

DISEASE STATUS OF *Clarias gariepinus* (Burchell, 1822) AND SOME FISH PONDS IN ASABA, NIGERIA

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Abstract

The disease status of Clarias gariepinus and some fish ponds in Asaba, were investigated. Fish and water samples were collected three times weekly from some fish ponds for a period of twelve weeks between May and July, 2012. Skin scrapping of fresh fish samples for external parasites was carried out and mucus obtained examined for parasites. Smears were stained while bacteria and fungi were isolated for confirmation. Nine parasites were identified in the fish ponds surveyed and these include Ichthyobodo, Trichodina, Tricophyra, Cryptobia, Chilodonella, Gyrodactylus, Nematode (Capillaria), Glochidium and Piscicola. The number of fish infested with parasites in all the fish ponds surveyed were not significantly ($P>0.05$) different. Bacteria isolated from the fish include Flexibacter, Escherichia coli and Staphylococcus. Five different fungi species were identified in the fish. These are Saprolegnia, Ichthyophonus, Candida albican, Achyla, and Aspergillus fumigatus. Bacteria and fungi isolated from the fish ponds differed significantly ($P<0.05$). Water quality parameters of fish ponds surveyed show that there were on significant ($P>0.05$) difference in the values obtained for temperature and pH. The disease status of fish and fish ponds in Asaba shows that fish and fish ponds are not free of pathogenic agents. All the fish examined and most fish ponds surveyed had pathogenic agents. Caution and routine checks of current status and prevalence of disease agents in fish ponds is desirable to forestall any future outbreaks of fish diseases.

Keywords: Disease status, *C. gariepinus*, fish ponds, Asaba.

Introduction

Fish is an excellent and relatively cheap source of animal protein. Fish production through

aquaculture is one of the fastest growing food producing sectors, providing an acceptable supplement and substitute to wild fish. Waseem (2007) reported that fish is important in bridging the gap between the demand and supply of protein because of its multifarious economic advantages and nutritional significance. Fish like any other animal are subject to diseases and as such require adequate health care. Diseases are considered as the most limiting factor in aquaculture production.

Disease is a great threat to profitable fish production (Ibiwoye, 2002). Fish diseases have been classified as infectious which fish can acquire from the environment and non-infectious which are normally due to deficiencies in nutritional requirements of fish. Infectious diseases are caused by pathogenic organisms (parasites, bacteria, virus and fungi) present in the environment. It has been estimated that 10% of all cultured aquatic animals are lost as a result of infectious disease (Leong and Fryer, 1993). Environmental circumstances such as poor water quality, fluctuations in temperature, poor nutrition, overcrowding, poor handling and transportation which are common in intensive fish farming poses stressful conditions to the fish making the fish more susceptible to a wide variety of pathogens. Francis- Floyd (2005) noted that with diseases fish is not in balance with itself or with its environment. Stress in fish results in poor feeding, deformity and cannibalism, reduced growth and survival rate. All these predispose the fish to infection and disease leading to a reduction in fish health status and eventual mortality.

In view of the recent preponderance of fish ponds in Asaba owned and managed by mostly unskilled personnel, this study examines the disease status of *C. gariepinus* (one of the most cultured catfishes in the area) and fish ponds in Asaba.

Materials and Methods

Study Area

The study area is Asaba, located on longitude $6^{\circ}42'19''$ E and latitude $6^{\circ}11'04''$ N (Collins Map, 2011). Asaba is the capital city of Delta State and is situated in Oshimili South Local Government Area of Delta State which is bounded on the east by River Niger with a few streams

discharging into the river. The inhabitants of Asaba are mostly civil servants, traders, artisans and a few fishermen. Fish of various types are available in the area. Sources of fresh fish in the area are rivers, lakes, wild ponds and culture ponds. Rivers and lakes are the most regular sources of fresh fish while culture ponds are the most reliable source of fresh fish (Nwabueze and Nwabueze, 2010). All ten fish ponds surveyed for this study are culture ponds located in Asaba and include: Friday, J.K.C., Glamour, Tega, Delta Spring Nigeria limited, DELSU Investment, Obiorha, Morrison, Abraham and Faculty of Agriculture Research fish farms.

Collection of Water and Fish Samples

Water samples were collected three times a week from the ten fish ponds using sterile 75 cl plastic bottles for a period of twelve weeks spanning from May to July, 2012. Two bottles of water samples were collected per pond and were transported to the Fisheries Laboratory for analysis. Water quality parameters such as temperature, dissolved oxygen (DO) and pH were determined according to APHA (1985).

Five fresh fish samples each were obtained from the ten fish ponds three times a week for the twelve weeks of study. Fish samples were transported in coolers with ice packs to the Fisheries Laboratory for parasitological and microbial examination.

Examination of Fish for Disease Agents

Parasitological examination was performed according to Robert (1978). Skin scrapping of fresh fish samples for external parasites was carried out. The mucus obtained from the skin and fins were separately washed into sterile petri dish using physiological saline. The gills were extracted and also washed into sterile petri dish as with the mucus. The mixture obtained was stirred for consistency and a pipette was used to take 1 or 2 drops which was placed onto glass slide and covered with a cover slip. This was viewed under the microscope at x 100 magnification. Wet squash preparations were made of internal organs like the kidney, liver and intestine. The squash preparations were fixed for 60 sec in absolute methanol. Slides were then stained with parasitologic iodine and were placed on slide and viewed under the microscope.

For the examination of fish for the presence of bacteria, smears were stained according to (Robert, 1978). Bacteria were isolated for confirmation. MacConkey agar was prepared using the manufacturer's instructions. Twenty six grams of MacConkey agar was mixed into 500 ml of

distilled water after which it was stirred to mix properly. The mixture was autoclaved at 120 °C for 15 mins and then allowed to cool to 45 °C. The media was then poured into disposable Petri dish and allowed to solidify. The plate was inoculated with pasture pipette and incubated at 37 °C for 24 hours. A portion of the growth was sub-cultured and gram stained (Buchann and Gibson, 1974) and slide viewed under the microscope (x 100).

Saboraud dextrose agar was used for the isolation of fungi according to Alexopoulos and Mins (1979). The medium was prepared by dissolving 14 g/liter of distilled water by gentle heating and sterilized in an autoclave at 120 °C for 15 mins and then allowed to cool to 45 °C (Cruickshank *et al.*, 1975) and was poured into sterile disposable Petri dishes and allowed to solidify. Isolates were identified using gram staining and viewed at x 100 magnification.

Identification of Disease Agents

Parasites were identified using Roberts (1978), bacteria characterized and identified by methods described by MacFaddin (1980) and fungi isolated were identified according to (Rehulka, 1991).

Data Analysis

Data obtained were subjected to analysis of variance test at 95% confidence limit means were separated using Duncan Multiple Range Test (DMRT).

Results

The skin gills, fins, kidney, liver and gall bladder were all infested with parasites. Nine parasites were identified in the fish ponds surveyed and these include *Ichthyobodo Trichodina*, *Tricophyra*, *Cryptobia*, *Chilodonella*, *Gyrodactylus*, Nematode (*Capillaria*), *Glochidium* and *Piscicola*. Most of the parasites were ectoparasites except Nematode (helminth endoparasite) and *Cryptobia* (blood parasite). Eighty-five (56.7%) of 150 *C. gariepinus* examined were found to be infested with protozoan and nematode parasites. Glamour and Delsu Investment fish farms had the highest number of parasite-infested catfishes while Friday and Tega fish farms had the least infestation. Parasites of *C. gariepinus* in surveyed ponds are presented on Table 1.

Bacteria isolated from the *C. gariepinus* include *Flexibacter*, *Escherichia coli* and *Staphylococcus*. Bacteria were found on the skin, fins, gills, liver, kidney and intestine of fish samples examined. Out of 150 fish samples examined 80 (53.3%) had bacterial infestations. Glamour and Delsu

Table 1. Parasites of *C. gariepinus* in fish ponds surveyed.

S/N	Fish Ponds	Number of fish Examined	Number of fish With Parasites	Parts of Body of fish infested	Parasites found
1	Friday Farm	15	6	Skin	<i>Gyrodactylus</i> , <i>Chilodonella</i>
2	J.K.C. Farm	15	7	Gill	<i>Ichthyobodo</i> , <i>Piscicola</i>
3	Glamour Farm	15	12	Skin & gills	<i>Nematode</i> (<i>Capillaria</i>), <i>Ichthyobodo</i>
4	Tega Farm	15	6	Gills	<i>Glochidium</i> , <i>Piscicola</i>
5	Delta Spring	15	8	Skin & gills	<i>Glochidium</i> , <i>Piscicola</i>
6	DELSU Inv.	15	11	Skin & Gill	<i>Chilodonella</i> , <i>Gyrodactylus</i> , <i>Trichodina</i> , <i>Tricophyra</i>
7	Obiorha Farm	15	10	Skin, fins, liver, intestine	<i>Cryptobia</i> , <i>Ichthyobodo</i> , <i>Gyrodactylus</i>
8	Morrison Farm	15	7	Skin, fins, gill, intestine	<i>Cryptobia</i> , <i>Trichodina</i> , <i>Ichthyobodo</i> , <i>Chilodonella</i>
9	Abraham Farm	15	9	Skin & Gill	<i>Gyrodactylus</i> , <i>Chilodonella</i> , <i>Ichthyobodo</i>
10	Faculty Research Farm	15	9	Skin, liver, fins, intestine	<i>Cryptobia</i> , <i>Trichodina</i> , <i>Ichthyobodo</i> , <i>Chilodonella</i>
Total		150	85 (56.7%)		

Table 2. Bacterial flora of *C. gariepinus* in fish ponds surveyed.

S/N	Fish Ponds	Number of fish Examined	Number of fish With Bacteria	Parts of Body of fish infested	Bacteria isolated
1	Friday Farm	15	5	Skin, liver	<i>Staphylococcus</i>
2	J.K.C. Farm	15	6	Gill, skin	<i>Escherichia coli</i>
3	Glamour Farm	15	9	Liver	<i>Staphylococcus</i>
4	Tega Farm	15	7	Skin	<i>Escherichia coli</i>
5	Delta Spring	15	9	Skin	<i>staphylococcus</i>
6	DELSU Inv.	15	8	Skin, gill, liver	<i>Flexibacter</i> , <i>E. coli</i> , <i>Staphylococcus</i>
7	Obiorha Farm	15	9	Skin, fins, kidney, liver	<i>Flexibacter</i> , <i>E. coli</i> , <i>Staphylococcus</i>
8	Morrison Farm	15	10	gill, kidney, liver, intestine	<i>Flexibacter</i> , <i>E. coli</i> , <i>Staphylococcus</i>
9	Abraham Farm	15	10	Skin & Gill	<i>Flexibacter</i> , <i>E. coli</i> , <i>Staphylococcus</i>
10	Faculty Research Farm	15	7	Gill, liver	<i>Flexibacter</i> , <i>E. coli</i> , <i>Staphylococcus</i>
Total		150	80(53.3%)		

Inv. fish ponds had more bacterial infestations than the other fish ponds. Table 2 shows the bacterial flora of *C. gariepinus* in fish ponds surveyed.

Five different fungi species were identified in the fish. These are *Saprolegnia*, *Ichthyophonus*, *Candida albican*, *Achyla*, and *Aspergillus fumigatus*. Fungi spores were found on all the organs and the external body of the fish. In Glamour Farms more of the fish (12 out of 15) examined had fungi infestation. Faculty Research Farm had fewer numbers (5) of fish with fungi infestation. Out of 150 fish samples examined 84 (56.0%). Mixed infections

were also observed. Most of the fish examined had a combination of two or more organisms especially on the skin and gill surfaces. Fungi of *C. gariepinus* in fish ponds surveyed are presented in Table 3.

Table 3. Fungi of *C. gariepinus* in fish ponds surveyed.

S/N	Fish Ponds	Number of fish Examined	Number of fish With Fungi	Parts of Body of fish infested	Fungi found
1	Friday Farm	15	6	Skin, skin, gill, Kidney	<i>Saprolegnia</i> , <i>Ichthyophonus</i>
2	J.K.C. Farm	15	7	Skin, gill	<i>Saprolegnia</i> , <i>Candida albican</i>
3	Glamour Farm	15	12	Skin & gills	<i>Achyla</i>
4	Tega Farm	15	6	Gills	<i>Ichthyophonus</i> <i>Achyla</i>
5	Delta Spring	15	8	Skin & gills	<i>Ichthyophonus</i> , <i>Candida Albican</i>
6	DELSU Inv.	15	11	Skin & Gill	<i>Ichthyophonus</i> , <i>Saprolegnia</i> , <i>Aspergillus</i>
7	Obiorha Farm	15	11	Skin, gills, fins, kidney	<i>Ichthyophonus</i> , <i>Saprolegnia</i> , <i>Candida</i> ,
8	Morrison Farm	15	10	Kidney gill, liver intestine	<i>Ichthyophonus</i> , <i>Saprolegnia</i> , <i>Aspergillus</i>
9	Abraham Farm	15	8	Fins, intestine	<i>Ichthyophonus</i> , <i>Saprolegnia</i> , <i>Aspergillus</i>
10	Faculty Research Farm	15	5	Skin gill, liver, kidney	<i>Ichthyophonus</i> , <i>Saprolegnia</i> , <i>Candida</i> , <i>Aspergillus</i>
Total		150	84 (56.0%)		

Analysis shows that the number of fish infested with parasites in all the fish ponds surveyed were not significantly ($P>0.05$) different. The number of bacteria and fungi isolated from the fish ponds differed significantly ($P<0.05$). Friday, DELSU Inv., Faculty research farm differed from J.K.C, Glamour, Tega, Delta spring, Obiorha, Morrison and Abraham fish ponds (Table 4).

Table 4. Results of analysis of disease agents on *C. gariepinus*

S/N	Fish Pond	Parasites	Bacteria	Fungi
1	Friday Farm	4.00± 0.00 ^a	4.33± 0.33 ^b	4.33± 0.00 ^a
2	J.K.C. Farm	2.67± 0.00 ^a	3.00± 0.33 ^a	2.67± 0.67 ^a
3	Glamour Farm	2.67± 0.00 ^a	3.00± 0.00 ^a	2.33± 0.00 ^a
4	Tega Farm	3.33± 0.00 ^a	2.00± 0.33 ^a	3.00± 0.00 ^a
5	Delta Spring	2.33± 0.00 ^a	3.67± 0.00 ^a	3.67± 0.00 ^a
6	DELSU Inv.	4.00± 0.00 ^a	4.00± 0.00 ^b	4.33± 0.33 ^d
7	Obiorha Farm	1.00±0.00 ^a	1.00±0.00 ^a	1.00±0.00 ^a
8	Morrison Farm	3.00±0.00 ^a	3.00±0.00 ^a	3.33±0.33 ^c
9	Abraham Farm	2.00±0.00 ^a	2.00±0.00 ^a	2.00±0.00 ^b
10	Faculty Research Farm	5.00±0.00 ^a	4.33±0.67 ^b	4.66±0.33 ^d

Means± S.E (Standard Error of mean) with the same superscript on the same column are not significantly ($P<0.05$) different.

Water quality parameters of the fish ponds surveyed are presented in Table 5. During the period of study, temperature, DO and pH were highest in Glamour, Faculty Research Farm and Friday fish ponds respectively. Temperature, DO and pH were lowest in Faculty Research Farm, Morrison and DELSU Inv. fish ponds respectively. Results of analysis of water quality parameters of fish ponds surveyed show that there were on significant

($P>0.05$) difference in the values obtained for temperature and pH. However, DO was significantly ($P<0.05$) lowest in Friday, Tega and Morrison fish ponds and lower in Glamour and Delta spring fish ponds than in J.K.C, DELSU Inv., Obiorha, Abraham and Faculty Research Farm fish ponds which had significantly ($P<0.05$) higher DO (Table 6).

Table 5. Water quality parameters of fish ponds surveyed

S/N	Fish Pond	Temp. ($^{\circ}$ C)	DO	pH
1	Friday Farm	26	6.8	7.8
2	J.K.C. Farm	25	7.0	7.4
3	Glamour Farm	27	7.3	7.5
4	Tega Farm			
5	Delta Spring	23	5.8	7.3
6	DELSU Inv.	22	6.8	5.8
7	Obiorha Farm	21	7.5	6.4
8	Morrison Farm	20	5.0	6.0
9	Abraham Farm	24	7.5	7.2
10	Faculty Research Farm	19	8.8	7.3

DO= dissolved oxygen

Table 6. Results of analysis of water quality parameters of fish ponds surveyed.

S/N	Fish Pond	Temp. ($^{\circ}$ C)	DO	pH
1	Friday Farm	22.33 \pm 1.16 ^a	7.03 \pm 0.62 ^a	7.20 \pm 0.52 ^a
2	J.K.C. Farm	23.00 \pm 1.16 ^a	7.60 \pm 0.35 ^b	7.67 \pm 0.20 ^a
3	Glamour Farm	23.00 \pm 1.16 ^a	7.17 \pm 0.18 ^{ab}	7.47 \pm 0.47 ^a
4	Tega Farm	24.33 \pm 1.16 ^a	5.67 \pm 0.38 ^a	7.37 \pm 0.52 ^a
5	Delta Spring	22.00 \pm 1.16 ^a	6.33 \pm 0.21 ^{ab}	6.80 \pm 0.23 ^a
6	DELSU Inv.	20.00 \pm 1.15 ^a	8.00 \pm 0.62 ^b	6.63 \pm 0.52 ^a
7	Obiorha Farm	21.00 \pm 1.15 ^a	7.23 \pm 0.17 ^b	7.10 \pm 0.47 ^a
8	Morrison Farm	22.00 \pm 1.15 ^a	5.56 \pm 0.31 ^a	6.93 \pm 0.52 ^a
9	Abraham Farm	22.00 \pm 1.15 ^a	7.06 \pm 0.21 ^b	6.76 \pm 0.23 ^a
10	Faculty Research Farm	21.00 \pm 1.15 ^a	8.20 \pm 0.34 ^b	7.63 \pm 0.20 ^a

Means \pm S.E (Standard Error of mean) with the same superscript on the same column are not significantly ($P<0.05$) different.

DO= dissolved oxygen.

Discussion

A variety of parasites, bacteria and fungi were found on *C. gariepinus* examined in this study. A total of 150 samples of *C. gariepinus* studied harboured at least one parasite, bacterium or fungus. The presence of protozoans, nematodes, bacteria and fungi infections of *C. gariepinus* observed in this study is an evidence of poor culture conditions in the fish ponds incidence of undesirable bacteria, protozoa, viruses and helminthes of public health significance have been recorded from fish ponds fertilized with organic manure (Okaeme, 1990).

Ichthyobodo, *Trichodina* and *Chilodonella* were found on the skin, gills and the fins of *C. gariepinus*. Infections of fish body parts with these parasites have been reported particular the gills which have load of parasites relative to the other parts of the body due to the ability of the gill rakers to sieve and in the process trap some of these organisms (Omoniyi *et al.*, 2002, Emere and Egbe, 2006). Helminthes have also been noted as a predominant group of parasites that significantly affect fish in most of the fish ponds in Africa (Paperna, 1996, Hecht and Endemenn, 1998). Environmental circumstances such as poor water quality, changes in temperature, poor nutrition and

overcrowding which are common in intensive fish farming cause stress making the fish more susceptible to a wide variety of pathogens (Reno, 1998). Stress resulting from poor maintenance of ponds can lead to increase in parasitic organisms which on the other hand predispose fish to parasites (Salmon Society, 2009).

Flexibacter, *Escherichia coli* and *Staphylococcus* were isolated from all the fish ponds except *Staphylococcus* in only one fish pond. Ikpi and Offem (2008) observed that *Flexibacter columnaris*, *Pseudomonas*, *Aeromonas* and *Vibrio* species are common fish pathogens in fish farms in Nigeria. Outbreaks of *Flexibacter columnaris*, *Staphylococcus* and *Escherichia coli* in fish ponds have been reported to probably have resulted from the purchase of fingerlings from infected ponds, poor husbandry practices and environmental factors (Ventura and Grizzle, 1987, Clembor *et al.*, 1995, Noga, 2000).

Saprolegnia, *Ichthyophonus*, *Candida albican*, *Achyla*, and *Aspergillus fumigatus* were found on the body parts of *C. gariepinus*. The three most important fungal diseases that have been identified are saprolegniasis, branchiomycosis and *Ichthyophonus* disease (Tucker, 1985, Singh *et al.*, 1991, Klinger and Francis-Floyd, 1996). Klinger and Francis-Floyd, (2002) reported that fungi are everywhere and that they are the most common disease seen in fish because fungi spores are found in all fish tanks. It was also reported that fungi can be a problem if the fish are stressed by disease, poor environmental conditions, receive poor nutrition or are injured. Saprolegniasis is amongst the most common fungal infections of fish and fish eggs. *Saprolegnia* (water mold) can occur with poor water quality culture conditions such as low DO, high ammonia, high organic load and presence of dead eggs. Many other less common mycotic infections have been reported in fish, including the infections with *Achyla*, *Candida* and others (Merck Veterinary Manual, 2011). The mixed infections observed are similar to earlier reports of coexistence of bacterial infections of fish with fungal infections. *Flavobacterium* have been reported to coexist with *Saprolegnia* (Hart *et al.*, 2006).

Water quality is the most important factor involved in fish health management. This is based on the fact that water quality attributes are prime factors that influence fish survival, reproduction, growth performance and overall biological production (Boyd and Lichtkoppler, 1979). According to Boyd (1978) the recommended range of temperature, pH and DO are 20-30 °C, 5.0-9.0 and 5.0-9.5 mg/l respectively. In this study, the values of temperature, pH and DO obtained fall within the recommended range.

Conclusion

The disease status of fish and fish ponds in Asaba shows that fish and fish ponds are not free of pathogenic agents. All the fish examined and most fish ponds surveyed had pathogenic agents. Caution should therefore be exercised in fish farm operations for effective management of water quality. Also, a routine check and monitoring of the current status and prevalence of disease agents in fish ponds is desirable to forestall any future outbreaks of fish diseases.

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