

**OCCURRENCE AND DISTRIBUTION OF FISH PARASITES OF POTENTIAL
THREAT TO THE AQUACULTURE SECTOR ALONG THE KAVANGO RIVER,
NAMIBIA**

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BY

Victoria Mumba

200305158

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Supervisor: Dr Clinton Hay

Co-supervisors: Prof. Jo G. Van As

Prof. Liesl L. Van As

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Victoria Mumba

Date:

Student number: 200305158

ABSTRACT

Cichlids are subject to a wide range of diseases and parasites. Previously studies were conducted on fish parasites on the Okavango Delta, Botswana. This thesis is the first on fish parasites in the Kavango River, Namibia.

The study aims to identify parasites occurring on commercially farmed fish i.e.: *Oreochromis andersonii*, *Tilapia rendalli* and *Serranochromis robustus* and to determine if these parasites may have potential threat to aquaculture and humans.

For the purpose of identification and calculating prevalence (pr) and abundance (ab) parasites collected were fixed and stained using standard methods for each taxon. Kruskal-Wallis One Way Analysis of Variance was used to determine whether there were any significant differences in the number of fish parasites per zone, fish species and fish size. No significant differences were found in the number of parasites found in each zone, as well as the host fish species and group size of target fish species. A total of 205 specimens (91 *T. rendalli*, 89 *O. andersonii* and 25, *S. robustus*), were collected and examined for endo- and ectoparasites over twelve months. Of these 102 were infested with various parasites (total pr 49.7%), *T. rendalli*, 45%, *O. andersonii* 63% and *S. robustus* 13%. Parasites included *Trichodina* sp. (ab0.18) followed by *Dactylogyrus* sp. (ab0.17). Some *Tripartiella* and *Epistylis* spp. were collected in low levels as compared to other protozoan parasites. Helminths parasites such as *Contracecum* sp. were found in high abundance, whilst *Proteocephalus*, *Clinostomum* and *Acanthocephala* spp. were found in low abundance and infected mostly *S. robustus*.

Opistholernaea (ab0.02) occurred in high numbers on *T. rendalli* and *O. andersonii* in zone 1 (Katwitwi to Kasivi). *Dolops ranarum*, *Lernaea hardingi* and *Lamproglana monodi* were also

collected (ab0.02). Most of the infested fish species ranged from 1– 20.9 cm in total length, while the parasite *Contracecum* sp. was mostly found in fish ranging from 21 – 30.9 cm. Most of the infested fish were found in zones 3 (Mbambi to Popa Falls) and 4 (Popa Falls to Kwetze).

This study could be a useful management tool for fisheries researchers in the aquaculture industry. In conclusion some protozoans and parasitic crustaceans, if found in high abundance, may cause a threat to aquaculture. Other parasites such as *Contracecum* sp. could be a concern to human health when fish is eaten raw, half cooked or inadequately smoked.

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CHAPTER ONE

INTRODUCTION

Parasites are common among fish and affect them negatively in several ways, which poses a potential threat to the sustainability of fisheries, which is a source of protein, and income to several communities in the world (Douëllou, 1992; Paperna, 1996). In fish farming, parasites may be highly pathogenic, contributing significantly to high mortalities and economic losses, while in natural systems; parasites may threaten the abundance and diversity of indigenous fish species (Douëllou, 1992; Paperna, 1996).

Fish are subject to a wide range of diseases and parasites, which are well-known in southern Africa (Skelton, 2001). In the wild, deteriorating river conditions throughout Southern Africa are causing stress to fish populations with a consequent rise in parasite infestation and fish disease (Skelton, 2001). There are a number of parasites that are common and could negatively affect fish populations as documented by Skelton (2001). These include: Protozoans, Nematode worms, Trematode worms, tapeworms, leeches, and parasitic crustacean, which in extreme cases cause mortality in fish.

In this study, the occurrence and distribution of parasites in three fish species were investigated: Three spot tilapia, (*Oreochromis andersonii*), the *Redbreast tilapia (*Tilapia rendalli*) and the Nembwe, (*Serranochromis robustus*) as they are commonly farmed fish in the Caprivi and Kavango region (Van Der Waal, 1991). *This species name recently changed to *Coptodon rendalli*, but *T. rendalli* is still used in this dissertation as study was completed and submitted before the changes.

1.1. Objective of the study

Infectious fish diseases are caused by bacteria, fungi or viruses, but can also be parasitic borne. Parasite infection in fish refers to a diseased condition resulting from organisms living in or on the fish (Bassey, 2011); therefore, a thorough study on endo- and ectoparasites on farmed fish is essential.

The objectives of this study were:

1. to identify fish parasites that occurs on commercially farmed tilapia species along the Kavango River and at specific habitats in the river system.
2. to compile a data base on the occurrence and distribution of fish parasites.
3. to identify fish parasites that could potentially threaten the fish farming industry in the Kavango region.

1.2. Problem Statement

The lack of any documentation and epidemiology of most of the parasitic diseases, particularly intestinal helminths and tissue invading protozoans occurring in fresh water fish in the Kavango River has received relatively little attention. Therefore, this gap may result in Fisheries Officers/Researchers not having reliable knowledge and experience to make sound decisions based on scientific information to advise the inland fisheries and aquaculture sector with regards to parasitic borne diseases.

1.3. Hypothesis of study

Hypotheses of this study are:

H1 = There is a no significant difference in the number of infested fish in the four different zones.

H2 = There is no significant difference in the number of fish infested in the three different fish host species.

H3 = There is no significant difference in the number of fish infested with the group size of target fish species.

1.4. Introduction to Aquaculture

To understand aquaculture internationally and in a Namibian context, definitions need to be provided. Aquaculture being the farming of aquatic organisms including fish, mollusc, crustaceans and aquatic plants in freshwater, brackish-water and sea water environment (El-Sayed, 2006). This has become an important industry and fast growing sector worldwide. With a constant increase in consumer's demand for fish products, aquaculture is expanding in new directions, becoming more intensive with a diversity of species (Chakraborty & Hancz, 2011).

Cichlids, especially tilapia, are frequently used for aquaculture production (Blier *et al.*, 2010). Tilapia is one of the most common fish species cultured both in small and large-scale aquaculture farm operations, in developed and developing countries (Lim & Webster, 2006). Tilapia's, primarily of the genus *Oreochromis*, are the second most important group of farmed fish worldwide. In 2006, 2.4 million tonnes were produced from farms in over 100 countries including Africa's largest producer Egypt, with 250 000 tonnes (Blier *et al.*, 2010).

1.5. Global overview on Aquaculture

Globally, the demand of aquatic products has increased, while stock fish are over exploited, posing pressure on the environment and wild stock. Therefore, nations around the world are searching for alternatives to meet the local and international demands for fish and fisheries products. Aquaculture's contribution to the total global fisheries landings was very low during the 1950 – 1970, ranging from 3.2% in 1950 to 5.2% in 1970 (El-Sayed, 2006). In 2009 the global aquaculture production reached 73 million tonnes growing at an annual rate of 8 to 10 % (FAO, 2011). This is mostly by Asia contributing 91% amounting to 67 million tonnes of the total world aquaculture production in 2009 (FAO, 2011).

Figure 1.1. Illustrates the global aquaculture production which has been dominated by freshwater fishes with 41% and the remaining 59% is from marine aquaculture. From the marine environment, 75% constitutes aquatic plants and shellfish (FAO, 2011).

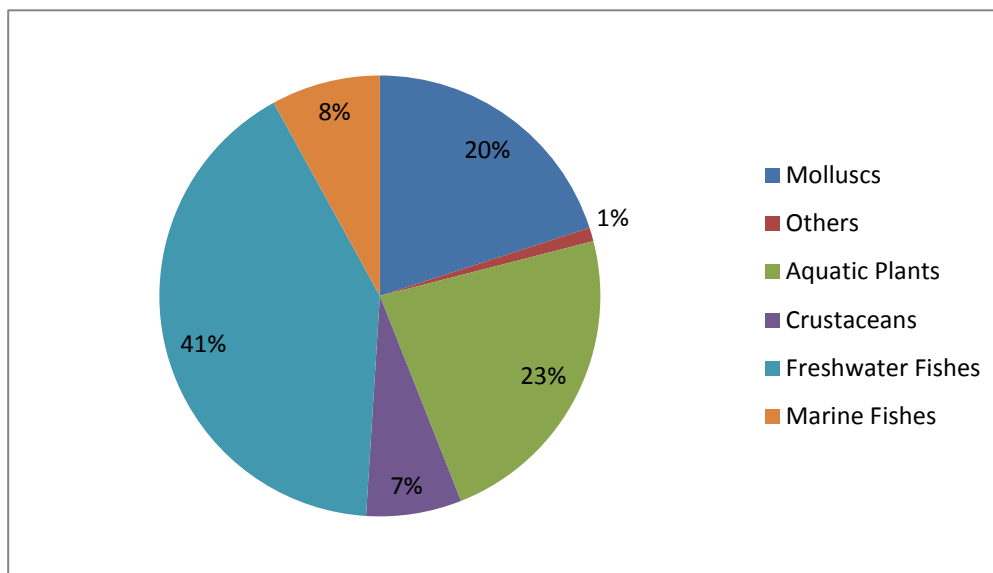


Figure 1.1: Global output of aquaculture for 2009 (FAO, 2011)

In the case of freshwater aquaculture it is mostly dominated by tilapia species (El-Sayed, 2006), as tilapia has characteristics such as high resistance to viral, bacterial and parasitic diseases compared to any other cultured species that make them ultimately good for aquaculture (El-Sayed, 2006). Aquaculture production of tilapia increased between 1970 and 2009 (El-Sayed, 2006), from 28 260 tonnes to 2 586 403 tonnes (Table 1.1). China, Egypt, the Philippines, Indonesia and Thailand are the countries that have contributed the most to aquaculture production and where trade mostly occurs internationally.

Table 1.1: Major cultured tilapia species production in tonnes during 2009 (from FAO, 2011)

Species	Total global Production in 2009 (tonnes)
Nile tilapia (<i>Oreochromis niloticus</i>)	2 542 960
Mozambique tilapia (<i>Oreochromis mossambicus</i>)	33 169
Three-spot tilapia (<i>Oreochromis andersonii</i>)	3090
Blue tilapia (<i>Oreochromis aureus</i>)	3 131
Red breast tilapia (<i>Tilapia rendalli</i>)	2 585
Longfin tilapia (<i>Oreochromis macrochir</i>)	1 174
Sabaki tilapia (<i>Oreochromis spilurusspilurus</i>)	165
Redbelly tilapia (<i>Tilapia zillii</i>)	129
Total	2 586 403

Source: (Adopted from FAO, 2011).

Aquaculture in Namibia is still in its infancy stage, although policies are being implemented for opening profitable investment opportunities in the aquaculture sector, such as the Eco-fish farm in Namibia.

1.6. Status of aquaculture in Africa

Africa contributed only 3 % of global aquaculture production for 2009, whereby most of this production is contributed by Egypt (FAO, 2011). However, in line with global trends, the aquaculture industry in Africa has also experienced considerable growth, although on a small scale.

History of aquaculture in Africa dates back to the 1920's with the introduction of trout in Kenya, Madagascar and Tanzania (Toguyeni, 2010). Recently, Southern African countries have started farming different types of fish species, although *Tilapia* species still remain the dominant farmed fish.

Contrary to other countries, the aquaculture industries in most Southern African countries are lagging behind due to lack of sufficient finances on national and regional aquaculture related programmes (Hempel, 2007). A large percentage of governmental aquaculture facilities are either abandoned or currently dysfunctional for various reasons (Gupta *et al.*, 2004). The greatest increase in production in Africa has been since 1998. Egypt is the most important aquaculture producer in Africa producing 705 tonnes in 2009/2010 (FAO, 2011).

Limited use of indigenous species for farming is one of the reasons for failure in aquaculture, as well as the insufficient knowledge of fish pathology. Many authors strongly suggest that governments should encourage aquaculture development based on native species (Toguyeni, 2010).

Southern Africa has a wide variety of native fish species and several of these have been studied and used in aquaculture production such as *Tilapia rendalli*, and *Oreochromis andersonii*. The decision about which species to use in aquaculture depends mainly on the environmental conditions and scale

of the aquaculture enterprise. Small-scale rural fish farmers mostly use species that are promoted by aquaculture authorities (Van der Mheen, 1994).

If exotic species are to be introduced, governments should ensure that adequate infrastructures are put in place, and have researchers with knowledge and skills on how to tackle possible outbreak of fish diseases (Stickney, 2000). Research in the past was focused on the adaptation of the culture of exotic species to local conditions, but has since gradually moved towards the development of culture methods for native species (Van der Mheen & Haight, 1994).

Integrated aquaculture might offer solution for small holding farmers in Southern African Development Community (SADC) as the potential for this type of farming is high. According to a FAO (2003) study based on Geographic Information Systems (GIS), seven of the 14 SADC countries have large areas in which warm water fish farming could be further developed.

Freshwater aquaculture in Angola to date has no statistical data (FAO, 2007). Aquaculture in Angola started before Independence, and was practiced in a basic way at the initiative of the private sector in some State facilities. Its development was interrupted by the difficulties derived from the civil war. It is only recently that it is attracting the interest of the private sector (FAO, 2005). However, available information indicates that it is being implemented with low level technology and without control. Tilapia production is carried out in some farms, but there are no records of harvest volumes or of relevant results (FAO, 2007).

Aquaculture sub-sector in Botswana has been one of the national goals in recent years focused on creating jobs and ensuring food security (ACP, 2011). Small-scale fish farming has been introduced in

some parts of the country. The government has supported development of a hatchery facility, to produce approximately 500 000 mixed fingerlings of tilapia and catfish for stock enhancement in dams and freshwater systems to supply to potential fish farmers (J. Choto, personal Communication, 2012). As in most Southern African countries, Botswana's aquaculture sector is still at a very early stage and development is limited by challenges such as relatively high capital costs and inadequate technical skills and experience in fish farming (ACP, 2011).

Aquaculture in Botswana started to receive attention during the 1980's and 1990's through the ALCOM (Aquaculture for Local Communities) Development Program, which undertook a number of activities, organized meetings and produced a number of publications on small scale rural aquaculture in Southern Africa (ALCOM, 1999). The driving force of ALCOM activities was technical, focusing on the dissemination of technology for smallholder farmers and low input farming in impounded water bodies (ALCOM, 1999). Fingerling production was underlined as the main technical constraint to aquaculture in Botswana at the time (ALCOM, 1996). The outcome of these activities has been the stocking of dams with fingerlings produced by the Government Fisheries Division hatchery at Mmadinare.

1.7. Status of freshwater aquaculture in Namibia

Freshwater aquaculture in Namibia is reported to have started in the late 1800's, but has been promoted and acknowledged only in 2003. Foreign aquaculture expertise have conducted studies after independence and reported that Namibia's aquaculture sector, mainly mariculture, was under the development of the socio-economical aspects of the country (Rana & Abban, 2011).

The initial promotion of Freshwater aquaculture in Namibia was to promote food security by facilitating the provision of fingerling production to farmers and rural communities for fish farming (MFMR, 2006) but this approach is shifting to economic activity in freshwater aquaculture.

Aquaculture species currently being farmed in the northern parts of Namibia is dominated by three main fish species namely; African Sharptooth catfish (*Clarias gariepinus*), Three spot tilapia (*Oreochromis andersonii*) and Redbreast tilapia (*Tilapia rendalli*). Most farmers are either farming with one or various combinations of the three species.

Four pilot fish farms are currently in operation in the North Eastern regions (Table 1.2). In the Zambezi region, a number of fish farmers are farming with Tilapia because the demand for catfish is very small due to cultural beliefs. An exception is the Likunganelo fish farm, which farms with both tilapia and catfish.

In the Northern region, two fish farms are involved in catfish and tilapia farming. Catfish is more common for fish consumer in the Northern region, as it is also found in the wild in floodplains (Oshana's). Tilapia production in the Northern regions is mostly produced by fish farming.

Table 1.2: Namibian Government pilot fish farms and farmed fish species in Northern and North Eastern regions according to MFMR (2011).

Number of farms/species farmed	Catfish and Tilapia	Catfish	Tilapia	Total
Zambezi/Kavango	2	0	2	4
Northern regions	2	0	0	2

Source: Adopted from Rana & Abban, (2011).

1.8. Development of aquaculture in Namibia

The government of Namibia has placed a high priority on the development of the aquaculture sector, particularly freshwater aquaculture to contribute towards poverty alleviation and unemployment in rural areas. This initiative was supported by the country's Vision 2030 as well as the National Development Plan (NDP2 and NDP3) (Rana & Abban, 2011).

Large investments have gone into Namibian aquaculture, as part of accomplishing Vision 2030. The government of Namibia have realized the potential in freshwater aquaculture, thus they have undertaken initiatives such as implementing pilot projects and making use of vast natural resources for aquaculture purposes. In 2000, the government of Namibia established an Aquaculture policy and legal framework to promote fish farming in Namibians. Following this the government commissioned a series of feasibility studies, marketing and value added studies, in order to obtain a national view on the potential and scope for aquaculture development (Rana & Abban, 2011).

Several pilot projects were established (Table 1.2). A hatchery was built at the Inland Aquaculture Center in Omahenene/Onavivi in the Northern region with the purpose to supply small scale farmers with fingerlings. Six community based pilot projects in the North-Eastern region, and a privately owned fish farm was established in the Hardap region.

Total inland aquaculture production was estimated at 85 and 95 tonnes during 2007 and 2008, respectively. This was mainly contributed by the Eco-Fish farm, while the government funded pilot projects fish farms contributed only 10 and 12 tonnes of the total production in 2007 and 2008 respectively.

1.9. Aquaculture challenges in Namibia

With the development of Aquaculture in Namibia, several challenges have been encountered such as the lack of:

- Demand in International Markets for Namibian farmed freshwater fish
- Human resources development (expertise in Freshwater aquaculture)
- Training, knowledge and skill guidelines for running fish farms
- Good quality of Fish feed
- Potential candidate species for freshwater aquaculture in Namibia
- Implementation of management/community based aquaculture concept.

A challenge related to this study is the lack of expertise and knowledge of freshwater fish related diseases. In both cultured and natural environments diseases have a serious impact on fish; these diseases can be parasitic borne (Fig. 1). The fisheries industry worldwide has a huge concern and awareness for aquatic diseases affecting fishery stocks (Noga, 2010).

1.10. Current and potential fish species farmed in Namibia

Cichlids that are commercially farmed in Namibia are Three spot tilapia, (*Oreochromis andersonii*) and the Redbreast tilapia (*Tilapia rendalli*). In terms of aquaculture, these species are the most commonly farmed species in both the Kavango and Caprivi Regions (Van der Waal, 1991), while the Nembwe (*Serranochromis robustus*) is still under investigation as a future potential fish species.

1.11. Impacts of fish parasites in aquaculture in Namibia

Most cichlids such as tilapia are characterized by resistance to stress and diseases and withstanding poor water conditions, unlike other cichlid species such as *Serranochromis* species. Although tilapia has been classified as disease resistant (El-Sayed, 2006), water quality plays an important role in the process of controlling diseases.

Diseases are globally reported as one of the many serious threats to the success of commercial aquaculture (Noga, 2010). One of the most significant diseases in tilapia around the world, and particularly in closed culture systems, is caused by *Streptococcus* bacteria. This bacterium is known to be opportunistic as it mostly attacks fish that are exposed to stress conditions in a cultured system (El-Sayed, 2006). It has therefore been responsible for significant losses in tilapia fisheries.

Parasites is one of the causes of diseases that constitute a major threat in aquaculture production worldwide (Nicholas, 2000). The parasites usually affect the growth of commercially produced fish to market size, thus raising concern in public health, especially in areas where raw or smoked fish are eaten (Paperna, 1996). In fish farming or aquaculture, possibilities of high pathogen infection can occur, contributing to high fish mortalities and economic losses, while in natural systems, parasites may threaten the abundance and diversity of indigenous fish species (Douëllou, 1992; Paperna, 1996).

Disease and parasites infection of aquaculture fish species in Africa have been poorly studied in Africa as a whole and in sub-Saharan Africa in particular (South Africa being the exception) (Hecht & Endemann, 2007). It is mainly due to the low-level intensity of aquaculture in the region. At present the lack of research on fish diseases in Africa is not seen as a factor that will negatively impact on aquaculture development and as such is not a target research area (Hecht & Endemann, 2007). A study was performed to assess the impacts of pathogenic agents in fish and shellfish aquaculture in Sub-

Sahara Africa. The study revealed that bacterial infections as well as parasitic ectoproteozoans, Helminths and parasitic crustaceans are the most prevalent problems in African freshwater, brackish water and marine aquaculture (Hecht & Endemann, 2007).

Literature reported fish parasites such as protozoa and arthropods as being harmful to fish (Van As & Basson 1984), as stated in a joint symposium paper on aquaculture in South Africa. Once these parasites infest fish, they may cause serious outbreaks of diseases related to parasitic infestations and fish mortalities.

1.12. Outline of thesis: The different aspects (excluding the introduction that was dealt with in this chapter) of the current study are discussed in the following chapters:

Chapter 2: The Literature review of the target species is dealt with in this chapter. It contains the morphological description of the three studied species *Oreochromis andersonii*, *Tilapia rendalli* and *Serranochromis robustus*, and their geographic distribution caused by high water levels of the Kavango River and their history. This chapter also deals with the general morphological features (on genus level) of the fish parasites encountered during the present study.

Chapter 3: In the Methodology a description of the study area is provided including the seven main sampling areas along the Kavango River, sampling methods as well as gear used. The research design is discussed including data recording. Methods used for parasite examination and preservation are provided.

Chapter 4: Contains the overall results of the present study.

Chapter 5: Comprises the discussion that was made from the results obtained.

Chapter 6: Includes some recommendations for future research on fish parasites in aquaculture as well as the conclusion.

Chapter 7: The thesis ends with the references chapter followed by the appendix.

CHAPTER TWO

LITERATURE REVIEW OF TARGET FISH SPECIES AND FISH PARASITES

2.1 Introduction

Although the main concerns of the present study is the effects of fish parasites on the target fish species and its threats to aquaculture, but the implications of fish parasites on human health as well as the social economic well being are of equal importance.

In southern Africa there has been an increase interest in aquaculture, particularly the development of inland fisheries and aquatic animal health. Countries such as South Africa and Botswana have conducted parasitological surveys the last 15 to 20 years by Basson *et al.*, (2002). The impacts of fish parasites in aquaculture systems are significant to the present study as they not only affect fish, but also affect humans on a social economic point of view (Noga, 2010). The most obvious effect of the occurrence of fish disease is mortality, followed by economic losses (Mazid & Banu, 2002). Although there is a lack of fish parasite scientific literature review, the impacts of diseases have been reported in social economic terms such as nutritious food supply, job creation, poverty alleviation, income generation and food security and consumer confidence (Mazid & Banu, 2002).

Mass mortalities of Tilapia fry and fingerlings are due to protozoan and metazoan parasites are frequently reported. A small initial infection gradually leads to a serious outbreak of disease, resulting in large mortalities and great economic loss for small-scale farmers (Lim & Webster, 2006). The most common disease problem in Namibia is epizootic ulcerative syndrome (EUS). It has affected the Inland fisheries since 2006; it was first reported in 2008 in the Kavango River. Its impact has directly affected inhabitants that relied on fish from the river as their source of income and source of proteins

as the fish infected cannot be sold or consumed. It is obvious that disease outbreaks in tilapia culture systems have a great impact on low-income groups (Mazid & Banu, 2002).

The Cichlidae is a large family of fresh and brackish water fishes native to Africa, South and Central America, Madagascar, the Levant, parts of Arabia and India (Skelton, 2001). They are the largest fish family in Africa with about 900 described species and many more are yet to be described. In Southern Africa, eight genera and 42 species are found. Cichlid species are extremely important for food and biologically important in aquaria thus making them scientifically important in fields related to evolution, behavior and physiology (Skelton, 2001).

Three spot tilapia, *Oreochromis andersonii*, Redbreast tilapia, *Tilapia rendalli* and the Nembwe, *Serranochromis robustus* are some of the most important species in the fisheries of the Kavango and Caprivi Regions (Van der Waal, 1991). *Oreochromis andersonii* comprises up to 25 % of the commercial gill net catches in the Kavango River. Historically, *T. rendalli* made up a much smaller proportion of the catch comprising a mere 3% of the commercial gill net catch in Kavango River. *Serranochromis robustus* comprised only 0.4% of the commercial gill net catches in the Kavango River (Van der Waal, 1991).

Oreochromis andersonii and *S. robustus* are mouth brooders, both species building nests in the form of simple saucer-shaped depressions in sandy substrates which are guarded by males (Van der Waal, 1985, Merron, 1991). After fertilization, the females will mouth brood the eggs, larvae and fry (Bruton & De Moor, 1988). *Tilapia rendalli* are guarders, breeding pairs clearing vegetation and excavating nests with tunnel-like brood chambers on sandy substrates in shallow water (Van der Waal, 1985). Eggs are deposited in the brood chambers where the eggs and larvae are defended and fanned by both parents (Van der Waal, 1985).

The two tilapiine species (*O. andersonii* and *T. rendalli*) are habitat generalists, occurring across a range of habitats in the Upper Zambezi River, although they are usually most abundant in lagoons (Winemiller & Kelso-Winemiller, 2003). Both tilapiines are omnivores, *O. andersonii* feeds predominantly on vegetative detritus while *T. rendalli* feeds mainly on aquatic macrophytes (Winemiller & Kelso-Winemiller, 2003). The piscivorous *S. robustus* is common in the main river channels of the Upper Zambezi and Kafue Rivers and specializes in feeding on small *Synodontis* species (Winemiller, 1991).

Studies of these three species on movement and habitat utilization have been conducted in the past. The movement and habitat utilization of *O. andersonii* were studied by Thorstad *et al.*, (2001) and Økland *et al.* (2003), and that of *S. robustus* were studied by Thorstad *et al.*, (2002). The effects of catch and release angling on *O. andersonii* and *S. robustus* have been studied by Thorstad *et al.*, (2004) and the objectives of the studies were to investigate the movements and habitat utilisation of the fish species in all seasons at low, middle and high water levels.

MORPHOLOGICAL DESCRIPTION OF STUDIED TARGET FISH SPECIES

Oreochromis andersonii (Fig 2.1)



Adults have blue-grey with light scale borders. The fins are blue-grey with light spots on the soft dorsal and anal. The margins of dorsal and anal are bright red. Juveniles are silvery with 8 to 9 irregular thin bars on the body and 3 to 4 mid lateral spots (Skelton, 2001).

Breeding males are blue-black with silvery mesh and have maroon flush on top of head. The outer dorsal and caudal fins

Figure 2.1: *Oreochromis andersonii* collected from the Kavango River, Namibia

Oreochromis andersonii occurs in the Upper Zambezi, Kafue, Okavango and the Kunene Rivers (Skelton, 2001). The species is indigenous to countries such as Angola, Botswana and Namibia (Okavango River Basin), Zambia and Zimbabwe (Fig 2.4). Introduction of *O. andersonii* was done in Kenya in 1980, from Botswana to the Motti Pan for aquaculture purposes (Tweddle & Hay, 2009).

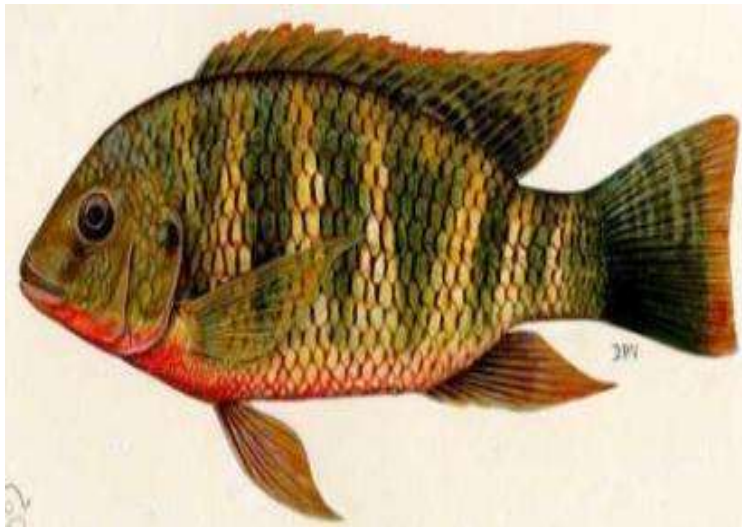
Oreochromis andersonii is an indigenous species in Namibia. It is found in rivers, streams, swamps and dams. Limited literature is available on *O. andersonii*, as studies have mostly been done on *O. niloticus* and *O. mossambicus* (El-Sayed, 2006).

The Three spot tilapia is the most preferred fish for local people residing along the Kavango River and in the northern parts of Namibia. It is also a preferred fish species for aquaculture in Namibia. Tilapia species are native to Africa and in Namibia the Three spot tilapia is an ideal candidate for freshwater

inland aquaculture because of fast growth and its tolerance to a wide range of environmental conditions (such as temperature, salinity, low dissolved oxygen).

Oreochromis andersonii tolerates fresh and brackish water, and prefers slow moving, standing waters, and backwater or floodplains lagoons. They feed mostly on detritus, diatoms and zooplankton (Skelton, 2001). *Oreochromis andersonii* is one of the most important fish species for subsistence and commercial fisheries (Hay *et al.*, 2000).

***Tilapia rendalli* (Fig 2.2)**



Head is typically convex with a mouth protruding with prominent bicuspid teeth. They have a bright red throat and chest; the extremities of the soft dorsal, anal and lower half of the caudal fin vary from yellow to red. Juveniles have a rounded head, beak like mouth, with few broad body bands (Skelton, 2001)

Figure 2.2: *Tilapia rendalli*, adopted from Skelton (2001).

Geographic location of *Tilapia rendalli* in Southern Africa is from the Kunene, Okavango, Zambezi systems including Lake Malawi, and east coastal rivers south to the Phongolo and coastal lakes to Lake Sibaya (Skelton 2001), as well as estuaries in Mozambique and KwaZulu-Natal (Fig 2.4).

Tilapia rendalli is a fish species tolerant to a wide temperature range, from 11 to 37 °C and salinities up to 19 parts per thousand. They inhabit well vegetated water along river littorals and backwaters,

floodplains and swamps. They usually feed on water plants and algae but will also eat aquatic invertebrates and even small fish (Skelton, 2001).

Tilapia rendalli is popularly used for angling and is valued in aquaculture and fisheries and sometimes used for weed control in cultured ponds (Skelton, 2001).

***Serranochromis robustus* (Fig 2.3)**



The mouth is large, with large well-spaced conical teeth. It is olive to bright green, with a deep olive band along the mid body. Two subspecies are described mainly on the basis of male breeding colours (Skelton, 2001)

Figure 2.3: *Serranochromis robustus* collected from the Kavango River, Namibia.

Serranochromis robustus is known from the Democratic Republic of Congo to Namibia and Botswana. In Southern Africa it is found in the Upper Zambezi, Okavango, Kafue and Zambian Congo systems (Skelton, 2001). Its native range in Zimbabwe is the Zambezi River above the Victoria Falls although two specimens were taken from Lake Kariba in 1968 (Balon, 1974) (Fig 2.4). It has been widely translocated by anglers in Zimbabwe and could be expected in almost any river or impoundments on the central plateau (Skelton, 2001).

Serranochromis robustus is a fish species that mainly feeds on small fish, including squeakers such as *Synodontis* sp., insects and other invertebrates (Jackson, 1961). Juvenile species of *Serranochromis robustus* mainly feed on minnows, and occur in, open and closed lagoons, and small tributaries (Skelton, 2001).

2.2.The influence of flooding on species distribution in the Kavango River

The fish communities are diverse, and their population dynamics, migration patterns, habitat use and production rates are poorly known. This means there is a high risk of resource exploitation and species distribution.

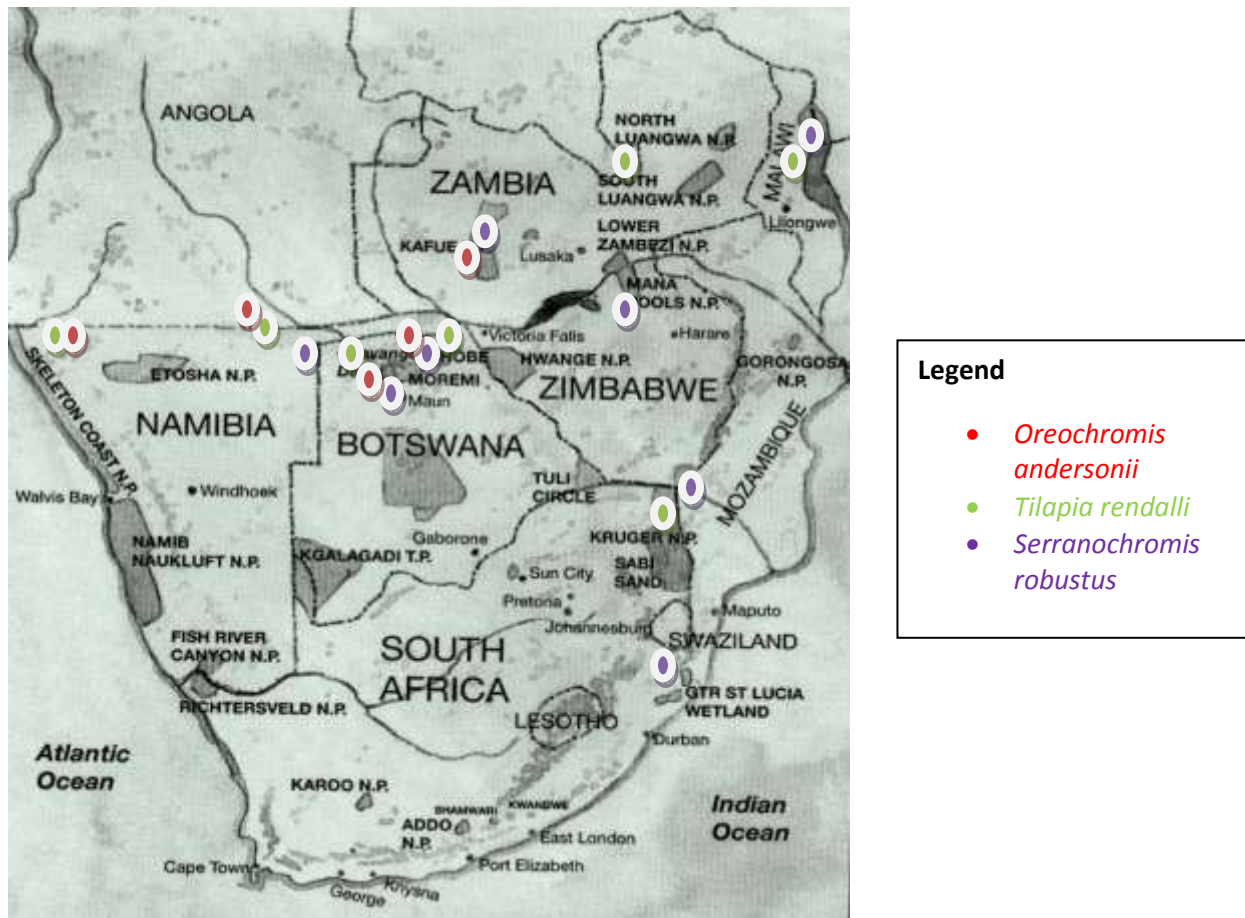


Figure 2.4: Distribution of *Oreochromis andersonii*, *Tilapia rendalli*, and *Serranochromis robustus* in Southern Africa.

The annual flood in the Namibian portion of the Okavango starts during December and reaches its peak in March – April where after it recedes during May. However, the intensity, timing and duration of the flood depend on the rainfall in Angola. The annual discharge of the Okavango River system in Rundu is between 5000 and 6000 million m³ (MAWF, 2011). This runoff is increased by the inflow from the Cuito River so that the annual discharge in the Okavango at Mukwe has nearly doubled to over 10 000 million m³. The water discharge is largest in April, both at Rundu and at Mukwe (MAWF, 2011).

The Okavango Delta situated in north-western Botswana, in the midst of the Kalahari Desert, is the largest wetland and if not one of the remaining untouched areas in Africa (Basson *et al.*, 2002). The Delta has an estimated annual inflow of approximately 11 000 million m³ to date, which is an increase of 5000 million m³ of rainfall annually (Hay *et al.*, 2000). Of this, 15 400 million m³ is lost to the atmosphere through evapo-transpiration annually while approximately 2% of the input appears as output at the distal end of the Delta at Mohembo (Hay *et al.*, 2000).

The dynamic ecosystem that is driven by an annual flood, takes approximately 6 months to flow the distance of around 250 km from Mohembo (at the Namibian border) to the town of Maun in the south. During this extended flood, water starts rising gradually from the end of December at Mohembo, reaching the Gumare fault around March to April and Maun by the end of July.

During these winter months, water temperatures fluctuate, dropping at night and rising during the sunny, warm winter days. These fluctuations, as well as crowding of fish in the relatively shallow water, stress the fish populations. One of the consequences of this stressed condition is a weakened immune system coupled with extremely high loads of parasites, specifically ectoparasites. During flooding seasons, the floodplains in the Delta become vast pools of water increasing the diversity of parasites in the Delta (Basson & Van As, 2006).

As the floodplains start receding in September, fish from the floodplain pools move into the main streams, backwaters and to a lesser extent the lagoons. Fish that remain in these habitats are exposed to a notable range of ectoparasites present. This and the ever present threatening low oxygen levels in the Okavango system, results in fish having higher level of parasites infestation than those expected in a natural system (Basson & Van As, 2006).

MORPHOLOGICAL DESCRIPTION OF FISH PARASITES ENCOUNTERED DURING THE STUDY.

2.3 General description of parasites

***Tripartiella* sp. and *Trichodina* sp. (Ciliophora)**

Members of the family Trichodinidae (Fig. 2.5 A-D) are ectoparasites, with most of the species reported from freshwater environments. The distribution of specialized host-specific parasite species such as trichodinids and sessiline ciliophorans follows that of their fish hosts, but may also be more restricted, sometimes to only one or a few watersheds (Fryer, 1961). Studies conducted on African fish reported a broad taxonomic diversity of trichodinids in Southern Africa, in particular in the Zambezi river system (Van As & Basson, 1989). The body size is about 40 - 90 µm in diameter.

General description of Ciliophora

Their cup- to cylinder-shaped structure is a distinct taxonomic feature (Van As & Basson, 1987). Small trichodinids, predominantly from the gills, are bell shaped (*Tripartiella* body diameter about 20 µm) and often settle on the tips of the gill lamellae (Van As & Basson, 1989). The ray (the inner extension of the denticle) is totally reduced (Van As & Basson, 1987).

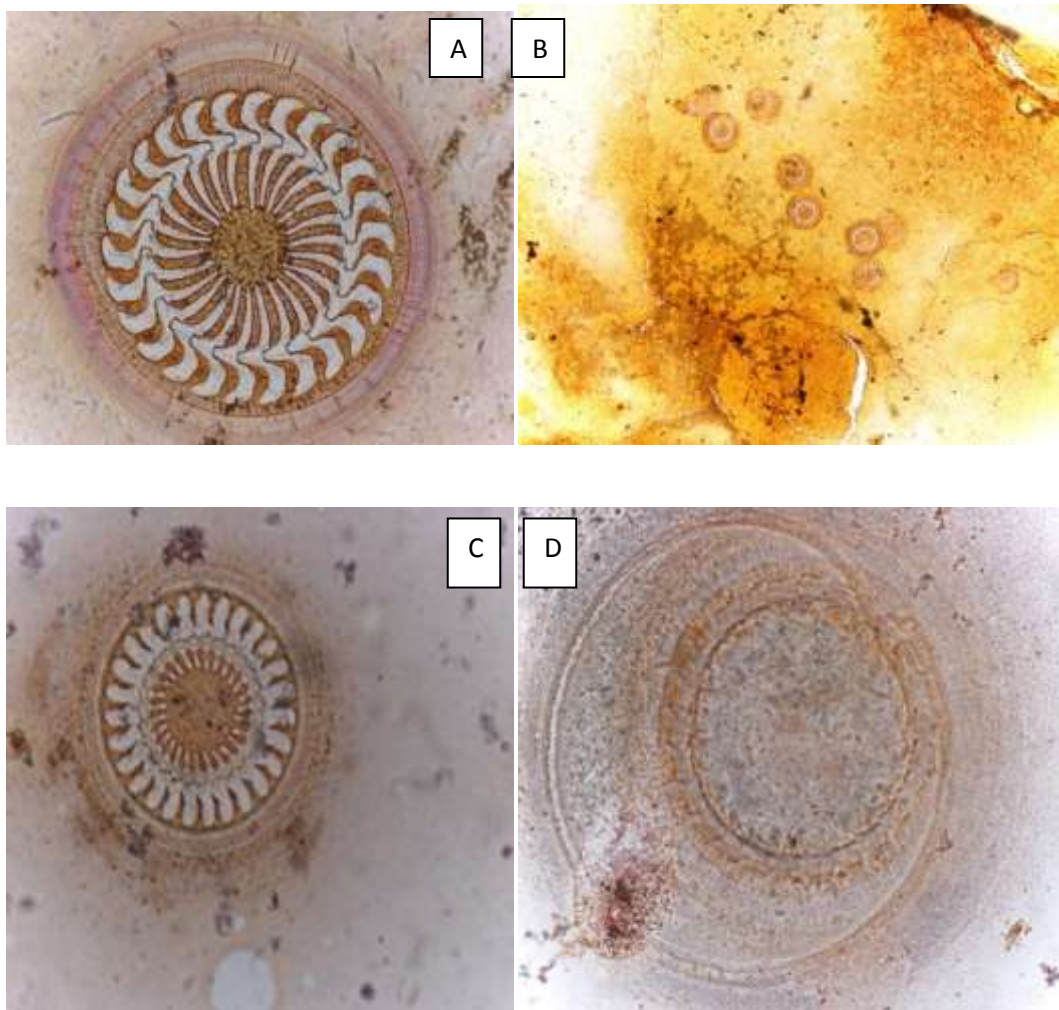


Figure 2.5: Light micrograph of silver impregnated smears of A: the adhesive disc of *Trichodina* sp. B: Infection of *Trichodina* sp. C: adhesive disc of *Tripartiella* sp. D: cup shaped body with of *Trichodina* sp. (magnification A, C&D 100x; B 40xc) (Photos by V. Mumba)

***Epistylis* sp. (Ciliophora)**

Members of the genus *Epistylis* (Fig. 2.6 A&B) may be extremely pathogenic under some circumstances such as those in cultured environments (Noga, 2010). *Epistylis* has been reported to

attach on the body of fish, causing erosion of scales and hard-fins and sometimes bone, hyperplasia and haemorrhaging of epithelial tissue as well as inflammation on the body (Hoffman, 1967).

General description of *Epistylis* sp.

Epistylis sp. is cup-shaped with horse shoe-shaped macronucleus. Body width and length is about 20X60 μm . Other general features of *Epistylis* sp. are;

- They are single-celled, but live in stalked colonies.
- The branching stalks are rigid and do not contract;
- Contains cells at the ends of the stalks called zooids.
- They contain cilia around the oral opening and contract when feeding.

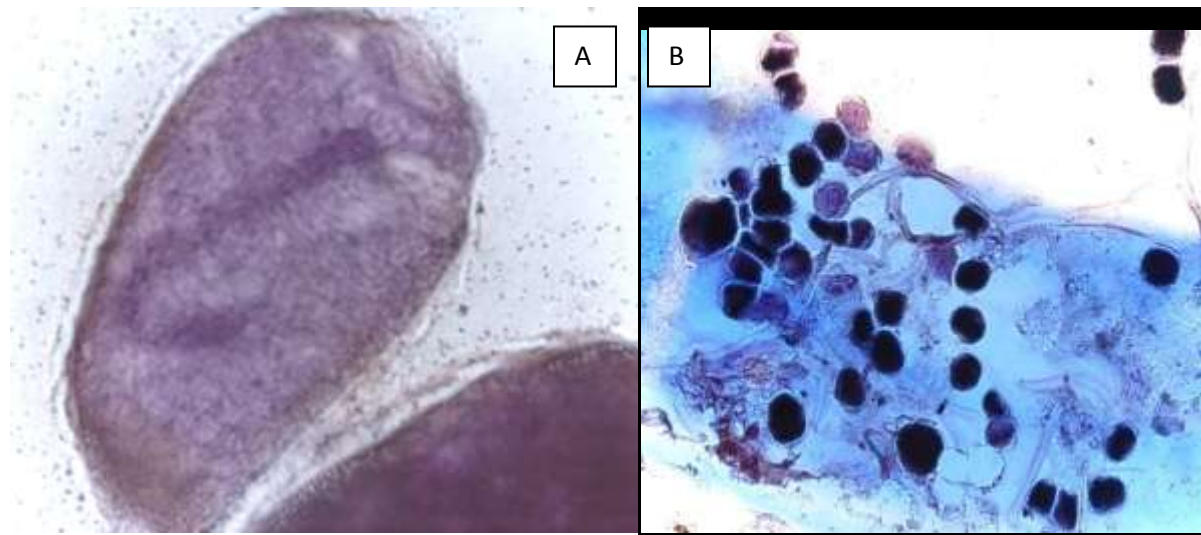


Figure 2.6: Light micrographs of Harris Haematoxylin stained micrograph of A: *Epistylis* sp. zooid. B: *Epistylis* sp. infestation. (Magnification A 100x; B 10x) (Photos by V. Mumba)

***Dactylogyrus* sp. (Monogenea)**

Monogenea is a parasite that affects freshwater and brackish water fish in most families including cichlids (Khalil, 1969). Exotic monogeneans such as *Dactylogyrus* sp. (Fig. 2.7A&B) were introduced with the fish hosts into Southern Africa such as the Okavango river basin (Paperna, 1980). Monogeneans are subdivided into several major taxa; *Dactylogyroidea*, *Caspaloidea* and *Polyopisthocotylea*. Most monogeneans found in inland water fish are *Dactylogyroidea* Monogeneans (Paperna, 1996).

General description of *Dactylogyrus* sp. (Monogenea)

Monogeneans are flatworms (Platyhelminthes), ectoparasitic and attached by special posteriorly positioned attachment organs to their host's skin or gills.

Dactylogyroidea have the following features;

- Usually 0.3–1.5 mm long
- Usually have one or two anterior-dorsal pairs of eyes
- A posterior-ventral attachment organ (the opisthaptor).
- This disk-like organ contains centrally positioned sclerotinoid anchors, connected to support bars and marginally located hooklets.



Figure 2.7: Light micrographs of A: live specimen of *Dactylogyrus* sp. with hooklets (arrow) on the opisthaptor for attachment. B: *Dactylogyrus* sp. entire specimen, arrow indicate eye spots. (Magnification 100x) (Photos by V. Mumba)

***Acanthocephala* sp. (Fig. 2.8 A&B)**

The number and arrangement of the hooks on the proboscis are the main criteria for differentiation of species. A wider range of anatomical details are considered for determination of higher taxa (Kabata, 1985). The worms can be up to 10 mm in length.

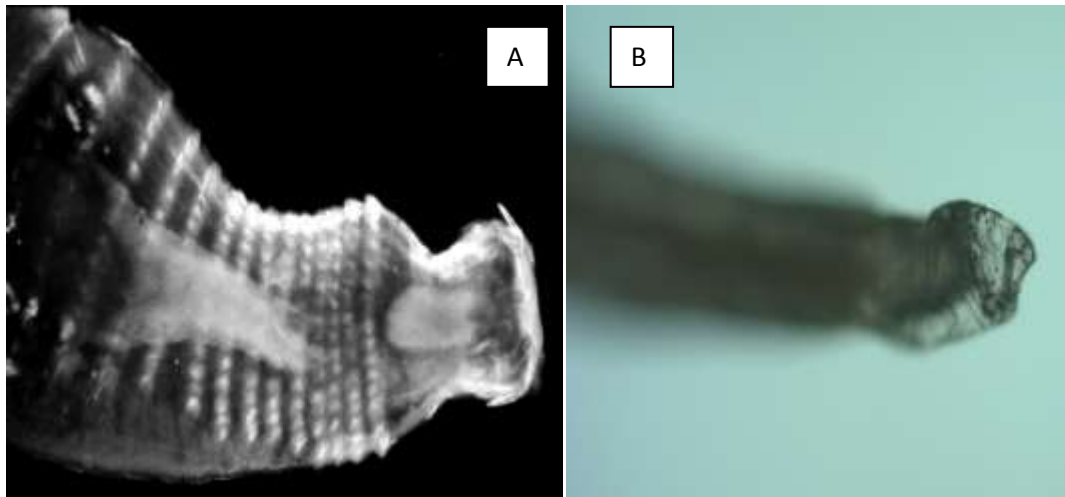


Figure 2.8: Light micrographs of head region of *Acanthocephala* sp. A: head with hooklets protruded, B head drawn inwards. (Magnification A&B: 100x ;) (Photos by V. Mumba)

Cestodes (Fig. 2.9)

In histology, the worms collected in the present study resemble the description that of *Proteocephalus glanduliger* by Mashego (2001) from South African dams. With records of abundance and diversity of proteocephalid cestodes in African freshwater fish (Khalil& Polling 1997), only one species, *Proteocephalus glanduliger* has been recorded in South Africa from *Clarias gariepinus* (Van As & Basson, 1984).

General description of *Proteocephalus* sp.

General characteristics used by Moravec (1975) to describe *Proteocephalus* sp. are as follows;

- Scolex unarmed
- Four cup –shaped suckers arranged symmetrically around a protrusible rostellum

- Neck region not differentiated into well-formed proglottids
- Body length 2 mm

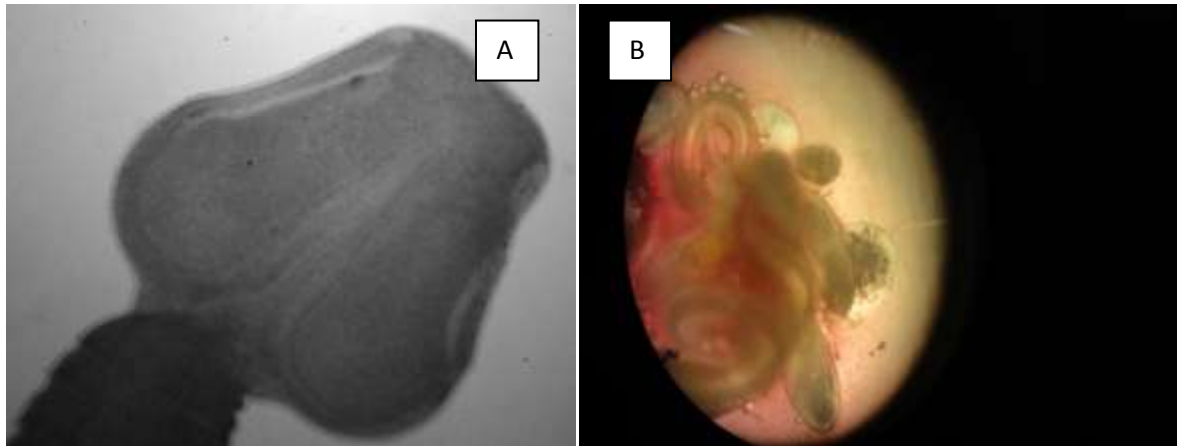


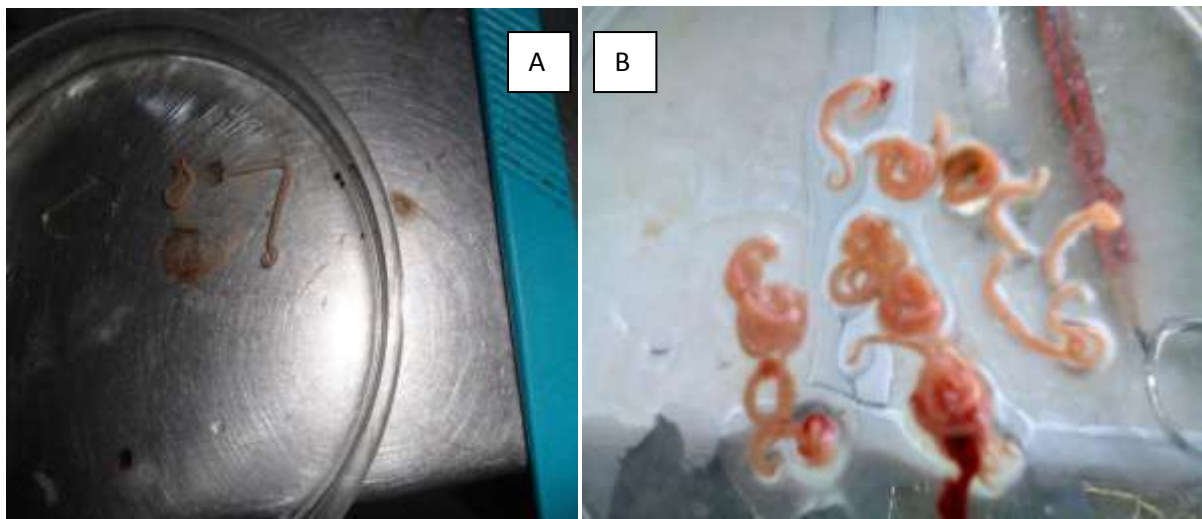
Figure 2.9: Light micrographs of A: Scolex of custody, B: Live specimen of custodies (Magnification A 100x; B 20x) (Photos by V. Mumba)

***Contracecum* sp. (Nematode)**

Nematode (round worms) are very distinctive in shape, with a solid cuticle. Because of their resistant cuticle these worms last longer than flatworms in post-mortem conditions. Most adult forms are large enough to be visible to the naked eye; body size ranges from 40 – 60 mm. Khalil (1971) reported 40 species of adult nematodes, representatives of nine families from fish in Africa. *Contracaecum* sp. larvae have been recorded from catfish and other species from South Africa, mainly from *Clarias gariepinus* (Mashego, 1977).

Contracecum sp.(Fig. 2.10 A-D) are larval nematodes occurring in fresh and brackish water, with heavier infections occurring in fish occupying higher positions in the food-chain, such as the predatory fish Nembwe with average lengths ranging from 15 to > 30 cm.

Identification of nematodes at larval stage is quite difficult, since they lack genital systems and several other features that are used as taxonomic identification criteria which are found on the adult stages of nematodes (Paperna, 1996). Although, the third stage has two blind caeca branching off from the intestinal canal between the oesophagus and the midgut, it has a shorter Venticular appendix and a longer intestinal caecum (Aliyu& Solomon, 2012).



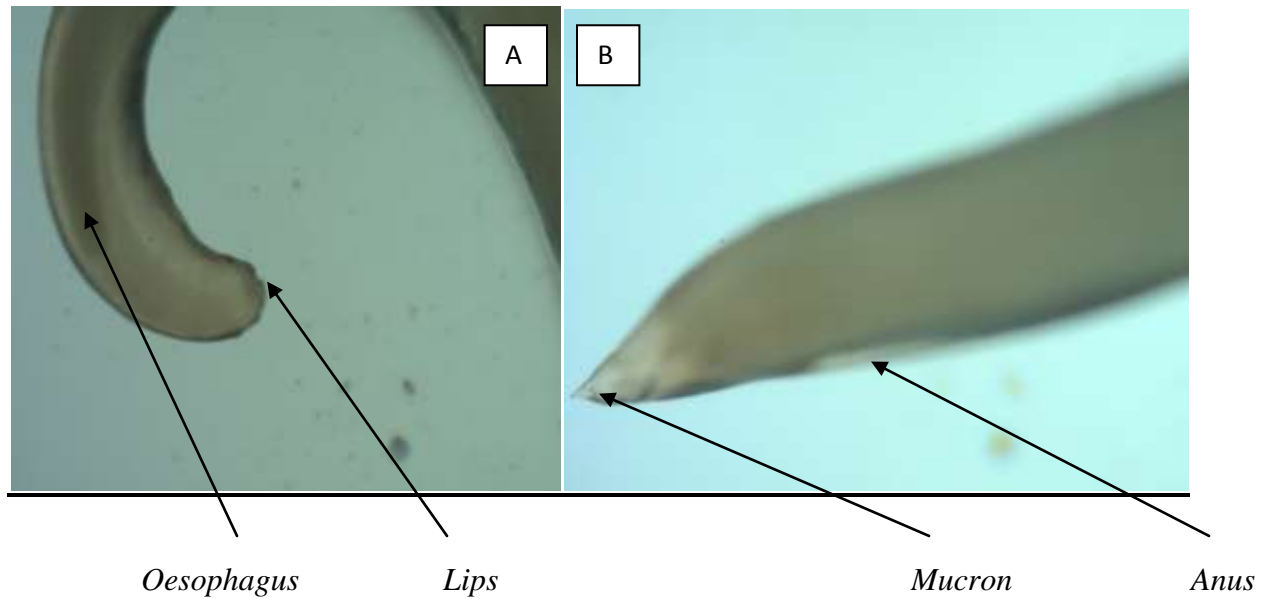


Figure 2.10: A & B: Micrographs of *Contracecum* specimens. Light micrograph of *Contracecum* sp. illustrating body features on head and tail region respectively, indicated by arrows. (Magnification A&B 100x) (Photos by V. Mumba)

***Clinostomum* sp. (Trematoda) (Fig. 2.11A&B)**

According to Khalil, (1971) 50 species of trematodes were listed from 15 families which all occur in freshwater fish in Africa. Clinostomatid cysts and worms are the largest (up to 5 mm in diameter and 10×3 mm in size) and the worm's intestine is loaded with a yellow to orange substance (Paperna, 1996)

General characteristics of *Clinostomum* sp. (Trematoda)

Adult-stage digeneans usually are;

- dorso-ventrally flattened,
- Oval body with a smooth, spiny or corrugated surface,

- A sucker around the antero-ventral mouth, (for attachment and locomotion).
- Additional ventral sucker or acetabulum (for attachment and locomotion).
- The digestive system consists of a pharynx connected to the mouth opening, a short esophagus and two blind intestinal caeca.
- Eggs are evacuated to the genital opening, and are usually oval and operculated
- Body size 8 mm

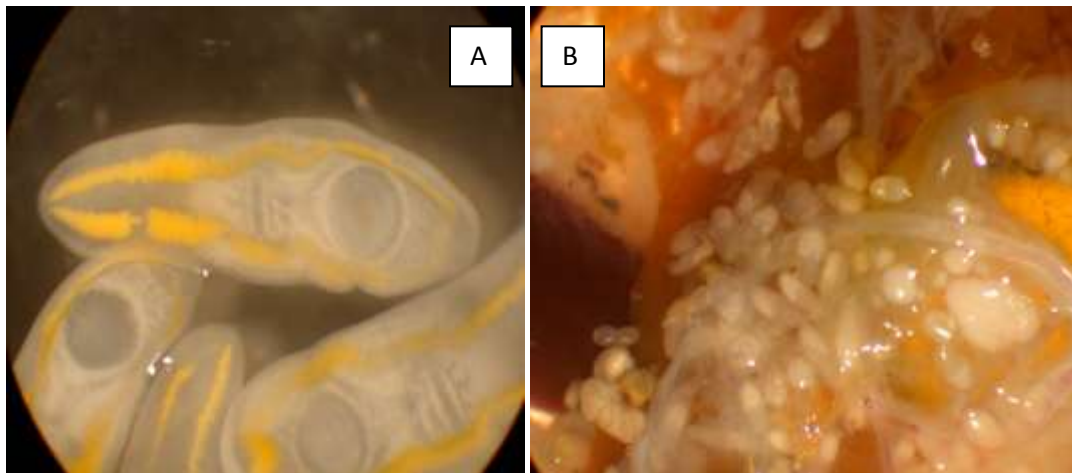


Figure 2.11: Light micrograph of Trematode specimens found in the intestines and tissues of fish hosts (Basson, 2010) Adapted by the University of Free State, Department of Zoology and Entomology. (Magnification A 100x; B 10x)

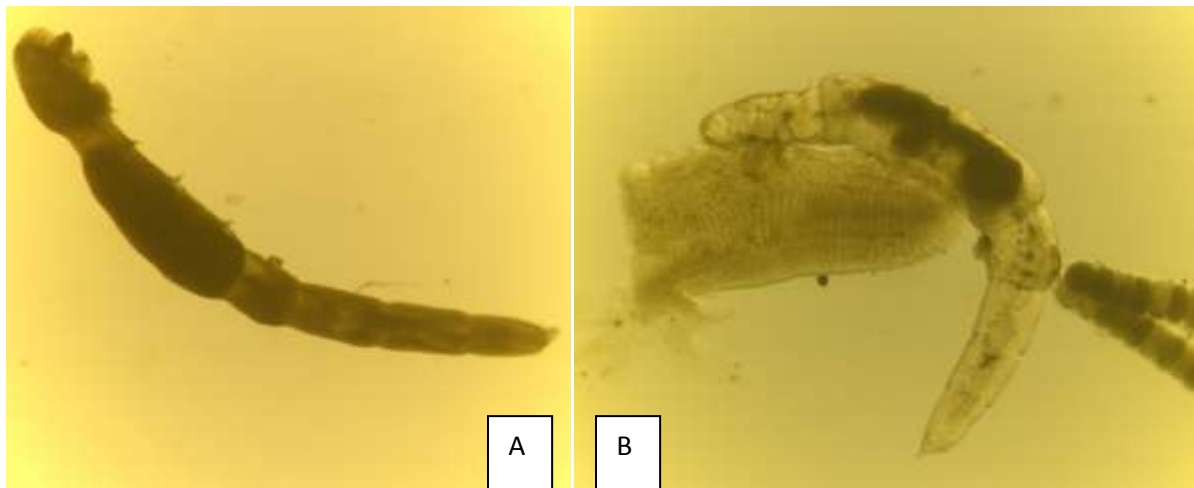
***Lamproglena monodi* (Copepoda)**

The species described was identified as *Lamproglena monodi* (Fig. 2.12 A-D) because it shows evidence of the characteristics described by Fryer (1961) in the original description.

General characteristics of *Lamproglena monodi*

General characteristics used by Fryer (1961) to describe *Lamproglena monodi* which is usually attached to gills are as follows;

- Body is Segmented and large mouth parts
- Cephalothorax always less than half total length,
- Second antenna small and weak,
- Maxillipeds with terminal claws,
- Thoracopods present, but sometimes fewer than 4 pairs.
- Average body length of 4 mm.



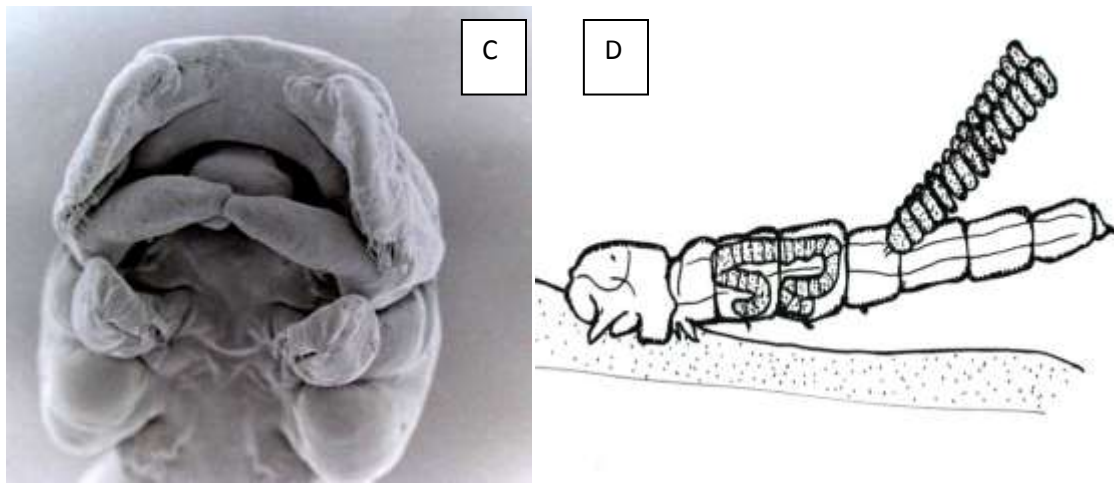


Figure 2.12: Light micrograph of *Lamproglena monodi* found on *Serranochromis robustus* in Botswana, Adapted by the Aquatic Ecology, University of the Free State, South Africa. A: Specimen removed from gills, B: *Lamproglena monodi* attaching to the gills of Nembwe (same here). C: Scanning electron micrograph of head region of *Lamproglena monodi*. D: Line drawing of *Lamproglena monodi* as described by Fryer (1961) (Magnification A-C 10x)

***Lerner herding* (Copepoda)**

Host preferences may differ in populations from different geographical regions; *L. hardingi* (Fig 2.13) throughout Africa is associated with Cichlidae, however in Lake Bangweulu and Lake Mweru they principally infect *Synodontis nigromaculatus*.

General characteristic of *Lernaea hardingi* (Copepoda):

- head with 2–3 (rarely 4) pointed or swollen horns
- The trunk may thicken gradually towards its posterior end.

- Average body length of 10 mm



Figure 2.13: Light micrograph of *Lernaea hardingi* found on fish in Botswana, adapted from the Department of Aquatic ecology, University of the Free State, South Africa. (Magnification 10x)

***Opistholernaea* sp. (Copepoda) (Fig. 2.14 A-D)**

General characteristics of *Opistholernaea* sp. (Copepoda);

- Head with 4 horns, with 2 posterior horns directed symmetrically backward to form a 90 degree angle.
- A lateral outgrowth extends from the “neck” region; it may be, however, located in very close proximity to the head to become functionally part of the anchor complex.
- Average body length of 12 mm

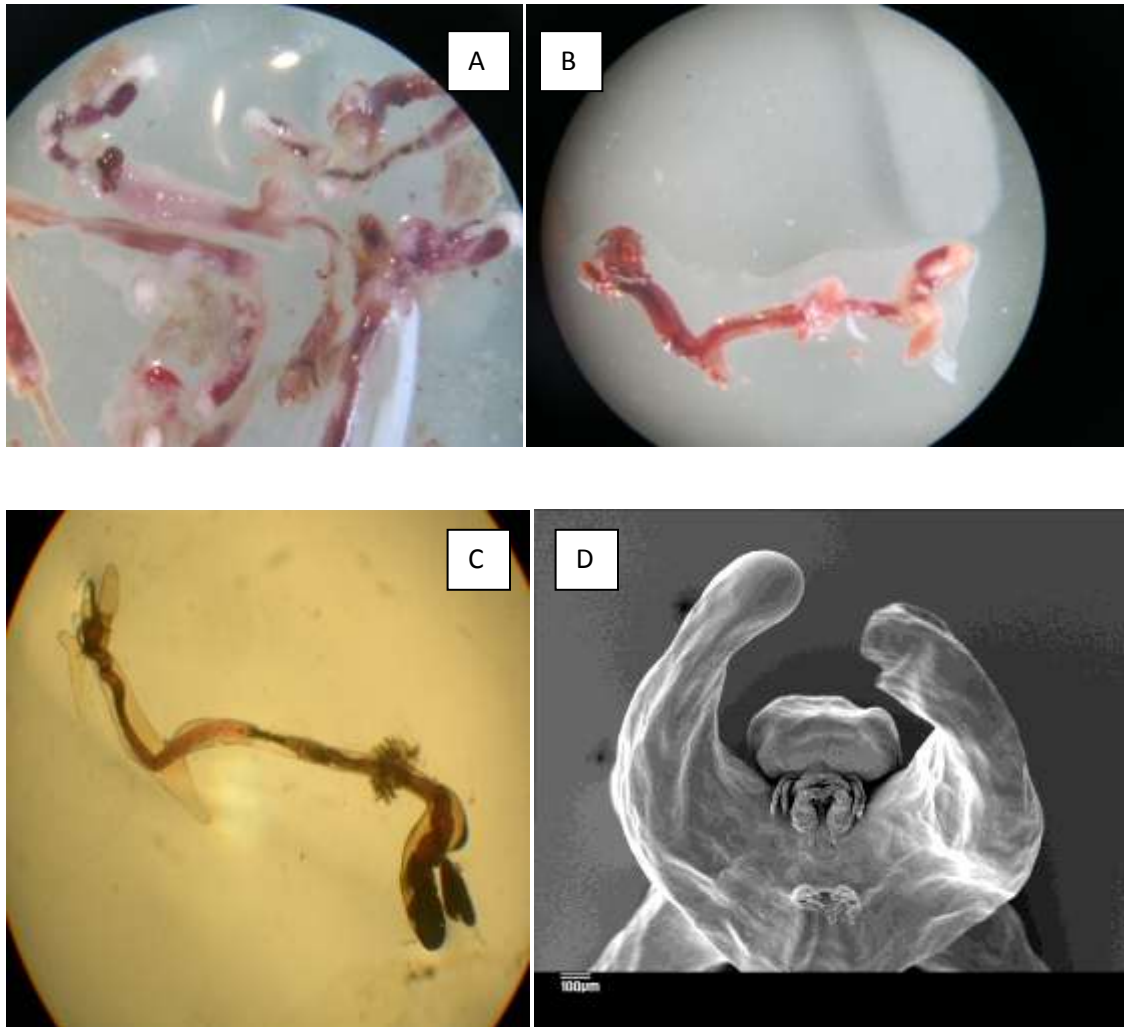


Figure 2.14: Light micrographs (A-C) of live specimen of *Opistholernaea* sp. found on fish in Botswana, D Scanning electron micrograph of head of *Opistholernaea* sp. Adapted from the Department of Aquatic Ecology, University of the Free State, South Africa.(Magnification A-C 10x; D 100x)

Dolops ranarum

Only one species of *Dolops* is present in Africa, it differs from *Argulus* in having the second maxilla armed with a hook rather than a sucker (Paperna, 1996).

Dolops ranarum (Fig. 2.15 A&B) are parasitic throughout its life, but leave the host to lay eggs, and during this process will also change hosts. Both males and females may survive free living for as long as 15 days (Hoffman, 1977).

Dolops ranarum have a preference for smooth-skinned fish the same species may, however, infect buckle and opercula mucosal integuments of scaly fish, notably cichlids (Fryer, 1968). These opportunistic argufies spread all over the body or the skin in smooth skinned hosts, while in scaled fish they occur only on the buckle and bronchial cavity mucosa (Avenant & Van As, 1985).

General characteristics of *Dolops ranarum*

The body consists of three regions:

- The head is un-segmented and bears two pairs of antennae, which puncture the skin of a fish for attachment.
- A pair of variable sized eyes and a mouth,
- The maxillae covers all other mouth parts,
- Average body length of 6 - 10 mm

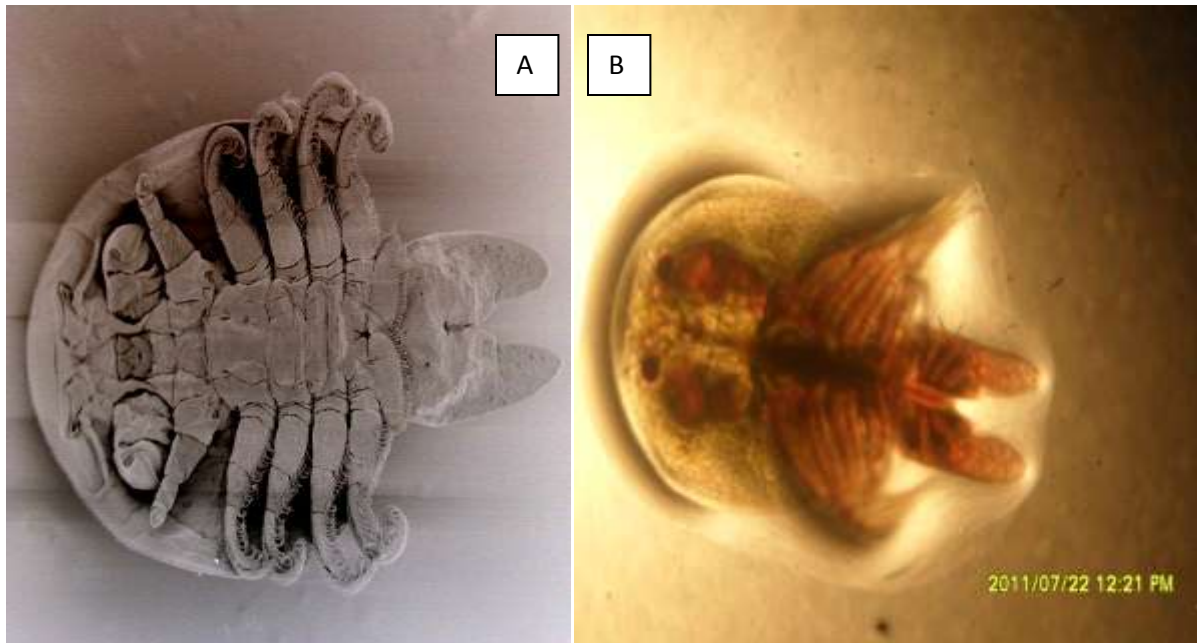


Figure 2.14: A: Scanning electron micrograph of *Dolops ranarum* found on fish in Botswana. Adapted from the Department of Aquatic Ecology, University of the Free State, South Africa). B: Light micrograph of live specimen of *Dolops ranarum* (Magnification A&B 10x)

CHAPTER THREE

METHODOLOGY

3.1 Study area

Sample collection

Monthly surveys were conducted between July 2011 and June 2012, with a total of 12 surveys. The entire Kavango River from Katwitwi through to Kwetze including the seven main sampling sites (Fig. 2.1) was judgmentally sampled.

Kavango River

The Kavango River located in the Kavango region is situated in the North-eastern part of Namibia boarding Botswana and Angola. The region is populated with five ethnic groups: the Kwangali, Mbundza, Sambyu, Gciriku and Mbukushu. Traditional boundaries separate the distinct territories of each group. While related, the languages of the groups have dialectical differences, with Mbukushu being the most distinct (Tvedten *et al.*, 1994).

The region has a variety of vegetation types associated with the drainage systems, including floodplains and open water, riverine forests, and the Omatako drainage system (Obeid & Mendelsohn, 2001). Life in the Kavango region is dependent on the Kavango River. Thus, this river is the central element of the social and cultural identity as it is in other regions with river systems such as the Zambezi region.

The area is characterized by several interconnected perennial river systems and floodplains including the Kavango River (460 km) turning south towards Botswana, where it drains into the swamps of the

Okavango Delta (Hay *et al.*, 2000). The river and its floodplains form the life line of the Kavango region and its people. It is estimated that 90% of the population (approximately 182 000 people) live within 10 km of the river, at a subsistence level. Of these, approximately 32% of the inhabitants are known to catch fish as a source of food and income. Most of these people still use traditional methods such as baskets, funnel traps and fences constructed of plant materials. However, the use of more modern gear such as gill nets, seine net as well as hook and line have been on the increase (Hay *et al.*, 2000).

The river is shared between three countries namely Angola, Botswana and Namibia called Rio Cuban go, Okavango Delta and Kavango River, respectively. The river catchment area covers approximately 88 700 km² with an annual rainfall of 900 mm originating in the central highlands of Angola from where it flows southwards for 600 km before entering Namibia at Katwitwi close to Nkurenkuru (Hay *et al.*, 2000).

It is a very unique river in that it is the only river in Africa that flows eastwards without reaching the ocean. With a distance of more than a thousand kilometres, it drains in the endless sand of the Kalahari basin forming the magical Okavango swamps in Botswana (Hay *et al.*, 2000).

The Kavango River is subject to annual flooding with two main tributaries. The Cuito which is the major tributary entering the Kavango River at Katter, approximately 100 km east from Rundu which nearly doubles the annual flow of the Okavango, thus playing a major role for the fish population downstream. The only southern tributary of any significance is the Omarabad Omatako, which drains the North-Eastern parts of Namibia (Hay *et al.*, 2000).

According to Skelton (2001), the Kavango River with its rich fauna and flora has 86 different fish species. The fish populations in the Kavango River are much smaller compared to many other

freshwater systems and this is primarily because of the low nutrients levels, which would provide food for fish (Mendelsohn & Obeid, 2004).

In less than 30 years, the Okavango River Basin has become a model for successful tourism in countries like Namibia and Botswana. The Okavango Delta has been the main international tourism destination in Botswana since the early 1990's (Maida, 2009). It has an estimate of about 120 000 tourists that visit annually. Tourism in the Delta is based on wildlife tourism which include hunting safaris, photographic safaris, and self drive tourism and fixed lodge tourism.

The tourism sub sector contributes 5 to 9% to Botswana's Gross Domestic Product (GDP) and is rated the second largest economic sector. It is mentioned to be one of the backbone sectors of the economy in Ngami land and Botswana as a whole (Maida, 2009).

Namibian tourism is widely spread in the country, from the ocean, to the desert and the rivers and lakes. The Kavango River has attracted tourists both locally, regionally and internationally not only for the unique lodges and camps that are situated on the river side, but for its diversity in natural resources of the river and the diverse river habitats. Most locals in the Kavango region generate income from river resources and the tourist sector.

The Kavango River system has potentially a high tourism value and it attracts people who treasure wild places and healthy environments. Habitants living close to the Kavango River obtain most resources such as fish, water and building resources from the river, as a source living and employment in the tourist industry (Mendelsohn & Obeid, 2004).

Sampling zones

Data collection was conducted along the Kavango River which is partitioned into four sampling habitat zones by Hocutt *et al.*, (1994) (Fig 3.1). Zone 1, from Katwitwi to Kasogi is characterized by shallow waters with sandy and rocky substrates. Zone 2, which stretches from Kasogi to Mamba is characterized by developed floodplains with large oxbow lakes and backwater habitats. Zone 3, from Mamba to Pope Falls, is characterized by many rapids and a substratum of sand and gravel with large boulders. Zone 4, stretches from Pope Falls to the Namibian/Botswana border and forms the beginning of the Okavango Delta panhandle and features large floodplains.

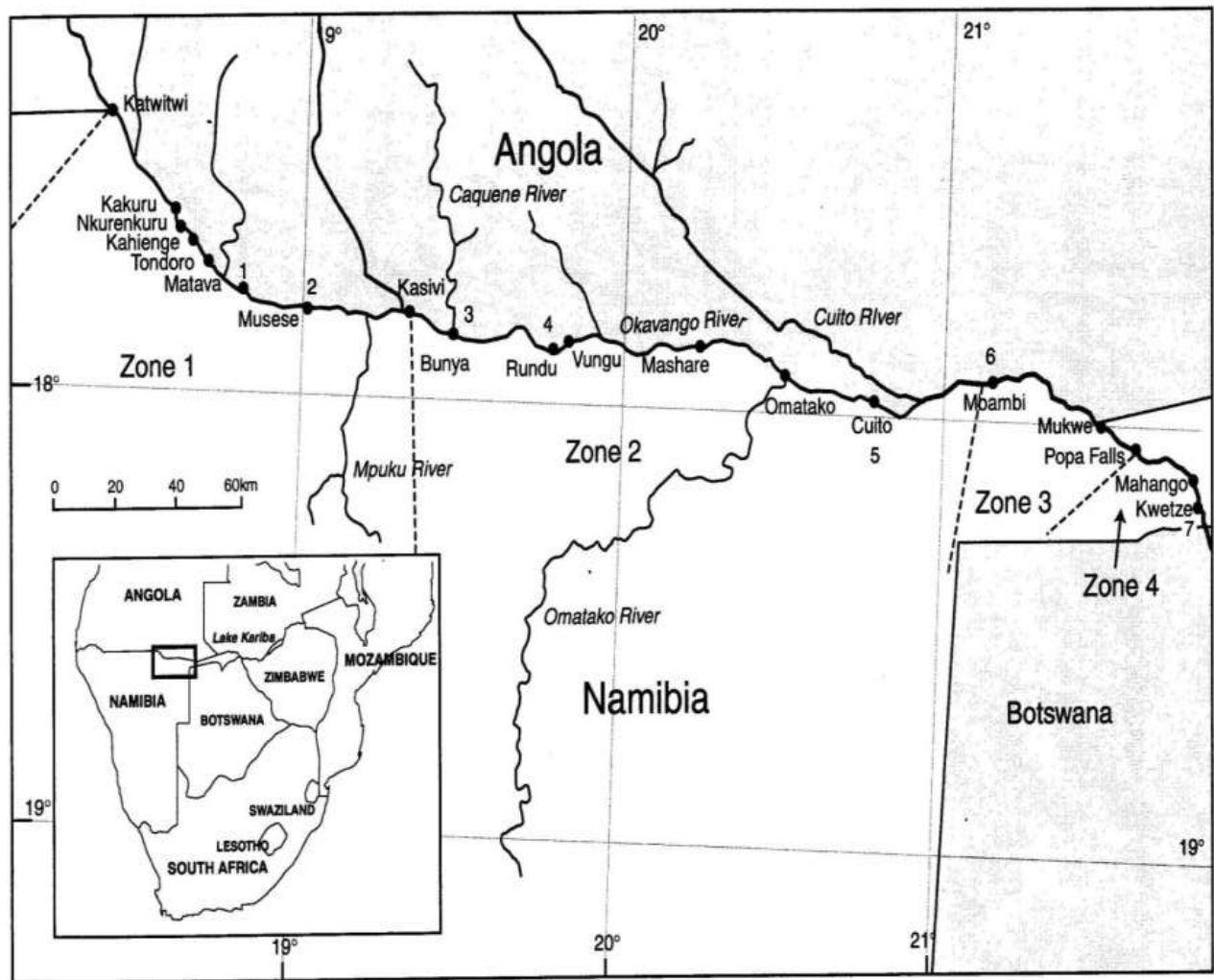


Figure 3.1: The Okavango River in Namibia with the four zones and the main sampling localities numbered 1-7. Adapted from Hocutt *et al.*, (1994).

Seven sampling sites.**Motiva (Fig 3.2)**

It is characterized by a large floodplain area during the flood season and high water which is between December and May. The aquatic vegetation, reeds and grass on the banks form backwater habitats. In this area the water is usually slow flowing especially when water levels are high. The substrate is sandy with little muddy soil present. The channel opens up into the main river where large rock boulders are present.

Vegetation is almost absent in the main stream with only minimum reeds, shrubs and small trees on the river banks.



Figure 3.2: Main stream of the Kavango River (Namibia) and side vegetation at Motiva, in Zone 1. (V. Mumba, 2012).

Muses

This area is mainly characterized by the main stream habitat, with small floodplains present. The substrate in the main stream is sandy with no vegetation. The water is clear with a strong current. The small floodplain has aquatic vegetation with stagnant or slow moving waters with a clay substrate. The river bank has reeds that sometime form shallow isolated pools. In some areas backwaters with reeds are present during the high water season. An agriculture irrigation scheme is nearby with large pumps extracting water near this site.

Bunya (Fig 3.3)

Bunya is characterized by the main stream with clear fast flowing water, sandy substrate with no aquatic vegetation. Reeds are present on the river banks. During the flooding season, stagnant backwaters are present. Rocky areas are also present in Bunya with some reeds between the rocks with no other vegetation.



Figure 3.3: Stagnant backwaters with aquatic vegetation at Bunya along the Kavango River in Namibia. (V. Mumba. 2012)

Rundu

Rundu is the largest established town along the river which is characterized by a large floodplain where the main stream runs along it, with sandy substrate and reeds along the shore. Rundu sampling area has back channels that are characterized by marginal vegetation, stagnant waters and clay substrate. During low water seasons, rocky substrates are present in the main stream.

Cuito (Fig 3.4)

The main stream in this sampling area is characterized by sandy substrate with reeds along the river bank. Large rocks are present in the main stream. There is little or no aquatic vegetation in the deep water, but it is present in the shallow waters. Stagnant backwaters are present with aquatic vegetation.



Figure 3.4: Retrieval of a multifilament net, set at the backwaters in Cuito on the Kavango River in Namibia. (V. Mumba, 2012).

Mamba

The main stream at Mamba has a sandy substrate with minimum vegetation of mainly grass and reeds. Rocks are present in the main stream. The floodplain has stagnant waters with grass and aquatic vegetation with a clay substrate. Reeds are present in the backwaters.

Kwetze

Kwetze is in a reserved area situated in the Bwabwata/Mohembo National Park. The mainstream has clear flowing water with a sandy substrate, with reeds along the shoreline. There are stagnant backwaters with reeds along the shore. Aquatic plants are present in the backwater.

3.2. Sampling methods

Sampling gear used

Fish were collected in the zones mentioned above using a number of different methods. The different sampling methods were determined by the habitat in each sampling zone. Fish were collected using cast nets, seine nets (Fig 3.5A), multifilament gillnets (Fig 3.5B) and scoop nets. Due to lack of man power during some sampling months, fishermen were hired to collect fish by hook and line.

Table 3.1. Summary of sampling gears used during the study period.

Gear used	Description
Cast nets	A two meter cast net with 22mm also known as a throw net was mainly used in shallow areas with a sandy substrate such as areas in Zone 1. Target fish were mainly juveniles
Seine net	The drag net used was a 20 meter net with 22 and 28 mm mesh sizes to capture juveniles as well as medium sized fish. This net was used in shallow water habitats and floodplains mostly in zones 2 and 3.
Gillnet	Two multifilament nets were used with different mesh size, 118 and 150 mm (stretched mesh). These gear types targeted mainly large fish and were set in backwater habitats.
Scoop net	Scoop nets with 16 mm mesh size were used mainly when collecting juveniles or small sized fish in floodplains or in the main stream along the river edge/shoreline.
Hook and line	Hook and line was used for capturing large fish species in the main stream, as most of the nets used were for juveniles and small sized fish.

Source: (V. Mumba 2013).

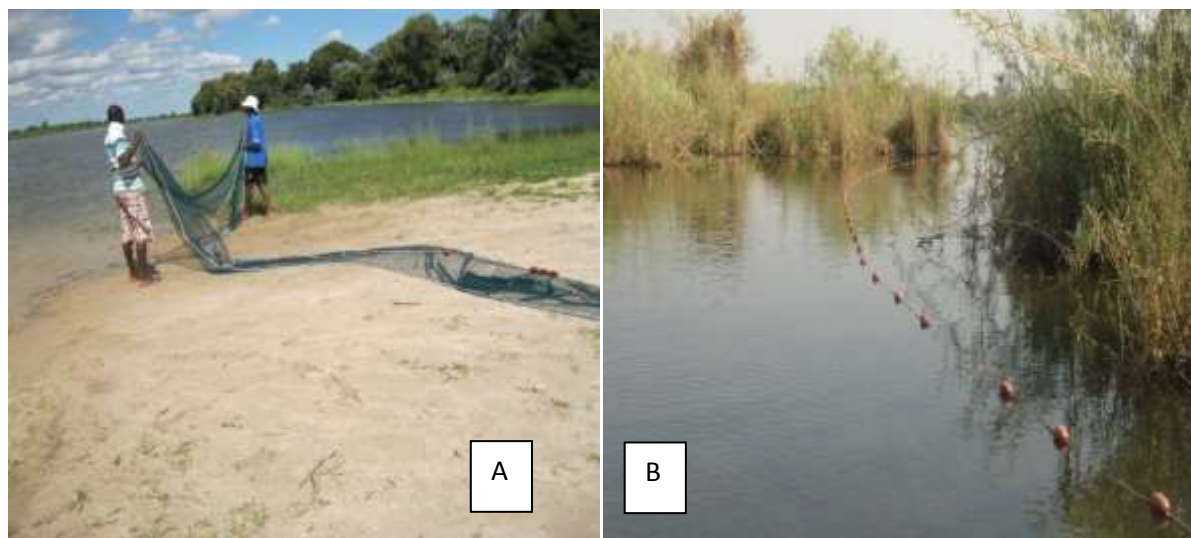


Figure 3.5: Seine net (A) and multifilament net (B) used during the present study to capture target host fish and snails along river banks of the Kavango River, Namibia. (V. Mumba, 2012).

3.3. Research Design

Since the population of target fish species are unknown in the Kavango River system, the population size was thus calculated as infinite therefore, the sampling population size for the present study was a minimum of 35 individuals of targeted fish species for each zone.

Data recording

Fish samples that were analysed were first recorded on a data sheet that documented the area of fish collection, date, fish host, water temperature, water pH, (Fig. 3.6) gear used and the type of parasites found as well as indicating the level of infestation for the number of parasites per host.



Figure 3.6: Water temperature and pH recorded in the field during present study along the Kavango River, Namibia. (V. Mumba, 2012).

The levels of infestation for *Trichodina* sp. and *Tripartiella* sp. (Protozoans) were recorded as shown in Table 3.1. The X (used as a representation of the infestation level, after a skin or gill smear was made on a microscope slide) in the table below was used as an index of infestation.

Table 3.1. Number of protozoans present on examined fish.

Number of protozoan's spotted per smear made on a microscope slide.	Index of infestation
0	Clean
1-5	X
6 - 10	XX
11 - 20	XXX
21-50	XXXX
>50	XXXXX

Source: (V. Mumba, 2012).

Some Protozoan parasites infestation levels were quantified using an infestation index (Table 3.1) as some fish host had high infection level of mobilise and sessiline peritrichs, and due to their size (only a few μm in diameter) it was impossible to count them individually.



Figure 3.7: Microscopes used during present study to examine possible fish parasites on target host fish species. (V. Mumba, 2012).

Fish sample examination

For an accurate prevalence and infestation level, fish were examined as soon as possible after being captured. Live fish specimens were transported to a mobile laboratory, in 5 litre buckets containing river water that was aerated using a battery- powered air pump to keep the fish alive.

Fish were identified to species level with the aid of Skelton (2001) and total length (TL) measured for comparison of parasite infestation in relation to length and size of fish. This was done for adult and juvenile fish. Fish were dissected and examined for potential parasite infestations. Gill arches (Fig 3.8) infested with *Trichodina* and monogeneans were fixed in 10 % formalin. Endo-parasites such as the helminths found were fixed by pouring warm water over them to relax them after which they were transferred to 70% ethanol. Each preserved specimen from the gut was stored in a specimen bottle for further analysis to identify to species.



Figure 3.8: Gill arches of a fish on a Petri dish before examination for possible parasites.

Preservation of parasites

Preserved parasites were further analysed and identified to species level of parasites at the University of Free State (UFS) and Kamutjonga Inland Fisheries Institute (KIFI) laboratories. Dried gill and skin smear slides were further analysed using standard silver impregnating staining techniques to study the internal features such as the identical ring of trichodinids while Harris Haematoxylin staining was used to identify sessile ciliophorans and to study the nuclear apparatus.

3.4. Staining techniques

Silvers staining technique

The techniques used were followed as instructed by (Professor L Basson, personal. Communication, 2012). Two staining methods trails were used for the silver nitrate. The dry smears were placed in mixed 2% silver nitrate for 10 minutes; rinsed in distilled water, transferred to a white slide try in doubled distilled water and placed under a black light for 15 minutes for impregnation. This method was not very successful, as the slides were not successfully impregnated. In a second method, the dried smears were placed in 2% silver nitrate for 15 minutes, rinsed in distilled water and placed under a UV light for 45 minutes. This impregnation method proved to be more successful. Finally the slides were mounted using Eukitt mounting medium and further analysed under a compound microscope for identification to species level

Harris Haematoxylin staining technique

Dried gills and skin smear with sessile ciliates were stained using Harris Haematoxylin. This staining is used to identify internal organs such as the nuclear apparatus.

Dried slides smears were placed in Haematoxylin for 30 minutes rinsed thoroughly with tap water and transferred to Scott's solution for 3 – 5 minutes and rinsed again with tap water. Slides were dehydrated through ethanol concentrations for one minute each. After drying the slides in 100% ethanol they were transferred to Xylene for 3 minutes. Finally the slides were mounted using Eukitt, and examined under a compound microscope.



Figure 3.9: University of Free State laboratory preparing the Harris Haematoxylin staining technique. (V. Mumba, 2012)

3.5. Statistical analysis

Statistical analysis

Data were analysed using Sigma Plot version 11.0. To determine the difference in number of fish parasite infestation in the four zones, the host fish species and the four fish size groups. A non parametric test, Kruskal-Wallis One Way was performed to test the significant differences in the values using Analysis of Variance (ANOVA).

Prevalence (%) and abundance of fish parasites on fish host species

Statistical data for the different parasites found was calculated according to Margolis *et al.*, (1982) for prevalence and relative abundance and are presented in Tables 4.2 and 4.3.

Prevalence (% of infestation)

$$\text{Prevalence (\%)} = \frac{\text{Number of fish infested with parasites}}{\text{Total number of fish examined}} \times 100$$

Relative Abundance

$$\text{Abundance of fish parasite} = \frac{\text{Total number of individual parasite in a sample of hosts}}{\text{Total number of individuals of the host species}}$$

CHAPTER FOUR

RESULTS

4.1. Temperature and pH

Water parameters were analyzed in each zone where host fish species were collected. Water temperature along the Kavango River did not show much variation as well as pH as shown in Table 4.1.

Table 4.1: Mean Temperature and pH values of the Kavango River, Namibia.

Zones	Mean temperature	Mean pH
One	$26.88 \pm 0.05^{\circ}\text{C}$	7.23 ± 0.01
Two	$27.03 \pm 0.06^{\circ}\text{C}$	7.25 ± 0.01
Three	$27.46 \pm 0.06^{\circ}\text{C}$	7.26 ± 0.01
Four	$27.00 \pm 0.05^{\circ}\text{C}$	7.28 ± 0.01

In Table 4.1 the average temperature and pH in the Kavango River are presented. On average the temperature ranged between 26°C to 29.5°C , whilst pH ranged between 6.92 to 7.90 therefore the presence of parasite infestation on target fish species are not contributed by water temperature or pH.

Table 4.2: Prevalence (%) level of fish parasites found on host fish species found in the Kavango River

Parasites species	<i>Tilapia rendalli</i>	<i>Oreochromis andersonii</i>	<i>Serranochromis robustus</i>
<i>Dactylogyrus</i> sp. (Monogenea)	11	23	0
<i>Proteocephalus</i> sp. (Cestoda)	0	1	1
<i>Contracecum</i> sp. (Nematoda)	4	4	4
<i>Clinostmum</i> sp. (Trematoda)	1	1	7
<i>Acanthocephala</i> sp. (Spiny headed worm)	0	1	0
<i>Epistylis</i> sp. (Ciliophora)	4	3	0
<i>Tripartiella</i> sp. (Ciliophora)	7	3	0
<i>Trichodina</i> sp. (Ciliophora)	10	26	0
<i>Lamproglena monodi</i> (Copepoda)	4	0	0
<i>Opistholernaea</i> sp. (Copepoda)	3	0	1
<i>Lernaea hardingi</i> (Copepoda)	0	1	0
<i>Dolops ranarum</i> (Branchiura)	1	0	0

Table 4.3: Relative abundance of fish parasites found on fish hosts along the Kavango River, Namibia

Fish Parasites	Abundance
<i>Dactylogyrus</i> sp. (Monogenea)	0.17
<i>Proteocephalus</i> sp. (Cestoda)	0.01
<i>Contracecum</i> sp. (Nematoda)	0.06
<i>Clinostmum</i> sp. (Trematoda)	0.04
<i>Acanthocephala</i> sp.(Spiny headed worm)	0.01
<i>Epistylis</i> sp. (Ciliophora)	0.04
<i>Tripartiella</i> sp. (Ciliophora)	0.06
<i>Trichodina</i> sp. (Ciliophora)	0.18
<i>Lamproglena monodi</i> (Copepoda)	0.02
<i>Opistholernaea</i> sp. (Copepoda)	0.02
<i>Lernaea hardingi</i> (Copepoda)	0.01
<i>Dolops ranarum</i> (Branchiura)	0.01

Table 4.4: Total number of parasite specimens (excluding Protozoa) collected on the three target species along the Kavango River, Namibia.

Fish Parasites	<i>Tilapia rendalli</i>	<i>Oreochromis andersonii</i>	<i>Serranochromis robustus</i>
<i>Dactylogyrus</i> sp. (Monogenea)	42	88	0
<i>Proteocephalus</i> sp. (Cestoda)	0	2	3
<i>Contracecum</i> sp. (Nematoda)	23	2	26
<i>Clinostomum</i> sp. (Trematoda)	2	1	31
<i>Acanthocephala</i> sp. (Spiny headed worm)	0	1	0
<i>Lamproglana monodi</i> (Copepoda)	7	0	0
<i>Opistholernaea</i> sp. (Copepoda)	16	0	7
<i>Lernaea hardingi</i> (Copepoda)	0	2	0
<i>Dolops ranarum</i> (Branchiura)	1	0	0

4.2. Target fish species (Sampling Zone/target fish species size)

The three target fish species in the present study were categorized in four size groups and analyzed for each zone. Each graph represents individual fish species collected in each zone and are grouped in 4 different size categories as show in Table 4.5.

Table 4.5: Frequency data of host fish species infested with parasites, prevalence (%) of parasite and host fish size (in cm) category of fish examined during study in the Kavango River, Namibia.

Fish size category, in cm (TL)	5 – 10.9	11-20.9	21 – 30.9	≥31
Number of fish species examined	51	93	54	7
Number of fish species infested with parasites	23	58	21	1
Prevalence (%)	45%	62%	38%	14%

Highest number of fish examined were those from the categories 11 – 20.9 and 21 – 20.9, which were fish ranging from developing stage into adults (Table 4.5). The reason for a high number of host fish in this category was the gear that was mostly used i.e. multifilament nets that ranged from mesh size 28 to 150mm.

In total 205 individual fish were examined from four demarcated zones along the Kavango River (see Tables 4.6 and 4.7). One hundred and two fish were infested or infected with either one or more parasites on 1 host, which means that 103 fishes examined were not infected with any parasites.

Table 4.6: Total number of fish examined and infested with parasites from the Kavango River, Namibia

Fish host	No. of fish examined	No. of fish infested
<i>Tilapia rendalli</i>	91	39
<i>Oreochromis andersonii</i>	89	49
<i>Serranochromis robustus</i>	25	14
Total	205	102

Table 4.7: Number of infested and not infested of the three host fish species examined from the four zones along the Kavango River, Namibia.

	<i>Tilapia rendalli</i>		<i>Oreochromis andersonii</i>		<i>Serranochromis robustus</i>	
	Infested	Not infested	Infested	Not infested	Infested	Not infested
Zone 1	17	14	12	10	4	3
Zone 2	11	11	11	8	2	3
Zone 3	4	18	14	9	2	4
Zone 4	7	9	12	13	5	2
Total	39	52	49	40	13	12

From the results, Table 4.6 and 4.7 indicate the total number of fish species (*Tilapia rendalli*, *Oreochromis andersonii* and *Serranochromis robustus*) that were examined and those found infested with parasites. In the case of *T. rendalli* and *O. andersonii* the highest numbers of fish species were recorded and this was due to the methods and gears used for fish collection and accessibility of the river in the various habitats.

Parasite infestation on *Oreochromis andersonii* in Zone 1 of the Kavango River, Namibia.

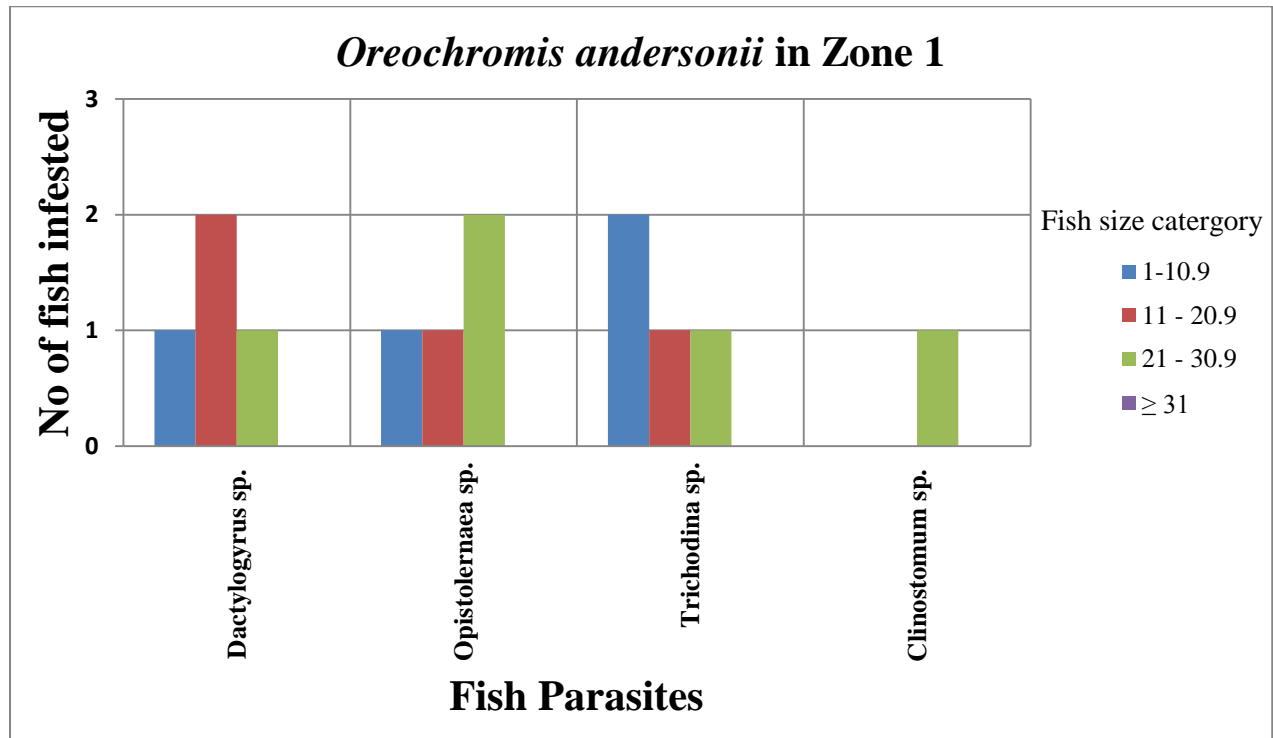


Figure 4.1: Histogram indicating the different parasites that were found from the various sizes of *Oreochromis andersonii* n= 22 that were examined from Zone 1 along the Kavango River, Namibia. (Take note that some individual fish had more than 1 parasite, thus showing more parasites in the figure than indicated in the Table 4.7 of the fish infested).

Parasite infestation on *Oreochromis andersonii* in Zone 2 of the Kavango River, Namibia.

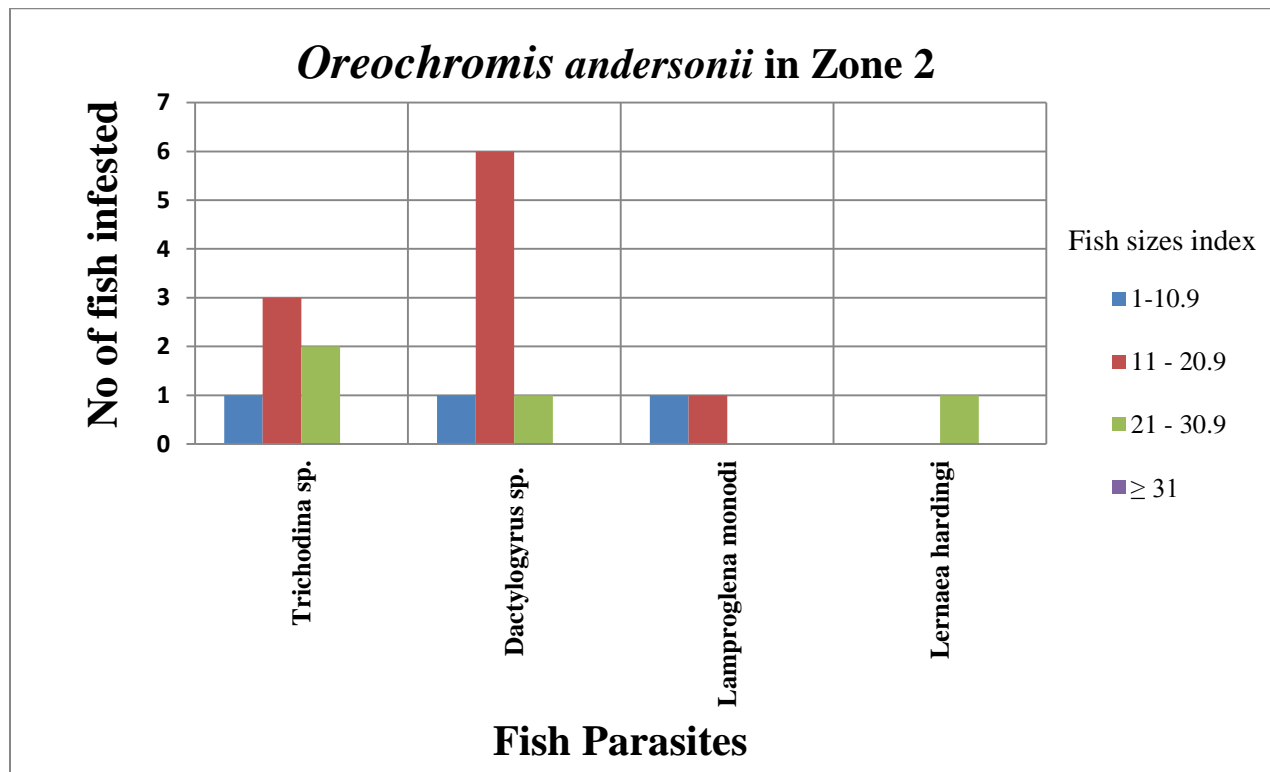


Figure 4.2: Histogram indicating the different parasites that were found from the various sizes of *Oreochromis andersonii* n = 19 that were examined from Zone 2, along the Kavango, River, Namibia. (Take note that some individual fish had more than 1 parasite, thus showing more parasites in the figure than indicated in the Table 4.7 of the fish infested).

Parasite infestation on *Oreochromis andersonii* in Zone 3 of the Kavango River, Namibia.

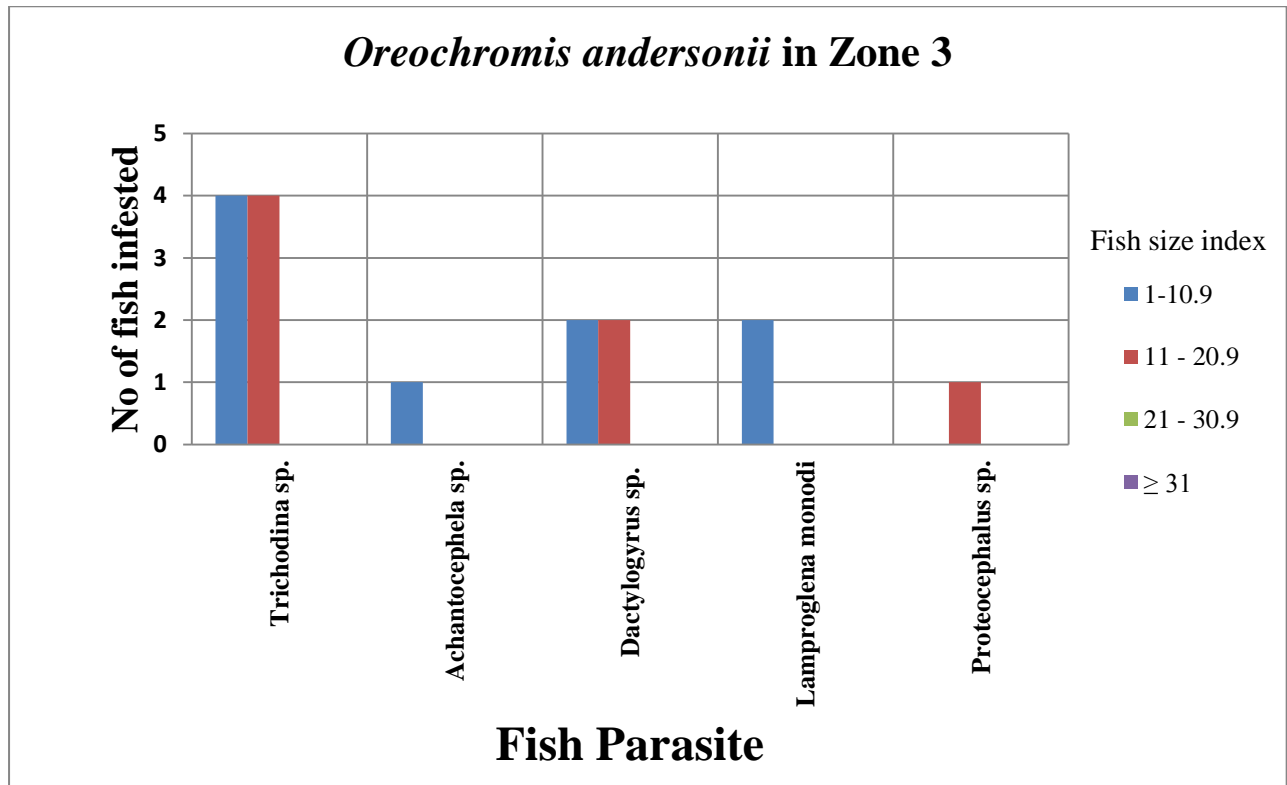


Figure 4.3: Histogram indicating the different parasites that were found from the various sizes of *Oreochromis andersonii* n = 23 that were examined from Zone 3, along the Kavango River, Namibia. (Take note that some individual fish had more than 1 parasite, thus showing more parasites in the figure than indicated in the Table 4.7 of the fish infested).

Parasite infestation on *Oreochromis andersonii* in Zone 4 of the Kavango River, Namibia.

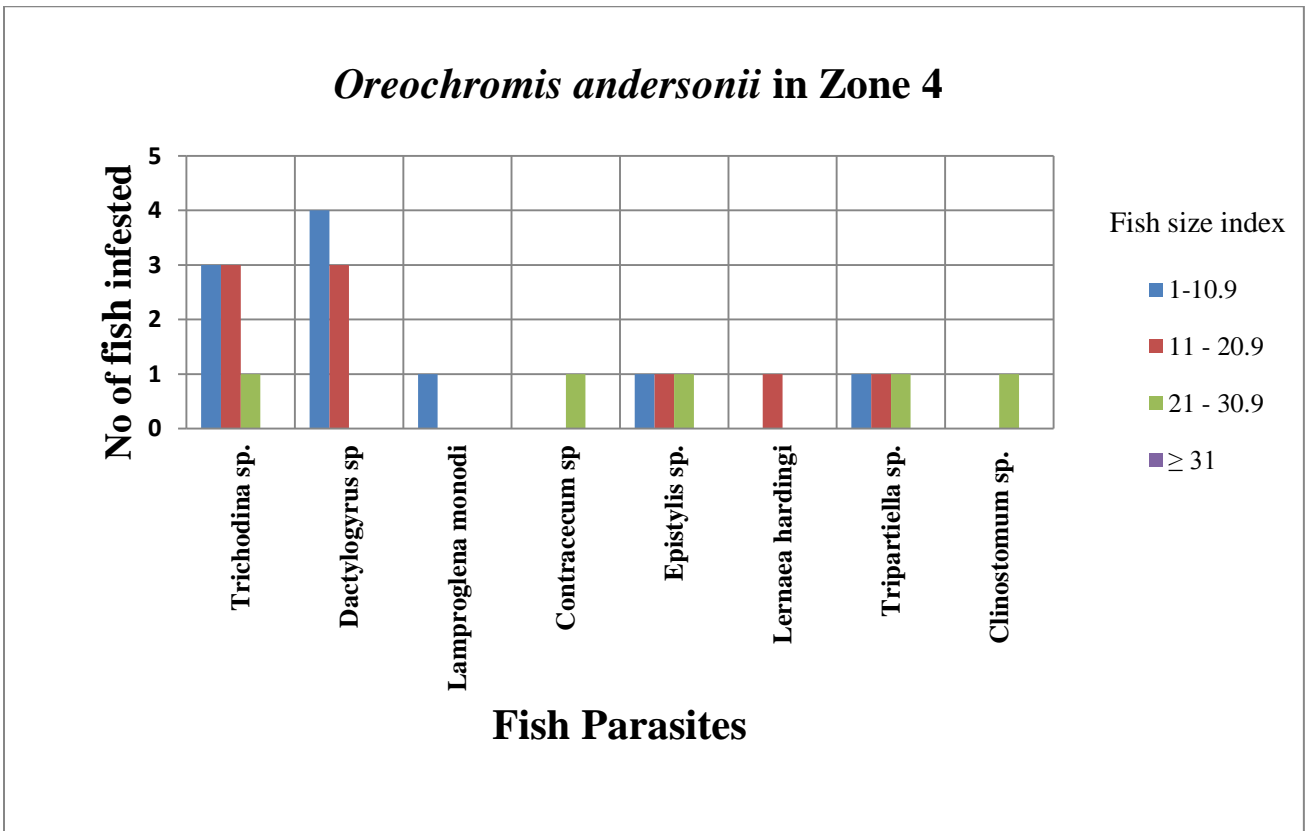


Figure 4.4: Histogram indicating the different parasites that were found from the various sizes of *Oreochromis andersonii* n = 25 that were examined from Zone 4, along the Kavango River, Namibia. (Take note that some individual fish had more than 1 parasite, thus showing more parasites in the figure than indicated in the Table 4.7 of the fish infested).

Parasite infestation on *Serranochromis robustus* in Zone 1 of the Kavango River, Namibia.

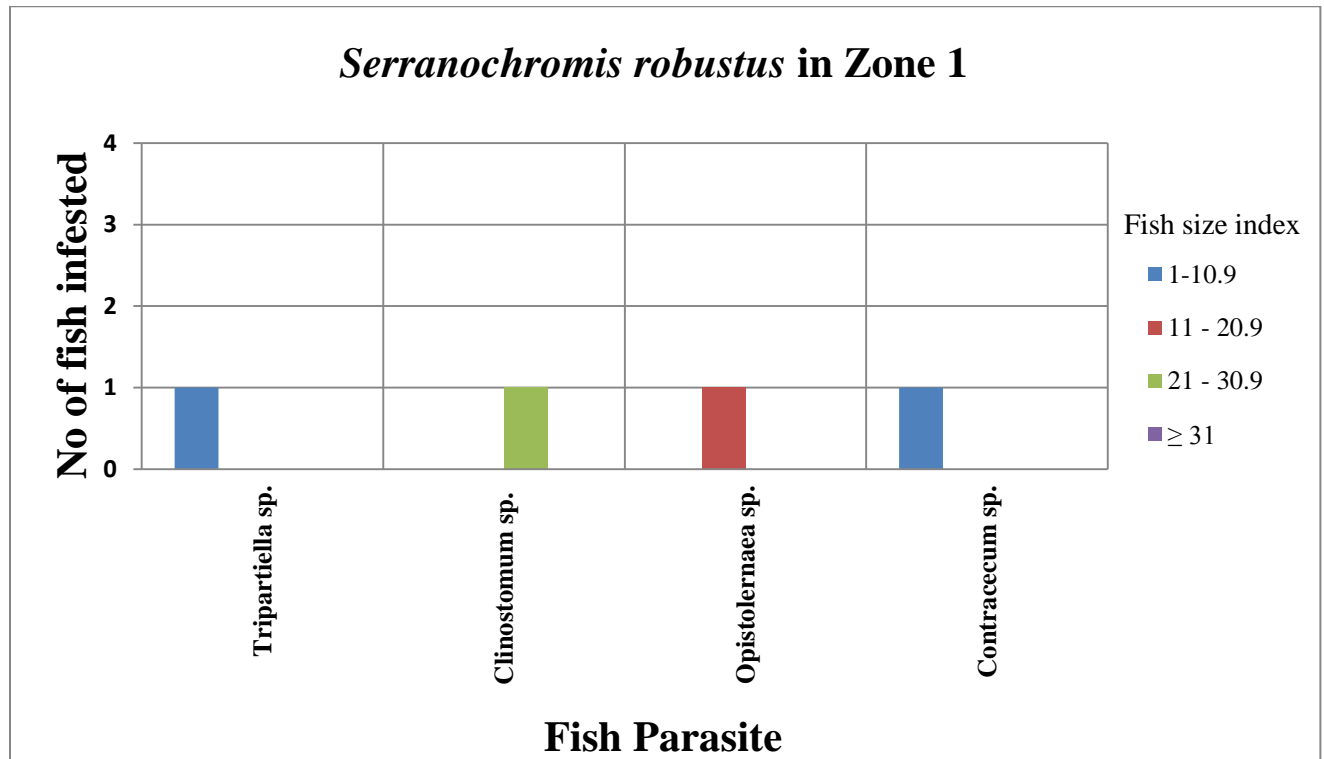


Figure 4.5: Histogram indicating the different parasites that were found from the various sizes of *Serranochromis robustus* $n = 6$ that were examined from Zone 4, along the Kavango River, Namibia. (Take note that some individual fish had more than 1 parasite, thus showing more parasites in the figure than indicated in the Table 4.7 of the fish infested).

Parasite infestation on *Serranochromis robustus* in Zone 2 of the Kavango River, Namibia

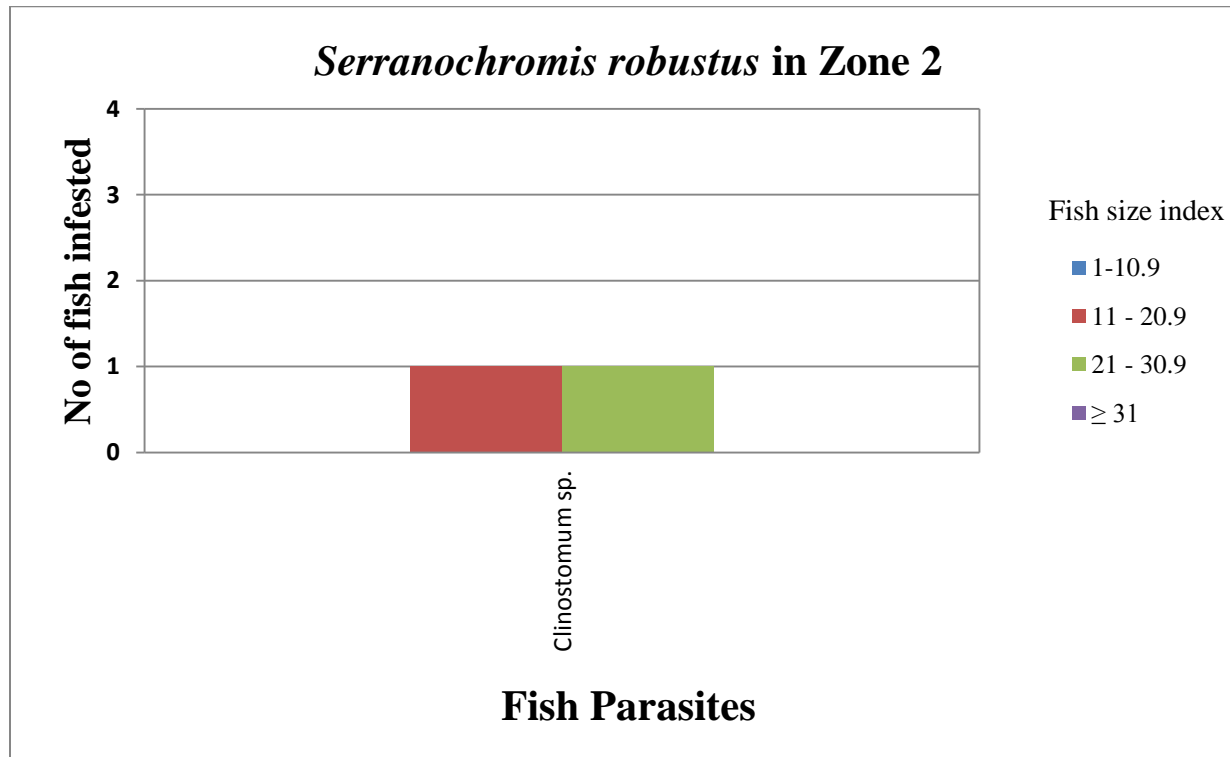


Figure 4.6: Histogram indicating the different parasites that were found from the various sizes of *Serranochromis robustus* n = 5 that were examined from Zone 4, along the Kavango River, Namibia.

Parasite infestation on *Serranochromis robustus* in Zone 3 of the Kavango River, Namibia.

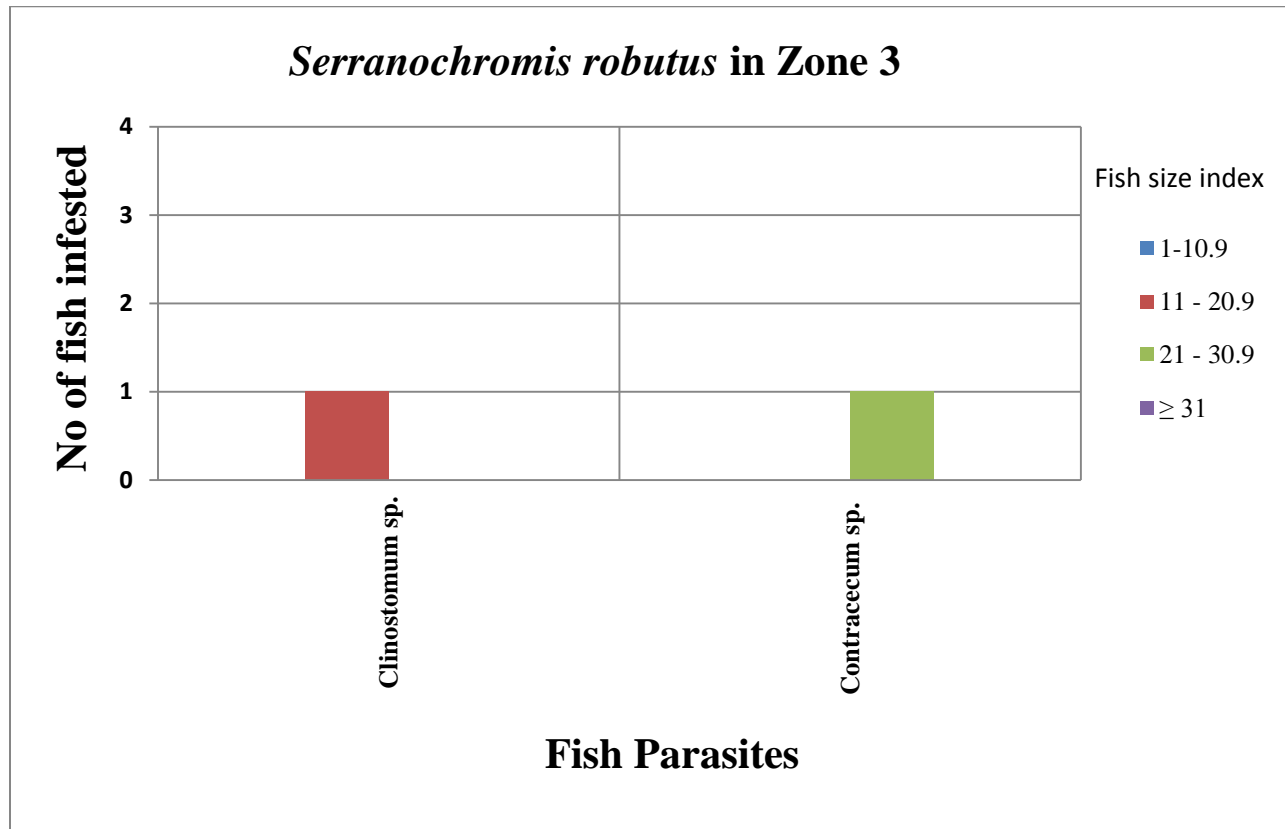


Figure 4.7: Histogram indicating the different parasites that were found from the various sizes of *Serranochromis robustus* n = 6 that were examined from Zone 4, along the Kavango River, Namibia.

Parasite infestation on *Serranochromis robustus* in Zone 4 of the Kavango River, Namibia.

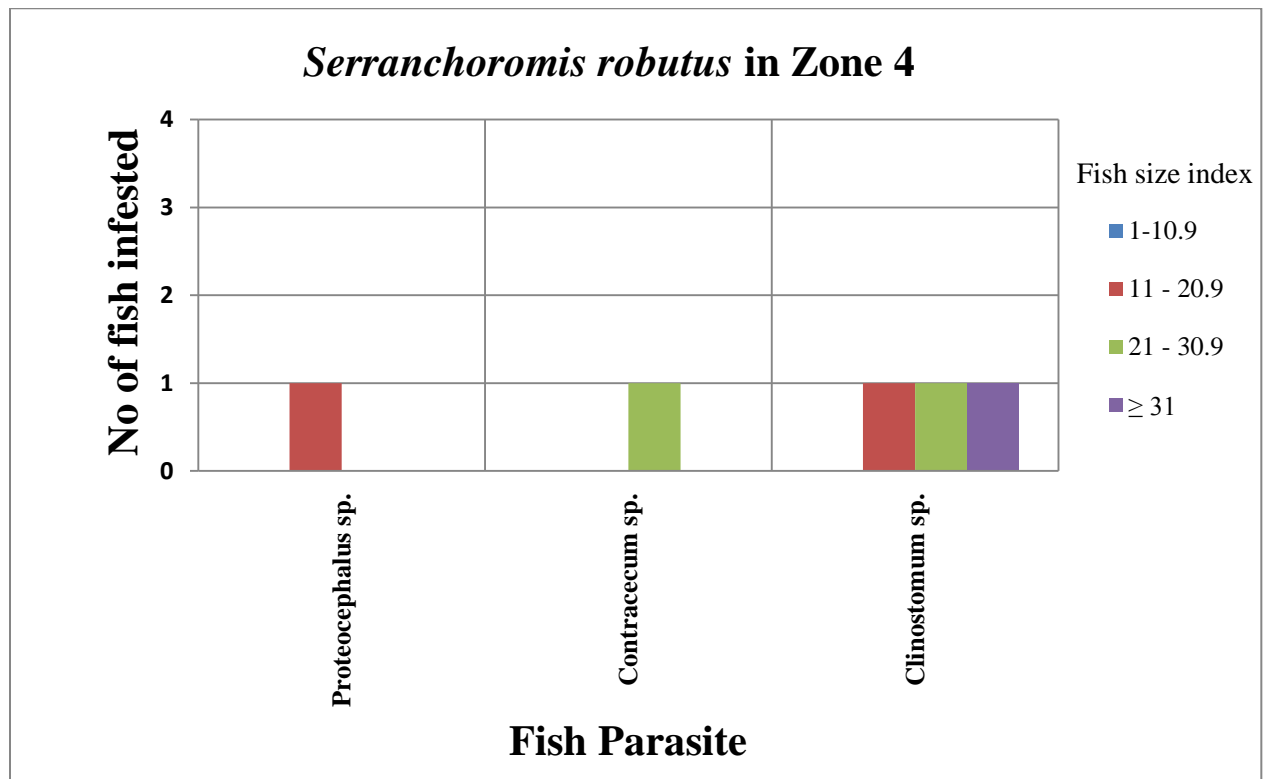


Figure 4.8: Histogram indicating the different parasites that were found from the various sizes of *Serranochromis robustus* $n = 8$ that were examined from Zone 4, along the Kavango River, Namibia. (Take note that some individual fish had more than 1 parasite, thus showing more parasites in the figure than indicated in the Table 4.7 of the fish infested.

Parasite infestation on *Tilapia rendalli* in Zone 1 of the Kavango River, Namibia.

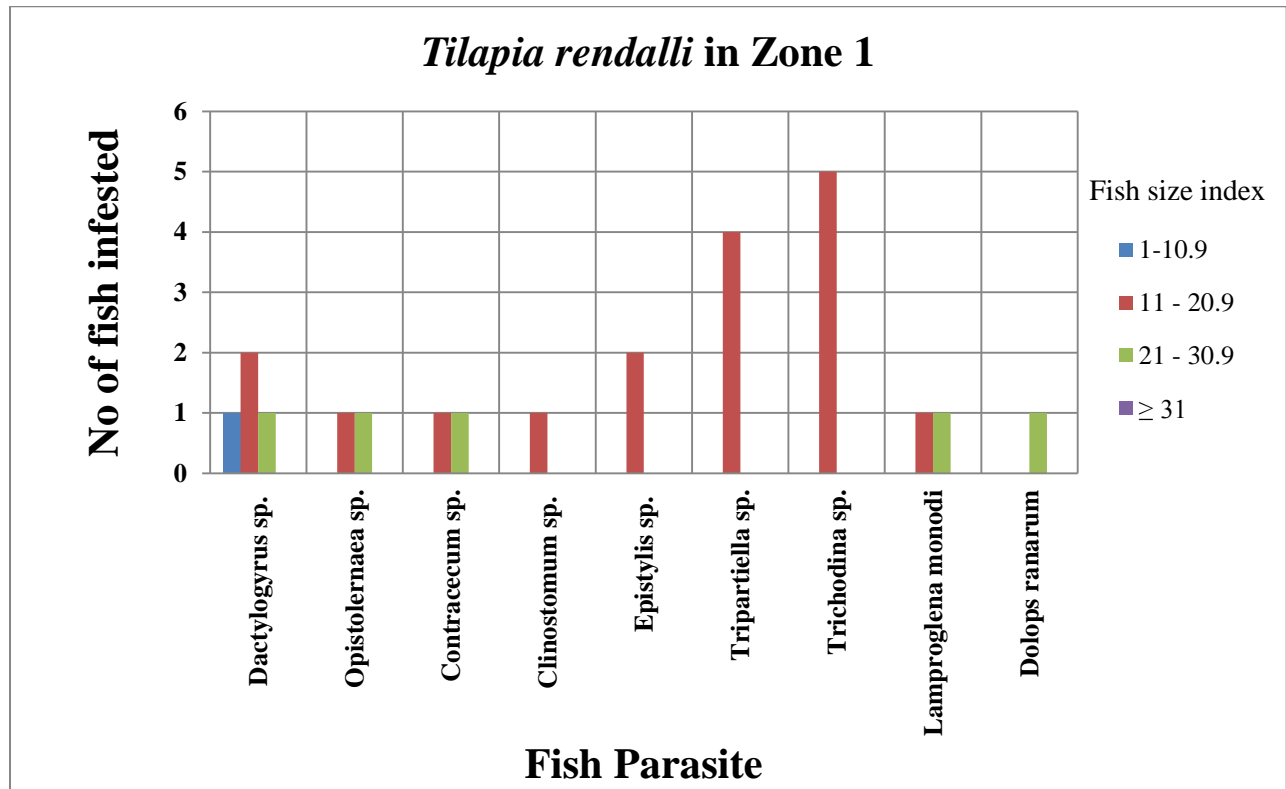


Figure 4.9: Histogram indicating the different parasites that were found from the various sizes of *Tilapia rendalli* n = 31 that were examined from Zone 4, along the Kavango River, Namibia.

(Take note that some individual fish had more than 1 parasite, thus showing more parasites in the figure than indicated in the Table 4.7 of the fish infested).

Parasite infestation on *Tilapia rendalli* in Zone 2 of the Kavango River, Namibia

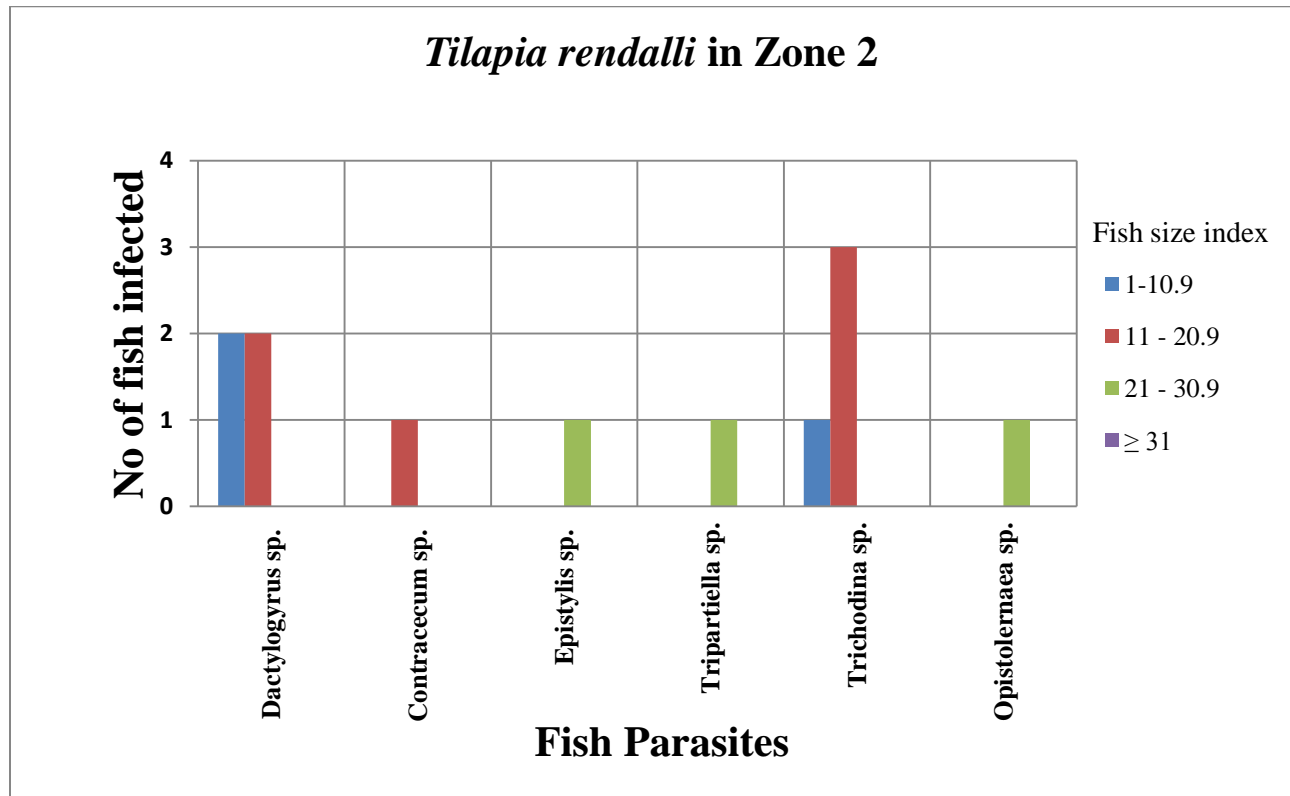


Figure 4.10: Histogram indicating the different parasites that were found from the various sizes of *Tilapia rendalli* n = 22 that were examined from Zone 4, along the Kavango River. (Take note that some individual fish had more than 1 parasite, thus showing more parasites in the figure than indicated in the Table 4.7 of the fish infested).

Parasite infestation on *Tilapia rendalli* in Zone 3 of the Kavango River, Namibia.

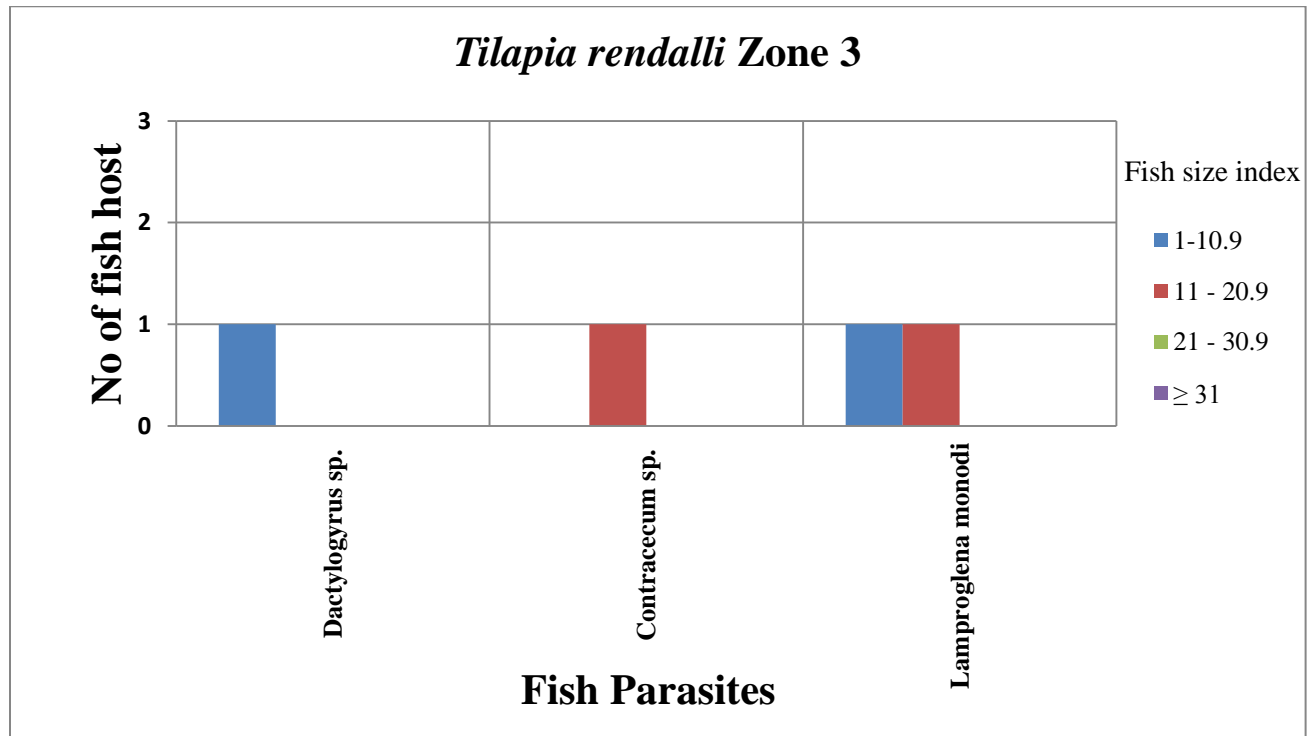


Figure 4.11: Histogram indicating the different parasites that were found from the various sizes of *Tilapia rendalli* n = 22 that were examined from Zone 4, along the Kavango River, Namibia.

Parasite infestation on *Tilapia rendalli* in Zone 4 of the Kavango River, Namibia.

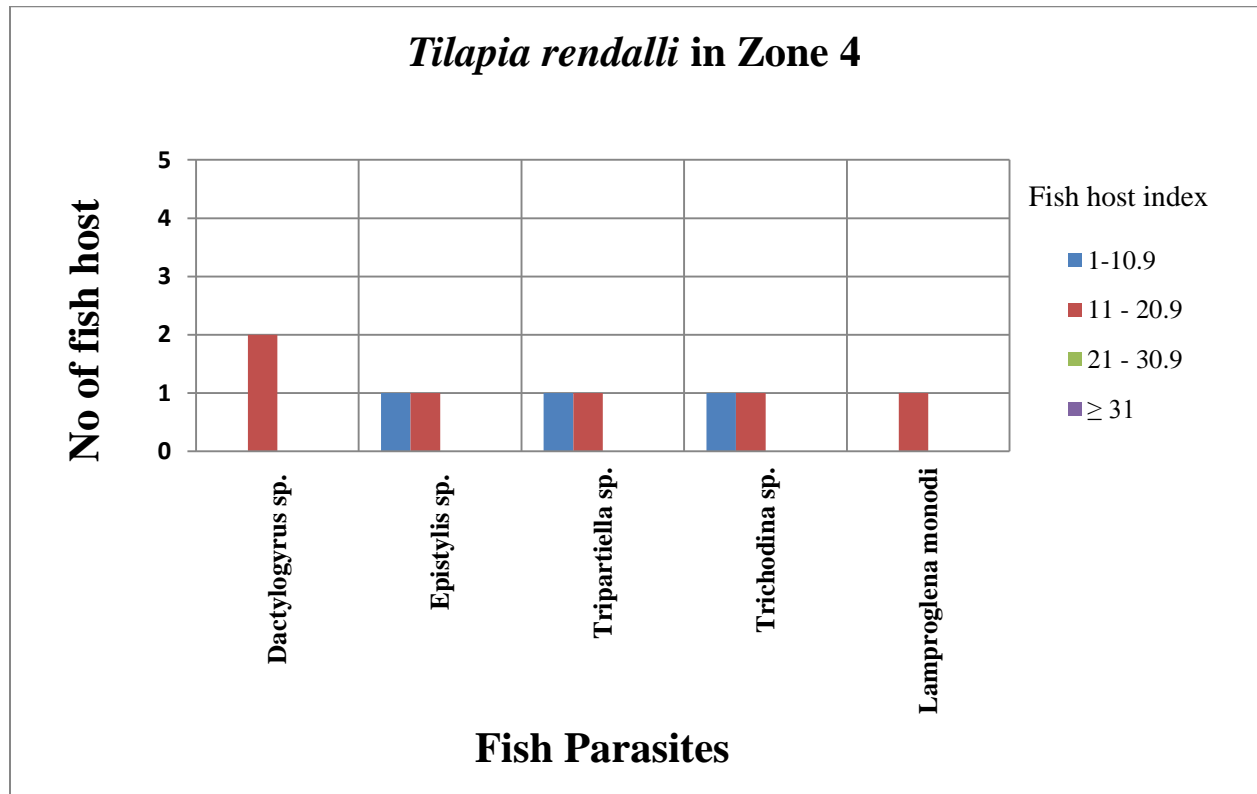


Figure 4.12: Histogram indicating the different parasites that were found from the various sizes of *Tilapia rendalli* n = 16 that were examined from Zone 4, along the Kavango River. (Take note that some individual fish had more than 1 parasite, thus showing more parasites in the figure than indicated in the Table 4.7 of the fish infested).

The different parasites that were found from the fish host species i.e. (Table 4.4) *O. andersonii* (Figs. 4.1-4.4), *S. robustus* (Figs. 4.5-4.8) and *T. rendalli* (Figs. 4.9-4.12), were analysed and are presented in the following histograms above. The fish host species and the parasites collected from each host from the four zones were kept separately. In some cases the number of infected fish illustrates more

parasites per zone, and the reason is that multiple infections of certain parasite groups or taxa were found on the same host.

4.3. Statistical results

Table 4.8: Kruskal-Wallis One Way Analysis of Variance on number of fish infested in the 4 different zones.

Zone	N	Missing	Median	25%	75%
Zone 1	12	0	0,500	0,000	4,000
Zone 2	12	0	1,000	0,000	3,000
Zone 3	12	0	1,000	0,000	2,500
Zone 4	12	0	1,000	0,000	2,000

Data source: (V. Mumba, 2014) $H = 0,0498$ with 3 (df) degrees of freedom ($P = 0,997$).

The differences in the median values among the treatment zones are not great enough to exclude the possibility that the difference is due to judgemental sampling variability; there is not a statistically significant difference ($P = 0,997$).

Table 4.9: Kruskal-Wallis One Way Analysis of Variance on number of fish infested of the 3 fish species.

Fish species	N	Missing	Median	25%	75%
<i>Oreochromis andersonii</i>	12	0	1,000	0,000	6,000
<i>Tilapia rendalli</i>	12	0	0,500	0,000	4,500
<i>Serranochromis robustus</i>	12	0	0,000	0,000	1,500

Data source: (V. Mumba, 2014).

H = 2,433 with 2 (df) degrees of freedom (P = 0,296).

The differences in the median values among the treatment groups are not great enough to exclude the possibility that the difference is due to random sampling variability; there is not a statistically significant difference(P = 0,296).

Table 4.10: Kruskal-Wallis One Way Analysis of Variance on number of fish infested in the 4 different size group of target fish species.

Fish size	N	Missing	Median	25%	75%
5-10.9cm	11	0	0,000	0,000	3,250
11-20.9	12	0	1,500	0,000	8,000
21-30.9	12	0	0,000	0,000	2,000
>31	12	0	0,000	0,000	0,000

Data source: (V. Mumba, 2014).

H = 7,670 with 3 (df) degrees of freedom (P = 0,053).

The differences in the median values among the zones are not great enough to exclude the possibility that the difference is due to judgemental sampling variability; there is not a statistically significant difference ($P = 0,053$).

Statistical analysis

The P values (Tables: 4.8 – 4.10) obtained from the number of fish infested with fish parasites over a period of 12 months in the different zones, fish sizes and fish host species were not significantly different ($H = 0,0498$ with 3 (df) $P = 0,997$), ($H = 2,433$ with 2 (df) $P = 0,296$) and ($H = 7,670$ with 3 (df) $P = 0,053$) respectively. Therefore further statistically analysis was not necessary.

According to the statistical analysis that were performed using the one way ANOVA, we failed to reject H1 as (p value > 0.05), therefore there is no significant difference in fish infested with fish parasites in the four different zones. The H2 failed to reject as there was no significant difference in fish species infested with fish parasites. We failed to reject H3, as there was no significant difference with fish infested with parasites in the four different fish size.

Protozoans

The results of the investigation from the above histograms (Figs. 4.1-4.12) on fish sizes and zones of parasites infestation of each fish species reveal the occurrence of *Dactylogyrus* sp. and *Trichodina* sp. being the highest on *O. andersonii* (Figs. 4.1-4.4) and *T. rendalli* (Figs. 4.9-4.12) in zone 2 and 4, in size group ranging between 1 – 10.9 cm and 11 – 20.9 cm in total length of the fish examined. *Dactylogyrus* sp. (Fig. 2.7 A&B) was found with a prevalence of 34% on *O. andersonii* and *T. rendalli* in all four zones. *Tripartiella* sp. (Fig 2.5C) and *Epistylis* sp. (Figs. 2.6 A&B, 5.2) were also found on

the fish that were examined. The prevalence levels were 10% and 7%, respectively. These two ciliophorans were abundant mostly on *Tilapia rendalli*.

Helminths

Prevalence and abundance level of Helminths found in this study were low, (Tables 4.2 & 4.3). whilst the same low levels were found in *Achantocephala* sp. (Spiny headed worms) (Fig. 2.8 A&B). Nematoda (Roundworms) (Fig. 3.10A-D) were found in moderate numbers although *Contracecum* sp. was found in all three of the target fish species (Figs. 4.4, 4.5, 4.7-4.11 and Table 4.2). *Clinostomum* sp. (Figs. 4.1, 4.4-4.9) was found mostly on *Serranochromis robustus* with a prevalence of 7% (Table 4.2) compared to the other two fish species where it was 1% for both *Proteocephalus* sp. (Fig. 2.9B) was very low in prevalence (see Table 4.2) and that could be because of the intermediate host (free living Copepoda) that are not abundant in the river site where fish were collected

Crustaceans

Crustaceans reported on host fish species were *Lamproglana monodi* (Fig. 2.12), *Dolops ranarum* (Fig. 2.15 A&B), *Opistholernaea* sp. (Fig. 2.14 A-D) and *Lerne hardingi* (Fig. 2.13) with prevalence levels of 4%, 1%, 4% and 1%, respectively (see Table 5.2). The parasites were abundantly found on *T. rendalli* followed by *S. robustus* and *O. andersonii* (Table 4.4).

Table 4.4 shows the high occurrence of *Opistholernaea* sp. on the fish examined. These parasites were mostly found in Zone 1 on *O. andersonii* (Figs 4.1, 4.9 & 4.10). With *Serranochromis robustus* most of the fish were especially infected with Nematodes. These occur in the fish size group 11 – 20.9 cm and 21 – 30.9 cm which are mostly adult fish. They were mostly collected in zone 4, which is a reserved area although few Nematodes occurred in Zone 1 and 3, but in low abundance (Tables 4.6 & 4.7).

CHAPTER FIVE

DISCUSSION

To date, there is no study that has been done to investigate the presence of fish parasite infection on wild fish stocks caused by farmed fish. It is important to take precaution that no water from the fish farms spill back into the river, as very little research has been done to examine the role of aquaculture, in the outbreak of diseases in wild stock. Van As & Basson, (1989) concludes that pathogens can be transferred from a fish that is farmed to wild stock, though research shows that infestation will be very low in prevalence of the pathogen agent.

Water parameters

Various physical freshwater parameters are either a direct or indirect factor contributing to the influence on fish growth and reproduction, but not to the pathological effects (Van As & Basson, 1984). Temperature is one of the most important factors affecting the physiology, growth, reproduction and metabolism of tilapia fish species (El-Sayed, 2006). In the wild it is unfeasible to control water parameters, whilst in culture environments water quality is very likely to be subjected to continuous change, especially in semi-intensive culture systems (Lim & Webster, 2006).

According to Oldewage and Van As (1987), a sudden change in water temperature can cause a prompt increase in the reproduction of invertebrates and most potential pathogens.

Most fish, including cichlids, have an upper and lower thermal tolerance and an optimum temperature requirement for growth, food conversion and resistance to specific diseases (Barson, 2004) including parasitic infections. Freshwater organisms including freshwater fish have an optimal range of pH to survive in, ranging between 6.5 to 8.5 (Van As & Basson, 1984). The same applies to the

developmental phase of freshwater parasites, free-swimming larvae and ectoparasites. However, some freshwater fish parasites can survive low pH values (Bauer, 1962).

For cichlids, mostly tilapia's ideal optimum water temperature ranges from 23°C to 30°C. Temperatures below 18°C and above 33°C are stressful to fish and should be avoided in aquaculture production (Lim & Webster, 2006). Cichlids are susceptible to disease if they are kept in particularly stressful environments. Disease results from the interaction of the environment, the fish and the presence of pathogenic organisms which can be parasites as well (Bauer, 1962). Particular care must be taken to monitor the quality of the aquatic environment such as pH, oxygen, temperature and turbidity which varies in natural systems such as rivers and cultured systems (Bauer, 1962).

Statistical analysis on number of fish infested (Zone/fish species/fish size)

Tilapia rendalli and *O. andersonii* are midwater slow swimmers (Ramberg *et al.*, 2006), thus the high numbers of fish collected as mostly multifilament gill nets were used, whilst in the case of *S. robustus*, which is a predator, they are mostly found along the banks where there is abundant vegetation and the setting of multifilament gillnets is difficult.

The different size group of target fish species and different host fish species showed no significant difference in the number of fish infested in parasite as well as the 4 different zones (Table 4.8 – 4.10). The results of the ANOVA analyses examining fish sizes, zones and host fish species were statically insignificant. Fish from the four body size groups had some presence of parasites, but it is clear that there is no clear difference in size class, although prevalence levels (Table 4.2 and Table 4.5) indicated that most medium fish sizes group were heavily infested with *Trichodina* and *Dactlogyrus* fish

parasite. The prevalence occurred mostly in *O. andersonii* and *T. rendalli*. Statistically it means that all sizes groups and fish species could have equal chance of encountering fish parasites.

Protozoans

Most of the parasites recorded in the present study have also been reported previously by Basson *et al.*, (2002) from Botswana in the Okavango Delta and river systems in South Africa. *Trichodina* sp. had a prevalence of 36 %, and in most cases co-exists with Monogenea (*Dactylogyrus* sp.) with a prevalence of 34%, on the host fish species examined. In numerous cases they were found on the same fish host.

Trichodina sp. has a simple direct life cycle. They reproduce via binary fusion, literally by cell splitting (Poulin, 2002). With such a direct life cycle multiplication in a cultured environment can be massive and cause high levels of mortality.

Trichodina sp. never occurs in high numbers on a healthy fish. A massive number of trichodinids can, by their constant attachment and rotating movements, seriously damage the epithelial or epidermal cells (Basson *et al.*, 2002). In the examined host fish species in this study the trichodinids occurred on the fins and skin. Basson *et al.*, (2002) conducted a study in the Okavango Delta, where five trichodinids genera were found associated with 68 of the 70 fish species examined; these were *Trichodina*, *Trichodinella*, *Tripartiella*, *Paratrichodina* and *Hemitrichodina*. Only six known species of the genus *Trichodina* are found worldwide exclusively on the skin and fins of fish hosts.

Trichodina sp., together with *Dactylogyrus* sp. (Monogenea) were found in high numbers on *O. andersonii* which is the most common fish species used for aquaculture farming in Namibia including the Kavango region as compared to *T. rendalli* and *S. robustus*. This implicates aquaculture, as the health of fish is of the utmost importance. The potential impact of heavy infestation of protozoan in

cultured systems is that they tend to lower the general health of the fish, thus fish become susceptible to other dangerous pathogens.



Figure5.1: Photograph of the host fish species i.e. *Oreochromis andersonii* and *Tilapia rendalli* that were examined for the presence of *Trichodina* sp. and *Dactylogyrus* sp. from the Kavango River, Namibia. (V. Mumba, 2012).

Infested fish shows signs of epithelial hyperplasia, and these areas are usually infested with bacteria as well as with protozoans (Van As & Basson 1984). Protozoan parasites mainly cause external infestations on most cichlid species. The high infestation level of protozoan may cause significant loss of fish in cultured environments as they affect the fish body surface causing wounds especially if they are in abundance (Lim & Webster, 2006).

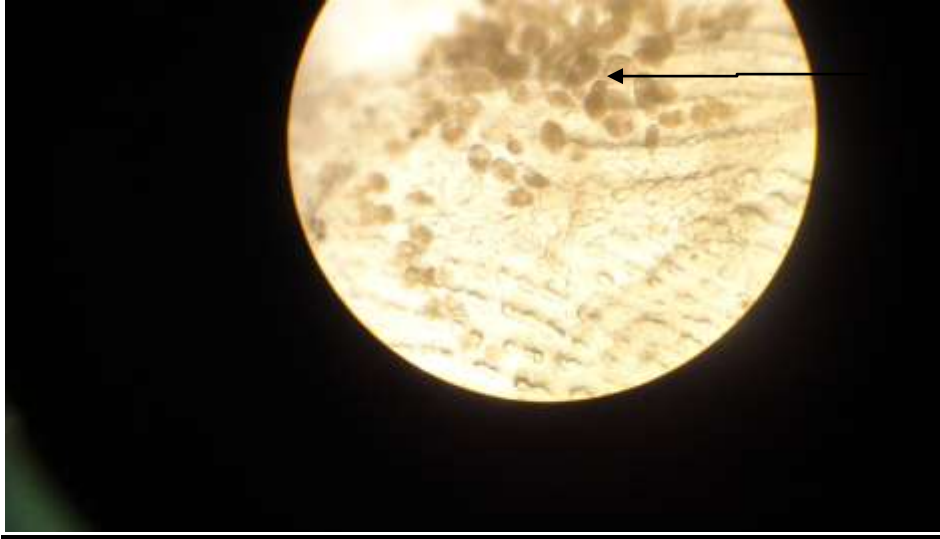


Figure 5.2: Light micrograph (Magnification B; 10x) of wet skin smear of *Tilapia rendalli* collected from the Kavango River, Namibia with *Epistylis* sp. infestation on the skin indicated by arrow. (V. Mumba, 2012).

Fish parasites that are found in abundance such as *Trichodina*, *Tripartiella* and *Trichodinella* species can cause a common disease called Trichodiniasis and can be found in the wild as well as in cultured fish species (Van As & Basson 1984). In the present study the relative abundance of these parasites that were found was not very high (less than 20%, Fig. 4.3). Therefore the threat in the Kavango River is minimal. However fish species infested with these ciliophorans in the wild can cause problems when transferred to cultured environments, as they can multiply very fast via binary fission. This happens when the fish host is being transported to a new environment, and during handling of the fish.



Figure 5.3: Red sores (arrow) on *Tilapia rendalli* collected from the Kavango River, Namibia, with possible infestation of *Epistylis* sp. on skin. (V. Mumba, 2012).

Epistylis sp. is the most common and pathogenic type of sessile, colonial ciliate (Esch *et al.*, 1975) and is usually found in the summer months. *Epistylis* sp. parasites can weaken and kill fish. Ulcers caused by *Epistylis* infections can make fish more vulnerable to bacterial infections. For example, red sore disease (Fig 5.3) involves the combination of *Aeromonas* bacteria and *Epistylis* (Durborow, 2003). Red sore lesions may become secondarily invaded by water moulds, they tend to be chronic but acute mortalities can occur, usually caused by systematic bacterial infection (Noga, 2010).

Helminths and other worms

Monogenea parasites are characterized by high species richness (Poulin, 2002). In general, Monogeneans have a direct life cycle with no intermediate host. The hermaphroditic adults are oviparous and produce eggs into the water which hatch prior to attaching to the gills of a fish host

(Poulin, 2002). Moreover, they are highly host-specific compared to other groups of parasites, thus may explain the high numbers found in *Tilapia rendalli* and *Oreochromis andersonii*.

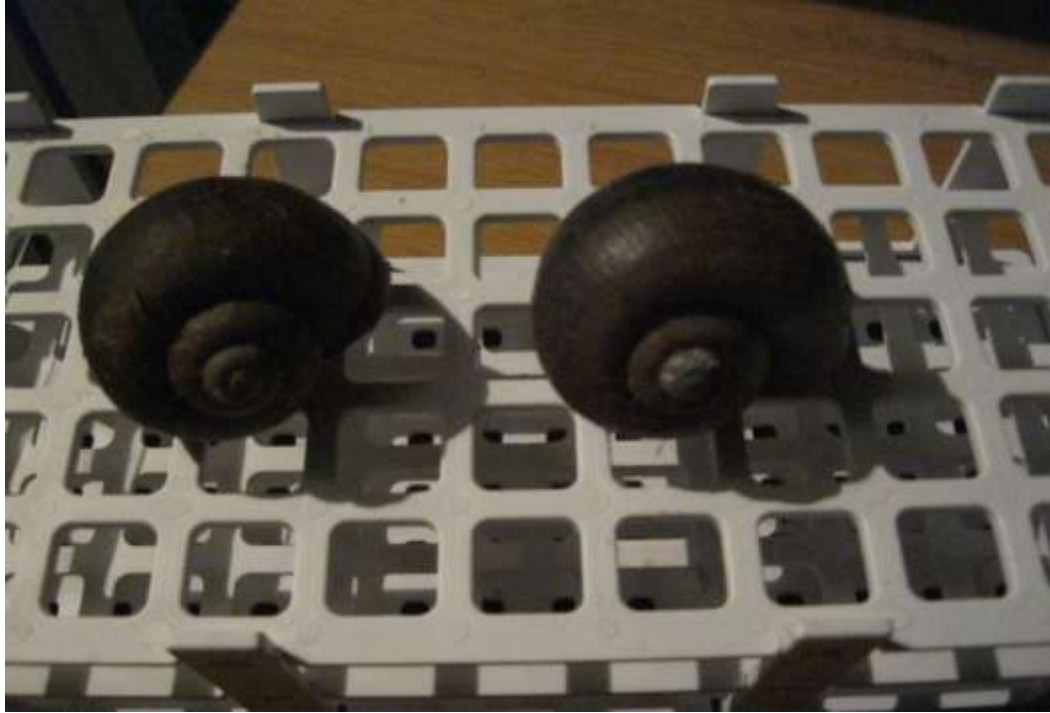


Figure 5.4: Intermediate host (*Pila occidentalis*) of some Trematode species. This Southern African snail is confined to the Okavango and Kunene River systems in Northern Namibia.

According to Ramberg *et al.*, (2006) most cichlids especially tilapia species are midwater slow swimmers and hover in open waters and in vegetation areas. They feed on plants, small invertebrates and detritus. They may be intermediate hosts of various parasites which inhabit the guts of the host after consumption, especially if the transmission is by a definite host such as birds. Clinostomatid trematodes can use mammals as a primary host (Britz *et al.*, 1984). This has not been reported in Africa yet, but has been recorded in India and Japan. A study was conducted in India and reported that 11 *Clinostomum* specimens were found in a cat attached to the upper gum in the region of the canine

teeth (Britz *et al.*, 1984). Another study in Japan reported a single specimen of *Clinostomum complanatum* in the pharynx of a woman (Yamashita, 1938). *Clinostomum* parasites can be a potential threat to humans if the fish is not properly cooked or partially sun dried.

Observing the prevalence of *Procamallanus* sp. in the target host fish in this study, shows that the intermediate in this case, copepods, are present in the habitat (Zone 4). This is due to the volume of vegetation (Zone 4) which is in abundance and gives rise to a more extensive habitat for the copepods therefore; fish are more exposed to greater concentrations of *Procamallanus* sp. than in Zone 2 and 3.

The presence of Nematoda in fish may lead to a decline in fish population in natural environments if found in abundance (Royce, 1972). Some nematodes such as *Anisakis* sp. can be serious public health problem to humans if ingested causing a parasitic diseases called Anisakiasis when nematodes attaching to human esophagus, stomach or intestines. (Noga, 2010).

Parasitic worms do not appear to present much of a problem in cultured environments of most cichlids such as tilapia (Plump, 1997). However, a significant population of adult helminths in the intestines may limit growth and nutrient uptake (Lim & Webster, 2006) in some cases the migrating larvae may cause problems or may encyst in muscles of fish resulting in a loss of value on the market place.

Helminths are problematic only in the production of fry, fingerlings and larvae stages of fish. In aquaculture systems, the most problematic parasitic worms are the monogeneans (Lim & Webster, 2006). They can cause mortalities and also contribute to the death of their hosts due to other secondary infectious diseases.

Parasitic Crustaceans

Most parasitic crustaceans in this study have been recorded in previous studies done in South Africa and Botswana (Basson *et al.*, 2002). The abundance of the entire parasitic crustacean group found in this study were very low, not exceeding 0.04 which gives an indication that the number of fish hosts with crustacean parasites were low.

The high number of *Opistholernaea* sp. was found in the river's backwaters and floodplains near Mpungu fish farm and in the fish farm ponds. This high number of fish parasites in this area could be due to the high number of human activities happening in that area, such as washing of dishes, cars, animals' drinking water and people bathing.

As with Karovo fish farm, which is found in Zone 3 and KIFI zone 4, where aquaculture is also practised just like at Mpungu, the infestation of *Opistholernaea* is close to zero.

The high number of *Opistholernaea* in Zone 1 could also be due to bad management practices of the fish farm waste water spillage from the farm. The waste water from the ponds is used for watering vegetables and trees at the fish farm but some excess water overflowing from the earth ponds end up in the river during the flooding season. The water might be a carrier of these parasites thus ending up in the wild. The study done in Botswana by Basson *et al.*, (2002) indicated that *Opistholernaea* usually occurs in low numbers in the wild, with a low potential threat to the fish stocks. This copepod was mostly found in the eyes and gills and can cause blindness in fish.

Approximately 80 crustacean ectoparasites species have been recovered from freshwater fish in Africa (Fryer, 1968). Most of these crustacean ectoparasites species are host specific (Noga, 2010), although some are widely distributed ubiquitous species found on cichlids such as *Lamproglana monodi* are totally opportunistic in choice of hosts for example *Lernaea hardingi* (Paperna *et al.*, 1981). Other

parasites such as *Dolops ranarum* is found only when a specific host fish species occurs in a river system or farmed.

One of the most significant parasitic diseases in cultured environments around the world is White Spot caused by *Ichthyophirius multifiliis*. Although this parasite was not found in the present study it could be a potential threat on culture systems in the Kavango Region. This parasite is an opportunist as it mostly attacks when the fish are exposed to stress conditions in culture system and has been responsible for significant losses in fish aquaculture.

Although EUS (Epizootic Ulcerative Syndrome) was not part of this study, it is seen as a potential disease threat in aquaculture and the wild stock in the Kavango and Caprivi Regions, as it has been reported in the Kavango River system, by the Ministry of Fisheries and the Government is dealing with the outbreak (Kibria Ghulum, personal communication, 2012). The disease is highly infectious, caused by fungi and spreads rapidly in water. It is identified by small grey or red lesions that can worsen into large ulcers resulting in loss of scales and fins and haemorrhaging oedema. Currently, there are no known causes for EUS, but environmental pollution or poor water quality coupled with chemicals used in irrigation.

Data on infestations in cultured fishes in tropical Africa are limited. However, these parasites cannot be overlooked as potential risk to cultured fishes; their pathogenic effect is evident even when infesting natural fish populations (Paperna, 1980).

CHAPTER SIX

RECOMMENDATIONS AND CONCLUSIONS

Africa is considered to be the sleeping giant of aquaculture and presently it is relatively disease free. It is vitally important that African countries take steps to safeguard this status. One method of accomplishing this is to restrict the importation of fishes into Africa. The importation of ornamental aquarium fish as well as hybrid and genetically improved fish strains pose the greatest threats. The smorgasbord of fish and shellfish parasites and diseases that occur in Israel, for example, can to a large extent be ascribed to uncontrolled translocations. Every effort needs to be made to prevent a similar occurrence in Africa.

Ten years ago it was thought that there were very few commercially important fish diseases in aquaculture. Now there are several important ones, some are new, whereas others are old, such as white spot disease. These diseases in tilapias are very likely related to the global adoption of intensive methods of fish farming. Tilapia species is characterized by withstanding adverse water conditions better than most aquaculture species. Although Tilapia has been classified as disease resistant, water quality plays an important role in the process of controlling diseases.

Fish parasites are considered to be ecological indicators of natural system. The fish parasites provide evidence about conditions within the ecosystem, therefore if a natural system such as the entire Okavango system is altered by human activities, it can cause some parasites to transform into extreme pathogens which may cause mass mortality. Studying fish parasites on an ecosystem like the Kavango River provides information that goes beyond parasites alone; it could also give useful information on managing the ecosystems and its value in coping with potential problems in aquaculture in the Kavango region. Very little to no information exist regarding fish parasites in aquaculture on the

Kavango River, with the exception of studies done in the Okavango Delta on fish parasites. With the MFMR (Ministry of Fisheries and Marine Resources) research centre and pilot fish farm projects in aquaculture that is developing in Namibia, it is imperative that this critical information be collected to be incorporated into these fish farms and research centres.

Results obtained from the ANOVA analysis (Tables 4.8 – 4.10) showed no significant difference in number of fish infested between fish species, zones and different group size of target fish species. This clearly indicates that the fish infestation of parasites does not differ in fish species, sizes and zones, there any significant difference. There was no indication of any introduced fish parasites, specifically on the three target fish species; therefore there is less or no danger of parasites on aquaculture, regarding the current and potential fish which is farmed if fish health aquaculture practices are implemented correctly.

Fish from different habitat zones along the entire Kavango River were collected and examined. This highlights the four different types of habitats found in the river system as well as the population of the three target fish species, along the entire river system during the period of one year. The other important aspect in the habitat zone was the prevalence and abundance of fish parasites found on the fish hosts and its relation to the habitats. On average the water temperature and pH in the river does not vary that much between the four zones, although in the floodplain water and backwater the temperature fluctuates with the seasons. Therefore fish in these habitats need to cope with slight fluctuations of water temperature as well as pH levels. In most cases, fish farmers collect brood stock in backwaters during the low water periods and during the high water periods they collect on the floodplains. Thus, the constant fluctuation in water temperature may result in high stress level in fish, resulting to an increase in parasites infestation especially protozoans found on the gills and on the skin.

Findings in this study concluded a high level of prevalence of protozoans on host fish species, which is an ecosystem indicator on how low and fluctuating water temperatures can affect the fish species.

Most fish parasites are harmless to humans as we are not an intermediate or final host to any of these parasites under natural circumstances. However, most of the helminths and roundworms may affect humans at the larval stage, if fish are ingested that are not properly cooked or eaten raw. In the Kavango Region, the risk of people getting infected by any of these worms is very low as the eating of raw fish is not common and dried fish are boiled before eaten. The boiling process kills all possible worms that were not killed during the drying of the fish.

This information is useful to aquaculture and with the current emphasis on aquaculture in Namibia, it is imperative that the Ministry be in a position to deal with any outbreak of fish parasites. It is a known fact that parasites can cause heavy losses in aquaculture ventures that will also relate to financial losses. The Ministry must therefore have a fisheries scientist specializing in this field to deal with such cases and implement good health management plans.

It is recommended that follow up surveys be done every two years, to identify any change in the trends of possible fish parasites that could affect the wild stock in the Kavango River. This will ensure that up to date information on fish parasites is available, that might affect the aquaculture sector in the Kavango Region and Namibia at large.

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Appendix 1

Appendix 1: The following protocol is recommended to be used by fish farmers when they encounter any sign of fish disorders.

1. First check the environmental water quality samples <ul style="list-style-type: none"> a. Oxygen b. Temperature c. pH
2. Place infested fish in aerated container
3. Check for possible physical abnormalities e.g. red sores, mucus on body, blood, red or pale gills and eyes
4. Check for possible behaviour abnormalities e.g. breathing and swimming
5. Perform skin and gills smear to check for possible parasites
6. Training in Identifying any: <ul style="list-style-type: none"> a. Protozoan parasites b. Worms c. Parasitic crustaceans
7. Identify any problematic parasites and prioritize the problem
8. Treatment: <ul style="list-style-type: none"> a. Short term treatment is medication (consult a specialist) b. Long term treatment is management of the environment especially in cultured earth ponds.

Appendix 2: Data sheet used during surveys

Host:

Place:

Dates:

GPS:

Skin smear:	<table border="1"> <tr><td><i>Trichodina</i></td><td></td></tr> <tr><td><i>Scyphidia</i></td><td></td></tr> <tr><td><i>Ichthyophthirius</i></td><td></td></tr> <tr><td>Monogenea</td><td></td></tr> <tr><td><i>Apiosoma</i></td><td></td></tr> </table>		<i>Trichodina</i>		<i>Scyphidia</i>		<i>Ichthyophthirius</i>		Monogenea		<i>Apiosoma</i>												
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Methods of Collection	Type and Number of fish species
Gill nets	
Scoop nets	
Cast nets	
Anglers	

COMMENTS
