PARASITES OF THE BLACK SEA BASS, CENTROPRISTIS STRIATA

by

LESLIE ANN TOOKE

(Under direction of Dr. Randal L. Walker)

ABSTRACT

A one-year study (November 2000 – November 2001) of internal and external parasites of the black sea bass, Centropristis striata from a wild population offshore Sapelo Island examined the seasonal dynamics of parasites in relationship to black sea bass health. Over a twelve-month period, parasite abundance and prevalence was recorded and compared to the physiology of the fish. At least fifteen fish were collected monthly and sampled for internal and external parasites. This study on black sea bass from Georgia revealed nine species (two internal and seven external) of parasites which may have the capability of causing epizootics in aquacultural operations. The biology of the black sea bass is reviewed including age, growth, and reproduction; and biological aspects relevant to management of future aquaculture of the black sea bass are discussed.

INDEX WORDS: Black sea bass, Centropristis striata, Parasites, Copepods, Nematodes, Trematodes, Aquaculture

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DEDICATION

This thesis is dedicated to my husband, Jay Tooke. His support and encouragement throughout this process has been instrumental and sustaining. I thank him for listening to my complaints and devoting his time to help me. He has always stood by my side and encouraged me along the way. I owe so much to him, and I thank him for being patient with me these past two years. Most of all, I thank him for being my husband and the father of our beautiful daughter, Ryleigh.

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CHAPTER 1

INTRODUCTION

The black sea bass, Centropristis striata (Linnaeus, 1758), ranges along the Atlantic coast of the United States from the Gulf of Maine to the Florida Keys and is an important commercial and recreational species (Miller, 1959). Over the past twenty years, commercial landings have more than doubled in dollar value; however, a trend of decreasing metric tons of catch has been established (Personal communication from the National Marine Fisheries Services, Fisheries Statistics and Economics Division, Silver Spring, MD). This indicator of over fishing by both recreational and commercial fisheries illustrates the need for better management of this species. As natural stocks dwindle, aquaculture is viewed as a viable means for producing seafood in the United States. The goal of this study is to monitor parasite abundance in healthy black sea bass. Such data will provide pertinent information about potential health threats in the aquaculture of black sea bass.

Life History

The black sea bass is a quasi-catadromous, marine pelagic fish (Grosslin and Azarovitz, 1982). Spawning varies within their range depending on the range of latitude. Spawning occurs earlier in the southernmost region (Schwartz, 1964), but the number of times fish spawn during a season is unknown (Hoff, 1970). According to a research review by Mercer (1989), sexual succession is mainly a post-spawning process indicated by the frequency of occurrence of transitional individuals, which increases after spawning periods. Spawning of C. striata occurs offshore during the spring and early summer months (March-June) (Cupka et al., 1973).

As the female fish matures, it becomes more rotund and a yellow-green discoloration occurs, which occasionally darkens. A male 6 years old or older during the spawning season develops a bright blue nuchal hump. Tips of the male's rays also develop filaments on the median fins and coloration intensifies to vibrant fluorescent blues and greens (Lavenda, 1949). Color changes, affiliated with seasonal spawning, become more prominent with increasing age (Kendall, 1977).

Spawning of the black sea bass may occur in inshore estuaries or on offshore reefs. Little is known about the early life history stages, except that the eggs are pelagic. The diameter of eggs are about 1 mm. Larvae greater than a length of 13 mm have not been collected in pelagic samples from offshore areas, which suggests a short pelagic stage and a switch to a demersal or estuarine existence (Furse, 1995). Pelagic larvae are transported inshore via currents/tides to assume demersal estuarine habits. In the estuarine nursery, larvae prefer areas associated with high salinity and live bottoms (Cupka et al., 1973). Juveniles are observed within the estuaries around jetties, wrecks, piers, and shell bottoms (Mercer, 1989). Juveniles vacate estuaries once the water temperature drops below 10˚C and move offshore to warmer waters (Cupka et al., 1973).

As a protogynous hermaphrodite, the black sea bass exhibits sexual reversal over a wide age and size range (Low, 1982). Cupka et al. (1973) recorded a predominance of females ranging in size from 130mm to 290mm standard length while males predominate from 290mm to 370mm. His findings were recorded in deep-water populations (>30 m), and age > 4 years (Low, 1982). Cupka et al. (1973) observed this lower value in the male predominance to the size of a five-year-old fish, which Lavenda (1949) suggested as the age of sex reversal in this protogynous species. Females mature at about 2 years of age while male gonadal tissue is first seen around 3 years of age (Reinboth, 1965). The largest female has been recorded as 34 cm and 8 years old, while males reach 45 cm and at least 12 years (Lavenda, 1949).

Habitat and Biogeographic Region

Adult black sea bass are predominately found at offshore depths of 20 to 60 m, however, some may occur at depths of 10 to 120 m (Mercer, 1989). South of Cape Hatteras, adult fish reside in offshore areas year round. In contrast, black sea bass move northward and inshore during spring months and south and offshore in fall months in the Middle Atlantic Bight. Differences in temperature regime appear to be the cause of migratory and seasonal patterns between the South Atlantic and Mid-Atlantic Bights. Black sea bass in the middle Atlantic Bight have been caught in temperatures as low as 6˚C, but are most abundant in waters greater than 8-10˚C. Tagging studies have shown that black sea bass are nonmigratory in the areas of South Carolina, Georgia and along the east coast of Florida (Mercer, 1989). Although seasonal migration was not observed in the waters of South Carolina, Cupka et al. (1973) noted that offshore movement occurred as the sea bass increased in age and growth.

Diet

According to the current research reviewed by Mercer (1989), the black sea bass are carnivorous bottom-feeders that consume fish, mollusks, crustaceans, and echinoderms. Juveniles primarily eat shrimp, isopods, and amphipods, while adults feed on crabs and fish (Mercer, 1989). Based on the frequency of occurrence, the diet of black sea bass primarily consisted of crustaceans (91%), mollusks (28%), and fish (28%) (Link, 1980). Sessile organisms such as barnacles and colonial tunicates in the diet were indicators of grazing activity by adult fish (Cupka et al., 1973).

Parasites

About 10,000 parasite species are known to live in fish. Approximately one-third of those species are trematodes. About 4,200 species are ectoparasites including Monogenea, Crustacea, Hirudinea, Coelenterata, and several groups among the Protozoa. Digenea, Cestoda, Nematoda, and Acanthocephala are intestinal parasites of fish. They

enter the fish as larval stages in food organisms. Most ectoparasites have direct development; their eggs either attach to the bottom substrate or are shed into the water. The hatched larvae develop into a short free-swimming stage. After further developmental stages, they attach to a host and mature. Platyhelminthes and Nemathelminthes are the two groups that make up the endoparasitic helminthes. The eggs are shed into the water in which the first larval stages develop inside each egg. The first intermediate host usually swallows the first larva within the egg of both the nemathelminth and platyhelminth species. However, in several platyhelminth species, the first larva hatches from the egg to form a free-swimming larval stage. The larva is then ingested by the first intermediate host (Anders and Möller, 1986).

The life cycle of several parasite groups involves more than one intermediate host with the exception of Acanthocephala, which has only one intermediate host. The Digenea has the most complicated development. For all the helminth species, the final host is infected after it ingests the second (or first in Acanthocephala) intermediate host. The eggs are produced in the organs with a connection to the intestines or the intestines themselves; however, there are a few exceptions. The eggs are then shed into the water with the feces (Anders and Möller, 1986).

The body of the host is the primary habitat and source of nourishment for parasites. Its second environment is the surrounding conditions that affect the parasite and its host. This secondary environment also governs the development, epizootics, and the population dynamics of the parasite. Environmental factors such as temperature and salinity are particularly important. Both factors control the development of the fish parasites (Grabda, 1989).

Parasites adapt their mode of life in several ways. The first adaptation is the shape of their body, depending on the parasite's location on the host. External parasites are usually clypeate in shape and flattened dorso-ventrally. Internal parasites are elongated or ribbon-like to allow for free flow of the intestinal contents. For adaptation

of the parasite's location, various organs of attachment are used. Hooks, suckers, and clamps allow the external parasites to cling to the host's body or gills or embedding of anterior portion into the mucosa. Suckers, attachment grooves, and hooks assist the internal parasites to adhere to the intestinal walls of the host. Physiological adaptations are also necessary for parasites. Certain intestinal parasites lack digestive systems and depend on the host to digest the food. Also, the dark conditions of their environment have led to a disappearance of sense organs in the parasites, mainly the eyes. Other sensory organs are restricted to a small number of sensory cells for internal parasites. The restricted spots are in certain areas of the body: on suckers, nematode labia, and on the tail (in male nematodes) (Grabda, 1989).

To meet their energy demand, external parasites use the oxygen dissolved in the water, while internal parasites decompose glycogen in their cells. Carbon dioxide and fatty acids are released as a result (Grabda, 1989).

The invasions of parasites occur after direct contact has been made between healthy fish and an infected one. When there is a high density of the host population, it enables the parasite to spread rapidly. Usually the parasitic larvae are the invaders. Fish parasites are transmitted from one host to another via various pathways depending on the species. In the first pathway, infection via alimentary tract, occurs after eggs, larvae, or spores have been ingested with food. In the second pathway, parasites are transported from host to host by different auxilliary animals, which is known as phoresis. In the third pathway, parasites invade a host is through the penetration of the fish's skin (Grabda, 1989).

Immune Response

Fish immune responses protect fish from attacks by microorganisms and animal parasites, but each environment triggers a different immune response. Fish living solitarily in cold water do not require as reactive an immune response, because cooler temperatures slow a pathogen's growth and reproduction. However, fish living in

schools, where pathogens can pass from fish to fish and where warmer temperatures are more optimal for rapid generation of microorganisms, need a highly responsive immune system (Post, 1983).

Potential pathogens of all fishes contain antigens: bacteria or bacterial toxins, viral particles, animal parasites, fungi or fungal toxins. These antigens stimulate immunological responses in the animal only when they are within the body or tissues of the host. There are two types of immune responses: humoral and cellular responses. Both cellular immune responses and humoral antibodies protect the animal from agents of disease. The skin of each fish produces a mucus layer to act as a chemical and physical barrier against potential pathogens. The mucus contains a bactericidal substance to reduce bacterial flora on the skin (Post, 1983). Once infected, fish develop a protective immunity. Disease outbreaks occur only when there is a large population of susceptible fish. As the infection increases, the more resistant fish develop immunity while those that are highly susceptible die (Dawe and Dickerson, 1995).

Factors of Outbreak

Temperature is the critical condition for an outbreak to occur. Usually, as the temperature rises (up to 25-28° C) parasite activity increases. Temperature increase also accelerates the reproductive process and initiates the parasite's life cycles (Dawe and Dickerson, 1995).

Stress also plays a role in an outbreak. Several types of stress affect the endocrine systems of fish which causes the release of steroids from the adrenal glands and results in suppression of immune systems, thereby decreasing the fish's ability to fight infections, parasites, and diseases (Collette et al., 1997). Several factors can exacerbate stress, including low dissolved oxygen, high temperature, crowding, chemical pollutants in the water, spawning, (Dawe and Dickerson, 1995) pathogens, and handling by humans (Collette et al., 1997). A fish's response to stress may include the production of excess mucus and thickening of the gill epithelium. In both responses, gas exchange, electrolyte

and pH regulation are inhibited and gill function is impaired. Stress can also cause separation of epithelial layers and gill hemorrhaging thus open the fish to further infection (Collette et al., 1997). Disease outbreaks occur when fish are spawning and when water temperature is rising in the spring (Dawe and Dickerson, 1995). By increasing water temperature, a decrease in oxygen availability in the water occurs causing the destruction of physiological proteins such as enzymes and hemoglobin (Collette et al., 1997).

Signs of Disease

The fish's epidermis becomes inflamed around the parasite cluster (encysting parasites) and will eventually peel off. The gills become broom-shaped due to the destruction of the soft tissue. The fish becomes depressed and the color often darkens in seriously ill fish. The swimming patterns become abnormal and the fish begin to rub their bodies against available substrates. The skin is further injured and hemorrhage occurs. The greatest harm occurs when an excessive numbers of parasites crowd the gills. This causes excessive mucus to be excreted in the gills, increased thickness of the epithelium, and necrosis with subsequent obstruction of gas exchange. Respiratory difficulties occur due to filling of the space between the gill lamellae. As a result, the fish can become hypoxic, weak, and die (Egusa, 1992).

As already mentioned, many parasites use attachment organs to stay on their host. The attachments often cause mechanical damage to the host’s body. Next, as the parasites feed at the expense of their host, the host is deprived of a considerable amount of nutrition. As a result, the fish will lose weight and develop anemia during a heavy invasion. Finally, toxic and lytic effects can occur in the host as a result of the parasite’s glandular secretions and metabolites may be toxic for the host (Grabda, 1989).

Commercial and Recreational Fisheries

In the Mid-Atlantic and South Atlantic ports, black sea bass is an important recreational and commercial fish. Over the last decade, commercial landings have continued to rise rapidly as seen in table 1 below.

Table 1: Annual Commercial Landings of Black Sea Bass, *C. striata* in the United States

Year	Lbs	$
1980	3,978,009	2,713,838
1981	3,735,229	2,787,092
1982	3,639,454	2,659,957
1983	4,000,659	2,981,856
1984	4,871,108	3,803,946
1985	4,024,853	3,760,034
1986	4,917,469	4,721,132
1987	4,656,217	4,709,586
1988	4,831,463	5,114,408
1989	3,945,473	4,654,195
1990	4,935,747	5,506,886
1991	3,907,751	4,655,747
1992	4,048,415	4,142,641
1993	4,150,007	4,216,652
1994	3,285,215	3,458,635
1995	2,814,075	3,701,037
1996	4,259,445	4,835,357
1997	3,518,964	5,001,400
1998	3,273,942	5,196,032
1999	3,626,789	5,888,343

The Mid-Atlantic fishery for black sea bass operates from May to December, using unbaited wooden slat lobster traps. Traps are set over the live-bottom reefs for a maximum 45 minutes. They are also an incidental catch in the snapper-grouper handline fishery. In the South Atlantic Bight, the fishery consists of a handline and a small trawl fishery. However, along the 18 m contour, a year-round fishery exits in the South Atlantic Bight. Highest landings for black sea bass occur during the winter and spring due to the additional participation of off-season shrimp fishermen (Furse, 1995).

Furse (1995) concluded that along the Middle Atlantic Bight, economical gain and commercial landings are higher due to the productivity of nearshore natural reefs. In the South Atlantic Bight, commercial fishing is more restricted, because the majority of reefs are artificial and built specifically for recreational fishermen.

Increased recreational fishery of black sea bass also has occurred due to the rising popularity of charter boats and the development of more sophisticated boats. Black sea bass landings for charter boats and party boats make up the majority of the North Carolina offshore reef fishery (64% by weight and 75% by number) (Struhsaker 1969; Chester et al. 1984). In Georgia, black sea bass are considered the dominant offshore recreational reef fish (Ansley and Harris 1981; Johnson et al. 1985).

Management of Black Sea Bass

In the South Atlantic Region, the Fishery Management Plan for the snapper-grouper fishery established the management regulations for the black sea bass that are caught in the Fishery Conservation Zone, which extends from 3 to 200 nautical miles offshore (South Atlantic Fishery Management Council 1983 after Furse, 1995). All species in the snapper-grouper complex are used in recreational and commercial fisheries exclusively for human consumption. The minimum size limit for black sea bass is 254 mm (10 inches). The minimum mesh size and trapping areas for fish traps are 25.4 mm by 50.8 mm (rectangular) or 38.1 mm (hexagonal). Commercial fishing is also prohibited

on marine sanctuaries, including artificial reefs and Gray's Reef National Marine Sanctuary in Georgia (South Atlantic Fishery Management Council 1988).

The reproductive history (females changing to males) of the black sea bass ensures that the majority of the harvest will be males. A minimum-legal-size limit of 254 mm allows fish to spawn at least one year as females. A potential future problem is that heavy fishing may alter the sex ratio, which favors females thus leaving an insufficient number of males for adequate reproduction (Musick and Mercer 1977).

Aquaculture

Selective breeding, controlled and induced spawning, larviculture, disease control, and nutrition are being researched intensively. Today the increased demand for high quality seafood is turning toward aquaculture for a higher intensification of productivity and profitability economic justification. Many countries have recognized the crucial need to produce high protein foods for dense human populations. The technology for aquaculture is changing rapidly and is following the analogous methods of intensive husbandry practiced with terrestrial animals. There is a range of techniques being used including low density populations, closed, high-technology systems in the form of water recycling and in the form of feed. Also, the environmental parameters for these techniques are controlled fully (Kimble, 1985).

Many countries are looking for alternative methods to obtain seafood, and aquaculture may well provide an economically viable means to increase seafood supplies. Not only does aquaculture help to meet the high demands for food, but also it may become an integral component of fishery management programs. Aquaculture enhances fisheries by producing mass quantities of juveniles of key species to stock lakes and coastal areas. In several cases, these restocked fish help to supersede those displaced by habitat alteration and overfishing.

Aquaculture is very diversified; for instance, aquatic farming also includes the production of special purpose fish for biological control, ornamental fish, and baitfish.

Aquaculture has advanced as a result of the development of technology. Technology has enabled scientists to induce spawning by hormonal injection and has made it possible to mass rear many valuable species. The application of these new technologies is still in its infancy which limits mariculture to mollusks and seaweeds as the chief products in saltwater (Ackefors et al., 1994)

In marine systems, less than 1% of all marine fish are cultivated and 17% of crustaceans are harvested. However, in fresh-water systems, 60% of all fish food production is cultivated. This ratio of marine to fresh-water aquaculture should change dramatically as the new technologies become more widely used in that marine systems should contribute more (Ackefors et al., 1994).

Many experts are concerned that the world's natural fisheries are in trouble due to the decline in natural fish stocks. Scientists and policy makers have recognized that seafood production could be supported by aquaculture in the future. Wholesale prices for black sea bass average from $1.25 to $1.50 per 0.45 kilogram. But, a live fish weighing 907 to 1134g can range from $3.50 to $4.50 per 0.45 kilogram on the American and Canadian wholesale sushi markets. At one point, live fish were being sold for $8.00 per 0.45 kilogram in 1997 (University of Georgia Marine Extension Service, personal communications). This new market for live black sea bass in sushi markets of the northeast has intensified the need to develop aquacultural methods for hatchery reared fingerlings of C. striata in order to protect the native stock while satisfying the demand (Cotton, 2002).

Studies have been initiated to address some of the techniques of fish culture. Culturing small black sea bass fringerlings was investigated by the Shellfish Aquaculture and Fisheries Laboratory in Savannah, Georgia. Several experiments were performed in order to determine the optimal temperature and commercial diet for rearing black sea bass fingerlings. In the temperature experiment, after two weeks, fish being raised at a temperature of 30 C doubled in size but most succumb to mycobacterial infections soon

after that time. At week 10, all fish raised at 30 C had succumbed to disease. Significant differences in weight were also found in fish at other temperatures. Fish reared at 25 C, by week 10 were equal in size to those fish raised at 20 C. Both of these temperature treatments produced larger fish than those raised at 15°C (Cotton, 2002).

Parasites can play an important role in the success or failure of aquaculture. It is the purpose of this study to examine the parasite population that exists in the natural black sea bass population and identify potential threats to aquaculture of this species. Presently, fish grower's pot-trap a minimum legal size fish (25.4 cm) and place them in recirculating fish tanks inland, growing them to a 907g size. Fishes densely stocked in cages or tanks are susceptible to parasitic epizootics, especially those caused by external parasites, which have a direct life cycle. Parasites can be transmitted from stocking centers, from fishes living in the wild, or fish feeds (Grabda, 1989). It is imperative that the grower is cognizant of various parasites and the susceptibility of aquaculture systems infiltration. Once parasites are introduced into a recirculating system, they can rapidly procreate causing a loss of fish crop.

CHAPTER 2

MATERIALS AND METHODS

Objective

The goal of this study is to monitor parasite abundance seasonally in healthy fish. Also, since black sea bass aquaculture is presently dependent upon minimal-legal-size wild caught fish, this parasite information may identify potential health problems in black sea bass aquaculture. Such information will be useful as a tool in maintaining the health of fish in hatcheries, aquacultures, etc. by understanding the biological role of the fish parasite and what is necessary to maintain good fish health. The pertinent data includes information such as water quality, nutritional diet, and the life cycles of fish. By coupling the study of the biology of Black Sea Bass and its parasites, one can determine when and how fish become infested and eventually become susceptible to disease and parasites. This knowledge will help prevent fish kills and illnesses in aquaculture operations and improve the management of aquacultural systems.

Materials and Methods

The field procedure for sampling parasites consisted of collecting Black Sea Bass. Fish were collected monthly from November 2000 to November 2001 with fish pots deployed from the R/V Spartina along the Altamaha artificial reef approximately 7.2 km offshore of Sapelo Island, Georgia (31º 18' 54" North; 81º 09' 19" West) (figure 2.1). The pot traps used were standard fish traps with mesh size and trapping areas of 25.4 mm by 50.8 mm (rectangular). A permit was obtained from the National Marine Fisheries Service (NMFS) in order to collect the fish samples. The artificial reef is made up of pallet balls and concrete rubble. Sampling took place within a radius of 1.6 km between the different sets of structures. Once in the reef area, four pot traps baited with menhaden

were deployed for thirty to forty-five minutes. Fishing continued until approximately twenty specimens were caught.

Once the fish were collected, the whole fish were placed in plastic containers in a solution of 1:4,000 of buffered formalin to seawater (35 ppt surface water at the sampling site). As of May 2001, MS222 was used in the sample solution to anesthetize the fish. Fish remained in the containers approximately 45 minutes. Containers were then shaken vigorously for 2 to 3 minutes to dislodge external parasites from the fish. The fish were removed and the formalin solution with external parasites was placed into a pre-labeled (External with fish number) 500 ml plastic jar. An additional tag identifying the fish was placed in the 500 ml plastic container. Formalin was added to the jar to make a 5% solution of formalin (Beverly-Burton, 1994).

For the whole fish, the gills were clipped from each side and placed in the corresponding individually labeled "external" jars containing the external parasites, which contained 5% formalin. Next, fish were clipped ventrally from the anus to pectoral fin then up behind the opercular cavity to the backbone. If needed, a second horizontal cut was made from the anus. The internal organs from the esophagus to the anus were removed and fixed. The body cavity was inspected for any unusual structures or wounds. If any abnormalities were found, a sample was obtained for pathological inspection. The internal organs were placed in a 5% formalin solution. The internal organs were stored in pre-labeled (Internal with fish number) 800 ml plastic containers. An individual fish identification tag was placed within the container.

The remaining portion of the fish was placed in a pre-labeled plastic bag for candling and for measuring length. The total length of the fish was obtained using a fish measuring board in centimeters. Each fish was filleted to inspect for parasites within the meat. This procedure is called candling in which the light penetrates through the translucent material and allows any foreign objects in the meat to be seen.

In the laboratory, formalin was exchanged for 70% ETOH for both internal and external samples. The exchange from formalin to 70% ETOH took several dilutions and decanting. The excess fluid that was decanted off from both internal and external samples was properly disposed. After each sample was prepared, inspection of the material began.

Analysis of the external samples began with the gills from each container. The primary lamellae were cut off the gill arch and placed into a petrey dish. Each lamella was inspected under a dissecting scope for parasites. Any parasites found were removed and added to the corresponding external sample. Material in the external jars was then inspected under a dissecting scope. The parasites collected from each external jar (gill sets and solution in which the fish was submersed) were sorted into species, counted and then placed in vials of 70% ETOH containing an identifying label as to individual host fish and time of collection.

Internal samples began with sorting out the feces and excess material that collected in the bottom of each jar. The internal organs were then placed into a petri dish where an incision was made from the stomach down the intestinal tract. Using a squirt bottle filled with 70% ETOH, the internal cavity was rinsed into the petri dish. The same procedure was applied to all the setae and liver. The material in the dish was then sorted out under the dissecting microscope. All collected parasites were placed in a pre-labeled vial containing 70% ETOH. Once all fish were processed, each vial was then reanalyzed to determine the number and type of parasite species. Each species was then sorted and numerically tallied.

Parasite means (number of) were analyzed by One-way Analysis of Variance (ANOVA), Tukeys HSD test using SPSS data editor v10.1 software. Descriptive statistics for total external, total internal, and total length were analyzed using Microsoft Excel 2000 software.

In months November 2000, January 2001, and April 2001, only gill arch lengths were determined. In the remaining months, different measurements were taken at different times resulting in some months with standard length (SL) and some months with total length (TL). In order to standardize the measurements, all lengths (SL and TL) were converted to total length by using Waltz et al. (1979) functional regression equation 2.1 of SL to TL (sexes combined). To convert gill arch sizes to total length, an additional 45 black sea bass were collected to develop our own conversion equation (equation 2.2). All final data were analyzed as total length (TL).

(2.1) SL = 8.179 + 0.7387 (TL) r = .97, n = 1903

(2.2) y = 8.5745x + 75.574 r = .77, n = 45

No data were collected for months December 2000, March 2001, and September 2001 due to ship technical difficulties or adverse weather conditions. In addition, for the month of May 2001, internal parasite data were collected, but external parasite data were not due to an unexplainable white substance that hindered the analysis process.

Environmental data was collected from the Skidaway Institute of Oceanography's SABSOON tower data (R2) and from Sapelo Islands National Estuarine Research Reserve (NERR) Marsh Landing site for the duration of the experiment, November 2000 –November 2001 (figure 2.1). However, the data for Marsh Landing were only collected from November 2000 through June 2001. Water temperature and salinity were collected as bottom water data. Our collection site is located approximately 6e-005 km offshore of the Marsh Landing site and 1.2e-005 km inshore of the SABSOON Tower.

In addition to this study, a histological overview of the black sea bass was preformed in conjunction with the University of Georgia College of Veterinary Medicine to look at microorganisms and tissue lesions. The procedure began by collecting tissues from the fish at necropsy and preserving them in 10% neutral buffered formalin solution. The tissues were trimmed and placed in plastic cassettes for routine processing. The tissues were processed in successive changes of 10% formalin, through graded alcohols

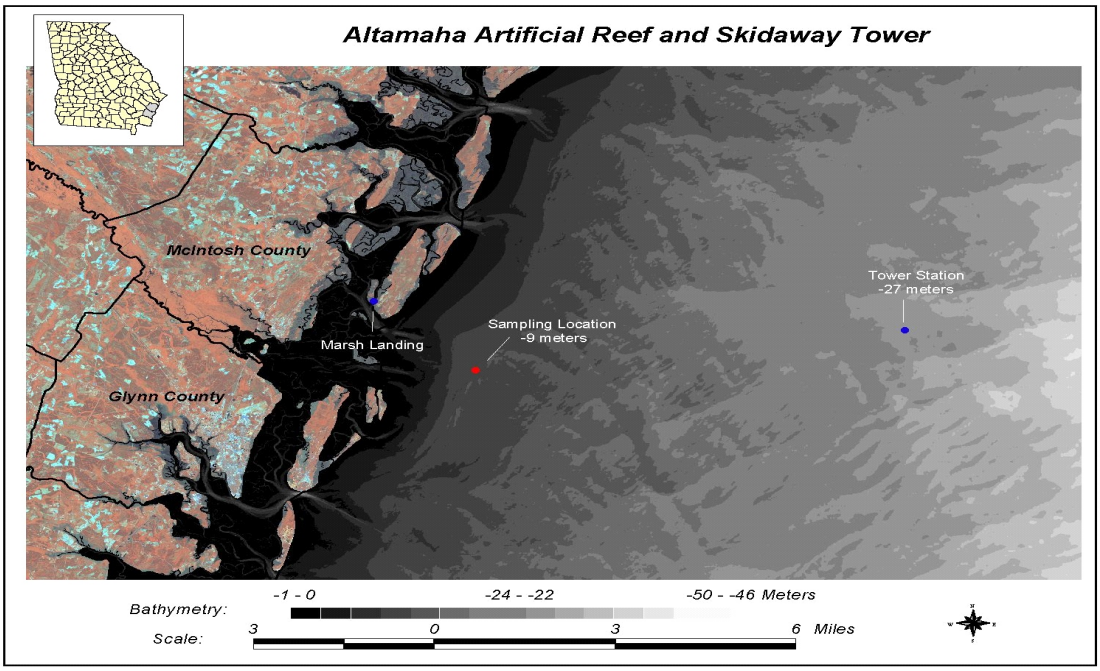

Figure 2.1 – Sampling location on the Altamaha artificial reef offshore of Sapelo Island, Georgia. Water quality sites: Marsh Landing and SABSOON Tower.

to 100% ethanol, xylene and infiltrated with paraffin wax. The paraffin tissue blocks were sectioned at 3µm using a microtome, placed in a water bath to remove any wrinkles, and collected on 1 x 3 inch glass slides. The tissue sections were stained with hematoxylin and eosin using an automated stainer. The tissue sections were coverslipped and examined microscopically for lesions.

CHAPTER 3

RESULTS

From November 2000 to November 2001, a total of 188 fish were collected and analyzed for parasites. Descriptive statistics for this study are presented in Table 3.1 giving the number of (N) of fish sampled, minimum and maximum number for both parasites per fish and total length of fish, mean number for both parasites per fish and total length of fish, and standard deviation for total external parasites, total internal parasites, and total fish length (TL). Figure 3.1 presents mean size (TL in mm) of fish each month in which data was collected for the duration of the study, November 2000 – November 2001. Monthly mean fish size ranges from 210mm – 354mm with individual fish ranging from 141mm to 410mm.

Table 3.1 – Descriptive statistics for the number of Black Sea Bass sampled from November 2000 to November 2001.

	Total Number of Fish Examined	Minimum Number of Parasites and Fish Size	Maximum Number of Parasites and Fish Size	Mean No. of Parasites and Fish Size Across Sample	Std. Deviation
External parasites	168	0	256	19.80	33.84
Internal parasites	188	0	4	0.39	0.720
Fish Total Length	188	141.00	409.98	255.78	69.35

Internal Parasites

Only two species of internal parasites occurred in black sea bass. Species A, a nematode of the genus *Capillaria* (figure 3.2), occurred every month that data was collected (figure 3.3); however, species AA, (an undescribed adult digenean) (figure 3.4) occurred only during the months of November 2000, February 2001, October 2001, and November 2001. Figure 3.3 presents the prevalence of occurrence for each month with ranges from 10% in June to 40% in February for species A and 5% in October to 30% in February for species AA. The mean number (±SE) of parasite per fish over time is graphed in figure 3.5 and ranges from 0.10 in June 2001 to 0.50 in November 2000 for species A and 0.05 in October to 0.45 in February for species AA.

The total number of internal parasites found each month in which data were collected for the study is graphed in figure 3.6 with a minimum of 2 parasites collected in January and June 2001 and a maximum of 18 collected in February 2001. Figure 3.7 presents a regression for the total number of internal parasites per fish versus total fish length in mm. A regression coefficient value of 0.0012 indicates that there is no relation between numbers of internal parasites and size of fish.

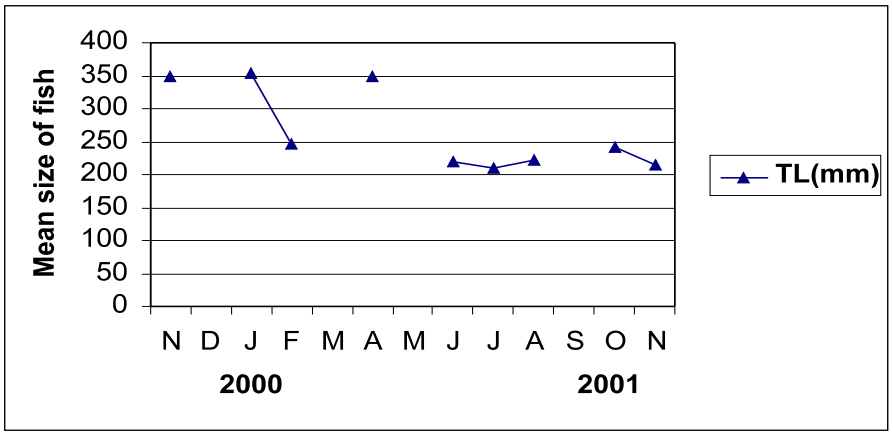

Figure 3.1 – Mean size (total length) of fish each month data was collected for the duration of the study, November 2000 – November 2001.

Figure 3.2 – Species A, a nematode (*Capillaria*)

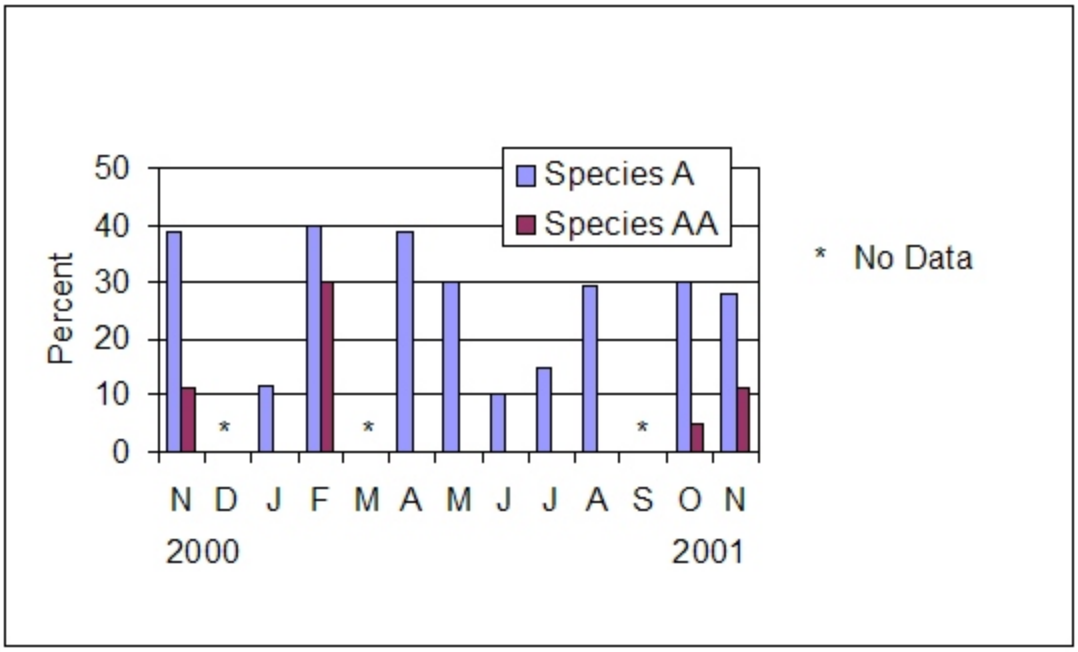

Figure 3.3 – Monthly prevalence for the two internal parasites of the black sea bass identified as species A, a nematode (*Capillaria*), and species AA, an undescribed trematode (adult digenean).

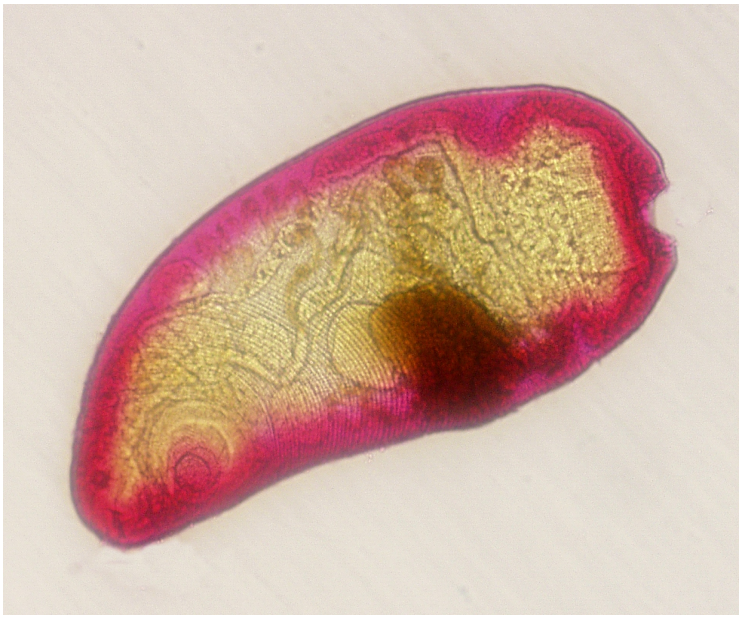

Figure 3.4 – Species AA, an undescribed trematode (adult digenean)

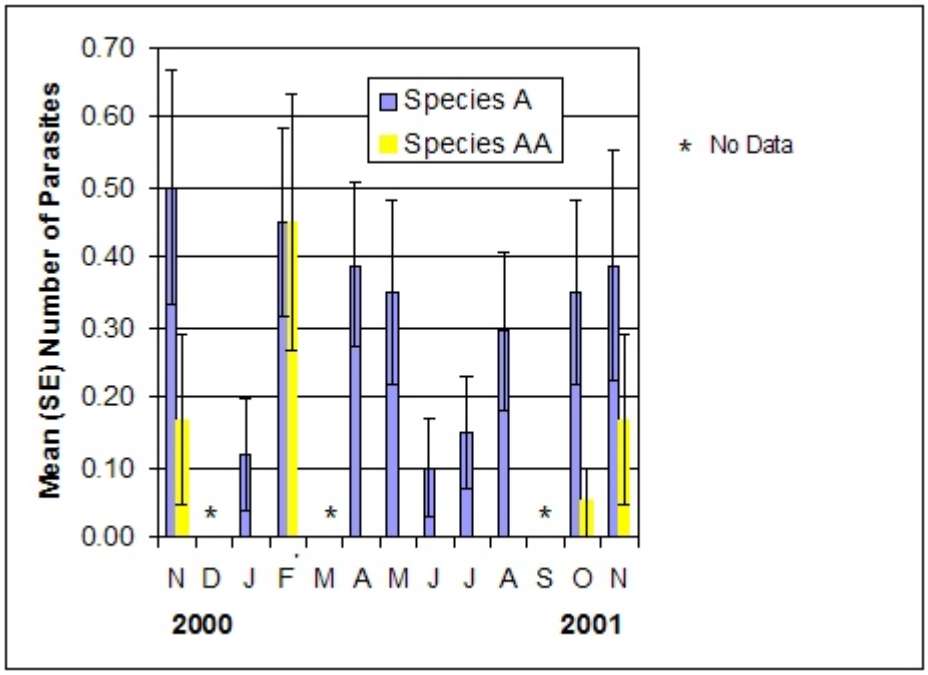

Figure 3.5 – Monthly mean number (±SE) of parasites per fish, versus time for the two internal parasites of the black sea bass identified as species A, a nematode (*Capillaria*), and species AA, an undescribed trematode (adult digenean).

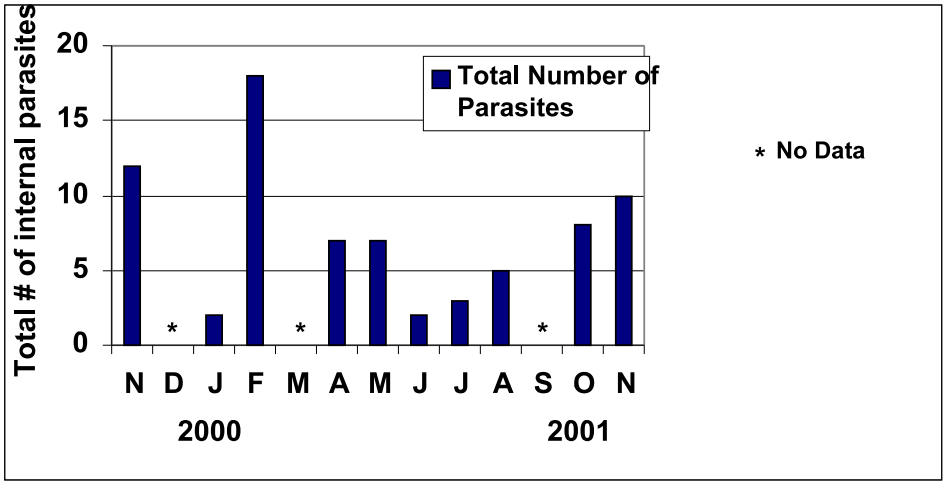

Figure 3.6 - The total number of internal parasites found each month data was collected for the duration of the study, November 2000 – November 2001.

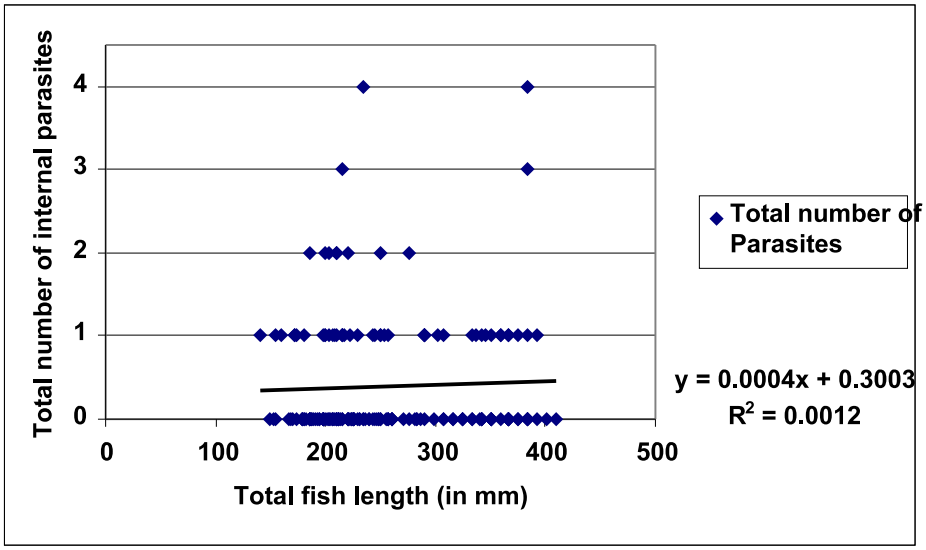

Figure 3.7 - A regression of total number of internal parasites in relation to total fish length (in mm).

External parasites

Species B, a copepodid stage in the Family *Lernaeidae* (figure 3.8), occurred every month in which data was collected in the sampling period. Figure 3.9 presents the monthly prevalence of the parasite which ranges from 47% in August 2001 to 100% in November 2000, January, February, and October 2001. The mean number (±SE) over time is graphed in figure 3.10 and ranged from 1.6 in August to 62.3 in April. A second organism, which is presumably the same species of *Lernaeidae* but in a different copepodid stage, a mature prematamorphosis female is Species C (figure 3.11). This stage occurred only in the months of November 2000, February, April, June, and October 2001. The prevalence of occurrence for each month is graphed in figure 3.12 with ranges from 10% in February 2001 to 78% in November 2000. Figure 3.13 presents the monthly mean number (±SE) of parasites per fish over time with ranges from 0.15 in February 2001 to 2.28 in April 2001.

Species D, a copepod identified as *Caligus elongates* (figure 3.14), occurred during the months of November 2000, January, August, and October 2001. Prevalence of occurrence is presented in figure 3.15 with ranges as low as 5% in October and high as 41% in August. Mean number (±SE) ranges from 0.05 in October to 0.41 in August and is graphed in figure 3.16.

Species E, Family *Bomolochidae* (a mature female copepod) (figure 3.17), occurred only in the months of November 2000 and June 2001 during this study. Figure 3.18 presents the prevalence of occurrence for this species with two percentages of 15 % in June and 22% in November 2000. Mean numbers (±SE) of 0.15 and 0.28 over time are presented in figure 3.19. Species F, also *Bomolochidae* (a copepod) (figure 3.20), also occurred only in the months of November 2000 and June 2001. Figure 3.21 graphs the prevalence of occurrence with percentages of 5% and 11%. Figure 3.22 presents the mean numbers (±SE) of 0.05 and 0.11 over time.

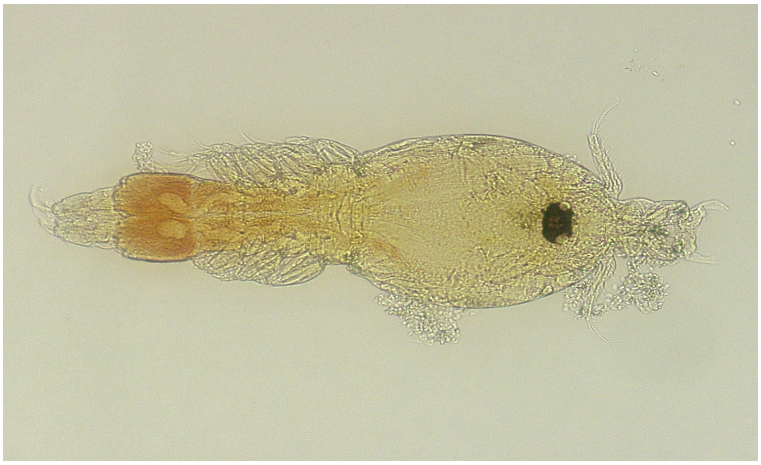

Figure 3.8 – Species B, *Lernaeidae* (Copepodid stage)

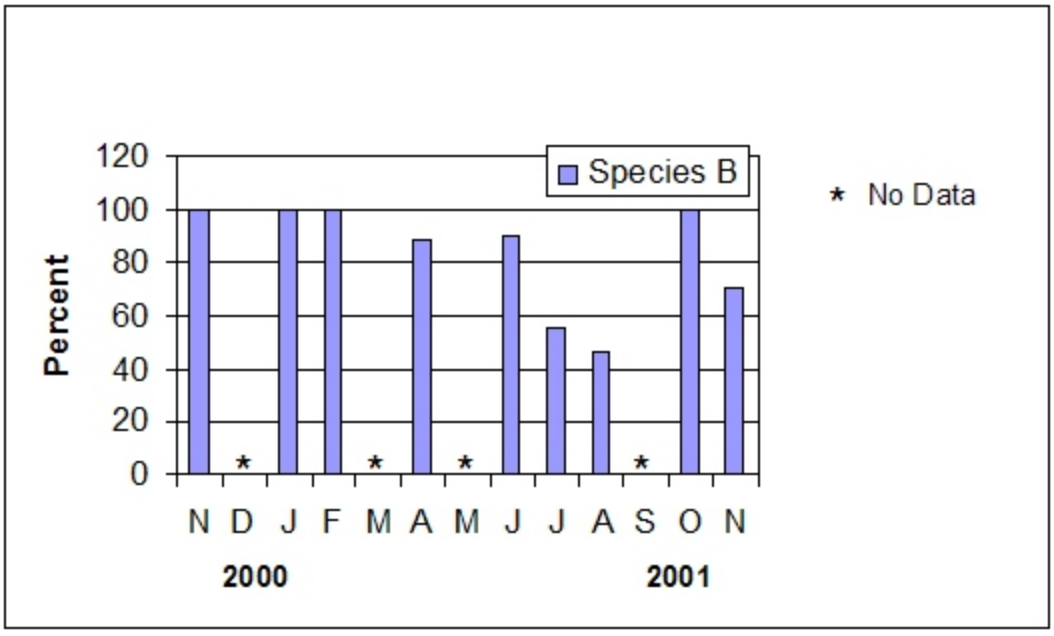

Figure 3.9 – Monthly prevalence for the external parasite species B, an undescribed copepodid stage of the family *Lernaeidae*, of the black sea bass.

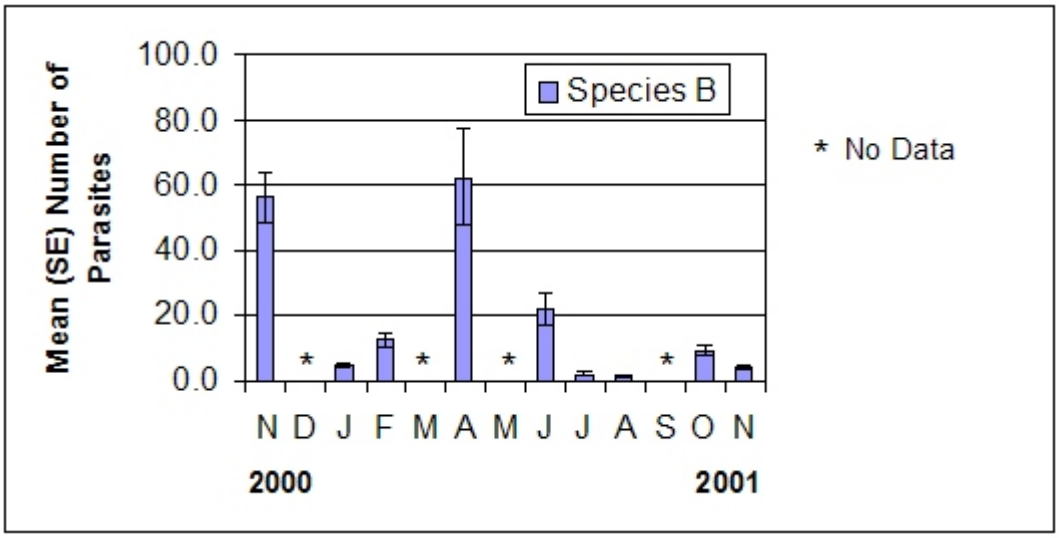

Figure 3.10 – Monthly mean number (±SE), versus time for the external parasite species B of the black sea bass identified, B = *Lernaeidae* (Copepodid stage).

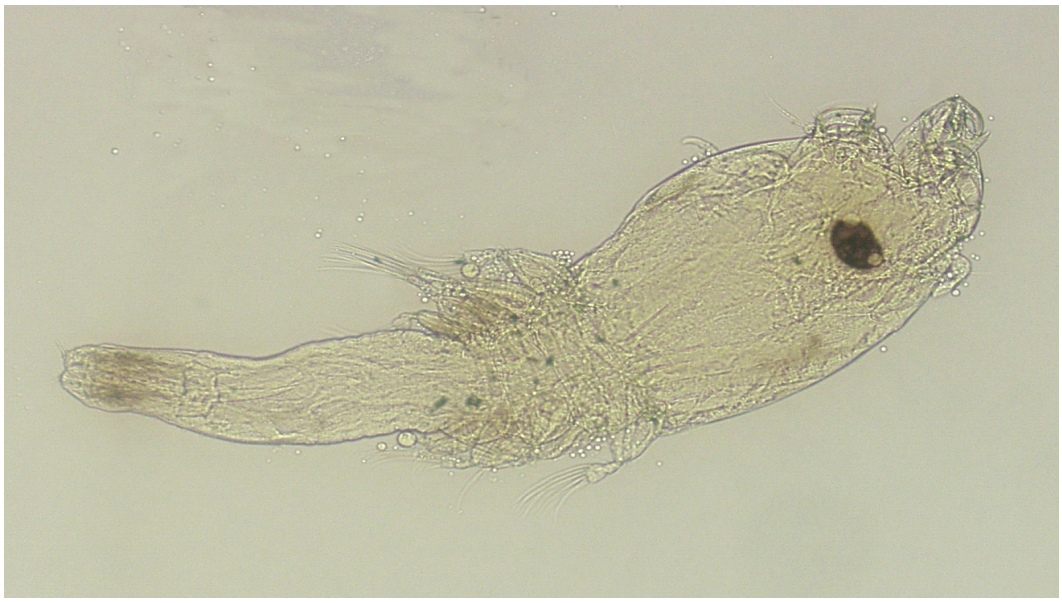

Figure 3.11 – Species C, Lernaeidae (mature prematamorphosis female)

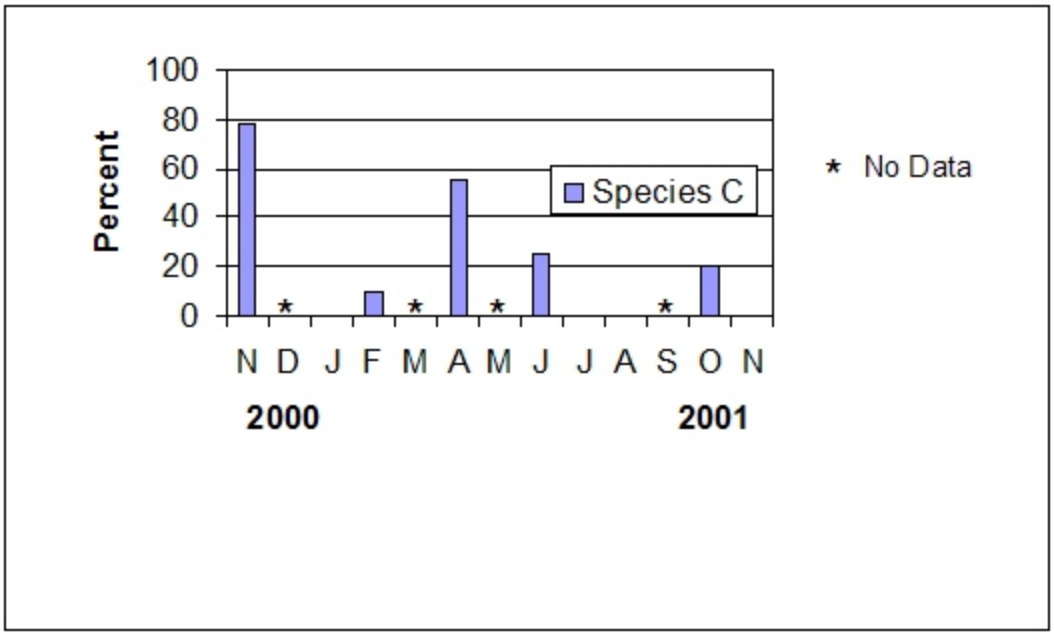

Figure 3.12 – Monthly prevalence for the external parasite species C, an undescribed mature prematamorphosis copepod female of the family *Lernaeidae* of the black sea bass.

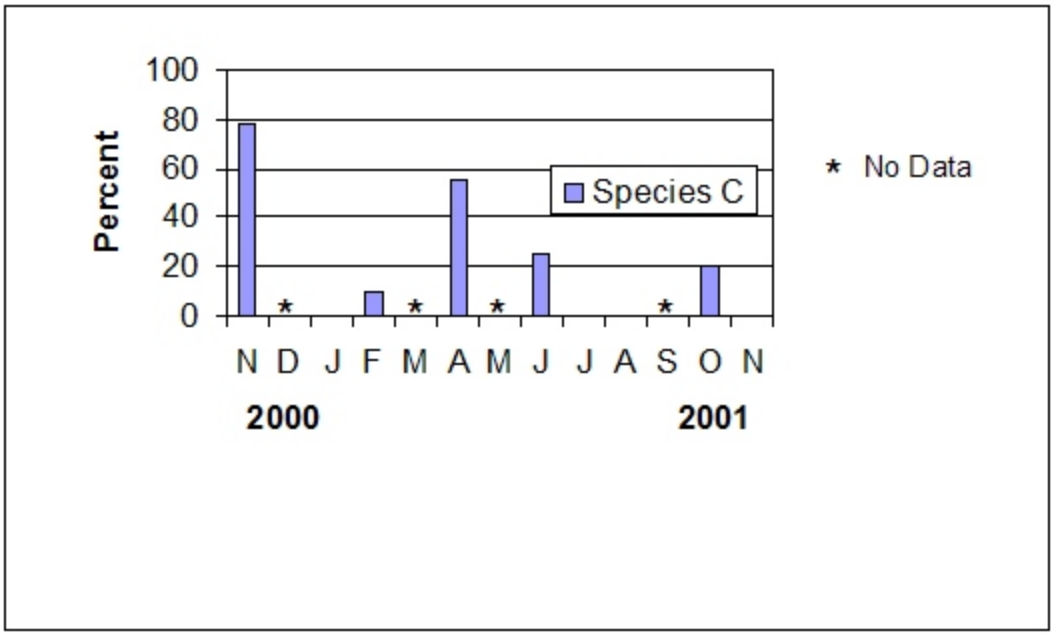

Figure 3.13 – Monthly mean number (±SE), versus time for the external parasite species C of the black sea bass identified, C = *Lernaeidae* (mature prematamorphosis female).

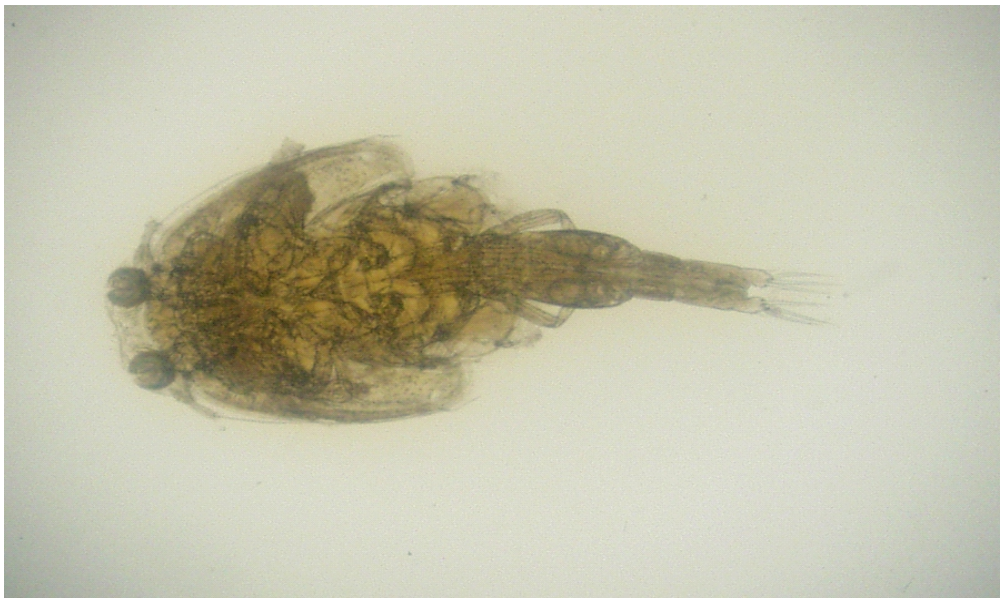

Figure 3.14 – Species D, *Caligus elongatus*

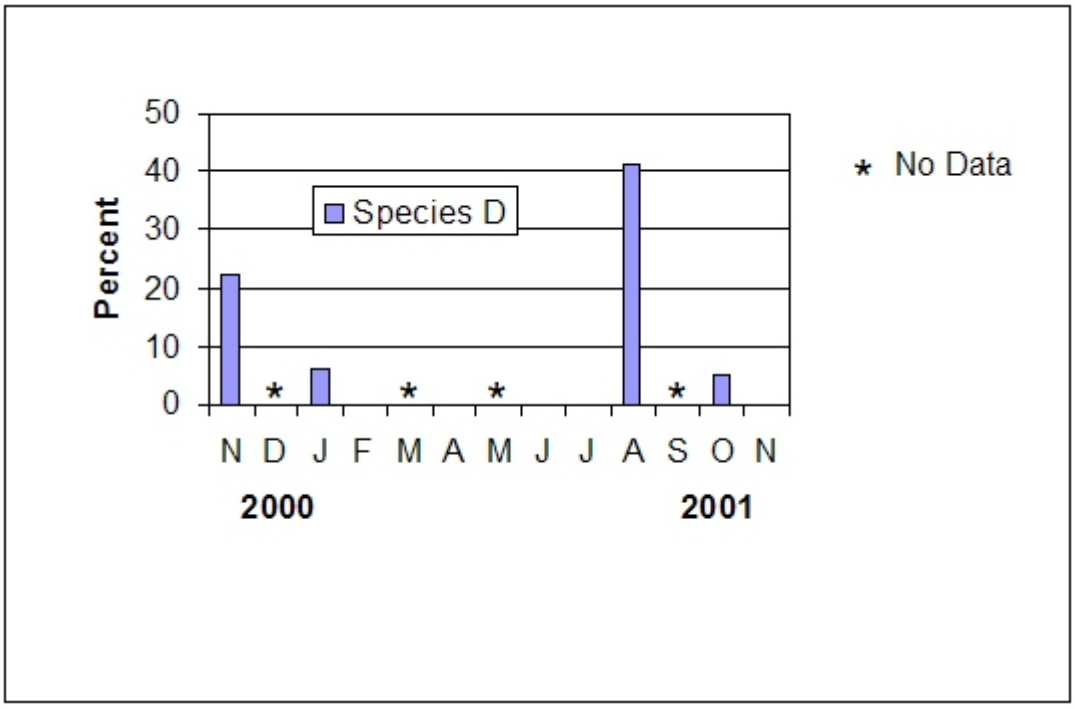

Figure 3.15 – Monthly prevalence of the external parasite species D, *Caligus elongates*, of the black sea bass.

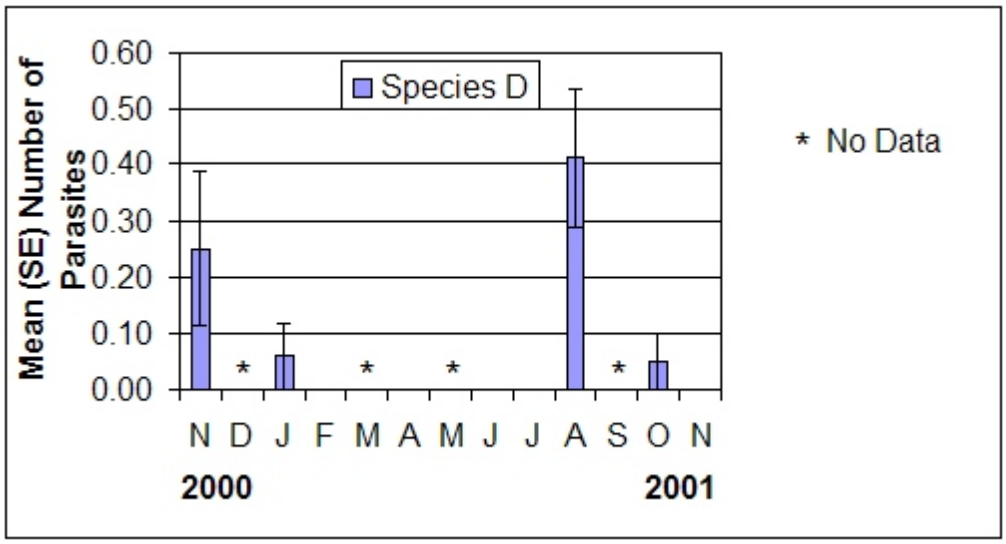

Figure 3.16 – Monthly mean number (±SE), versus time for the external parasite species D of the black sea bass identified, D = *Caligus elongatus*.

Figure 3.17 – Species E, a mature female copepod of the Family *Bomolochidae*

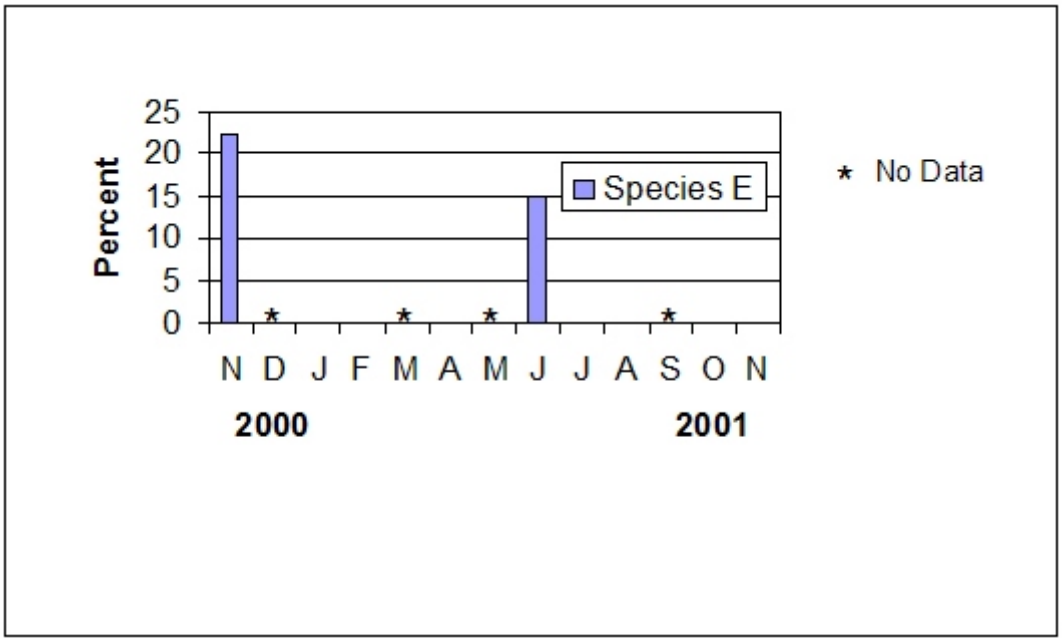

Figure 3.18 – Monthly prevalence for the external parasite species E, a mature female copepod of the family *Bomolochidae,* of the black sea bass.

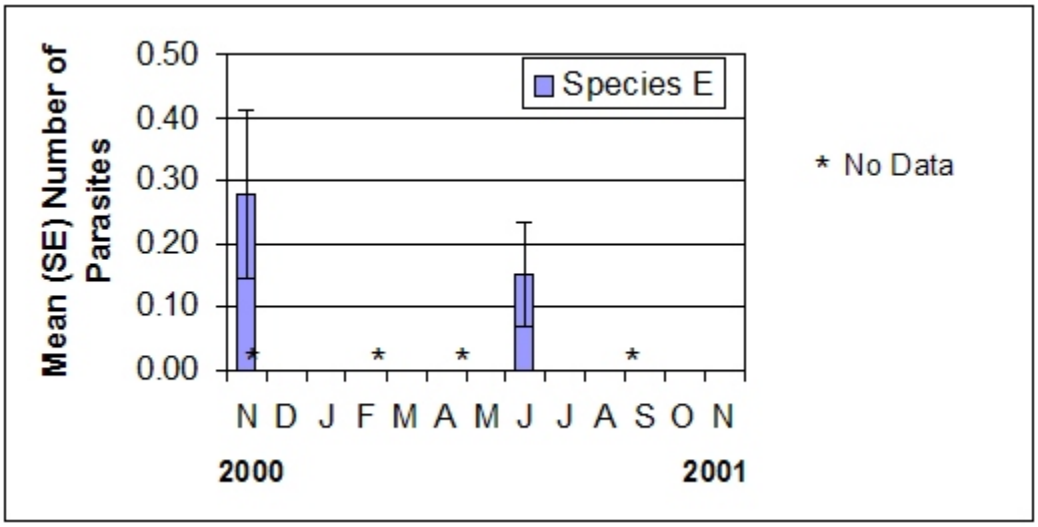

Figure 3.19 – Monthly mean number (±SE), versus time for the external parasite species E of the black sea bass identified, E = Family *Bomolochidae* (mature female copepod).

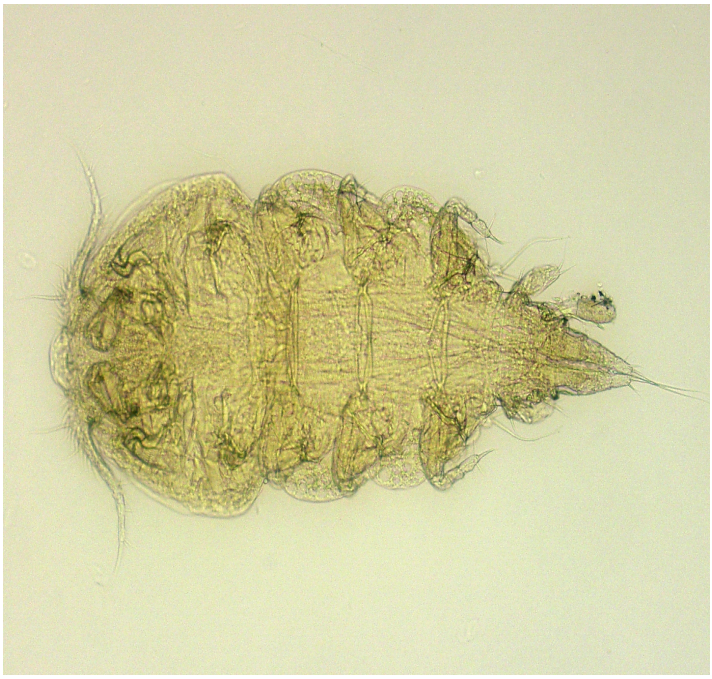

Figure 3.20 – Species F, a copepod of the Family *Bomolochidae*

Species G, *Bomolochidae* (figure 3.23), occurred during the months of November 2000, April, June, July, and October 2001. Figure 3.24 presents the prevalence of occurrence for this species with ranges from 1% in November 2000 to 6% April 2001. In figure 3.25, the mean number (±SE) is graphed with ranges from 0.05 in June, July, and October 2001 to 0.06 in November 2000 and April 2001.

Species H, a copepod of the genus *Argulus* (figure 3.26), occurred during the months of November 2000, February 2001, and June 2001 during the collecting period. The prevalence of occurrence is presented in figure 3.27 with ranges from 1% in November 2000 to 10% in June 2001. The mean number (±SE) over time is graphed in figure 3.28 with ranges from 0.06 in November 2000 to 0.10 in February and June 2001.

The total number of external parasites found each month in which data was collected for the study is graphed in figure 3.29 with a minimum of 34 in August 2001 and a maximum of 1164 in April 2001. Figure 3.30 presents a regression for the total number of external parasites per fish versus total fish length in mm. A regression

coefficient value of 0.1556 shows that there is no significant relation between number of external parasites and size of fish.

Water temperature and salinity data for this study are presented in Figures 3.31 and 3.32. Water temperature and salinity ranged from 13.1˚ C – 27.6˚ C and from 35.2 ppt – 36.2 ppt for the Skidaway Tower site (figure 3.31), respectively. In figure 3.32, the Marsh Landing site, water temperature and salinity ranged from 10.4˚ C – 27.8˚ C and 22.0 ppt – 31.8 ppt, respectively.

A multi-comparison of the mean number of external and internal parasites each month for each species was run against dates (every month sampled) using a one-way ANOVA and a Tukeys HSD test (table 3.2). The comparison showed that external species B was significant in months four (April 2001) and eleven (November 2000) and external species D was significant during months eight (August 2001) and eleven (November 2000). The comparison for internal species revealed no significance during any sampled months for species A and only month two (February 2001) was significant for species AA.

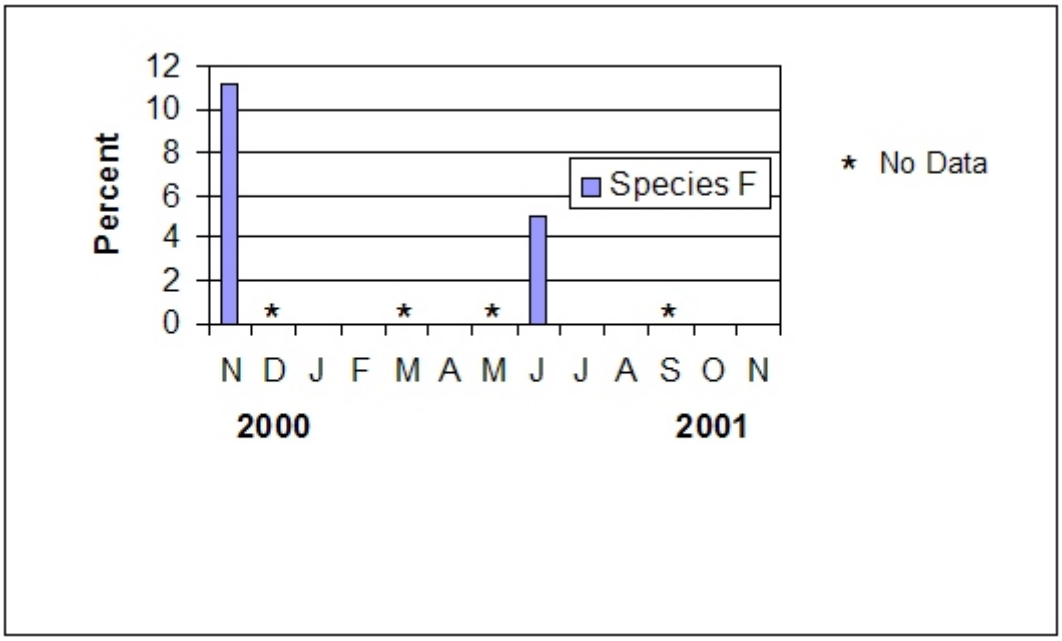

Figure 3.21 – Monthly prevalence for the external parasite species F, a copepod of the Family *Bomolochidae,* of the black sea bass.

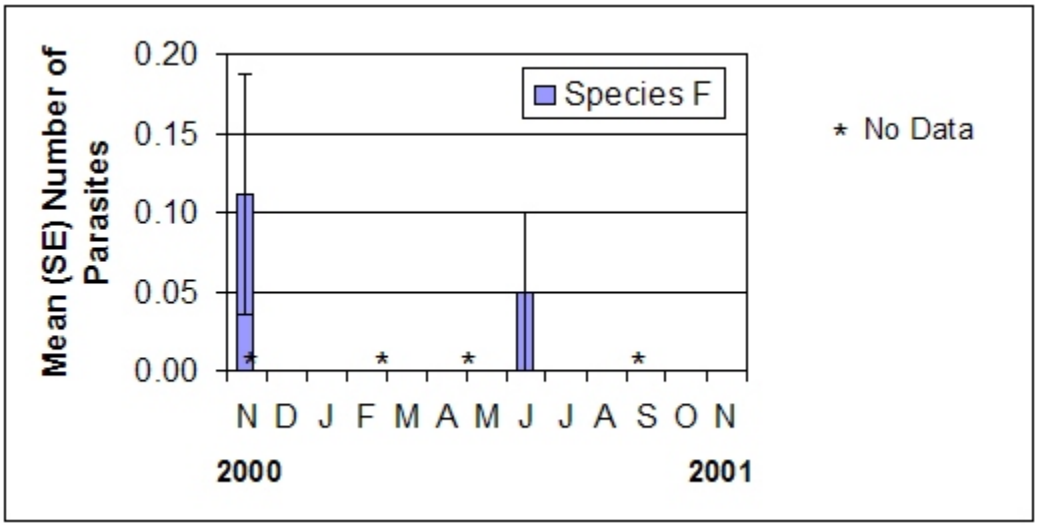

Figure 3.22 – Monthly mean number (±SE), versus time for the external parasite species F of the black sea bass identified, F = Copepod; Family *Bomolochidae*.

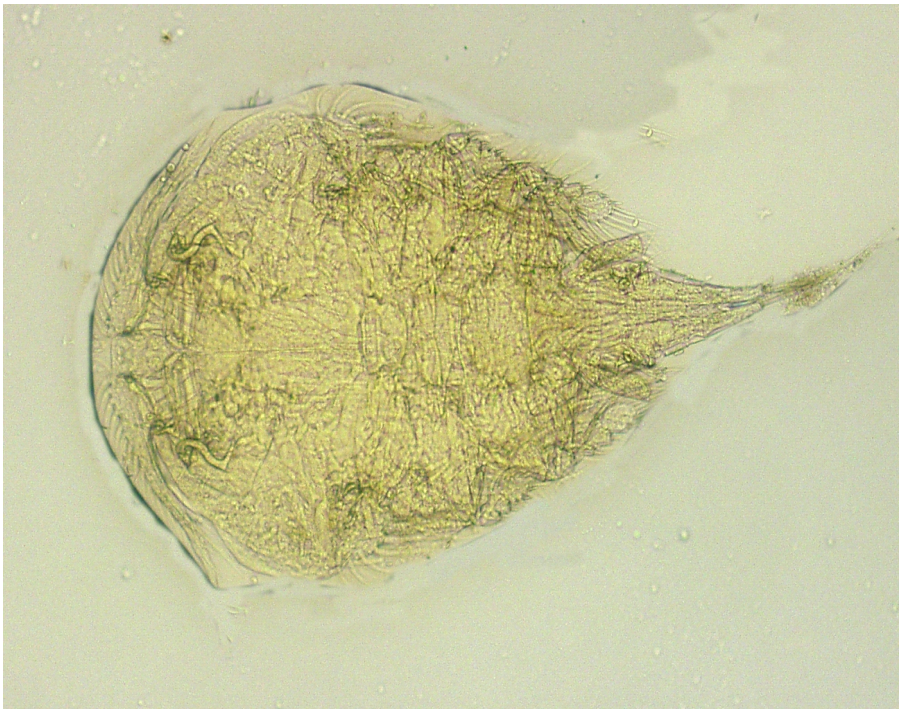

Figure 3.23 - Species G, a copepod of the Family *Bomolochidae*

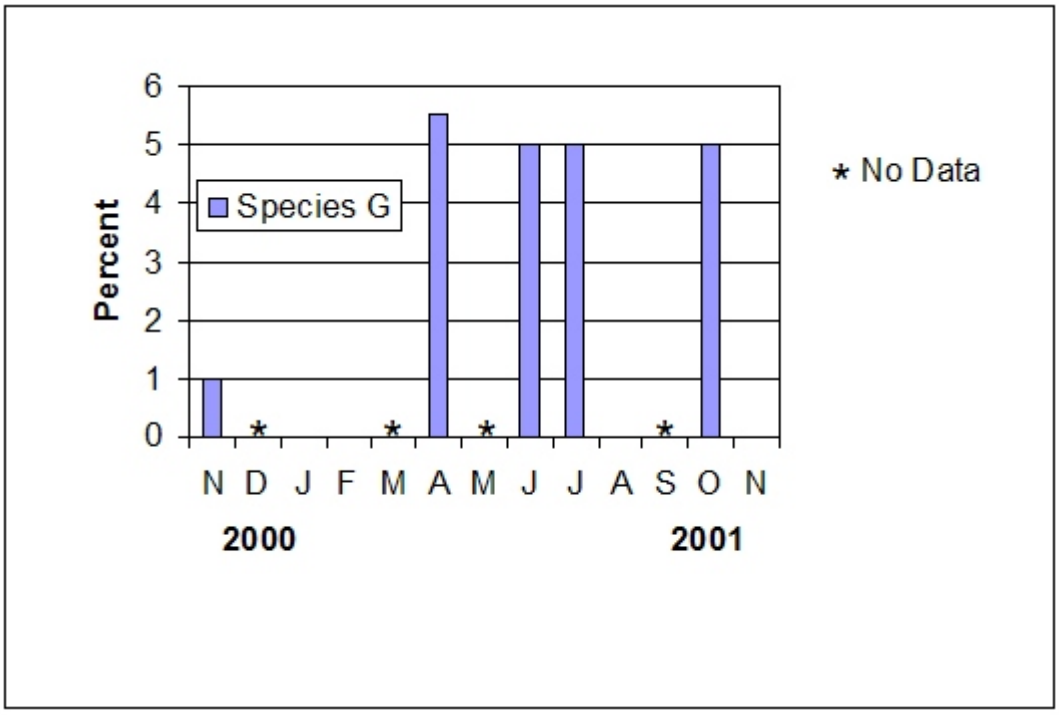

Figure 3.24 – Monthly prevalence for the external parasite species G, a copepod of the Family *Bomolochidae*, of the black sea bass.

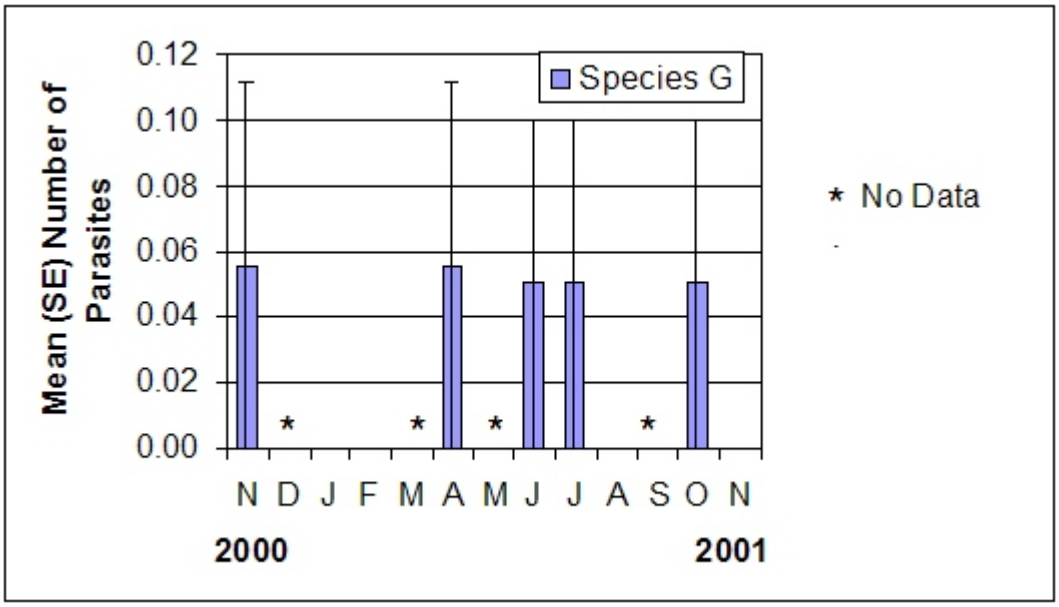

Figure 3.25 – Monthly mean number (±SE), versus time for the external parasite species H of the black sea bass identified, G = Copepod; Family *Bomolochidae*.

Figure 3.26 – Species H, a copepod of the genus *Argulus*

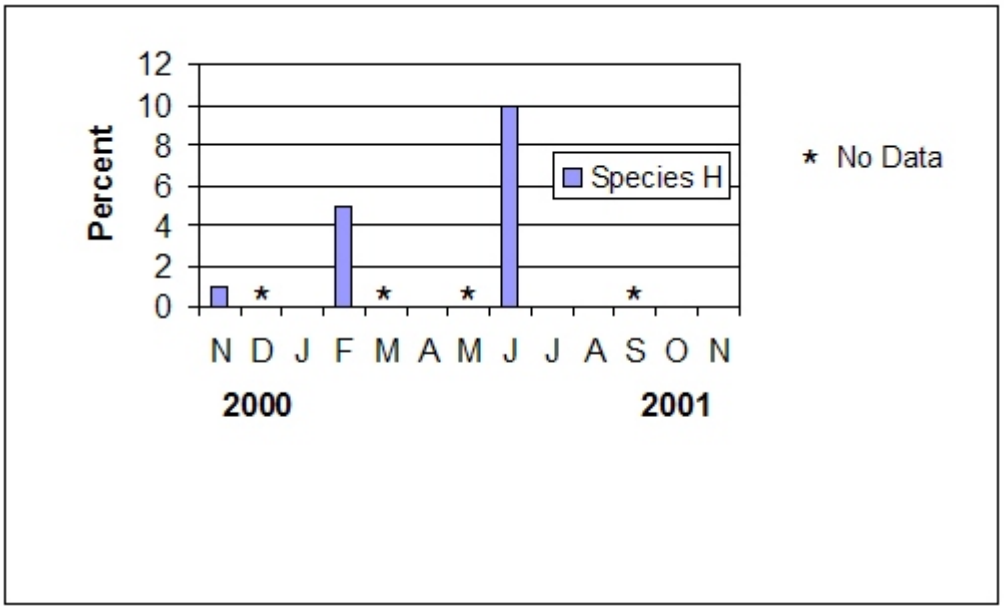

Figure 3.27 – Monthly prevalence for the external parasite species H, *Argulus*, of the black sea bass.

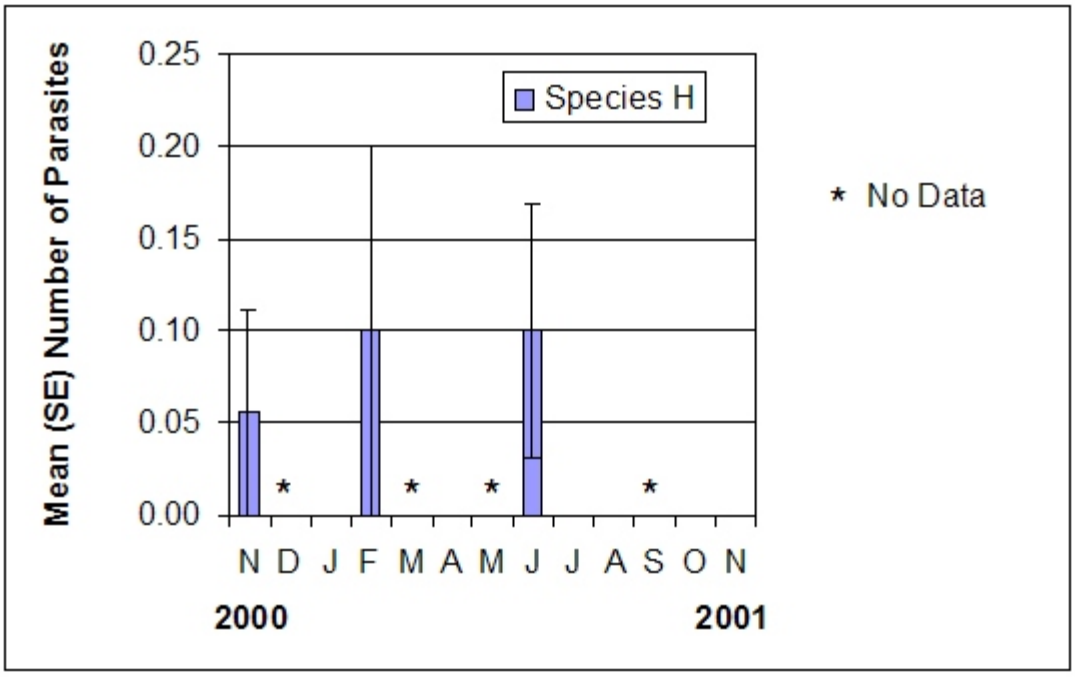

Figure 3.28 – Monthly mean number (±SE), versus time for the external parasite species H of the black sea bass identified, H = *Argulus*.

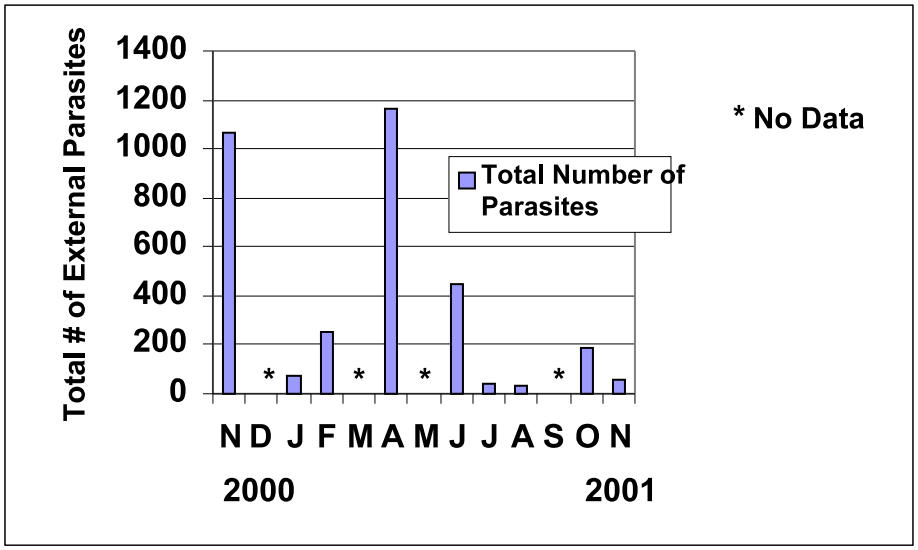

Figure 3.29 – The total number of external parasites found each month data was collected for the duration of the study, November 2000 – November 2001.

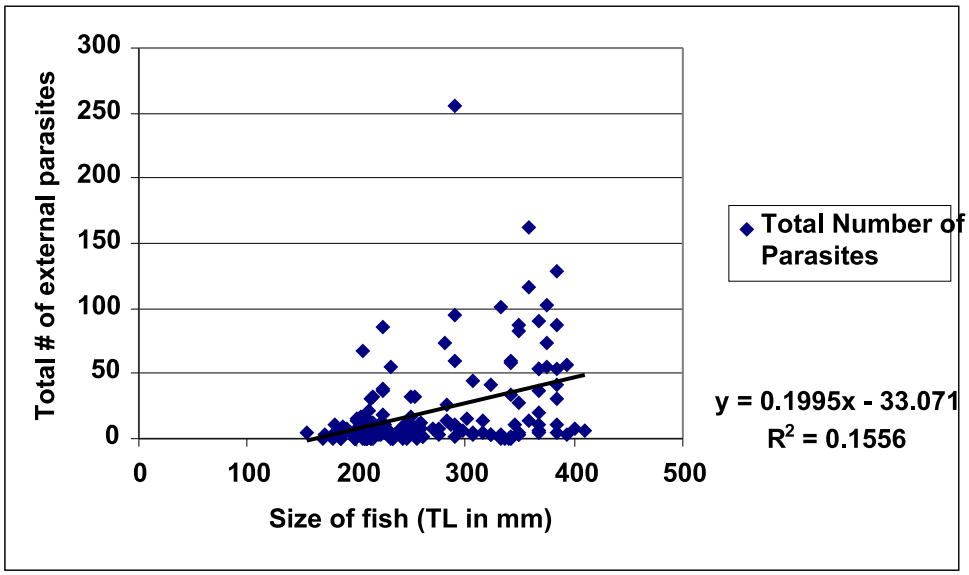

Figure 3.30 – A regression of total number of external parasites in relation to size (total length in mm) of fish.

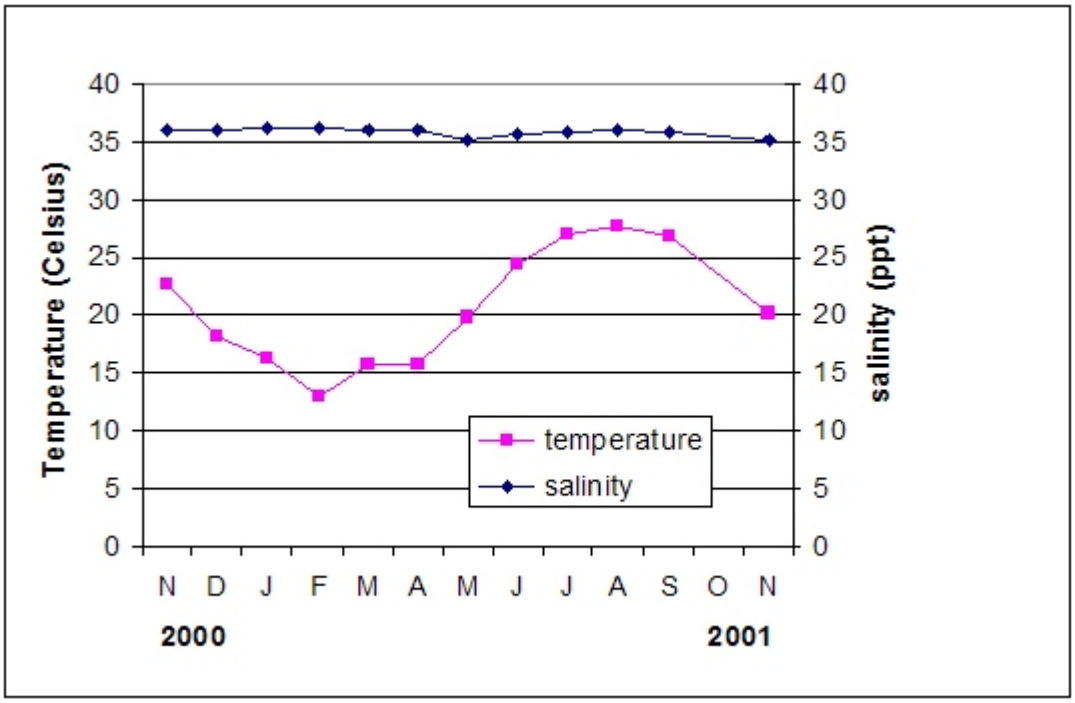

Figure 3.31 – Water temperature and salinity of the Skidaway Institute of Oceanography's SABSOON Tower Data (R2) for the duration of the parasite experiment, November 2000 – November 2001.

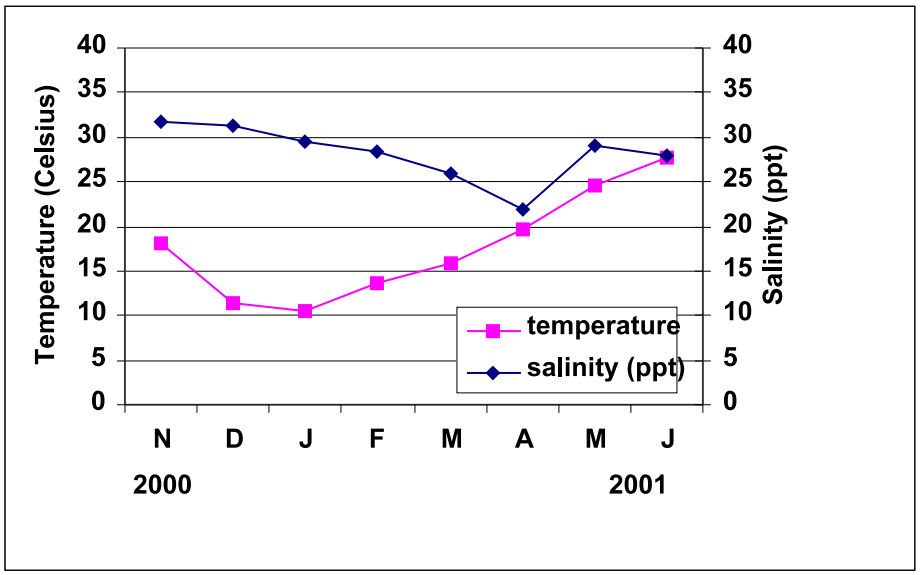

Figure 3.32 – Water temperature and salinity of the Sapelo Island, NERR Marsh Landing site on the Lower Duplin River from November 2000 – June 2001.

Table 3.2 – Multiple Comparisons of individual species against each month sampled.

Dependent Variable	(l)Date	(J)Date	Mean Difference(l-J)	Std. Error	Sig.
EXB	4	1	57.81*	8.387	.000
		2	49.93*	8.057	.000
		6	40.43*	8.057	.000
		7	60.13*	8.057	.000
		8	60.69*	8.387	.000
		10	53.38*	8.057	.000
		11	6.17	8.267	.998
		13	59.00*	8.267	.000
	1		51.64*	8.387	.000
	2		43.76*	8.057	.000
		4	6.17	8.267	.998
		6	34.26*	8.057	.001
		7	53.96*	8.057	.000
		8	54.52*	8.387	.000
		10	47.21*	8.057	.000
		13	52.83*	8.267	.000
EXD	8	1	.35*	.093	.006
		2	.41*	.089	.000
		4	.41*	.091	.000
		6	.41*	.089	.000
		7	.41*	.089	.000
		10	.36*	.089	.002
		11	.13	.091	.870
		13	.41*	.091	.000
	1		.22	.091	.294
	2		.28*	.088	.048
		4	.28	.090	.060
		6	.28*	.088	.048
		7	.28*	.088	.048
			-.13	.091	.870
		8	.23	.088	.198
		10	.28	.090	.060
		13			
INAA	2	1	.45*	.118	.007
		4	.45*	.117	.006
		5	.45*	.113	.004
		6	.45*	.113	.004
		7	.45*	.113	.004
		8	.45*	.118	.007
		10	.40*	.113	.019
		11	.28	.117	.314
		13	.28	.117	.314

CHAPTER 4

DISCUSSION

Life History of Parasites Identified

Parasites live in equilibrium with the communities of plants and animals that they inhabit. However, when unusual events occur in the environment, either natural or human induced, the equilibrium between parasites and the host is disturbed. An epizootic of one or more species can occur (Hoffman, 1967). This study of the black sea bass from Georgia revealed nine species of parasites that have the capability of causing epizootics. A brief life history of the parasites identified in this study is detailed below.

Nematodes

Species A, *Capillaria*, is a nematode, also known as a roundworm. There are about 650 species of nematodes that parasitize fish as either final or intermediate hosts. Nematodes are most common in wild caught fish, because most cultured fish are not exposed to either the adult nor larval nematodes in other hosts of the life cycle (Noga, 2000).

The sexes of nematodes are separate. Most nematodes that infect fish are oviparous, with females releasing the eggs in the water and hatching free-swimming larva. However, some are viviparous and live young are released from the female. In both cases, nematode larvae are ingested by an intermediate host, most likely a crustacean, which in turn is consumed by a fish. Once ingested in a fish, it either encysts or matures to an adult. Encysted larvae in fish must be ingested by a mammal, bird, or another fish as a final host (Rohde, 1984; Hoffman, 1967). Typically, there are four larval stages, followed by the adult of the nematode life cycle. In the fourth larval stage,

the definitive host is infectived. They maintain a hold on their hosts by using an apparatus called the spicule with a terminal claw. They have well-developed alimentary canals and no supporting skeleton of any strength (Kabata, 1985).

Adult nematodes are almost always located in the digestive tract, but some adults inhabit the peritoneal cavity, gonads, or swim bladder (Noga, 2000). In this study, the nematodes were found in the digestive tract. The body of *Capillaria* possesses a simple mouth, with or without bacillary bands. The esophagus is long and increases in size posteriorly. The life cycle of *Capillaria* is unknown (Hoffman, 1967).

Trematodes

There are two types of trematodes: digenean (more than one host) or monogenean (one host). Species AA, is an adult digenean fluke that was found in the digestive tract. The trematode life cycles are diverse and highly complex, involving one or more changes in the host. Trematodes are equipped with two sucker-like attachment organs, one anterior and one ventral (either sometimes absent). The body is unsegmented, flat and the gut is present (usually bifurcated). Digenetic trematodes are predominantly endoparasites. The life cycle of the digenetic trematode involve more than one host (often three) and include several biologically and morphologically dissimilar stages. "The most elaborate type of cycle includes eggs, miracidia, sporocysts, cercariae, metacercariae, as well as the adult. However, one or more of these stages are left out in most cycles (Kabata, 1985).

Fish serve either as second intermediate or as definitive hosts of trematodes. Where fish are the intermediate host, the trematodes are encysted in various tissues and organs. Only if the fish or invertebrate is ingested by the definitive host can the cycle be completed and the next generation of adults produced. As a definitive host, fish harbour adult worms ingested with the second intermediate host. Adult trematodes are often found in the lumen of the alimentary canal. They attach by their acetabula, but are mobile; however, in post-mortem examinations, they are commonly found detached.

Identification of trematodes requires examination of their internal organs; larval stages are often difficult to identify (Kabata, 1985).

Copepods

Lernaea spp. are among the most harmful fish parasites because of its relatively large size and its mode of feeding and attachment. These copepods undergo a profound metamorphosis following attachment to fish. When they metamorphose, they assume a vermiform shape with an anterior holdfast organ. This organ allows these parasites to bury its anterior end into the host tissue. Some 40 species of *Lernea* have been described, mainly based on the shape of the hold fast organ; however, the shape of the hold fast is largely determined by the consistency of the fish tissues (Kabata, 1985).

There are a total of eleven developmental stages in the life cycle of *Lernaea* (Kabata, 1985). Specimen B and C are both in the suborder *Cyclopoida*, most likely family *Lernaeidae*. Species B is in a copepodid stage and possesses a paired eye structure in the center anterior half. Species C, identical to species B but in a different developmental stage, appears to be a mature premetamorphosis female. Some of the features of the pre-metamorphosis female include a truncated posterior end, rounded anterior margin, a cephalothorax oval, and a paired eye in the center of anterior half. According to Kabata (1979), there are two host cycles to complete the life cycle. The intermediate host is where the copepodid stage and premetamorphosis stage of the life cycle are completed.

Species D (*Caligus elongatus*) belongs to the family Caligidae. *C. elongatus* can cause major problems in cultured and wild populations. Temperature seems to determine the infestation levels of *C. elongates*. They cause skin erosion and if heavily infected, fish often die (Lester and Roubal, 1995). *C. elongatus* has been found on over 80 species of fish in 17 orders and 43 families. Hogans and Trudeau (1989a) found that the generation time for cultured *C. elongatus* was 5 weeks at 10 C. Stuart (1990) found that this parasite leaves the host at temperatures lower than 6 C. Wootten et al. (1982)

responded that relatively few copepods seem to mature and it is not known whether the parasite dies or adult parasites enter the plankton.

Sexual dimorphism exists in adult caligids; the female is larger than the male. During copulation, male appendages, particularly the first maxillae and second antennae, are modified for attachment (Lester and Roubal, 1995). The adult males of *C. elongatus* are about 5 mm long and have a slimmer genital segment. Females are 6-8 mm long. The parasite is a golden-brown or yellow color (Hogans and Trudeau, 1989b). The female egg sac contains about 30 eggs each (Mackinnon, 1992). Kabata (1972) proposed that five phases and ten stages occurred during the life cycle of caligid copepods. The copepodid stage of *C. elongatus* lasts 50 hours at a temperature of 30ºC. The N1 stage lasts 15-30 hours and 35 hours for the N2 stage, both at a temperature of 10ºC, respectively. In addition, there are no moults below 3ºC (Hogans and Trudeau, 1989b).

Adults and larvae of *Caligus elongatus* attach to most areas of the body of salmon, but adults prefer the lateral and dorsal surfaces of the head, between the opercula, and the base of the caudal fin (Hogans and Trudeau, 1989b). Other sites of attachment include the ventrolateral surfaces near the pelvic fin, the base of the dorsal fin and around the anus. The parasite also leaves a 'grazing trail' over the surface of the fish from the point of origin during attachment to its 'preferred site' of attachment (Hogans and Trudeau, 1989a).

Species E, F, and G belong to the family *Bomolochidae*. The structure of the bomolochid is segmented and greatly resembles the taeniacanthids. The segmentation has been clearly preserved during the process of adaptation to parasitism. There are 11 bomolochid genera. There are three types of facies, linked by intermediate forms, in aspect to the dorsal. These include: the "cyclopoid", "elongate", and "atracolax" facies (Kabata, 1979). The nearly 110 species of Bomolochidae are exclusively marine. Its parasitic members predominantly attach on teleosts. They are small in character, females rarely surpassing a length of 2 mm. As ectoparasites, they are fairly mobile. Though

they are normally restricted to defined habitats such as the inner surface of the operculum and surface of the eyeball, they are capable of moving over the surface of the host. The mode of feeding is unknown, though, presumed to be a browser, feeding off the mucus and superficial epithelial debris. Attachment is mainly by suction, however, the second antennae and the maxillipeds might play a role in auxiliary attachment organs (Kinne, 1984).

Branchiura

The species labeled as H belongs to the genus *Argulus* Muller, 1785. There are about 150 species of Branchiura; about 100 of them are in the genus *Argulus*. *Argulus* has both marine and freshwater species worldwide. These species are poorly known and are not easily identified (Kabata, 1985).

The body of *Argulus* has three regions: cephalothorax, thorax and abdomen. Its flattened dorsoventral plane is covered by a broad dorsal shield. The shape of the shield varies with sex and age. The most obvious appendage of *Argulus* is the sucker. The sucker is a modification of the first maxilla and is adapted to prehension and sliding over the host. There is a lower occurrence of male parasites compared to females (Kabata, 1985).

Without a host, *Argulus* cannot survive for a prolong period of time, however, it can freely swim to search for an alternate host. Female parasites leave the host to search for suitable objects to deposit its eggs. Eggs are deposited in clusters consisting of several egg strips arranged in parallel rows. The larvae that emerge are at the copepodid stage, or the larva I stage; there are six larval stages (Kabata, 1985).

The harmful effects of *Argulus* range from no ill effects to death. Injuries to the host caused by *Argulus* may include the point of attachment, but also more extensive injuries result from its feeding activities (Kabata, 1970). *Argulus'* feeding habits involves secretion and injection of large quantities of digestive fluids. In addition, tissues are torn by the buccal apparatus and attachment appendages. Prolonged attachment

causes extensive pathological changes in the skin such as red coloration or raised epithelium (Kabata, 1970). Ulcerations occur around the feeding area and epithelial mucous cells increase. The highly toxic secretion of the buccal glands causes an inflammatory response. The scales are loosened due to cytolytic activity that can lead to their loss. The toxins secreted into the fish can be defined as poisoning and the intensity of the effect depends on the fish size, the injection site, and amount of toxin secreted. *Argulus* can promote secondary infections, and can act as a vector for flagellates, viruses, and bacteria (Kabata, 1985).

Histology

A small study also was performed to evaluate the microscopic presence of parasitism and associated tissue lesions. Parasites were found in the gills, intestinal tract, mesentery and gastric wall. There were two types of trematodes found internally, adult and encysted flukes. The adult flukes were digenean.

Copepods were the only external parasites revealed. A histological section of the gills illustrate a copepod juxapositioned between two primary lamellae with subsequent attachment to the secondary lamellae (fig 4.1). The gill arch is made up of primary and secondary lamellae, which serves for respiration. Teleosts obtain oxygen from water through diffusion via the gills. The gills are located along the pharyngobranchial bones of the gill arch. These arches are situated within the pharyngeal cavity and are enclosed by opercula, which protect the gills and regulate water flow (Groman, 1982). This species, a copepodid stage of the copepods, is identical to species B of the gross parasite study (fig 4.2).

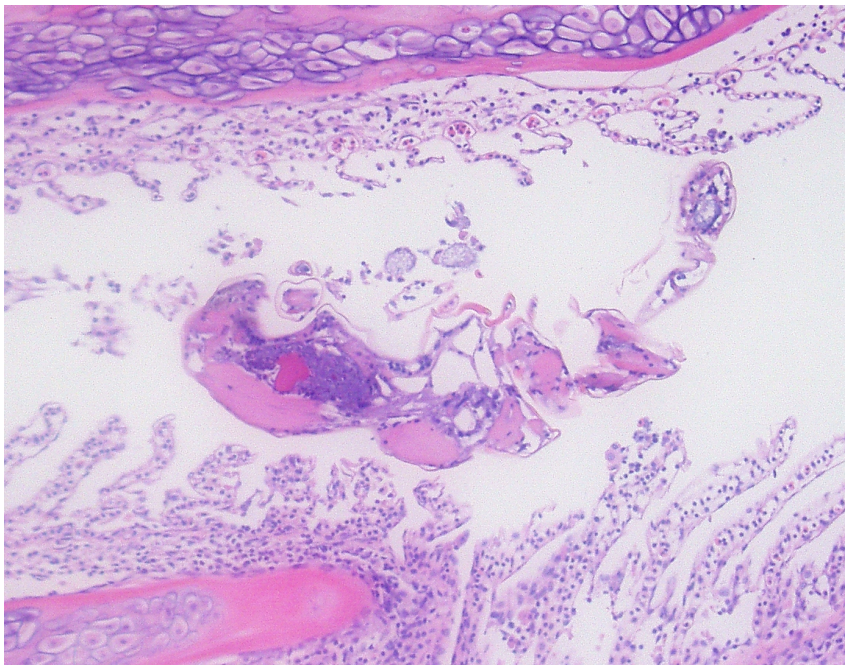

Figure 4.1 – A histological section of a copepod between two sets of primary lamellae

Figure 4.2 – A copepod (copepodid stage)

The adult digenean trematodes (fig 4.3) were found in the intestinal tract of the black sea bass. The ventral sucker, noted to be much larger than usually observed in other species, is used for attachment to the host and is located on the anterior portion of the parasite. A histological section of an adult fluke is depicted in figure 4.4. Figure 4.5 is another histological crossection of an adult digenean trematode found abnormally in the liver of the fish.

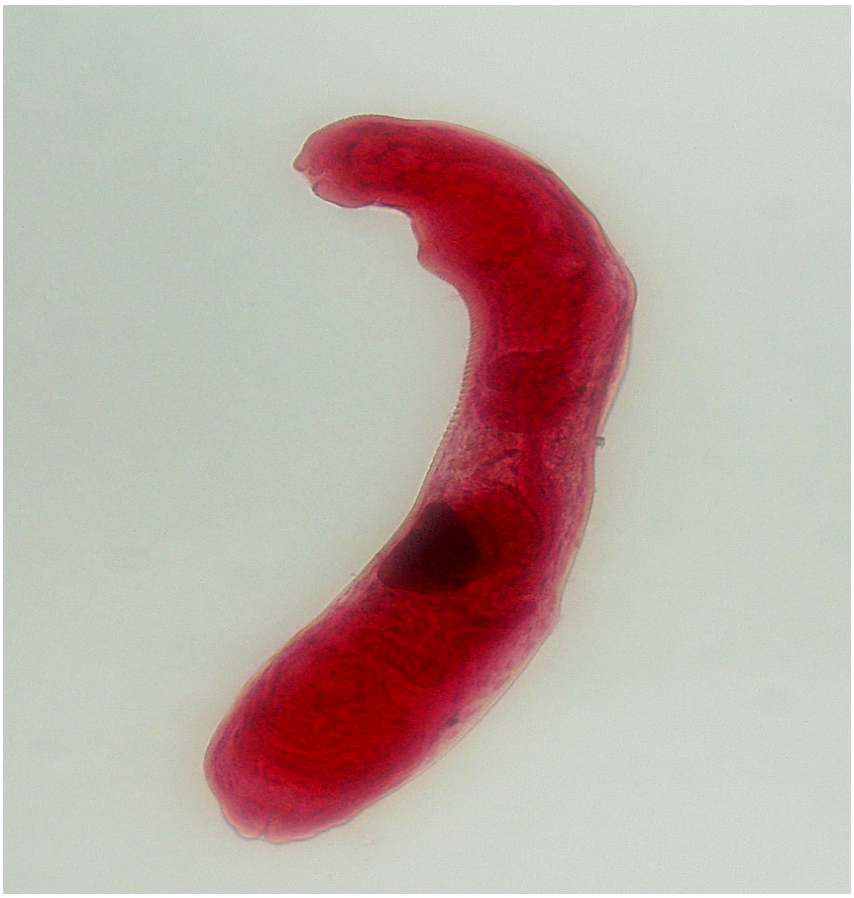

Figure 4.3 - A digenean trematode

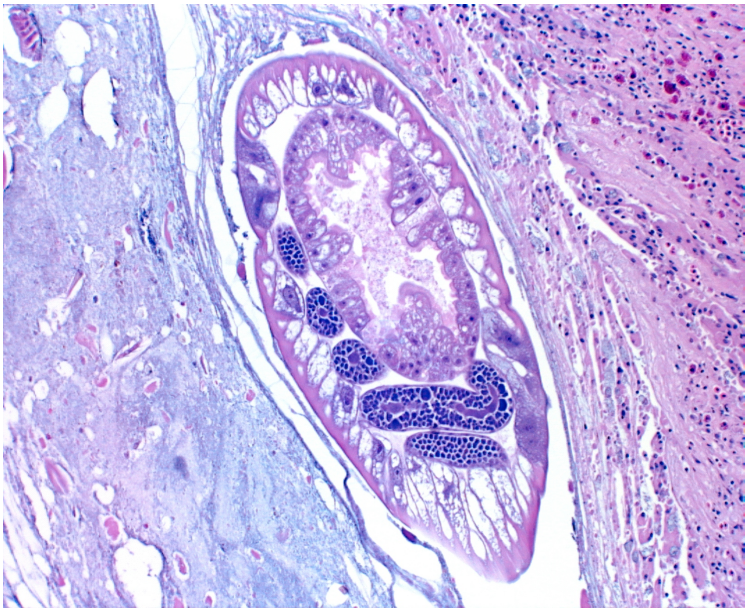

Figure 4.4 – (In order from left to right) A histological section of ingesta, an adult digenean fluke, and the gastric wall.

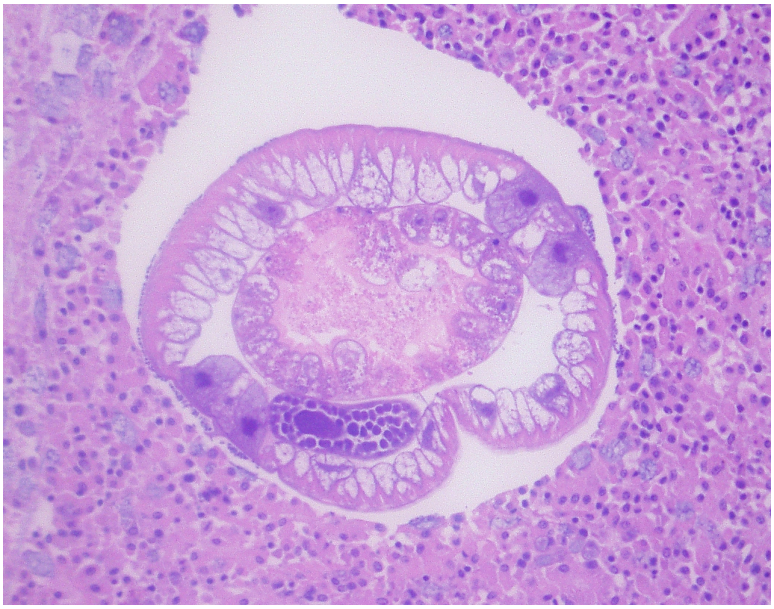

Figure 4.5 – A histology section of an adult fluke in the liver.

In addition to the adult digenean trematodes, metacercariae or encysted flukes were also found by histologic examination. A histological section of encysted trematodes surrounded by mesenteric fat is depicted in figure 4.6. Most fish acquire metacercariae by being attacked by the cercariae stage that penetrates the skin and migrates to its target sites. Fish or small invertebrates may act as a second intermediate hosts or definitive hosts for this parasite. The metacercarial stage encysts in various tissues and organs of their host and only when the intermediate host is eaten by the definitive host can the life cycle be completed (Kabata, 1985). Metacercariae have most characteristics of adult digeans, yet mature reproductive organs are usually absent. Since the shape and size of the reproductive organs are used for identification of species, it is extremely difficult to key metacercariae to species (Noga, 2000).

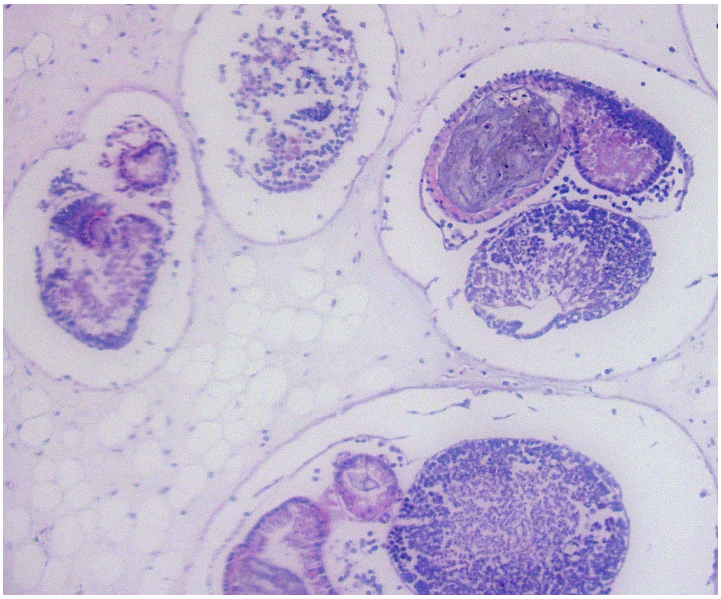

Figure 4.6 – A histological section encysted metasacarian flukes in mesenteric fat.

Last, granulomas were detected in the gastric wall of the black sea bass. A granuloma is a chronic inflammatory response due to foreign bodies, bacterial and fungal infections, or parasites. A granuloma consists of a coarse granular eosinophilic core surrounded by macrophages and multinucleate giant cells. It is encased by fibroblasts and collagen, which encapsulate and isolate the material (fig 4.7). There are several types of special stains used to detect pathogens and material within the granuloma. These include: giemsa stain for the general detection of bacteria in general, acid-fast stain for the specific detection of mycobacteria, and the periodic acid-schiff (PAS) technique to detect fungi. All three staining procedures were performed in the histologic examination with negative results. Histologically, parasites, bacteria, and fungi could not be ascertained as the immediate injury resulting in granuloma formation.

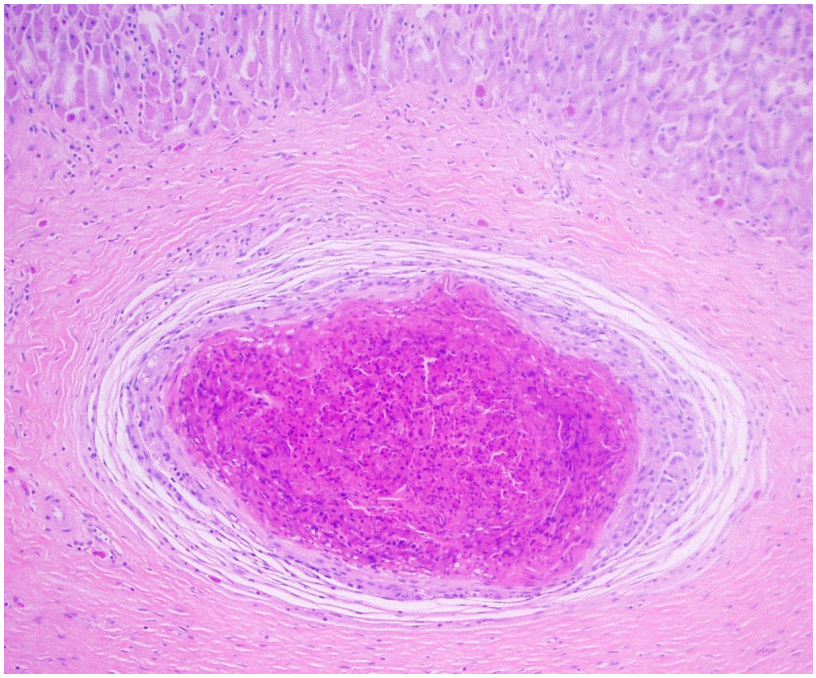

Figure 4.7 – A histological section of a granuloma encapsuled by the gastric wall.

Results from Other Black Sea Bass Studies

The results of this study indicate that internal and external parasites are prevalent on the black sea bass, *Centropristis striata*. This is not in consistent with past studies of the black sea bass. Linton (1901) found various acanthocephalans, cestodes, and nematodes, encysted in the walls of the digestive tract. Cupka et al. (1973) found that the black sea bass are generally free of external parasites. However, Cupka et al. (1973) did report that two fish were found with a single isopod, *Nerocila*, attached to the caudal region. In addition, another fish had an ophichthid eel in its coelomic cavity. Cupka et al. (1973) also reported that other authors recorded snake eels as pseudoparasites in various demersal fish species. Breder (1953) recorded an ophichthid eel, *Omochelys cruentifer*, from the coelom of a black sea bass.

Related Marine Fish

The gray snapper (*Lutjanus griseus*) has been reported to have many endoparasites and ectoparasites. According to Starck and Schroeder (1971), a number of trematodes, one acanthocephalan worm, three nematodes and one cestode have been reported as endoparasites. Four ectoparasites have been recorded: one isopod and three copepods (Starck and Schroeder, 1971).

A biological review on the Nassau Grouper, *Epinephelus striatus*, reported that parasites occurred predominantly in the viscera and gonads. A reddish-brown nematode occurred in the gonads and encysted larval tapeworms are prevalent in the viscera. Parasitic isopods were also identified in the nostrils of the fish. In addition, the digenetic trematodes *Lecithochirum parvum* and *L. microstomum* (from the stomach), *Helicometra torta* (found in the pyloric caeca), and *Sterrhurus musculus* (also found in the stomach) were identified in fish caught in Florida (Sadovy and Eklund, 1999).

Treatments to Prevent Transmission

Parasites that utilize intermediate hosts in their life cycles may be controlled, particularly in aquaculture conditions. By preventing the access of definitive or intermediate hosts to the water or by eliminating these hosts, parasites can be prevented (Dick and Choudhury, 1995). These intermediate and final hosts are usually a specific snail or fish-eating birds (Noga, 2000).

Another treatment of parasites is the use of drugs. For many drugs, a range of doses are given. Water quality can greatly affect efficacy and ichthyotoxicity for water-borne treatments. When utilizing pharmacological means, it is best to start at the lower recommended dose and repeat with a higher dose if the disease is unresponsive. Dosages vary with feed intake for oral medications. For example, fish eating less require a higher drug dosage. However, there are legal limitations and restrictions on the permissible dosage of a drug if approved. FDA regulates the use of drugs in all fish in the United States, whether for human consumption or other purposes. There are some drugs that are not approved for treating food fish in the United States, such as Malachite green, a parasiticide. In addition, specific drugs (i.e., formalin, oxytetracycline) are approved for treating certain pathogens and particular fish, yet these drugs require research before use. For example, Paraziquantel orals, injections, and baths are used to treat digenean trematode, cestode, and monogenean infections while anthelminthics can control adult nematodes (Noga, 2000).

Treatment of infected fish is best done in a separate “isolation” tank to remove contagions from susceptible fish and avoid exposing healthy fish to unnecessary and potentially toxic medications. Also, a smaller amount of drug can be used in a smaller “hospitalization” tank. However, the capturing of fish and foreign environments can also stress fish (Noga, 2000).

Chemotherapeutics such as formaldehyde, carbaryl, organophosphate insecticides, pyrethrum, and hydrogen peroxide are marginally effective against sea lice. Insecticides are also commonly used to control fish with argulosis. Anesthesia of infected broodstock or removal of parasites with forceps is another measure of control (Lester and Roubal, 1995).

The use of oral medications is one of the best ways to treat fish because it is least stressful and very effective if consumed in the proper dosage. However, sick fish have a propensity to decrease oral consumption and some commercially prepared oral medications are not available. An example of an oral medication is piperazine sulfate, which eliminates nonencysted nematodes in the gastrointestinal tract. The use of injectable medications is another route of administration. The advantage is delivering a precise dosage while the disadvantages include stress imposed by capturing the fish and handling of the fish. Levamisole hydrochloride is an example of an injectable drug used for treating nonencysted nematodes (Noga, 2000).

Examine all wild fish for internal and external parasites before stocking. Fresh water baths are also known to eliminate marine parasites, particularly *Trichodina* and other protozoans, Monogenea, and some crustaceans. Other chemical baths, where fish are exposed to a concentrated drug solution for a short time, are another type of treatment. These water-borne treatments are best used for external parasites dwelling on the skin and gills of the fish. For example, formalin is commonly used as an effective parasiticide for most ectoparasitic protozoa and monogeneans, but can be irritating to the gills and water should be well aerated during treatment. Organophosphate is another pharamalogical treatment of monogeneans, leeches, and crustacean ectoparasites (copepods, branchiurans, and isopods). All drugs must be completely mixed and dissolved before adding fish. For fish that are weak or sensitive, multiple treatments at the lower dosage are best.

Some parasites cannot be treated. For instance, there are no proven treatments for encapsulated forms of nematode larvae or encysted nematodes. Also, cestode larvae have no proven treatment regimen (Noga, 2000).

From an aquacultural perspective, prevention and control of infestation is imperative. A short and long-term plan is vital in maintaining this imperative. The short-term plan should include the means to control the immediate problem (usually involving various medications to control the infections). The long-term plan is management for prevention and is the most important. Water quality is the most important step in preventing and controlling parasites. These parameters include proper levels of dissolved oxygen (DO), ammonia, temperature, nitrite, pH, hardness, and salinity. These measurements can be taken using commercially available water quality test kits. If possible, disinfection and quarantine of wild fish before stocking is the best safety measure (Noga, 2000).

In conclusion, nine species of parasites were found in this study, two internal and seven external. These species include trematodes, nematodes, copepods, and branchiurans. However, in an aquacultural setting, nematodes and trematodes tend not to be a problem due to the fact that the intermediate hosts are missing to complete the life cycles. The primary problem area is the external parasites which include the copepods and the branchiurans, because they have a direct life cycle. Hence, prevention should be focused on external parasites.

WORKS CITED

Ackefors, H., J. V. Huner, and M. Konikoff. *Introduction to the General Principles of Aquaculture*. Food Products Press. New York. 1994.

Anders, K. and H. Möller. *Diseases and Parasites of Marine Fishes*. Kiel: Moller. Kiel, FRG. 1986.

Ansley, H. L. H. and D. Harris. *Migration and standing stock of fish associated with artificial and natural reefs on Georgia's outer continental shelf.* Georgia Department of Natural Resources Final Report, Project Number F-31-4. 1981.

Beverly-Burton: *Parasitic Diseases of Fishes Ch. XI: Monogenean Diseases. In Blue Book: Suggested Procedures for the Detection and Identification of Certain Finfish and Shellfish Pathogens.* John C. Thoesen. 4th ed. Version 1. Fish Health Section, American Fisheries Society. 1994.

Breder, C. M. "An ophichthid eel in the coelom of a sea bass." Zoologica, N.Y. 1953: 38: 201-203.

Chester, A. J., G. R. Huntsman, P. A. Tester, and C. S. Manooch III. "South Atlantic Bight reef fish communities as represented in hook-and-line catches." Bulletin of Marine Science 34 (2): 1984: 267-279.

Collette, B.B., Facey, D.E., and G.S. Helfman. *The Diversity of Life*. Blackwell Science, Inc. 1997.

Cotton, Charles. "Optimizing Growth for Aquaculture of Juvenile Black Sea Bass Centropristis Striata L.: Effects of Temperature, Salinity, Commercial Diet and Feeding Ration." Masters Thesis. University of Georgia. 2002. 83 pp.

Cupka, D. M., R. K. Dias and J. Tucker. "Biology of the black sea bass, *Centropristis striata* (Pisces: Serranidae), from South Carolina waters." South Carolina Wildlife and Marine Resources Department. Charleston**,** South Carolina. 1973. 93 pp.

Dawe, D.L. and H.W. Dickerson. "*Ichthyophthirius multifiliis* and *Cryptocaryon irritans* (Phylum Ciliophora). In Fish Diseases and Disorders, Vol 1: Protozoan and Metazoan Infections." P.T.K. Woo. CAB INTERNATIONAL. Wallingford, UK. 1995. 181-227 pp.

Dick, T.A. and A. Choudhury. "Phylum Nematoda." In Fish Diseases and Disorders, Vol 1: Protozoan and Metazoan Infections. P.T.K. Woo, CAB INTERNATIONAL. Wallingford, UK. 1995. 415 – 446 pp.

Egusa, S. *Infectious Diseases of Fish*. A.A. Balkema Publishers**.** Brookfield, USA. 1992.

Furse, J. B., Jr. "Seasonal Variations In The Population Structure Of Black Sea Bass, *Centropristis Striata*, On Gray's Reef National Marine Sanctuary." Masters Thesis. University of Georgia. 1995. 80 pp.

Grabda, J. *Marine Fish Parasitology: An Outline*. Polish Scientific Publishers, Warszawa. 1989.

Groman, D. B. Histology of the Striped Bass. American Fisheries Society. Monograph No. 3. Bethesda, Maryland. 1982.

Grosslin, M. D. and T. R. Azarovitz. "Fish distribution. MESA New York Atlas Monograph 15." New York Sea Grant Institute. Albany, New York. 1982. 80, 82-83pp.

Hoff, F. H., Jr. "Artificial spawning of black sea bass, *Centropristes striatus melanus* Ginsburg, aided by chorionic gonadiotrophic hormones." Florida Department of Natural Resources Special Scientific Report No. 25, St. Petersburg, Florida. 1970. 17pp.

Hoffman, G. L. "Parasites of North American Freshwater Fishes." University of California Press. Los Angeles, CA. 1967.

Hogans, W. E. and D.J. Trudeau "*Caligus elongatus* (Copepoda: Caligoida) from Atlantic salmon (*Salmo salar*) cultured in marine waters of the lower Bay of Fundy." *Canadian Journal of Zoology.* 1989a. 67:1080-1082.

Hogans, W.E. and D.J. Trudeau "Preliminary studies on the biology of sea lice, *Caligus elongatus, Caligus curtus* and *Lepeophtheirus salmonis* (Copepoda: Caligoida) parasitic on cage-cultured salmonids in the lower Bay of Fundy." *Canadian Technical Report of Fisheries and Aquatic Sciences.* 1989b. 1715, 1-14.

Johnson, C., J. Harding, and J. Whitted. *Fishing guide to offshore Georgia: Gray's Reef National Marine Sanctuary.* University of Georgia Marine Extension Service. Savannah, Georgia. 1985.

Kabata, Z. *Diseases of Fishes, Book 1: Crustacea as Enemies of Fishes.* T.F.H.: Neptune City, NJ. 1970. 171 pp.

Kabata, Z. "Developmenal stages of *Caligus clemensi* (Copepoda: Caligidae)." *Journal of the Fisheries Research Board of Canada. 1972*. 29:1571-1593.

Kabata, Z. *Parasitic Copepoda of British Fishes*. The Ray Society. London, England. 1979.

Kabata, Z. *Parasites and diseases of cultured fish in the tropics***.** Taylor & Francis, London. 1985.

Kendall, A. W. Jr. "Biological and fisheries data on black sea bass, *Centropristis striata* (Linnaeus)." National Marine Fisheries Service-National Oceanic and Atmospheric Administration Technical Service Report #7. Highlands, N.J. 1977. 29 pp.

Kimble, C. E. *Aquaculture: Public Health, Regulatory, and Management Aspects***.** 2nd ed. Zarcon Press. Silver Spring, Maryland. 1985.

Kinne, O. *Diseases of Marine Animals*. *Vol IV, Part 1: Introduction*. Pisces. Biologische Anstalt Helgoland, Hamburg. 1984.

Lavenda, N. "Sexual differences and normal protogynous hermaphroditism in the Atlantic sea bass, *Centropristes striatus*." Copeia. 1949. (3):185-194.

Lester, R.J.G. and F.R. Roubal: "Phylum Arthropoda. In *Fish Diseases and Disorders*, Vol 1: Protozoan and Metazoan Infections." P.T.K. Woo, CAB INTERNATIONAL. Wallingford, UK. 1995. pp. 475-598.

Link, G.W., Jr. "Age, growth, reproduction, feeding and ecological observations on the three species of *Centropristis* (Pisces: Serranidae) in North Carolina waters." Ph.D. Dissertation: University of North Carolina**.** Chapel Hill, North Carolina. 1980. 277 pp.

Linton, E. "Fish parasites collected at Woods Hole in 1898." Bull. U.S. Fish Comm. (1899) 1901. 19: 267-304, pls. 33-43.

Low, R. A., Jr. "Mortality rates and management strategies for black sea bass off the southeast coast of the United States." North American Journal of Fisheries Management 1981. 1:93-103.

Low, R. A., Jr. "The South Carolina fishery for black sea bass (*Centropristis striata*), 1977-1981." South Carolina Wildlife and Marine Resources Center Technical Report 53**.** Charleston, South Carolina. 1982. 17pp.

Marine Extension Service. The University of Georgia, Personal Interview. 2001.

Mackinnon, B.M. "Egg production in sea lice, *Caligus elongatus."* Bulletin of the Canadian Society for Zoology 1992. 23, 79.

Mercer, L. P. "Species profiles: life histories and environmental requirements of coastal fishes and invertebrates (South Atlantic)--black sea bass." United States Fish and Wildlife Service Biological Report 82 (11.99). U.S. Army Corps of Engineers, TR EL-82-4. 1989. 16 pp.

Miller, R. J. "A review of the seabasses of the genus *Centropristes* (Serranidae)." Tulane State Zoology 1959. 7(2) 35-68.

Musick, J. A., and L. P. Mercer. "Seasonal distribution of black sea bass, *Centropristis striata*, in the mid-Atlantic Bight with comments on the ecology and fisheries of the species." Transactions of the American Fisheries Society 106 (1): 1977. 12-25.

National Marine Fisheries Service, Fisheries Statistics and Economics Division. Personal Interview. Silver Spring, Maryland.

Noga, E. J. *Fish Disease: Diagnosis and Treatment*. Iowa State University Press. Ames, Iowa. 2000.

Overstreet, R.M. "Marine Maladies? Worms, Germs, and Other Symbionts From the Northern Gulf of Mexico." Mississippi-Alabama Sea Grant Consortium and R.M. Overstreet. 1978. 140pp.

Post, G.W. *Textbook of Fish Health.* TFH Publication, Inc. Ltd. Neptune City, New Jersey. 1983.

Reinboth, R. Sex reversal in the black sea bass *Centropristes striatus*. Anat. Rec. 1965. 151 : 403.

Rohde, K. "Diseases caused by metazoans: Helminths. In Kinne O, editor: *Diseases of Marine Animals*, vol 4, Hamburg, Germany, Pisces, Biological Anstalt Helgoland, 1984. pp. 193-320.

Sadovy, Y. and A.-M. Eklund. "Synopsis of Biological Data on the Nassau Grouper, *Epinephelus striatus* (Bloch, 1792), and the Jewfish, *E. itajara* (Lichtenstein, 1822). NOAA Technical Report NMFS 146." A Technical Report of the *Fishery Bulletin*; FAO Fisheries Synopsis 157. Seattle, Washington. 1999.

Schwartz, F. J. "Effects of winter water conditions on fifteen species of captive marine fishes." Am. Midl. Nat. 1964. 71:434-444.

South Atlantic Fishery Management Council. "Amendment number 1 and environmental assessment and regulatory impact review to the fishery management plan for the snapper-grouper fishery of the South Atlantic region." South Atlantic Fishery Management Council. National Marine Fishery Service: 1988. 46.

Starck II, W. A. and R. E. Schroeder. "Stud. Trop. Oceanogr." Miami 10: 1971. 224 pg., November, 1970.

Struhsaker, P. "Demersal fish resources: composition, distribution, and commercial potential of the continental shelf stocks off southeastern United States." Fisheries Independent Research 4. 1969. (7):261-300.

Stuart, R. "Sea lice, a maritime perspective." Bulletin of the Aquaculture Association of Canada. 1990. 90:18-24.

Waltz, W., W.A. Roumillat, and P.K. Ashe. "Distribution, age structure, and sex composition of the black sea bass, *Centropristis striata*, sampled along the southeastern coast of the United States." South Carolina Marine Resources Center Technical Report No. 43. Charleston, South Carolina. 1979. 18pp.

Wooten, R., J.W. Smith and E.A. Needham. "Aspects of the biology of the parasitic copepods *Lepeophtheirus salmonis* and *Caligus elongatus* on farmed salmonids, and their treatment." *Proceedings of the Royal Society of Edinburgh* 81B (1982): 185-197.