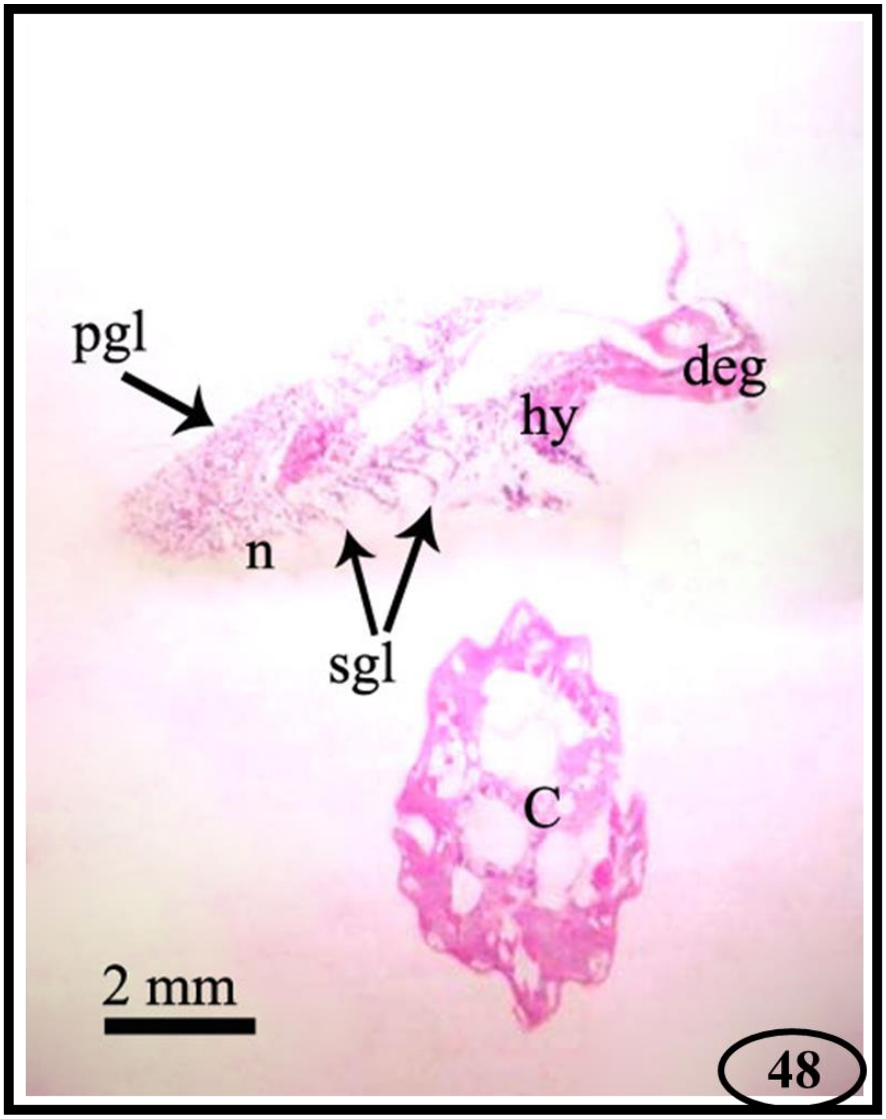


Figure 48: Photomicrograph of a paraffin wax section, stained with haematoxylin and eosin of a primary gill lamella of tilapia galilae, *Sarotherodon galilaeus* infested with a copepodian *Lamproglena minuta* (c) causing degeneration (deg) and hyperplasia (hy) of the primary gill lamella (pgl), necrosis (n) of the secondary gill lamellae (sgl).

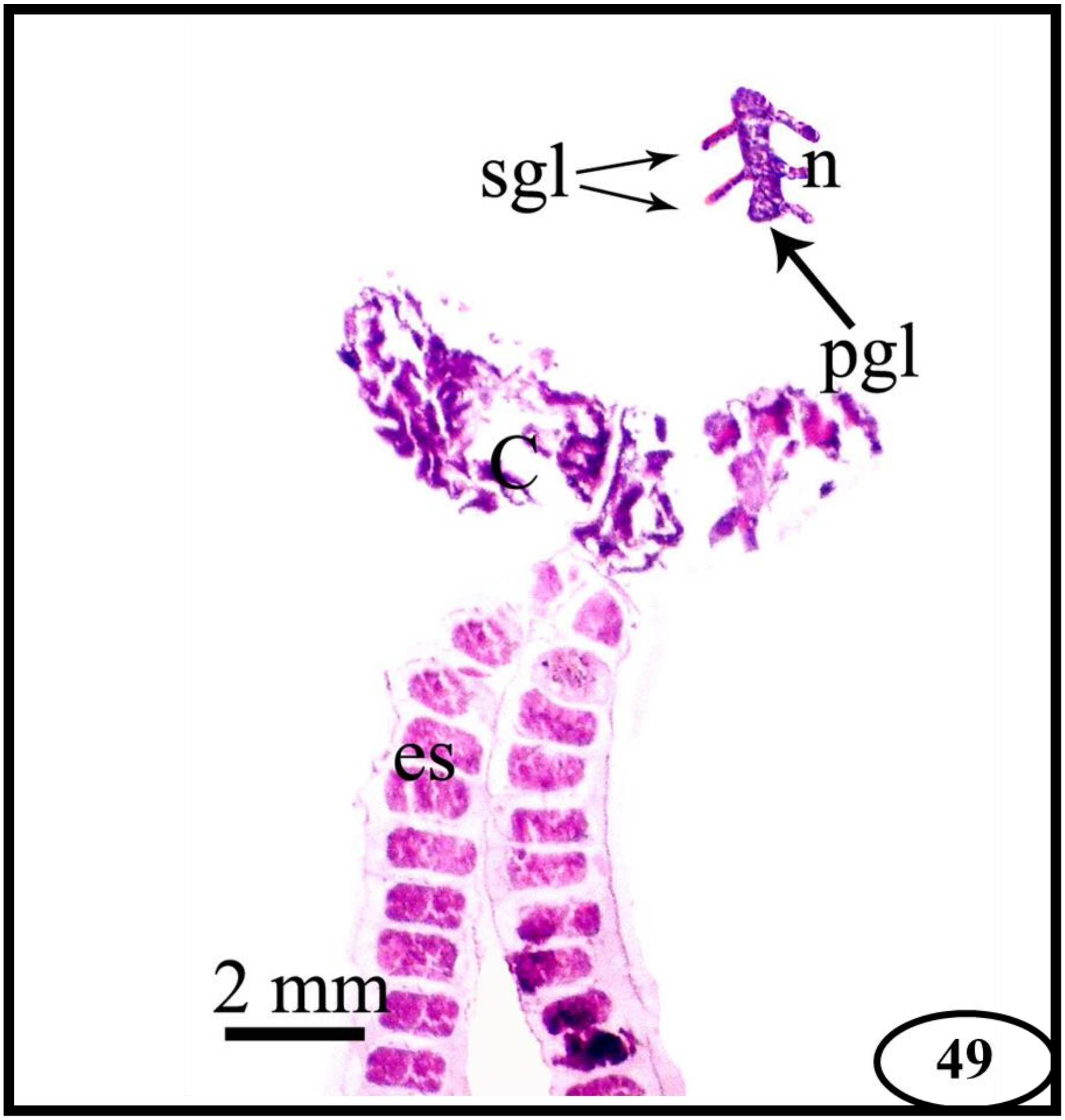


Figure 49: Photomicrograph of a paraffin wax section, stained with haematoxylin and eosin of a primary gill lamella of tilapia galilae, *Sarotherodon galilaeus* infected with female copepod *Lamproglena minuta* (c) causing necrosis (n) of the secondary gill lamellae (sgl). (es), egg sac.

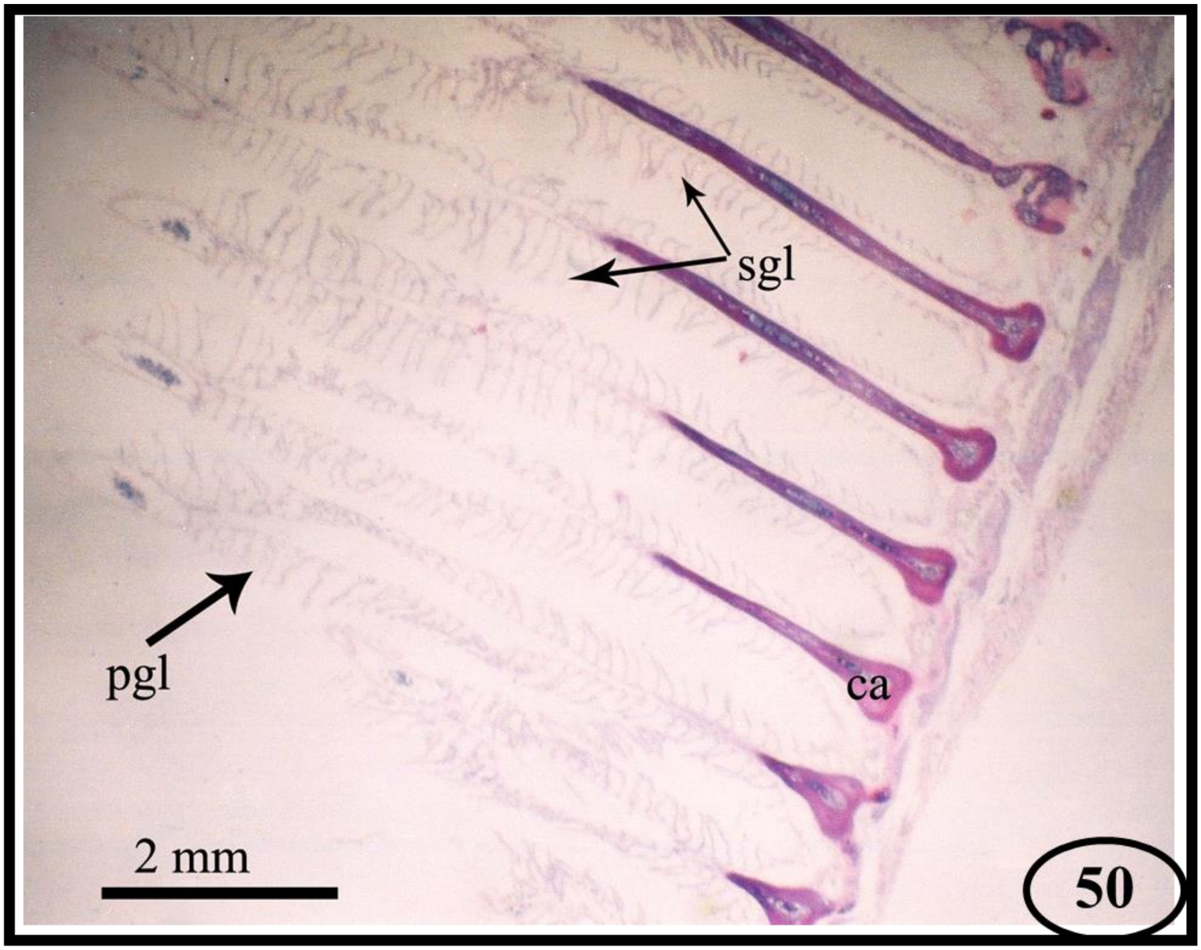


Figure 50: Photomicrograph of a paraffin wax section, stained with haematoxylin and eosin through a gill arch of non-infested flathead grey mullet, *Mugil cephalus* showing the normal histology. ca, cartilage, pgl, primary gill lamella, sgl, secondary gill lamellae.

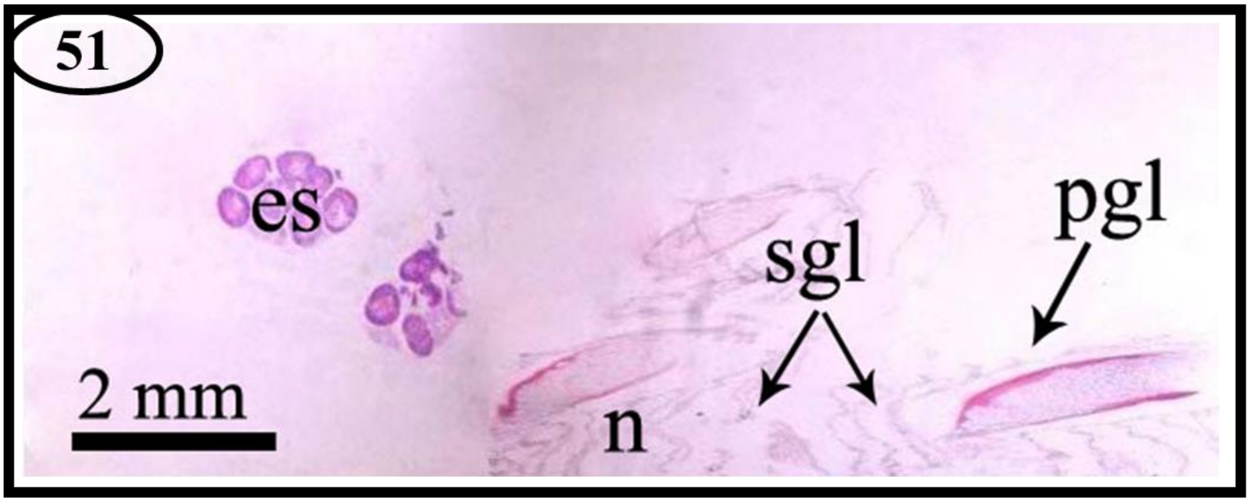


Figure 51: Photomicrograph of a paraffin wax section, stained with haematoxylin and eosin of a primary gill lamella (pgl) of flathead grey mullet *Mugil cephalus* showing the secondary gill lamellae (sgl) infested with female copepod *Ergasilus briani*. egg sac (es) and necrosis (n).

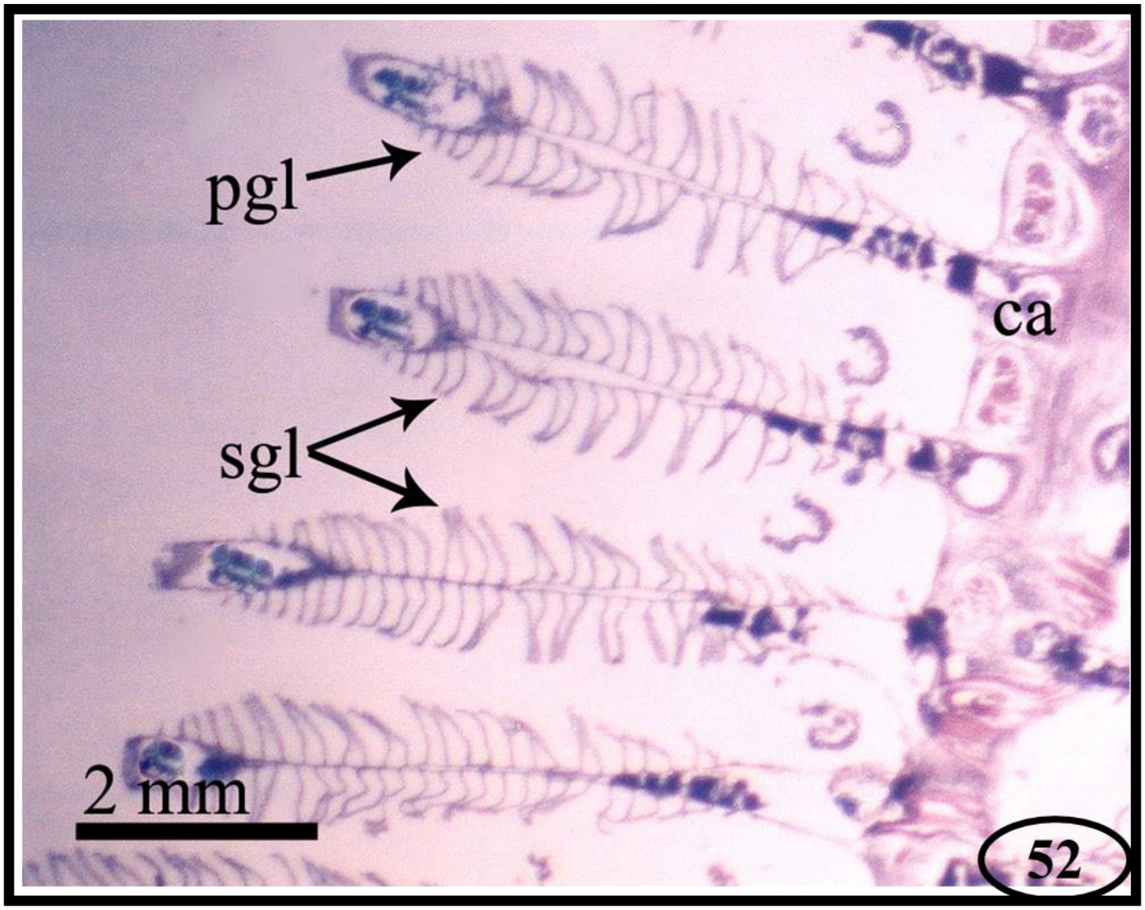


Figure 52: Photomicrograph of a paraffin wax section, stained with haematoxylin and eosin through a gill arch of non-infested *Valamugil seheli* showing the normal histology. ca, cartilage, pgl, primary gill lamella and sgl, secondary gill lamellae.

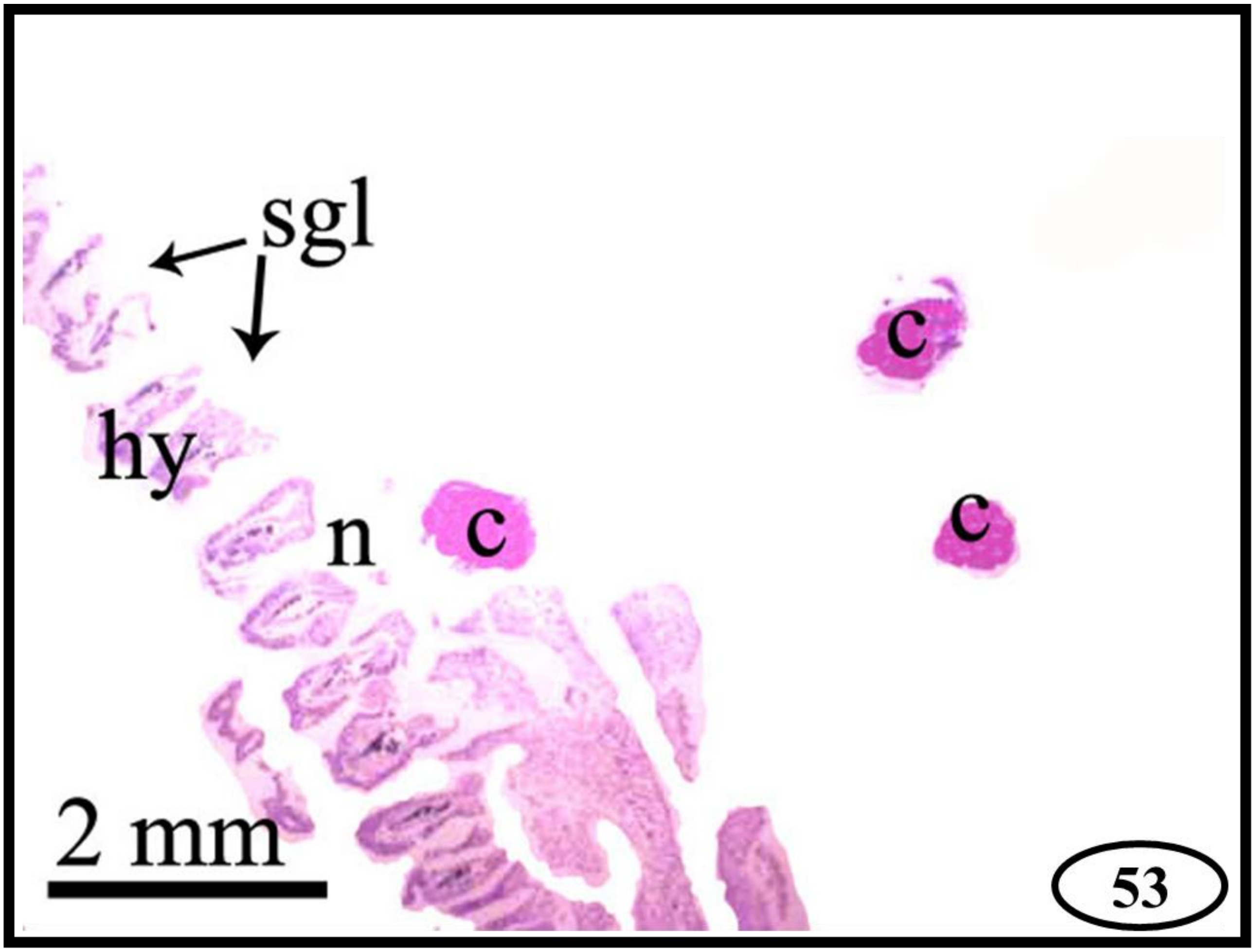


Figure 53: Photomicrograph of a paraffin wax section, stained with haematoxylin and eosin of secondary gill lamellae (sgl) of *Valamugil seheli* infested with female copepods *Ergasilus lizae* (c) induce hyperplasia (hy) and necrosis (n).

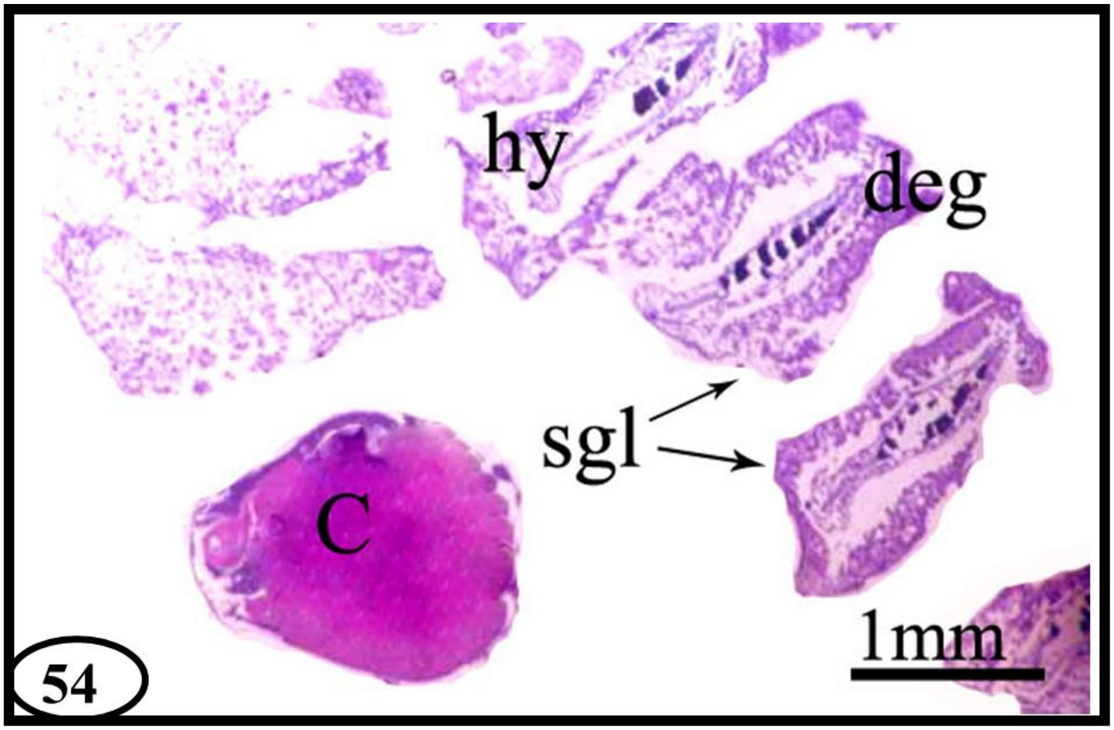


Figure 54: Photomicrograph of a paraffin wax section, stained with haematoxylin and eosin of secondary gill lamellae (sgl) of *Valamugil seheli* infested with female copepod *Ergasilus lizae* (c) induce hyperplasia (hy), degeneration (deg) and necrosis (n).

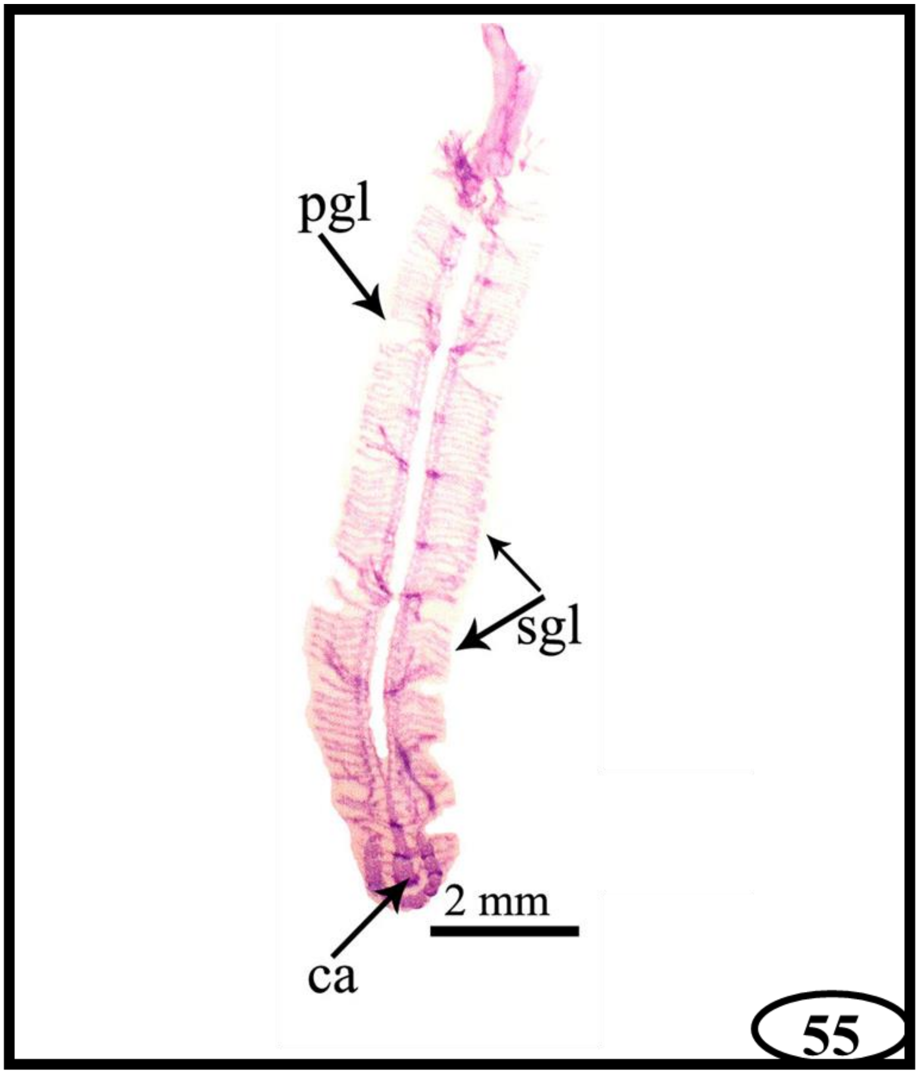


Figure 55: Photomicrograph of a paraffin wax section, stained with haematoxylin and eosin, through a primary gill lamella of non-infested white sea bream, *Diplodus sargus* showing the normal histology. ca, cartilage, pgl, primary gill lamella and sgl, secondary gill lamellae.

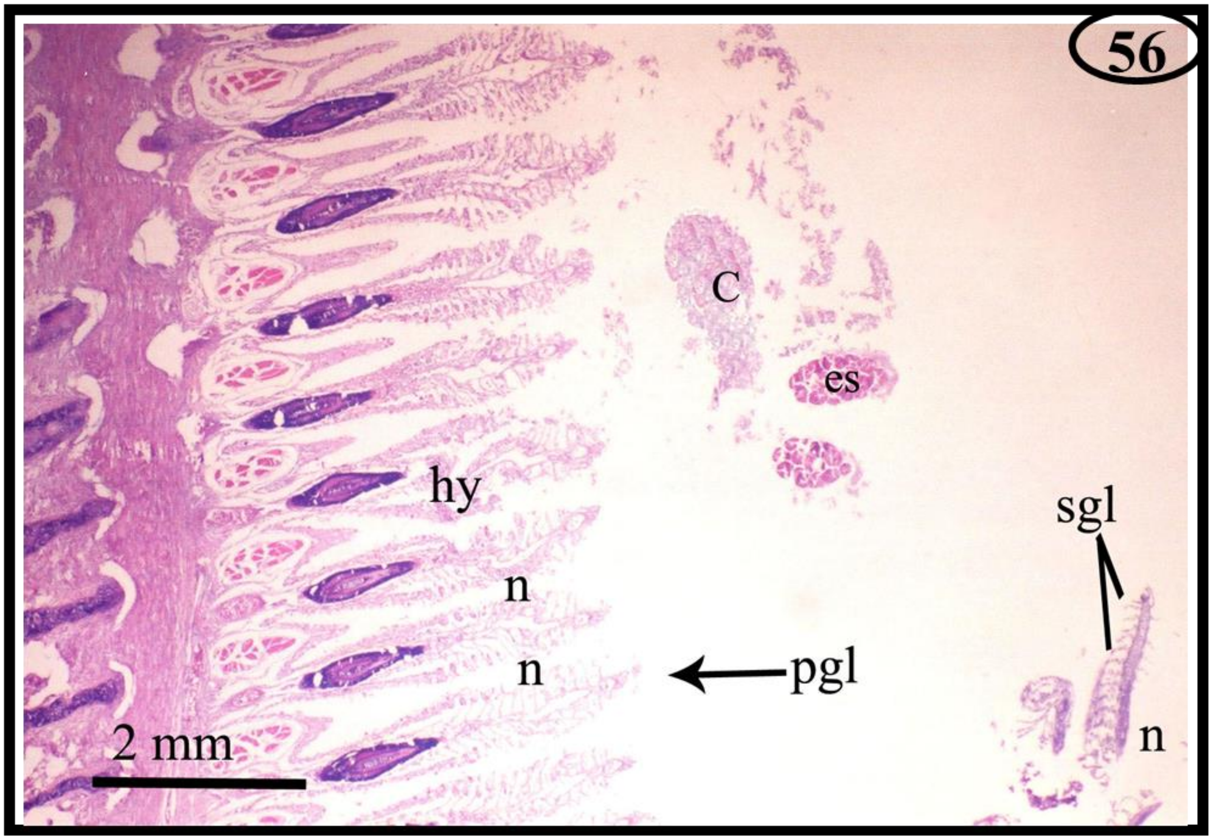


Figure 56: Photomicrograph of a paraffin wax section, stained with haematoxylin and eosin of a gill arch of white sea bream, *Diplodus sargus* infested with female copepod *Ergasilus australiensis* (c) induce hyperplasia (hy) and necrosis (n) of the secondary gill lamellae (sgl). egg sac (es) and primary gill lamella (pgl).

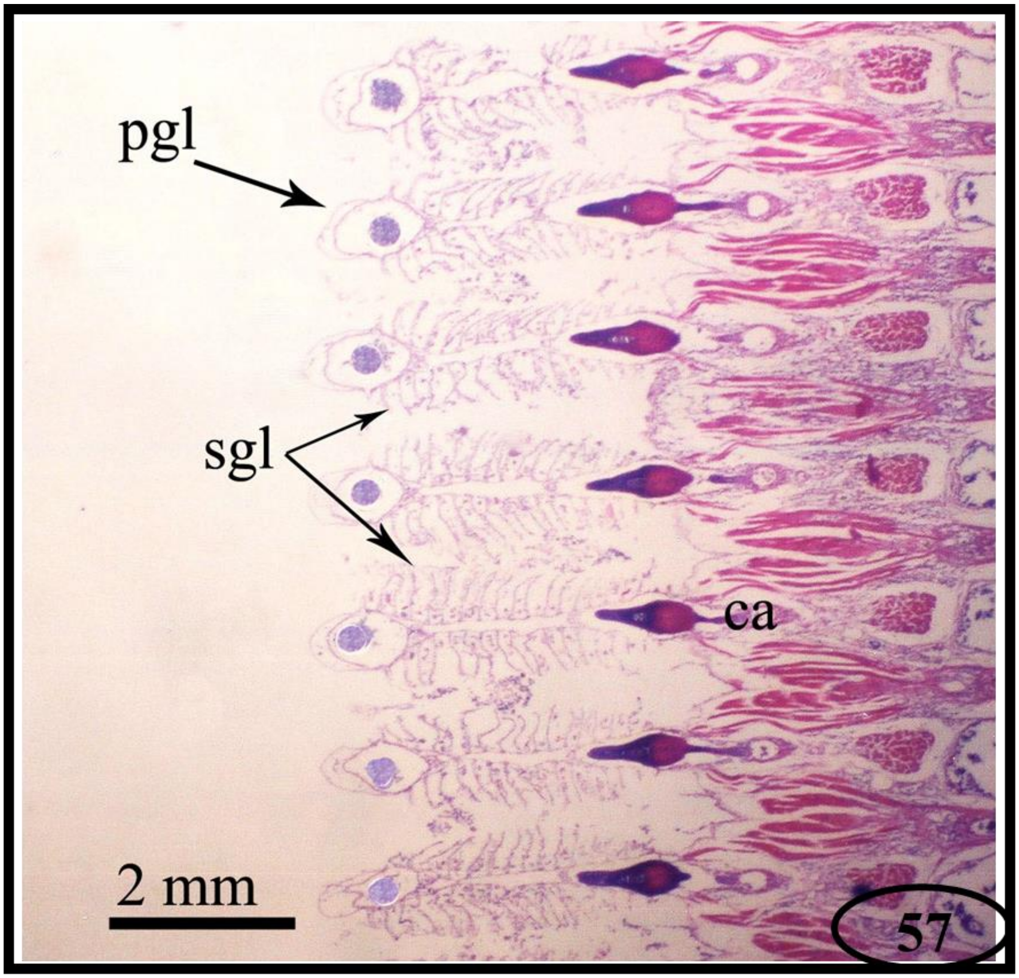


Figure 57: Photomicrograph of a paraffin wax section, stained with haematoxylin and eosin, through a gill arch of non-infested thinlip mullet, *Liza ramada* showing the normal histology. ca, cartilage, pgl, primary gill lamella, and sgl secondary gill lamellae.

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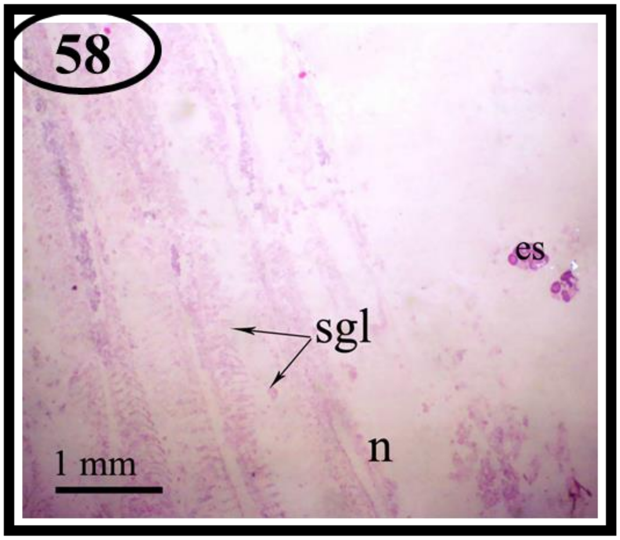

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Figures 58&59: Photomicrograph of a paraffin wax sections, stained with haematoxylin and eosin of a gill arch of thinlip mullet, *Liza ramada* infested with female copepod *Ergasilus mosulensis* (c) induce hyperplasia (hy), degeneration (deg) and necrosis (n) of the secondary gill lamellae (sgl). Es, egg sac and pgl, primary gill lamella.

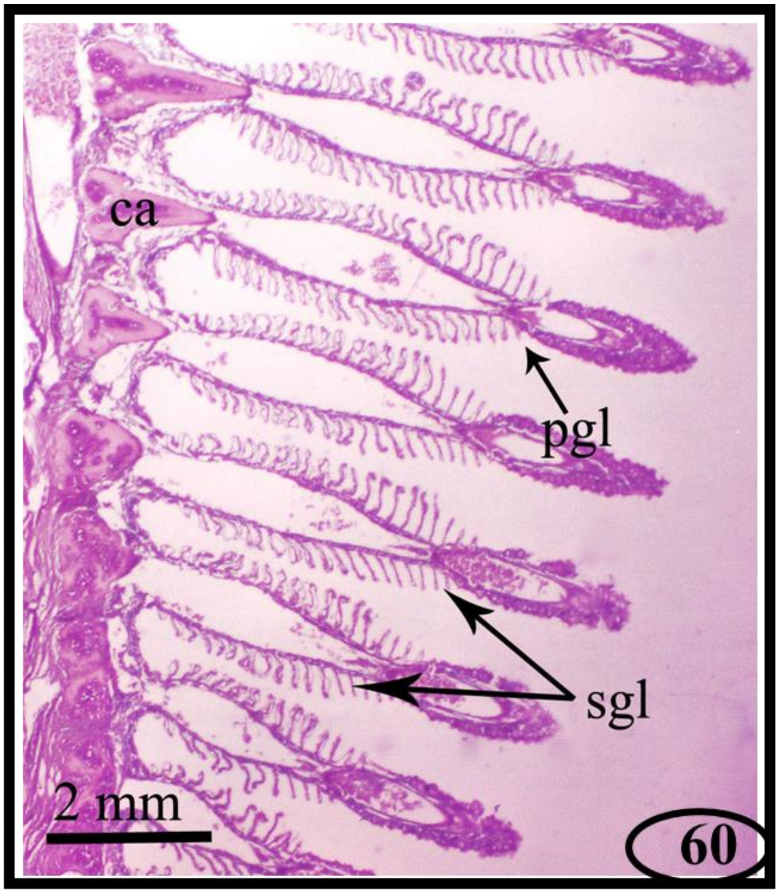


Figure 60: Photomicrograph of a paraffin wax section, stained with haematoxylin and eosin, through a gill arch of non-infested common eel, *Anguilla anguilla* showing the normal histology. ca, cartilage, pgl, primary gill lamella and sgl, secondary gill lamellae.

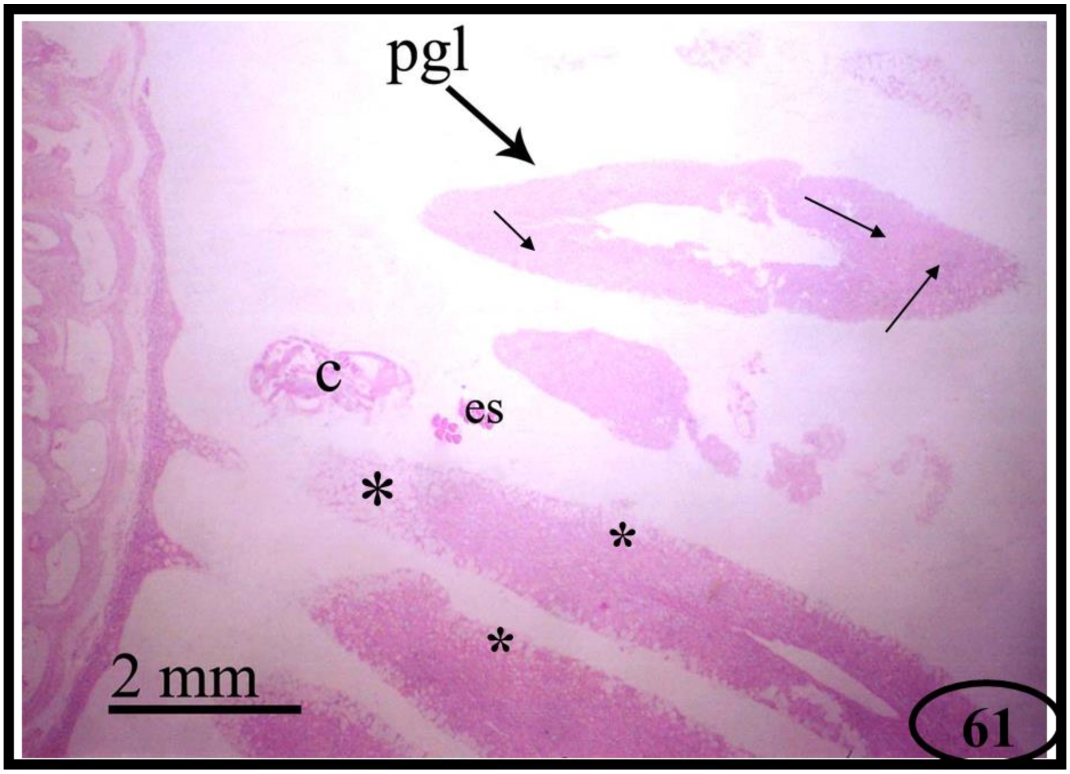


Figure 61: Photomicrograph of a paraffin wax section, stained with haematoxylin and eosin of a gill arch of common eel, *Anguilla anguilla* infested with female copepod *Ergasilus celestis* (c) causing chronic proliferation of primary gill epithelium (arrows) and epitheliocystis-likeinclusions (*). es, egg sac and pgl, primary gill lamellae.

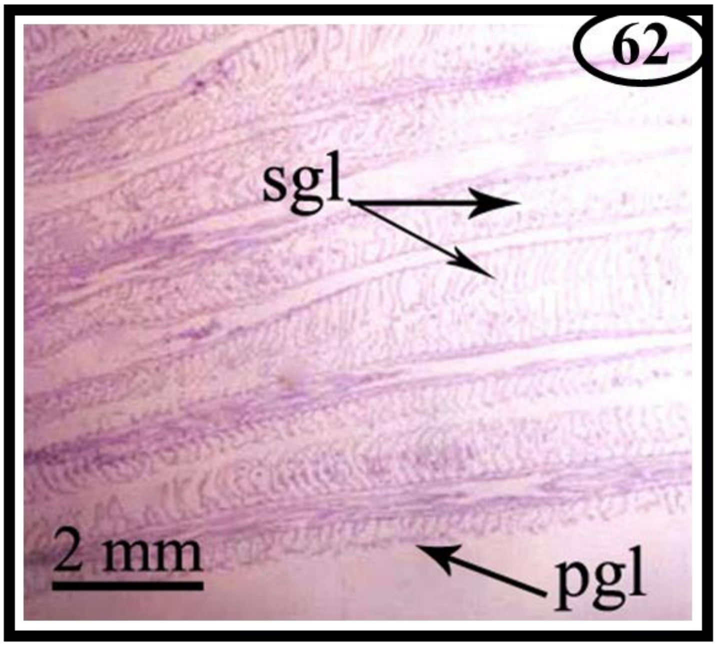


Figure 62: Photomicrograph of a paraffin wax section, stained with haematoxylin and eosin, through a gill arch of non-infested sea bass, *Dicentrarchus labrax* showing the normal histology. pgl, primary gill lamella and sgl, secondary gill lamellae.

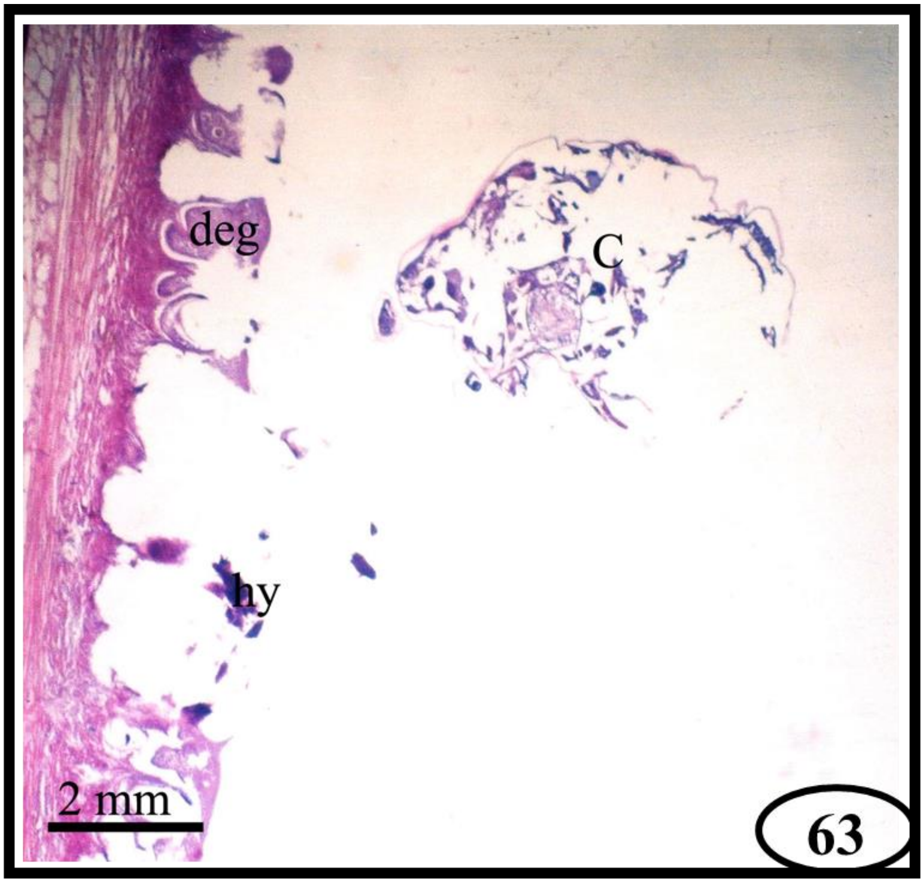


Figure 63: Photomicrograph of a paraffin wax section, stained with haematoxylin and eosin of a gill arch of sea bass, *Dicentrarchus labrax* infested with male copepod *Caligus elongatus* (c) induce hyperplasia (hy) and degeneration (deg).

2. Discussion:

The fish gill is a multifunctional organ involved not only in respiration but also in a variety of homeostatic activities such as osmoregulation, metabolism of circulating hormones, nitrogen excretion and acid base balance **(Erik, 2002)**. A varity of factors, including environmental pollutants and many parasites can induce morphological anomalies in fish gills. The anomalies commonly include hyperplasia with lamellar fusion, epithelial hypertrophy, telangiectasia, oedema, with epithelial separation from basement memberanes, necrosis and epitheliocystis-like inclusion. The economic losses caused by the parasites are considered to be large, primarly due to reduced growth of the host fish medication costs **(Erik, 2002)**.

The histology of the normal (non-infested) gills (gill arch and primary gill lamella) of the collected fishes **(***Dicentrarchus punctatus*, *Sarotherodon galilaeus*, *Mugil cephalus*, *Valamugil seheli*, *Diplodus sargus*, *Liza ramada*, *Anguilla anguilla* and *Dicentrarchus labrax***)** has been studied in detail using light microscope in the present study. The local impacts of the parasitic copepods **(***Lernanthropus kroyeri*, *Lamproglena minuta*, *Ergasilus briani*, *Ergasilus lizae*, *Ergasilus australiensis*, *Ergasilus mosulensis*, *Ergasilus celestis* and *Caligus elongatus***)** infesting gills of the studied fishes as well as the host response are cosidered in detail.

In the current study, the adult parasitic copepods *Lernanthropus kroyeri* and *Lamproglena minuta* were observed attaching themselves to the primary gill lamellae of *Dicentrarchus punctatus* and *Sarotherodon galilaeus* by second antennae causing epithelial hyperplasia with disorganization of epithelial layers, degeneration, necrosis and massive loss of filament tissues. In this respect, **Abu**

Samak (2005) reported that the parasitic copepod *Lernanthropus kroyeri* anchored themselves to the primary lamellae through the piercing action of their cephalothoracic appendages having terminal subchela (hook) such as second antennae and maxilliped. Moreover, **Manera and Dezfuli (2003)** reported also female *L. kroyeri* anchored themselves to the primary lamellae through the piercing action of their second antennae.

Similar results were observed that *Lernanthropus kroyeri* causes some pathological effects such as hyperplasia, degeneration, necrosis and massive loss of filament tissues by some authors **(Kabata, 1979; Luque and Farfan, 1990; Deets and Kabata, 1991; Timi and Etchegoin, 1996; Olivier *et al*., 1997 and Özel *et al*., 2004)** on the gill filaments of the sea bass *Dicentrarchus labrax*. In addition to the previous, there are similar findings were reported between this parasite and *Lamproglena minuta*. The latter causes some pathological effects such as hyperplasia, degeneration, necrosis and massive loss of filament tissues as reported by **Sproston *et al.* (1950)** on gills of *Ophiocephalus spp*.**.** Moreover, **Khalifa and Post (1976), Shields and Goode (1978), Shariff (1981), Uehara *et al.* (1984) and Berry *et al.* (1991)** studied these lesions on the gills filaments of *Cbanos cbanos* infesting with the copepodian *Lernaea cyprinacea*

Also, the present study has revealed that the five species of the parasitic copepods of genus *Ergasilus* (*Ergasilus briani*, *Ergasilus lizae*, *Ergasilus australiensis, Ergasilus mosulensis* and *Ergasilus celestis)* cause major lesions at the site of attachment of the gill lamellae such as hyperplasia, degeneration, chronic proliferation, epitheliocystis-like inclusions, necrosis and massive loss of filament tissues. In this respect, **Barker and David (2000)** reported that parasitic copepod *Ergasilus celestis* and monogenean parasite *Pseudodactylogryus anguillae* have caused serious problems of the gill filaments of the eel fish *Anguilla rostrata* often result in gill hyperplasia, degeneration, chronic proliferation and respiratory failure. Moreover, **Valtonen *et al*. (1997)** reported that parasitic copepods *Ergasilus briani* and *Ergasilus sieboldi* caused massive necrosis, epitheliocystis-like inclusions and swelling of one or a few primary lamellae on the gills of roach and perch. Similarly, *Ergasilus sieboldi* and other *Ergasilus* spp. caused histopathological changes at the site of attachment of the gill lamellae such as hyperplasia, sever hemorrhaging and gill inflammation associated with attachment and feeding of the parasite (**Barker *et al.*, 2000)**.

Similar results were described by **Dogiel *et al.* (1961)** that reported extensive gill damage and sever haemorrhage with inflammation and hyperplasia associated with the attachment and feeding of the parasitic copepod *Ergasilus sieboldi* on gills of *Tinca tinca*. **Rogers (1969)** reported that increase in number of mucous cells, fusion of lamellae and filaments due to epithelial proliferation and degeneration for *Ergasilus cyprinaceus*. **Kabata (1970)** have been reviewed that the effects of *Ergasilus* spp.**.** **Sarig (1971)** reported that severely damage gill tissue of *Ergasilus* spp.**.** **Nakajima *et al*. (1974)** reported that sever lesions of *Pseudoergasilus zacconis*. **Paperna (1975)** reported that several hundered *Ergasilus lizae* were caused massive lesions of *Mugil cephalus*. **Rogers and Hawke (1978)** found that massive lesions of *Dorosoma* from *Ergasilus sieboldi*. **Paperna and Zwerner (1981)** reported that an increase of mucous cells, lymphocytes and epithelial hyperplasia in gill filaments of *Morone saxatilis* infected by *Ergasilus labracis*. **Roubal (1986, 1989)** found that the parasitic copepod *Ergasilus australiensis* causes epithelial hyperplasia in the basal half of the gill filaments and pseudobranchs of *Acanthopagrus australis*. **Hogans (1989)** reported that sever lesions of *Ergasilus labracis*. **Cone *et al.* (1996)** reported that sever problems of *Ergasilus celestis* on gills of *Anguilla rostrata*. **Barker *et al.* (2000)** reported that epithelial hyperplasia and necrosis of the gill copepod, *Ergasilus gibbus* infesting eels, *Anguilla rostrata*. **Dezfuli *et al.* (2003)** studied that the pathological effect of *Ergasilus sieboldi* infesting the gills of *Abramis brama* induce lamellar fusion due to hyperplasia of the interlamellar epithelium, massive mucous cell proliferation as well as accumulation of many types of immune cells in the gill tissue at the site of attachment.

The present study has demonstrated also that the parasitic copepod *Caligus elongates* cause major lesions and produce hyperplasia, degeneration and necrosis at the site of attachment of the gills of the sea bass, *Dicentrarchus labrax*. In this respect, **Erik (2002)** reported massive lesions of the parasitic copepod, *Caligus minimus* on the gill lamellae of *Dicentrarchus labarax* such as epithelial hyperplasia and necrosis. In addition, other examples are *Lepeopheirus salmonis* and *Caligus elongatus* cause severe problems like hyperplasia, necrosis and degeneration with massive loss of filament tissues of *Dicentrarchus labarax* **(Pike and Wodsworth, 2000)**. Similar findings were observed that lesions found on the head and operculum by the infestation with lice reported by some workers **(White, 1940; Wootten *et al.*, 1982)**. Caligid copepods are among the most problematic parasites in cultured marine species, including Mediterranean sea bass and sea bream **(Paperna, 1980; Rodgers and Furones, 1998)**. Salmon lice infections cause severs osmoregulatory problems for salmonids due to mechanical disruption of the fish skin **(Wootten *et al.*, 1982)**. Oedema, hyperplasia, sloughing of epidermal cells and inflammation are caused by attachment and feeding of pre-adult and adult *Lepeopheirus salmonis* **Onsdottir, (1992)**.

CHAPTERIV

Effect of Anti-parasitic agents

Effect of Anti-parasitic agents

1. Results:

Experimental trials (in vitro) were conducted to determine the effect of anti-parasitic agents; natural plant extracts (Curcumin, *Curcuma longa* and Myrrh, *Commiphora molmol*) in addition to human anti-parasitic synthetic drug, (Mebendazole) against the parasitic copepods (*Lamproglena minuta*) infesting *Sarotherodon galilaeus* and (*Ergasilus celestis*) infesting *Anguilla anguilla* in which these parasites are more abundant and have the highest intensity on their fish hosts among the different seasons than the other parasitic copepods that were collected during the present study.

I: Effect of the natural plant extracts Curcumin, *Curcuma longa* and Myrrh, *Commiphora molmol* and the synthetic drug Mebendazole on the fish hosts *Sarotherodon galilaeus* and *Anguilla anguilla*.

A. Effects of the *Curcuma longa*:

The effect of different concentrations of *Curcuma longa* on the mortality rate (%) of the fish hosts *Sarotherodon galilaeus* and *Anguilla anguilla* compared with the control ones was illustrated in table 25 and showed in figure 64.

At exposure time 10 hours, the mortality rate (%) of the cichlid fish *Sarotherodon galilaeus* increases from 10% at concentration of 100ppm to 100% at concentration of 17500ppm compared with the control ones. And at the same exposure time 10 hours, the mortality rate (%) of the common eel *Anguilla anguilla* increases from 20% at concentration of 100ppm to 100% at concentration

Table (25): Effect of different concentrations of the plant extract Curcumin, *Curcuma longa* on the mortality rate (%) of the cichlid fish *Sarotherodon galilaeus* and common eel *Anguilla anguilla* in comparison with the control ones.

Fish hosts		Exposure time (hrs)	Control	Concentrations of the extract (ppm)				
				10ppm	100ppm	500ppm	1000ppm	17500ppm
Sarotherodon galilaeus	Mortality rate %	10	0%	0%	10%	40%	50%	100%
Anguilla anguilla		10	0%	0%	20%	50%	80%	100%

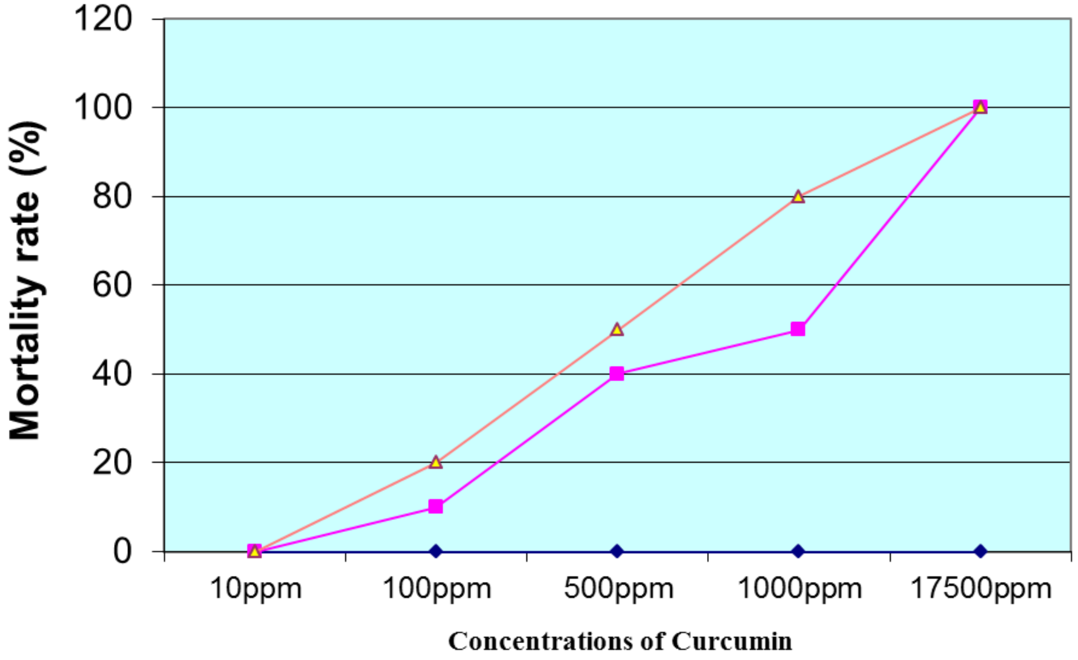


Figure (64): Effect of different concentrations of Curcumin on the mortality rate (%) of the cichlid fish *Sarotherodon galilaeus* and common eel *Anguilla anguilla* in comparison with the control ones.

of 1000ppm compared with the control ones (Table 25 and figure 64).

In general, table (25) has revealed that *Curcuma longa* is safe on the cichlid fish *Sarotherodon galilaeus* and on the common eel *Anguilla anguilla* at low concentration 10ppm at exposure time 10 hours where the mortality rate (%) is at the lowest value of 0 (%). Also, the LC_{50} of *Curcuma longa* for the cichlid fish *Sarotherodon galilaeus* is shown to be 1000ppm and 500 ppm for the common eel *Anguilla anguilla* at exposure time 10 hours (Table 25).

B. Effects of the *Commiphora molmol*:

The effect of different concentrations of *Commiphora molmol* on the mortality rate (%) of the fish hosts *Sarotherodon galilaeus* and *Anguilla anguilla* compared with the control ones was illustrated in table 26 and showed in figure 65.

At exposure time 10 hours, the mortality rate (%) of the cichlid fish *Sarotherodon galilaeus* increases from 20% at concentration of 100ppm to 100% at concentration of 1000ppm compared with the control ones. And at the same exposure time 10 hours, the mortality rate (%) of the common eel *Anguilla anguilla* increases from 30% at concentration of 100ppm to 100% at concentration of 8700ppm compared with the control ones (Table 26 and figure 65).

In general, table (26) has revealed that *Commiphora molmol* is safe on the cichlid fish *Sarotherodon galilaeus* and on the common eel *Anguilla anguilla* at low concentration 10ppm at exposure time 10 hours where the mortality rate (%) is at the lowest value of 0 (%). Also, the LC_{50} of *Commiphora molmol* for the cichlid fish *Sarotherodon galilaeus* and the common eel *Anguilla anguilla* is shown to be 500 ppm at exposure time 10 hours (Table 26).

Table (26): Effect of different concentrations of the plant extract Myrrh, *Commiphora molmol* on the mortality rate (%) of the cichlid fish *Sarotherodon galilaeus* and common eel *Anguilla anguilla* in comparison with the control ones.

Fish hosts		Exposure time (hrs)	Control	Concentrations of the extract (ppm)				
				10ppm	100ppm	500ppm	1000ppm	8700ppm
Sarotherodon galilaeus	Mortality rate %	10	0%	0%	20%	50%	100%	100%
Anguilla anguilla		10	0%	0%	30%	50%	90%	100%

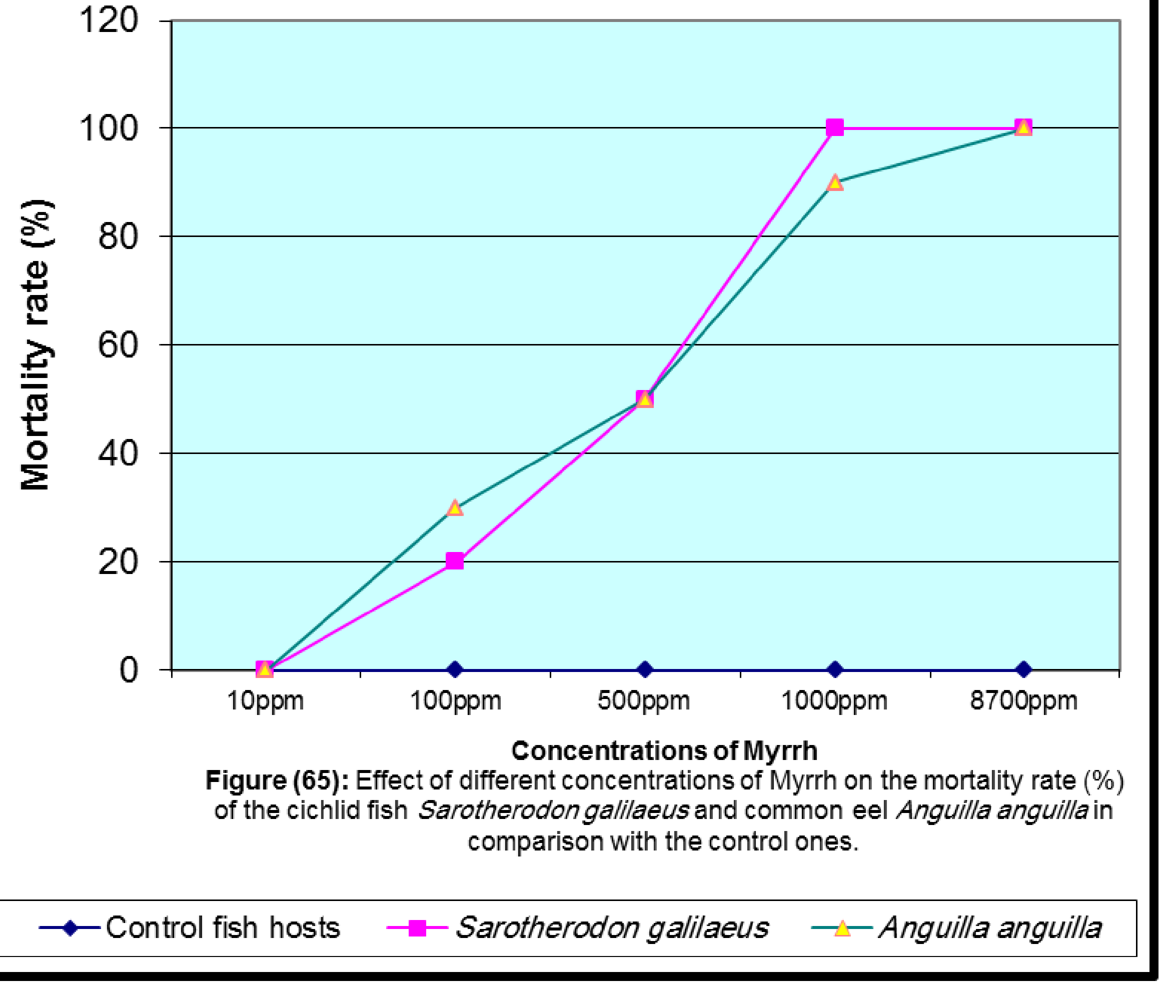


Figure (65): Effect of different concentrations of Myrrh on the mortality rate (%) of the cichlid fish *Sarotherodon galilaeus* and common eel *Anguilla anguilla* in comparison with the control ones.

C. Effects of the Mebendazole:

The effect of different concentrations of Mebendazole on the mortality rate (%) of the fish hosts *Sarotherodon galilaeus* and *Anguilla anguilla* compared with the control ones was illustrated in table 27 and showed in figure 66.

At exposure time 10 hours, the mortality rate (%) of the cichlid fish *Sarotherodon galilaeus* increases from 20% at concentration of 100ppm to 100% at concentration of 30000ppm compared with the control ones. And at the same exposure time 10 hours, the mortality rate (%) of the common eel *Anguilla anguilla* increases from 20% at concentration of 100ppm to 100% at concentration of 30000ppm compared with the control ones (Table 27 and figure 66).

In general, table (27) has revealed that Mebendazole is safe on the cichlid fish *Sarotherodon galilaeus* and on the common eel *Anguilla anguilla* at low concentration 10ppm at exposure time 10 hours where the mortality rate (%) is at the lowest value of 0 (%). Also, the LC_{50} of Mebendazole for the cichlid fish *Sarotherodon galilaeus* is shown to be 1000ppm and 500 ppm for the common eel *Anguilla anguilla* at exposure time 10 hours (Table 27).

Table (27): Effect of different concentrations of the medical drug Mebendazole on the mortality rate (%) of the cichlid fish *Sarotherodon galilaeus* and common eel *Anguilla anguilla* in comparison with the control ones.

Fish hosts		Exposure time (hrs)	Control	Concentrations of the extract (ppm)				
				10ppm	100ppm	500ppm	1000ppm	30000ppm
Sarotherodon galilaeus	Mortality rate %	10	0%	0%	20%	40%	50%	100%
Anguilla anguilla		10	0%	0%	20%	50%	70%	100%

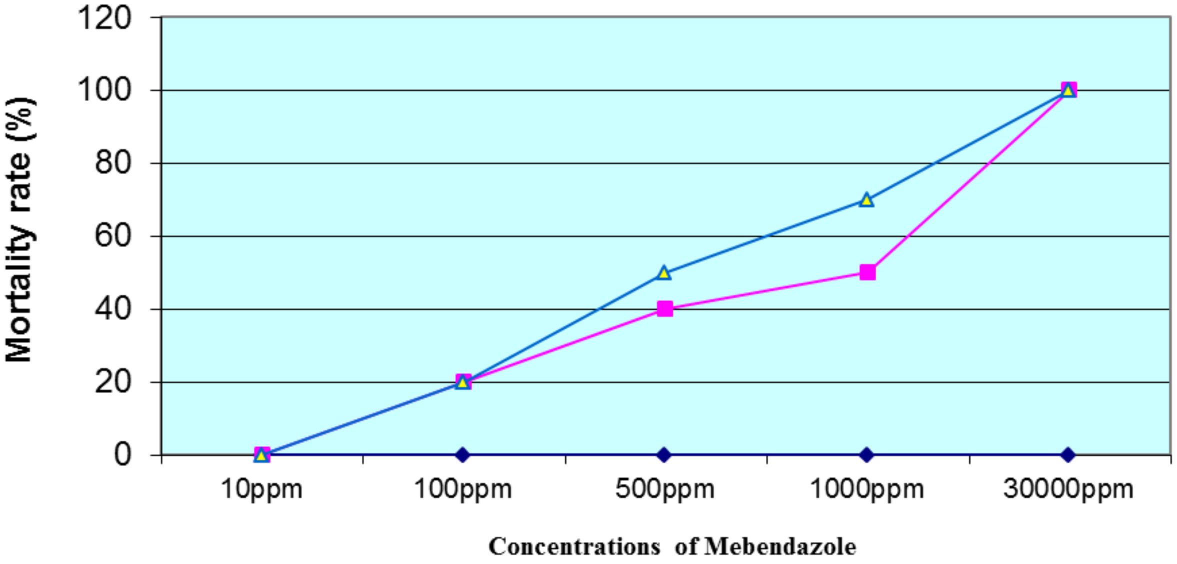


Figure (66): Effect of different concentrations of Mebendazole on the mortality rate (%) of the cichlid fish *Sarotherodon galilaeus* and common eel *Anguilla anguilla* in comparison with the control ones.

II: Effect of the natural plant extracts Curcumin, *Curcuma longa* and Myrrh, *Commiphora molmol* and the synthetic drug Mebendazole on the parasitic copepods *Lamproglena minuta* and *Ergasilus celestis.*

A. Effects of the *Curcuma longa*:

Effect of the natural plant extract, *Curcuma longa* was studied by using of different concentrations against the surviving duration or life span of the parasitic copepods (*Lamproglena minuta*) infesting *Sarotherodon galilaeus* and (*Ergasilus celestis*) infesting *Anguilla anguilla*. The efficacy of treatment by *Curcuma longa* was demonstrated as the reduction in the treated parasites life span per minutes, compared with that in the control groups, at water temperature 27±2ºC. The statistical results were illustrated in tables 28 & 31 respectively and showed in figures 67, 68 & 69.

I. On the crustacean parasite (*Lamproglena minuta*):

In general and comparing with the control groups, the life span of the treated parasites, *Lamproglena minuta* decreased with the increase of drug concentration. At the level of the drug effect, there was unstable decreasing in the life span of the treated parasites with the increasing of drug concentrations (Table 28 & figure 67). Differences in the life span of the treated *Lamproglena minuta* with *Curcuma longa* may be due to the susceptibility of *Lamproglena minuta*. Moreover, no abnormal or unusual behaviour was observed in any of the treated crustacean parasite.

Statistical analysis using linear regression (Table 31 and Figure 69) has revealed the presence of a very highly significant positive correlation between the concentrations of the natural plant extract (*Curcuma longa*) and life-span of the crustacean parasite *Lamproglena minuta* (*t*-value = 6.188, P = 0.000).

Table (28): Effect of different concentrations of the plant extract Curcumin, *Curcuma longa* on the survival time (Life span) of the crustacean parasites *Lamproglena minuta* infesting *Sarotherodon galilaeus* and *Ergasilus celestis* infesting *Anguilla anguilla*, at temperature 27±2ºC.

Parasitic copepods	Mean control life span/min.	Concentrations of the extract (ppm)				
		10ppm	100ppm	500ppm	1000ppm	17500ppm
Lamproglena minuta	180	50	55	75	30	15
Ergasilus celestis	135	85	55	70	45	11

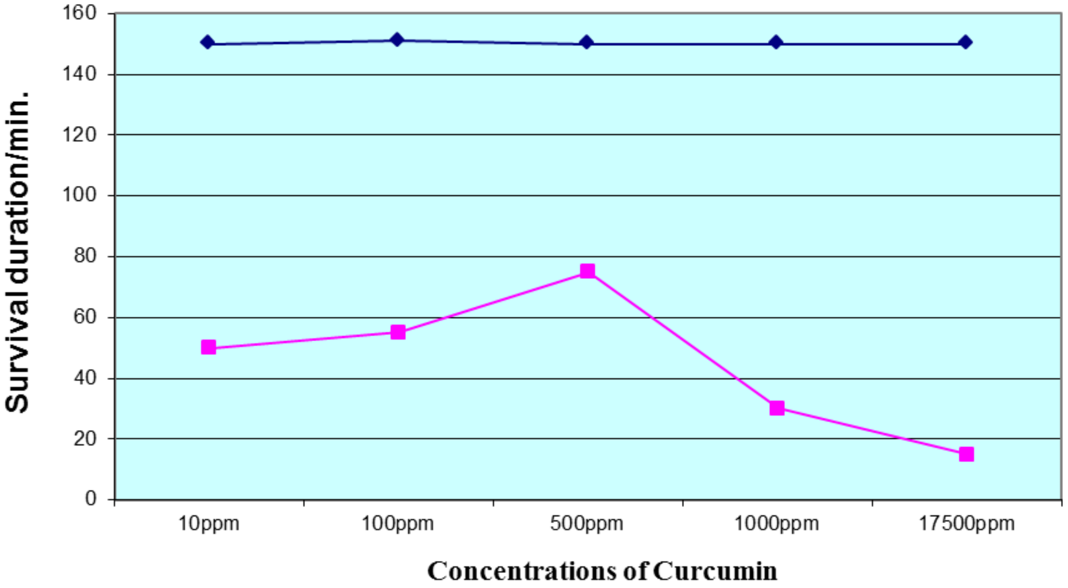


Figure (67): Efficacy of the plant extract, Curcumin on the copepodian parasite *Lamproglena minuta*, comparing with the control parasites.

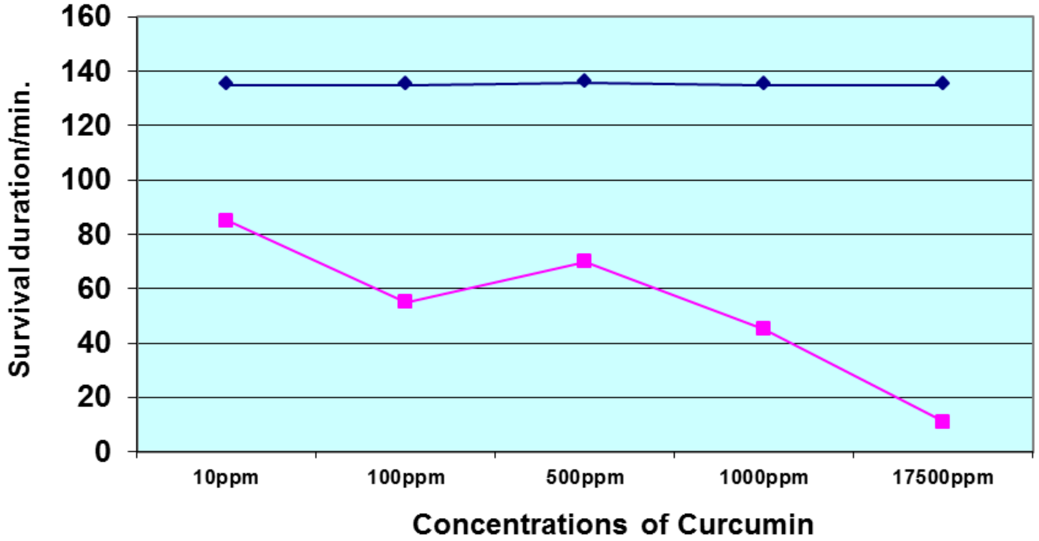


Figure (68): Efficacy of the plant extract, Curcumin on the copepodian parasite *Ergasilus celestis*, comparing with the control parasites.

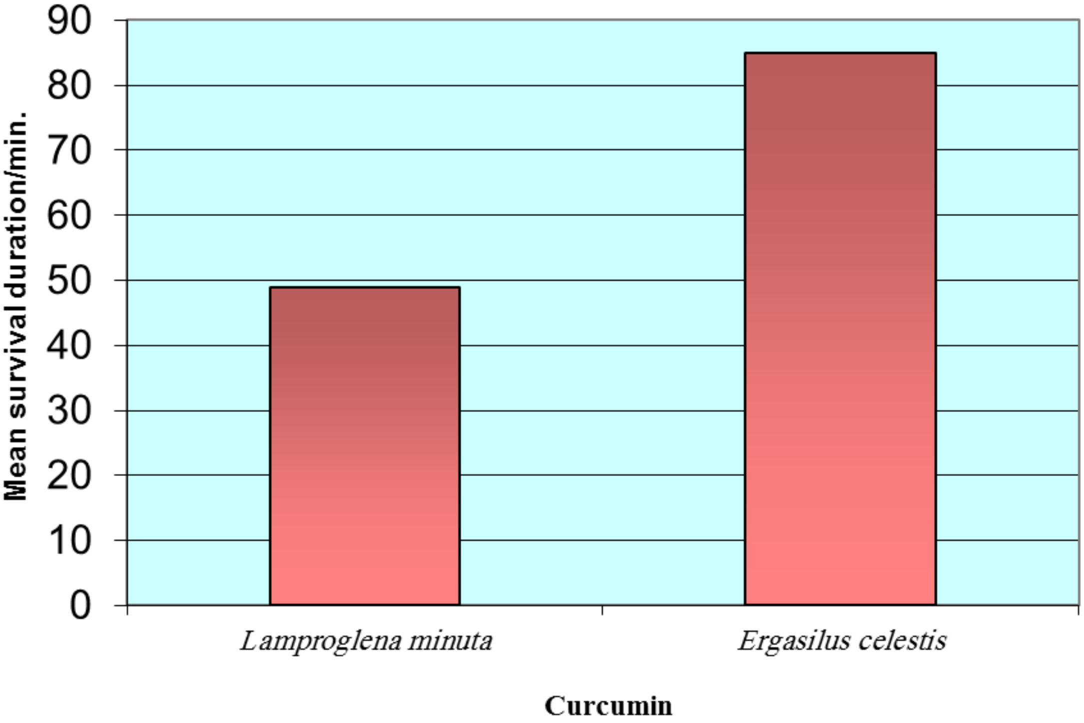


Figure (69): The relationship between the mean of survival time of each parasite group and the natural plant extract (Curcumin).

II. On the crustacean parasite (*Ergasilus celestis*):

In general and comparing with the control groups, the life span of the treated parasites, *Ergasilus celestis* decreased with the increase of drug concentration. At the level of the drug effect, there was unstable decreasing in the life span of the treated parasites with increasing of drug concentrations (Table 28 & figure 68). Differences in the life span of the treated *Ergasilus celestis* with *Curcuma longa* may be due to the susceptibility of *Ergasilus celestis*. Moreover, no abnormal or unusual behaviour was observed in any of the treated crustacean parasite, *Ergasilus celestis*.

Statistical analysis using linear regression (Table 31 and Figure 69) has revealed the presence of a highly significant positive correlation between the concentrations of the natural plant extract (*Curcuma longa*) and life-span of the crustacean parasite *Ergasilus celestis* (*t*-value = 2.786, P = 0.015).

Figure 69 has revealed that the parasitic copepod, *Lamproglena minuta* was more sensitive for treatment with *Curcuma longa* than the other parasite, *Ergasilus celestis.*

B. Effects of the *Commiphora molmol*:

Effect of the natural plant extract, *Commiphora molmol* was studied by using of different concentrations against the surviving duration or life span of parasitic copepods (*Lamproglena minuta*) infesting *Sarotherodon galilaeus* and (*Ergasilus celestis*) infesting *Anguilla anguilla*. The efficacy of treatment by *Commiphora molmol* was demonstrated as the reduction in the treated parasites life span per minutes, comparing with that in the control groups, at water temperature 27±2ºC. The statistical results were illustrated in tables 29 & 31 respectively and showed in figures 70, 71 & 72.

I. On the crustacean parasite (*Lamproglena minuta*):

In general and comparing with the control groups, the life span of the treated parasites, *Lamproglena minuta* decreased with the increase of drug concentration. At the level of the drug effect, there was unstable decreasing in the life span of the treated parasites with increasing of drug concentrations (Table 29 & figure 70). Differences in the life span of the treated *Lamproglena minuta* with *Commiphora molmol* may be due to the susceptibility of *Lamproglena minuta* to the extract. Moreover, no abnormal or unusual behaviour was observed in any of the treated crustacean parasite, *Lamproglena minuta*.

Statistical analysis using linear regression (Table 31 and Figure 72) has revealed the presence of a highly significant positive correlation between the concentrations of the natural plant extract (*Commiphora molmol*) and life-span of the crustacean parasite *Lamproglena minuta* (*t*-value = 2.929, P = 0.012).

II. On the crustacean parasite (*Ergasilus celestis*):

In general and comparing with the control groups, the life span of the treated parasites, *Ergasilus celestis* decreased with the increase of drug concentration. At the level of the drug effect, there was unstable decreasing in the life span of the treated parasites with increasing of drug concentrations (Table 29 & figure 71). Differences in the life span of the treated *Ergasilus celestis* with

Table (29): Effect of different concentrations of the plant extract Myrrh, *Commiphora molmol* on the survival time (Life span) of the crustacean parasites *Lamproglena minuta* infesting *Sarotherodon galilaeus* and *Ergasilus celestis* infesting *Anguilla anguilla*, at temperature 27±2ºC.

Parasitic copepods	Control life span/min.	Concentrations of the extract (ppm)				
		10ppm	100ppm	500ppm	1000ppm	8700ppm
Lamproglena minuta	180	100	55	90	95	25
Ergasilus celestis	135	120	100	95	85	35

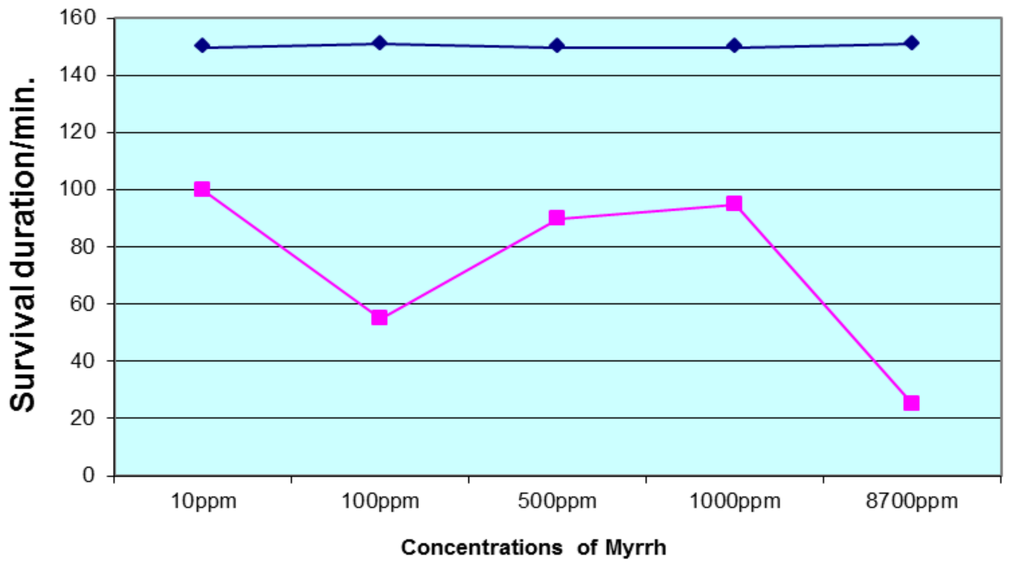


Figure (70): Efficacy of the plant extract, Myrrh on the copepodian parasite *Lamproglena minuta* by comparison control parasites.

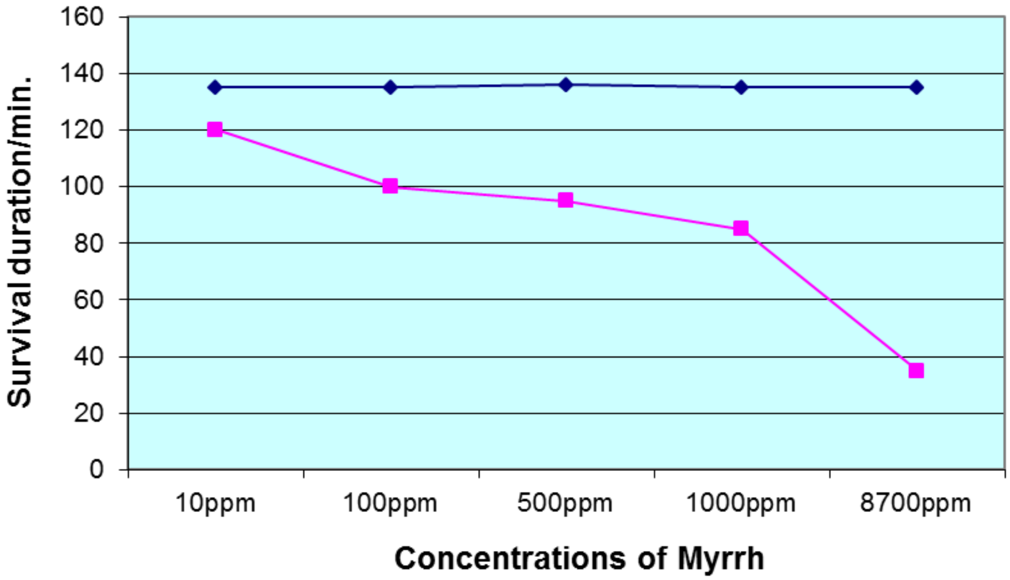
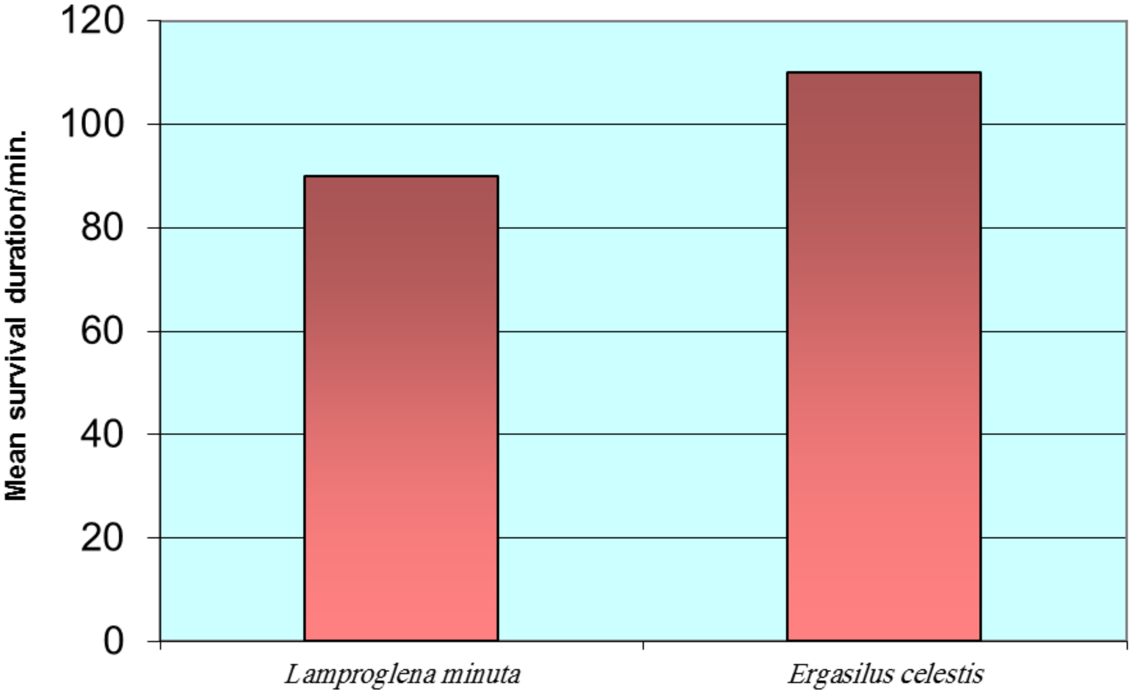


Figure (71): Efficacy of the plant extract, Myrrh on the copepodian parasite *Ergasilus celestis* by comparison control parasites.



Myrrh

Figure (72): The relationship between mean survival time of each parasite group and the natural plant extract (Myrrh).

Commiphora molmol may be due to the susceptibility of *Ergasilus celestis* to the extract. Moreover, no abnormal or unusual behaviour was observed in any of the treated crustacean parasite, *Ergasilus celestis*.

Statistical analysis using linear regression (Table 31 and Figure 72) has revealed the presence of a highly significant positive correlation between the concentrations of the natural plant extract (*Commiphora molmol*) and life-span of the crustacean parasite *Ergasilus celesti*s (*t*-value = 2.496, P = 0.026).

Figure 72 has revealed that the parasitic copepod, *Lamproglena minuta* was more sensitive for treatment with *Commiphora molmol* than the other parasite, *Ergasilus celestis.*

C. Effects of the Mebendazole:

Effect of the synthetic drug, Mebendazole was studied by using of different concentrations against the surviving duration or life span of parasitic copepods; (*Lamproglena minuta*) infesting *Sarotherodon galilaeus* and (*Ergasilus celestis*) infesting *Anguilla anguilla*. The efficacy of treatment by Mebendazole was demonstrated as the reduction in the treated parasites life span per minutes, comparing with that in the control groups, at water temperature 30±2ºC. The statistical results were illustrated in tables 30 & 31 respectively and showed in figures 73, 74 & 75.

I. On the crustacean parasite (*Lamproglena minuta*):

In general and comparing with the control groups, the life span of the treated parasites, *Lamproglena minuta* decreased with the increase of drug concentration. At the level of the drug effect, there was unstable decreasing in the

Table (30): Effect of different concentrations of the medical drug Mebendazole on the survival time (Life span) of the crustacean parasites *Lamproglena minuta* infesting *Sarotherodon galilaeus* and *Ergasilus celestis* infesting *Anguilla anguilla,* at temperature 30±2ºC.

Parasitic copepods	Control life span/min.	Concentrations of the extract (ppm)				
		10ppm	100ppm	500ppm	1000ppm	30000ppm
Lamproglena minuta	99	47	50	45	40	9
Ergasilus celestis	80	45	40	75	70	15

Table (31): Statistical analysis of the different concentrations of the natural plant extracts (Curcumin and Myrrh) and synthetic drug Mebendazole on parasitic copepods *Lamproglena minuta* and *Ergasilus celelstis* infesting *Sarotherodon galilaeus* and *Anguilla anguilla.*

Parasitic copepods	Anti-parasitic agents	Mean control life span/min.	Mean life span during treatment/min.	*t-value*	SD	P
Lamproglena minuta	Curcumin	180	49	6.188	20	0.000***
	Myrrh	180	90	2.929	59	0.012**
	Mebendazole	99	39	6.983	15	0.000***
Ergasilus celestis	Curcumin	135	85	2.786	85	0.015**
	Myrrh	135	110	2.496	110	0.026**
	Mebendazole	80	67	1.603	51	0.133

P, Probability; SD, standard deviation; **, highly significant; ***, very highly significant.

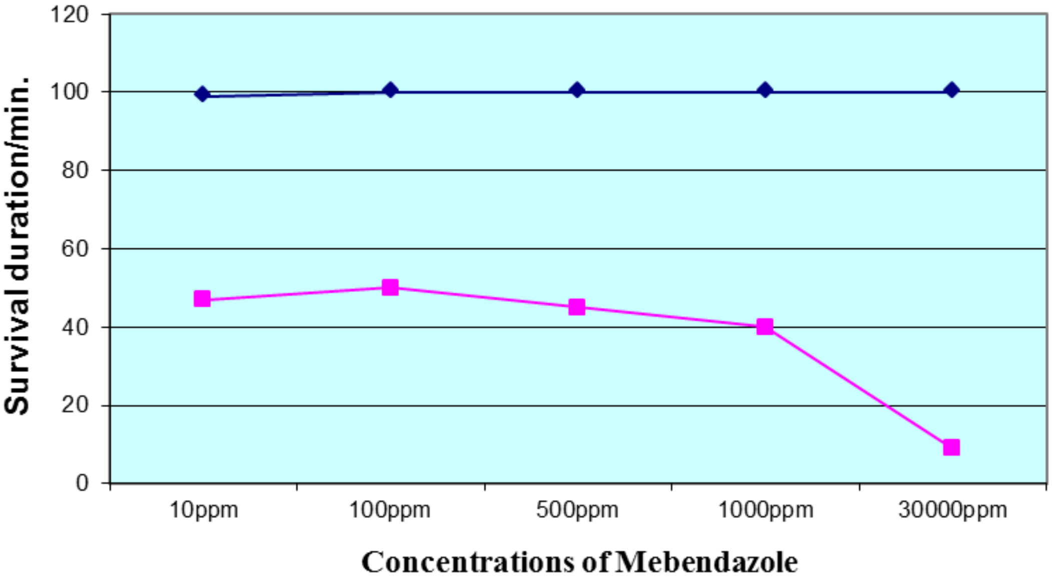


Figure (73): Efficacy of the medical drug, Mebendazole on parasitic copepod *Lamproglena minuta* by comparison control parasites.

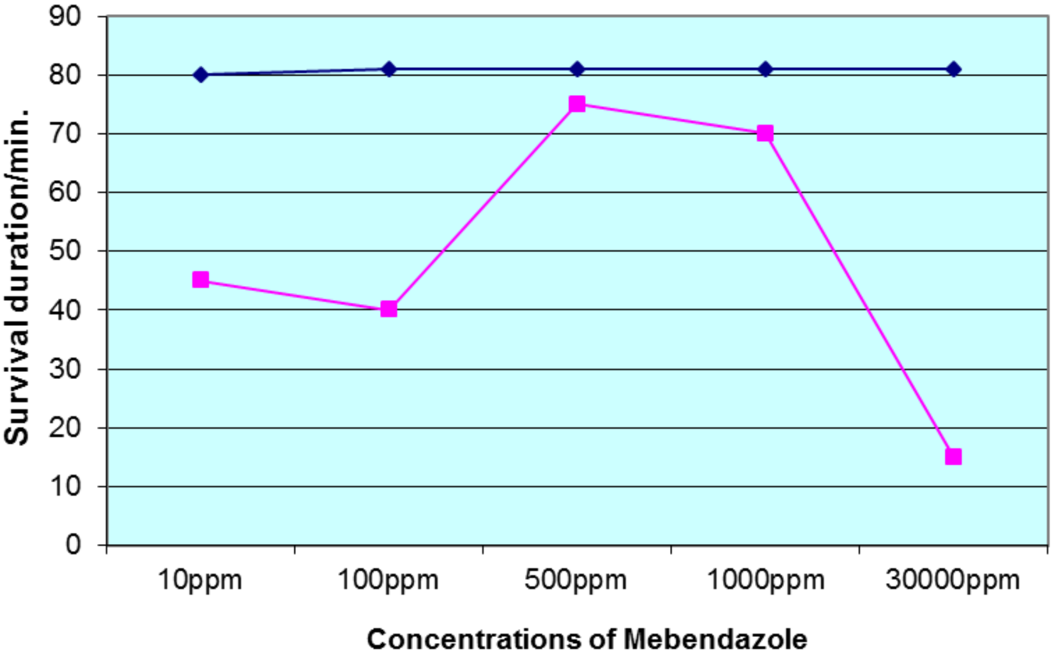


Figure (74): Efficacy of the medical drug, Mebendazole on the copepodian parasite *Ergasilus celestis* by comparison control parasites.

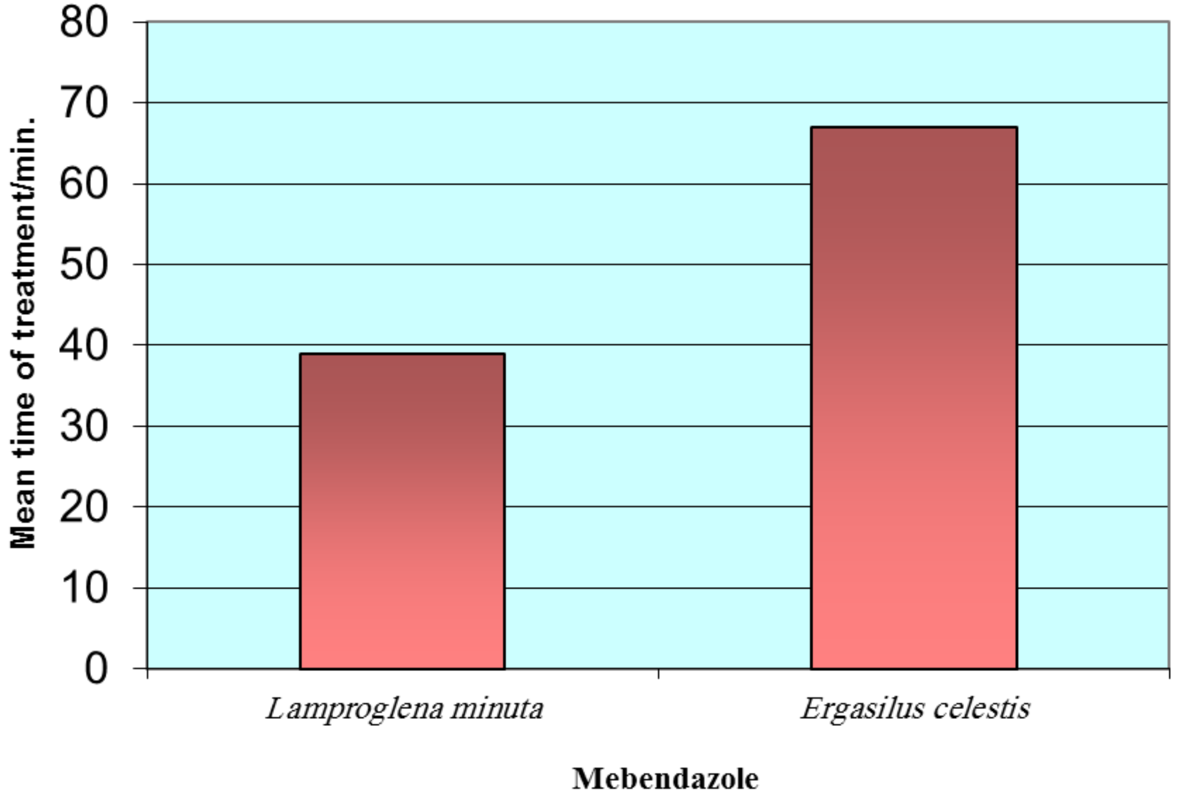


Figure (75): The relationship between the time of mortality of each parasite group and the concentrations of synthetic drug (Mebendazole).

life span of the treated parasites with increasing of drug concentrations (Table 30 & figure 73). Differences in the life span of the treated *Lamproglena minuta* with Mebendazole may be due to the susceptibility of *Lamproglena minuta*. Moreover, no abnormal or unusual behaviour was observed in any of the treated crustacean parasite, *Lamproglena minuta*.

Statistical analysis using linear regression (Table 31 and Figure 75) has revealed the presence of a very highly significant positive correlation between the concentrations of the synthetic drug (Mebendazole) and life-span of the crustacean parasite *Lamproglena minuta* (*t*-value = 6.983, P = 0.000).

II. On the crustacean parasite (*Ergasilus celestis*):

In general and comparing with the control groups, the life span of the treated parasites, *Ergasilus celestis* decreased with the increase of drug concentration. At the level of the drug effect, there was unstable decreasing in the life span of the treated parasites with increasing of drug concentrations (Table 30 & figure 74). The different of the life span of the treated *Ergasilus celestis* with Mebendazole may be due to the susceptibility of *Ergasilus celestis*. Moreover, no abnormal or unusual behaviour was observed in any of the treated crustacean parasite, *Ergasilus celestis*.

Statistical analysis using linear regression (Table 31 and Figure 75) has revealed the presence of a positive correlation between the concentrations of the synthetic drug (Mebendazole) and life-span of the crustacean parasite *Ergasilus celestis*, but the differences were not statistically significant (P > 0.05; Table 31).

Figure 75 has revealed that the parasitic copepod, *Lamproglena minuta* was more sensitive for treatment with Mebendazole than the other parasite, *Ergasilus celestis.*

2. Discussion:

The current study showed a striking discrepancy between antiparasitic copepods activity that observed in this study and that reported by previous investigators, **Ritchie *et al.* (2002)** that evaluated the clinical efficacy of teflubenzuron towards infestations of sea lice, *Lepeophtheirus salmonis* under commercial conditions at low water temperatures. Also, he recorded that the maximum efficacy of teflubenzuron towards the mean total number of lice per fish reached 77.5% at low temperature 5.4ºC. Moreover, **Branson *et al.* (2000)** found that the maximum efficacy of teflubenzuron towards the mean total number of *Lepeophtheirus salmonis* per fish reached 26% at 15 ºC. In this respect, **Botros, *et al.* (2004)** that proved the efficacy of the natural plant extract myrrh, *Commiphora molmol* on *Schistosoma mansoni* in experimental animals and they conducted that various extracts of myrrh, including the commercial product Mirazid, have clearly antischistosomal activity in infected mice or hamsters. They have tested different doses (from 180 mg/kg up to 10,000 mg/kg) of myrrh and Mirazid in mice and hamsters infected with different strains of *Schistosoma mansoni*. Animals were treated from the sixth to eighth weeks after infection. The significant worm reductions of 27% and 36% were recorded of mice receiving myrrh powder and Mirazid at the smaller dose tested. Moreover, **Leon *et al.* (2001)** used curcumin and some semi-synthetic derivatives against tripanosomatids and studied the activity of curcumin and some semi-synthetic derivatives in the literature against promastigotes (extracellular) and amatigotes (intracellular) forms of *Leishmania amazonensis*. They showed that curcumin has an excellent activity and the semi-synthetic derivatives have the best action against promastigotes forms. The literature showing the administration of *C. longa* powder in different patients with respiratory diseases, treated with 120 mg/day of the drug and a real improvement on them.

The present study clearly indicates that the parasitic copepods *Lamproglena minuta* and *Ergasilus celestis* sensitivity will be the most highly significant modulating factor in the efficacy of both natural plant extracts; Curcumin, *Curcuma longa* and myrrh, *Commiphora molmol.*

Similar findings were reported by **Massoud *et al.* (2004)** that reported that Myrrh plays an important role in the control of schistosomiasis. They determined the effect of exposing *Biomphalaria alexandrina* to sublethal dose (LC10 & LC20) of Myrrh, on its susceptibility to infection with *Schistosoma mansoni* miracidia. **Soliman *et al.* (2004)** who proved Mirazid (the oleo-resin extract from Myrrh of *Commiphora molmol* tree) is an effective fasciolicidal and schistosomicidal drug. They reported that parasitologic cure was 90.9% in fascioliasis and 100% in schistosomiasis at 4 weeks post treatment of 10 mg/kg/d given dose of Mirazid for 21 children with fasciolasis (8 males and 13 females) with mean age of 10.4 years, 8 children with schistosomiasis (6 males and 2 females) with mean age of 11.37 years. **Abo-Madyan *et al.* (2004)** who recorded that Myrrh proved to be safe and very effective in treatment of *Schistosoma haematobium* and *Schistosoma mansoni* infections under field conditions. They proved that most of 1019 patients with haematobiasis and mansoniasis were <15 years treated by Myrrh (Mirazid) as two capsules (600 mg) on an empty stomach an hour before breakfast for six consecutive days and the parasitological cure rate after three months was 97.4% and 96.2% for *Schistosoma haematobium* and *Schistosoma mansoni* cases without any side-effects. **Massoud and Habib (2003)** who found that Myrrh has molluscicidal effect on infected *Bulinus truncates* and *Biomphalaria alexandrina* snails at low concentrations (10 & 20ppm respectively) after 24 hours exposure. Also, they proved that all *Schistosoma* free cercariae were killed by 2.5ppm within 15 minutes. **Hassan *et al.* (2003)** who reported that Mirazid (the oleo-resin extract from Myrrh of *Commiphora molmol* tree) is effective in treatment of *Schistosoma mansoni* worms. They determined the effect of Mirazid on the muscle tension of single *Schistosoma mansoni* worm reached the highest response with 400 nM Mirazid and ten minutes exposure caused disruption of the tegument and collapse of tubercles. **Rasmussen *et al.* (2000)** reported the efficacy of an ethanolic extract from *Curcuma longa* against *Plasmodium falciparum* and *Leishmania major* and they found that this extract was able to inhibit the in vitro growth of these parasites.

The present study has revealed differences in the action of anti-parasitic agents against parasitic copepods. These differences may be due to the action of the effective compounds of both natural plant extracts; *Curcuma longa* and *Commiphora molmol* and synthetic drug Mebendazole and the resistance of the parasites against these compounds. In this respect, **Schmahl and Mehlhorn (1989)** found that the use of the same chemical compound might cause different actions on parasite species. **Lima *et al.* (1994)** reported that the anatomies of the same chemical compound show different actions on both sexes of the same parasite species at the exposure time. **Walker *et al*. (1996)** reported that toxicity of any compound depends on the respect, properties of the chemicals used, tested organisms and the environmental factors. **Yoshinaga, (2000)** noted that some chemicals had different effects on adult parasite and larvae even at the same concentration of the compound.

In contrast with the present study, synthetic drug Mebendazole had no significant effects on the parasitic copepod *Ergasilus celestis* that infesting *Anguilla anguilla*. On the other hand, Mebendazole had very significant effects on the parasitic copepod *Lamproglena minuta* that infesting *Sarotherodon galilaeus*. In this respect, **Roth *et al.* 1993 and Costello, 1993** used Mebendazole against sea lice and reported that Mebendazole had no effects on the parasitic copepod *Lepeoptheirus salmonis*. Moreover, **Roth *et al.* (1993)** reported that Mebendazole had no effects on the parasite copepod *Caligus elongatus*. **Stuart (1990)** reported that Mebendazole was effective against preadult and adult lice, *Lepeophtheirus salmonis* but not chalimus stages. On the other hand, Mebendazole has been used as an alternative against human nematodes and cestodes infections and has been widely used in veterinary medicine, as it is active against flukes and cestodes **(Yanong and Watson, 2005).** Also, he recorded that antibiotics are very useful additions to any fish health manager, and the ability of antibiotics to help eliminate a fish disease depends on:

1-Sensitivity of parasites to the antibiotics.

2-The intervals of treatment and dosage used.

Moreover, Mebendazole is commonly used in bath treatment; it is probably most effective against superficial infections like *Aeromonas hydrophila* on sea bream, *Sparus aurata* **Darwish and Hobbs (2005)**. Nevertheless, Mebendazole showed better efficacy (79.6 and 81.4%, respectively) against infections with *Anacanthorus penilabiatus*, gill parasites of *Piaractus mesopotamicus* at 100 mg MBZ/1 (10 min.) and 10 mg MBZ/1 (24h.) **Maurício *et al.* (2001)**.

English Summary and Conclusion

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Due to the economic importance of the marine and fresh water fishes in Egypt, it was necessary to investigate the important parasitic copepods that infesting some marine fishes; *Dicentrarchus punctatus*, *Mugil cephalus, Valamugil seheli*, *Diplodus sargus*, *Liza ramada*, *Anguilla anguilla* and *Dicentrarchus labrax* inhabiting Lake El-Manzala in Egypt and the important fresh water cichlid fishes inhabiting the Nile River, *Sarotherodon galilaeus*.

The present study was planned to study the following:

1- Detailed morphological and anatomical studies have been made to collect parasitic copepods.

2- To study of the effect of the mean intensity of parasitic copepods on the host biotic factors: length, weight and sex, as well as on the different seasons and the effect of the environmental parameters on these parasitic copepods.

3- To study of the histology of the normal (non-infected), histopathological changes of infected gills and histopathological impacts of the parasitic copepods.

4- Clinical trials were made to determine the effect of anti-parasitic agents; Curcumin, Myrrh and Mebendazole against the more abundance and intensity crustacean parasites; *Lamproglena minuta* and *Ergasilus celestis*.

First: Morphological and anatomical studies

The present study recorded that eight species of parasitic copepods belonging to four genera *Lernanthropus kroyeri, Lamproglena minuta*, *Ergasilus briani, Ergasilus lizae, Ergasilus australiensis, Ergasilus mosulensis, Ergasilus celestis* and *Caligus elongatus* were collected during the period of investigation from the gill filaments of their fish hosts, *Dicentrarchus punctatus, Sarotherodon* *galilaeus*, *Mugil cephalus*, *Valamugil seheli*, *Diplodus sargus*, *Liza ramada*, *Anguilla anguilla* and *Dicentrarchus labrax* respectively.

Second: Ecological studies

Statistical analysis was conducted using **SPSS** (version **11.5**). A mixed model analysis of variance **(ANOVA)** containing ***student-t* test** and linear regression for all the results recorded. The present study has revealed the following:

1- There is a strong positive linear correlation between numbers of *Lernanthropus kroyeri*, *Lamproglena minuta*, *Ergasilus briani*, *Ergasilus lizae*, *Ergasilus australiensis*, *Ergasilus mosulensis*, *Ergasilus celestis* and *Caligus elongatus* and increasing length and weight of their hosts. *Lernanthropus kroyeri*, *Lamproglena minuta* and *Ergasilus briani* are particularly common on small fishes **(class I)**, also, *Ergasilus lizae*, *Ergasilus australiensis*, *Ergasilus mosulensis*, *Ergasilus celestis* and *Caligus elongatus* are particularly common on small fishes **(class I)** and as well as on longer fishes **(class II and III)**.

2- In the present study, individuals of *Lamproglena minuta*, *Ergasilus australiensis*, *Ergasilus celestis* and *Caligus elongatus* were found to prefer female fishes, whereas individuals of *Lernanthropus kroyeri*, *Ergasilus briani*, *Ergasilus lizae* and *Ergasilus mosulensis* were found to prefer male fishes.

3- The present study proved that the copepodian parasite, *Ergasilus celestis* was the most abundant species, attained the highest prevalence (**58%**) and mean intensity (**37.300**) at the site **1** and **2** (especially in spring) while *Lamproglena minuta* was the most abundant species, attained the highest prevalence (**71%**) and mean intensity (**75.29**) but at site **3** (especially in summer).

4- Environmental parameters (**temperature**, **pH**, **salinity** and **oxygen content**) were recorded and possible negative correlations between these parameters and the mean intensity levels of infestation with the parasitic copepods were investigated at the three sites.

Third: Histopathological studies

The present study represents the first report of the histopathological impacts of the parasitic copepods; *Lernanthropus kroyeri, Lamproglena minuta, Ergasilus briani, Ergasilus lizae, Ergasilus australiensis, Ergasilus mosulensis, Ergasilus celestis* and *Caligus elongatus* that infesting *Dicentrarchus punctatus*, *Sarotherodon galilaeus*, *Mugil cephalus*, *Valamugil seheli*, *Diplodus sargus*, *Liza ramada*, *Anguilla anguilla* and *Dicentrarchus labrax* respectively, at the level of the light microscope. The histopathological impacts of parasitic copepods *Lernanthropus kroyeri* and *Lamproglena minuta* were limited to epithelial hyperplasia with disorganization of epithelial layers, degeneration, necrosis and massive loss of filament tissues. The five species of the genus *Ergasilus* (*Ergasilus briani*, *Ergasilus lizae*, *Ergasilus australiensis*, *Ergasilus mosulensis* and *Ergasilus celestis)* cause major lesions at the site of attachment of the gill lamellae such as hyperplasia, degeneration, chronic proliferation, epitheliocystis-like inclusions, necrosis and massive loss of filament tissues. The parasitic copepod, *Caligus elongates* cause major lesions and produce hyperplasia, degeneration and necrosis at the site of attachment of the gills of the sea bass, *Dicentrarchus labrax*.

Fourth: Effect of Anti-parasitic agents

Clinical trials were conducted to determine the effect of anti-parasitic agents the natural plant extracts; **Curcumin**, *Curcuma longa* and **Myrrh**, *Commiphora molmol* in addition to human anti-parasitic synthetic drug, **Mebendazole** against the parasitic copepods; (*Lamproglena minuta*) infesting

Sarotherodon galilaeus and (*Ergasilus celestis*) infesting *Anguilla anguilla* in which these parasites are more abundant and have highest intensity on their fish hosts among the different seasons than other parasitic copepods which were collected during the present study. The efficacy of treatment by **Curcumin**, **Myrrh** and **Mebendazole** was demonstrated as the reduction in the life span of treated parasites, compared with of control groups. The present study has revealed that parasitic copepod; *Lamproglena minuta* was more sensitive for treatment with *Curcuma longa, Commiphora molmol* and Mebendazole than the other parasite, *Ergasilus celestis.*

The present study shows a **very highly significant positive correlation** between the concentrations of anti-parasitic agents; **Curcumin**, **Myrrh** and **Mebendazole** and mortality time of the parasitic copepods, *Lamproglena minuta* and *Ergasilus celestis*.

The present study provides a contribution to our understanding of the parasitic crustacean fauna infesting fish hosts in Egypt. Since crustacean parasites are potential pathogens in fish farms, the knowledge gained by these studies may be valuable in any attempt to raise these fishes on a commercial basis for food. There are still many areas of parasite/host biology that require study. We need to know more about different life stages of the parasite, behaviour of the free-swimming larva, sites of invasion, origin and evolution of parasitic copepods and the route of migrations on the host.

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Arabic Summary

الملخص العربى

عنوان الرسالة: دراسات بيولوجية على قشريات طفيلية تصيب أنواع مختلفة من الأسماك البحرية والمياه العذبة وتأثيراتها الهستوباثولوجية

رسالة ماجستير مقدمة من

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نظرا للأهمية الاقتصادية للأسماك البحرية والأسماك المستوطنة في مياه نهر النيل حيث تمثل مصدرا هاما من مصادر البروتين الحيواني في مصر وبالإضافة إلى التأثير الضار التي تسببه الطفيليات القشرية على صحة هذه الأسماك الأمر الذي يؤدي بالإضرار بالثروة السمكية فقد تم في هذه الرسالة عمل دراسات شملت أربعة مواضيع بالطفيليات القشرية والتي تصيب بعض الأسماك البحرية محل الدراسة وهي النقط (دايسنترارکس بنکتيتس)، البوري (ميوجل سيفالس)، السهيلي (فالاميوجل سهيلي)، الشرغوث (ديبليدوس سارجوس)، الطوبارة (ليزا رمادا)، الحنشان (أنجويلا أنجويلا)، القاروص (دايسنترارکس لابرکس) وأسماك مياه النيل والتي تتمثل في أسماك البلطي "البلطي الجاليلي" (ساروسيردون جاليلاس).

وإهتمت الدراسة الحالية بدراسة الأربع موضوعات التالية:

الموضوع الأول: دراسة النواحي التصنيفية والمورفولوجية والتشريحية للطفيليات القشرية التي تم عزلها من بعض الأسماك البحرية وأسماك المياة العذبة المستوطنة لمياه نهر النيل.

الموضوع الثاني: دراسة تأثير كثافة الإصابة للطفيليات القشرية على العوامل الحيوية للعوائل السمكية الثمانية ومتوسط كثافة الإصابة بتلك الطفيليات خلال الفصول الأربعة وكذلك تأثير العوامل البيئية المائية على هذه الطفيليات القشرية.

الموضوع الثالث: دراسة نسيجية لأسماك المياة البحرية والمياة العذبة محل الدراسة والآثار الهستوباثولوجية لبيان التأثير المرضي لهذه الطفيليات القشرية.

الموضوع الرابع: تجربة بعض المستخلصات النباتية الطبيعية مثل الكركم ومر البطارخ وكذلك المركبات الدوائية المصنعة على حيوية هذه الطفيليات لمعرفة كيفية مقاومتها.

في هذا الصدد تم تجميع 900 سمكة من أسماك المياة البحرية والمياة العذبة محل الدراسة من ثلاث مواقع بمحافظة دمياط بجمهورية مصر العربية؛ الموقع الأول: قرية شطا، الموقع الثاني: قرية الديبة، وقدتم تجميع أسماك البلطي فقط من الموقع الثالث: مدينة الزرقا وقد تم فحصها خلال الفترة من شهر إبريل 2005 حتى شهر مارس 2006 بحثا عن الطفيليات القشرية موضوع الدراسة.

وقد أمكن من هذه الدراسة التوصل إلى عدة نتائج من أهمها ما يلي:

أولا: الدراسات التصنيفية والمورفولوجية والتشريحية

أظهرت الدراسة الحالية أن هناك ثمانية أنواع من الطفيليات القشرية تم عزلها من الخيوط الخيشومية لأسماك المياة البحرية والمياة العذبة محل الدراسة وهم (ليرنناثروبث كرويري) والذي تم عزله من الخيوط الخيشومية لأسماك النقط، وطفيلي (لامبروجلينا منيوتا) الذي تم عزله من الخيوط الخيشومية لأسماك البلطي، والطفيلي (إرجسيلاس بيرياني) الذي تم عزله من الخيوط الخيشومية لأسماك البوري، والطفيلي (إرجسيلاس ليزي) الذي تم عزله من الخيوط الخيشومية لأسماك السهيلي، والطفيلي (إرجسيلاس أستيورلانسس) الذي تم عزله من الخيوط الخيشومية لأسماك الشرغوث، والطفيلي (إرجسيلاس موسيولينسس) الذي تم عزله من الخيوط الخيشومية لأسماك الطوبارة، والطفيلي (إرجسيلاس سيليستس) الذي تم عزله من الخيوط الخيشومية لأسماك الحنشان والطفيلي (كاليجس إلونجاتس) الذي تم عزله من الخيوط الخيشومية لأسماك القاروص وقد أثبتت الدراسة تسجيل هذه الطفيليات القشرية لأول مرة في مصر على خياشيم هذه الأسماك وقد تم عمل رسومات تفصيلية للأنواع الطفيلية سالفة الذكر.

ثانيا:الدراسات البيئية

قامت الدراسة الحالية بعمل دراسة شاملة لتأثير كثافة الإصابة للطفيليات القشرية على العوامل الحيوية للعوائل السمكية الثمانية مثل طول ووزن وجنس العائل؛ درجة السيادة و متوسط كثافة الإصابة بتلك الطفيليات خلال الفصول الأربعة وقد أظهرت الدراسة أيضا تأثير العوامل البيئية المائية على هذه الطفيليات القشرية.

جميع النتائج التي رصدت خلال الدراسة الحالية تم تحليلها إحصائيا بإستخدام البرنامج الإحصائي (SPSS (version 11.5 وطريقة إختبار *t* (*student-t* test) وعلاقة (Linear regression) وقد أسفرت الدراسة عن النتائج التالية:

1- أن أعلى متوسط إصابة بالطفيليات القشرية تم رصده إما على الأسماك الصغيرة أو متوسطة الطول.

2- وجود علاقة وثيقة بين متوسط كثافة الإصابة لجميع الطفيليات من جهة وجنس العائل من جهة أخرى حيث سجلت إناث العوائل السمكية كثافة إصابة أعلى من الذكور.

3- أثبتت الدراسة الحالية أن الطفيلي (إرجسيلاس سيليستس) هو الأكثر سيادة والأعلى كثافة على خياشيم العائل السمكي (أنجويلا أنجويلا) فى الموقع الأول والثاني خلال الفصول الأربعة بالإضافة إلى أن الطفيلي (لامبروجلينا منيوتا) كان غائبا في هذه المواقع. على الجانب الآخر كان الطفيلي (لامبروجلينا منيوتا) هو الأكثر سيادة والأعلى كثافة على خياشيم أسماك البلطي (ساروسيردون جاليلاس) في الموقع الثالث أيضا على مدار السنة.

4- وأن الطفيلي (إرجسيلاس سيليستس) هو الأكثر شيوعا محققا أعلى نسبة إصابة **58%** وكثافة **37.300** طفيلي/سمكة مصابة في الموقع الأول والثاني (خصوصا في فصل الربيع) بينما سجل الطفيلي (لامبروجلينا منيوتا) أعلى نسبة إصاية **71%** وكثافة **75.29**طفيلي/سمكة مصابة في الموقع الثالث (خصوصا في فصل الصيف).

5- أثبتت التحليلات الإحصائية أن العلاقة بين متوسط الكثافة لجميع الطفيليات في كل المواقع

والعوامل البيئية (درجة حرارة الماء، درجة الأس الهيدروجيني pH، الملوحة و محتوى الأكسجين) كانت سلبية ذات مغزى في المواقع الثلاثة.

ثالثا: الدراسات النسيجية والآثار الهستوباثولوجية

أوضحت الدراسة الحالية تركيب النسيج الطبيعي (الغير مصاب) لخياشيم العوائل السمكية الثامنية (النقط، البلطي، البوري، السهيلي، الشرغوث، الطوبارة، الحنشان و القاروص) موضع الدراسة بإستخدام الميكرسكوب الضوئي.

وقد بينت هذه الدراسة أن التأثيرات المرضية الناجمة عن الإصابة بالطفيليات القشرية (ليرنناثروبث كرويري)، (إرجسيلاس بيرياني)، (إرجسيلاس ليزي)، (كاليجس إلونجاتس)، (إرجسيلاس أستيورلانسس) و (إرجسيلاس موسيولينسس) قد اقتصرت علي تلف أو موت في خلايا النسيج الخيشومي، النمو الغير طبيعي لخلايا النسيج الخيشومي و كذلك تكون بثرات خلوية في مكان الإصابة بالطفيلي للعوائل السمكية النقط،البوري، السهيلي،القاروص، الشرغوث و الطوبارة. على الجانب الآخر كانت إستجابة النسيج الخيشومي بالإصابة بالطفيلي القشري (لامبروجلينا منيوتا) لأسماك البلطي متمثلة في تلف أو موت النسيج الخيشومي للخيوط الخيشومية الأولية والثانوية. وتمثلت الآثار الناجمة عن الطفيلي (إرجسيلاس سيليستس) في تقسيم الخلايا الطلائية للخيوط الخيشومية الأولية والثانوية وتضخم الخيوط الخيشومية وتكون مايشبه بأكياس داخل الخيوط الخيشومية عند قواعد الخيوط الخيشومية الثانوية وأطراف الخيوط الخيشومية الأولية.

رابعا: تأثير بعض المستخلصات النباتية الطبيعية و المركبات الكيميائية على حيوية الطفيليات القشرية وكمواد مقاومة لوجود هذه الطفيليات

خلال تلك الدراسة تم عمل إختبارات أولية لتحديد تأثير المستخلص النباتي لريزومات الكركم ومر البطارخ بالإضافة إلى عقار مضاد للطفيليات التي تصيب الإنسان (مبيندازول) على الطفيليات القشرية (لامبروجلينا منيوتا) و (إرجسيلاس سيليستس) التي تصيب العوائل السمكية البلطي والحنشان على التوالي حيث كانت هذه الطفيليات الأكثر إنتشارا والأعلى كثافة خلال الفصول الأربعة عن باقي

الطفيليات القشرية التي تم تجميعها أثناء البحث الحالي.

وقد تم إستخدام أربع تركيزات لكل مركب 10، 100، 500، 1000 جزء في المليون على التوالي بالإضافة إلى تركيز المحلول الأصل لكل مركب وأوضحت هذه الدراسة أن فاعلية كلا من الكركم ، ومر البطارخ و العقار (مبيندازول) تختلف على أساس حساسية الطفيلين (لامبروجلينا منيوتا) و (إرجسيلاس سيليستس) خلال معالجتهم بهذه الأدوية. وسجلت الدراسة فاعلية الكركم ومر البطارخ على فترة حياة الطفيلين عند درجة حرارة 27 ^{o}س وفاعلية المبيندازول عند30 ^{o}س. وأثبتت الدراسة أيضا أن فترة حياة هذه الطفيليات تقل مع زيادة تركيز الكركم ومرالبطارخ وكذلك المبيندازول. كما أظهرت الدراسة أن الطفيلي (لامبروجلينا منيوتا) كان أكثر حساسية لكلا من الكركم ومرالبطارخ وكذلك المبيندازول عن الطفيلي (إرجسيلاس سيليستس).

ساعدت الدراسة في توضيح بعض المفاهيم الهامة عن بيئة هذه الطفيليات القشرية علي عوائلها السمكية في مياه بحيرة المنزلة ومياه نهر النيل بمصر. أوضحت الدراسة أيضا بعض الحقائق التي تساعد على فهم كيفية حدوث الإصابة بالطفيليات القشرية. بالإضافة أنها أكدت بعض المفاهيم القيمة التي يمكن الاستفادة منها في عملية استزراع هذه الأسماك.

كما أنه لا يزال يوجد بعض الثغرات التي لم يتم دراستها في علاقة الطفيل بالعائل حيث نود معرفة المزيد عن أطوار الحياة المختلفة ، و سلوك اليرقات السابحة، و أماكن الإصابة بهذه اليرقات في النسيج الخيشومي، منشأ وتطور أو تحول القشريات الطفيلية و كيفية هجرتها علي جسم العائل السمكي.

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قسم علم الحيوان

دراسات بيولوجية على قشريات طفيلية تصيب أنواع مختلفة من الأسماك وتأثيراتها الهستوباثولوجية

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الماجستير في علم الحيوان

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