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SHORT COMMUNICATION

OCCURRENCE OF STREPTOCOCCOSIS IN AFRICAN CATFISH (*Clarias gariepinus*) FINGERLINGS RAISED IN A COMMERCIAL FARM IN AKURE, NIGERIA

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Abstract

This investigation reports the outbreak of Steptococcosis in *Clarias gariepinus* fingerlings in a commercial farm in Akure, Nigeria. The clinical signs observed on affected fish samples were haemorrhages in their ventral parts and mucous on the gills. The causative agent was found to be *Streptococcus iniae*. The predisposing factors for the outbreak were high stocking density and highwater temperature, the bio-film formations on the wall of the rearing tanks, as well as growing young fish in untreated rearing water that was previously stocked with adult catfish and tilapia. The infection lasted ten days during which all the fish died.

Keywords: infection, fingerlings, antibiotics, outbreak.

Introduction

Fish farming activities have developed at a rapid rate in Nigeria over the last two decades, with Clarid catfishes being the mostly cultured species due to its hardness and good commercial value (Adewumi, 2005; Olabode and Ogunshe, 2009). However, current production intensification has in turn brought about several infectious diseases which were initially unknown to farmers, similar to what is being witnessed in other aquaculture-producing nations (Winton, 2001; Munn, 2004).

Bacterial diseases were reported to be the major cause of mortality of farmed catfishes in Africa, leading to serious economic losses to fish farmers (Ikpi and Offem, 2011). Apart from few obligate fish pathogens, most of these disease-causing microorganisms are essentially opportunistic pathogens, which invade the tissue of a fish rendered susceptible to infection by stress factors or other disease processes (Oladele *et al.*, 2015). One of such opportunistic pathogen is *Streptococcus iniae*. This bacterium was initially isolated from an Amazon freshwater dolphin, *Inia geoffrensis* (Pier and Madin, 1976). The association with fish diseases came when it was described as a cause of mortality in tilapia hybrids *(Tilapia nilotica x T. aurea*) (Perera *et al.* 1994) and later in dusky spinefoot (*Siganus fuscenscens)* (Sugita 1996) and hybrid striped bass (Stoffregen *et al.* 1996). *Str. iniae* has since then been reported as one of the most relevant streptococcal pathogens of cultured and wild fish globally (Shoemaker *et al* 2001; Figueiredo *et al* 2012).

Information on conditions associated with outbreak of fish diseases in commercial fish

farm is important in ensuring that appropriate steps are taken to minimal losses. According to Ezeri (2007), infectious disease conditions often exhibit noticeable signs which are either physical or behavioural in nature. Although behavioural signs or symptoms can be helpful to identify or suspect the occurrence of diseases in fishes, appropriate diagnosis which involves identification of the causative agents and examination of predisposing environmental factors are most essential before treatment measures can be implemented.

The Nigerian aquaculture industry is one which is characterized by poor documentation of fish-associated pathogenic and parasitic diseases (Oni, 2009), as less attention is given to the veterinary aspects of aquaculture. However, there were some reported cases of high mortality losses, especially during the early developmental stages of cultured fish, which could be traced to bacterial infections (Ezeri, 2007; Oladele *et al.*, 2015). As a growing aquaculture-producing country, it is necessary to build research and diagnostic capacities to deal with fish disease problems, using data from disease outbreaks in commercial fish farms (Hecht, 2000; Diyaolu, 2015). This information can be obtained when incidence of mass mortalities of cultured fish are properly investigated and reported. The report investigated here is one of such occurrences of sudden mortalities in fish farms in Nigeria.

Materials and Methods

In October 2019, an episode of mortality occurred in African catfish (*Clarias gariepinus*) fingerlings (average weight of 7.92 g) raised in a commercial farm in Akure, Nigeria. The farm has a hatchery unit (where catfish fingerlings are produced), the holding tanks (where the fingerlings are maintained prior to being sold to grow-out farmers) and a reservoir (which stores water for use in the holding tanks). The farmer reported that the reservoir was used for growing table-sized catfish and tilapia in the previous year. At the time of the outbreak being investigated, the reservoir was only used to store water for pumping into holding tanks.

The catfish fingerlings, grown in the indoor, water flow-through hatchery, were transferred into six, 10 m^3 hexagonal, polythene-fabricated holding tanks half-filled with water. The water was pumped directly from the farm's reservoir into these tanks without any treatment. Each of these holding tanks was stocked with 150 fish/m^3. The water in each tank were partially drained and replaced with the reservoir water every day to ensure that the rearing water was maintained at 27^{0}C, pH 6.7 and dissolved oxygen 5.3 mg/L.

After two days of stocking, few fish were found dead at the base of each holding tanks and responses to feeding were generally poor. Thereafter, fish began to show erratic swimming at the water surface, followed by slow downward movement from water surface to the bottom and finally lateral recumbence at the base of the tanks. The number of fish with these signs increased after fourth day, with similar effects noticed in all holding tanks. The remaining fish were immediately restocked into other tanks prepared by washing with salt (NaCl) solution before filling with water. The daily mortalities continued unabated despite this treatment.

On the sixth day of transfer from the hatchery to the holding tanks, affected fish (both freshly dead and moribund) samples were taken to the laboratory for microbiological analysis in order to determine the cause of infection. Clinical signs and gross external lesions were examined. The abdominal cavity of the sampled fish was split open under

aseptic condition and examined for internal lesions. Swabs from affected organs (gills) and external lesions on the ventral part were streaked on tryptone soya agar (TSA, Oxoid). The plates were incubated at temperature of 30 ºC for 48 hours.

Three representatives of colonies from different plates for fish organ and lesions were subjected to biochemical characterization and antibiotic sensitivity testing. The biochemical tests were performed following standard microbiological procedures recommended by American Fisheries Society – Fish health Section (2007). Drug sensitivity to antimicrobials was carried out on the isolates using disc diffusion method recommended by National Council on Clinical Laboratory Standard (NCCLS, 2003). Antibiotic discs (perfloxacin, gentamycin, ampicillin, amoxiciilin, zinnacef, rocephin, ciprofloxacin, streptomycin, tetracycline and erythromycin) obtained from Maxicare, Nigeria, were placed on the seeded plates and incubated overnight.

Results

The clinical signs observed during gross external examination of affected fish samples were haemorrhages in their ventral parts and mucous on the gill. During internal examination, there was ascitic fluid in the peritoneal cavity and intestines, and the liver was observed to be pale. During bacteriological analysis of the affected areas, mucoid, whitish colonies of 1 – 2 mm in diameter dominated all the plates. These colonies were found to be Gram positive, motile, catalase negative, oxidase negative, indole negative and β haemolytic on blood agar. It fermented glucose and sucrose but not lactose, arabinose and inositol. Comparing these results with biochemical characteristics described by Noga (2010), this organism was identified as *Streptococcus iniae*. In terms of susceptibility to antibiotics, *Str. iniae* identified in this study showed high sensitivities to gentamycin, ciprofloxacin, perfloxacin and rocephin (Figure 1). *In-vivo* trials on the efficacies of these drugs on the fish could not be conducted, as all the fish died within the ten days of infection.

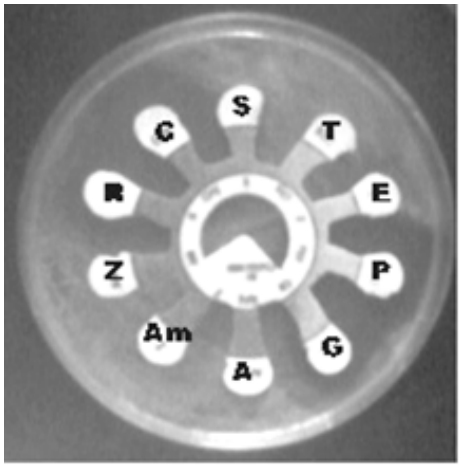

Figure 1. Drug sensitivity of *Streptococcus iniae* isolated from infected *Clarias gariepinus* fingerlings; The symbols in the plate are as follows: A – Ampicilin; P – Perfloxacin; Am – Amoxicilin; R – Rocephin; C – Ciprofloxacin; S – Streptomycin; E – Erythromycin; T – Tetracycline; G – Gentamycin; Z – Zinnacef

Discussion

The disease conditions observed during the present investigation were similar to the description of streptococcosis given by previous researchers (Agnew and Barnes 2007; Austin and Austin, 2016). Streptococcosis has been implicated for significant mortalities resulting in considerable losses to the aquaculture industry, with many species from the family Streptococcaceae (including *S. iniae*, *S. agalactiae*, *S. parauberis*) identified as aetiological agents in cultured fish (Noga, 2010). This investigation identified *Streptococcus iniae* as the causative agent responsible for mass mortalities of *C. gariepinus* fingerlings.

Some predisposing factors might have contributed to the Streptococcosis outbreak

investigated in this study. African catfish fingerlings were stocked at the rate of 150 fish per cubic metre of water in each holding tank, indicating high stocking densities capable of predisposing fish to infection. The water temperature (27 ^{0}C) was maintained within the limits recommended by Britz and Hecht (1989), but these values may likely favour the survival of *Str. iniae* within the culture facilities. Earlier reports on *Str. iniae* outbreaks in tilapia commercial fish farms by Agnew and Barnes (2007) showed that high stocking densities and highwater temperatures (above 25 ^{0}C) were the main predisposing factors. The water used for growing fish in this study was pumped from the reservoir into the holding tanks without any treatment. The reservoir was reportedly used to raise table size catfish and tilapia in the previous year, which might be the source of infection in this study. Hence, there is need to exercise caution when untreated water from previously stocked pond is used to raise young fish whose immunity is yet to be fully developed.

Additionally, bio-film formations (dense aggregates of surface-adherent microorganisms embedded in an exopolysaccharide matrix) may occur on the wall of the tanks and serve as shield for the pathogen, providing protection from the host defence factors and from external treatments, such as antibiotics (Lazado *et al.*, 2010). Many aquatic bacteria are reported to be capable of forming bio-films on solids immersed in water (Branda *et al.*, 2005; Cvitkovitch *et al.*, 2003). According to several authors, intra-community (i.e. biofilm, auto-aggregation) and inter-community interactions (i.e. co-aggregation) are common mechanisms of bacterial survival in nature and have been identified to play a part in the virulence of pathogens, including the streptococci (Cvitkovitch *et al.*, 2003; Khemaleelakul *et al.*, 2006). It is therefore paramount to ensure continuous cleaning of tanks used for growing young fish under intensive culture conditions in order to prevent the establishment of bio-films that may accumulate harmful microorganism within the rearing environment.

The present investigation was a serious infection in which all fish died within 10 days of infection, so it was impossible to conduct in-vivo trials of the drugs on the fish to determine their efficacies. This calls for adoption of rapid diagnostic and treatment techniques in Nigerian aquaculture in order to avert economic loss associated with fish infection and subsequent mortalities.

Conclusion

This study identified *Streptococcus iniae* as a causative organism responsible for mass mortality of African catfish fingerlings in the investigated outbreak. The clinical signs observed on affected fish samples were haemorrhages in their ventral parts and mucous on the gills. The streptococcosis reported here was likely induced by high stocking densities and high water temperature, untreated water used for growing fingerlings and the bio-film formation on the wall of the rearing tanks. The study presented a list of possible antibiotics for treating infected fish if the infection is identified and its treatment initiated on time. Future investigations should employ rapid, culture independent, molecular techniques to properly ascertain causative organisms in fish disease outbreaks.

References

Adewumi, A. A. (2005). The effect of heating time of soyabean for broodstock nutrition on the reproduction performance of *Clarias gariepinus* (Burchell 1822). *Ph. D. Thesis,* Obafemi Awolowo University, Nigreria. Pp 160

Agnew, W. and Barnes, A. C. (2007) *Streptococcus iniae*: An aquatic pathogen of global veterinary significance and challenging candidate for reliable vaccination. *Veterinary Microbiology* 122:1–15.

American Fisheries Society – Fish health Section (AFS-FHS) (2007). *FHS Bluebook suggested procedures for the detection and identification of certain finfish and shellfish pathogens. 2007 edition.* AFS Bethesda, MD.

Austin, B. and Austin, D. A. (2016) *Bacterial disease pathogens of wild and cultures fish. Sixth edition.* Springer International Publishing, Switzerland

Branda, S. S., Vik, A., Friedman, L. and Kolter, R. (2005) Biofilms: The matrix revisited. *Trends in Microbiology* 13: 20–26.

Britz, P. J and Hecht, T. 1987. Temperature preferences and optimum temperature for growth of African sharptooth catfish *Clarias gariepinus* larvae and postlarvae. Aquaculture 63:205 – 214

Cvitkovitch, D. G., Li, Y. H. and Ellen, R. P. (2003) Quorum sensing and biofilm formation in Streptococcal infections. *The Journal of Clinical Investigation* 112: 1626–1632.

Diyaolu, D. O. (2015) Aerobic bacterial flora and survival of African catfish *Clarias gariepinus* during early life stages in the hatchery. *International Journal of Fisheries and Aquatic Studies* 3 (1): 380-384

Ezeri, G.N.O. 2007. Tumor and other epidermal anomalies of *Clarias gariepinus. Aquafield* 3(1): 1 – 7

Figueiredo, H. C. P., Netto, L. N., Leal, C. A. G., Pereira, U. P. and Mian, G. F. (2012) *Streptococcus iniae* outbreaks in Brazilian Nile tilapia (*Oreochromis niloticus* L:) farms. *Brazilian Journal of Microbiology* 43(2): 576–580.

Hecht, T. (2000) Considerations on African Aquaculture. *World Aquaculture* 31 (1):12-19

Ikpi, G. and Offem, B.(2010). Bacterial infection of mudfish *Clarias gariepinus* (Siluriformes: Clariidae) fingerlings in tropical nursery ponds. *Revista de Biologı´a Tropical* 59 (2): 751–759.

Khemaleelakul, S., Baumgartner, J. C. and Pruksakom, S. (2006). Autoaggregation and coaggregation of bacteria associated with acute endodontic infections. *Journal of Endodontics* 32: 312–318.

Lazado, C. C., Caipang, C. M. A., Rajan, B., Brinchmann, M. F. and Kiron, V. (2010). Characterization of GP21 and GP12: Two potential probiotic bacteria isolated from the gastrointestinal tract of Atlantic cod. *Probiotics and Antimicrobial Proteins*, 2, 126–134.

Munn, C. B. (2004) *Marine Microbiology: ecology and application.* USA, Garland Science/BIOS Scientific Publishers. 282p

National Council on Clinical Laboratory Standards (NCCLS) (2003) Performance standard for antimicrobials disc susceptibility tests. *NCCLS* 23: 37 – 50

Noga, E. J, (2010) *Fish disease: diagnosis and treatment. Second edition.* Wiley-Blackwell, USA, 538p

Olabode, P. O and Ogunshe, A. A. (2009) Antimicrobial potentials of indigenous Lactobacillus strains on gram-negative indicator bacterial species from *Clarias gariepinus* (Burchell, 1822). Microbial inhibition of fish-borne pathogens. *African Journal of Microbiology Research* 3(12): 870 – 876

Oladele, O. O., Olarinmoye, A. O., Ntiwunka, U. G. and Akintomi, T. O. (2015) Survey of bacterial isolates from cases of fish disease outbreaks and their antibiotic

susceptibility patterns. *Nigerian Journal of Fisheries* 12 (2): 901 – 906

Oni, T. A. (2009). Characterization of bacteria and fungi associated with *Clarias gariepinus* (Burchell 1822) fingerlings. *M. Sc.Thesis*, Obafemi Awolowo University, Nigeria. Pp 108.

Perera, R. P., Johnson, S. K., Collins, M. D. and Lewis, D. H. (1994) *Streptococcus iniae* associated with mortality in *Tilapia nilotica* x *T. aurea* hybrids. *Journal of Aquatic Animal Health* 6:335–340

Pier, G. B. and Madin, S. H. (1976) *Streptococcus iniae* sp. - a beta-hemolytic streptococcus from an Amazon freshwater dolphon, *Inia geoffrensis*. *International Journal of Systematic Bacteriology* 26:545–553

Salinas, I., Diaz-Rosales, P., Cuesta, A., Meseguer, J. and Chabrillo, M. (2006) Effect of heat inactivated fish and non-fish derived probiotics on the innate immune parameters of a Teleost fish (*Sparus aurata* L). *Veterinary Immunology and Immunopathology* 111: 279-286.

Shoemaker, C. A., Klesius, P. H. and Evans, J. J. (2001) Prevalence of *Streptococcus iniae* in tilapia, hybrid striped bass, and channel catfish on commercial fish farms in the United States. *American Journal of Veterinary Research* 62:174–177

Stoffregen, D. A., Backman, S. C., Perham, R. E., Bowser, P. R. and Babish, J. G. (1996) Initial disease report of *Streptococcus iniae* infection in hybrid striped (sunshine) bass and successful therapeutic intervention with the fluoroquinolone antibacterial enrofloxacin. *Journal of World Aquaculture Society* 27:420–434

Sugita, A. (1996). A case of streptococcicosis in dusky spinefoot. *Fish Pathology* 31:47–48

Winton, J. R. (2001). Fish health management. In: Wedemeyer, G. A. (ed) *Fish Hatchery Management. Second edition*. USA, American Fisheries Society. Pp560-639