# EFFECTS OF EUSTRONGYLIDES AFRICANUS ON CLARIAS GARIEPINUS FROM SOME PARTS OF NIGER STATE, NIGERIA.

## BY

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## A THESIS SUBMITTED TO THE POST GRADUATE SCHOOL, FEDERAL UNIVERSITY OF TECHNOLOGY, MINNA IN PARTIAL FULFILLMENT OF THE REQUIREMENT FOR THE AWARD OF M.TECH FISHERIES TECHNOLOGY.

# DEPARTMENT OF WATER RESOURCES, AQUACULTURE AND FISHERIES TECHNOLOGY, FEDERAL UNIVERSITY OF TECHNOLOGY, MINNA, NIGER STATE.

# AUGUST, 2007.

## DEDICATION

This thesis is dedicated to my late parents and my wife Mrs. Christiana Jiya, Children: Solomon Suman Jiya, Samuel Yisa Jiya, Elizabeth kakaraba Jiya and James Babakeke Jiya (Junior).

## DECLARATION

I hereby declare that this thesis is original work done by me and has not been previously presented for any qualification at any other institution.

All information that emanated from other works either published or unpublished have been duly acknowledged.

30th Aug, 2007

James Jiya

Date

## **CERTIFICATION**

This thesis titled Effects of Eustrongylides africanus on Clarias gariepinus from some parts of Niger state, Nigeria by Jiya. James (M.Tech/SAAT/2001/792) meets the regulation governing the award of the degree of M. Tech. fisheries technology from the Federal University of Technology Minna and is approved for its contribution to scientific knowledge and literary presentation.

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V

#### ABSTRACT

This study was undertaken to investigate the effects of *Eustrongylides africanus* on *Clarias gariepinus* from some parts Niger state, Nigeria between November, 2004 and April, 2005.

Three hundred and sixty (360) samples of *Clarias gariepinus* of various sizes, sex and age were used for the study. The fish sampled were caught by fishermen fishing within Rivers and man-made dams. The sampling/landing sites were: Shiroro dam, Tagwai dam, Suleja dam, Wushishi dam, Kagara dam and Matandi dam, Kakakpangi, Wuya kede and Katcha. Morphometric measurement recorded for the fish sampled include: Total length (TL) standard length (SL) parasite count (pc) and liver weight (L.wt). The colour of the liver of both infected and uninfected fish sampled did not show any difference, it was observed to be reddish brown. The mean values recorded for body weight, standard length, liver weight and condition factor for the three geopolitical areas are as follows:- 181.2+100.8, 25.24+5.31, 1.86+1.10, 1.414; 182.5+98.6, 23.88+4.24, 1.73+1.03, 1.334; 173.3+3.10, 1.62+0.73, 1.289. Conditions factor for the three areas ranged between 1.289 - 1414. Mean values for body weight standard length and condition factor for the infected and uninfected fish sampled were 220 + 110g, 25.3 + 5.4cm, 2.8 and 169.8 + 83g, 23.5 + 3.5cm and 2.0 respectively.

The length –weight relationship between the uninfected and infected fish sample in the study area showed allometric and isometric growth represented by the formulae Y=4.90+3.15 and Y=2.07+2.28. *Eustrongylides africanus* found on the fish samples were quite few (1-11) and did not have any adverse effect on the condition of the *Clarias gariepinus* sampled.

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#### **CHAPTER ONE**

#### **1.0 INTRODUCTION**

#### 1.1 FISH DISEASE CONCEPT

Fish disease is said to be an illness affecting fish population and it is a reflection of improper environmental management. The interaction between the host (fish), environments (water) and pathogen (parasites) cause disease (Sniesko, 1983). Meyer (1983) defined parasite as an organism that lives in or on another organism (the host) and depends on the host for it's nutrition and is suspected of harming the host. Fish disease is caused by parasites, fungi, environmental (Paperna, 1996) or predatory (Hine, 1993). Mbuthia (1993) and Hine (1993) observed that like other animals, fish reproduction, growth, appearance and condition are affected by parasites as they are usually potential sources of discomfort.

Basically, fish is an important source of cheap animal protein to man. It is however, worthy to note that parasitic infection reduces the relative value and palatability of fish (Onusiriuka, 2001). French (1965) recognized that studies on the parasites of fish are very important as these parasites may effect fish production.

### **1.2 JUSTIFICATION**

The physical presence of *Eustrongylides africanus* in *Clarias* species is of concern to fishermen and fish consumers in Niger state. According to Anthony (1982), *Clarias gariepinus* is one of the most resistant, widely accepted and highly valued fish that could be cultivated apart from being harvested from the wild.

Fish production from the capture and recently from fish culture, fisheries in Niger state show there is the need to acquire knowledge about the nature and effect of fish parasites. Fish of the genus *Clarridae* are very common throughout the year in Niger state and they form important commercial catches. They are constantly disposed to infection by parasites particularly *Eustrongylides* species (Paperna, 1996). The reasons for their susceptibility could be as a result of their uncovered soft skin. *Clarias gariepinus* is a hardy fish, fast growing and widely acceptable. Inspite of their relative importance as one of the major source of fish protein in the state, documented information on their conditions due to *Eustrongylide* infection is limited and scanty. The above reasons, therefore forms the basis for this investigation of pathological effects and conditions of the infected fish.

The importance of animal protein particularly fish to human beings cannot be over-emphasized as it provides the body with essential nutrients required for growth and other related functions. In order to investigate the effects of *Eustrongylides* parasite infected *Clarias gariepinus* three geopolitical areas of Niger state were used for this study.

Prevalence of *Eustrongylides africanus* infected *Clarias gariepinus* has been observed in the state and investigation has only been restricted to water bodies and fadama within Bida area.

#### **1.3 OBJECTIVES OF THE STUDY**

## **General objectives**

1. To investigate the effects of *Eustrongylide africanus* on *Clarias gariepinus* in three geopolitical areas of Niger state

## **Specific Objectives**

- 1. To identify the type of Eustrongylides that affect Clarias gariepinus
- 2. To determine body part mostly affected in the fish.
- To determine the level of infection of *Eustrongylides africanus* on *Clarias gariepinus* in Niger state.
- 4. To Investigate the spread of *Eustrongylides africanus* in some water bodies from three geopolitical areas of Niger state.

## 1.4 SCOPE AND LIMITATION

The research is only limited to *Eustrongylides africanus* infection of *Clarias gariepinus* species from some water bodies in the three geopolitical areas of Niger state. The scope is only to investigate level of infection and effects on fish production from these areas.

Different parts of the fish body, both internal and external are known to harbour parasites, however, the parts which this study limited its investigation and examinations on are the skin, fins, gill region, abdominal cavity, liver and muscles. Live fish were examined individually for parasites according to the method described by Brent R. Dixon (2006).

#### **CHAPTER TWO**

#### 2.0 LITERATURE REVIEW

#### 2.1 VULNERABILITY OF FISH TO PARASITES

Many phyla of the animal kingdom have representatives which are parasitic on fish. The number of species of fish parasites are in thousands and many more remained to be discovered. Very few are seriously harmful to fish. Most individual fish in the wild or cultivated populations are infested with parasites, but in the great majority of cases, no significant harm appears to be caused to the host fish. Although, there are surprisingly few reports of parasites causing mortality or serious damage to wild fish populations, such effects often go unnoticed. Catfish are vulnerable to fish parasites, because of favourable environmental factors, there soft and uncovered skin and as parasites form parts of the food consumed by them. Parasites in the wild fish are only remarked upon when they are so obvious as to lead to rejection of fish by fishermen or consumers (Paperna, 1980).

## 2.2 GEOGRAPHICAL SPREAD

Paperna (1996) reporting and quoting observation of other workers on geographical spread of nematodes in Africa listed the following occurrences: *Contracaecum* in Egypt, Mali, most large and small East African (Rift Valley) lakes (including lakes kivu, Edward, and Albert –campana – Rouget 1961). Zaire, Mali (Niger), (Khalil, 1971) and South Africa, where it was reported from brackish water hosts (Boomker, 1982; Van As and Basson, 1984).

Infections of the pericardia in cichlid fish occur in lakes Victoria, George, Nakuru, Naivasha, Baringo and Magadi (Paperna, 1974a, Malvestuto and Ojambo Ongoma, 1978). *Amplcaecum* was reported from Sudan (Khalil, 1969) and Diyardinascaris from lakes Chad and Tangayika. *Eustrongylides* has, thus far, only been found in the East Africa lakes, including lake Tangayika (Campana-Rouget, 1961; Khalil, 1971; Paperna, 1974b) and freshwater Fadama around Bida, Niger state (Ibiwoye et al 1996).

## 2.3 OUTBREAK AND SIGNS OF FISH DISEASE

Disease outbreak according to Rogers and Plumb (1977) are more common during certain times of the year, due to various environmental factors or due to the life cycle of the causative agent. They observed that the peak times of the year for this outbreak are in spring, summer and fall when fishermen are most likely to be enjoying the sports. There is much overlap of clinical signs (symptoms) and outward manifestations of different diseases, but some disease can be readily identified.

In general, the most common signs of a disease include:- haemorrhage, lesions (scores, cysts or erroded fins) discoloration, fluid accumulation in the body cavity, popeyes, sliminess of the skin, irregular swimming or sluggishness and gasping near the surface. Many atimes, a combination of these signs could be present.

## 2.4 FISH DISEASE TRANSMISSION

Maar *et al* (1966) and Huet (1972), observed that it would be almost impossible for parasites found in infected fish flesh to infect man, except through raw, improperly cooked or partially sun dried fish and parasites such as tapeworms, but the thought of eating these parasites is most unlikely.

A lot of parasites are found abundantly in fish where water birds, the final host congregate. Nematodes and tapeworms are commonly found in the visceral organs of fish. One of the most common is a large nematode: *contracaecum*, often found coiled like a watch spring encysted in the mesenteries. Another nematode *philometra* usually found behind the eye may sometimes be located in the body cavity. Generally, report have it that parasites infect fish through the fish's food or through the gills or via attachment of skin, before gaining entrance into the viscera of fish (Oniye, 2000).

#### 2.5 PARASITES AND PARASITE LIFE CYCE

Ahmed (1997) described parasitic diseases as diseases that are caused by organisms, which lived in or on other animals known as hosts. Fish parasites include: protozoans, worms and crustaceans. They could be internal or external and feed on the fluid or the tissues of their hosts. Ricker (1970) reported that fish may contain many parasites yet seen perfectly healthy. He also observed that under certain circumstances, any parasitic species may increase the number and directly or indirectly cause the death of a fish. In nature, majority of common parasitic species appear to have no serious effect on their host fish. He further said that the experience of most parasitologist thus lead them to believe empirically that fish can harbour considerable numbers of most kinds of parasites without being adversely affected. He pointed out that the fish biologist on finding the gut of his fish full of worms may be equally convinced that such parasites must be harming the fish.

Dick and Harvey (1986) reported that fishes often find themselves acting as host to some very unwholesome guests. Most parasites feed on the mucus layer on the skin and gills, larger ones penetrate the tissues and feed on blood and tissue fluids. Similarly, Maar et al (1966) noted that under natural condition in the wild (rivers and lakes) most fish species have parasites and that fish however have "learnt to live' with them and consequently do not suffer greatly, if at all from the infection. Huet (1972) corroborated these statement and further said that fish are widely dispersed and diseases are often not noticed as the risk of contamination are fewer and the losses are low.

Petrushevski and Shulman (1970) reported that in freshwater environments severe infestation of fishes with nematodes- *Rhaphidascaris acus* (Bloch, 1779) leading to mass moartality occurred in lake Sudochye in the neighbourhood of Muynak, the Delta of the Amu-Daria (Osmanov, (1953) in Petrushevski and Shulman (1970). The worms were found in *Esox lucius, leuciscus idus, Scardinus erythrophthalmus, Aspius aspius, Tinca tinca, Carassius carassius, Lucioperca lucioperca* and *Cyprinus carpio.* The most severely infested fish was *Abramis brama* in which the intensity reached 1,035. The worms were localized in the intestinal walls and in the liver, which was seriously damaged. All the functions of the liver, gut, the gonads and other organs were made totally impossible, it was also reported that among marine fishes, the disease due to nematode infestation are very common.

In another investigation, Okaeme (1991) quoting Hoffman and Bauer (1971) reported that the prevalence of parasites of freshwater fishes under natural conditions is usually low when compared to that of fish intensely managed because of the large expanse of water, rapid rate of water exchange and reduced risk of contact between parasites and fish under natural conditions. A number of factors, he observed have been found to be responsible for the prevalence of freshwater parasite in lakes and reservoirs. These include availability of intermediate hosts responsible for sustenance of parasites life cycle, host population changes and the ecology of the water body (Hoffman, 1976).

Ibiwoye et al (1996) reported that just like animals, fishes are subjected to parasites, diseases and predation, and these hampers their reproduction, growth, appearance and well being. In Africa, under culture system, fish are raised in extensive system at low stocking densities with reduced problems of parasites and pathogens, due to inadequate diagnostic facilities. Most of the early observations on diseases of tilapia were related to parasitic infection, often from the wild fish and at low levels. These have generally, shown no evidence of clinical effects on the fish. While in most wild populations of tilapia, it seems that parasitism is a normal occurrence of little consequence.

Ricker (1970) reported that many of the definitely serious fish parasites are not adult in fish and need to be eaten by a predator to complete their life cycle. Thus, it is to their advantage to weaken or even kill the fish. Thomas (1964); Richer (1970), gave a useful survey of the literature of harmful larval parasites of fishes and stated that the stage which are found in piscivorous vertebrate are apparently not pathogenic. He further reiterated that, it is useful to simply assume that all parasites deprive the fish host of its anabolic materials and thus the total parasitic burden is viewed as a measure of the degree to which the fish is weakened below its optimal condition. Paperna (1996) observed that the life histories of some parasites which (at the adult stage) infect African fish have so far not been studied and their first molluscan host and other invertebrate hosts remain unknown. Davies (1946): Hoffman (1967) and Schell (1970): in Paperna (1996) further remarked that investigations carried out could be summarized as: eggs of gut dwelling digeneans are released via defecation: while eggs of those living in the gall bladder are evacuated into the gut with bile. Eggs produced by digeneans in the kidney or gonads are evacuated from their host with the respective organs products. If they are located in the tissue or closed internal cavities they can only be liberated following death of the hosts or predator.

Work on fish parasites in Nigeria, especially those prevalent in Niger state includes: Awachie-(1966) Parasities of fish in the area of Kainji reservoir; Ukoli-(1966) Helminth infection of fish in the River Niger; Ibiwoye et al (1996) Prevalence of endoparasite in some commercially important freshwater fishes of Bida area, Niger state, Nigeria. Ibiwoye et al (1996) Prevelance, intensity and abundance of *Eustrongylides africanus* larvae in Clarias species of freshwater fadama of Bida area, Nigeria; Okaeme (1991) Helminth fauna of tilapia of lake Kainji in the pre and post impoundment conditions.

### 2.6 DESCRIPTION OF NEMATODES

Nematodes are fairly frequent parasites and occurs worldwide in all animals (Reichenbach- Klinke's 1973). They are encased in a very tough and impermeable transparent or semi-transparent cuticle. The cuticle is not chitin like the cuticle of arthropoda since it is soluble in potassium hydroxide, but have true chitin in the egg shell. Paperna (1996) described all freshwater and brackish water fish as being infected, with heavier infection in predatory fish especially species also utilizing fish as intermediate or transient host.

Duijn (1973) reported that nematodes live in fish during their larval stage, whilst the adult form is produced when the fish is eaten by the definite host. The larvae, he said have a size of a few millimeter and live for a short period in the skin or in the internal organs then they encyst. Generally, the cyst are the size of a pinhead, although larger ones may occur. They are formed outside the intestine or in the peritoneum, pancreas, liver or other internal organs of a fish may occur.

Kabata (1985) apparently observed that nematodes maintain their hold on their hosts by using buccal apparatus when not encysted in the tissues. They also have well developed alimentary canals and that they can use fish either as intermediate or definitive hosts, living in them as encysted larvae or as lumendwelling adults.

Nematodes vary generally in body structure ranging from long, round, threadlike, flat, unsegmented smooth bodied. The cylindrical body tapers at both ends. The mouth is terminal anteriorly. The gut is clearly divided into oesophagus and an intestine. In the species parasitic on fish, at least one host is required. Since females are spindle-shaped or twisted when gravid, there are no appendages in the parasitic species. sexes are separate with females generally larger than the males.

A pseudocal or false body cavity is present, the tail consists of that part of the body posterior to the anus. The anterior end of the body forms the head, the mouth is terminal and associated with it are various structures such as lips, pseudoobia, odontia, cephalic papillae, amphids, collarettes and cordons. The primitive number of lips is six (6) of which two (2) are dorsal two (2) lateral and two (2) ventral. One of these triangular lips are three sensory papillae. One at the tip near the opening of the mouth and two at the base. One on eachside, papillae at the tips of the lips form the inner circle and those at the base constitute the outer circle (Paperna 1996).

## 2.7 IDENTIFICATION OF LARVAL NEMATODES

Identification of larval nematodes, particularly to species level is not usually feasible, since the larvae lack genital systems and several other features of adult stages which are used as taxonomic criteria. In recent years, a method of identification of larval stages of *Anisakidae* by biochemical (Isoenzyme) method utilizing multilous electrophoresis analysis has been developed (Paperna 1996; Orechia et al, 1986).

*Rhabdochona* and *Spnitectus*. Paperna (1996) said are very small (<10mm in length) the former shows dentaton in its mouth opening, while the cuticle of the latter bears circular rows of spines.

*Eustrongylides* are large long red worms, 18-70mm long. 0.3-0.8mm thick, with a long oesophagus merging with an indistinct ventriculum (Paperna. 1996).

### 2.8 LIFE CYCLE OF NEMATODES

Paperna (1996) reported that larval nematodes are potentially pronounced in all fresh and brackish water fish, with heavier infection occurring in fish occupying higher positions in the food-chain e.g predatory fish.

The life cycle of nematodes are of two basic types: direct or monogenous (i.e with only one host in the cycle) and indirect or heterogenous (ie with two or more host in the cycle). The basic pattern of development is similar whether the life cycle is direct or indirect. Larvae hatching from the eggs progress through a series of stages in their development. Beginning with the first stage, each one is separated by a molting of the cuticle (Paperna, 1974). There are four larval stages i.e 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup> and 4<sup>th</sup> followed by the adult. The 3<sup>rd</sup> stage is infective to

the final host. The pattern of growth of the larvae (L) and occurrence of the successive molt (m) may be expressed as follows:-

$$\operatorname{Egg} \to L_1 + M_1 \to L_2 + M_2 \to \overline{L_3 + M_2} \to L_4 + M_4 \to \operatorname{Adult}$$

In many parasitic nematodes (orders *Rbabditida strongylida*) with a direct life cycle, the 1<sup>st</sup>, 2<sup>nd</sup> and 3<sup>rd</sup> stages are free in the soil. The 2<sup>nd</sup> stage feed on organic material. The 3<sup>rd</sup> stage which retains the shed cuticle of the 2<sup>nd</sup> stage as enclosing sheath is unable to ingest food. In some species the 1<sup>st</sup> (Ascaris) and 2<sup>nd</sup> molts some hookworms, *strongyles* and *trichestrongyles* take place inside the egg shell. The 3<sup>rd</sup> stage larvae enter the final host when swallowed with food or by penetrating the skin, as in the case of hookworms. The 3<sup>rd</sup> and 4<sup>th</sup> molts are completed inside the final host.

Reichenbach Klinke's (1973) observed that in an intermediate host, parasites are present as larval or juvenile forms. Such life cycle are often necessary to ensure the dissemination of infective stages of parasites to the final host in which maturity will occur. Fish are utilized as intermediate hosts by parasites. Intermediate host often form part of the diet of the final host or the next intermediate host in the life cycle. In other cases, free living stages may be released from the intermediate host which actively invade, or are eaten by a further host. Many fish parasites spend atleast some part of their life cycle outside a host. The parasite species with a direct life cycle, infect other hosts or by means of free swimming larvae, which often actively invade the host or by spores or eggs which are ingested. Paperna (1996) further disclosed that parasites with an indirect life cycle have the same stage, but reach the final host through the means of a vector or intermediate host, usually an anthropod, but other invertebrates are utilized. Vectors are transmitters of parasites, if the transmitter is essential in the life cycle of the parasite, it is a biological vector, if it is unessential, it is a mechanical vector.

In Oviparous forms with the indirect cycle, the eggs are embryonated, when laid and in most cases hatch only when ingested by the intermediate host in whose body the 3<sup>rd</sup> or infective larval stage is reached through the necessary molts. In some of the lungworms the egg hatch on the ground or in the faeces of the final host and the larvae penetrate land snails, which serve as the intermediate host. Infection of the final host occurs when the intermediaries containing the infective larvae are swallowed. The 3<sup>rd</sup> and 4<sup>th</sup> molt take place in the final host.

The knowledge of parasite life cycle is often essential. If successful preventive measures are to be achieved as it allows the parasite to be attacked at the most vulnerable point of its life cycle.

#### 2.9 NEMATODE INFECTION

Nematode infection are common in predatory fishes, lesions and gonadal deformation occurs in cichlids of the genus *haplochromis*, larval nematodes (*Eustrongylides* and *contracaecum*) causes lesions in the dermis and deformation of gonads of *Bagrus docmac* and *Clarias species* (Oniye, and

Annune 1993). They observed that good example of nematodes that are found in wide range of fishes are *Procamallanus laevionchus*. Paperna (1996) described nematodes as being able to infect all organs of the host and particularly causing loss of function of the damaged area. Signs of nematodiasis, they said include:- anaemia, emaciation, unthriftness and reduced vitality.

#### 2.10 LARVAL NEMATODES

Most notorious larval nematodes, are representatives of the anisakdae (Haeterocheilidae): genera Ampicaecum, contracaceum and perrocaecum, dioctophymidae, the genus Eustrongylides and Rhabdochonidae, the genera Rhabdochona and Spinitectus. Furthermore, larval nematodes, occur either encysted in tissues or free in body cavity most often in the abdominal or pericardial cavity. Larvae of contracaecum and Eustrongylides tend to escape from their cyst and out of their host body after its death Paperna (1996).

Nematode eggs are released via defecation by definitive hosts e.g pelican, herons and cormorants. They are also released into water when whole nematodes are vomited from the stomach by regurgitation. Eggs are released from such discharged nematodes by oviposition or after death following their decomposition. Eggs hatch within 2-3 days at  $24^{\circ}$ C; 5 – 7 days at  $21^{\circ}$ C; hatching is not simultaneous and is further delayed in some eggs. Free living infective (second) stage larvae can survive in water for several months. Larvae

become firmly attached by their posterior end to a substrate in the aquatic habitat (Paperna 1996).

The intermediate hosts of *Eustrongylides*, Paperna (1996) reported are unknown, although Oligochaetes are the first intermediate hosts of a related dioctophymatiod from the genus *Hystrichia*. However, the later, he said do not develop via fish. In fish, larval infection passes from prey (Cichid fish, mainly *Haplochromis*) to predator, finally accumulating in the predatory catfish *Bagrus docmac*, *Clarias gariepinus*\_and also in the lung fish *Protopterus aethiopicus*. Numerous adult *Eustrongylides* species were found attached to the stomach of cormorants (*Phalacrocorax africanus*) obtained from the same habitat (in Entebbe, Lake Victoria) where fish were heavily infected. In conclusion he said herons, snake birds (*anhinya rufa*) and pelicans in the Sudan are host to *Eustrongylides africanus*.

#### 2.11 EUSTRONGYLIDES LARVAE

Kabata (1985) reported that *Eustrongylides* are nematodes that uses fish as its intermediate host. The definitive host, they observed are the wadding bird, a common visitor to ponds and water bodies. The worm encysts in the peritoneum or muscle of the fish and appear to cause little damage. Because of the large size of the worms, infected fish may appear unsuitable for retail sales. Protecting fish from wading birds and eliminating the intermediate host Oligochaete or Tubiflex (soft bodied worm) are the best means to prevent infection. The larvae of *Eustrongylides* species are found encysted within the body cavity, muscles and peritoneum of many freshwater species world wide. As the larvae are large upto 10cm in length and blood red in colour, they are rather unsightly (Paperna 1974). Paperna (1974) further observed that if encysted in the gonads particularly ovary they can cause severe damage. The first intermediate hosts according to Paperna (1974) are probably tubificid oligochaetes and the final hosts are piscivorous birds.

Rechenbach – Klinke's (1973) in his work, observed that the main characteristics of these species of nematodes is the sucker of the male at the caudal end of the body, forming a bursa copulatrix. The definitive hosts are the ducks. Invertebrates intermediate host has not yet been established. Juvenile form of *Eustrongylides ignotus* can also be found in perch and European catfish, while sexually matured worms are found in herons.

Aydoghu et al (2000) reported that larval stages of *Eustrongylides* species have been found, but infestation and prevalence in fish was very low. They also observed that *Eustrongylides* could not be identified to species level. Yamaguti (1961) in Aydoghu et al (2000) further reported that *Eustrongylides* species are found in birds, but their larval stages have been recorded in fish, frogs and reptiles living close to water.

### 2.12 EPIZOOTIOLOGY

Paperna (1996) reported that epizootiology of the pericardium inhibiting contracaecum is linked with migration of piscivorous birds particularly (or even only) pelicans, between Europe and tropical East Africa. Infection of ponds in Israel occurred after they have been visited by pelicans during spring migration. Definitive hosts of the other forms of *Contracaecum* (piscivorous birds). *Amplicaecum* (aquatic reptiles) or *Eustrongylides* (cormorants) are apparently sedentary as infection is geographically localized.

*Eustrongylides* larvae, he said, if ingested by another fish, will re-encyst in its new hosts, this causes larvae to accumulate in predators at higher trophic levels. These usually large catfish and lungfish are beyond the reach of cormorants and are therefore a dead end for the parasites transmission cycles. Accumulation of nematode larvac in the large predator fish may have considerable ecological importance in moderating parasite populations in lake fish. Among *Haplochromis* species of Northern lake Victoria, incidence of infection ranged from 17 to 52% (mean 27%) with a mean burden of 5.1(SD=9.3) and up t 17 per fish. A quantitative study of *Bagrus docmac* from the same fishing area in the lake revealed a 77% prevalence of infection with a mean burden of 26 (SD-29 overdispered) up to 125 worms per fish.

Castration resulting from invasion of the gonads with a prevalence of infection ranging from 5 to 17% was found in 6 out of 15 representatives of *Haplochromis* and *Haplochromis* related species from lake Victoria. Incidence of castration was more abundant in species demonstrating an overall higher prevalence of infection. Although, *Eustrongylides* infection occurred in *Haplochromis* from lake George, in none of these were gonads involved (Paperna 1996 quoting Paperna 1974b).

Petrushevski and Shulman (1970) reported that a severe epizootic caused by protozoan (*Myxobolus exiguus Thelohan* 1895) was recorded for *Mugil cephalus* of the black and Azor seas. The gill filaments of the infected fishes were seriously affected. Some of the cysts were situated along the course of the blood vessels and the tips of the affected filaments were swollen. In many instances, the entire filaments were filled with the cysts, which were also found in the organs of the fish (the base of the gill arches, the upper lip, the intestinal wall, the mysentary etc). These sites, however, showed no deflection from normal. Rapture of the cysts resulted in damage to the tissues, causing haemorrhages, which were sometimes quite extensive. Some fish were seen with blood literally pouring from under the opercula. Death can follow either asphyxia caused by damage to a large number of gill filament or from severe loss of blood.

#### 2.13 TRANSMISSION TO INVERTEBRATES

The nematode eggs or larvae are passed into the water with the faeces of the fishes. These eggs or larvae actively or passively enter invertebrates which are their intermediate hosts and at the same time components of the diet of fish. Ingestion of infected invertebrates causes infestation of fish, which becomes the secondary intermediate host for many species of parasites (Paperna, 1974).

## 2.14 MODE OF TRANSMISSION OF PARASITES TO MAN

Reports have it that man and other animals become infected with parasites by eating raw, improperly cooked or partially sun dried fish. Fish are generally said to be intermediate host of parasites. However, Ibiwoye et al (1996) reported that no case of human disease had been documented from eating nematode infected fishes.

Oniye (2000) in Paperna (1980) reported that several trematodes, cestodes, nematodes and acanthocephalans that are naturally parasites of fish, amphibians, birds, marine and terrestrial mammals have been found in humans, who have accidentally ingested these worms or their larval stages through consumption of improperly cooked, sun dried or raw fish. Some symptoms, he observed in man associated with infection by these helminthes include abdominal discomfort, nausea, dysentery and lesions in the heart, liver, lungs, central nervous system and deficiency of certain vitamins. Control of these parasites, he concluded can be achieved by proper cooking (above 70%) and freezing (-18°C) of fish before consumption; though, these may have far reaching consequences on eating habits and economy of societies that consume raw fish.

## 2.15 SITE OF INFECTION

*Eustrongylides* larval, encysted in white cysts, 4-6mm diameter, were found predominantly in the mesenteries on the surface of the stomach and in the spleen of *Bagrus docmac*, *Clarias mossambicus* and *Haplochromis* species, larvae were also encysted in the gonads of *Haplochromis* species (Paperna, 1974). Unencysted worms were found embedded in the somatic muscles and the connective tissue of the ventral abdominal wall of *Bagrus docmac*.

#### 2.16 CONDITIONS AND EFFECTS

Bauer (1970) reported that by damaging the surface of the body and internal organs of fishes and producing various wounds and ulcerations, parasites favour the penetration of other pathogenic organisms mainly fungi and bacteria.

Quoting Petrushevski and Kogteva (1950) Petrushevski (1970) observed that the influence of parasites on the conditions of fish depend on the site of infestation, for example, larval tetracotyle when in the heart has a considerable influence on the condition of the host; no such influence is exerted by the parasite in the peritoneum. The infestation of the heart and liver, he said have stronger effects than from any other organ, gut in particular.

The pathogentic activity of parasites affects the growth rate and the condition of the host. Lechler (1935) quoted by Bauer (1970) described mass infestation of coregonids with *Ergasilus sieboldi*. He also observed during the five years of investigation that this infestation resulted in a marked lowering of the growth rate of the fish.

Quoting Paperna (1980); Okaeme (1989) reported that fish diseases are important because they can affect the productivity of fish in several ways, such as: lethargic effects by reducing food intake and thereby retardation and reduced growth rate. Furthermore, they are said to cause pathological lesions ranging from the lesion of skin to those vital body organs. The pathological lesions cause some undesirable influence which affects the physiological performance of such organs, reduced immunity to infection and eventual mortality (Ribelin and Migaki, quoted by Okaeme, 1989).

Parasites and diseases affect fish production by decreasing their aesthetic value, marketability, palatability and reproductive potentials. Paperna (1996) observed that *Eustrongylides* larvae in Cichilds when encysted migrate under the skin and in the muscle causing extensive inflammation and necrosis. Encysted worms in the viscera, particularly liver, spleen and the gonads cause severe pathological changes in the adjoining tissue. In the spleen, the tissue is replaced by lipid cells. Infection in the testes and ovaries causes sever pressure necrosis, degeneration of the spermatogenous and follicular tissues being either replaced by lipid cells or undergoing complete necrosis, ultimately resulting in castration.

The incidence and the degree of damage to the gonads was positively correlated with the overall burden of infection in the fish.

In large catfish and lungfish, larvae are numerous (often over 100)., but they encyts only in mesenteries. Even heavy infection induces localized inflammatory response, while essential visceral organs are unaffected. Only heavy infected *Bagrus docmac* was emaciated, but otherwise fish condition (determined using weight indices) did not seem to be affected (Paperna 1974b).

Robert (1978) reported that lesions renders the fish unsightly and the fish with even a single parasite feed poorly and loose weight. Onive and Annune (1993) remarked that by virture of the economic importance of fish to man, as the cheapest source of animal protein as an alternative source of income to farmers and fishermen, it's increased demand as population increases, fish has become a subject of study and information of fish parasites in particular has become important as these may affect the production of fish in culture or wild conditions.

#### 2.17 CONTROL AND PREVENTION

Unlike disease diagnostic in other vertebrates, clinical symptoms in fish are not easily discernible at a glance with gross pathology. Dujin (1973) observed that the prospects of curing fish suffering from infection of the internal organ are poor. In most cases, too much damage has been caused to important organs before the disease is recognized.

Drugs and clinical treatment under tropical (Kabata, 1985) and temperate conditions (Paperna, 1980) have shown that the response to treatment when disease is established is limited and thus in general, preventive measure are recommended in the management of diseases. However, fish leave in an aquatic environment that serves as an important vehicle for the transmission of pathogens. It is therefore necessary that preventive procedures are adopted in the production of fish. Dick and Harvey (1986) recognized that microscopic parasites are easy to treat using remedies added to the water, although a few have a cyclical lifestyle and are resistant to treatment at some stages of the cycle. Most large adult parasites, he agreed could be removed with the hand.

In an effort to check this problem, Paperna (1996) reported that the best control of digenean trematodes is to break the life cycle of the parasite. Elimination of the first intermediate host, the freshwater snail is often recommended. Copper sulfate, they said has been used in ponds with limited success and is most effective against snails when applied at night due to their nocturnal feeding activity.

Kabata (1985) reported that in Russia, chlorofos has been used to eliminate copepods as a control measure against *philometra*. Masoten has been used to control nematodes in East Germany (one prolonged application for 10 days at 1-2ppm.)

Ibiwoye et al (1996) stated that the control of disease is of considerable importance as different fish farming situations may call for different approaches. Traditional method of disease control, they remarked are not always effective because it is not cost-effective and may cause environmental pollution. Many drugs and chemicals used as chemotherapy of fishes are the same as for higher animals. Drugs, they said should be inexpensive and not in competition with human and other valuable farmed animals.

"Prevention an old adage say is better than cure," however, Paperna (1996) quoting Bejeramo (pers. Comm.) observed that prevention of larval nematode infection by keeping away piscivorous birds is impracticable, not only in fishing areas in natural habitats or man-made impoundments but even in fish ponds.

In fish ponds, preventive treatment of *Contracaecum* by elimination of copepod (by insecticide such as masoten or Bromex). Bromex applied at a level of 2ppm, killed free living larvae in vitro, but such a dose is about or beyond tolerance limit of fish. Experiments with helminthicides (Levamizol, mebendazol or ivermectin) have not so far produced satisfactory results and it is not certain, if costs of treatment (by use of medicated feeds) will be economical. Kabata (1985) advised that in case involving nematodes pathogenic to man the effectiveness of fish product must be eliminated by cooking or other treatments (salting, drying, freezing etc) to kill nematodes encysted in the flesh.

General treatment for fungi, bacterial and parasitic infections as reported by Ahmed (1997) is presented in Table 1.

Chemical	Dosage in Bath	Dosage in pond	Infection
Formalin	25ppm for 30min	5ppm	Worms
	250ppm for 5min.	15ppm	Protozoa
Malachite green	1.25ppm for 10- 15min	0.5ppm	All types

1ppm

3ppm

Fungal an bacteria

treatment of fish

Protozoa

crustacean

**Bacterial** 

Protozoa and general

and

Table. 1 General treatment of fungi, bacterial and parasite infections.

500ppm for 1-2min.

3,000-5,000ppm for

10ppm

3-5min.

50ppm

Source: Ahmed H.U(1997)

**Copper Sulphate** 

Potassium

Permanganate

Sodium Chloride

Chloramphenicol

### CHAPTER THREE

#### 3.0 MATERIALS AND METHODS

### 3.1 STUDY AREA

This study was conducted between November, 2004 and April, 2005 in the natural rivers and man-made dams spread across the three geopolitical areas represented as A, B and C (Niger South, Niger East and Niger North) of Niger state, Nigeria.

## 3.2 FISH SAMPLES

Niger state is blessed with abundant water bodies ranging from rivers, flood plans, natural and man-made dams. Fishing gears used by fishermen are baited long lines and traditionally made traps which are confirmed to be very active and efficient in harvesting *Clarias species*. *Clarias gariepinus* used for this study were purchased from fishermen, fishing on the water bodies mentioned above. The fish sample (*Clarias gariepinus*) was identified by methods described by Reed et al (1967). *C. gariepinus* are characterized by elongated smooth body, flattened head, which are rough and granular and wide mouth. They have strong, barbed spine in front of each of the pictorial fins, but none preceding the dorsal fin (Holden and Reed 1972). Landing sites from where *C. gariepinus* were purchased are shown on Fig. 1 (Hydrological map of Niger state, Nigeria).

### 3.3 LOCATION OF SAMPLING SITES

Using the existing three geopolitical areas (Niger South, Niger East and Niger North) of the state, three water bodies were earmarked for the collection

of fish samples from each area. The location (water bodies) from where the fish samples were collected are as follows:

Area	Location	Water body
A (Niger South)	Kakakpangi	River Gbako
	Katcha	River Niger
	Wuya	River Kaduna
B (Niger East)	Shiroro	River Kaduna/Shiroro dam
<i>t</i> *	Suleja	Suleja Dam
	Tagwai	Tagwai dam
C (Niger North)	Kagara	Kagara dam
	Matandi	Kontagora dam
	Wushishi	Tunga kawo dam

### 3.4 COLLECTION OF FISH SAMPLES

*Clarias gariepinus* is one of the commercial important fish species in most water bodies in Niger state. Irrespective of size, age and sex, forty (40) samples of *Clarias gariepinus* were randomly purchased from fishermen at landing sites and altogether one hundred and twenty (120) samples from each geopolitical area. Three hundred and sixty (360) fish samples were purchased from the three geopolitical areas.

Collection of the fish samples was done weekly i.e. three sites within each areas was visited. Rotationally, the three areas were visited and all the fish samples required were collected.

The fish samples were transported in a 60cm x 60cm flask to the fisheries laboratory, of the Federal University of Technology, Minna where morphometric parameters, dissection were carried out.

# 3.5 MEASUREMENT OF MORPHOMETRIC PARAMETERS

Total and standard length in grams of individual fish were taken from the snout to the tip of the caudal fin and peduncle respectively using measuring rule. Weights were measured by a portable kitchen weighing scale (Cap 5kg model). These parameters were measured using methods described by Olatunde (1977). Data collected and condition were used to determine length- weight relationship and conditions factors, (K) of the fish sample.

$$K = \frac{100w}{l^3}$$

where W = Weight of fish (kg)

L = Length of fish (cm)

As the fish sample were dissected, *Eustronglylides africanus* were visibly seen lodged in the muscles and abdominal cavity. However, none was found on the liver. The liver weights was determined with the aid of a laboratory sensitive weighing scale (Mettler P.M 2000).

### 3.6 PARTS OF BODY EXAMINED

Fish sample were examined individually, observation of the skin, gill region and fins were done with the aid of hand lens for presence of parasites. The fish samples were dissected and the skin, gill, abdominal cavity, muscles and liver were observed thoroughly for presence of parasite. Different portion of the above parts were separated and put in normal saline.

### 3.7 DATA ANALYSIS

Data obtained were subjected to:

T – test and analysis of variance (ANOVA) at 5% significance level, where there was a difference, Duncan multiple Range test for variance was used. They were also subjected to correlation regression analysis at 5% significance level.

# 3.8 ISOLATION OF PARASITES, IDENTIFICATION AND PRESERVATION

Parasites were isolated from the musculature and abdominal cavity of the fish sample manually according to procedure described by Brent R. Dixon (2006). All the *Eustrongylides africanus* isolated were observed to be alive as they exhibited movement. *Eustrongylides africanus* identified were characterized by reddish color, long, cylindrical and tapering at both ends confirming description made by Paperna (1996). They were counted and preserved in 40% ethanol.

### 3.9 CONDITION FACTOR

The condition factor is used to compare the condition which is an index of fitness or well being of the fish (Bagenal,1978) fultons condition factor K is calculated using the formula.

 $K = \frac{100w}{l^3}$  where W = Total weight (kg)

L= Total length (cm)

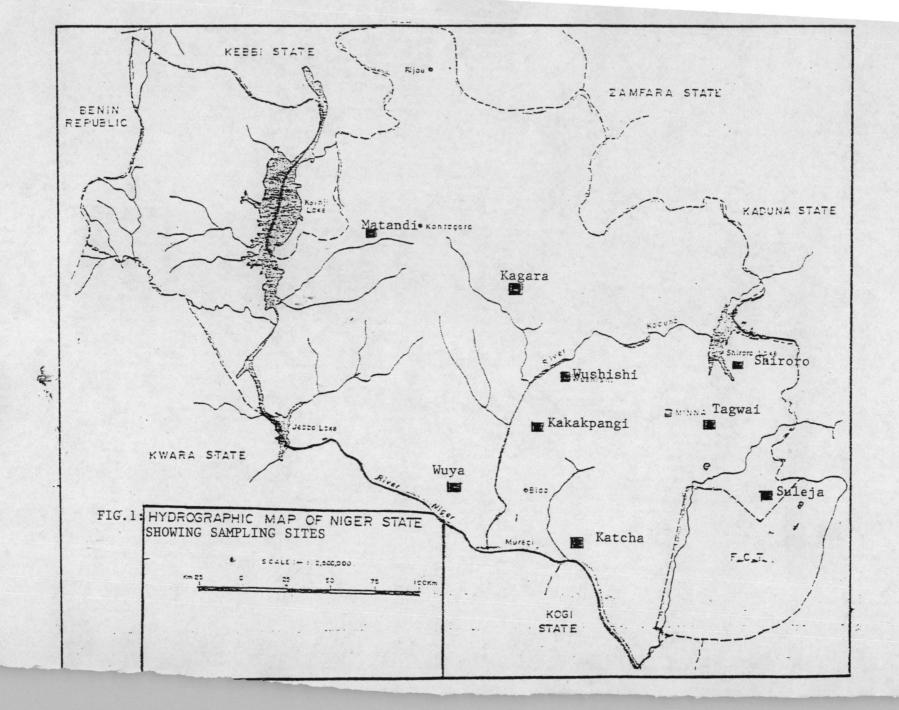
### **CHAPTER FOUR**

### 4.0 RESULTS

### 4.1 MORPHOMETRIC PARAMETERS

Morphometric parameters recorded as mean for the fish weight, liver weight and standard length in the three geopolitical areas A, B & C are presented in Tables 4.1, 4.2 and 4.3. In the same vein the parameters (fish weight, liver weight and standard length) recorded for the whole study area as an entity are presented in Table. 4.4. The result of mean of the parameters recorded in the tables listed above shows that there was no significant difference (P>0.05) in the fish weight, standard length, liver weight and condition factor within each geopolitical area and the entire study area.

The mean value recorded as fish weight, total length, standard length and liver weight for the uninfected and infected fish samples in area A and C did not show any significant difference (P>0.05) as presented in Table 4.5 & 4.6. Similarly, the same parameters recorded for the study area showed that there was significant difference (P<0.05) in the fish weight as shown in Table 4.7.



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Table 4.1 Mean values of fish weight. standard-length and-liver weight of fish samples in Area A'.

Parameter	Site i	Site ii	Site iii
Mean fish weight(g)	$196.1 \pm 147.1^{\circ}$	187.1 <u>+</u> 62.5 <sup>c</sup>	158.8 <u>+</u> 69.6 <sup>b</sup>
Mean SL (cm)	23.63± 5.93 <sup>b</sup>	24.09 <u>+</u> 2.99 <sup>b</sup>	27.80 <u>+</u> 25.82 <sup>c</sup>
Mean L.wt (g)	$1.83 \pm 1.31^{\circ}$	2.03 <u>+</u> 0.71 <sup>b</sup>	1.71 <u>+</u> 1.21 <sup>b</sup>

Row mean data carrying the same superscript do not differ significantly from each other (P>0.05).

Note: g= gram ; SL=standard length: cm=centimeter; L.wt=liver weight

Table 4.2 Mean values of fish weight, standard length and liver weight of fish sample in Area B

Parameter	Site i	Site ii	Site iii
Mean Weight (g)	198.63 <u>+</u> 105.17 <sup>b</sup>	164.88 <u>+</u> 92.64 <sup>a</sup>	185.00 <u>+</u> 95.5 <sup>b</sup>
Mean S L(cm)	24.54 <u>+</u> 4.42 <sup>°</sup>	23.28 <u>+</u> 3.34 <sup>b</sup>	23.89 <u>+</u> 4.74 <sup>b</sup>
Mean L.wt(g)	1.92 <u>+</u> 1.13 <sup>c</sup>	1.56±0.94 <sup>b</sup>	1.74 <u>+</u> 0.99 <sup>b</sup>

Row mean data carrying the same superscript do not differ significantly from each other (P>0.05).

Note: g= gram ; SL=standard length; cm=centimeter; L.wt=liver weight

Table 4.3 Means values of fish weight, standard length and liver weight of fish samples in Area C

Parameter	Site i	Site ii	Site iii
Mean fish Weight (g)	183.13 <u>+</u> 62.57 <sup>b</sup>	167.75 <u>+</u> 83.27 <sup>a</sup>	169.13 <u>+</u> 74.33 <sup>a</sup>
Mean SL (cm)	23.30 <u>+</u> 2.16 <sup>a</sup>	23.63 <u>+</u> 3.63 <sup>a</sup>	24.08 <u>+</u> 3.39 <sup>b</sup>
Mean L.wt (g)	1.73 <u>+</u> 0.64 <sup>a</sup>	1.57 <u>+</u> 0.80 <sup>a</sup>	1.55 <u>+</u> 0.75 <sup>a</sup>

Row mean data carrying the same superscript do not differ significantly from each (P>0.05)

Note: g= gram ; SL=standard length; cm=centimeter; L.wt=liver weight

Table 4.4 Mean values of fish weight, standard length and liver weight of fish samples in Area A,B,&C.(combined)

Mean of parameter	A	В	С
Mean fish weight (g)	181.2 <u>+</u> 100.8 <sup>a</sup>	182.5 <u>+</u> 98.6 <sup>a</sup>	173.3 <u>+</u> 73.3 <sup>a</sup>
Mean SL (cm)	25.24 <u>+</u> 5.31 <sup>a</sup>	23.88 <u>+</u> 4.24 <sup>a</sup>	23.66 <u>+</u> 3.10 <sup>a</sup>
Mean L. wt (g)	1.86 <u>+</u> 1.10 <sup>b</sup>	1.73 <u>+</u> 1.03 <sup>a</sup>	1.62 <u>+</u> 0.73 <sup>a</sup>
K	1.414	1.334	1.289

Row mean data carrying-the same superscript do not differ significantly among

the areas (P>0.05)

Note: g= gram ; SL=standard length; cm=centimeter; L.wt=liver weight:

K = condition factor

-

Table 4.5. Mean values of fish weight, total length, standard length and liver weight of uninfected infected fish samples in Area A

Uninfected	Infected	
130.1+48.5	224 <u>+</u> 111	
25.31+2.22	29.40+6.2	
22.02+1.92	25.43+5.46	
1.3±0.42	2.4+1.23	
	130.1 <u>+</u> 48.5 25.31 <u>+</u> 2.22 22.02 <u>+</u> 1.92	$130.1\pm48.5$ $224\pm111$ $25.31\pm2.22$ $29.40\pm6.2$ $22.02\pm1.92$ $25.43\pm5.46$

Note: g= gram ; cm=centimeter;

Table 4.6 Mean values of weight, total length, standard length and liver weight of uninfected and fish sample in Area C

Parameter	Uninfected	Infected
Mean fish weight (g)	173 <u>+</u> 73	164 <u>+</u> 95.6
Mean total length(cm)	27.33+3:4	27 <u>+</u> 4.2.
Mean standard length(cm)	23.7±3.2	23.3+4
Mean liver weight (g)	1.60+0.7	<u>1.84+1.2</u>

Note: g= gram ; cm=centimeter;

Table 4.7 Mean values for uninfected and infected fish sample from the three Areas. (A, B &C)

Parameter	Uninfected	Infected	
Mean fish weight (g)	169.8+83	220+110	
Mean total length(cm)	27.1 <u>+</u> 3.9	29.2 <u>+</u> 6.1	
Mean standard length(cm)	<b>23.5</b> <u>+</u> 3.5	25.3+5.4	
Mean parasite count (No)	0.00±0.0	2.31 <u>+</u> 1.2	
K	2.0	2.8	

Note: g= gram ; cm=centimeter;  $\overline{K}$  = condition factor

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## 4.2 PARTS OF THE BODY EXAMINED

The most infected fish samples were those obtained from area A i.e. Katcha, Kakakpangi and Wuya samplings areas (fig. 6). Seventy two (72) fish samples (20%) were infected with *Eustrongylides africanus*. Sixty eight (68) samples of *Clarias gariepinus* were discovered to have *Eustrongylides africanus* encysted in the musculature and four (4) in the abdominal cavity as presented in fig. 6.

Observation of a few sample shows undulation on the skin surface of the fish sampled. As the fish sampled were dissected *Eustrongylides africanus* were spotted underneath the skin sheath and at different depth of the muscles as shown in Plate 1, Plate 2 shows red coloured *Eustrongylides africanus* that emerged from a cyst under the muscle. The length of the *Eustrogylides africanus* found was in the range of 5 cm - 14 cm.

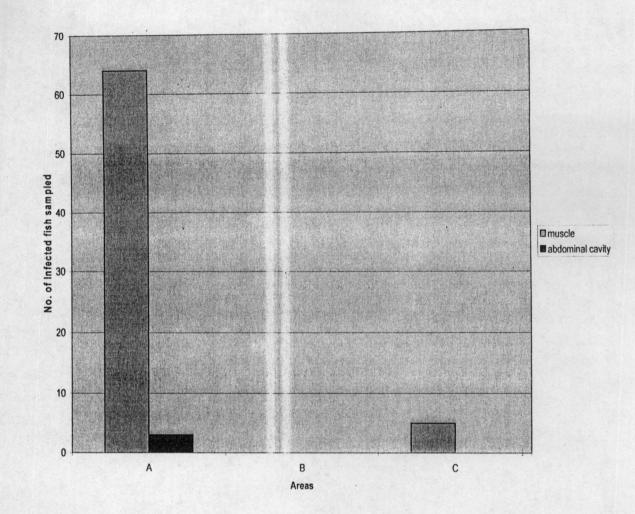


Fig. 6. *Eustrongylides africanus* infestation of *Clarias gariepinus* from three geopolitical Areas of Niger state.

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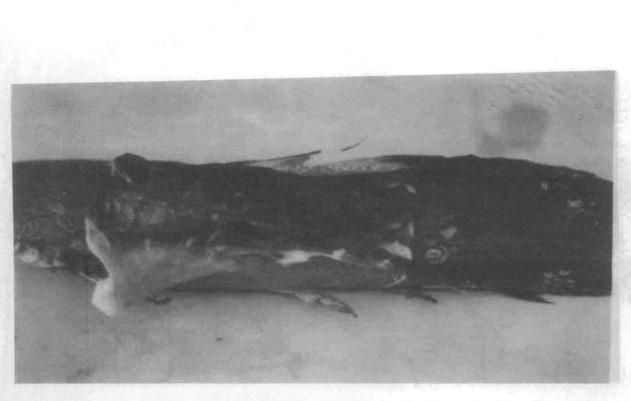


Plate 1: Photograph of sample Clarias gariepinus infested with Eustrongylides

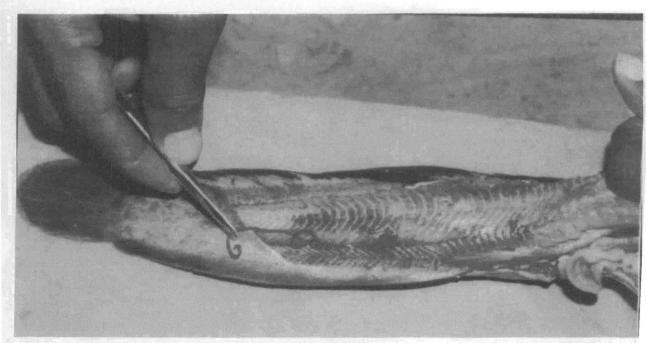


Plate 2: Photograph of sample *Clarias gariepinus* showing *Eustrongylides* that emerged from a sheath under the musculature.

Mean number of *Eustrongylides africanus* in the fish muscle, abdominal cavity and liver in Area A and C are presented in Tables 4.8 and 4.9. Amongst the Areas A, B & C the mean value shows a significant difference (P<0.05) demonstrated in table 4.10

Table 4.8 Mean number of *Eustrongylides africanus* in fish muscle, abdominal cavity and liver of fish sample area A.

Site of infestation	Site i	Site ii	Site iii
Muscle	2.68 <u>+</u> 3.10 <sup>b</sup>	2.90 <u>+</u> 3.02 <sup>b</sup>	0.43 <u>+</u> 0.90 <sup>a</sup>
Abd. Cavity	0.10 <u>+</u> 0.39 <sup>a</sup>	0.00 <u>+</u> 0.00 <sup>b</sup>	0.00 <u>+</u> 0.00 <sup>a</sup>
Liver	0.00 <u>+</u> 0.00 <sup>a</sup>	0.00±0.00 <sup>a</sup>	0.00±0.00 <sup>a</sup>

Values in the same row carrying different superscript differ significantly (P<0.05).

Table 4.9 Mean number of *Eustrongylides africanus* in fish muscle, abdominal cavity and liver of fish sample with in area C.

Site of infestation	Site i	Site ii	Site iii
Muscle	0.25 <u>+</u> 0.81 <sup>a</sup>	0.00 <u>+</u> 0.00 <sup>a</sup>	0.00 <u>+</u> 0.00 <sup>a</sup>
Abd. Cavity	$0.00\pm0.00^{a}$	0.00 <u>+</u> 0.00 <sup>a</sup>	0.00 <u>+</u> 0.00 <sup>a</sup>
Liver	$0.00\pm0.00^{a}$	0.00 <u>+</u> 0.00 <sup>a</sup>	0.00 <u>+</u> 0.00 <sup>a</sup>

Values in the same row carrying the same superscript do not differ significantly

(P>0.05).

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Table 4.10 Mean number of *Eustrongylides africanus* in fish muscle, abdominal cavity and liver of fish sample in areas A,B&C.

Area A	Area B	Area C
1.98 <u>+</u> 2.76 <sup>c</sup>	0.00 <u>+</u> 0.00 <sup>a</sup>	0.00 <u>+</u> 0.00 <sup>a</sup>
0.033 <u>+</u> 0.22 <sup>b</sup>	$0.00\pm0.00^{a}$	0.00 <u>+</u> 0.00 <sup>a</sup>
$0.00\pm0.00^{a}$	0.00 <u>+</u> 0.00 <sup>a</sup>	0.00 <u>+</u> 0.00 <sup>a</sup>
	1.98 <u>+</u> 2.76 <sup>c</sup> 0.033 <u>+</u> 0.22 <sup>b</sup>	$1.98\pm2.76^{c}$ $0.00\pm0.00^{a}$ $0.033\pm0.22^{b}$ $0.00\pm0.00^{a}$

Values in the same row carrying different superscript differs significant (P<0.05).

The length-weight relationship of the fish sample from area A are shown in a regression graph characterized by allometric growth pattern as shown in Fig. 2. In area B and C the growth exhibited was allometric and is presented in Fig. 3 and Fig. 4. The relationship however amongst the area A, B and C is isometric as shown in Fig.5.

T- test value = T < P is not significant (P<0.05) for uninfected and infected fish sample in geopolitical area A & C and the entire study area for total length, fish weight, standard length and liver weight respectively.

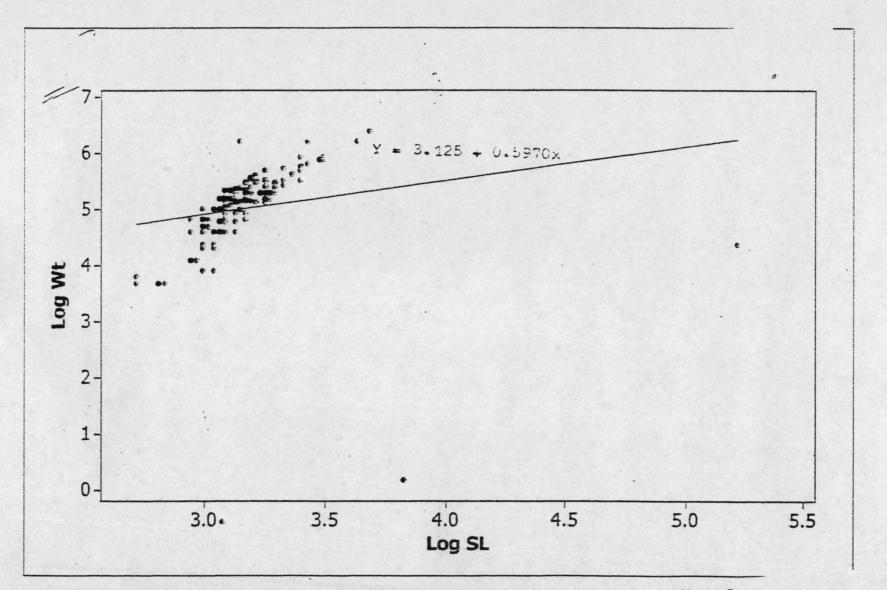
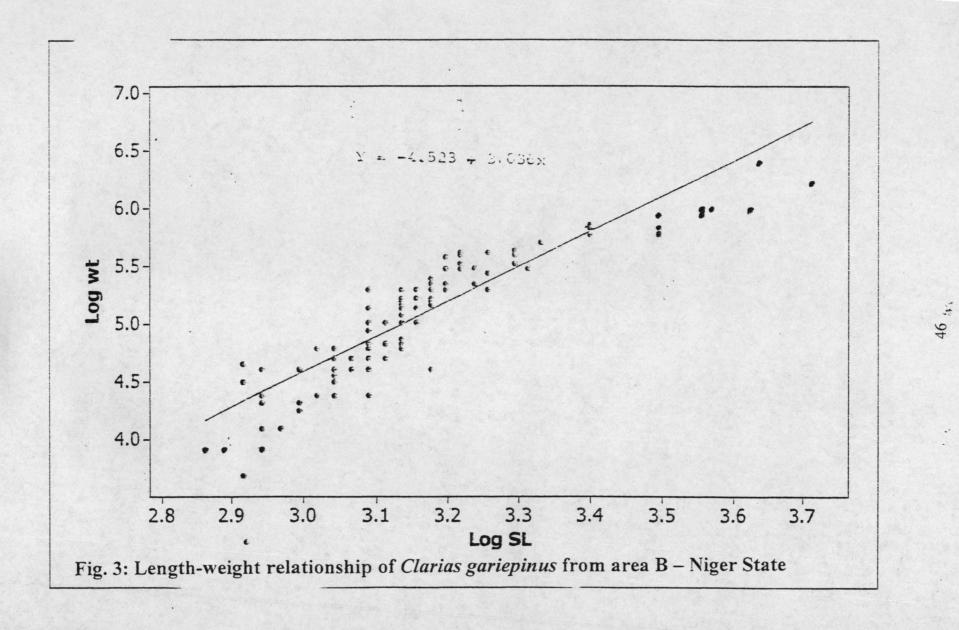
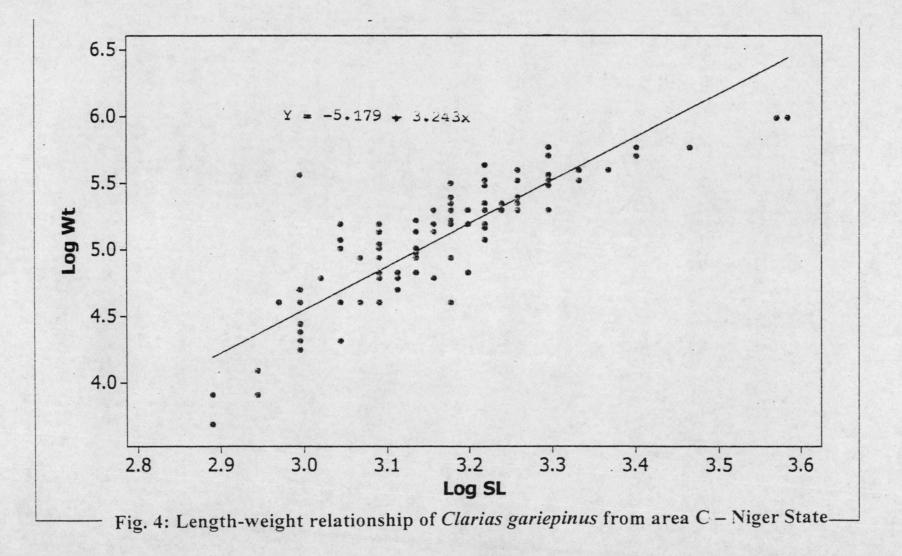
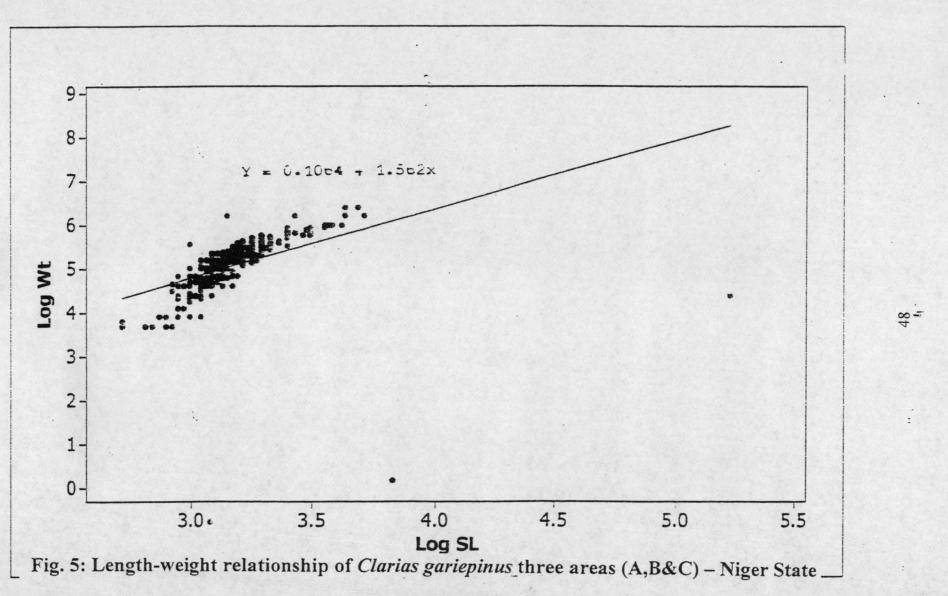


Fig. 2: Length-weight relationship of Clarias gariepinus from area A - Niger State



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The correlation of parameters in zone A correlated significantly (p>0.05) between liver weight, fish weight, standard length and muscle as shown in Table 4.11. In Table 4.12 and 4.13, the correlation was significant (p<0.05) between standard length, fish weight and liver weight in area B and C. However the correlation of parameters for the uninfected fish sample from the state was significant (p<0.05) between the fish weight, total length, standard length and liver weight and liver weight and likewise for the infected fish sample, it was significant (p<0.05) between fish weight, total length and liver weight total length and liver weight and liver weight.

	Wt(g)	SL	L.wt	Muscle	Abd. cavity	
SL	0.112					
L.wt	0.699*	0.161*				
Muscle 0.259*		-0.040	0.255*		· ·	
Abd. Cavity	0.044	-0.007	0.072	-0.055		
Liver 0.00		0.00	0.00	0.00	0.00	

Table 4.11: Correlation Matrix of Parameters for Area A

• Significant difference (p< 0.05)

Note: g= gram ; SL=standard length; cm=centimeter; L.wt=liver weight: Abd. cavity = Abdominal cavity

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# Table 4.12: Correlation Matrix of Parameters for Area B

	Wt (g)	SL	L.wt	Muscle	Abd. cavity
SL	0.942*	3 3396			
L.wt	0.990*	0.942*			
Muscle	0.00	0.00	0.00		
Abd. Cavity	0.00	0.00	0.00	0.00	
Liver 0.00		0.00	0.00	0.00	0.00

• Significant difference (p< 0.05)

Note: g= gram ; SL=standard length; cm=centimeter; L.wt=liver weight: Abd. cavity = Abdominal cavity

# Table 4.13: Correlation Matrix of Parameters for Area C

	Wt (g)	SL	L.wt	Muscle	Abd. Cavity
SL	0.896*				
L. wt	0.971*	0.868*			
Muscle	e -0.061 -0.100		0.010		
Abd cavity	0.00	0.00	0.00 0.00		
Liver 0.00		0.00	0.00	0.00	0.00

• Significant difference (p< 0.05)

Note: g= gram ; SL=standard length; cm=centimeter; L.wt=liver weight:

Abd. cavity = Abdominal cavity

Table 4.14 Correlation matrix of parameters for uninfected and infected fish samples from Niger state.

	Wt1	TL1	SL1	PC1	Lwt1	Wt2	TL2	SL2	PC2
TLI	0.926*								
SL1	0.923*	0.975*							
PC1	0.00	0.00	0.00					•	
Lwt1	0.969*	0.917*	0.915*	0.00					
Wt2	-0.052	-0.036	-0.047	0.00	-0.12				
TL2	0.146	0.177	0.172	0.00	0.141	0.751*			
SL2	0.126	0.161	0.155	0.00	0.119	0.755*	0.994*		
PC2	-0.103	-0.044	-0.041	0.00	014	0.032	0102	0.098*	
Lwt-2	0.102	0.145	0.153	0.00	0.107	0.688*	0.905*	0.897*	-0.074

• Significant (p<0.05)

1. Uninfected fish sample

2. Infected fish sample

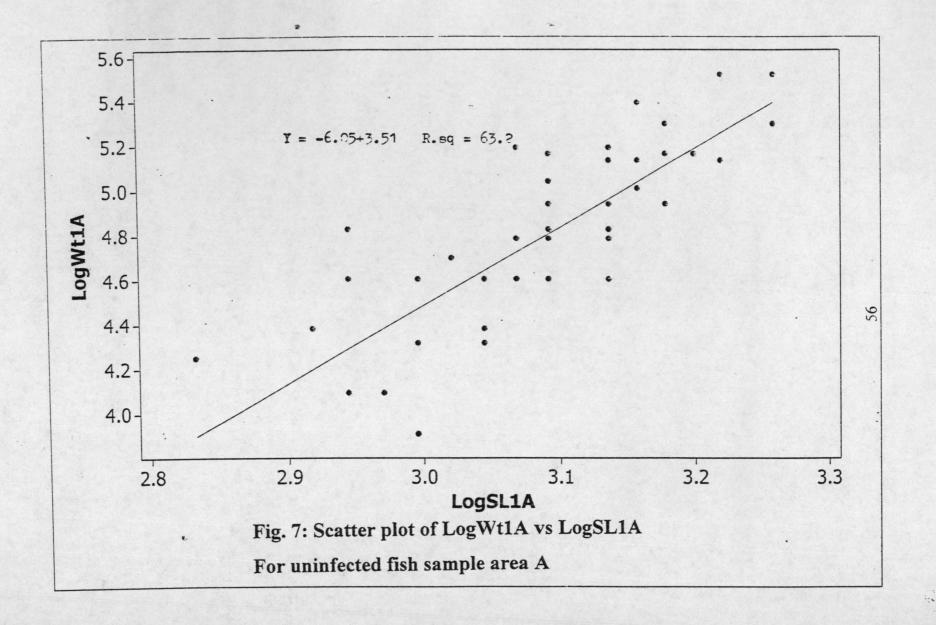
Note: g= gram ; SL=standard length; cm=centimeter; L.wt=liver weight:

Abd. cavity = Abdominal cavity

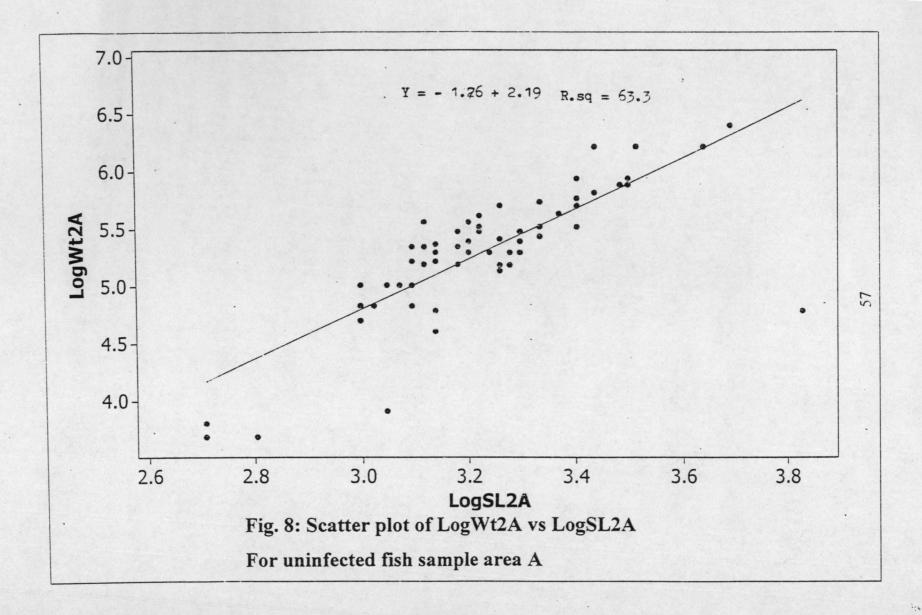
Regression analysis equations for fish weight and fish length relationship of uninfected and infected fish sample in the three geopolitical areas A, B & C and in the study area as an entity are presented in Table 4.15. The length-weight relationship connotes allometric and Isometric growth for the uninfected and infected fish samples in area A represented in Fig. 7 and Fig. 8. Uninfected in area B exhibited allometric growth shown in Fig. 9. Allometric growth was observed for the uninfected and infected in area C as is illustrated in Fig. 10 and Fig. 11. The relationship exhibited in Niger state for the uninfected and infected fish sample shows allometric and isometric growth respectively represented in Fig 12 and Fig. 13. Table 4.15 Regression analysis equations for log wt and log SL relationship of uninfected and infected fish sample in area A, B and C (combined).

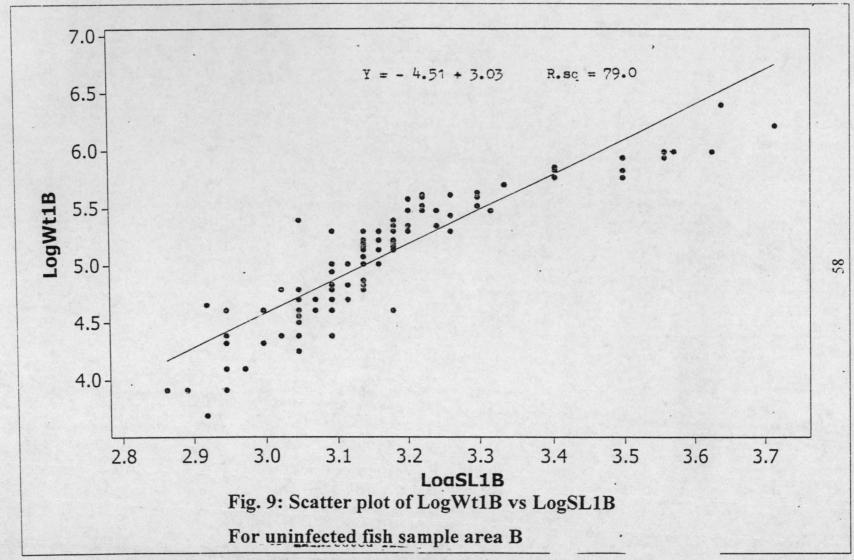
Condition of fish Sample	Regression	R-sq. 63.2 63.3	
Uninfected area A	Y=-6.05 + 3.51		
Infected area A	Y=-1.76 + 2.19		
Infected area B			
Uninfected area B	Y=-4.51 +3.03	79.0	
Uninfected area C	Y=-5.01 + 3.19	78.6	
Infected area C	Y=-8.01 + 4.12	94.6	
Uninfected area A, B &C (combined)	Y=-4.90 + 3.15	77.7	
Infected area A, B &C(combined)	Y=-2.07 + 2.28	64.5	

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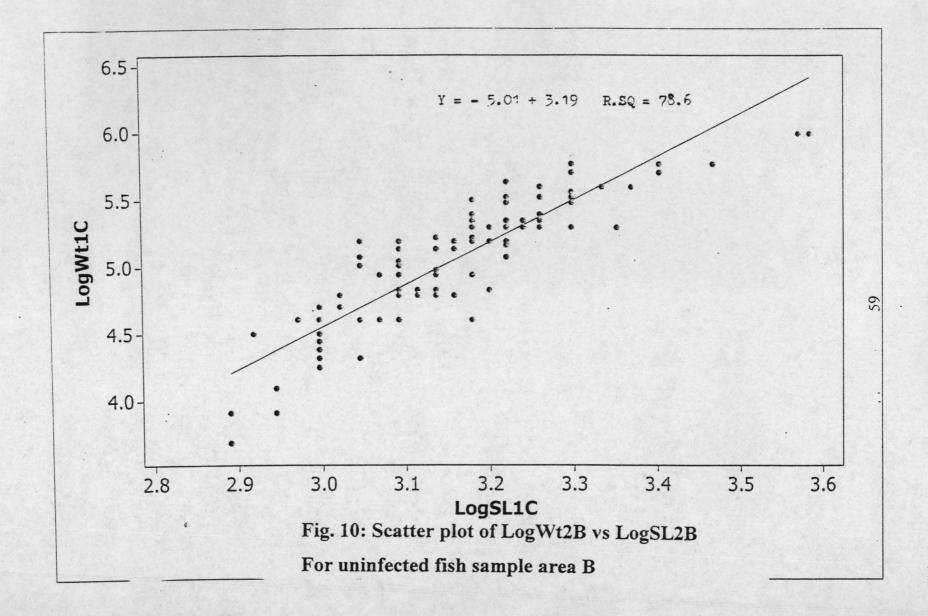
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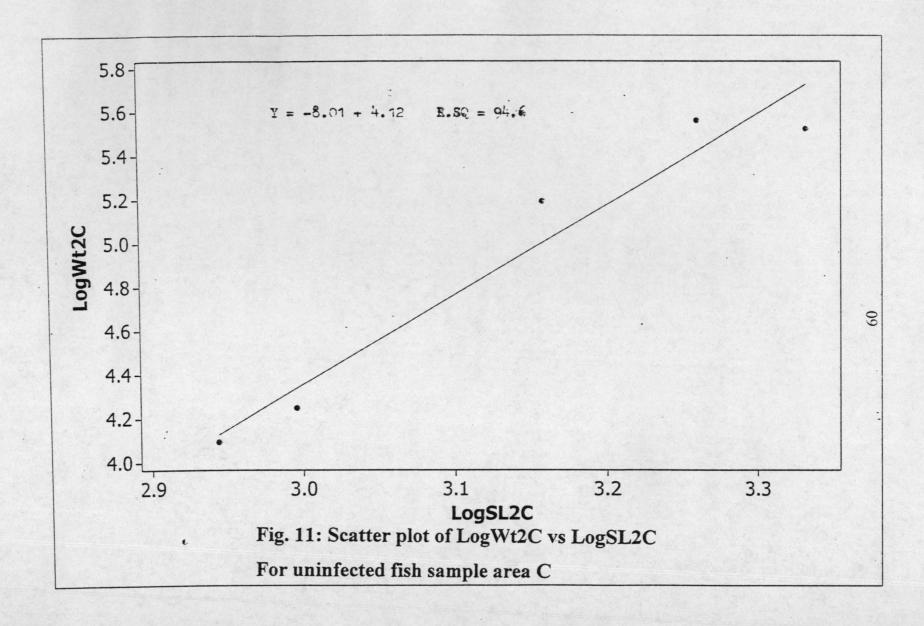




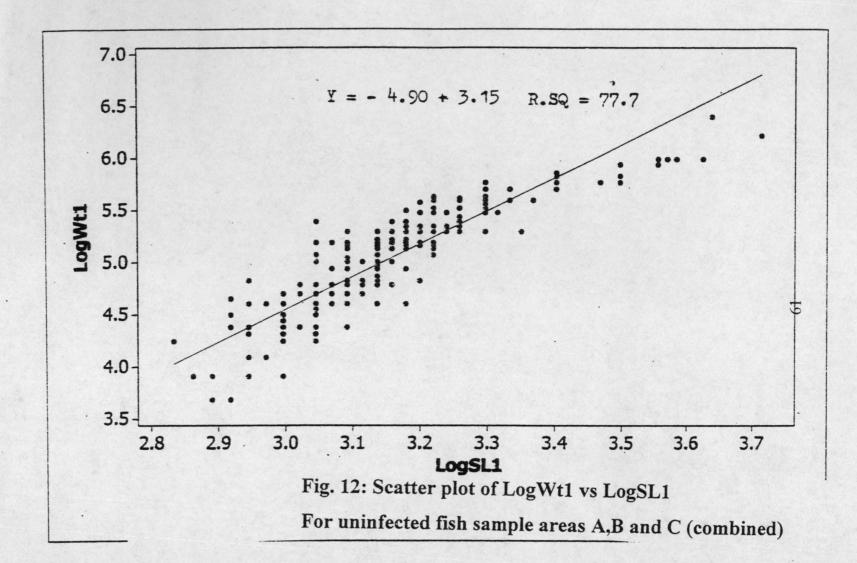
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5.4

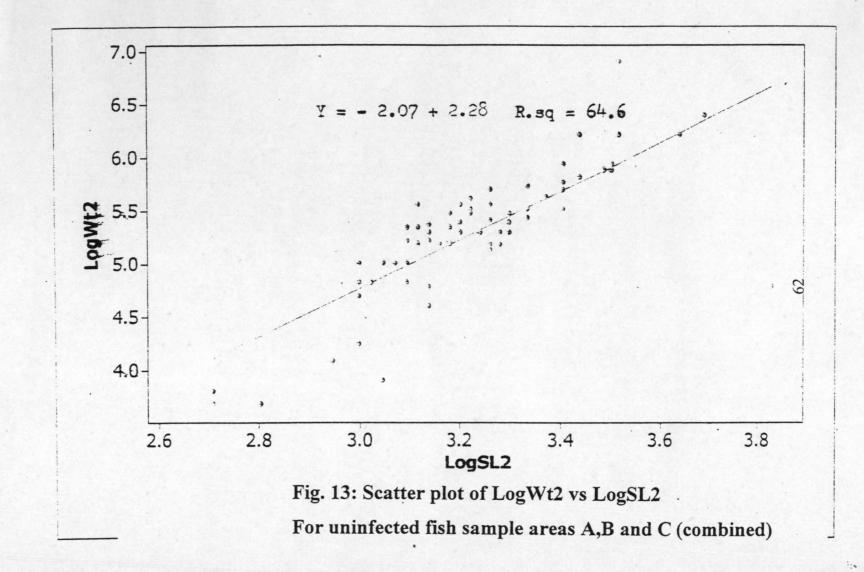




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# CHAPTER FIVE

## 5.0 DISCUSSION

# 5.1 INFESTATION LEVEL OF SAMPLES

Three hundred and sixty (360) samples of *Clarias gariepinus* were used for this study. Out of which 20% (72) were infested with *Eustrongylides africanus*. Out of the seventy two (72) infected samples, sixty seven (67) had the parasite located in the muscles and the remaining five (5) had the parasite located in the abdominal cavity. The total number of parasite (*Eustrongylides africanus*) isolated was two hundred and fifty one (251).

There was no size specificity in the infestation, as they were found in different size of the fish sampled, which confirms Ibiwoye et al (1996) report that *Clarias* species of not less than 5cm long, but of different sexes, weights and ages have been observed to develop natural infestation with *Eustrongylides africanus* larvae within the freshwater Fadama of Bida area.

A distinct external characteristic observed on the samples obtained from geopolitical area A and which was not found on the samples from the other geopolitical areas was undulation on the skin which is said to be a symptom of infection as reported by Paperna (1996). Out of the one hundred and twenty (120) *Clarias gariepinus* sampled from geopolitical area A, 67 (56%) were infected with *Eustronglides africanus* while 53 (44%) were not infected. Sixty four (64) of the infected fish sampled had the parasite lodged in the muscles and three (3) in the abdominal cavity. The parasite was not found in samples from geopolitical area B, however, in geopolitical area C five (5) were infected and localized in the muscles.

The geopolitical areas A and C where the *Eustrongylides africanus* were found in the fish sample was characterized by rivers which were during the sampling time static and cut-off from main water course and the water bottom contained clayish and loamy soils (muddy soils) and high organic debris, which is said to be favourable to the survival of snails and frogs that are intermediate hosts to these parasites (Paperna, 1974; Ibiwoye et al., 1996).

## 5.2 MORPHOMETRIC PARAMETERS

Observations made of the sampled fish from the recorded parameters within the sites in each area and amongst the areas were very similar, however, the difference observed was in the situation where the samples were grouped into uninfected or infected in geopolitical area A and in the study area as an entity where the mean fish weight for the infected differ significantly. The reasons for this could probably be due to the sizes that were randomly sampled.

# 5.3 PARTS OF THE BODY EXAMINED

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The external part of some of the fish samples examined showed undulation, which depicts the presence of worms (plate 1.). As the fish samples were dissected and the skin, gill region, muscles, abdominal cavity and liver closely observed, the worms were found encysted in the muscles and abdominal cavity. The worms were seen coiled like a watch-spring in the cyst and located at different depth of the fish muscle and body cavity which corroborate the work earlier done by Oniye, (2000).

*Eustrongylides africanus* found in the muscles and abdominal cavity of the *Clarias gariepinus* sampled appeared reddish in colour which suggest that they must have been feeding on the blood and other materials of their host as reported by Dicks and Harvey (1988).

This study has confirmed the prevalence of *Eustrongylides africanus* on *Clarias gariepinus* in Niger state however it is not widespread and the occurrence is quite low. The parasite load ranged between 1 - 11. No serious inflammation of the internal organs was observed as the *Eustrongylides africanus* found were only located in the muscles and abdominal cavity and not in vital internal organs of the fish that could have caused any considerable influence on the condition of the fish negatively. This corroborate the earlier work done by Petrushevski (1970).

# 5.4 CONDITION FACTOR

Begenal (1978) reported that condition factor is used to determine "Condition", "fitness" or "well-being" of an organism. It is also said to be used in comparing difference related to season, sex or habitat of a species. The result obtained in this study indicated that the condition of the fish was good and that *Eustrongylides africanus* found did not in any way affect the condition of the fish as *Eustrongylides africanus* found on the effect was not adverse as revealed in the statistical analysis and work carried out by Ibiwoye et al (1996).

# 5.5 CONCLUSION

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The finding of this study showed that *Clarias gariepinus* from Niger state are infested with *Eustrongylides africanus*, however, it is not widespread. The level of infestation is also very low, compared to other reports around the world.

The condition of the fish samples was not adversely affected by the infestation of *Esutrongylides africanus* based on the number found on the fish sample and part of the body where they were sited.

Studies have shown that eradicating the prevalence of *Eustrongylides africanus* sp in the wild could be a difficult task, however, certain measures to reduce their prevalence includes: eradication of the intermediate hosts, scaring away migratory birds etc.,

In conclusion, it is important to note that the following measures, if put in place could remedy the situation of transmission to human beings:-

- Properly examine catfish before buying. Those with swollen or undulated body surface should be avoided.
- Catfish bought and cut into pieces should be thoroughly examined and worms found be removed.
  - Catfish bought must be thoroughly cooked before consumption.

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Welcome to Minitab, press F1 for help.

# Results for: james1.MTW

# One-way ANOVA: Wt(g) versus Des

SourceDFSSMSFPDes230375151871.500.227Error117118460410125Total1191214979

s = 100.6 R-Sq = 2.50% R-Sq(adj) = 0.83%

				Individual 95% CIs For Mean Based on Pooled StDev
Level	N	Mean	StDev	
1	40	196.1	147.1	()
2	40	187.1	62.5	()
3	40	158.8	69.6	()
				150 180 210 240

Pooled StDev = 100.6

# One-way ANOVA: SL(cm) versus Des

Source	DF	SS	MS	F	Р	
Des	2	395	198	0.83	0.437	
Error		27728	237			
Total	119	28123				
S = 15.	39	R-Sq =	1.418	R-S	q(adj) =	0.003

				Pooled StI			
Level	N	Mean	StDev		+		
1	40	23.83	5.93	(	*	)	
2	40	24.09	2.99	(		)	
3	40	27.80	25.82		(	+	
				21.0	24.5	28.0	31.5

Pooled StDev = 15.39

. .:

# One-way ANOVA: L.Wt(g) versus Des

Source	DF	SS	MS	F	Р
Des	2	2.16	1.08	0.88	0.416
Error	116	141.61	1.22		
Total	118	143.77			

S = 1.105 R-Sq = 1.50% R-Sq(adj) = 0.00%	S	= 1.105	R-Sq =	= 1.50%	R-Sq (	(adj)	= 0.00%
--	---	---------	--------	---------	--------	-------	---------

				Individual Pooled StDe		or Mean Ba	sed on
Level	N	Mean	StDev		+	+	
1	39	1.827	1.310	(	*	)	
2	40	2.034	0.710		(	*	)
3	40	1.710	1.207	(	*	)	
				+	+	+	+
				1.50	1.80	2.10	2.40

# Results for: james2.MTW

# One-way ANOVA: Wt(g) versus Des

 Source
 DF
 SS
 MS
 F
 P

 Des
 2
 23063
 11531
 1.21
 0.303

 Error
 117
 1118424
 9559
 9559

 Total
 119
 1141487

s = 97.77 R-Sq = 2.02% R-Sq(adj) = 0.35%

				Individual			sed on
				Fooled StDe	y /		1 -
Level	N	Mean	StDev				
1	40	198.63	105.17.		(		
2	40	164.88	92.64	(			
3	40	185.00	95.05				
					· · · · · · · · · · · · · · · · · · ·		
				150	175	200	225

Pooled StDev = 97.77

## One-way ANOVA: SL(cm) versus Des

Source	DF	55	MS	F	P
Des	2	32.0	16.0	0.90	0.408
Error		2071.4	17.7		
Total	119	2103.4			

S = 4.208 R-Sq = 1.52% R-Sq(adj) = 0.00%

Individual 95% CIs For Mean Based on Pooled StDev

Level	N	Mean	StDev	+			
1	40	24.538	4.416		(-		- * )
2	40	23.275	3.340	(	4		)
3	40	23.838	4.739	() ·			
				+			
				22.0	23.0	24.0	25.0

Pooled StDev = 4.208

## One-way ANOVA: L.Wt(g) versus Des

Source	DF	SS	MS	F	Р
Des	2	2.63	1.31	1.26	0.288
Error	1.17	122.24	1.04		
Total	119	124.86			

S = 1.022 R-Sq = 2.10% R-Sq(adj) = 0.43%

				Individual 95% CIs For Mean Based on Pooled StDev
Level	N	Mean	StDev	
1	40	1.919	1.126	(
2	40	1.557	0.944	* (+
3	40	1.739	0.988	(
				1.50 1.80 2.10 2.40

Pooled StDev = 1.022

.;

# Results for: james3.MTW One-way ANOVA: Wt(g) versus Des

 Source
 DF
 SS
 MS
 F
 P

 Des
 2
 5790
 2895
 0.53
 0.590

 Error
 117
 638526
 5457

 Total
 119
 644317

S = 73.87 R-Sq = 0.90% R-Sq(adj) = 0.00%

Individual 95% CIs For Mean Based on Pooled StDev

)

				TOOTCA				
Level	N	Mean	StDev	+	+	+	+	
1	40	183.13	62.57		(	,	*	;
2	40	167.75	83.27 .	1				
3	40	169.13	74.33	(		*	)	
				+	+	+	+	
				144	160	176	192	

Pooled StDev = 73.87

## One-way ANOVA: SL(cm) versus Des

Source	DF	SS	MS	F	P	
Des	2	12.12	6.06	0.62	0.540	
Error	117	1143.55	9.77			
Total	119	1155.67				

S = 3.126 R-Sq = 1.05% R-Sq(adj) = 0.00%

Individual 95% CIs For Mean Based on Pooled StDev

Level	N	Mean	StDev	-+	+	+	+
1	40	23.300	2.160	(	*.		)
2	40	23.625	3.625		(	*	)
3	40	24.075	3.394		(		*)
				-+	+	+	
				22.40	23.10	23.80	24.50

Pooled StDev = 3.126

# One-way ANOVA: L.Wt(g) versus Des

S = 0.7341 R-Sq = 1.28% R-Sq(adj) = 0.00%

			2.2.1.	Individua Pooled St	1 95% CIs Dev	For Mean I	Based on
Level	N	Mean	StDev	+	+	+	+
1	40	1.7325	0.6414		(	*	) .
2	40	1.5698	0.8026	(	*	)	
3	40	1.5470	0.7491	(	*	)	
				+	+	+	+
				1.40	1.60	1.80	2.00

Pooled StDev = 0.7341

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.....

# Results for: james4.MTW One-way ANOVA: Wt(g) versus Des

Source	DF 2	SS 5897	MS 2948	F F 0.35 0.704
Error Total	357 359	3000835	8406	
s = 91.	. 68	R-Sq = 0	.20%	R-Sq(adj) = 0.00%
				Individual 95% Cls For Mean Based on Pooled StDev
Taual		Mean	StDev	
		181.16	100 77	()
1 2		182.46	98 62	(*
3		173.31	72 20	()
3	121	1/3.31	13.20	+++++
				168 180 192 204
Pooled	StDe	v = 91.68	B	
One-w	ay Al	VOVA: SI	_(cm) v	ersus Des
Source	DF	S	s MS	F P
				1.00 0.370
		31381.0		
		31556.		
rocar	555	51550.	1000	
S = 9.	376	R-Sq = 0	0.56%	R-Sq(adj) = 0.003
				Individual 95% CIs For Mean Based on
				Pooled StDev

				Pooled	StDev			
Level	N	Mean	StDev	+				
1	121	25.236	15.309		(		)	
2	118	23.881	4.238	(		)		
3	121	23.661	3.104	(	+	)		
				+	+		+	
				22.5	24.0	25.5	27.0	

Pooled StDev = 9.376

# One-way ANOVA: L.Wt(g) versus Des

Source	DF	SS	MS	F	Р
Des	2	3.615	1.807	1.94	0.146
Error	356	332.354	0.934		
Total	358	335.969			

S = 0.9662 R-Sq = 1.08% R-Sq(adj) = 0.52%

				Individual 95% CIs For Mean Based on Pooled StDev
Level	N	Mean	StDev	++++++
1	120	1.8614	1.1002	(+++++++
2	118	1.7338	1.0313	(
3	121	1.6165	0.7296	()
				++++++
				1.50 1.65 1.80 1.95

Pooled StDev = 0.9662

....

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# **One-way ANOVA: Muscle versus Des**

SourceDFSSMSFFDes2149.8574.9311.500.000Error117762.156.51Total119912.00

s = 2.552 R-Sq = 16.43? R-Sq(adj) = 15.00\*

Individual 951 CIs For Mean Pased on Fooled Sthev Level N Mean StDev ---+----

1	40	2.675	3.100			(	
2	40	2.900	3.020			(*-	
3	40	0.425	0.903	.(*-	)		
					in a second in a second		and the start
				0.0	1.2	2.4	3.6

Pooled StDev = 2.552

## **One-way ANOVA: Abd Cavity versus Des**

DesDFSSMSFPDes20.26670.13332.790.066Error1175.60000.0479Total1195.8667

S = 0.2188 R-Sq = 4.55% R-Sq(adj) = 2.91?

Individual 95% Cls For Mean Based on Pooled StDev

Level	N	Mean	StDev	-+				
1	40	0.0000	0.0000	(		)		
2	40	0.1000	0.3789			(		)
3	40	0.0000	0.0000	(				
				-+				
				-0.060	0.000	0.060	0.120	

Pooled StDev = 0.2188

....

## **One-way ANOVA: Liver versus Des**

Source	DF	SS	MS	F	P
Des	2	0.0000000	0.0000000	+	ł
Error	117	0.0000000	0.0000000		
Total	119	0.0000000			

S = 0 R-Sq = \*? R-Sq(adj) = \*?

Individual 95° CIs For Mean Based on Pooled StDev

				a contract a state of	
Level.	N	Mean	StDev	4	
1	40	0.000000000	0.000000000	*	
2	40	0.000000000	0.000000000	*	
3	40	0.000000000	0.000000000	+	
				0.000000 0.000010 0.000020 0.000030	

Fooled StDev = 0.000000000

# One-way ANOVA: Muscle versus Des

 Source
 DF
 SS
 MS
 F
 P

 Des
 2
 0.0000000
 0.0000000
 \*
 \*

 Error
 117
 0.0000000
 0.0000000
 \*
 \*

 Total
 119
 0.0000000
 \*
 \*
 \*

S = 0 R-Sq = \*\* R-Sq(adj) = \*\*

Individual 957 CIS For Mean Based on Fooled StDev

1

4

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+

Level	N	Mean	Strev
1	40	0.000000000	0.000000000
2 .	40	0.000000000	0.0000000000
3	40	0.000000000	0.000000000

t.....t....t...t....t. 0.000000 0.000010 0.000020 0.000030

Pooled StDev = 0.000000000

# One-way ANOVA: Abd. Cavity versus Des

Source	DF	SS	MS	F	F
Des	2	0.0000000	0.0000000	+	+
Error	117	0.0000000	0.0000000		
Total	119	0.0000000			

S = 0 R-Sq = \*% R-Sq(adj) = \*%

Level	N	Mean	StDev
1	40	0.000000000	0.000000000
2	40	0.000000000	0.0000000000
3	40	0.000000000	0.000000000

Individual	951 CI	s For	Mean	Based	on
Fooled StDe	v				
• • • • • • • • • • • • •	. 1		+		
+					

Pooled StDev = 0.000000000

## **One-way ANOVA: Liver versus Des**

Source	DF	SS	MS	F	P
Des	2	0.0000000	0.0000000	+	+
Error	117	0.0000000	0.0000000		
Total	119	0.0000000			

S = 0 R-Sq = \*% R-Sq(adj) = \*%

Individual 95° CIs For Mean Based on Fooled StDev

Level	N	Mean	StDev
1	40	0.000000000	0.000000000
2	40	0.000000000	0.000000000
3	40	0.000000000	0.000000000

0.000000 0.000010 0.000020 0.000030

Pooled StDev = 0.000000000

....

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#### **One-way ANOVA: Muscle versus Des** DF SS MS F P 2 1.667 0.833 3.82 0.025 117 25.500 0.218 Source Des 117 Error 119 27.167 Total S = 0.4668 R-Sq = 6.13% R-Sq(adj) = 4.53% . Individual 95% CIs For Mean Based on Fooled StDev ' +---..... -----StDev Level N Mean 40 0.2500 0.8086 1 2 (---40 0.0000 0.0000 ---1 40 0.0000 0.0000 3. +-----0.15 0.00 0.15 0.30 Pooled StDev = 0.4668One-way ANOVA: Abd. Cavity versus Des F P Source DF SS MS 2 0.0000000 0.0000000 117 0.0000000 0.0000000 \* Des Error 119 0.0000000 Total R-Sq = \* R-Sq(adj) = \*S = 0Individual 95% CIs For Mean Based on Pooled StDev StDev +----+----+----Level N Mean 40 0.00000000 0.00000000 1 40 0.00000000 0.00000000 2 40 0.00000000 0.00000000 3 +--0.000000 0.000010 0.000020 0.000030 Pooled StDev = 0.000000000One-way ANOVA: Liver versus Des DF Source SS MS. F P DF SS MS. 2 0.0000000 0.0000000 117 0.0000000 0.0000000 + Des Error 119 0.0000000 Total S = 0 R-Sq = \*% R-Sq(adj) = \*% Individual 95% CIs For Mean Based on Pooled StDev Level N StDev Mean 40 0.00000000 0.00000000 1 40 0.00000000 0.00000000 40 0.00000000 0.00000000 2 3 +----0.000000 0.000010 0.000020 0.000030 Pooled StDev = 0.000000000

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....

 
 One-way
 ANOVA:
 Muscle versus
 Des

 Source
 DF
 SS
 MS
 F
 ·P

 Des
 2
 303.25
 151.62
 57.39
 0.000

 Error
 357
 943.14
 2.64

 Total
 359
 1246.39
 S = 1.625 R-Sq = 24.33% R-Sq(adj) = 23.91% Individual 95% Cls For Mean Based on Fooled StDev manufa un annam forman or a famale coundress a N Mean StDev 121 1.983 2.763 Level ( 1 2 3 Pooled StDev = 1.625**One-way ANOVA: Abd Cavity versus Des**  
 Source
 DF
 SS
 MS
 F
 P

 Des
 2
 0.0878
 0.0439
 2.67
 0.071
 Des 357 5.8678 0.0164 Error Total 359 5.9556 S = 0.1282 R-Sq = 1.47% R-Sq(adj) = 0.92% Individual 95° CIs For Mean Based on Pooled StDev N Mean StDev 121 0.0331 0.2211 StDev Level (.....) 1 118 0.0000 0.0000 2 -0.020 0.000 0.020 0.040 121 0.0000 0.0000 3 Pooled StDev = 0.1282**One-way ANOVA: Liver versus Des**  
 DF
 SS
 MS
 F
 P

 2
 0.0000000
 0.0000000
 \*
 \*

 357
 0.0000000
 0.0000000
 \*
 \*
 Source DF Des Error 359 0.0000000 Total R-Sq = \* R-Sq(adj) = \*S = 0 
 N
 Mean
 StDev

 121
 0.000000000
 0.000000000

 118
 0.000000000
 0.000000000
 Level 1 2 3 121 0.00000000 0.00000000 Individual 95% CIs For Mean Based on Pooled StDev +----+-Level. 1 2 + 3 + +----0.000000 0.000010 0.000020 0.000030 Pooled StDev = 0.000000000

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# **Results for: JAMES.MTW**

# Two-Sample T-Test and CI: Wt1A, Wt2A

Two-sample T for Wt1A vs Wt2A

	N	Mean	StDev	SE Mean	
Wt1A	53	130.1	48.5	6.7	
Wt2A	67	224	111	14	

Difference = mu (Wt1A) - mu (Wt2A) Estimate for difference: -93.846095% CI for difference: (-123.7731, -63.9188)T-Test of difference = 0 (vs not =): T-Value = -6.23 P-Value = 0.000 DF = 94

### Two-Sample T-Test and CI: TL1A, TL2A

Two-sample T for TL1A vs TL2A

	N	Mean	StDey	SE	Mean
TL1A	53	25.31	2.22		0.30
TL2A	67	29.40	6.15		0.75

Difference = mu (TL1A) - mu (TL2A) Estimate for difference: -4.08420 95% CI for difference: (-5.69585, -2.47255) T-Test of difference = 0 (vs not =): T-Value = -5.04 P-Value = 0.000 DF = 86

### Two-Sample T-Test and CI: SL1A, SL2A

Two-sample T for SL1A vs SL2A.

 N
 Mean'
 StDev
 SE
 Mean

 SL1A
 53
 22.02
 1.92
 0.26

 SL2A
 67
 25.43
 5.46
 0.67

Difference = mu (SL1A) - mu (SL2A) Estimate for difference: -3.41397 95% CI for difference: (-4.84102, -1.98691) T-Test of difference = 0 (vs not =): T-Value = -4.76 P-Value = 0.000 DF = 85

### Two-Sample T-Test and CI: PC1A, PC2A .

\* ERROR \* All values in column are identical.

## Two-Sample T-Test and CI: LWt1A, LWt2A

Two-sample T for LWt1A vs LWt2A

N Mean StDev SE Mean LWt1A 53 1.262 0.421 0.058 LWt2A 67 2.35 1.23 0.15

4

Difference = mu (LWt1A) - mu (LWt2A) Estimate for difference: -1.08811 95% CI for difference: (-1.40742, -0.76880) T-Test of difference = 0 (vs not =): T-Value = -6.78 P-Value = 0.000 DF = 84

# Two-Sample T-Test and CI: Wt1C, Wt2C

Two-sample T for Wt1C vs Wt2C

	N	Mean	StDev	SE Mean	
Wt1C	116	172.9	73.0	6.8	
Wt2C	5	164.0	95.6	43	

Difference = mu (Wt1C) - mu (Wt2C) Estimate for difference: 8.9310395% CI for difference: (-111.19446, 129.05652) T-Test of difference = 0 (vs not =): T-Value = 0.21 P-Value = 0.847 DF = 4

### Two-Sample T-Test and CI: TL1C, TL2C

Two-sample T for TL1C vs TL2C

	N	Mean	StDev	SE	Mean	
TL1C	116	27.33	3.40		0.32	
TL2C	5	26.90	4.22		1.9	

Difference = mu (TL1C) - mu (TL2C) Estimate for difference: 0.427586 95% CI for difference: (-4.883688, 5.738860) T-Test of difference = 0 (vs not =): T-Value = 0.22 P-Value = 0.834 DF = 4

## Two-Sample T-Test and CI: SL1C, SL2C

Two-sample T for SL1C vs SL2C

 N
 Mean
 StDev
 SE
 Mean

 SL1C
 116
 23.72
 3.16
 0.29

 SL2C
 5
 23.30
 3.83
 1.7

Difference = mu (SL1C) - mu (SL2C) Estimate for difference: 0.415517 95% CI for difference: (-4.414183, 5.245217) T-Test of difference = 0 (vs not =): T-Value = 0.24 P-Value = 0.823 DF = 4

## Two-Sample T-Test and CI: PC1C, PC2C

\* ERROR \* All values in column are identical.

### Two-Sample T-Test and CI: LWt1C, LWt2C

Two-sample T for LWt1C vs LWt2C

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	N	Mean	StDev	SE Mean
LWt1C	116	1.603	0.724	0.067
LWt2C	5	1.84	1.24	0.55

Difference = mu (LWt1C) - mu (LWt2C) Estimate for difference: -0.232724 95% CI for difference: (-1.779424, 1.313975) T-Test of difference = 0 (vs not =): T-Value = -0.42 P-Value = 0.698 DF = 4

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# Results for: JAMES1B.MTW

# Two-Sample T-Test and CI: Wt1, Wt2

Two-sample T for Wt1 vs Wt2

	N	Mean	StDev	SE Mean
Wt1	288	169.8	83.0	4.0
Wt2	72	220	. 110	13

Difference = mu (Wt1) mu (Wt2) Estimate for difference: -49.968795% CI for difference: (-77.5214, -22.4161)T-Test of difference = 0 (vs not =): T-Value = -3.60 P-Value = 0.001 DF = 92

## Two-Sample T-Test and CI: TL1, TL2

Two-sample T for TL1 vs TL2

	N	Méan	StDev	SE Mean
TL1	288	27.07	3.86	0.23
TL2	72	29.22	6.05	0.71

Difference = mu (TL1) mu (TL2) Estimate for difference: -2.1493195% CI for difference: (-3.63664, -0.66198)T-Test of difference = 0 (vs not =): T-Value = 2.87 P-Value = 0.005 DF = 85

## Two-Sample T-Test and CI: SL1, SL2

Two-sample T for SL1 vs SL2

	. N	Mean	StDev	SE	Mean
SL1	288	23.50	3.52		0.21
SL2	72	25.28	5.37		0.63

Difference = mu (SL1) mu (SL2) Estimate for difference: -1.78299 95% CI for difference: (-3.10751, -0.45847) T-Test of difference = 0 (vs not =): T-Value = -2.68 P Value = 0.009 DF = 86

## Two-Sample T-Test and CI: PC1, PC2

\* ERROR \* All values in column are identical.

## Two-Sample T-Test and CI: LWt1, LWt2

Two-sample T for LWt1 vs LWt2

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	N	Mean	StDev	SE Mean
LWt1	288	1.606	0.835	0.049
LWt2	72	2.31	1.23	0.14

Difference = mu (LWt1) - mu (LWt2) Estimate for difference: -0.70795195% CI for difference: (-1.011075, -0.404828)T-Test of difference = 0 (vs not =): T-Value = -4.64 P-Value = 0.000 DF = 88

# Regression Analysis: LogWt1A versus LogSL1A

The regression	equation is	
LogWt1A = -6.0	5 + 3.51 Log	JSL1A

Predictor	Coef	SE Coef	Т	E	
Constant	-6.052	1.159	-5.22	0.000	
LogSL1A	3.5130	0.3753	9.36	0.000	

S = 0.239961 R-Sq = 63.23 R-Sq(adj) = 62.5%

Analysis of Variance

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Source	DF	SS	MS	F	F
Regression	1	5.0457	5.0457	87.63	0.000
Residual Error	51	2.9367	0.0576		
Total	52	7.9824			

Unusual Observations

Obs	LogSL1A	LogWt1A	Fit	SE Fit	Residual	St	Resid
15	2.94	4.8283	4.2919	0.0632	0.5364		2.32R
17	3.00	3.9120	4.4721	0.0478	-0.5600		-2.38R
18	3.00	3.9120	4.4721	0.0478	-0.5600		-2.38R
21	2.83	4.2485	3.9011	0.1012	0.3474		1.60 X

R denotes an observation with a large standardized residual. X denotes an observation whose X value gives it large influence.

# Regression Analysis: LogWt2A versus LogSL2A

The regression equation is LogWt2A = -1.76 + 2.19 LogSL2A

Predictor	Goef	SE Coef	Ŧ	р
Constant	-1.7633	0.6658	-2.65	0.010
LogSL2A	2.1895	0.2067	10.59	0.000

S = 0.350123 R-Sq = 63.33 R-Sq(adj) = 62.83

Analysis of Variance

Source	DF	SS	MS		F	Р
Regression	1	13.757	13.757	112.	2.2	0.000
Residual Error	65	7.968	0.123			
Total	66	21.725				

Unusual Observations

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Obs	LogSL2A	LogWt2A	Fit	SE Fit	Residual	St Resid
23	2.71	3.6889	4.1659	0.11.31	-0.4771	-1.44 X
36	3.69,	6.3969	6.3135	0.1070	0.0835	0.25 X
40	2.80	3.6889	4.3746	0.0951	-0.6857	-2.04R
41	3.04	3.9120	4.9026	0.0553	-0.9906	-2.87R
47	2.71	3.8067	4.1659	0.1131	-0.3593	-1.08 X
54	2.71	3.6889	4.1659	0.1131	-0.4771	-1.44 X
58	3.83	4.7875	6.6195	0.1340	-1.8320	-5.66RX

R denotes an observation with a large standardized residual. X denotes an observation whose X value gives it large influence.

## Regression Analysis: LogWt1B versus LogSL1B

The regression equation is LogWt1B = -4.51 + 3.03 LogSL1B

4

 
 Predictor
 Coef
 SE Coef
 T
 P

 Constant
 -4.5116
 0.4583
 -9.85
 0.000

 LogSL1B
 3.0326
 0.1447
 20.96
 0.000
 S = 0.247491 R-Sq = 79.0% R-Sq(adj) = 78.8% Analysis of Variance 
 Source
 DF
 SS
 MS
 F
 P

 Regression
 1
 26.904
 26.904
 439.23
 0.000

 Residual Error
 117
 7.166
 0.061
 0.061

 Total
 118
 34.070
 0
 0
 , Unusual Observations JSL1BLogWt1BFitSEFitResidualStResid3.565.94026.27050.0611-0.3304-1.38 X3.565.99156.27050.0611-0.2791-1.16 X Obs LogSL1B LogWt1B 3.56 1 2 

 6.3969
 6.5199
 0.0723
 -0.1230
 -0.52 X

 5.9915
 6.3135
 0.0630
 -0.3221
 -1.35 X

 4.6052
 5.1263
 0.0228
 -0.5212
 -2.11R

 3 3.64 3.57 4 50 3.18 2.94 3.9120 4.4179 0.0389 -0.5058 -2.07R 61 2.94 3.9120 4.4179 0.0389 -0.5058 69 -2.07R 81 3.71 6:2146 6.7504 0.0828 -0.5358 -2.30RX 5.9915 6.4798 0.0705 -2.06RX 82 -0.4883 3.62 105 2.92 3.6889 4.3370 0.0421 -0.6481 -2.66R 109 3.04 5.3936 4.7214 0.0284 2.73R 0.6722

R denotes an observation with a large standardized residual. X denotes an observation whose X value gives it large influence.

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## Regression Analysis: LogWt1C versus LogSL1C

The regression equation is LogWt1C = -5.01 + 3.19 LogSL1C

 Predictor
 Coef
 SE Coef
 T
 P

 Constant
 -5.0097
 0.4928
 -10.17
 0.000

 LogSL1C
 3.1878
 0.1559
 20.44
 0.000

S = 0.213669 R-Sq = 78.6% R-Sq(adj) = 78.4%

Analysis of Variance

Source	DF	SS	MS	E,	Р	
Regression	1	19.083	19.083	417.98	0.000	
Residual Error	114	5.205	0.046			
Total	115	24.287				

Unusual Observations

Obs	LogSL1C	LogWt1C	Fit	SE Fit	Residual	St Resid
37	3.58	5.9915	6.4137	0.0693	-0.4222	-2.09RX
43	3.18	4.6052	5.1211	0.0201	-0.5160	-2.43R
45	2.89	3.6889	4.2041	0.0462	-0.5152	-2.47R
47	2.94	3.9120	4.3764	0.0387	-0.4644	-2.21R
50	2.94	3.9120	4.3764	0.0387	-0.4644	-2.21R
57	3.04	5.1930	4.6955	0.0266	0.4975	2.35R
105	3.57	.5.9915	6.3691	0.0672	-0.3776	-1.86 X
106	3.47	5.7683	6.0382	0.0519	-0.2699	-1.30 X

R denotes an observation with a large standardized residual. X denotes an observation whose X value gives it large influence.

## Regression Analysis: LogWt2C versus LogSL2C

The regression equation is LogWt2C = - 8.01 + 4.12 LogSL2C

5 cases used, 216 cases contain missing values

 Predictor
 Coef
 SE Coef
 T
 P

 Constant
 -8.010
 1.783
 -4.49
 0.021

 LogSL2C
 4.1223
 0.5677
 7.26
 0.005

S = 0.188473 R-Sq = 94.6% R-Sq(adj) = 92.8%

Analysis of Variance

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Source	DF	SS	MS	F	P
Regression	1	1.8727	1.8727	52.72	0.005
Residual Error	3	0.1066	0.0355		
Total	4	1.9793			

# Regression Analysis: LogWt1 versus LogSL1

The regression equation is LogWt1 = -4.90 + 3.15 LogSh1

 Predictor
 Coef
 SE Coef
 T
 P

 Constant
 -4.9048
 0.3143
 -15.60
 0.000

 LogSL1
 3.15318
 0.09979
 31.60
 0.000

S = 0.232749 R-Sq = 77.7° R-Sq(adj) = 77.7°

Analysis of Variance

 Source
 <sup>5</sup>
 DF
 SS
 MS
 F
 P

 Regression
 1
 54.090
 54.090
 998.48
 0.000

 Residual Error
 286
 15.493
 0.054
 1000

 Total
 287
 69.583
 0.054
 0.054

.

# Regression Analysis: LogWt2 versus LogSL2

The regression equation is LogWt2 = -2.07 + 2.28 LogSL2

72 cases used, 216 cases contain missing values

 Predictor
 Coef
 SE Coef
 T
 P

 Constant
 -2.0731
 0.6505
 -3.19
 0.002

 LogSL2
 2.2821
 0.2023
 11.28
 0.000

S = 0.350831 R-Sq = 64.5° R-Sq(adj) = 64.0°

Analysis of Variance

Source	DF	SS	MS	F	P
Regression	1	15.662	15.662	127.25	0.000
Residual Error	70	8.616	0.123		
Total	71	24.278			

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## Results for: james1a.MTW

### Descriptive Statistics: PC1A, LWt1A

 Variable
 N
 N\*
 Mean
 SE Mean
 StDev
 Minimum

 PC1A
 53
 0
 0.00000000
 0.00000000
 0.00000000
 0.00000000
 0.00000000
 0.00000000
 0.00000000
 0.00000000
 0.00000000
 0.00000000
 0.00000000
 0.00000000
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 Variable
 Q1
 Median
 Q3
 Maximum

 PC1A
 0.00000000
 0.00000000
 0.00000000
 0.00000000

 LWt1A
 0.9600
 1.3100
 1.4050
 2.5500

## **Descriptive Statistics: PC2A**

 Variable
 N
 Mean
 SE Mean
 StDev
 Minimum
 Q1
 Median
 Q3
 Maximum

 PC2A
 67
 0
 3.627
 0.341
 2.795
 1.000
 1.000
 3.000
 5.000
 11.000

## **Descriptive Statistics: PC1C**

 Variable
 Q1
 Median
 Q3
 Maximum

 PC1C
 0.000000000
 0.000000000
 0.000000000
 0.000000000

## **Descriptive Statistics: PC2C**

....

 Variable
 N
 Mean
 SE Mean
 StDev
 Minimum
 Q1
 Median
 Q3
 Maximum

 PC2C
 5
 216
 2.000
 0.632
 1.414
 1.000
 1.000
 3.500
 '4.000

Results for: James1b.mtw Descriptive Statistics: PC2 Variable N N\* PC2 73 216 3.514 SE Mean St Dev Minimum 1.000 1.000 1.000 Median 3.000 4.250 Variable Maximum PC2 11 000 Descriptive Statistics: PC1