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Assessment of the Bacteriological Profile of Water and Fish Samples from Hadejia Reservoir in Jigawa State

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ABSTRACT

Hadejia reservoir is a floodplain complex in northeastern Nigeria. This area has long been noted for its importance in fish production. It is also important to both year round native birds and European water birds that travel over the Sahara desert to this wetland to spend their winters. Quantitative and qualitative profile of bacteria in the reservoir was carried out. Quantitative analysis of bacteria in the water revealed that the water Contained Total Heterotrophic Count (THC) of 3.1×10^8 to 3.5×10^6 CFU mL⁻¹ and Total Coliform Count (TCC) ranging from 1.4×10^2 to 1.4×10^3 CFU mL⁻¹. Bacteria load in the fish intestines are 3.5×10^8 and 3.1×10^4 CFU g⁻¹ for total coliform and total heterotrophic count, respectively. Fish gills has less count $(2.1 \times 10^2$ and 3.4×10^3 CFU g⁻¹ for TCC and THC, respectively) than the fish intestines. Bacteria species such as *Aeromonas hydrophila*, *Vibrio cholera*, *Shigella* species were isolated from the water samples. *Escherichia coli*, *Salmonella* sp. and *Shigella* sp. were isolated from the fish samples. So, the present study aims to assess the bacteriological profile of water and fish samples from Hadejia reservoir.

Key words: Bacteriological profile, Jigawa state, Hadejia reservoir

INTRODUCTION

Hadejia reservoir is one of the inland water bodies in Nigeria. Hadejia reservoir is a floodplain complex on the southern edge of the Sahel savanna in the northeastern Nigeria. Hadejia and Jamare rivers that supply this floodplain originate on the Jos Plateau and flow seasonally into lake Chad. This area has long been noted for its importance to both year round native birds and European water birds that travel over the Sahara desert to this wetland to spend their winters. Hadejia-Nguru wetland has at least 89 species of freshwater fish (USAID, 2008). Due to all these, Hadejia wetland had an international attraction. As a natural water body it contains wide variety of microbial flora that originates from living and non-living plant and animals. Some of these microorganisms may be disease causing organisms to aquatic lives and to fish in particular (Cheesbrough, 2006; Agarwal, 2005).

It has been estimated that 25,000-deaths in a day are caused in developing countries either by direct consumption of polluted water or indirectly by contraction of diseases like malaria and bilharzia through disease vectors that live in polluted water (UNEP, 1991).

Biological pollutants may also introduce pathogens into the aquatic environment that could lead to the death of aquatic food organisms such as fish and snails. These pathogens may be transferred to human beings through fish consumption (Agarwal, 2005). This study will help to provide baseline information on biological pollution status of Hadejia reservoir.

MATERIALS AND METHODS

Sample collection: Water samples for bacteriological study were collected by deeping the sample bottle into the lake to a depth of 30 cm below water surface. The bottle is opened, allowed to fill up with water and then corked while still under water (Greenberg, 1985). This was labeled, kept in ice chest box and taken to the laboratory for bacteriological analysis. Fish samples were collected into a wide mouth sterile container and this was covered and kept in an ice chest box before taken to the laboratory for microbial analysis.

Analysis: Quantitative analysis of bacterial populations in the fish and water samples were carried out. Primary isolation which was followed by biochemical analysis was done on the isolates to identify the pathogens present in the water body and fish samples.

Enumeration of total heterotrophic and total coliform bacteria in fish samples: The fish sample used was *Oreochromis niloticus*. This fish species was chosen because it is one of the commonest fish caught from the reservoir. The fish sample was washed thoroughly with sterile distilled water and dissected with the help of a sterile dissecting set. Fish gills and intestines were chosen on the basis that they have direct contact with water from the reservoir.

A 1.0 g portion each of the gills and the intestines were pulverized in 9.0 mL of sterile 0.1% peptone water using sterilized pestle and mortar. A serial dilution of each of these homogenates was prepared. A 0.1 mL aliquot of the serially diluted homogenate was inoculated into sterilized plate count agar and Macconkey agar (for total heterotrophic bacterial count and total coliform count respectively), spread with a sterile bent glass rod (spread plates) and incubated at 37°C for 24 h as previously done by Okaeme *et al.* (1991) and Jones (1979). After incubation, the number of colonies were counted, calculated and recorded as colony forming units per gram (CFU g⁻¹) as shown in the following equation:

Colony Forming Units (CFU)= $\frac{\text{Colony count} \times \text{Dilution factor}}{\text{Inoculum volume}}$

Enumeration of total heterotrophic and total coliform bacteria in water samples: The 1.0 mL of water sample was serially diluted with sterile distilled water and 0.1 mL of each serial dilution was plated on sterilized media (standard plate count technique) as described by Jones (1979). This was spread with a sterile bent glass rod and incubated at 37°C for 24 h. Media used were, plate count agar and Macconkey agar for total heterotrophic bacterial count and total coliform count, respectively. After incubation for 24 h the number of colonies were counted using colony counter, calculated and recorded as colony forming units per ml (CFU mL⁻¹) as shown in the above equation.

RESULTS

Bacterial enumeration of water samples: Enumeration of water samples show that at the inlet, the microbial load was low $(4.2\times10^4 \text{ and } 1.2\times10^2 \text{ CFU mL}^{-1})$ for total heterotrophic and total coliform count, respectively. These load witness a sharp increase $(2.4\times10^6 \text{ and } 1.3\times10^8 \text{ CFU mL}^{-1})$ for total heterotrophic and total coliform count, respectively) at sample point 2. Microbial load at the outlet is lower $(3.1\times10^5 \text{ and } 1.4\times10^2 \text{ CFU mL}^{-1})$ than that at the upper coarse. The result are shown in Table 1.

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Table 1: Bacterial enumeration of water samples from Hadejia reservoir

	THC	TCC
Sampling points	CFU :	mL ⁻¹
Inlet	4.2×10^4	1.2×10^{2}
landing site	$2.4\!\! imes\!10^6$	1.3×10^{3}
landing site	$3.5\!\! imes\!10^6$	1.4×10^{3}
Damsite	2.5×10^{5}	2.2×10^{2}
Outlet	3.1×10^{3}	1.4×10^{2}

THC: Total heterotrophic count, TCC: Total coliform count, CFU mL⁻¹: Colony forming unit per milliliter

Table 2: Bacterial enumeration of fish samples from Hadejia reservoir

	THC	TCC
Samples	CFU	mL ⁻¹
Fish Gill (FG)	3.4×10^{3}	2.1×10^{2}
Fish Intestines (FI)	3.1×10^4	3.5×10^3

THC: Total heterotrophic count, TCC: Total coliform count, CFU g⁻¹: Colony forming unit per gram

Table 3: Results of biochemical characterization of isolates from water samples

Isolates	Gram stain	Ind	MR	VP	CU	MO	H ₂ S on TSI	Amylase	OX	Organisms
1	-	+	+	-	+	+	-	+	+	A.h
2	-	+	+	-	-	+	-	-	-	E.c
3	-	+	+	-	-	+	-	-	-	E.c
4	-	-	+	-	+	-	+	-	-	S. t
5	-	+	-	+	+	+	-	+	+	A.h
6	-	+	-	-	+	+	-	+	+	V.c
7	-	+	+	-	-	+	-	-	-	E.c
8	-	+	+	-	-	+	-	-	-	E.c
9	-	+	+	-	-	-	-	-	-	S
10	-	+	+	-	-	+	-	-	-	E.c
11	-	+	+	-	-	+	-	-	-	E.c
12	-	+	+	-	-	+	-	-	-	E.c
13	-	+	+	-	-	+	-	-	-	E.c
14	-	+	+	-	-	-	-	-	-	E.c

Ind: Indole, MR: Methyl red, VP: Voges proskauer, CU: Citrate utilization, MO: Motility, OX: Oxidase, A.h: Aeromonas hydrophila, E.c.: Escherichia coli and S.t: Salmonella typhi, S: Shigella species, V.c: Vibrio cholerae

Bacterial enumeration of fish samples: Bacterial load in the fish intestines is higher $(3.1\times10^4 \text{ and } 3.5\times10^3 \text{ CFU g}^{-1} \text{ for total heterotrophic and total coliform count, respectively) than that of gills <math>(3.4\times10^3 \text{ and } 2.1\times10^2 \text{ CFU g}^{-1} \text{ for total heterotrophic and total coliform count, respectively)}$. The results are shown in Table 2.

Biochemical characterization of isolates from water samples: Result of biochemical analysis shows that nine (64%) isolates out of fourteen isolates were *Escherichia coli*. Two (14%) out of the fourteen isolates were *Aeromonas hydrophila*. Vibrio cholera and Salmonella typhi were also isolated from the water samples. Although one isolate (7%) each of these bacteria was isolated, it still calls for concern because the duo are strict pathogens of public health importance. The results are shown in Table 3.

Table 4: Results of biochemical characterization of isolates from fish gills

Isolates	Gram stain	Ind	MR	VP	CU	MO	H_2S on TSI	Amylase	OX	Organisms
1	-	+	+	-	-	+	-	-	-	E.c
2	-	-	+	-	+	+	+	-	-	S.sp
3	-	+	+	-	+	+	-	+	+	A.h
4	-	+	+	-	+	+	-	+	+	A.h
5	-	+	+	-	-	+	-	-	-	E.c
6	-	-	+	-	+	+	+	-	-	S.sp
7	-	+	+	-	-	+	-	-	-	E.c
8	-	+	+	-	-	+	-	-	-	E.c
9	-	+	+	-	+	+	-	+	+	A.h
10	-	+	+	-	-	-	-	-	-	S
11	-	+	+	-	-	+	-	-	-	E.c

A.h: Aeromonas hydrophila, E.c.: Escherichia coli, S: Shigella species, S.sp: Salmonella species, Ind: Indole, MR: Methyl red, VP: Voges proskauer, CU: Citrate utilization, MO: Motility and OX: Oxidase

Table 5: Result of biochemical characterization of isolates from fish intestines

Isolates	Gram stain	Ind	MR	VP	CU	MO	H ₂ S on TSI	Amylase	OX	Organisms
1	-	+	+	-	-	+	-	-	-	E.c
2	-	+	+	-	+	+	-	+	+	A.h
3	-	-	+	-	+	+	+	-	-	S.sp
4	-	+	+	-	+	+	-	+	+	A.h
5	-	+	+	-	+	+	-	+	+	A.h
6	-	-	+	-	+	+	+	-	-	S.sp
7	-	+	+	-	-	+	-	-	-	E.c
8	-	+	+	-	+	+	-	-	-	E.c
9	-	+	+	-	-	-	-	-	-	S
10	-	+	+	-	+	+	-	-	-	E.c
11	-	+	+	-	+	+	-	+	+	A.h
12	-	+	+	-	-	+	-	-	-	E.c
13	-	+	+	-	-	+	-	-	-	E.c

A.h: Aeromonas hydrophila, E.c.: Escherichia coli, S: Shigella species, S.sp: Salmonella species, Ind: Indole, MR: Methyl red, VP: Voges proskauer, CU: Citrate utilization, MO: Motility and OX: Oxidase

Biochemical characterization of isolates from fish gills: Biochemical analysis of isolates from fish gill shows that five (46%) out of eleven isolates were *Escherichia coli*, three (27%) were *Aeromonas hydrophila*, two (18%) were *Salmonella* species and one of the isolate is *Shigella* species (Table 4).

Biochemical characterization of isolates from fish intestines: Table 5 shows the results of biochemical characterization of isolates from the fish intestines. Six (46%) isolates were *Escherichia coli*, four (31%) were *Aeromonas hydrophila*. Two (15%) out of the thirteen isolates were *Salmonella* species and one isolate is *Shigella* species.

DISCUSSION

Quantitative analysis of bacterial isolates at the reservoir reveals lower load of bacteria at the inlet (sampling point 1) compared to the load at sampling point 2 and 3 (Table 1). This may be due to the fact that much anthropogenic activities e.g., washing, bathing are carried out at sampling

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point 2 and 3. This load tends to decrease as the water flows down the lower course. The outlet (sampling point 5) after the dam site has the lowest bacterial load (Table 5). These differences in bacterial load agrees with the fact that the presence of bacteria in natural aquatic ecosystem is dependent upon the rate of contamination and the equilibrium that is established between bacterial proliferation in that environment and the rate of their elimination (Lejeune *et al.*, 2001).

Total coliform count in all the sampling point is higher than that of WHO standard (1.0×10⁸ CFU/100 mL). This may be because most people in this part of the world see reservoirs, streams, rivers as dumping ground.

Fish samples were seen to harbor *Aeromonas hydrophila*, *Escherichia coli*, *Salmonella* species and *Shigella* species. This result agrees with Mitchel (1972) who states that fresh water fish may harbor human pathogens after exposure to contaminated water or food sources.

Fish intestines harbour more bacterial load (3.1×10⁴ CFU g⁻¹) than fish gills (3.4×10⁸ CFU g⁻¹). This result also agrees with Apun *et al.* (1999) who observed in his study that the intestines of all fish examined harbored most number of bacterial species.

CONCLUSION

Bacteriological analysis of the reservoir reveals high load of bacteria and some of these are known to be pathogenic to both fish and humans. It is therefore recommended that fish caught from the reservoir should be properly boiled before consumption to avoid zoonotic diseases. High percentage of *Escherichia coli* calls for further study to identify the different strains of *E. coli* involved as some are known to be pathogenic e.g., *E. coli* 0157:H7. Contamination of water body should be discouraged through public enlightenment.

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