# ASSESSMENT OF MICROBIAL QUALITY AND SAFETY OF SOME SELECTED FISH PONDS IN YAMALTU/DEBA L.G.A OF GOMBE STATE

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## ABSTRACT

Water quality entails the undesirable growth of microorganisms whose metabolic compounds are easily detected by human nose or mouth, while its safety could be determine by the presence of pathogenic microbes. This study aimed to analyze both the microbiological quality and safety of water from some selected fish ponds in Yamaltu-Deba Local Government area of Gombe State, Nigeria. Samples were aseptically collected using sterile sampling bottles, subjected to isolation, Gram staining, biochemical tests, microscopic and macroscopic identification using atlas. The bacterial strains isolated include *Escherichia coli*, *Streptococcus sp, Salmonella sp, Klebsiella sp and Enterobacter sp* while the identified fungal isolates include *Aspergillus sp*, *Penicillum sp*, *Cladosporium sp*, *Fusarium sp* and *Mucor sp*. *E. coli* was the most predominant organism with 26.3% occurrence while *Salmonella sp* had the least (14.5%). *Streptococcus sp, Klebsiella sp and Enterobacter sp* had occurrences of 15.8%, 23.7% and 19.7% respectively. However, *Aspergillus sp* had the highest incidence of 40.9%, then *Penicillum sp* (32.7%), *Cladosporium sp* (14 %), *Fusarium sp* (6.5%) and finally *Mucor sp* (4.9 %). The study revealed that the ponds were grossly contaminated with pathogenic microorganisms which poses a risk to the health of the consumers and thus of significant public health concern. The occurrence of *Pseudomonas sp, Shigella sp, Salmonella sp* and *Enterobacter sp* in the pond water if not properly checked could endanger both the fish and the ultimate consumers especially if the fish harvested from these farms are undercooked.

**Keywords**: Water quality, safety, bacteria, fungi, fish pond, Yamaltu-Deba

## INTRODUCTION

Fish is one of the major sources of animal protein, providing over half of the world total nutrition (FAO, 2013). In Nigeria, fish cultivation is a very important source of income and provides employment opportunities to a significant number (70%) of a work force. Nigeria is the largest African aquaculture producer, produced

about 15,489 tonnes per year (Adedeji *et al.*, 2011).

Fishes are reared in different culture media or controlled environment which could be ponds (concrete or earthen), vats (wooden or fiber glass) and plastics (Osawe, 2004). Among these culture systems, concrete and earthen ponds are widely used (Ezenwa, 2006). Fishes cultivated in these controlled environments has been found to be contaminated by microorganisms (pathogenic and opportunistic organisms) (Fafioye, 2011; Nguyen *et al.*, 2007). This contamination has been attributed to questionable water quality and high stocking densities (Okpokwasili and Ogbulie, 2008). The feed used for the fish in these ponds contain organic materials and introduces a wide variety of microorganisms into the ponds (Okpokwasili and Ogbulie, 2009). The sources of water and the feed used for fish that is usually generated from animal manure are usually the factors that determine water quality of the ponds (Adebami *et al*., 2020).

Most diseases of humans are caused by opportunistic enteric pathogens, which are prevalent in the rearing environment (Jayasne *et al.*, 2003). Water is the most important resource for aquaculture and can be a significant source of contamination. The conditions that fishes are cultured may be potentially stressful, causing existing infections to become more severe and precipitate disease outbreaks which may also compromise the fitness of such fish for human consumption. The temperature of water supplied to a fish pond ranges from 25 to 35$^{o}$C and this support the growth of fishes as well as microorganisms (Torimiro *et al*., 2014).

The presence of some members of the enteric bacteria such as *Salmonella sp.* and the coliforms in fish pond is an indication that the water is unsafe, a food safety concern. This is in addition, an indication of faecal contamination. However, the presence of fungi such as *Penicillium sp.* and *Mucor sp.* indicates poor quality of the water because such organisms are also involved in food spoilage (Stephen, 2010). Reports on the increase number of pathogenic microorganisms related to aquaculture and discharged fish ponds waste waters are on the increase (Abu and Wondikom, 2018). In this study Ponds water contamination with microorganisms have negative impact on the public health. There are limited literatures that investigate the effect of microorganism in fishes of ponds of Yamaltu-Deba Local Government Area. Hence, there is need for research to detect the microbial quality of the ponds water . Therefore, the aim of this study was to assess the microbial quality and safety of some selected fish ponds within Yamaltu-Deba L G A of Gombe State..

## MATERIALS AND METHODS

### Study Area

The study area is located in Yamaltu-Deba Local Government Area (LGA) which is located in the southern part of Gombe State. The population of the area is about 255,726 according to population census 2006 (NPC 2006) and lies and lies on latitudes 10$^{o}$00’ to 10$^{o}$30’ N and longitudes 11$^{o}$ 15’ to 11$^{o}$ 45’ E. It covers a total land area of

1,981km$^2$ (Samuel *et al*., 2021), with a temperature range between 20$^o$C – 30$^o$C (Ndaghu *et al*., 2011). The ponds are located in Kwadon, Yamaltu-Deba LGA, Gombe State. It lies between latitude 10$^o$ 17'24.9"N to 10°17'74.7"N and longitude 11°17'51.3"E to 11°18'28.0"E. (Fig. 1).

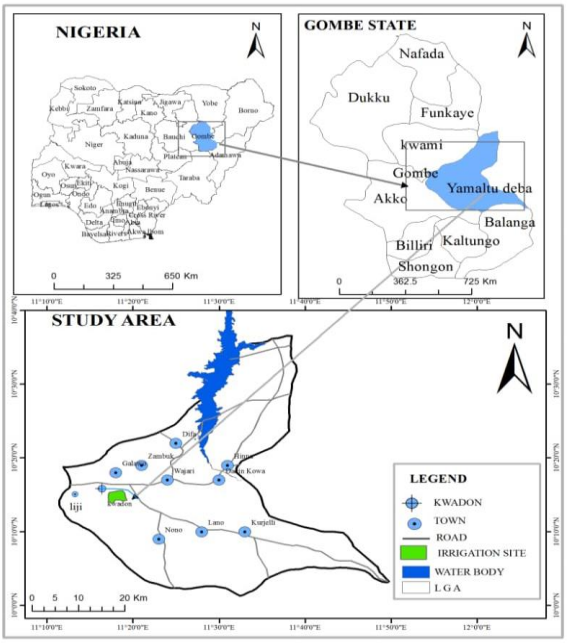


**Figure 1:** Map of Gombe State displaying the the study area in Yamaltu-Deba L.G.

## Sample Collection

Forty pond water samples were collected aseptically from the selected fish ponds at regular intervals of seven days using sterile sampling bottles and brought to Microbiology departmental laboratory, Gombe State University. Upon collection, each sample was labeled with the temporal fish pond's code number and transported in ice pack coolers containing ice cubes within 2-3 hours to the research laboratory for analysis.

## Sample processing

All samples were processed and prepared on a supporting media based on manufacturers description as described by Cheesbrough (2010).

## Glass Wares/Sterilization

All glass wares were washed and sterilized (using dry heat sterilization) in hot air oven at 160°C for 2 hours while wire loop and needles were sterilized by flaming to red heat using naked flame.

## Media Preparation

Media used for this study include: Nutrient Agar, MacConkey Agar, Salmonella-Shigella Agar (SSA) and Potato dextrose agar (PDA). All media used were prepared according to manufacturer's instruction and sterilized in an autoclave at 121°C for 15 minutes.

## Bacterial Isolation

A 0.1ml of each of the pond water sample was inoculated onto the sterile nutrient agar plates using spread plate technique. The plates were then incubated at 35$^0$C for 24 hours under aerobic condition (Waddell, 2016).

## Isolation of Salmonella and Shigella

The Salmonella-Shigella Agar (SSA) was prepared according to the manufactures instruction for selective identification of only *Salmonella spp.* and *Shigella spp*. A 0.1ml of the water sample was inoculated onto the medium as described above. The plates were then incubated at 37 $^o$C for 24 - 48h. Thereafter, pure cultures were obtained and subjected to biochemical reactions for identification.

## Gram staining

A loop full of the colony was picked and emulsify on a drop of normal saline

dropped on a grease-free slide which was allowed to air-dry, the smear was heat fixed. Crystal violet was applied for a minute on the smear; it was rapidly washed off with clean water and the water was tipped after, Lugol's iodine was added and allowed to stay for a minute and was washed off with clean water. It was then decolorized (for few seconds) with acetone-alcohol and was washed immediately with distilled water. The smear was then counter stained with Safranin for two minutes and the stain was washed off. Back of the slide was wiped clean and clean and placed in a draining rack to air-dry and was examined microscopically first with the 40x objective lens to check the staining and see the distribution of the material and then with oil immersion objective (100x) for examination (Cheesbrough, 2010).

**Biochemical Tests**

The confirmatory tests such as catalase, coagulase, citrate utilization, oxidase and urease tests were carried out to identify the isolate microorganisms. The results obtained were then compared with Bergy's manual for determinative bacteriology for confirmation.

**Identification of Fungi**

Fungal identification was conducted using microscopic and macroscopic examination of the fungal growth of hyphae using fungal Atlas.

**RESULTS**

The bacteriological analysis of the water samples showed five different genera (Table 1). These include *Escherichia coli, Staphylococcus aureus, Salmonella* sp., *Klebsiella* sp., and *Enterobacter* sp. *E. coli* had the highest occurrence of 26.3% (n=20) followed by *Klebsiella sp* with 23.7% (n=18), then *Enterobacter* sp. with 19.7% (n=15) occurrence, *Streptococcus* sp. with 15.8% (n=12) occurrence and finally *Salmonella* sp. with 14.5% (n=11) with the least occurrence

**Table 1**: Distribution of bacterial isolates from the different ponds

| Organisms | Number of Occurrence in Ponds | Percentage % |
|---|---|---|
| *E. coli* | 20 | 26.3 |
| *Salmonella* | 11 | 14.5 |
| *Streptococcus* sp | 12 | 15.8 |
| *Enterobacter* sp | 15 | 19.7 |
| *Klebsiella* | 18 | 23.7 |
| **Total** | **75** | **100** |

The fungal genera isolated are presented in Table 2. *Aspergillus* sp. had the highest incidence of 40.9% (n=25). This is followed by *Penicillum sp*. 32.7% (n=20), then *Cladosporium sp*. with 14% (n=9), *Fusarium sp.* 6.5% (n=4) and finally *Mucor sp*. 4.9% (n=3).

**Table 2**: Occurrence of fungal isolates within the ponds

| Organisms | No. of Occu | Percentage (%) |
|---|---|---|
| *Aspergillus* | 25 | 40.9 |
| *Penicillum* | 20 | 32.7 |
| *Cladosporium* | 9 | 14.1 |
| *Fusarium* | 4 | 6.5 |
| *Mucor* | 3 | 4.9 |
| **Total** | **61** | **100** |

## DISCUSSION

The result of the microbiological characteristic showed that Gram negative bacteria were dominant in the bacteria isolated from the ponds. The microorganisms isolated were *E-coli*, *Salmonella* sp. *Klebsiella* sp. *Enterobacter* sp., and *Streptococcus* sp. The coliforms isolated were an indication of the contamination of the pond water with fecal matter which may result to the presence of pathogenic organisms. The fecal matter may be as a result of fertilization of the ponds with animal manure which is discharged directly into the fish ponds, or excreted by the fish into the ponds or through runoff (Kay *et al.*, 2008). The diverse groups of bacteria isolated from these ponds are in line with the report of Okpokwasili and Ogbulie (1999) who worked on pond water suggesting that allochthonous bacteria from feed added to the ponds are the principle source of bacteria of public health importance, and Dabbor (2008) who reported similar organisms in the microbiological study of El-quanter fish pond. The presence of pathogenic microorganisms especially *E.coli* and *Salmonella* can lead to the transmission of water borne diseases such as, Typhoid fever, Cholera, food poisoning and gastroenteritis (Piet, 2009) on consumption of improperly cooked fish cultivated in these ponds. *E. coli* was the most dominant organism occurring in both concrete and earthen ponds. The presence of *E. coli* in water or food indicates the possible presence of causative agents of many gastro intestinal diseases (Ampofo and Clerk, 2010). *Staphylococcus* species have been implicated in food poisoning (Oni *et al.*, 2013). This organism is one of the most opportunistic pathogen for fresh water fish and he main etiological agents in disease outbreak were several mortalities were recorded (Das and Mukheyce, 1999).

Fungal infection is an important economic and limiting factor in intensive fish production. The fungi genera isolated from the ponds were *Aspergillus* sp., *Penicillum* sp., *Cladosporium* sp., *Mucor* sp., and *Fusarium* sp. *Aspergillus* and *Penicillum* species formed the dominant group of fungi in this study. The observation is consistent with the work of Obire and Anyanwu, (2009) who noted that *Aspergillus* and *Penicillum* species are believed to penetrate into the environment through dead plants materials and remains for long period of time. Similarly, Eze and Ogbaran, (2010) cited *Penicillum* sp. as the most abundant fungi during his study on the microbiological and physiochemical of fish pond water in Ugheli Delta state Nigeria. In contrast to the present result, Fafioye, (2011) reported *Cladosporium* sp. as the dominant fungi species. The occurrence of *Fusarium* sp. and *Mucor* sp. in earthen ponds could be

attributed to the fact that the earthen ponds were a more conducive environment for their growth and proliferation due to the presence of soil and plants in the earthen ponds.

## CONCLUSION

This study revealed that both ponds under study were grossly contaminated with food spoilage and pathogenic bacteria that could endanger the health status of consumers particularly if fish harvested from these ponds are under-cooked. It is thus recommended that the environment where fish ponds are located should be protected from pollutants and weeds which can harbour microorganisms that can find their way into fish ponds, themselves or by passive process through wind, rainfall Dam water supply to the fish pond should be examined in the laboratory for its microbiological quality and safety before stocking, this would give insight to the possible presence of certain types of microorganisms, hence provide enabling environment for the purpose of aquaculture. Fish handlers with open wounds should avoid contact with water from the fish ponds and fish should be properly cooked prior to consumption.

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