SAPROLEGNIASIS IN FRESHWATER CATFISHES SOLD IN FISH MARKETS IN ASABA, SOUTHERN NIGERIA

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ABSTRACT
The prevalence of saprolegniasis in freshwater catfishes sold in fish markets in Asaba was investigated. A total of two hundred and sixty-five freshly caught table-sized freshwater catfishes namely: *Clarias anguillaris* (Pellegrin), *Heterobranchus bidorsalis* (Geoffrey St. Hillarie), *Synodontis clarias* (Linnaeus), *Schilbe mystus* (Linnaeus) and *Bagrus bayad* (Ruppell) were used for the study which lasted for three months. Fish samples bought on a weekly basis from two major markets (Ogbeogonogo and Cable point) were transported to the laboratory and examined by smear scraping of mucus on skin for saprolegniasis and possible secondary bacterial and protozoal infections using routine procedures. Isolates from cultures revealed the presence of saprolegniasis, scyphidian flora and bacterial infections of *Staphylococcus aureus* and *Escherichia coli*. Forty-six (17.4%) of the fish examined had saprolegniasis (P > 0.05). The number of fungal zoospores from ulcerative lesions were however significantly (P < 0.05) higher than those in control fish which were free from *Saprolegnia* infection. A significant (P < 0.05) number, 25 (54%) of catfishes with *Saprolegnia* had scyphidian flora. *S. aureus* was isolated from all catfishes examined while *E. coli* was found in 193 (72.8%) of the fishes. Maintenance of good water quality and high standard of personal hygiene by fresh fish handlers is necessary to avoid the health implications of fish diseases.

KEYWORDS: Asaba, Catfishes, Fish markets, Nigeria, Saprolegniasis etc.

INTRODUCTION
Fish diseases constitute one of the most important problems militating against the growth of fish farming, especially in the tropics. Diseases of fish have been reported to be common in intensive fish culture systems which are prone to continuous environmental fluctuations and poor management practices which render the fish susceptible to a wide variety of pathogens (Romney et al., 1995). Among the many pathogens of fish is *Saprolegnia*, which has been reported to be the major fungus infecting freshwater fishes (Willoughby, 1976; Roberts, 1978; Singh et al., 1991). The fungus causes the diseases saprolegniasis, which has a characteristic ulcerative skin lesion commonly noticed on the head and gill regions. Primary infection with saprolegniasis has been documented (Hoshina et al., 1960). Recently however, saprolegniasia is well recognized as a secondary infection of all types of piscine skin lesions (Arearat et al., 1981). It was observed that most fishes which are subjected to stress due to handling injuries, malnutrition, temperature shock, external parasitism and frequent spawning are susceptible to infection by *Saprolegnia* (Romney et al., 1995). The inhabitants of Asaba especially the indigenes have a special delight for fresh fish which is abundant due to proximity to the River Niger and other sources of fresh fish including cultured fishes. *Saprolegnia* infected fishes have been recently observed in fish markets in Asaba and this may soon become a source of concern to some consumers of fresh fish in the area. This paper reports the prevalence of saprolegniasis in fresh water catfishes sold in fish markets in Asaba.

MATERIALS AND METHODS
Study area
Asaba is the state capital of Delta State of Nigeria located on latitude 6°11′N and longitude 6°45′E. Asaba is bounded on the East by the River Niger. There are also smaller rivers such as River Anwai which discharge into the R. Niger. In addition, there are numerous streams, lakes and culture ponds in and around Asaba.

Collection of samples
Freshly caught table-sized freshwater catfishes were bought on a weekly basis from the two major markets in Asaba: Ogbeogonogo and Cable point markets. A total of 265 apparently healthy freshwater catfishes were bought and transported to the Fisheries laboratory of Delta State University, Asaba Campus, in open plastic buckets containing little amount of water. The catfishes used were mud catfish, *Clarias anguillaris* (L.), catfish, *Heterobranchus bidorsalis* (Geoffrey Saint Hilaire), upside down catfish, *Synodontis clarias* (L.), butterfish, *Schilbe mystus* (L.) and silver catfish, *Bagrus bayad* (Ruppell). The study lasted for three months in 2012.

Examination of fish
Fish were examined for possible infection. A smear scraping of mucus from the skin and the ulcerative portions of the skin of each catfish was done separately and scrapings cultured using sabouraud dextrose agar medium. Isolation and identification of *Saprolegnia* fungus was carried out (Alexopoulos and Mines, 1979). Also, smear scrapings were cultured for possible bacterial infection (ICMSF, 1986). Characterization and identification of bacterial isolates were done following standard methods (Buchanan and Gibbson, 1974,
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Macfaddin, 1980). For protozoal investigation, smear scrapings of mucus from the fish skin as well as the ulcerative portions of the skin were taken, mounted on slide in saline and examined for protozoal invaders (Roberts, 1978). Apparently healthy freshwater catfishes without ulcerative lesions obtained from the University Research Farms were treated with disinfectant by immersing the fish in a solution of 10g/l dosage of malachite green for 30 minutes according to (Shaoqi, 1989) and used as control. These were also examined for Saprolegnia, bacterial and protozoal infections.

Data analysis
Data obtained was subjected to student’s t test statistics at 95% confidence limit.

RESULTS
Out of 265 freshwater catfishes examined for saprolegniasis, 46 (17.4%) were infected. Prevalence of infection was observed to be higher in C. anguillaris than in any other catfish examined. Results are presented in Table 1. Of the 150 catfishes obtained from Ogbeogonogo market, 28 (18.7%) were infected while 18 (15.7%) of 115 from Cable Point market were infected (Figure 1). Table 2 shows the prevalence of saprolegniasis in catfishes in the two fish markets surveyed. The number of fungal zoospores from the saprolegniasis infected ulcerative lesions was significantly (P<0.05) higher than those of the control fish which had no Saprolegnia infection (Table 3). However, there was no significant difference in the number of fishes examined and the number of fishes with saprolegniasis. Laboratory investigations also revealed the presence of scyphidian flora such as Vorticella and Chilodonella. Twenty five (54%) of the catfishes with saprolegniasis had scyphidian flora. This was found to be significantly higher (P<0.05) than in catfishes without saprolegniasis. All freshwater catfishes with or without ulcerative lesions examined had bacterial infections of S. aureus and E. coil. S. aureus was isolated from all the catfishes while E. coil was isolated from 193(72.8%) of the 265 catfishes examined. Double bacterial infections of S. aureus and E. coil were isolated from only C. anguillaris and S. clarias. The other fish species had only S. aureus. Saprolegnia was not found on skin of fish without ulcerative lesion. Table 4 shows the pattern of occurrence of scyphidian flora and bacterial infections in catfishes examined. It was observed that most of the ulcerative lesions on the skin of the catfishes were as a result of handling injuries. In the markets, it was noted that fresh fish with the characteristic ulcerative skin lesions of Saprolegnia attracted lower prices than fresh fish without ulcerative lesions. Buyers of fresh fish were observed to prefer fishes with smoother skin, even though at a higher cost.

| Catfishes | Number Examined | Number Infected | Percentage (%) Prevalence |
|-----------|----------------|----------------|--------------------------|
| 1 C. anguillaris | 68 | 23 | 33.8 |
| 2 H. bidorsalis | 37 | 3 | 8.1 |
| 3 S. mystus | 53 | 2 | 3.8 |
| 4 B. bayad | 45 | 6 | 13.3 |
| 5 S. clarias | 62 | 12 | 19.4 |
| Total | 265 | 46 | 17.4 |

**FIGURE 1.** Percentage infection of saprolegniasis in catfishes sold in fish markets in Asaba.
TABLE 2: Prevalence of Saprolegniasis in Catfishes in Two Markets in Asaba.

| Catfishes        | Ogbeogonogo Market | Cable Point Market |
|------------------|--------------------|--------------------|
|                  | Number Examined | Number Infected | % Prevalence | Number Examined | Number Infected | % Prevalence |
| C. anguillaris   | 48               | 13               | 27.1        | 26             | 10            | 38.5        |
| H. bidorsalis    | 15               | 2                | 13.3        | 22             | 1             | 4.6         |
| S. mystus        | 27               | 0                | 0.0         | 30             | 2             | 6.7         |
| B. bayad         | 25               | 4                | 16.0        | 20             | 2             | 10.0        |
| S. clarias       | 35               | 9                | 25.7        | 17             | 3             | 17.7        |
| Total            | 150              | 28               | 18.7        | 115            | 18            | 15.7        |

TABLE 3: Occurrence and mean number of saprolegniasis zoospores (zoospores/litre) in freshwater catfishes surveyed.

| Catfishes | Control | Ogbeogonogo market | Cable Point market |
|-----------|---------|--------------------|--------------------|
| C. anguillaris | 0       | 3.7 x 10^4        | 4.5 x 10^4        |
| H. bidorsalis | 0       | 2.2 x 10^3        | 1.8 x 10^3        |
| S. mystus  | 0       | 0.1 x 10^4        | 1.1 x 10^2        |
| B. bayad   | 0       | 2.5 x 10^5        | 2.3 x 10^5        |
| S. clarias | 0       | 4.7 x 10^4        | 2.1 x 10^3        |

TABLE 4: Pattern of Occurrence of Scyphidian Flora in Saprolegniasis and Bacterial Infections of Fresh Water Catfishes.

| Catfishes | Number Examined | Number with Saprolegnia | Number with Scyphidian flora (%) | Number with bacterial infections (%) |
|-----------|-----------------|-------------------------|----------------------------------|-------------------------------------|
| C. anguillaris | 68             | 23                      | 21 (91.3)                        | 68 (102)                            |
| H. bidorsalis    | 37             | 3                       | 0(0)                            | 37 (0)                              |
| S. mystus       | 53             | 2                       | 0(0)                            | 53 (0)                              |
| B. bayad        | 45             | 6                       | 1(16.7)                         | 45 (0)                              |
| S. clarias      | 62             | 12                      | 3(25.0)                         | 62 (91)                             |
| Total           | 265            | 46                      | 25(54.3)                        | 165(100.0)                          |

DISCUSSION

Fresh water catfishes examined had 17.4% prevalence of Saprolegnia infection. The presence of saprolegniasis in the catfishes is not uncommon as several cases of the fungal infection have been reported in many freshwater bodies, especially in intensive culture systems (Fedoruk, 1981; Muir, 1981; Hart et al., 2006). In Asaba, cases of protozoal, bacterial and fungal pathogens in fish and fish ponds have been reported (Nwabueze, 2012). Fungi spores are everywhere and a common occurrence in fish tanks (Klinger and Francis-Floyd, 2002). A high prevalence of infection was observed for C. anguillaris than in any other fish. C. anguillaris is a well known cultured fish in this part of the country and the fish species has the ability to withstand several common parasitic infections (Holden and Reed, 1978). This ability to tolerate parasitic infection may be the reason for the higher prevalence of saprolegniasis observed in the catfishes. Saprolegnia infection was observed in fish from the two fish markets surveyed. This is expected since the markets depend on the same sources for fish supply in Asaba. Ogbeogonogo had a higher prevalence of infection than Cable Point market. This may be due to the fact that Ogbeogonogo currently has a higher sale outlet for freshwater fishes in Asaba. The number of fungal zoospores from the Saprolegnia infected ulcerative lesions was significantly higher (P <0.05) than in control fish which had no Saprolegnia infection. This finding agrees with previous work (Romney et al., 1995) which reported that Saprolegnia is an opportunistic fungus, which takes advantage of the stress condition of the fish to cause an infection. It was observed that majority of the catfishes infected with saprolegniasis had handling injuries. This may have increased the susceptibility of the catfishes to infection by Saprolegnia. Saprolegnia has also been described as either a primary (Hoshina et al., 1960) or secondary infection (MacPag et al., 1981; Arearat et al., 1981) of all types of piscine skin lesions. The presence of saprolegniasis is also indicative of a sewage or organic polluted environment which increased the conduciveness of the water bodies to saprotrophs. Saprotrophs are water moulds which breakdown cells and tissues enabling them to absorb nutrients such as proteins and carbohydrates (Willoughby, 1976; Roberts, 1978). According to earlier reports (Romney et al., 1995), there is a potential for infection whenever fungal zoospores are present in excess of 23,000 spores / liter. Although the prevalence of saprolegniasis was not significant, it has been reported (Roberts and Wootten, 2005) that there may be a danger in underestimating saprolegniasis. The number of fungal zoospores from infected ulcerative Saprolegnia lesions was however significant (P<0.05) higher than those of the control fish without saprolegniasis. This shows that the infection when present could rapidly spread in the fish population since the fungal zoospores may be abundant in the water bodies. It shows that any stressed fish in the water body is liable to having the infection. Poor maintenance of fish ponds can lead to an increase in parasitic organisms which the fish become predisposed to when they are stressed up (Salmon Society, 2009). Scyphidian flora was observed in 54.3% of Saprolegnia infected cases. The presence of scyphidian flora along
with saprolegniasis has been reported as a common occurrence (MacPag et al., 1981). *S. aureus* and *E. coli* were isolated and identified from catfishes examined with or without ulcerative lesions. Coexistence of *Flavobacterium* with *Saprolegnia* has been reported. The duo of *S. aureus* and *E. coli* has also been reported in fresh fish samples as part of the natural micro-flora of fish. *S. aureus* contamination of fresh fish has been found to be from human handlers while *E. coli* is indicative of human waste polluted water body (Clucas and Ward, 1996). This implies that wild and culture ponds in and around Asaba may be polluted with organic matter as the fishers in fish markets get regular and most reliable supplies of fish from wild and culture ponds respectively (Nwabueze and Nwabueze, 2010). This is not healthy for the well being of the fishes in the water as well as for human consumers of fresh fish in Asaba. Maintenance of clean water bodies free of untreated sewage and other organic waste is highly recommended. Fish culturist and fresh fish handlers should maintain good water quality for cultured fishes and a high standard of personal hygiene to avoid the health implications of fish diseases.

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