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# Antibiotics Susceptibility Profile of *Listeria* species Isolated from Untreated Abattoir Wastewater in Akinyele, Ibadan, Nigeria and Its Implication on Public Health

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# Authors' contributions

This work was carried out in collaboration between all authors. Authors OIF and OEF designed the study and the protocol. Authors OIF and AGR managed literature search, data acquisition and wrote the first draft. All authors read and approved the final manuscript.

## Article Information

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# **ABSTRACT**

**Introduction:** The demand for meat to meet the need of increasing human population has led to increase in the abattoir activities. Untreated abattoir wastewaters which may constitute a public health threat due to the possibility of transmission of pathogens to humans are constantly being discharged into water bodies. *Listeria* is an emerging pathogen that is commonly associated with food infection and has been found to survive in water.

**Aim:** This study was carried out to determine the occurrence and antibiotic resistant pattern of *Listeria* species isolated from abattoir wastewater in Akinyele, Ibadan.

**Materials and Methods:** Wastewater samples were collected from the slaughter slab and drainage channels between May and June, 2015. *Listeria* species were isolated using *Listeria* Selective Agar Base with *Listeria* Selective Supplement and identification of the isolates was done using conventional methods. Antimicrobial susceptibility testing was carried out using the Kirby-Bauer disk diffusion assay using single antibiotic disks including ampicillin, streptomycin, chloramphenicol, amoxicillin/clavulanate, ceftriazone, cloxacillin, ciprofloxacin, ofloxacin, tetracycline and

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trimethoprim/sulfamethoxazole.

**Results:** A total of 82 *Listeria* spp. were isolated comprising 58.5% and 41.5% from the slaughter slab and drainage respectively; all the isolates were resistant to ampicillin while 86.6%, 68.3% and 65.5% were resistant to ceftriaxone, amoxicillin/clavulanate and tetracycline respectively. In addition, 12.2% of the total isolates were resistant to a combination of six antibiotics (ampicillin, tetracycline, amoxicillin, ceftriazone, streptomycin, and trimetoprim/sulfamethozaxole) while only one isolate was resistant to eight or more antibiotics.

**Conclusion**: This study revealed that the Akinyele abattoir wastewater is a potential medium for the transmission of multi-drug resistant bacteria to humans. Hence, adequate measures should be put in place for the treatment of the abattoir wastewater before discharge into the environment.

Keywords: Listeria species; antibiotics resistance; abattoir wastewater; emerging pathogen.

## 1. INTRODUCTION

Listeria is an emerging pathogen which is commonly associated with food borne infection [1]. Out of the eight generally recognized species that includes the following: L. monocytogenes, L. ivanovii, L. seeligeri, L. innocua, L. welshimeri, L. grayi, L. marthii and L. rocourtiae [2,3], only L. monocytogenes and L. ivanovii are both humans and animals pathogens while L. ivanovii cause disease mostly in ruminants animals, others are regarded as nonpathogenic [2,4]. However, there have been reported cases of human infection as a result of L. ivanovii, L. seeligeri, L. innocua and L. welshimeri [5-7]. For instance, L innocua has been reported to be occasionally associated with encephalitis in ruminants [8]. L. monocytogenes, etiologic agent of listeriosis continues to be a pathogen with significant impact in the public economy all over the world because of the food safety concern [9]. The pathogen has also been reported to be responsible for infections such as meningitis, encephalitis and septicemia in immune-compromised individuals, abortion in pregnant women and neonatal infections with lethality around 20%-30% of all cases [7,10].

While *Listeria* is reported to be responsible for several food borne outbreaks in the developed world, little is known about such outbreak in the developing countries [10,11]. Although, food is the primary route of transmission of *Listeria* infections, studies have shown that this pathogen is able to survive in water and that wastewater is a potential reservoir for *Listeria* species and possible source of its transmission [1,12-14]. Transmission of this pathogen via wastewater has a serious public health implication especially in the developing world in which the greater percentage of the population depends on surface water for drinking and other domestic purposes.

Most of the surface water is negatively impacted as they are constantly receiving wastewater that is untreated or not properly treated from operations like the abattoir [15,16].

The continuous drive to increase production to meet the ever increasing protein demand of the increasing world population goes along with pollution problem [17,18]. Abattoir wastewater generated during the slaughtering activities are of great concern because like other discharged sewage, they eventually enter natural bodies of water like ground water, streams, rivers, lakes and oceans [19,20]. These water bodies are used by humans for different purposes especially for drinking and recreation; such usage has been implicated in the transmission of pathogens to humans [21,22]. Previous reports has shown that bacteria exists either as free-living or attached cells [23] in other to resist the action of disinfection and enhance their ability to develop resistance to antimicrobial agents [1]. More so Listeria has been reported to be resistant to antimicrobial agents which were as a result of certain resistance genes that encode proteins and act to either inhibit or reduce the effect of the antimicrobials on the pathogen [1,24].

Reports have shown that the mortality rate of *Listeria* infections is as high as 50% among the foodborne pathogens [10], an indication that Nigerian public is vulnerable because of the high HIV/AIDS prevalence in the country coupled with the rate of drug and alcohol abuse just like some other low income countries [1,25]. However, only limited information on the prevalence and possible outbreak of listeriosis in Nigeria is available. More importantly, *Listeria* is not considered as a waterborne pathogen despite reports in literature that it is well established in water supply chain [12,14]. The first prevalence study of *Listeria* was a serological study, but the

first isolation was in 1969 [26,27] in a female adult with leptomeningitis being treated with chloramphenicol and prednisone. The first neonatal listeriosis from which L. monocytogenes was isolated was from a 2-day old baby who developed Listeria meningitis after the contact with the mother. [28]. Since then the pathogen has been isolated from neonatal septicemia patients in the country [29,30]. Listeria monocytogenes was also reported to be prevalent in cattle and other animals characterised by still birth, abortion and nervous signs before death [31].

Resistance of Listeria spp. to antimicrobial agents has been reported, the most common among clinical isolates is to chloramphenicol and ampicillin [26]. A high level of resistance to clindamycin, daptomycin, fluoroquinolone and to third- and fourth-generation cephalosporins was observed in a study on *Listeria* spp. isolated from humans and slaughterhouse [32]. In another study, some strains of L. monocytogenes, L. innocua, L. seeligeri, and L. welshimeri were found to be resistant to oxacillin, clindamycin and daptomycin which are widely used hospitals for the treatment of Gram-positive infections [33]. The indiscriminate discharge of untreated abattoir wastewater into environment may portend grave danger to the health of the public because the wastewater may harbour pathogens with high level of antibiotics resistance and even multiple drug resistant organisms capable of crossinfecting human and cattle due to Increasing evidence that the use of antimicrobial in animals selects for resistant pathogens [34]. In Nigeria, there is a need to investigate emerging waterborne pathogen that is being overlooked or not investigated though they have tendency to survive and be distributed in water supply chain [1]. This study was carried out to determine the prevalence of Listeria species in untreated abattoir wastewater as well as determine the profile of the isolated strains to selected antibiotics.

# 2. MATERIALS AND METHODS

# 2.1 Study Site

The study site was a commercial abattoir located in Akinyele-Ibadan, Oyo State, Nigeria. Ibadan is the largest city in West Africa [35] with land size covering an area of 240 km<sup>2</sup>, and a human

population of about 3 million. The city is located on geographic grid reference longitude  $35^1$  E and latitude  $720^1$  N [36].

# 2.2 Sample Collection

Wastewater samples were aseptically collected into sterile polythene bottles from slabs where animals are being slaughtered and also from the drainage channel which is about 15 meters away from the slab. The samples were transported immediately to the laboratory in ice packs for microbiological analyses. Samples were collected bi-weekly over a period of six weeks (May and June, 2015).

# 2.3 Isolation and Identification of *Listeria* species

Listeria species were isolated using the method described by Akano et al. [37]. The wastewater samples analysis was done using the Listeria Selective Agar Base (Oxoid) which was prepared according to manufacturers' instruction and supplemented with a vial of Listeria Selective Supplement. Serial dilutions of the abattoir wastewater samples were done and standard pour plate technique was used by plating out 1 ml of the appropriate dilution on the listeria selective media. The plates were then incubated at 35℃ for 24-48 hrs. Typical colonies of Listeria species, gray coloured isolates with dark background, were sub-cultured on Listeria Selective Agar media to obtain pure isolates. Identification of Listeria strains were carried out in accordance with standard methods of identification of bacteria of medical importance through microscopy, Gram staining biochemical tests.

# 2.4 Antibiotics Susceptibility Test of the Isolates

The antibiotics susceptibility test for the Listeria isolates was done using standard disk diffusion technique based on the recommendation of Clinical Laboratory Standards Institute [38] on Mueller-Hinton agar. The antibiotics used were obtained from Oxoid, U.K. and include: Ampicillin (10 µg), streptomycin chloramphenicol (30 μg), amoxicillin/clavulanate (30 µg), ceftriazone (30 μg), cloxacillin (5 μg), ciprofloxacin (10 μg), ofloxacin (5 µg), tetracycline (25 µg) and trimethoprim/sulfamethoxazole (25 µg).

Colonies of 18-24 hour old culture was picked and suspended in a tube containing sterile normal saline (0.85% NaCl) and the turbidity adjusted to 0.5 McFarland standards. With the aid of a sterile swab stick, the suspension was uniformly spread over already prepared Mueller Hinton agar plates and with the aid of sterile forceps, the antibiotics were placed carefully on the plates which were inverted and incubated at 37°C for 18-24 hours. After the incubation period, the zones of inhibition were measured, recorded and compared/interpreted to the CLSI standards [38].

# 3. RESULTS

A total of 82 *Listeria* spp. were recovered from the Akinyele abattoir wastewater samples and comprises of 48 (58.5%) and 34 (41.5%) from the slaughter slab and the drainage respectively (Table 1). The identification showed that 22 (26.8%) were *Listeria monocytogenes*; 15 (18.3%) were *L. ivanovii*, 17 (20.7%) were *L. innocua* while 28 (34.2%) belongs to other *Listeria* species. From the slaughter slab, the highest was *L. monocytogens* (17.1%) while from the drainage, it was other *Listeria* sp. (18.3%) (Table 1).

The results of the antibiotic susceptibility test showed that all the isolates (100%) were resistant to ampicillin. However, all the L. monocytogenes (100%) and L. ivanovii (100%) were susceptible to ofloxacin while all the other Listeria species were susceptible to ciprofloxacin. Furthermore, resistance of the isolates to ceftriaxone (86.6%), amoxicillin (68.3%) and tetracycline (65.5%) was high. Among the isolates that were resistant to ceftriaxone, the highest was from other Listeria spp. (92.9%), followed by L. monocytogenes (90.9%), L. innocua (82.4%) and L. ivanovii. (73.3%) while among the different isolates that showed resistant to amoxicillin, the highest was among the other Listeria spp. (85.7%), followed by L. innocua (70.6%) then L. monocytogenes (63.6%) and L. ivanovii (40%). More so resistance to tetracycline was 71.4% (other Listeria species), 70.6% (L. innocua), 60.0% (L. ivanovii) and 59.1% (L. monocytogenes).

Moreover, resistance of the isolates to ofloxacin (6.1%), cloxacillin. (11.0%) and streptomycin (36.6%) was low (Table 2). Furthermore, the result of the antibiotypes (pattern of resistance to combination of antimicrobials) showed that the highest (12.2%) was to the combination of six

different drugs that included tetracycline, amoxicillin/ clavullanate, ceftriaxone, streptomycin and Trimethoprim/ sulfamethoxazole. Out of these, 25.0% Listeria were resistant while it was 4.5% (L. monocytogenes), 6.7% (L. ivanovi) and 5.9% (L. innocua). In addition the result of this study also revealed that 9.6% of the resistant isolates exhibited resistance to a combination of four drugs including: Ampicillin, tetracycline amoxicillin and ceftriaxone with the highest (17.6%) being among the L. innocua followed by the L. monocytogen (13.6%) and other Listeria species but none of the L. ivanovvi showed resistance to the four antibiotic combination. Resistance to a combination of five drugs including ampicillin, tetracycline, ceftriaxone amoxicillin/clavullanate, chloramphenicol was 7.3% which was among L. monocytogenes (13.6%) and other Listeria species (10.8%) while none of the L ivanovvi and L. innocua showed resistant to the combination of the drugs. More so, the result of the study also showed that one (1.2%) isolates each among the other Listeria spp. showed resistant to eight or more antibiotics (Table 3).

#### 4. DISCUSSION

Isolation of Listeria species from wastewater portending the organism as emerging pathogen with wastewater associated has reported [39]. This organism has also been isolated from ruminant animals [40,41] and in 2008; strains of *L. monocytogenes* were isolated from broiler abattoir in Spain [42] and from pork products in India [9]. The highest occurrence of isolates from this study which (26.8%)L. monocytogenes followed bν L. innocua (20.7%) and L. ivanovii (18.3%) is similar to the report of the study on pork and pork where products the occurrence L. monocytogenes was higher than the other species [9]. However, the occurrence rate of these isolates was higher in this study compared to the same study; this may be because other animals including pigs are being slaughtered in the sampling site for this present study. This observation was not in agreement with another study carried out on animal faeces from the northern part of Nigeria in which the occurrence of L. monocytogenes was reported to be the least of all the listerial isolates [40].

The sensitivity of *L. monocytogenes* to ampicillin (0%), chloramphenicol (59.1%), streptomycin (59.1%) and trimethoprim/ sulfamethoxazole

(63.6%) in this study is not in agreement with the report of another study carried out on isolates from ready to eat food from South Africa in which all the isolates (100%) were reported to be sensitive to the combination of the four antibiotics [34]. However, the observation that all the Listeria isolates were fully sensitive (100%) to ciprofloxacin is the same with the report of Ndip et al. [34]. This observation is however higher but comparable to the 91% resistant reported in South Africa [1]. The reason for the slight difference may be as a result of the studied sample. While the present study was on abattoir wastewater, the later study was on municipal wastewater. Comparing the antibiotics resistance patterns of the L. monocytogenes form this study with the same strain in another study in the country, resistance to cloxacillin at the same concentration (5 µg) was much higher (80.7%) in the study [39] than the 4.5% in the present study. Resistance of the *L. monocytogenes* in this study was lower (63.6%) but comparable to the 77.4% in the other study [39]. The reason for the slight difference may be due to the different concentration of the antibiotics used in the studies which was 30 µg (present study) and 25 µg [39]. With the same drug concentration, resistance of L. monocytogenes to ampicillin (100%).chloranphenicol (36.4%)and streptomycin (40.9%) in this study was higher than the one reported by Enurah and his colleagues which was 64.5% (ampicillin), 1.2% (chloramphenicol) and 35.5% (streptomycin). Resistance to tetracycline in this study was 59.1% while it was 19.36% in the other study. However, the concentration used in this study was 30 µg while that reported in the latter study was 10 µg.

Also in another study carried out on the tissues of slaughtered cattle in a neighboring abattoir in Ibadan, all the Listeria isolates were reported to exhibit 100% resistance to all the antibiotics tested [41] which is not in agreement with the result of the present study except for resistance of the isolates to ofloxacin. The reason for this may be as a result of the samples studied and some of the concentration of the antibiotics used which was higher in the present study, although, the same concentration of antibiotic was used for amoxicillin and streptomycin. Moreover, there were discrepancies in the resistance of the isolates to ciprofloaxin, amoxicillin, tetracycline. Resistance of L. monocytogenes to the respective antibiotics was 50%, 100% and 50% [43] which is not in agreement with the observation from the present study. Furthermore, while resistance of L. innocua to tetracycline and ceftriaxone was high in both studies; varying resistance rate was observed in this study which is not in agreement with full susceptibility reported in the other study [43]. The resistance of all the Listeria isolates to ceftriazone in this study and that of Akano et al. [43] is similar with 86.6% and 85.2% respectively. However, resistance of the isolates in this study was lower to amoxicillin, chloramphenicol, ciporofloaxin and ofloxacin. But the resistance of all the listeria isolates in this study which was 100% (ampicillin), 41.5% (chloramphenicol), 36.6% (streptomycin and 65.5% (tetracycline) was much higher than the 12% (ampicillin), 15% (chloramphenicol), 15.8% (streptomycin) and 23.3% (tetracycline) obtained from the study of listeria isolates from open-air fish market [44]. More so, the observation was similar to the resistance of *L. monocytogenes* isolates in which all the strains in this study is higher than those of the later study. The reason for these discrepancies may be due to the studied samples. The observation of the resistance patterns of L. monocytogenes to ampicillin, chloramphenicol, tetracycline and streptomycin in this study is not in agreement with another study from Gaborone, Botswana where resistance to the respective drugs was reported to be 0%, 28.3%, 67.8% and 80.7% [45] as against the 100%, 36.4%, 59.1% and 40.9% respectively. These discrepancies could be as a result of the samples studied which may impact on their adaptation and response to different chemicals and antimicrobial agents. While the samples for this study were from abattoir wastewater, the Botswana samples were from various foods. The high resistance of the isolates to some of the tested antibiotics shows the probability of natural resistance to third/fourthgeneration cephalosporin. This is an indication that the isolates showed resistance to antibiotics that are currently being used in the treatment of listeriosis infection such as ampicillin and trimethoprimsulfamethoxazole [34]. Consequently, these isolates potentially constitute high risk when these traditional treatments are used. Multiple drug resistance in organisms might be attributed to antimicrobial selective pressure and gene transfer mechanisms between and among the Listeria species and close relatives of the bacteria such as Enterococcus, Streptococcus and Staphylococcus species [46,47].

Table 1. Number and percentage occurrence of isolated organisms from the abattoir wastewater sample in Akinyele, Ibadan

Isolates		Total (%)	
	Slaughter slab (%)	Drainage (%)	
Listeria monocytogenes	14 (17.1)	8 (9.6)	22 (26.8)
Listeria ivanovii	12 (14.6)	3 (3.7)	15 (18.3)
Listeria innocua	9 (11)	8 (9.8)	17 (20.7)
Listeria spp.	13 (15.9)	15 (18.3)	28 (34.2)
Total	48 (58.5%)	34 (41.5%)	82 (100%)

Table 2. Antibiotic susceptibility pattern of Listeria species isolated from the abattoir wastewater. Number (%)

Antibiotics	L. m.	L. iv	L. in	L. spp	L. m.	L. iv	L. in	L. spp	TR
	n=(22)	(n=15)	(n=17)	(n=28)	n=(22)	(n=15)	(n=17)	(n=28)	N=82
	Sensitive				Resistance				_
Ampicillin (10 μg)	0 (0)	0 (0)	0 (0)	0 (0)	22 (100)	15 (100)	17 (100)	28 (100)	82 (100)
Amoxicillin/Clavullanate (30 µg)	8 (36.4)	9 (60)	5 (29.4)	4 (14.3)	14 (63.6)	6 (40)	12 (70.6)	24 (85.7)	56 (68.3)
Ceftriaxone (30 µg)	2 (9.1)	4 (26.7)	3 (17.6)	2 (7.1)	20 (90.9)	11 (73.3)	14 (82.4)	26 (92.9)	71 (86.6)
Ciprofloxacin (5 µg)	22 (100)	15 (100)	17 (100)	28 (100)	0 (0)	0 (0)	0 (0)	0 (0)	0
Ofloxacin (5 µg)	22 (100)	15 (100)	16 (94.1)	24 (85.7)	0 (0)	0 (0)	1 (5.9)	4 (14.3)	5 (6.1)
Cloxacillin (5 µg)	21 (95.5)	13 (86.7)	15 (88.2)	24 (85.7)	1 (4.5)	2 (13.3)	2 (11.8)	4 (14.3)	9 (11.0)
Chloramphenicol (30 µg)	14 (63.6)	8 (53.3)	12 (70.6)	14 (50)	8 (36.4)	7 (46.7)	5 (29.4)	14 (50)	34 (41.5)
Tetracycline (30 µg)	9 (40.9)	6 (40)	5 (29.4)	8 (28.6)	13 (59.1)	9 (60)	12 (70.6)	20 (71.4)	54 (65.5)
Trimethoprim/sulfamethozaxole (25 μg)	14 (63.6)	8 (53.3)	8 (47.1)	13 (46.4)	8 (36.4)	7 (46.7)	9 (52.9)	15 (53.6)	39 (47,6)
Streptomycin (10 µg)	13 (59.1)	9 (60)	11 (64.7)	19 (67.9)	9 (40.9)	6 (40)	6 (35.3)	9 (32.1)	30 (36.6)

L. m., Listeria monocytogenes; L. iva, L. ivanovii; L. inn, L. innocua;, L. spp, other Listeria species; TR Total Resistance

Table 3. Antibiotypes of all *Listeria* species isolated from abattoir wastewater

Antibiotypes	L. monocytogenes	L. ivanovii	L. innocua	Other Listeria.	Total n/%
	22 (26.8)	15 (18.3)	17 (20.7)	spp. 28(34.1)	(82/100)
AMP	2 (9.1)	3 (20.1)	1 (5.9)	2 (7.1)	8 (9.6%)
AMP~OB	0 (0)	1 (6.7)	1 (5.9)	2 (7.1)	4 (4.9%)
AMP~T	0 (0)	0 (0)	1 (5.9)	0 (0)	1 (1.2%)
AMP~AMC~OB	1 (4.5)	1 (6.7)	0 (0)	1 (3.6)	3 (3.7%)
AMP~T~OB	1 (4.5)	1 (6.7)	0 (0)	0 (0)	2(2.4%)
AMP~S~SXT	0 (0)	0 (0)	1 (5.9)	0 (0)	1 (1.2%)
AMP~OB~SXT	1 (4.5)	0 (0)	0 (0)	1 (3.6)	2 (2.4%)
AMP~T~OB~C	0 (0)	1 (6.7)	0 (0)	1 (3.6)	2(2.4%)
AMP~T~OB~S	1 (4.5)	1 (6.7)	0 (0)	0 (0)	2(2.4%)
AMP~OB~S~SXT	2 (9.1)	1 (6.7)	0 (0)	0 (0)	3 (3.7%)
AMP~AMC~OB~SXT	2 (9.1)	0 (0)	1 (5.9)	1 (3.6)	4 (4.9%)
AMP~T~AMC~OB	3 (13.6)	0 (0)	3 (17.6)	2 (7.1)	8 (9.6%)
AMP~T~OB~SXT	0 (0)	0 (0)	1 (5.9)	1 (3.6)	2(2.4%)
AMP~T~AMC~OB~C	3 (13.6)	0 (0)	0 (0)	3 (10.8)	6 (7.3%)
AMP~T~CRO~OB~SXT	0 (0)	0 (0)	1 (5.9)	0 (0)	1(1.2%)
AMP~T~AMC~OB~SXT	1 (4.5)	0 (0)	1 (5.9)	1 (3.6)	3(3.7%)
AMP~T~AMC~OB~C~SXT	1 (4.5)	1 (6.7)	2 (11.8)	0 (0)	4 (4.9%)
AMP~T~AMC~OB~S~SXT	1 (4.5)	1 (6.7)	1 (5.9)	7 (25.0)	10 (12.2%)
AMP~T~AMC~OB~OFX~SXT	0 (0)	0 (0)	0 (0)	1 (3.6)	1(1.2%) ´
AMP~T~AMC~OB~C~S	2 (9.1)	0 (0)	1 (5.9)	0 (0)	3(3.7%)
AMP~T~AMC~CRO~OB~C~SXT	0 (0)	1 (6.7)	0 (0)	0 (0)	1(1.2%)
AMP~T~AMC~OB~C~S~SXT	0 (0)	2 (13.3)	0 (0)	1 (3.6)	3(3.7%)
AMP~T~AMC~CRO~OB~C~S	1 (4.5)	0 (0)	0 (0)	0 (0)	1(1.2%)
AMP~T~AMC~CRO~OB~S~SXT	0 (0)	0 (0)	1 (5.9)	0 (0)	1(1.2%)
AMP~T~AMC~OB~OFX~C~S~SXT	0 (0)	0 (0)	1 (5.9)	1 (3.6)	2(2.4%)
AMP~T~AMC~CRO~OB~C~S~SXT	0 (0)	1 (6.7)	0 (0)	1 (3.6)	2(2.4%)
AMP~T~AMC~CRO~OB~OFX~C~SXT	0 (0)	0 (0)	0 (0)	1 (3.6)	1(1.2%)
AMP~T~AMC~CRO~OB~OFX~C~S~SXT	0 (0)	0 (0)	0 (0)	1 (3.6)	1(1.2%)

CIP, Ciprofloxacin; AMP, Ampicillin; T, Tetracycline; OFX, Ofloxacin; CRO, Cloxacillin; OB, Ceftriaxone; C, Chloramphenicol; S, Streptomycin; SXT, Trimethoprim/sulfamethoxazole; AMC, Amoxicillin/clavullanate; A, Slaughter slab; B, drainage

The results indicated alarming multi-resistance frequencies to at least three or more of the tested antibiotics. This could constitute public health concern because the abattoir wastewater can act as reservoir of resistant strains which could be transmitted to humans. Similar patterns of resistance have been reported by Srinivasan et al. [24] who found that all *L. monocytogenes* strains isolated from a dairy farm environment were resistant to ampicillin, tetracycline and chloramphenicol. Therefore, the emergence of resistance to these antibiotics demands attention due to the risk of increasing multidrug resistance in *Listeria* and the possibility of its transference to other bacteria [24].

## 5. CONCLUSION

In conclusion, this study has further confirmed a high prevalence of Listeria species especially L. monocytogenes in wastewater samples. This is an indication that such wastewater could serve as a public health threat especially among the immune compromised individuals like the HIV/AIDS patients that are at greater risk as it may lead to the outbreak of listeriosis disease. It was also evident that L. monocytogenes exhibited high resistant to ampicillin, ceftriaxone, amoxicillin/clavulanate and tetracycline which is suggestive that the misuse of antimicrobial drugs for therapeutic purposes in both animals and humans may lead to antibiotic resistance development. It is therefore recommended that adequate measure should be put in place for the proper treatment of the abattoir wastewater before they are discharged into the environment and further study is needed to provide better understanding of the epidemiology of listeriosis in Nigeria.

# COMPETING INTERESTS

Authors have declared that no competing interests exist.

#### REFERENCES

- Odjadjare EEO, Obi CL, Okoh Al. Municipal wastewater effluents as a source of listerial pathogens in the aquatic milieu of the Eastern Cape Province of South Africa: A concern of public health importance. International Journal of Environmental Research and Public Health. 2010;7:2376-2394.
- 2. Liu D. Identification, subtyping and virulence determination of *Listeria*

- monocytogenes and important food-borne pathogen. Journal of Medical Microbiology. 2006;55:645-659.
- Den Bakker HC, Cummings CA, Ferreira V, Vatta P, Orsi RH, Deogoricija L, Baker M, Petrauskene O, Furtado MR, Wiedmann M. Comparative genomics of the bacterial genus *Listeria*: Genome acquisition and limited gene loss. Biomed Central (BMC) Genomics. 2010;11:688-693.
- 4. Brugere-Picoux O. Ovine listeriosis. Small Ruminant Research. 2008;76:12-20.
- 5. Allenberger F. *Listeria*: Growth, phenotypic differentiation and molecular microbiology. FEMS Immunology of Medical Microbiology. 2002;35:183-189.
- Cocolin L, Rantsiou K, Iacumin L Cantoni C, Comi G. Direct identification in food samples of *Listeria* spp. and *Listeria* monocytogenes by molecular methods. Applied Environmental Microbiology. 2002; 68:6273-6282.
- Perrin M, Bemer M, Delamare C. Fatal case of *Listeria innocua* bacteremia. Journal of Clinical Microbiology. 2003; 41:5308-5309.
- 8. Walker JK, Morgan JH, McLauchlin J, Grant KA, Shallcross JA. *Listeria ivanovii* isolated from a case of ovine meningoencephalitis. Veterinary Microbiology. 1994;59:193-202.
- Raorane AV, Doijad SP, Poharkar1 KV, Pathak A, Bhosle S, Barbuddhe SB. Isolation and genotypic characterization of *Listeria monocytogenes* from pork and pork products International Journal of Current Microbiology and Applied Science. 2015;4:788-798.
- Rocourt J, Jacquet CH, Reilly A. Epidemiology of human listeriosis and seafoods. International Journal of Food Microbioogy. 2000;62:197-209.
- Siegman-Igra Y, Levin R, Weinberger M, Golan Y, Schwartz D, Samra Z, Konigsberger H, Yinnon A, Rahav G, Keller N, Basharat N, Karpuch J, Finkelstein R, Alkan M, Landau Z, Novikov J, Hassin D, Rudnicki C, Kitzes R, Ovadia S, Shimoni Z, Lang R, Shohat T. Listeria monocytogenes in Israel and review of case worldwide. Emerging Infectious Diseases. 2002;8(3):205-210.
- Paillard D, Dubois V, Thiebaut R, Nathier F, Hoogland E, Caumette P, Quentin C. Occurrence of *Listeria* spp. in effluents of French urban wastewater treatment plants.

- Applied Environmental Microbiology. 2005; 71:7562-7566.
- Arslan S, Ozdemir F. Prevalence and antimicrobial resistance of *Listeria* species in homemade white cheese. Food Control. 2008:19:360-363.
- Odjadjare EEO, Okoh AI. Prevalence and distribution of *Listeria* pathogens in the final effluents of a rural wastewater treatment facility in the Eastern Cape Province of South Africa. World Journal of Microbiology and Biotechnology. 2010;26: 297-307.
- Adekanmbi AO, Falodun OI. Heavy metals and antibiotics susceptibility profiles of Staphylococcus aureus isolated from several points receiving daily input from the Bodija Abattoir in Ibadan, Oyo State, Nigeria. Advances in Microbiology. 2015; 5:871-880.
- Falodun OI, Adekanmbi AO. Antibiogram of Escherichia coli and Pseudomonas strains isolated from wastewater generated by an abattoir as it journeys into a receiving river. Advances in Microbiology. 2016;6:303-309.
- Laukova A, Marekova M, Vasilkova Z, Papajova I, Juris P. Selected microbial consortium of raw and digested slurry and its susceptibility to enterocins. World Journal of Microbiology and Biotechnology. 2002;18:11-15.
- Amisu KO, Coker AO, Isokpehi RD. Arcobacter butzlieri strains from poultry abattoir effluent in Nigeria. East Africa Medical Journal. 2003;80:218-221.
- Madigan M, Martink JM, Parker J. Biology of microorganism, 8th Ed. New York Prentice Hal; 1997.
- Pelczar MJ, Chan ECS, Kreig NR. Microbiology 5th Ed. New Delhi. Tata McGraw-Hill; 2002.
- Muniesa M, Jofre JG, Aljaro C, Blanch AR. Occurrence of Eshcerichia coli 0157: H7 and other enterohaemorragic E. coli in the environment. Environmental Science and Technology. 2006;40:7141-7149.
- Sehgal R, Kumar Y, Kumar S. Prevalence and geographical distribution of Escherichia coli 0157 in India; a 10-year survey. Royal Society of Tropical Medicine and Hygiene. 2008;102:380-383.
- Djordjevic D, Wiedmann M, McLandsborough LA. Microtitre assay for assessment of *Listeria monocytogenes* biofilm formation. Applied Environmental Microbiology. 2002;68:2950-2958.

- 24. Srinivasan V, Nam HM, Nguyen LT, Tamilselvam B, Murinda SE, Oliver SP. Prevalence of antimicrobial resistance genes in *Listeria monocytogenes* isolated from dairy farms. Foodborne Diseases. 2005;2:201-211.
- 25. Obi CL, Onabolu B, Momba MNB, Igumbor JO, Ramalivahna J, Bossong PO, van Rensburg EJ, Lukoto M, Green E, Mulaudzi TB. The interesting cross-paths of HIV/AIDS and water in Southern Africa with special reference to South Africa. Water South Africa. 2006;32:323-343.
- Nwaiwu O. An overview of *Listeria* species in Nigeria. International Food Research Journal. 2015;22(2):455-464.
- Eyo EE, Senbanjo AO, Babatunde EO. Listeria monocytogenes meningitis in an adult Nigerian female. The West African medical Journal and Nigerian Practitioner. 1969;18:121-124.
- Onyemelukwe GC, Lawande RV. Listeriosis in a neonate and the mother. Tropical and Geographical Medicine. 1982; 34:87-89.
- Ako-Nai AK, Adejuyigbe EA, Ajayi FM, Onipede AO. The bacteriology of neonatal septicaemia in Ile-Ife, Nigeria. Journal of Tropical Paediatrics. 1999;45:146-151.
- Adejuyigbe EA, Adeodu OO, Ako-Nai KA, Taiwo O, Owa JA. Septicaemia in highrisk neonates at a teaching hospital in Ile-Ife, Nigeria. East African Medical Journal. 2001;78:540-543.
- 31. Akpavie SO, Ikheloa JO. An outbreak of listeriosis in cattle in Nigeria. Revue d'Elevage et de Medecine Veterinaire des Pays Tropicaux. 1992;45:263-264.
- 32. Moreno LZ, Paixão R, Gobbi DDS, Raimundo DC, Ferreira TP, Moreno AM, Hofer E, Reis CMF, Matté GR Matté MH. Characterization of antibiotic resistance in *Listeria* spp. isolated from slaughterhouse environments, pork and human infections. Journal of Infections from Developing Countries. 2014;8(4):416-423.
- Conter M, Paludi D, Zanardi E, Ghidini S, Vergara A, Ianieri A. Characterization of antimicrobial resistance of foodborne *Listeria monocytogenes*. International Journal of Food Microbiology. 2009;128: 497-500.
- Ndip RN, Nyenje ME, Tanih NF, Green E. Current status of antibiograms of *Listeria* ivanovii and Enterobacter cloacae isolated from ready-to-eat foods in Alice, South Africa. International Journal of

- Environmental Research and Public Health. 2012;9:3101-3114.
- 35. Filani MO, Akintola FO, Ikporukpo CO. Ibadan region. Recharles Publications and Connel Publication. University of Ibadan, Ibadan. 1994;271.
- Coker AO, Olugasa BO, Adeyemi AO. Abattoir wastewater quality in South Western Nigeria, Proceedings of the 27th WEDC Conference, Loughborough University, United Kingdom. 2001;329-331.
- Akano SO, Moro DD, Deji-Agboola AM, Oluwadun A. Public health implication of Listeria species and other bacteria isolates of abattoir effluent in Lagos, Nigeria. International Research Journal of Microbiology. 2013;4(7):162-167.
- Clinical and Laboratory Standards Institute (CLSI), performance standards for antimicrobial disk susceptibility tests; 9th ed.; Document M2-A9; Clinical and Laboratory Standards Institute (CLSI): Wayne, PA, USA; 2014.
- Enurah LU, Aboaba OO, Nwachukwu SCU, Nwosuh CI. Antibiotic resistant profiles of food (fresh raw milk) and environmental (abattoir effluents) isolates of Listeria monocytogenes from the six zones of Nigeria. Afican Journal of Microbiology Research. 2013;7:4373-4378.
- Lawan FA, Tijjani AN, Raufu AI, Ameh JA, Ngoshe IY, Auwal MS. Isolation and characterisation of *Listeria* species from ruminants in Maiduguri North

   Nigeria.
   African Journal of Microbiology Research.
   2013;12:6997-7001.
- 41. Adetunji VO, Adesokan HK, Agada CA, Isola TO. Bacterial load and antimicrobial

- profile of *Escherichia coli* and *Listeria* spp. Isolates from muscle tissues of slaughtered cattle at a Major Abattoir in Ibadan, South-Western Nigeria Journal of Basic & Applied Sciences. 2014;10:299-305.
- López V, Ortiz S, Corujo A, López P, Poza D, Navas J, Moreno R, Martínez-Suárez JV. Different contamination patterns of lineage i and ii strains of *Listeria monocytogenes* in a Spanish Broiler Abattoir Poultry Science. 2008;87:1874–1882.
- 43. Akano SO, Deji-Agboola AM, Oluwadun A. *Listeria* species and antimicrobial resistance profiles of HIV/AIDS patients in Lagos, Nigeria. New York Science Journal. 2014;7(4):46-52.
- Jamali H, Paydar M, Ismail S, Looi CY, Wong WF, Radmehr B, Abedini A. Prevalence, antimicrobial susceptibility and virulotyping of *Listeria* species and *Listeria* monocytogenes isolated from open-air fish markets. BMC Microbiology. 2015;15:144
- 45. Morobe IC, Obi CL, Nyila MA, Gashe BA, Matsheka MI. Prevalence, antimicrobial resistance profiles of *Listeria* monocytognes from various foods Journal of Biotechnology. 2009;8:6383-6387.
- Safdar A, Armstrong D. Listeriosis in Gaborone, Botswana African patients at a comprehensive cancer center, 1955–1997. Clinical Infectious Disease. 2003; 37:359-364
- 47. Arslan S, Ozdemir F. Prevalence and antimicrobial resistance of *Listeria* species in homemade white cheese. Food Control. 2008;19:360-363.

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