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Occurrence of bacterial infection in two commonly cultured fish species on two fish farms in southern Ghana

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Abstract

Bacterial infection in fish is very important to the fish, the fish farmer and the consumer. The study evaluated the presence of pathogenic and non-pathogenic bacterial species in cultured *Oreochromis niloticus* and *Clarias gariepinus*. A total of Fifty-five (55) individuals *O. niloticus* and *C. gariepinus* with weight between 14.1 to 389.5 g and standard length between 7.0 to 35.4 cm were sampled from two farm at Ashaiman and Akosombo for the examination of presence of pathogenic bacteria in their tissues. Tissues from the gill, stomach and liver were excised under aseptic conditions. These were inoculated in Brain Heart Infusion broths (BHI) and incubated for 24 hours at 37 ^{o}C then subsequently streaked on MacConkey Agars and Blood Agars (Supplemented with 5% sheep blood). The MacConkey and Blood Agars were also incubated for 24 hours at 37 ^{o}C. Bacteria colony that grew on the MacConkey and Blood Agars were picked with an inoculation loop and taken through a series of standardized test for bacteria identification. The test used were gram staining and biochemical test such as Indole, Catalase, Oxidase, Urease, Simmons Citrate and Sulphur Indole Motility tests. Bacterial species belonging to the genus *Shigella*, *Streptococcus, Klebsiella, Pasteurella, Neisseria*, *Edwardsiella*, *Bacillus*, *Pseudomonas*, *Staphyloccocus*, *Yersinia, Enterobacter,Citrobacter, Escherichia, Flavobacterium, Streptobacillus,* and *Proteus* were isolated from the two fish species sampled from the two sites. The presence of these pathogens means the potential outbreak of serious fish disease exists for fish farms, consumers in Ghana and worldwide.

Keywords: bacterial infection, fish farm, fish disease, *O. niloticus*, *C. gariepinus*, Ghana

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Introduction

Aquaculture has become one of the fastest developing source of animal protein to humans and animals due to dwindling wild fish stocks around the world and in particular Ghana (Ashitey and Flake, 2010; Al-Harbi and Uddin, 2005). Ghana's fish production has been fluctuating but generally on the decline since 2000 from 460,000 metric tons down to 436,000 metric tons in 2008. While the national average fish requirement is approximately 800,000 metric tons annually, the domestic fish catch (production) and imports only provide about half of this requirement (Ashitey and Flake, 2010). It is estimated that fish provides at least 50% of total animal protein intake in some small island developing states, as well as in Bangladesh, Cambodia, Equatorial Guinea, French Guiana, thpdfe Gambia, Ghana, Indonesia and Sierra Leone (FAO, 2009). In Ghana the species of fish that are commonly cultured are *Oreochromis niloticus* and *Clarias gariepinus* and sometimes *Heterotis niloticus* (FAO, 2010). Pathogenic bacteria are of most importance in fish health since their activities could lead to mass mortality of cultured finfish or shellfish species. Diseases associated with cultured fish can be divided into infectious and non-infectious.

Non-infectious disease occurs due to hereditary, nutritional deficiencies and poor management practices (Lucas and Southgate, 2003). Infectious disease may be caused by viral, bacterial, fungal or parasites pathogens (Lucas and Southgate, 2003). Infectious and non-infectious diseases pose serious threats to the fish in terms of their survival and growth rates. The World Bank reported that financial losses associated with fish disease were in the range of US$ 3 million per annum (Faruk *et al.*, 2004).

Effective water management in a fish holding facility is one of the important factors contributing to the success of fish culture (Hossain *et al.,* 2006). Ahmed *et al.* (2009) found that seasonal variations in pH, temperature and dissolved oxygen play important roles in the multiplication of pathogens thus leading to diseases in fish. Low or rapid changes in water temperature, rapid or prolonged depression of pH, low alkalinity and low dissolved oxygen are seasonal aggregations of fish diseases (Lilley *et al.,* 1992).

Majority of water borne pathogenic microorganisms enter water courses as a result of faecal and waste water contamination (Adewoye and Adegunlola, 2010). Bacteria present in the aquatic environment influences the composition of the gut microbiota and at the same time, those in the gut influence the environment microbiota (Apún-Monila *et al*., 2009). Tilapias are known to harbour bacterial flora in their guts (Sugita *et al.,* 1985). Al-Harbi and Uddin, (2005) showed that bacteria species isolated from the intestine of a tilapia species are predominantly Gram-negative rods (87%).

High proportion of *Bacillus* spp. in the intestinal of fish species may show that intestinal environment is suitable for the given probiotic to settle and grow and also lead to harbour a great number of microbial cells of host intestine (Bagheri *et al.*, 2008).

The types of bacteria contaminants in the feed given to the cultured animal also influence the diversity of bacteria species found in guts of the fish. Bacterial infection also represents a limiting factor for the further development of aquaculture (Austin & Austin, 1999). Fish diseases caused by Aeromonads and Pseudomonads are considered to be the major bacterial problems facing aquaculture development causing mass mortalities, reduced production and low quality of aquatic organisms, both *Aeromonas spp*. (*A. hydrophila, A. sobria and A. caviae*) and *Pseudomonas spp.* (*P. fluorescens, P. putida and P. aeruginosa*) have been implicated in severe outbreaks among *O. niloticus* in fish hatcheries (Ahmed and Shoreit, 2001).

Bacterial species isolated from fish could have serious health issues for humans who consume or ingest them. Some of the bacteria species that are isolated from the gut of fish are faecal coliforms; and this is especially characteristic of farms where there is little or no biosecurity. Contamination of hands and surfaces during cleaning and evisceration of fish is a common route for pathogenic infection in humans (Adedeji *et al.*, 2011). The objectives of this research were to determine the presence of bacterial species in *O. niloticus* and *C. gariepinus*, and which organ(s) was more susceptible to bacterial infection.

Materials and methods

1.1. Experimental area and fish species

Ashaiman Demonstration Farm of Ministry of Food and Agriculture (Site A) (N 05°41.986'and W 000°03.226') is located in the Ashaiman Metropolis of the Greater Accra Region of Ghana and is about 5 km from the Ashaiman main market. Aquaculture Research Development Centre of Water Research Institute (Site B) (N 06° 16.982' and E 000° 03.365') is located at Akosombo in the Eastern Region close to Lake Volta Dam, about 100 km north-east of Accra. The research was carried out on the two fish species (*O. niloticus* and *C. gariepinus*) commonly cultured on these two sites.

1.2. Physico-chemical characteristics

Temperature (°C), turbidity (NTU), pH and dissolved oxygen (mgl^{-1}) of the ponds were measured insitu with a handheld thermometer, turbidimeter, pH meter and oxygen meter, respectively. All these measurement were done ones a month at 10 am.

1.3. Biological sampling and analysis

1.3.1. Fish sampling

Cast net were used to collect fish samples from two earthen ponds each at Akosombo and Ashaiman. Five tilapia and catfish each were randomly selected at each site and sampling time. The length and weight were measured with measuring board and electronic balance respectively. After which they were placed in a waterproof plastic bag and transported at 4°C in an ice chest to the laboratory.

1.3.2. Fish tissue sampling and analysis

Tissues from gills, stomach and liver were sampled aseptically within six (6) hours of fish sampling. The excised tissues were placed into Brain Heart Infusion broths (BHI) and then incubated for 24 hrs at 37 °C. These were inoculated unto sterile MacConkey (MA) and Blood agars (BA) and incubated for 24 hrs at 37 °C with control media. Bacteria colonies and cells were identified by physical characterization and staining. Pure colonies were then taken through a series of standardized biochemical tests including SIM, Urease and Simmon citrate, oxidase, catalase, Galactose, D-Glucose, D-Sorbitol, I (meso) Inositol, L-Arabinose, L-Rhamnose, Salicin, Dulcitol, Raffinose, Dextrose and Fructose) which have been sterilized by tindalization.

1.3.3. Bacterial identification

Bergey's Manual of Determinative Bacteriology (Buchanan and Gibson, 1974), Quinn *et al.* (1994)'s Clinical Veterinary Microbiology and Bacteria diseases of Fish by Inglis *et al.* (1994) were used to analyze the various biochemical reactions in order to classify the bacteria to the genus and species levels.

1.4. Statistical analysis

Calculated means of water quality variables were tabulated. Mean condition factors (K) were estimated from mean weight-length-relationship ($K = 100 \times W/L^3$). Student-*t* test was used to test the mean significant difference of bacterial occurrence between the Akosombo and Ashaiman, gill, stomach and liver, the significant level was set at $P < 0.05$.

Results

1.5. Physicochemical parameters

From Table 1a the pH ranged from 5.2 ± 0.1 to 6.2 ± 0.1 at Akosombo while those of Ashaiman ranged between 6.9 ± 0.1 and 7.3 ± 0.1. Temperature varied from 29.3 ± 0.1 to 31.1 ± 0.1 °C for *O. niloticus* at Akosombo while those of Ashaiman varied from 27.0 ± 0.1 to 28.0 ± 0.2 °C. The pond turbidity value ranged from 21.00 ± 0.10 to 90.00 ± 1.00 NTU at Akosombo while those of Ashaiman varied from 7.14 ± 0.01 to 10.00 ± 1.00 NTU. Dissolved oxygen levels in the *O. niloticus* varied from 2.95 ± 0.05 to 8.34 ± 0.01 mgl^{-1} at Akosombo while those for Ashaiman ranged from 4.06 ± 0.02 to 7.11 ± 0.02 mgl^{-1}.

Table 1a: Mean monthly water quality values of *O. niloticus* pond

Site/ month	pH	Temperature (^oC)	Turbidity (NTU)	Dissolved oxygen (mgl^{-1})
Akosombo				
October	5.2±0.1	29.3±0.1	21.00±0.10	2.95±0.05
November	6.2±0.1	31.1±0.1	37.10±0.10	4.43±0.01
December	6.2± 0.1	29.8± 0.1	90.00±1.00	8.34±0.01
Mean±se	**5.9± 0.3**	**30.1±0.5**	**49.40±20.80**	**5.24±1.61**
Ashaiman				
October	7.3±0.1	27.0±0.1	10.00±1.00	7.11±0.02
November	7.3±0.1	27.0±0.1	7.14± 0.01	7.11± 0.02
December	6.9±0.1	28.0±0.2	8.13±0.03	4.06±0.02
Mean±se	**7.2±0.1**	**27.3± 0.3**	**8.42±0.84**	**6.09±1.02**

From Table 1b the pH ranged from 6.4 ± 0.1 to 6.4 ± 0.1 at Akosombo while those of Ashaiman ranged between 6.8 ± 0.1 and 7.0 ± 0.1. Temperature varied from 28.0 ± 0.1 to 29.3 ± 0.1 ^oC for *C. gariepinus* at Akosombo while those of Ashaiman varied from 27.3 ± 0.1 to 27.5 ± 0.1 ^oC. The pond turbidity value ranged from 228.00 ± 4.00 to 370.00 ± 10.00 NTU at Akosombo while those of Ashaiman varied from 200 ± 10.00 to 320.00 ± 5.00 NTU. Dissolved oxygen levels in the *C. gariepinus* varied from 4.32 ± 0.02 to 5.41 ± 0.01 mgl^{-1} at Akosombo while those for Ashaiman ranged from 3.05 ± 0.01 to 8.12 ± 0.02 mgl^{-1}.

Table 1b: Mean monthly water quality values of *C. gariepinus* pond

Site/ month	pH	Temperature (^oC)	Turbidity (NTU)	Dissolved oxygen (mgl^{-1})
Akosombo				
October	6.4±0.1	28.0±0.1	370.00±10.00	4.98±0.01
November	6.4±0.1	29.3±0.1	228.00±4.00	4.32±0.02
December	6.2±0.1	28.6±0.1	280±6.00	5.41±0.01
Mean±se	**6.3±0.1**	**28.6±0.4**	**292.70±41.50**	**4.90±0.32**
Ashaiman				
October	6.9±0.1	27.5±0.1	270±1.00	4.06±0.02
November	7.0±0.1	27.3 ± 0.1	200.0±10.00	8.12±0.02
December	6.8±0.1	27.5±0.1	320±5.00	3.05±0.01
Mean±se	**6.9±0.1**	**27.4±0.1**	**263.30±34.80**	**5.08±1.55**

1.6. Bacterial species isolated

From Table 2a, *E. faecalis, S. saprophytica* and *E. aerogenes* were not isolated from the gill and stomach of *O. niloticus* from Ashaiman but was isolated from the liver of *O. niloticus*. *S. iniae, Bacillus brevis, P. gallinarum, F. devorans, C. diversus* and *E. ictaluri* were not isolated from any of the three tissues of the fish from Ashaiman. In comparison to Akosombo *S. saprophytica, P. haemolytica, E. coli, P. mirabilis* and *P. mallei* are the species of bacteria that were also isolated from the gill, stomach and liver of *O. niloticus*. Bacteria species such as *E. cloacae* was isolated from the gill and stomach of *O. niloticus* from both Ashaiman and Akosombo.

Table 2a: Bacteria diversity in the tissues of *O. niloticus*

Bacterial species	**Akosombo**						**Ashaiman**					
	Gill		Stomach		Liver		Gill		Stomach		Liver	
	No.	%	No.	%	No.	%	No.	%	No.	%	No	%
S. iniae			3	37.5	2	22.2						
E. faecalis	1	10									1	14.3
S. saprophytica											1	14.3
Bacillus brevis					1	11.1						
E. cloacae	2	20	1	12.5			4	44.4	1	10		
E. aerogenes	1	10									1	14.3
P. gallinarum					2	22.2						
P. haemolytica									1	10		
S. rubidaea					1	11.1						
Shigella sp			1	12.5					1	10	1	14.3
K. pneumonia	3	30			1	11.1	4	44.4	2	20	2	28.6
F. devorans	1	10	1	12.5	1	11.1						
C. diversus	2	20										
E. coli							1	11.1	3	30		
P. mirabilis									1	10	1	14.3
E. ictaluri			2	25	1	11.1						
P. mallei									1	10		
Total	**10**		**8**		**9**		**9**		**10**		**7**	

In Table 2b, *S. iniae*, *Staphylococcus sp., Bacillus laterosporus, P. haemolytica, Shigella sp., E. coli, S. moniliformis, E. ictaluri* and *P. mallei* were isolated only at Ashaiman, while *Neisseria sicca, E. aerogenes, Y. pestis, Y. enterocolitica, Flavobacterium sp.* and *P. mirabilis* were isolated only for Akosombo.

Table 2b: Bacteria diversity in the tissues of *C. gariepinus*

Bacterial species	**Akosombo**						**Ashaiman**					
	Gill		Stomach		Liver		Gill		Stomach		Liver	
	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
S. iniae									1	14.3	2	22.2
Staphylococcus sp											1	11.1
Neisseria sicca	3	25										
B. laterosporus									1	14.3	1	11.1
E. cloacae	6	50	6	60	6	66.7	3	42.9				
E. aerogenes			1	10								
P. haemolytica							1	14.3	2	28.6		
Y. pestis			1	10								
Y. enterocolitica	1	8.3	1	10								
Shigella sp											1	11.1
K. pneumoniae	1	8.3			2	22.2	1	14.3				
Flavobacterium sp.	1	8.3										
E. coli							2	28.6	1	14.3	1	11.1
S. moniliformis											1	11.1
P. mirabilis			1	10	1	11.1						
E. ictaluri									2	28.6	1	11.1
P. mallei											1	11.1
Total	**12**		**10**		**9**		**7**		**7**		**9**	

1.7. Biochemical reaction of bacterial isolates

Table 3: Biochemical reactions of identified bacterial species

Species/Test	Indole	S. Citrate	Urea	Motility	H_2S	Catalase	Oxidase	D-Glucose	D-Mannitol	Dulcitol	Salicin	Dextrose	Inositol	D-sorbitol	L-Arabinose	L-Rhamnose	Raffinose	Fructose	Galactose	Gram reaction	Shape	Haemolysis
S. iniae	-	-	-	-	-	-		+	+	-	+	+	-	-	-	-	-	+	+	+	c	+
E. faecalis	-	-	-	+	-	+		+	+	-	+		-	+	-	+	+	+	+	+	c	+
Staphylococcus sp.	-	-	+	+	-	+		+	+	-	+	+	-	-	-	+	-	+	+	+	c	-
S. saprophytica	-	-	+	+	-	+		+	d	-	d	+	-	-	-	-	-	+	-	+	c	-
Neisseria sicca	-					+		+	-	+	+	+	-	+	-	+	-	+	+	-	c	+(α)
Bacillus brevis	-					(-)		+	+	-	+		-	+	-	+	+	+	+	+	b	+
B. laterosporus	-	-	-	-	-	+		+	-	+	+	-	+	+	+	-	-	+	+	+	b	-
Enterobacter cloacae	-	+	d	+	-	+	-	+	+	-	+	+	+	+	-	+	+	+	+	-	cb	-
***E. cloacae*[2]**	-	+	+	+	-	+	-	+	+	-	-	+	(-)	+	-	+	+	+	(+)	-	b	-
***E. cloaccae*[3]**	-	+	+	+	-	+	-	+	+	+	+	+	+	+	(+)	+	+	+	+	-	b	-
E. areogenes	-	+	-	(-)	-	+	-	+	+	(+)	+	+	+	+	+	+	-	+	+	-	b	+(α)
***E.aerogenes*[2]**	-	+	-	+	-	+	-	+	+	-	+	+	+	+	+	+	+	+	+	-	b	+(α)
***E. aerogenes*[3]**	-	+	-	+	-	+	-	+	+	-	+	+	+	+	(-)	+	+	+	+	-	b	+(α)
Pseudomonas mallei	-	-	-	-	-	(+)	+	+	+	-	-		-	-	-	-	-	+	-	-	b	+

LEGEND: b = bacillus, + = most strains positive, - = most strains negative, (+) = few positive, (-) = few negative, c = cocci, cb = cocco bacilli, dc = diplococci, d = delayed reaction, [2,3] = same species but with some different reactions, (α) = alpha haemolysis and (β) = beta haemolysis

Table 5- Biochemical reactions of identified bacteria species (Continued)

Species/Test	Indole	S. Citrate	Urea	Motility	H_2S	Catalase	Oxidase	D-Glucose	D-Mannitol	Dulcitol	Salicin	Dextrose	Inositol	D-sorbitol	L-Arabinose	L-Rhamnose	Raffinose	Fructose	Galactose	Gram reaction	Shape	Haemolysis
Pasteurella gallinarum	-	+	-	-	-	+	+	+	(+)	d	+	+	+	+	+	+	+	+	+	-	b	-
P. haemolytica	-	(-)	-	+	-	+	+	+	(+)	+	+	+	-	+	+	+	d	+	+	-	b	+(β)
***P. haemolytica*[2]**	-	(-)	-	+	-	+	+	+	(+)	+	+	+	+	+	+	+	d	+	+	-	b	+(β)
P. haemolytica	-	-	-	-	-	+	+	+	+	+	+	+	+	+	+	+	d	+	+	-	b	+
Yersinia pestis	-	-	-	-	-	+	-	-	+	+	+	-	+	(+)	-	+	+	+	+	-	b	-
Y. enterocolitica	+	-	-	-	-	+	-	+	+	d	d		d	+	+	+	+	+	+	-	b	-
Serratia rubidaea	-	+	-	(+)	-	+	-		(-)	-	-		+	-	-	-	d	-	-	-	b	-
Shigella sp.	-	-	-	-	-	-	-		-	-	-		-	-	-	-	-	-	-	-	b	-
***Shigella sp.*[2]**	-	-	-	-	-	-	-		-	-	d		-	-	-	-	-	+	-	-	b	-
Klebsiella pneumoniae	-	+	(-)	-	-	+	-	+	+	d	+		+	+	-	+	+	+	d	-	b	+(α)
***K. pneumoniae*[2]**	-	+	d	(+)	-	+	-	+	+	+	+		+	+	-	+	+	+	+	-	b	-
Flavobacterium devorans	-	+	-	+	-	+		+	-	d	+	-	-	+	+	-	-	+	+	-	cb	+(β)

LEGEND: b = bacillus, + = most strains positive, - = most strains negative, (+) = few positive, (-) = few negative, c = cocci, cb = cocco bacilli, dc = diplococci, d = delayed reaction, [2,3] = same species but with some different reactions, (α) = alpha haemolysis and (β) = beta haemolysis

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Flavobacterium sp	-					+		+	-	-	+	+	-	-	+	-	-	+	+	-	cb	-
Citrobacter diversus	+	+	(-)	+	-	+	-		+	(-)	-		-	+	-	+	+	+	+	-	b	-
Escherichia coli	+	-	-	(-)	-	+	-	+	+	d	+	+	-	+	+	+	+	+	+	-	b	-
***E. coli*[2]**	+	-	-	-	-	+	-	+	+	+	-	+	-	+	+	+	-	+	+	-	b	+(α)
***E. coli*[3]**	+	-	-	-	-	+	-	+	+	-	d	+	-	+	-	+	+	+	+	-	b	-
Streptobacillus moniliformis	-	-	-	-	-	-		+	-	+	+	-	-	+	+	+	-	+	+	-	b	-
Proteus mirabilis	-	+	+	+	(-)	+	-	+	-	-	-	-	-	-	-	-	-	+	+	-	dc	-
Edwardsiella ictaluri	-	-	-	-	-	+	-	+	-	-	-		(+)	-	-	-	-	-	-	-	b	+

LEGEND: b = bacillus, + = most strains positive, - = most strains negative, (+) = few positive, (-) = few negative, c = cocci, cb = cocco bacilli, dc = diplococci, d = delayed reaction, [2,3] = same species but with some different reactions, (α) = alpha haemolysis and (β) = beta haemolysis

Discussions

Water quality parameters are indicators of excellent and poor living conditions of any fish. When pond conditions are poor they inhibit and reduce the suitability of environment for the growth and reproduction of fish (Bolorunduro and Abdullah, 1996). Good or optimum pond conditions promote growth and reproduction for fish and reduce their susceptibility to diseases (Rakocy and McGinty, 1989). Mean temperatures of 30.1 ± 0.5 and 28.6 ± 0.4 for the tilapia and catfish ponds, respectively from Akosombo were within the suitable range (25-32 ^{o}C) for the culture of these species (Bolorunduro and Abdullah, 1996). In comparison with those from Ashaiman the mean temperatures for the entire period of the study were 27.3 ± 0.3 ^{o}C and 27.4 ± 0.1 ^{o}C for tilapia and catfish ponds, respectively, which were also within the suitable temperature range but lower than those recorded from Akosombo. The mean pH, 5.9 ± 0.3 and 6.3 ± 0.1 of *O. niloticus* and *C. gariepinus* ponds respectively (Akosombo) for the period of study were moderately acidic. These are below the optimum (6.9 to 9.9) levels for tropical fish culture (Bolorunduro and Abdullah, 1996); therefore productivity of the fish species will be lower. In comparison with Ashaiman, mean pH of 7.2 ± 0.1 and 6.9 ± 0.1 were recorded for *O. niloticus* and *C. gariepinus* ponds respectively for period of study. The pH was within optimum range desirable for high fish production. The mean dissolved oxygen levels (Table 1a & b) for period from both tilapia and catfish ponds (both Akosombo and Ashaiman) were not at levels (less than 1 mgl^{-1}) that would have been lethal to the fish. Desired levels (above 5 mgl^{-1}) for response to fast feeding, growth and reproduction were recorded in *O. niloticus*, and *C. gariepinus* ponds from Akosombo (5.24 ± 1.61 mgl^{-1}) and Ashaiman (6.09 ± 1.02 mgl^{-1}), respectively. The mean turbidity values (Table 1a & b) for the entire period showed that phytoplankton production and sediment loads in the water were above the desired amount (turbidity value of less than 5 NTU) for fish production (Bolorunduro and Abdullah, 1996). Gram-negative bacterial species dominated the isolates from the gill, stomach and liver of *O. niloticus* and *C. gariepinus* for Akosombo and Ashaiman. *Flavobacterium spp.* is known to cause Bacteria Gill Disease (BGD) in salmonids. BGD has been reported to have caused significant annual mortalities of salmonid in many European countries (Inglis *et al.,* 1994). The isolation of *Flavobacterium sp.* from the catfish suggests that there might be another species belonging to the genus in the internal and external environment of the fish but since the water environment was not posing serious threats to fish as a result of the mean excellent condition of fish few isolates were made (Inglis *et al.*, 1994; Barnham and Baxter, 2003). Studies by Ampofo and Clerk (2010) also reported the presence of *Flavobacterium spp.* in cultured fish species in Ghana. Flick (2008) indicated that *S. iniae* bacteria pose the most serious disease threat to the tilapia industry. Therefore the isolation of this well-known pathogenic bacterium of fish from the stomach and liver of tilapia and catfish from Akosombo and Ashaiman is of concern. *S. iniae* is known to cause haemorrhagic lesions in fish. *Pasteurella spp.* are known to be pathogenic to fish and have mostly been isolated from the fish feed, freshwater, marine and brackish environment (Inglis *et al.,* 1994; Jones, 1999; Ampofo and Clerk, 2010). Therefore from the result, the possibility that *P. gallinarum* and *P. haemolytica* are peculiar to the environment from which the fishes were sampled exists. The presence of these species in fish sampled from the two areas could pose threats to the health of the fish in the case of physiological and environment imbalance. *C. diversus* was the only species from the genus that was isolated from the organs of the *O. niloticus* from Akosombo in December, which does not suggest the absence of *C. freundii* in the environment. Currently, there is no information on the pathogenicity of *C. diversus* to fish therefore they might be considered as commensals in the fish's environment.

According to Versschuere *et al*. (2000) gram-positive *Bacillus* spp. are generally more efficient in converting organic matter back to CO_2 than are gram-negative bacteria, which would convert a greater percentage of organic carbon to bacterial biomass or slime. Therefore the occurrence of *Bacillus spp.* (*B. brevis* and *B. laterosporus*) in high numbers in the pond environments of Akosombo and Ashaiman could be of immense benefit to the fish by changing the host-related or ambient microbial community (Sanders *et al*., 2003; Gullian *et al*., 2004; Apún-Monila *et al*., 2009). *Shigella spp.*, and *Klebsiella pneumoniae* are considered as commensals in the environment of the fish but might pose serious health threat to humans who may consume fish that have high contamination of this species (Ampofo and Clerk, 2010). *E. aerogenes* and *Enterobacter cloacae* were the two species that were isolated from the fishes sampled. Ampofo and Clerk (2010) also reported the presence of *Enterobacter spp* in cultured fish species in Ghana. *Enterobacter cloacae* had the most occurrences throughout the period of the study from both farms. The isolation of bacteria species from the genus *Citrobacter*, *Enterobacter*, *Streptococcus*, *Escherichia, Pseudomonas* and *Klebsiella, Staphylococcus* and *Bacillus* from the fishes suggests that these potential human pathogens are present on fish farms in Ghana. This means that fish and fish products from these farms could pose serious health threat to humans when these bacteria are consumed in large quantities (Ampofo and Clerk, 2010). There is lack of biosecurity on both farms hence farm workers are usually seen handling fish and farm equipments without concern for their own health and that for the farmed animals. Lack of biosecurity on fish farms could have also contributed to the introduction of some of the human pathogens that were isolated on the farms and there is the potential for cross contaminations between humans and the fish. Presence of *E. coli* in water or food indicates the possible presence of other causative organisms of many gastro-intestinal diseases (Frazier and Westhoff, 1985; DHSS, 1991). *Shigella spp.* is also known to cause shigellosis in humans. Raj and Liston (1961) found that some pathogenic and potentially pathogenic microorganism including *E. coli, Staphylococcus* and some anaerobes survive when uncooked and precooked fish foods were stored at freezing point. With the advent of grilled tilapia and 'banku' joints and the demand for fresh tilapia and catfish in Ghana and other part of the world there is the danger of the transfer of these pathogens to both human and animals. There was no significant difference in the occurrence of both pathogenic and non-pathogenic bacteria between the gill, stomach and liver. This is an indication that all the organs are susceptible to bacterial infections. There were also no significant differences in the occurrences of bacteria (pathogenic and non-pathogenic) between the two farms. From the results presented there were high prevalence and diversity of bacterial species in the two species of fish examined. The institution of biosecurity as a standard for the establishment of fish farm is highly recommended.

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