Prevalence and Antibiogram of *Listeria Monocytogenes* Contamination of Liver, Spleen, Ruminal Content and Effluent in Jos, Nigeria

HI Dunka[1,2]*; M Bello[2]; MK Lawan[2]

[1]*Department of Veterinary Public Health and Preventive Medicine, Faculty of Veterinary Medicine, University of Jos, Nigeria.*

[2]*Department of Veterinary Public Health and Preventive Medicine, Faculty of Veterinary Medicine, Ahmadu Bello University, Zaria, Nigeria.*

*Corresponding Author(s): Dunka HI

Department of Veterinary Public Health and Preventive Medicine, Faculty of Veterinary Medicine, University of Jos, Nigeria.

Email: dunkah@unijos.edu.ng

Abstract

Objective: This study was designed to determine the prevalence and antibiogram of *L. monocytogenes* from the liver, spleen, ruminal content of cattle and goats and effluent from abattoir/slaughter slabs in Jos, Nigeria.

Methods: A cross-sectional study was conducted and 360 samples were collected which include raw liver, spleen, ruminal content and effluent. Statistical test using Pearson's chi-square test for independence was done to determine the frequency of isolation of *L. monocytogenes.*

Results: Confirmation of the isolates using conventional biochemical characterization showed that 9 (2.5%) were positive for *L. monocytogenes.* Distribution of *L. monocytogenes* by sample type showed effluent (6.7%) had the highest prevalence compared to ruminal content (2.2%) and liver (1.1%) and the association was statistically significant ($p < 0.05$). Distribution of *L. monocytogenes* based on location showed Yan-Shanu (6.9%) had the highest prevalence although, the association was not statistically significant ($p > 0.05$). Antibiotic susceptibility testing revealed all (100%) of the *L. monocytogenes* isolates were resistant to ampicillin and colistin sulphate while none of the isolates showed resistant to ciprofloxacin. The sensitivity regimens were 8 (88.9%) for chloramphenicol as the most effective while the least sensitive were 1 (11.1%) for nalidixic acid and penicillin G each. Antimicrobial resistant profile of the isolates revealed that they were all resistant to at least four antimicrobial agents and the mean multiple antibiotic resistance index was 0.47±0.034.

Conclusions: The study showed that *L. monocytogenes* occur in all the study sites therefore, adequate precaution should be taken in order to avoid cross-contamination when dressing meat and meat by-product both for human and animal consumption. Also, the antimicrobial resistance pattern shown by the isolates is an indication that adequate measures need be taken to regulate drug use in both humans and animals in order to minimize the risk of increasing antimicrobial resistance.

Introduction

Listeria monocytogenes is an opportunistic foodborne pathogen belonging to the genus *Listeria,* a Gram-positive, non-spore forming, facultative anaerobic rod-shaped bacterium [1]. The genus *Listeria* consists of other different recognized species namely *L. seeligeri, L. ivanovii, L. welshimeri, L. marthii, L. innocua, L. grayi, L. fleischmannii, L. floridensis, L. aquatica, L. newyorkensis, L. cornellensis, L. rocourtiae, L. weihenstephanensis, L. grandensis, L. riparia,* and *L. booriae* [2]. Only two of these species are considered pathogenic to humans (*L. monocytogenes*) and animals (*L. monocytogenes* and *L. ivanovii*) [3]. However, sporadic cases of human infections caused by *L. ivanovii, L. innocua* and *L. seeligeri* in immunocompromised individuals have been reported [4,5,6] *L. monocytogenes*, the agent of listeriosis has been recognized as an important public health problem due to food safety concerns [2].

Infection with listeriosis is acquired through consumption of contaminated food by susceptible individuals [7] primarily pregnant women, neonates, elderly people, and those with weakened immune systems [8]. In Nigeria, there have been limited reports documenting outbreaks of listeriosis in the country [9]. Recently, South Africa witnessed the world's largest outbreak of human listeriosis attributed to the consumption of ready-to-eat processed meat products from a food production facility in which were 1060 confirmed laboratory cases with 216 deaths [9,10]. *L. monocytogenes* which is ubiquitous in nature has been isolated from several food and environmental sources including fresh raw and frozen meat and fish products [11,12,13], veg- etables [14], several ready-to-eat foods [15,16], milk and milk products [17,18,19], cattle faeces [20], food-associated envi- ronment [21,22] and in untreated abattoir wastewater/effluent [23,24]. However, less information is available on the occur- rence of *L. monocytogenes* in abattoir by-product from slaugh-tered animals.

In the recent past decade, extensive administration of antibiotics has led to misuse of these drugs in humans and animals which has contributed to the widespread antibiotic resistance among foodborne pathogen, *Listeria monocytogenes* inclusive [25]. Presently, antibiotic resistance is a signifi-cant public health problem, for instance, in the United States of America thousands of people die from infections caused by antibiotic-resistant bacteria [17,26]. Hence, this study was con-ducted to get an overview on the occurrence, distribution and antibiogram of *L. monocytogenes* in the liver, spleen, ruminal content of slaughtered cattle and goats and effluent from abat- toir/slaughter slabs in Jos, Nigeria, as a threat to food safety since the organism has the tendency to form biofilms in food processing environment and thereby cross-contaminate food product as seen in many ready-to-eat foods [27,22].

Materials and methods

Study site

The study was carried out in Jos, Plateau State, Nigeria located in the North-Central geo-political zone of Nigeria (Figure 1). It lies between latitude 9°56' North and longitude 8°53' East, having a total land area of 1,821km². Plateau State shares geographic boundaries with four of the thirty-six States of the Federation, Bauchi, Kaduna, Nasarawa and Taraba States at its North-Eastern, North-Western, South-Western and South-Eastern boundaries respectively. It's comprised of three local governments: Jos East, Jos North and Jos South. Plateau State is the 12th largest State in Nigeria with a population of about three million people. The State capital has only one registered abattoir and many un-registered slaughter slabs [28]; Personal communication) that all serve the populace. The Jos abattoir was established in 1975.
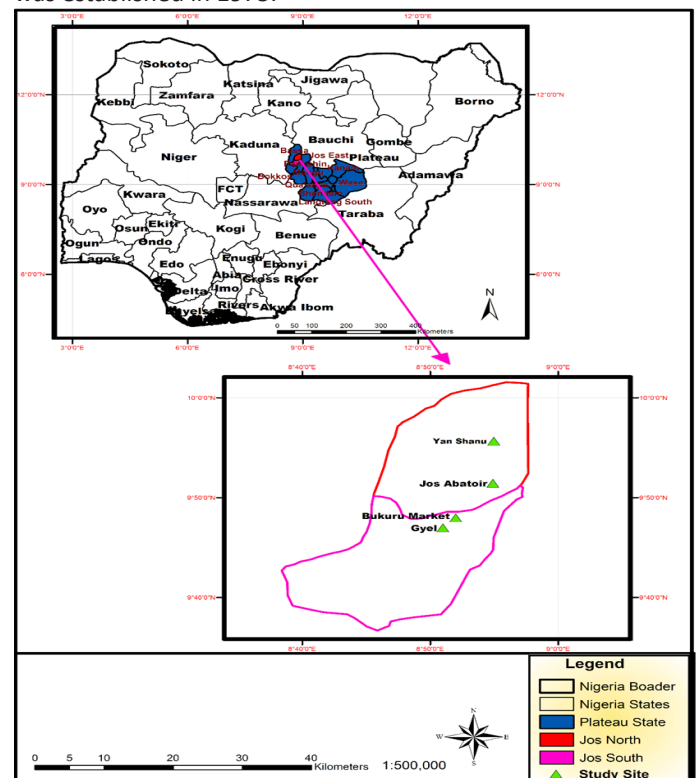

Figure 1: Map of Nigeria showing Plateau State and the Study Areas (Source: ArcAGIS).

Study design

A cross-sectional study was conducted in which one regis-tered abattoir and three un-registered slaughter slabs were se-lected for this study i.e. two each from Jos North (Jos Abattoir and Yan-Shanu slaughter slab) and Jos South (Bukuru and Gyel slaughter slabs) Local Government Areas. Three hundred and sixty (360) samples were collected from the study sites based on convenient sampling method. These are Bukuru (cattle) 72, Gyel (cattle) 72, Jos abattoir (cattle) 72, Jos abattoir (goat) 72 and Yan-Shanu (cattle) 72. Samples collected were 90 Liver, 90 spleen, 90 ruminal content of cattle and goats and 90 effluent samples from abattoir/slaughter slabs were collected, that is, 18 of each of these by-product samples from each of the sampling location.

Sampling

Pooled cattle and goat cut samples of liver, spleen and ruminal content were collected in polythene bags. Two hundred and fifty millilitres (250ml) of effluent samples from the abattoir/slaughter slabs were collected at the discharge point in the slaughter hall. The samples were kept on ice packs at 4°C in Coleman box and transported to the laboratory for analysis. Sampling was performed three times weekly for six weeks.

Isolation of *listeria* species

The ISO 11920-1 method for the quantitative isolation and identification of *L. monocytogenes* was used as described by Indrawattana *et al.* [29] with modifications. Aseptically, 10g of each sample of spleen, liver and ruminal content were and transferred into sterile stomacher bags and 90ml of 0.1% peptone water was added. They were homogenised using a stomacher (Stomacher Lab Blender 400) for 4-5 minutes followed by incubation at 37°C for 24 hours. One ml of each sample (effluent and homogenized liver, spleen and ruminal content) was suspended into 9ml of prepared *Listeria* enrichment broth incorporated with *Listeria* selective supplement and incubated at 37°C for 24 hours. A loopful of each sample was streaked onto the surface of *Listeria* selective agar incorporated with *Listeria* selective supplement and were incubated at 37°C for 24 hours. A loopful of each positive culture identified by the colonial morphology of greyish colonies surrounded by black halos (esculin hydrolysis) with sunken centres were sub-cultured by plating individually onto Tryptone Soya Agar (Oxoid, CMO 131) supplemented by 0.6% Yeast Extract Powder (Oxoid, LP002 1) (TSAYE) and were incubated at 37°C for 24 hours. The suspected colonies were inoculated onto prepared nutrient agar slant and incubated at 37°C for 24 hours and stored at 4°C in the refrigerator for further identification. Phenotypic characterization to species level was performed by subjecting presumptive colonies to Gram stanning and various biochemical test such as catalase, oxidase, haemolysis test, esculin hydrolysis, sugar fermentation tests (sucrose, arabinose, mannitol, rhamnose and xylose), methyl red, Voges Proskauer and the use of Microgen™ *Listeria* -ID by following manufacturer's instruction.

Antimicrobial susceptibility testing (AST)

The antimicrobial susceptibility test of the *Listeria monocytogenes* isolates was determined by the Kirby-Bauer disc diffusion method on Mueller Hinton agar (MHA, Oxoid, Basingstoke, UK). Standard discs were applied using a disc dispenser following the procedures recommended by the Clinical and Laboratory Standards Institute [30]. Three to five well distinct colonies of the confirmed *Listeria* isolates were transferred into 5ml Brain Heart Infusion Broth (BHIB, Oxoid) and were incubated at 37°C for 24 h. The overnight broth culture was diluted using sterile normal saline to a turbidity equivalent to 0.5 McFarland standard (10^8cfu/ml) and inoculated onto the dried surface of Muller Hinton agar (MHA, Oxoid) plate. Sterile cotton-tipped swab was used to inoculate the plate using the lawn inoculation technique. The inoculated MHA plates were allowed to dry at room temperature and paper discs impregnated sulphamethoxazole-Trimethoprim (25µg), Kanamycin (30µg), Gentamycin (30µg), Ciprofloxacin (5µg), Nalidixic Acid (30µg), Penicillin G (10 IU), Clindamycin (2µg), Chloramphenicol (30µg), Amoxicillin/Clavulanic Acid 2:1 (30µg), Ampicillin (10µg), Tetracycline (30µg) and Colistin Sulphate (10µg) where placed using antibiotic disc dispenser and incubated at 37°C for 24 hours. After incubation, the diameters (mm) of clear zones of inhibition around each disc was measured and interpreted in accordance with the Clinical and Laboratory Standards Institute guidelines [27] with Staphylococcus aureus ATCC® 25923 (*Pseudomonas aeruginosa* ATCC® 27853 for colistin sulphate) used as control.

Results

Antimicrobial susceptibility testing (AST)

Out of the 360 samples of by-product examined, 152 (42.2%) were positive for *Listeria* species while 208 (57.8%) samples showed no growth on *Listeria* agar. Distribution of the 152 *Listeria* species following biochemical characterization showed that 9 (2.5%) were *L. monocytogenes* while 16 (4.4%), 121 (33.6%) and 6 (1.7%) were *L. grayi, L. ivanovii* and *L. seeligeri* respectively (Table 1).

Table 1: Distribution of *Listeria* species based on total sample collected in Jos North and Jos South LGA of Plateau State using conventional biochemistry testing (n=360).

Species	Frequency	Percentage (%)
L. monocytogenes	9	2.5
L. grayi	16	4.4
L. ivanovii	121	33.6
L. seeligeri	6	1.7
Total	152	42.2

Distribution of *L. monocytogenes* isolates by sample type showed that effluent had the highest level of contamination, 6 (6.7%) which was followed by ruminal content, 2 (2.2%) and the least contamination in the liver sample, 1 (1.1%). The result of the chi-square test showed that there was a statistically significant association between the prevalence of *Listeria monocytogenes* and the sample type in this study (p<0.05) (Table 2).

Table 2: Distribution of *Listeria monocytogenes* Based on Sample Type in Jos North and Jos South LGA.

Sample Type	No. Tested	Number Positive (%)	χ^2	p-value
Effluent	90	6 (6.7)	98.448	0.0001
Liver	90	1 (1.1)		
Ruminal content	90	2 (2.2)		
Spleen	90	-		
Total	360	9 (2.5)		

The level of contamination of the samples with *L. monocytogenes* showed that Yan-Shanu slaughter slab had the highest occurrence, 5 (6.9%) this was followed by Jos Abattoir cattle with 2 (2.8%) and the least contamination in Gyel slaughter slab and Jos abattoir goat with 1 (1.4%) each. There was, however, no statistically significant association between the prevalence of *Listeria monocytogenes* and sampling locations (p>0.05) (Table 3).

Table 3: Distribution of *Listeria monocytogenes* Based on Sampling Location in Jos North and Jos South L.G.A.

Location	No. Tested	Number Positive (%)	χ^2	p-value
Bukuru cattle	72	-	21.498	0.1601
Gyel cattle	72	1 (1.4)		
Jos Abattoir cattle	72	2 (2.8)		
Jos Abattoir goat	72	1 (1.4)		
Yan Shanu cattle	72	5 (6.9)		
Total	**360**	**9 (2.8)**		

Antibiotic sensitivity testing of *listeria monocytogenes*

The antibiotic sensitivity test (Figure 2) showed a high susceptibility to chloramphenicol 88.9% followed by ciprofloxacin 55.6%, amoxicillin and clavulanic acid 55.6% and tetracycline 55.6%. the least susceptibility was obtained for nalidixic acid 11.1% and penicillin G 11.1%. In contrast, a high resistance to ampicillin and colistin sulphate (100%) was observed followed by penicillin G (88.9%). The least resistance was obtained for ciprofloxacin (0%) followed by chloramphenicol (11.1%)
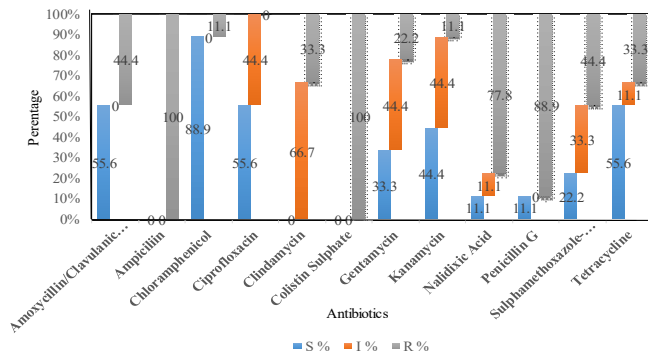

Figure 2: Distribution of antimicrobial susceptibility testing of *Listeria monocytogenes* isolates.
S: Sensitive; I: Intermediate; R: Resistant.

The antimicrobial resistance profile showed that all the isolates were resistance to at least four antimicrobial agents although the majority (88.9%) of the *L. monocytogenes* isolates were resistant to more than four antibiotics (Table 4). The mean multiple antibiotic resistance(MAR) index was 0.47±0.034 with the minimum being 0.33 and maximum 0.67 (Table 5).

Table 4: Antimicrobial resistance pattern of *Listeria monocytogenes* isolates from abattoir/slaughter slabs by-products.

Isolate	Source	Antibiogram	MAR Index
JCEF07	Effluent	SXT, NA, TE, CT, AMP	0.42
YSEF28	Effluent	NA, P, CT, AMP	0.33
JCEF47	Effluent	SXT, CN, P, TE, CT, AMP	0.50
JCRC54	R. content*	SXT, CN, NA, P, TE, DA, CT, AMP	0.67
JCLV71	Liver	SXT, NA, P, DA, CT, AMP	0.50
YSEF78	Effluent	K, P, AMC, C, DA, CT, AMP	0.58
YSEF85	Effluent	NA, P, AMC, CT, AMP	0.42
YSEF96	Effluent	NA, P, AMC, CT, AMP	0.42
GYRC152	R. content*	NA, P, AMC, CT, AMP	0.42

*Ruminal content, SXT: Sulfamethoxazole-Trimethoprim; NA: Nalidixic Acid; TE: Tetracycline; CT: Colistin Sulphate; AMP: Ampicillin; P: Penicillin G; CN: Gentamycin; DA: Clindamycin; K: Kanamycin; C: Chloramphenicol; AMC: Amoxicillin and Clavulanic Acid.

Table 5: Multiple drug resistance of the *Listeria monocytogenes* isolates from abattoir/by-products.

Number of antibiotics	Frequency	Percent (%)
Resistance to three antibiotics	-	-
Resistance to four antibiotics	1	11.1
Resistance to more than four antibiotics	8	88.9
Total	9	100

Discussion

This study established an overall prevalence of 42.2% of *Listeria* species in the abattoir by-product samples in Jos North and Jos South LGA of Plateau State. Similar prevalence of 40% was reported in Iran on chicken carcass by Zeinali *et al*. [31], 40.8% in Sudan on fresh raw dressed broiler by Alsheikh *et al*. [32]. A slightly higher prevalence of 58.2% in North-Central Nigeria on raw beef was reported by Chuku *et al*. [12] and a lower findings of 11.3% in Kano, North-Western Nigeria on some ready to eat foods by Aisha and Kawo [33]. This is probably due the fact that *Listeria* species are ubiquitous in nature and especially found in temperate regions [34], also, domestic ruminant are known to sustain the micro-organisms in their ruminal environment through a repeated faecal-oral enrichment cycle [35]. *Listeria* species have been reported in salad vegetables and vegetable salads [14], milk and milk products [36], raw and processed meat [37], abattoir effluent [24] to mention a few.

The prevalence of individual *Listeria* species was recorded as 2.5%, 4.4%, 33.6% and 1.7% for *Listeria monocytogenes*, *Listeria grayi*, *Listeria ivanovii* and *Listeria seeligeri* respectively. The results of this study was lower than the report of Omogbai and Esokpunwu [13] in frozen beef and chicken samples sold in Benin city, Nigeria except for *Listeria ivanovii* which was 30.43% as compared to 33.6% found in this study. Also, *Listeria innocua* was isolated in their study as while *Listeria seeligeri is* found in this study.

The overall prevalence of 2.5% obtained for *Listeria monocytogenes* is higher than the 1.8% observed by Aisha and Kawo [33] from some ready to eat foods in Kano, North-Western Nigeria and also the 1.29% findings of Eruteya *et al*. [38] from raw cow and goat meat in Port Harcourt, Nigeria. This prevalence is lower than the 36.73% observations of Ebakota *et al*. [15] from ready to eat foods in Southern Nigeria, also it is lower than the 26.8% reports of Falodun and Amusan [23] from untreated abattoir wastewater in Akinyele, Ibadan, Nigeria as well as lower than the 6.55% findings of Usman *et al*. [36] from raw milk and milk products in Northern Nigeria. The inconsistences in the prevalence of *Listeria* species in both raw and processed food as seen in this and other reports cited here could be attributed to the fact that the pathogen is present in the environment and could be as a result of cross contamination from the meat processing plants and after food processing as seen in ready to eat (RTE) foods [39].

The prevalence of *Listeria monocytogenes* was highest in effluent (6.7%) as compared to ruminal content (2.2%) and Liver (1.1%) samples. The association was statistically significant (p=0.0001). The relatively higher prevalence in effluent could be attributed to the fact that most Nigerian abattoirs discharge the effluent untreated through drainages unto water bodies that contaminates surface and ground waters with pathogenic micro-organisms that are most time invariably used in washing the carcass and slaughter hall after the slaughtering activities [40,41,42,43]. The relatively high prevalence in the ruminal content may be because of the fact that food animals are a major reservoir of a lot of foodborne pathogens including *Listeria monocytogenes* [44]. Studies have reported the isolation *Listeria* species and *Listeria monocytogenes* in faecal samples of animals that were fed from infected feedlot [21,45]. The prevalence in liver could be attributed to cross-contamination of meat and by-product by pathogenic organisms during processing activities such as washing of carcass with contaminated water sources [46,47], cutting of carcass on bare floor in the

abattoirs/slaughter slabs due to non-existent, non-functional or refusal of butchers to use the line/railing system and by further polluting the environment with theirs as well as the customers shoes by moving in and out of the slaughter hall [48,49,50].

Antibiotics are extensively used in animals to prevent, control and treat illnesses, they are also enormously used as growth promoters in animal production (25). In the recent past decade, extensive administration of antibiotics has led to misuse of these drugs in humans and animals which has contributed to the widespread antibiotic resistance among foodborne pathogen, *Listeria monocytogenes* inclusive [25]. Antimicrobial resistance has been demonstrated to develop in bacteria either through the bacteria exhibiting some intrinsically resistance to certain antibiotics due to their general physiology, through mutations or other kinds of genetic alteration while others by adapting to environmental stresses [51] such as exposure to heat, salt and anti-oxidants during food production and processing [52].

The high number of *Listeria monocytogenes* isolates (100%) resistant to ampicillin and colistin sulphate observed in this study is similar to the findings of Usman *et al*. [36], Falodun *et al*. [53] and Şanlıbaba *et al*. [16] where 100% of *Listeria monocytogenes* isolates in raw milk and milk products in Northern Kaduna State, Nigeria, from untreated abattoir wastewater in Akinyele, Ibadan, Nigeria and in ready to eat foods in Turkey respectively were reported to be resistant to ampicillin. *Listeria monocytogenes* is naturally susceptible to penicillins (example ampicillin) in combination to aminoglycosides (example gentamycin) to give a synergistic effect as a therapy of choice in the treatment of human listeriosis [54,51]. In this study, *Listeria monocytogenes* isolates displayed resistance to β-lactam antibiotic (ampicillin) which is highly significant from public health perspective because no β-lactamase enzyme has been isolated from environmental isolates before 2010 [55]. These, however does not agree with the findings of Chuku *et al*. [12], Osman *et al*. [27] and Aksoy *et al*. [17] from different ecological niches in Egypt, also from raw beef and chevon in North-Central Nigeria and in raw milk and dairy products in Turkey respectively where 100% of *Listeria monocytogenes* isolates were reported to be sensitive to ampicillin. The high level of *Listeria monocytogenes* isolates resistant to ampicillin suggest there is a misuse of the drug in this region. The low resistance of the isolates to the aminoglycosides: gentamycin 22.2% and kanamycin 11.1% were similar to the 21.4% reports of Ieren *et al*. [14] in salad vegetables and vegetable salads sold in Zaria, Nigeria, 20.9% finding of Chuku *et al*. [12] in raw beef and chevon in North-Central Nigeria and 26.67% observation of [56] from different food sources in Enugu, Nigeria were found to be resistant to gentamycin. Al-Mashhadany [11] also observed a similar resistance of 11.5% to kanamycin. A higher resistance of 52.94% and 76.5% to kanamycin was observed by Vaidya *et al*. [57] and Şanlıbaba *et al*. [16] from slaughtered goats and pigs of Nagpur, Central India and in ready to eat (RTE) foods in Turkey respectively. However, the 33.3% susceptibilities of the isolates to gentamycin and 44.4% to kanamycin was lower than the 100% and 91.1% respectively report of Owusu-Kwarteng *et al*. [19].

The resistance to colistin sulphate (100%) observed in the study is similar to the findings obtained by Neamatallah *et al*. [58] where all the *Listeria monocytogenes* isolates were resistant to colistin sulphate. This however, does not agree with the report of Olaniran *et al*. [59] in which 79.48% of *Listeria monocytogenes* were susceptible to colistin sulphate.

The high resistance of *Listeria monocytogenes* to penicillin G 88.9% and nalidixic acid 77.8% was similar to the work of Şanlıbaba *et al*. [16] where 100% of the isolates were both resistant to penicillin G and nalidixic acid from ready to eat foods in Turkey. Welekidan *et al*. [60] also reported a similar resistance of 76.47% among pregnant women in Tigray region and Vaidya *et al*. [57] observed a resistance of 66.7% from slaughtered goats and pigs in Nagpur, Central India to penicillin G. This finding however does not agree to the report of Osman *et al*. [27] from different ecological niches in Egypt and Aksoy *et al*. [17] in raw milk and dairy products in Turkey where 100% and 93.3% of the isolates were susceptible to penicillin G respectively. Chuku *et al*. [12] also reported a 100% resistance to nalidixic acid while Girma and Abebe [18] a resistance of 30.5%. The 44.4% resistance to sulfamethoxazole-trimethoprim was similar to the findings of Osman *et al*. [27] and Welekidan *et al*. [60] that reported a resistance of 44.4% and 33.3% respectively. The 33.3% resistance to clindamycin observed in this work was lower than the 66.7% and 82.3% findings of Welekidan *et al*. [60] and Owusu-Kwarteng *et al*. [19] respectively while Osman *et al*. [27] and Şanlıbaba *et al*. [16] each reported a much higher resistance of 100% to clindamycin.

Multiple Antibiotic Resistance (MAR) index of the *Listeria monocytogenes* isolates revealed that the mean MAR index was 0.47±0.034. Wong *et al*. [61] stated that a greater than 0.2 MAR index points to the fact that the isolates originate from high-risk area where the rate of antibiotic usage is much. This study is one of such where the minimum MAR index was 0.33. The high level of resistance of *L. monocytogenes* to these drugs should be a great concern to relevant health agencies, regula- tors and the general public. The implication of a multidrug re- sistant pathogen as seen in this study is that, it becomes more pathogenic compared to non-multidrug resistant pathogens[62]. This situation makes treatment regimen unsuccessful in infected patients [63] and there is additional cost of treatment, as well as prolonged hospital stay [64].

Multiple Antibiotic Resistance (MAR) index of the *Listeria monocytogenes* isolates revealed that the mean MAR index was 0.47±0.034. Wong *et al*. [58] stated that a greater than 0.2 MAR index points to the fact that the isolates originate from high-risk area where the rate of antibiotic usage is much. This study is one of such where the minimum MAR index was 0.33. The high level of resistance of *L. monocytogenes* to these drugs should be a great concern to relevant health agencies, regulators and the general public. The implication of a multidrug resistant pathogen as seen in this study is that, it becomes more pathogenic compared to non-multidrug resistant pathogens [59]. This situation makes treatment regimen unsuccessful in infected patients [60] and there is additional cost of treatment, as well as prolonged hospital stay [61].

Conclusion

The results obtained from this study have shown that *Listeria monocytogenes* is at 6.7% in effluent, 2.2% in ruminal content and 1.1% in liver abattoir by-products. These are of public health significance based on the fact that the meat processing environment is contaminated with this pathogenic organism and this have been shown to cross contaminate meat by-products (liver). These could result in listeriosis in animals when grazing in the abattoir/slaughter slabs environment which could invariably infect susceptible humans.

The result also showed that 100% of the isolates displayed resistance to more than three antimicrobial agents to which they were tested. The results also showed that ciprofloxacin is the best drug for the treatment of listeriosis since no (0%) isolate exhibited resistance to the drug. This is followed by chloramphenicol and kanamycin where only 11.1% of the isolates each showed resistance to the drugs.

It was discovered from this study that effluent and ruminal content that are discharged untreated into stagnant drainages and piled up in the environment respectively may pose a greater public health threat. This requires that effluent and ruminal content be treated before further use.

References

1. Dortet L, Veiga-Chacon E, Cossart P. Listeria Monocytogenes. In Encyclopedia of Microbiology. 2009; 182–198.

2. Orsi RH, Wiedmann M. Characteristics and distribution of Listeria spp., including Listeria species newly described since 2009. Applied Microbiology and Biotechnology. 2016; 100: 5273–5287.

3. Scallan E, Hoekstra RM, Angulo FJ, Tauxe RV, Widdowson MA, et al. Foodborne illness acquired in the United States-major pathogens. Emerging Infectious Diseases. 2011; 17: 7–15.

4. Favaro M, Sarmati L, Sancesario G, Fontana C. First case of Listeria innocua meningitis in a patient on steroids and eternecept. JMM case reports. 2014; 1: e003103.

5. Guillet C, Join-Lambert O, Le Monnier A, Leclercq A, Mecha F, et al. Human listeriosis caused by Listeria ivanovii. Emerging Infectious Diseases. 2010; 16: 136–138.

6. Perrin M, Bemer M, Delamare C. Fatal case of Listeria innocua bacteremia. Journal of Clinical Microbiology. 2003; 41: 5308–5309.

7. Centre for Disease Control and Prevention (CDC). Listeria (listeriosis): Multistate Outbreak of Listeriosis Linked to Soft Raw Milk Cheese Made by Vulto Creamery (Final Update). 2017.

8. Hof H. Miscellaneous Pathogenic Bacteria. In Medical Microbiology. 1996; 4th Edition.

9. Tchatchouang CDK, Fri J, De Santi M, Brandi G, Schiavano GF, et al. Listeriosis outbreak in south africa: A comparative analysis with previously reported cases worldwide. Microorganisms. 2020; 8: 1-18.

10. Thomas J, Govender N, McCarthy KM, Erasmus LK, Doyle TJ et al. Outbreak of Listeriosis in South Africa Associated with Processed Meat. New England Journal of Medicine. 2020; 382: 632–643.

11. Al-Mashhadany DA. Occurrence and antibiogram of Listeria monocytogenes Isolates from Retail Meat Shops at Erbil City, Kurdistan Region, Iraq. Italian Journal of Food Safety. 2019; 8: 8451: 194–198.

12. Chuku A, Obande GA, Eya SB. Listerial contamination of raw beef and chevon in north-central Nigeria. IMC Journal of Medical Science. 2019; 13: 1–8.

13. Omogbai BA, Esokpunwu DE. Molecular Characterization and Antibiotic Resistance Patterns of Listeria species in Frozen Beef and Chicken Sold in Benin City, Nigeria Barry. Food and Applied Bioscience Journal. 2019; 7: 68–86.

14. Ieren II, Bello M, Kwaga JKP. Occurrence and antibiotic resistance profile of Listeria monocytogenes in salad vegetables and vegetable salads sold in Zaria, Nigeria. African Journal of Food Science. 2013; 7: 334–338.

15. Ebakota DO, Abiodun OA, Nosa OO. Prevalence of Antibiotics Resistant Listeria monocytogenes Strains in Nigerian Ready-to-eat Foods. Food Safety. 2018; 6: 118–125.

16. Şanlıbaba P, Tezel BU, Çakmak GA. Prevalence and antibiotic resistance of Listeria monocytogenes isolated from ready-to-eat foods in Turkey. Journal of Food Quality. 2018; 2018: 1–9.

17. Aksoy A, Sezer Ç, Vatansever L, Gülbaz G. Presence and antibiotic resistance of Listeria monocytogenes in raw milk and dairy products. Kafkas Universitesi Veteriner Fakultesi Dergisi. 2018; 24: 415–421.

18. Girma Y, Abebe, B. Isolation, Identification and Antimicrobial Susceptibility of Listeria Species from Raw Bovine Milk in Debre-Birhan Town, Ethiopia. Journal of Zoonotic Diseases and Public Health. 2018; 2: 1–7.

19. Owusu-Kwarteng J, Wuni A, Akabanda F, Jespersen L. Prevalence and Characteristics of Listeria monocytogenes Isolates in Raw Milk, Heated Milk and Nunu, a Spontaneously Fermented Milk Beverage, in Ghana. Beverages. 2018; 4: 40.

20. Oyinloye MA, David OM, Famurewa O. Pathogenic potential of Listeria monocytogenes isolated from cattle faeces in Adoekiti. African Journal of Clinical and Experimental Microbiology. 2018; 19: 104.

21. Matto C, Varela G, Braga V, Vico V, Gianneechini RE, et al. Detec- tion of Listeria spp. in cattle and environment of pasture-based dairy farms. Pesquisa Veterinária Brasileira. 2018; 38: 1736–1741.

22. Ferreira V, Wiedmann M, Teixeira P, Stasiewicz MJ. Listeria monocytogenes Persistence in Food-Associated Environments: Epidemiology, Strain Characteristics, and Implications for Public Health. Journal of Food Protection. 2017; 77: 150–170.

23. Falodun O, Amusan M. Antibiotics Susceptibility Profile of Listeria Species Isolated from Poultry Wastes and Fishpond Water from Private and Institutional Farms in Ibadan, Nigeria. Journal of Advances in Biology & Biotechnology. 2017; 13: 1–8.

24. 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: 2141–5463.

25. Wilson A, Gray J, Scott Chandry P, Fox EM. Phenotypic and genotypic analysis of antimicrobial resistance among Listeria monocytogenes isolated from Australian food production chains. Genes. 2018; 9:2.

26. Frieden T. Antibiotic resistance threats in the United States. Centers for Disease Control and Prevention, US Department of Health and Human Services. 2013; 23: 11-28.

27. Osman KM, Kappell AD, Fox EM, Orabi A, Samir A. Prevalence, Pathogenicity, Virulence, Antibiotic Resistance, and Phylogenetic Analysis of Biofilm-Producing Listeria monocytogenes Isolated from Different Ecological Niches in Egypt: Food, Humans, Animals, and Environment. Pathogens. 2020; 9: 5.

28. Alao O, Sadiq L. Jos: Rise of Illegal Slaughter Slabs. Daily trust. 2014.

29. Indrawattana N, Nibaddhasobon T, Sookrung N, Chongsa-nguan M, Tungtrongchitr A, et al. Prevalence of Listeria monocytogenes in raw meats marketed in Bangkok and characterization of the isolates by phenotypic and molecular methods. Journal of Health, Population and Nutrition 2011; 29: 26–38.

30. Clinical and Laboratory Standards Institute (CLSI). Performance Standards for Antimicrobial Susceptibility Testing. CLSI supplement M100S. Wayne, PA. 2016; 26th ed.

31. Zeinali T, Jamshidi A, Bassami M, Rad M. Isolation and identification of Listeria spp. in chicken carcasses marketed in Northeast of Iran. International Food Research Journal. 2017; 24: 881-887.

32. Alsheikh ADI, Mohammed GE, Abdalla MA. First isolation and identification of Listeria monocytogenes from fresh raw dressed broiler chicken in Sudan. Research Journal of Microbiology. 2012; 7: 319–326.

33. Aisha B, Kawo A. Isolation of Listeria monocytogenes recovered from some ready-to-eat foods sold in Kano, north-western Nigeria. Bayero Journal of Pure and Applied Sciences. 2015; 7: 8.

34. Dhama K, Karthik K, Tiwari R, Shabbir MZ, Barbuddhe S, Malik SVS, et al. Listeriosis in animals, its public health significance (food-borne zoonosis) and advances in diagnosis and control: a comprehensive review. Veterinary Quarterly. 2015; 35: 211–235.

35. Sasakawa C. (Ed.). Listeria as an enteroinvasive gastrointestinal pathogen. In Molecular Mechanisms of Bacterial Infection via the Gut. 2009; (337) Springer.

36. Usman UB, Kwaga JKP, Kabir J, Olonitola OS. Isolation and Antimicrobial Susceptability of Listeria monocytogenes from Raw Milk and Milk Products in Northern Kaduna State, Nigeria. Journal of Applied & Environmental Microbiology. 2016; 4: 46–54.

37. Ohue LA, Enurah LU, Abaoaba OO, Aboaba O. Prevalence and antibiotic susceptiblity profile of Listeria monocytogenes isolated from processed and unprocessed meat products in Lagos, Nigeria. Science & Technology. 2015; 1: 12–18.

38. Eruteya OC, Odunfa SA, Lahor J. Listeria spp. in Raw Cow and Goat Meat in Port Harcourt, Nigeria. British Biotechnology Journal. 2014; 4: 204–214.

39. Tabit FT. Contamination, Prevention and Control of Listeria monocytogenes in Food Processing and Food Service Environments. In Listeria Monocytogenes. 2018; 71–85.

40. Egesi CO, Ezebuiro V, Ekeleme AC, Oblukwu CE. Compliance Monitoring of Microbiological and Physicochemical Parameters of Abattoirs' Effluents Discharged into Water Bodies in Owerri, Nigeria Constance. Microbiology Research Journal International. 2019; 28: 1–16.

41. Bello M, Lawan MK, Aluwong T, Sanusi M. Management of slaughter houses in northern Nigeria and the safety of meat produced for human consumption. Food Control. 2015; 49: 34–39.

42. Akinro AO, Ologunagba IB, Yahaya O. Environmental Implications of Unhygienic Operation of a City Abattoir in Akure, Western Nigeria. ARPN Journal of Engineering and Applied Sciences. 2009; 4: 60–63.

43. Adeyemo OK. Unhygienic operation of a city abattoir in South Western Nigeria: Environmental implication. African Journal of Environmental Assessment and Management. 2002; 4: 23-28.

44. Heredia N, García S. Animals as sources of food-borne pathogens: A review. Animal Nutrition. 2018; 4: 250–255.

45. Skovgaard N, Morgen CA. Detection of Listeria spp. in faeces from animals, in feeds, and in raw foods of animal origin. International Journal of Food Microbiology. 1988; 6: 229–242.

46. Nafarnda WD, Ajayi IE, Shawulu JC, Kawe MS, Omeiza GK, et al. Bacteriological Quality of Abattoir Effluents Discharged into Water Bodies in Abuja, Nigeria. ISRN Veterinary Science. 2012; 2012: 1–5.

47. Bello M, Lawan MK, Kwaga JKP, Raji MA. Assessment of carcass contamination with E. coli O157 before and after washing with water at abattoirs in Nigeria. International Journal of Food Microbiology. 2011; 150: 184–186.

48. Kwage AV, Adesokan HK, Ameh JA, Ambali A, Ndahi MD, et al. The Debilitating State of the Maiduguri Main Abattoir, Poor Sanitary and Waste Disposal Methods; the Way Forward. Nature and Science. 2016; 14: 38–48.

49. Lawan MK, Bello M, Kwaga JKP, Raji MA. Evaluation of physical facilities and processing operations of major abattoirs in Northwestern states of Nigeria. Sokoto Journal of Veterinary Sciences. 2013; 1: 56–61.

50. Adeyemo OK, Adeyemi IG, Awosanya EJ. Cattle cruelty and risks of meat contamination at Akinyele cattle market and slaughter slab in Oyo State, Nigeria. Tropical Animal Health and Production. 2009; 41: 1715-1721.

51. Olaimat AN, Al-Holy MA, Shahbaz HM, Al-Nabulsi AA, Abu Ghoush MH, et al. Emergence of Antibiotic Resistance in Listeria monocytogenes Isolated from Food Products: A Comprehensive Review. Comprehensive Reviews in Food Science and Food Safety. 2018; 17: 1277–1292.

52. Wesche AM, Gurtler JB, Marks BP, Ryser ET. Stress, Sublethal Injury, Resuscitation, and Virulence of Bacterial Foodborne Pathogens . In Journal of Food Protection. 2009.

53. Falodun OI, Rabiu AG, Fagade OE, Blaiotta G, Miguel HKC. Antibiotics Susceptibility Profile of Listeria species Isolated from Untreated Abattoir Wastewater in Akinyele, Ibadan, Nigeria and Its Implication on Public Health. British Microbiology Research Journal. 2016; 16: 1–10.

54. Alonso-Hernando A, Prieto M, Garc´ıa-Fernandez C, Alonso-Calleja C, Capita R. Increase over time in the prevalence of multiple antibiotic resistance among isolates of Listeria monocytogenes from poultry in Spain. Food Control. 2012; 23: 37–41

55. Collins B, Curtis N, Cotter PD, Hill C, Ross RP. The ABC transporter AnrAB contributes to the innate resistance of Listeria monocytogenes to nisin, bacitracin, and various β-lactam antibiotics. Antimicrobial Agents and Chemotherapy. 2010; 54: 4416–4423.

56. Chiamaka E, Emeka N, Ifeanyichu E. Incidence and Antimicrobial Susceptibility of Listeria monocytogenes Isolated from Different Food Sources in Enugu, Nigeria. Asian Journal of Biological Sciences. 2019; 12: 671–676.

57. Vaidya GR, Chaudhary SP, Zade NN, Khan WA, Shinde SV, et al. Prevalence, Virulence and Antibiotic Susceptibility of Listeria monocytogenes Recuperated from Slaughtered Goats and Pigs of Nagpur, Central India. Int.J.Curr.Microbiol.App.Sci. 2017; 7: 1566–1578.

58. Neamatallah AAN, Dewar SJ, Austin B. An improved selective isolation medium for the recovery of Listeria monocytogenes from smoked fish. Letters in Applied Microbiology. 2003; 36: 230–233.

59. Olaniran AO, Nzimande SBT, Mkize NG. Antimicrobial resistance and virulence signatures of Listeria and Aeromonas species recovered from treated wastewater effluent and receiving surface water in Durban, South Africa. BMC Microbiology. 2015; 15: 234.

60. Welekidan LN., Bahta YW, Teklehaimanot MG, Abay GK, Wasihun AG, Dejene TA, et al. Prevalence and drug resistance pattern of Listeria monocytogenes among pregnant women in Tigray region, Northern Ethiopia: a cross-sectional study. BMC Research Notes. 2019; 12: 538.

61. Wong WC, Pui CF, Chai LC, Lee HY, Ghazali FM et al. Biosafety assessment of Listeria monocytogenes in vegetarian burger patties in Malaysia. International Food Research Journal. 2011; 18: 459–463.

62. Foley SL, Lynne AM. Food animal-associated Salmonella challenges: Pathogenicity and antimicrobial resistance. Journal of Animal Science. 2008.

63. Adzitey F, Huda N, Ali GRR, Manuel CS. Isolation and characterization of unidentified Listeria strains showing phenotypic and genetic similarities to a novel group of Listeria species from Malaysian Pekin Ducks. Journal of Biological Sciences. 2013; 13: 614–620.

64. Buzby JC, Roberts T. The Economics of Enteric Infections: Human Foodborne Disease Costs. Gastroenterology. 2009; 136: 1851–1862.