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Distribution and Factors Associated with Occurrence of Listeria monocytogenes in Table Size African Catfish, Pond Water and Sediment in Two Ecological Zones in Nigeria

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Abstract

Occurrence of *Listeria monocytogenes* (LM) and associated factors affecting its distribution in Oyo state, Nigeria was investigated in 2016. A total number of 972 (756 fish organs, 108 water and 108 sediments) samples were collected and examined for LM using molecular method. Descriptive statistics, t-test, Manny-Whitney U test and ANOVA were employed to analysed the data generated. It was found out, among other that 61 out of 972 samples had listeria infection. Forest zone had higher percentage of infection (66.67%) than the derived savannah zone (25.93%). However, there was no significant difference in LM occurrence between the sexes (p=0.333) of the fish. Although, there was significant difference in body weight (p=0.005). Out of 61 biochemically characterized isolates, only 18(29.51%) using *iap* primer and 12(19.67%) using *hly-A* primer were confirmed to be *L. monocytogenes* by PCR method. There was significant difference in the occurrence of *L. monocytogenes* in organs (p=0.001) with the spleen having the highest occurrence. Thus, the study concluded that LM was well distributed in fish farms in the ecological zones. Also, the highest occurrence was observed in forest zone revealing body weight and fish length as possible predisposing factors.

Keywords: Listeriosis; fish farms; infection; predisposing factors

INTRODUCTION

Fish is one of the cheapest and major sources of animal protein with high polyunsaturated fatty acid and essential amino acids for riverine dwellers and consumers in general [1]. The highly perishable nature of fish encourages the growth of microorganism including food poisoning pathogens such as *Listeria monocytogenes* leading to mortality, economic losses and infections. African catfish, *Clarias gariepinus* is the most cultured fish in Nigeria and is widely accepted as "common man" fish in the market. Its wide acceptance is connected to some important features which include high quality flesh, hardy nature, high tolerance level of water characteristics, production performance and attraction of appreciable market values. Table size fish (adult) is often consumed in many homes, hotels and relaxation centres hence it has become the most important stage of the fish with greater contact with human populace [2].

Listeriosis is a serious foodborne disease caused by bacteria in the genus *Listeria,* which has ten species: *L. grayii, L. innocua, L. ivanovii, L. monocytogenes, L. seeligeri,* and *L. welshimeri, L. marthii, L. rocourtiae, L. weihenstephanensis,* and *L. fleischmannii* [3]. Among these species, only *L. monocytogenes* is capable of causing illness in both human and animal [4]. *Listeria monocytogenes* is a gram positive, facultative, non-sporing, rod-like, motile and flagellated bacterium. It is an ubiquitous organism capable of surviving at refrigerating temperature [5] and it is accountable for serious illnesses like septicaemia, central nervous system infection, intra-uterine infection, and meningitis in matured human, severe systemic infection in new born and/or unborn child while pregnant women suffer a mild influenza-like illness. *Listeria monocytogenes* has been reported to account for mortality rate of about 20% to 50% of infection in human or even 100% in laboratory animals [6-7]. However, investigation of incidence of *Listeria monocytognees* in fish is highly necessary.

Listeria monocytogenes has been isolated from several environmental samples such as soil, vegetation, and human and animal faeces [8-9], signifying the pervasive occurrence of the pathogen in nature. Its occurrences in fish and fish products varied with respect to the form of the products. Occurrence ranges slightly above 4% to as high as 88% in fishes [10], 0 to 51% in shelf fish [11], 7.5% in fresh fish in Iran [6, 12] and 25% in smoked fishes in Nigeria [13]. Although the occurrence of *Listeria monocytogenes* in several foods including fish and fish products have been investigated in some countries, there is dearth of information on the *L. monocytogenes* occurrence and associated factors in cultured fish in Oyo State, Nigeria hence, the need for this study.

The aim of the study was to investigate the distribution and factors associated with occurrence of Listeria species especially *L. monocytogenes* in table size African catfish raised in pond earthen Oyo State, Nigeria and highlight any differences in the occurrence and associated factors.

MATERIALS AND METHODS

Study area

The study was carried out in Oyo State, in the South-West geopolitical zone of Nigeria. It lies within Latitudes $7^{o}3'$ S and $9^{o}12'$ E and longitudes $2^{0}47'$ and $4^{0}23''$ [14]. The two ecological-vegetation division (forest and derived savannah zones) of Oyo State were adopted for this study [14-15]. The forest zone comprises 11 local government areas

in Ibadan metropolis in the southern part of the state while derived savannah zone comprises 22 local government areas in the northern part of the state [16].

Water and Fish organs collection

Farms in each zone were sub-grouped into Large, Medium and Small scales using stratified methods [17]. A total number of thirty-six (36) farms (12 Large, 12 Medium and 12 Small scales farms) were used [2]. Water, soil, and fish samples were collected from these farms and analyzed for *Listeria* species. The protocol of the study was subjected to ethical consideration and was approved by Animal Care Use and Research Ethics Committee, University of Ibadan, Ibadan, Oyo state, Nigeria with reference number UI-ACUREC/App/03/2017/008.

Water samples were collected using plastic bottles, fixed with Winkler A and B and transported to the Department of Aquaculture and Fisheries Management, University of Ibadan. A total number of one hundred and eight (108) (three fish per farm) fish were collected in early hours of the day (6:00 to 7:00 am) and transported to the laboratory in different plastic containers containing water from the respective ponds to avoid contamination. The fish were caught, condition (males and females were kept separate) to avoid stress and transported to the laboratory immediately. Seven (7) organs (liver, kidney, gill, intestine, flesh/muscle and spleen) were aseptically collected per fish and weighed into sterile universal bottles while skin samples were collected using skin swab [5]. Fish with body weight ranges between 300g to 1300g were used for this study as the group comprised stage of fish that are consumed by most individuals or processed for economic value. Organo-somatic index were determined using .

Water quality parameters

The water samples were taken and parameters were measured at an early hour (6:00hrs) of the day. Dissolved oxygen (DO) was determined by analytical method [18]. Water samples were fixed with 2 ml of Winkler A and B each. Exactly 100 ml of the fixed water was neutralized by addition of 2 ml of 10 sulphuric acid. Four drops of starch were added and titrated with 60% sulphuric acid. The DO level were calculated using , conductivity ($ms.cm^{-1}$) was measured with the aid of conductivity meter (LABTECH (R)] Model AVI-660 (Power: 220V, AC: 50 Hz: Sr./No. 376), nitrite (ppm) was measured using Auasol Nitrite kit (Nitrite High Level Test Kit; AE-207; MRP(Rs):1120; Rakiro Biotech Systems PVT Ltd, USA), pH was measured with the aid of a digital pH meter [LABTECH (R)] Model Photoic 20 (Power: 230V AC: 50 Hz: Sr./No. 1223) and temperature was measured using mercury thermometer.

Isolation of Listeria species

Sampled organs were pre-enriched in half-strength Fraser broth. One gram of each sample was added to universal bottles containing 9 mL of sterile half strength Fraser broth and supplements and incubated at 37°C for 24 hours. From the pre-enrichment culture media 0.1mL was transferred into 10 mL of full strength Fraser broth with supplements added and was incubated at 37°C for 24-48 hours (secondary enrichment) [4, 19]. The culture media were then streaked on PALCAM agar plates using wire loop and incubated for 48 hours. The wire loops were sterilized on spirit lamp before usage. The plates with a black sunken center and a black halo on a cherry-red background, following aesculin hydrolysis and mannitol fermentation were suspected to be listeria species colonies and selected for further confirmation and identification [4, 11]. The suspected colonies were sub-cultured on PALCAM agar and incubated for 24-48 hours to obtain pure colonies.

Macroscopic examination of culture plates

The presumptive identification of *L.* species was based on the cultural and evidence of aesculin hydrolysis or black – halo formation on PALCAM plates. The suspected organisms were kept on glycerol and PALCAM slants and stored in a freezer and refrigerator respectively, until use.

Biochemical test for Listeria species isolates

Biochemical tests adopted include gram stain reaction, beta-haemolysis, catalase reaction, sugar or carbohydrate fermentation (Rhamnose, Xylose, Lactose, Fructose and Mannitol) and motility at room temperature [20].

Confirmation of *L. monocytogenes*

Sixty-one (61) biochemically characterized isolates were further subjected to PCR method for identification of *L. monocytogenes* using primers specific to invasive associated protein (*iap*) and haemolysis (hlyA) [21] listed in table 1.

Table 1. Primers used for *Listeria monocytogenes* identification

Primers	Target gene	Length	Primer sequence	Amplification product bp
iap-F	Iap	20	5' ACAAGCTGCACCTGTTGCAG 3'	131
iap-R	Iap	20	5' TGACAGCGTGTGTAGTAGCA 3'	131
hlyA-F	hly	24	5'GCAGTTGCAAGCGCTTGGAGTGAA3	456
hlyA-R	hly	24	5'GCAACGTATCCTCCAGAGTGATCG 3	456

Source: adapted from Swetha *et al.* (2012).

DNA extraction

The *Listeria* isolates were suspended in 1.5 mL of enriched Fraser broths, grown on a shaker for 48 hours at 48°C, 4600 x g for 5 minutes. The pellets were re-suspended in 520 µL of TE buffer (10 mMTris-HCl, 1 mM EDTA, pH 8.0). Fifteen microliters of 20% SDS and 3 µL of Proteinase K (20 mg/ml) were added. The mixture was incubated for 1 hour at 37ºC, then 100 µL of 5 M NaCl and 80 µL of a 10% CTAB solution in 0.7 M NaCl were added and mixed [4]. The suspension was incubated for 10 minutes at 65 ºC and kept on ice for 15 minutes. An equal volume of chloroform: isoamyl alcohol (24:1) was added, follow by incubation on ice for 5 minutes and centrifugation at 7200 x g for 20 minutes. The aqueous phase was transferred to a new tube, isopropanol (1: 0.6) was added and DNA was precipitated at –20 ºC for 16 hours. The DNA was collected by centrifugation at 7200 x g for 10 minutes, washed with 500 µL of 70% ethanol, air-dried at room temperature for approximately 3 hours and finally dissolved in 50 µL of TE buffer [4].

Polymerase Chain Reaction (PCR)

PCR reaction cocktail consisted of 2.5 µl of 5x Go-Taqgreen reaction buffer, 0.75 µl of $MgCl_2$, 0.25 µl of 10 mM of dNTPs mix, 0.25 µl of 10 pmol each of forward and backward primers and 0.06 µl of Taq DNA polymerase (8,000U) (Promega USA) made up to 10.5 µl with sterile distilled water then 2 µl DNA template. The PCR was carried out in a GeneAmp 9700 PCR System (Applied Biosystem Inc., USA) Thermalcycler while the cycling conditions are shown in Table 2 [21].

Table 2. Cycling conditions used for *iap* and *hly-A* primer

S/No	Step	iap (L. monocytogenes)	HlyA (L. monocytogenes)
1.	Initial denaturation	95^0C / 5 min	95^0C / 5 min
2.	Final denaturation	95^0C/30 sec	94^0C/60 sec
3.	Annealing	56^0C/30 sec	61^0C/45 sec
4.	Initial extension	72^0C/30 sec	72^0C/60 sec
5.	Final extension	72^0C/7 min	72^0C/7mins
6.	Hold	4^0C	4^0C

Integrity

The integrity of the amplified band for *iap* and *hlyA* genes fragment was checked on a 1.5% Agarose gel ran at 110V for 1 hour to confirm amplification and pictures were taken under gel electrophoresis to indicate amplification using *iap* and *hlyA* specific primers.

Statistical analysis

The data were analyzed using both descriptive statistics (percentage) to express the prevalence of bacterium occurrence. The values of water quality paremeter, organo-somatic index were analysed using one-way analysis of variance (ANOVA). The difference in the means were separated using student t-test for values between the zones, Mann-Whitney U test for values between the sexes of the fish and Duncan Multiple Range Test for values among the organs at p = 0.05 with the aid of Statistical Package for Social Science (SPSS) version 20.

RESULTS

Water quality

The result of the water quality of the selected farms were shown in Table 3. Farms in forest zone of the state had higher DO (6.04 ± 1.32 mg/l) and pH (7.28 ± 0.51) compared with savannah zone (5.78 ± 1.61 mg/l) and (7.27 ± 0.53) respectively. More so, savannah zone had higher conductivity (2.17 ± 1.33 $ms.cm^{-1}$), nitrite (46.85 ppm) and temperature (25.64 ± 1.41 ^oC) while farms from the forest zone had lower values 1.42 ± 1.07 $ms.cm^{-1}$, 42.78 ± 19.48 ppm and 25.31 ± 1.16 ^oC respectively. There were significant differences in conductivity, nitrite, pH and temperature ($p<0.05$) except DO ($p=0.84$)

Table 3. Water quality of the fish farms in Oyo State, Nigeria

Parameters	Forest zone (n=27)	Derived Savannah zone (n=81)	t-test (Sig)
DO (mgL^{-1})	6.04±1.32	5.78±1.61	0.84
Conductivity ($ms.cm^{-1}$)	1.42±1.07	2.17±1.33	-2.97
Nitrite (NO2-) (ppm)	42.78±19.48	46.85±20.30	-0.93
pH	7.28±0.51	7.27±0.53	0.05
Temperature (^{0}C)	25.31±1.16	25.64±1.41	-0.12

Values are represented as mean ± standard deviation (P < 0.05).

The organo-somatic index of the sampled fish were shown in Table 4. Organ-somatic index of gill and kidney were highest in fish from forest zone (4.30±1.15 and 0.11±0.04) while liver and intestine were highest in fishes from derived savannah (2.66±1.07 and 2.23±0.85). Organ-somatic index of gill and kidney were not significantly different ($p>0.05$) while there were statistically significant difference in organ-index of liver and intestine ($p<0.05$).

Table 4. Organ-somatic of table size Africa catfish in Oyo State, Nigeria

Parameters	Forest zone (n=27)	Derived Savannah zone (n=81)	t-test (Sig)
Gill	4.30±1.15	3.54±1.18	2.97
Liver	1.57±0.56	2.10±0.85	-3.70
Kidney	0.11±0.04	0.10±0.04	1.02
Intestine	1.30±0.50	1.85±0.70	-4.44

Vales are represented as mean ± standard deviation (P < 0.05).

Table 5 shows that the female fish from forest zone had higher percentage of infection (72.73%) and intensity of infection (0.73) than male (62.50% and 0.63 respectively). In derived savannah, male fish had higher percentage and intensity of infection (29.27 % and 0.29 respectively) than females (22.50% and 0.23 respectively). There was no statistically significant difference of *L. monocytogenes* occurrence between the males and females (p=0.333). In addition, fish with total length of 51-60cm had higher percentage and intensity of infection (100% and 1.00 respectively) in forest zone and least was observed in fish with total length of 30 – 40cm (46.15% and 0.46 respectively). In derived savannah zone, the highest percentage and intensity of infection (31.71% and 0.32 respectively) was observed in fish with 41 – 50 cm total length while the least (11.76% and 0.12) were recorded in fish with 30 – 40cm total length.

Table 5. Occurrence of *Listeria* by length, weight and sex of farm raised *C. gariepinus* in Oyo State, Nigeria

	Parameter	No. of samples		No. of infection		Percentage		Intensity of infection		p-value
		F	S	F	S	F	S	F	S	
Sex	Male	16	41	10	12	62.50	29.27	0.63	0.29	p=0.333
	Female	11	40	8	9	72.73	22.50	0.73	0.23	
	Total	27	81	18	21	66.67	25.93	0.67	0.26	
Total length (cm)	30-40	13	17	6	2	46.15	11.76	0.46	0.12	p=0.099
	41-50	12	41	10	13	83.33	31.71	0.83	0.32	
	51-60	2	23	2	6	100.00	26.09	1.00	0.26	
	Total	27	81	18	21	66.67	25.93	0.67	0.26	
Body weight (g)	300-600	17	43	10	12	58.82	27.91	0.59	0.28	p=0.005
	601-900	8	35	6	7	75.00	20.00	0.75	0.20	
	≥ 900	2	3	2	2	100.00	66.67	1.00	0.67	
	Total	27	81	18	21	66.67	25.93	0.67	0.26	

Note: F = Forest zone, S = Derived savannah zone

(i) Intensity of infection = $\frac{\text{Number of infected samples}}{\text{Total samples examined}}$

More so, from the forest zone, fish with body weight of 900 g and above had highest percentage (100%) and intensity (1.00%) of infection and least (58.82% and 0.59), while in derived savannah zone, the highest percentage (66.67%) and intensity (0.67) of infection were observed in fish with body weight of 900g and above and lowest percentage (20.00%) and intensity (0.20) of infection in fish with body weight 601 – 900g. Hence, body weight significantly affected the percentage and intensity of infection (p=0.005) while sex and total length did not (p>0.05).

Table 6 shows that fish gill, kidney, skin, flesh and spleen of fish from forest zone had higher percentages of infection (22.22%, 22.22%, 33.33%, 22.22% and 11.11% respectively) while derived savannah zone had lower infection (3.70%, 11.11%, 7.40, 11.00 and 0.00%) respectively. Overall, organs of fish in forest zone had higher infection 15.86% compared with derived savannah (4.76). There are differences in the values of infection but there is no significant difference (p>0.05) as shown in Table 6.

Table 6. Distribution of Listeria species in fish by organs in Oyo state, Nigeria

Organ	Forest zone				Derived savannah zone			
	No. of sample	No. of Positive	%	Intensity of infection	No. of sample	No. of Positive	%	Intensity of infection
Gill	27.00	6.00	22.22	0.22	81.00	3.00	3.70	0.04
Liver	27.00	0.00	0.00	0.00	81.00	0.00	0.00	0.00
Kidney	27.00	6.00	22.22	0.22	81.00	9.00	11.11	0.11
Intestine	27.00	0.00	0.00	0.00	81.00	0.00	0.00	0.00
Skin	27.00	9.00	33.33	0.33	81.00	6.00	7.41	0.07
Flesh	27.00	6.00	22.22	0.22	81.00	9.00	11.11	0.11
Spleen	27.00	3.00	11.11	0.11	81.00	0.00	0.00	0.00
Total	189.00	30.00	15.87	0.16	567.00	27.00	4.76	0.05
p-value		0.842				0.503		

i) Intensity of infection = $\frac{\text{Number of infected samples}}{\text{Total samples examined}}$

Table 7 shows that pond water from forest zone had higher percentage (3.70%) and intensity (0.04) of infection while no infection was recorded in derived savannah zone. In the pond sediments, forest zone had higher percentage (7.41%) and intensity (0.07) of infection than 1.00% and 0.01 percentage and intensity of infection recorded in derived savannah respectively. Overall, percentage (5.56%) and intensity (0.06) of infection were higher in forest zone than 1.0% and 0.00 recorded in derived savannah. There were no significant difference in percentage of infection in pond water (p=0.221) and sediment (p=0.254).

Table 7. Distribution of *Listeria* in pond water and sediment in two ecological zones

Material	Forest zone				Derived savannah zone				p=value
	No. of sample	No. of Positive %		Intensity of infection		No. of Positive %		Intensity of infection	
Pond water	7.00	1.00	3.70	0.04	81.00	0.00	0.00	0.00	p=0.221
Sediment	27.00	2.00	7.41	0.07	81.00	1.00	1.23	0.01	
	54.00	3.00	5.56	0.06	162	1.00	0.62	0.00	

Note: Intensity of infection = $\frac{\text{Number of infected samples}}{\text{Total samples examined}}$

Table 8 shows that out of 61 isolates examined, only 18(29.51%) were confirmed *L. monocytogenes* using *iap* primer. Among the organs, spleen had highest occurrence (100%), and least was observed in kidney (20.00%). When using *hly-A* primer, only 12(19.67%) were confirmed *L. monocytogenes*. However, among the organs, *L. monocytogenes* were only identified from the skin and muscles (40.00%). There is significant difference in the occurrence of *L. monocytogenes* among the organs and between the *iap* and *hly-A* primers ($p<0.05$).

Table 8. Occurrence of *L. monocytogenes* using *iap* primer in two ecological zones.

Organs	Number of samples	*L. monocytogenes* using *iap* (%)	*L. monocytogenes* using *hly-A* (%)	Other Listeria species (%)
Skin swab	15	6 (40.00)	6(40.00)	9(60.00)
Kidney	15	3(20.00)	0(0.00)	12(80.00)
Muscles/flesh	15	6(40.00)	6(40.00)	9(60.00)
Spleen	3	3(100)	0(0.00)	0(0.00)
Gill	9	0(0.00)	(0.00)	9(100.00)
Water	1	0(0.00)	(0.00)	1(100.00)
Sediment	3	0(0.00)	0(0.00)	3(100.00)
Total	61	18(29.51)	12(19.67)	43(70.49)
p-value		0.002	0.001	

Figure 1 shows the PCR product with positive band (131bp) from skin, kidney, muscles and spleen. Isolates on Lanes 1 and 5, 4, 9, 12 and 15 were the only isolates that had equivalent and desired band which were isolates from skin, muscle, kidney and spleen using specific primer invasive associated protein (*iap*) that is targeting *iap* gene confirming it to be *L. monocytogenes*. Figure 2 shows the PCR product with positive band (456bp). Isolates on Lanes 1, 4, 9, and 15 were the only isolates that had equivalent and desired band which are isolates from skin and muscle using specific primer haemolysis (*hlyA*) that is targeting haemolysis gene confirming it to be *L. monocytogenes*.

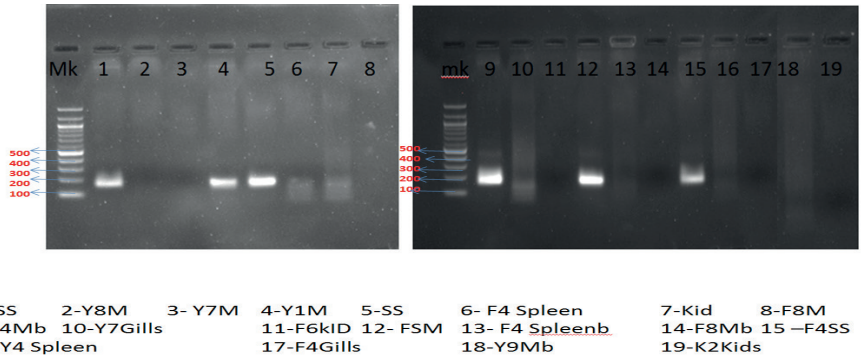

Figure 1. Standardization of PCR using assay for detection of Listeria monocytogenes (iap). Where: Lane 1= F2 Skin, Lane 2= F1 Muscle, Lane 3= F2 Gill, Lane 4 = Y1 Muscle, Lane 5 = O4 Skin, Lane 6 = F4 Skin, Lane 7 = F5 Kidney, Lane 8 = F6 Skin, Lane 9 = F6 Kidney, Lane 10 = F8 Muscle, Lane 11 = O9 Skin, Lane 12 = Y5 Muscle, Lane 13 = Y4 Kidney, Lane 14 = F4 Gill, Lane 15 = F4 Spleen, Lane 16 = Y7 Gill, Lane 17 = Y7 Muscle, Lane 18 = Y7 Kidney, Lane 19 = K7 Kidney.

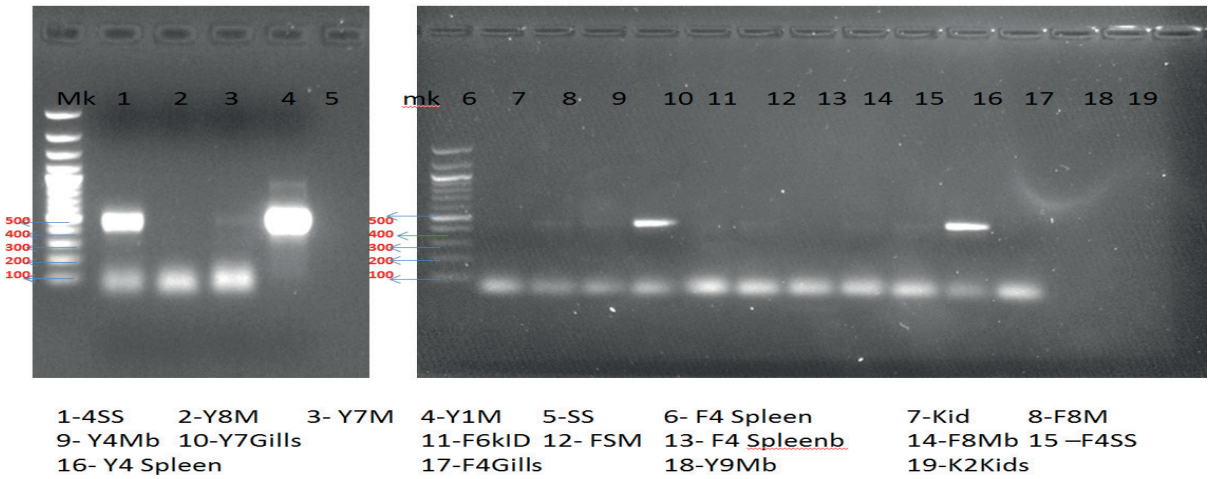

Figure 2. Standardization of PCR using assay for detection of *Listeria monocytogenes* (hly-A). Where: Lane 1= F2 Skin, Lane 2= F1 Muscle, Lane 3= F2 Gill, Lane 4 = Y1 Muscle, Lane 5 = O4 Skin, Lane 6 = F4 Spleen, Lane 7 = F5 Kidney, Lane 8 = F6 Skin, Lane 9 = Y5 Muscle, Lane 10 = F8 Muscle, Lane 11 = O9 Skin, Lane 12 = F6 Kidney, Lane 13 = Y4 Kidney, Lane 14 = F4 Gill, Lane 15 = F4 Skin, Lane 16 = Y7 Gill, Lane 17 = Y7 Muscle, Lane 18 = Y7 Kidney, Lane 19 = K7 Kidney.

DISCUSSION

This study describes the distribution and factors associated with occurrence of *Listeria spp* especially *L. monocytogenes* in table size African catfish, pond water and sediments in African catfish in Oyo state in Nigeria. Water is an important factor in fish farming as it predetermines the survival, growth and health of the fish since the entire life process of the fish wholly depends on the quality of its environment. DO, temperature and pH in this study fall within the recommend desirable values which is in agreement with works of [22-25]. The overall prevalence of *L. monocytogene*s in fish samples from Oyo State is similar to that reported for fresh fish [6, 26] but differ from 3% reported in fresh fish in Europe, 1%, and 56% of fresh fish on sale in Iceland [27]. The results further suggest that fish with longer total length have chance of being infected; this may be as a result of higher surface area. The infection is not sex related as there was no significant difference in the percentage of infection of the male to female. This suggests that the fish farms from the study area share similar management practices.

The result of the PCR showed that rate of occurrence of *L. monocytogenes* in fish sampled is high. The prevalence is in agreement with works of [26] who reported *L. monocytogenes* up to 21% from Salmon in USA, 22% from rainbow trout Switzerland, 30% in Salmon in USA and 23% from different species in USA fresh fish respectively. However, the present study observed lower percentage of *L. monocytogenes* compared to reports of [21, 28, 29] who reported up to 65% in India.

The significant difference in the occurrence with body weight and length further revealed that body weight and length of the fish are possible predisposing factors of *L. monocytogenes* occurrence in fish. However, sex may not have significant influence on the bacterium occurrence and therefore could not be used as an index to predict the presence of *L. monocytogenes* in fish. The insignificant difference in the farms also confirmed the ubiquitous nature of *L. monocytogenes* which is consistent with findings of [3, 6, 20, 26].

Also, the significant differences observed among the organs suggested that different organs are susceptible to *L. monocytogenes* than the other. In this study, spleen, skin and muscle have higher chance of being infected than others. This is in agreement with the work of [5] who reported significant variation in the occurrence of *L. monocytogenes* in gill, skin and muscle in rainbow trout. The higher intensity of infection observed in rainbow trout may be related to the small thinning scales in this fish which could serve as accumulation site with subsequent domiciling in gills, while the absence of scale in *C. gariepinus* may be responsible to the migration of the bacterium to the inner part such as spleen.

The low concentration of *Listeria* species in farm ponds water as compared to sediment may be as a result of precipitation, rainfall or direct addition of water by farmers when the water level is low. Although raw fish are not consumed in Nigeria, but the presence of *L. monocytogenes* in tissues of the fish across the chain pose a public health risk and the fact that the organism may be very difficult to eliminate.

From the result of this study it could be concluded that *L. monocytogenes* is well distributed in fish farms in fish farms in Oyo State, Nigeria. The prevalence by organs shows that fish flesh, skin, kidney and spleen are the affected organs which further buttresses the fact that infected fish may pose a public health risk to fish handlers and consumers. However, adequate awareness on significance of *L. monocytogenes* should be encourage within the fish handlers and consumers

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