



PRINT ISSN 1119-8362
Electronic ISSN 2659-1499

Full-text Available Online at
<https://www.ajol.info/index.php/jasem>
<https://www.bioline.org.br/ja>

J. Appl. Sci. Environ. Manage.
Vol. 30 (3) 1052-1064 March 2026

Phenotypic and Molecular Identification, Prevalence and Distribution of *Listeria* spp in *Lactuca sativa*, *Daucus carota* and *Brassica oleracea* Obtained from Selected Markets in Lagos State, Nigeria

***ILEVBARE, LO; UZEH, RE; ODUMOSU, BT**

Department of Microbiology, University of Lagos, Akoka, Lagos State, Nigeria

*Corresponding Author Email: lovethilevbare@gmail.com

*ORCID: <https://orcid.org/0009-0004-7063-1446>

*Tel: +2347031185415

Co-Authors Email: ruzeh@unilag.edu.ng; bodumosu@unilag.edu.ng

ABSTRACT: *Listeria monocytogenes* is a foodborne pathogen of significant public health concern particularly when associated with fresh vegetables that are consumed raw or minimally processed. A total of 354 vegetable samples were collected from five major markets and analysed using standard culture, morphological and biochemical methods, followed by polymerase chain reaction and sequencing for molecular confirmation. Mean bacterial counts varied significantly across markets with vegetables from Sabo Yaba recording the highest microbial loads while Idumota recorded the lowest. Molecular analysis confirmed the presence of *Listeria monocytogenes*, *Listeria innocua*, and *Listeria ivanovii*, with *L. monocytogenes* being the predominant species. Phylogenetic analysis revealed clear clustering of isolates according to species and demonstrated close genetic relatedness to reference strains. The overall prevalence of *Listeria* species ranged from 17.80 percent to 19.49 percent across the vegetables studied with cabbage showing the highest contamination rate. The findings highlight the widespread occurrence of *Listeria* species in fresh vegetables sold in open markets and underscore the need for improved hygiene practices and routine molecular surveillance to reduce the risk of listeriosis.

DOI: <https://dx.doi.org/10.4314/jasem.v30i3.39>

License: **CC-BY-4.0**

Open Access Policy: All articles published by **JASEM** are open-access and free for anyone to download, copy, redistribute, repost, translate, and read.

Copyright Policy: © 2026. Authors retain the copyright and grant **JASEM** the right of first publication. Any part of the article may be reused without permission, provided that the original article is cited.

Cite this Article as: ILEVBARE, L. O; UZEH, R. E; ODUMOSU, B. T. (2026). Phenotypic and Molecular Identification, Prevalence and Distribution of *Listeria* spp in *Lactuca sativa*, *Daucus carota* and *Brassica oleracea* Obtained from Selected Markets in Lagos State, Nigeria. *J. Appl. Sci. Environ. Manage.* 30 (3) 1052-1064

Dates: Received: 26 February 2026; Revised: 22 March 2026; Accepted: 27 March 2026; Published: 31 March 2026

Keywords: contamination, vegetables, *Listeria*, prevalence, markets

Listeria monocytogenes is a Gram-positive facultative anaerobic, ubiquitous, and persistent bacterium in food processing plants. Due to inadequate hygiene and manufacturing practices, this pathogen contaminates foods such as fresh or frozen fruits and vegetables, unpasteurized dairy products, sausages, and fish (Amajoud *et al.*, 2018). Cheeses, sausages, meats, and fish are the most frequently associated with outbreaks of this pathogen worldwide (Kurpas *et al.*, 2018; Ryser, 2021). *L. monocytogenes*

causes listeriosis, a disease characterized by low morbidity but high mortality in those who are infected, and the most at-risk groups are pregnant women, newborns, children, and older adults (Schlech, 2019). In Europe and North America, invasive listeriosis affects 0.3–0.6 persons per 100,000 inhabitants annually (Maertens de Noordhout *et al.*, 2017; European Centre for Disease Prevention and Control [ECDC], 2018). In Chile, *L. monocytogenes* has been under mandatory laboratory

*Corresponding Author Email: lovethilevbare@gmail.com

*ORCID: <https://orcid.org/0009-0004-7063-1446>

*Tel: +2347031185415

notification and surveillance since 2005 (Bustamante *et al.*, 2020). Adults aged 65 and older and pregnant women were the most affected groups.

The severity of *L. monocytogenes* infection is associated with several factors such as infecting dose, host immunity, and expression of virulence factors (adherence, invasion, immune modulator, intracellular survival, toxins, and mobile genetic elements), and the presence of CRISPR-Cas as a virulence regulator (Falavina dos Reis *et al.*, 2011; Louwen *et al.*, 2014; Pouillot *et al.*, 2016; Buchanan *et al.*, 2017; Kwon *et al.*, 2020). In addition, there are other factors such as resistance to disinfectants and antibiotics, especially beta-lactams (Olaimat *et al.*, 2018). The capacity to resist adverse environmental conditions (heat and cold stress) allows the persistence and colonization throughout the food chain by forming contamination reservoirs that are difficult to control (Bolocan *et al.*, 2016; Bucur *et al.*, 2018). The *Listeria* species can be categorized into different serotypes according to the serological reactions of the *Listerial* somatic antigen (O-antigen) and flagellar antigen (H-antigen) with specific antisera. *L. monocytogenes* can be classified into at least 13 serotypes (Orsi *et al.*, 2011), three of them (1/2a, 1/2b, and 4b) are involved in over 95% of human clinical cases, and serotype 4b exhibits the strongest epidemiological association with human listeriosis (Maury *et al.*, 2016; Lee *et al.*, 2018).

Ready-to-eat (RTE) foods are defined as any food in a raw state or one that is handled, processed, mixed, cooked, or prepared and is consumed without any further processing (Monteiro, 2010). The RTE foods are a practical alternative to meet daily food needs; however, they are not exempt from contamination by biological hazards such as *Salmonella* spp., pathogenic *Escherichia coli*, and *Listeria monocytogenes* (Becker *et al.*, 2019).

Although historically associated with ready-to-eat meats and dairy, evidence accumulated over the last decade demonstrates that plants and plant-derived foods are viable habitats and vectors for *L. monocytogenes*. Plants (including leafy greens and root vegetables) may be contaminated from natural reservoirs such as soil, irrigation water, manure, or during harvest and postharvest handling, and the bacterium's capacity to attach to plant surfaces and form biofilms increases the difficulty of eliminating it from both primary production environments and processing facilities (Truong *et al.*, 2021; Townsend *et al.*, 2021). These plant-associated reservoirs mean that commonly consumed vegetables such as lettuce (*Lactuca sativa*), carrot (*Daucus carota*), and

brassicas (*Brassica oleracea* and relatives) can act as vehicles for *L. monocytogenes*.

Fresh vegetables such as lettuce (*Lactuca sativa*), carrot (*Daucus carota*), and cabbage (*Brassica oleracea*) are widely consumed globally in Lagos, Nigeria, often in raw or minimally processed forms. While these foods offer essential nutrients, they also serve as potential vehicles for foodborne pathogens. One major pathogen of concern is *Listeria monocytogenes*, a psychrotrophic bacterium capable of causing severe and sometimes fatal listeriosis, especially among high-risk groups such as pregnant women, neonates, older adults and immunocompromised individuals. The burden of *L. monocytogenes* in developing countries is under-reported, yet available evidence suggests growing public-health relevance.

Ajayeoba *et al.* (2015) reported the detection of *L. monocytogenes* in lettuce, carrots, and cabbage sold in markets across southwestern Nigeria, with Lagos recording one of the highest contamination rates. Similarly, Ieren (2013) and Mumuney *et al.* (2021) found the pathogen in salad vegetables sold in Zaria and Sokoto, respectively, demonstrating widespread environmental and food-chain contamination. Given the complex vegetable supply chain in the Lagos metropolis—ranging from farms and irrigation systems to markets and roadside vendors—critical gaps remain in understanding where contamination occurs and how pathogenic, resistant strains spread. Without comprehensive and up-to-date data, effective risk assessment, prevention strategies and regulatory interventions remain difficult. This gap underscores the need for a detailed investigation into the prevalence, virulence potential, antimicrobial resistance patterns, and contamination routes of *L. monocytogenes* on commonly consumed vegetables in Lagos. Therefore, the objective of this paper is to investigate the phenotypic and molecular identification, prevalence and distribution of *Listeria* species in lettuce (*Lactuca sativa*), cabbage (*Brassica oleracea*) and Carrot (*Daucus carota*) obtained selected markets in Lagos State, Nigeria

MATERIALS AND METHODS

Sample Collection and Preparation: Fresh vegetables samples such as lettuce (*Lactuca sativa*), cabbage (*Brassica oleracea*) and Carrot (*Daucus carota*) were obtained from Ikotun (Alimosho LGA), Ajangbadi (Ojo LGA), Mile 12 (Kosofe), Sabo-Yaba (Lagos Mainland), and Idumota (Lagos Island) markets Lagos State. A total of 354 samples were obtained from the open market for a period of 4 weeks. The samples were placed separately in a sterile

polyethylene bags and taken immediately to the laboratory. All the samples obtained only included fresh vegetables and samples which showed any sign of spoilage were excluded. Fresh produce samples were packaged and stored at 4°C before the analysis.

Enumeration and Isolation of Bacteria: This was done according to Cheesbrough (2013). Five test tubes were labeled 10-1, 10-2, 10-3, 10-4 and 10-5. With the aid of a sterile pipette, 9 ml of distilled water was transferred into each test tube. The test tubes were sterilized in the autoclave and allowed to cool down. The intact vegetable samples were washed with water and placed in a stomacher bag containing 225 ml of peptone water. For the control, the vegetable samples were first washed with vinegar before being placed in a stomacher bag containing 225 ml of peptone water. The stomacher bag was then placed in the stomacher machine for vigorous shaking and mixing of the vegetable samples with the peptone water. 1ml of the mixed peptone water from the stomacher bag was measured using a 25ml syringe and added to the distilled water in test tube 10-1 and shaken vigorously to mix properly. 1ml was transferred from test tube 10-1 to test tube 10-2, this was done till the last test tube (10-5).

Inoculation and incubation: Aliquot (1ml) of the 10-1 dilution was introduced into 3 Petri dishes (in triplicate) and poured with PALCAM agar (Phenol Red, Acriflavine, Lithium Chloride, Cefazidime, and Aesculin with Mannitol) and Nutrient agar and were allowed to set and then incubated at 37°C for 24 h in an inverted position.

Identification of Bacterial Isolates: At the end of the incubation period, the culture plates were examined for growth. The population of the bacterial isolates was estimated by counting the number of discrete colonies in the plate which was expressed in colony forming unit (cfu) per gram. Morphological and cultural characteristics were observed. Discrete colonies were sub-cultured by streak plate method in order to obtain pure culture. Stock cultures of each colony were prepared in the agar slants which were incubated for 24 h to allow for growth after which they were stored at 4°C in the refrigerator for further use. Preliminary identification was carried out based on cultural and morphological characteristics. Polymerase chain reaction technique was then used to confirm the identity of the isolates

Bacteria DNA extraction: DNA was extracted using the protocol stated by Wilson (2001). Briefly, Single colonies grown on medium were transferred to 1.5 ml of liquid medium and cultures were grown on a

shaker for 48 h at 28 °C. After this period, cultures were centrifuged at 4600g for 5 min. The resulting pellets were resuspended in 520 µl of TE buffer (10 mM Tris-HCl, 1mM EDTA, pH 8.0). Fifteen microliters of 20% SDS and 3 µl of Proteinase K (20 mg/ml) were then added. The mixture was incubated for 1 hour at 37 °C, then 100 µl of 5 M NaCl and 80 µL of a 10% CTAB solution in 0.7 M NaCl were added and vortexed. The suspension was incubated for 10 min at 65 °C and kept on ice for 15 min. An equal volume of chloroform: isoamyl alcohol (24:1) was added, followed by incubation on ice for 5 min and centrifugation at 7200g for 20 min. The aqueous phase was then transferred to a new tube and isopropanol (1: 0.6) was added and DNA precipitated at -20 °C for 16 h. DNA was collected by centrifugation at 13000g for 10 min, washed with 500 µl of 70% ethanol, air-dried at room temperature for approximately three hours and finally dissolved in 50 µl of TE buffer.

Polymerase Chain Reaction Analysis: PCR sequencing preparation cocktail consisted of 10 µl of 5x GoTaq colourless reaction, 3 µl of 25mM MgCl₂, 1 µl of 10 mM of dNTPs mix, 1 µl of 10 pmol each 27F 5'-AGA GTT TGA TCM TGG CTC AG-3' and - 1525R, 5'-AAGGAGGTGATCCAGCC-3' primers and 0.3units of Taq DNA polymerase (Promega, USA) made up to 42 µl with sterile distilled water 8µl DNA template. PCR was carried out in a GeneAmp 9700 PCR System Thermalcycler (Applied Biosystem Inc., USA) with a Pcr profile consisting of an initial denaturation at 94°C for 5 min; followed by a 30 cycles consisting of 94°C for 30 s, 50°C for 60s and 72°C for 1 minute 30 seconds ; and a final termination at 72°C for 10 mins. And chill at 4°C. GEL (2,3)

Integrity: The integrity of the amplified gene fragment was checked on a 1% Agarose gel run to confirm amplification. The buffer (1XTAE buffer) was prepared and subsequently used to prepare 1.5% agarose gel. The suspension was boiled in a microwave for 5 minutes. The molten agarose was allowed to cool to 60°C and stained with 3µl of 0.5 g/ml ethidium bromide (which absorbs invisible UV light and transmits the energy as visible orange light). A comb was inserted into the slots of the casting tray and the molten agarose was poured into the tray. The gel was allowed to solidify for 20 minutes to form the wells. The 1XTAE buffer was poured into the gel tank to barely submerge the gel. Two microliter (2 l) of 10X blue gel loading dye (which gives colour and density to the samples to make it easy to load into the wells and monitor the progress of the gel) was added to 4µl of each PCR product and loaded into the wells

after the 100bp DNA ladder was loaded into well 1. The gel was electrophoresed at 120V for 45 minutes visualized by ultraviolet trans-illumination and photographed. The sizes of the PCR products were estimated by comparison with the mobility of a 100bp molecular weight ladder that was ran alongside experimental samples in the gel.

Purification of Amplified Products: After gel integrity, the amplified fragments were ethanol purified in order to remove the PCR reagents. Briefly, 7.6 µl of Na acetate 3M and 240 µl of 95% ethanol were added to each about 40µl PCR amplified product in a new sterile 1.5 µl tube eppendorf, mix thoroughly by vortexing and keep at -20°C for at least 30 min. Centrifugation for 10 min at 13000 g and 4°C followed by removal of supernatant (invert tube on trash once) after which the pellet were washed by adding 150 µl of 70% ethanol and mix then centrifuge for 15 min at 7500 g and 4°C. Again remove all supernatant (invert tube on trash) and invert tube on paper tissue and let it dry in the fume hood at room temperature for 10-15 min. then resuspend with 20 µl of sterile distilled water and kept in -20°C prior to sequencing. The purified fragment was checked on a 1.5% Agarose gel ran on a voltage of 110V for about 1hr as previous, to confirm the presence of the purified product and quantified using a nanodrop of model 2000 from thermo scientific.

Sequencing: The amplified fragments were sequenced using a Genetic Analyzer 3130xl sequencer from Applied Biosystems using manufacturers' manual while the sequencing kit used was that of BigDye terminator v3.1 cycle sequencing kit. Bio- Edit software and MEGA 6 were used for all genetic analysis

Data Analysis: Statistical analyses were carried out using SPSS 24. Descriptive statistics was used to present data as mean plus/minus standard error. Frequency distribution was used to analyse the distribution and prevalence of isolates while one way ANOVA was used to find significant differences among variables

RESULTS AND DISCUSSION

Table 1 presents the mean bacterial counts ($\times 10^3$ CFU/g) obtained from PALCAM agar for the three vegetables sampled across the five markets. The results show considerable variation in the microbial load among markets, although the differences among vegetables within the same market were not statistically significant ($p > 0.05$). Sabo-Yaba produced the highest bacterial counts across all three

vegetables, ranging from 7.87 to 8.75×10^3 CFU/g, suggesting a high degree of microbial contamination likely associated with environmental exposure, handling practices, or unhygienic market conditions. In contrast, Idumota recorded the lowest bacterial loads for all vegetables, with values between 2.30 and 2.56×10^3 CFU/g, indicating comparatively cleaner produce or better sanitary conditions in that market. Across all markets, lettuce, carrots, and cabbage exhibited similar levels of contamination within the same location, as shown by the non-significant p-values.

The bacterial counts obtained on PALCAM agar demonstrate clear differences in the microbiological quality of vegetables sold across the five markets and provide strong evidence that market related factors are the primary drivers of contamination. The substantial variation in mean bacterial loads among markets indicates that post-harvest handling practices environmental exposure and sanitation at the point of sale exert a greater influence on microbial burden than vegetable type. Similar observations have been reported in studies assessing fresh produce in open markets where differences in hygiene and infrastructure accounted for most of the variability in microbial counts rather than crop specific characteristics (Alegbeleye *et al.* 2018). The consistently high bacterial loads observed in vegetables from Sabo Yaba suggest significant exposure to microbial contamination. Counts ranging from 7.87 to $8.75 \log_{10}$ cfu per gram indicate heavy contamination and raise concerns regarding food safety. Open market environments characterized by overcrowding, poor waste management and limited access to potable water have been strongly associated with elevated bacterial loads on fresh produce (Abakari *et al.* 2018). In such settings vegetables are often displayed on bare surfaces or on the ground which increases contact with contaminated soil dust and wastewater. In addition, repeated handling by vendors and consumers without adequate hand hygiene further contributes to microbial transfer. These factors collectively create conditions that favor the persistence and proliferation of bacteria including *Listeria* species which are selectively supported by PALCAM agar and are known to survive under adverse environmental conditions (Carstens *et al.* 2019). In contrast, vegetables obtained from Idumota exhibited markedly lower bacterial loads across all three vegetable types. Mean values between 2.30 and $2.56 \log_{10}$ cfu per gram suggest comparatively better microbiological quality and reduced exposure to contamination. Lower microbial loads of this magnitude have been linked to improved market sanitation, better drainage systems and more frequent

washing of produce with cleaner water (Uyttendaele *et al.* 2015). The findings imply that vendors in this market may be employing basic hygienic practices that effectively limit bacterial contamination even in the absence of advanced food safety controls. This supports previous evidence that relatively simple interventions at the retail level can significantly improve the microbial safety of fresh vegetables (Gil *et al.* 2015). The absence of statistically significant differences in bacterial counts among lettuce carrots and cabbage within the same market further emphasizes the dominant role of shared environmental conditions. Although leafy vegetables such as lettuce are often considered more susceptible to contamination due to their large surface area and proximity to soil during cultivation the present findings indicate that once vegetables enter the same distribution and retail environment these intrinsic differences become less influential (Oliveira *et al.* 2015). When vegetables are washed with the same water displayed on similar surfaces and handled by the same vendors they are exposed to similar contamination pathways. This pattern has been reported in multiple studies which concluded that market hygiene and handling practices outweigh vegetable morphology in determining microbial load at the point of sale (Alegbeleye *et al.* 2018; Abakari *et al.* 2018).

Table 1: bacterial counts ($\times 10^3$ CFU/g) on PALCAM agar

Markets	Samples	N	Bacterial Counts
Ikotun	<i>Lactuca sativa</i>	24	7.54 $\pm$ 1.23
	<i>Daucus carota</i>	24	7.79 $\pm$ 0.47
	<i>Brassica oleracea</i>	23	7.95 $\pm$ 0.44
	p-value		0.936
Ajangbadi	<i>Lactuca sativa</i>	24	5.16 $\pm$ 0.58
	<i>Daucus carota</i>	23	4.73 $\pm$ 0.53
	<i>Brassica oleracea</i>	24	4.58 $\pm$ 0.57
	p-value		0.751
Mile 12	<i>Lactuca sativa</i>	23	7.00 $\pm$ 0.85
	<i>Daucus carota</i>	24	5.91 $\pm$ 0.70
	<i>Brassica oleracea</i>	24	5.79 $\pm$ 0.59
	p-value		0.436
Sabo-Yaba	<i>Lactuca sativa</i>	24	8.75 $\pm$ 0.51
	<i>Daucus carota</i>	23	8.08 $\pm$ 0.54
	<i>Brassica oleracea</i>	24	7.87 $\pm$ 0.65
	p-value		0.528
Idumota	<i>Lactuca sativa</i>	23	2.30 $\pm$ 0.41
	<i>Daucus carota</i>	24	2.41 $\pm$ 0.38
	<i>Brassica oleracea</i>	23	2.56 $\pm$ 0.35
	p-value		0.891

From a public health perspective the results are significant because vegetables are often consumed raw or minimally processed which limits the effectiveness of subsequent microbial reduction steps. High bacterial loads on retail vegetables have

been linked to foodborne disease outbreaks particularly those involving *Listeria monocytogenes* and other enteric pathogens (Carstens *et al.* 2019). The findings therefore highlight the need for routine microbiological surveillance of vegetables sold in open markets as well as targeted interventions focused on improving hygiene at the retail level. Provision of clean water enforcement of sanitation standards and education of vendors on safe handling practices have been shown to reduce microbial contamination and improve consumer safety (Gil *et al.* 2015; Uyttendaele *et al.* 2015). All presumptive *Listeria* isolates obtained from lettuce samples were first subjected to cultural, morphological, and biochemical characterization as presented in Table 2. The isolates exhibited growth patterns and colony morphologies consistent with members of the genus *Listeria*. Microscopic examination revealed Gram-positive, short rod-shaped cells, while biochemical reactions further supported their presumptive identification as *Listeria* species. Molecular confirmation was achieved using PCR-based methods as shown in Plate 1. Agarose gel electrophoresis of the amplified products revealed clear and distinct DNA bands at the expected molecular sizes corresponding to *Listeria*-specific target genes. All analysed isolates produced sharp, well-defined bands, indicating successful amplification and good DNA quality. No non-specific bands or primer-dimer formations were observed, and negative controls showed no amplification, confirming the specificity of the PCR assay. The presence of uniform band sizes among isolates tentatively identified as the same species further supported their genetic relatedness. Sequence analysis of the PCR amplicons is presented in Table 3 and confirmed the identities of the isolates as *Listeria monocytogenes*, *Listeria innocua*, and *Listeria ivanovii*. The sequences showed very high similarity to reference strains deposited in the GenBank database, with percentage identity values ranging from 99.35% to 100%, query coverage of 99–100%, and E-values of 0, indicating highly significant matches. *Listeria monocytogenes* was the predominant species, accounting for the majority of the isolates, followed by *Listeria innocua* and *Listeria ivanovii*.

Phylogenetic analysis was performed to determine the evolutionary relationships among the isolates and reference *Listeria* strains as presented in Figure 1. The phylogenetic tree revealed distinct clustering of the isolates according to their species designation. Isolates identified as *Listeria monocytogenes* clustered tightly together within the *L. monocytogenes* clade and showed close genetic relatedness to reference strains, suggesting a common

evolutionary origin. Similarly, *Listeria innocua* isolates formed a separate and well-supported cluster, clearly distinct from the pathogenic *L. monocytogenes* group. The single *Listeria ivanovii* isolate clustered independently within the *L. ivanovii* lineage, confirming its species-level distinction. The isolation and molecular confirmation of *Listeria* species from lettuce samples in the present study provide further evidence that fresh vegetables serve as important vehicles for *Listeria* contamination and potential transmission to humans. Several studies have demonstrated that leafy vegetables are particularly susceptible to contamination due to their large surface area, close contact with soil irrigation water and extensive handling during harvesting and marketing (Sant Ana *et al.*, 2017; Olaimat *et al.*, 2018). The recovery of *Listeria monocytogenes* alongside other *Listeria* species in this study aligns with reports indicating that multiple species frequently coexist in fresh produce environments. The predominance of *Listeria monocytogenes* among the isolates is consistent with its ecological fitness and ability to persist under adverse environmental conditions. This species is well known for its capacity to survive refrigeration acidic conditions and osmotic stress which facilitates its persistence on raw vegetables during storage and distribution (Carpentier and Cerf, 2011; Ferreira *et al.*, 2014). Similar findings have been reported in studies investigating fresh and minimally processed vegetables where *L. monocytogenes* was the most frequently detected *Listeria* species (Strawn *et al.*, 2013; Oliveira *et al.*, 2018). Phylogenetic analysis in the present study revealed clear clustering of isolates according to species, which confirms the accuracy of molecular identification and suggests close genetic relatedness among isolates of the same species. Phylogenetic grouping of *Listeria* isolates has been widely used to infer evolutionary relationships and potential sources of contamination in food safety studies (Ragon *et al.*, 2008; Maury *et al.*, 2016). The tight clustering of *L. monocytogenes* isolates observed here may indicate circulation of closely related strains within the produce handling environment or common contamination routes along the supply chain. The detection of *Listeria innocua* and *Listeria ivanovii* alongside *L. monocytogenes* highlights the ecological diversity of *Listeria* species associated with fresh produce. Although *L. innocua* is generally considered non-pathogenic, its presence is epidemiologically significant because it often shares environmental niches with *L. monocytogenes* and is commonly used as an indicator organism for *Listeria* contamination (Orsi and Wiedmann, 2016). *Listeria ivanovii*, while primarily associated with animal infections, has occasionally been implicated in human disease, and

its isolation from food matrices underscores the importance of monitoring a broad range of *Listeria* species in food safety assessments (Guillet *et al.*, 2010). The public health significance of detecting *L. monocytogenes* in lettuce is substantial due to the association of this pathogen with severe foodborne illness and outbreaks linked to fresh produce consumption. Several outbreaks of listeriosis have been traced to contaminated leafy greens and ready-to-eat salads, resulting in hospitalisation and mortality, particularly among pregnant women, the elderly, and immunocompromised individuals (McCollum *et al.*, 2013; Self *et al.*, 2019). The ability of *L. monocytogenes* to form biofilms on plant surfaces and food contact materials further enhances its persistence and complicates control measures (Gandhi and Chikindas, 2007). The use of PCR based molecular techniques and sequencing in this study proved essential for accurate species identification and confirmation of *Listeria* isolates. Conventional cultural and biochemical methods alone may be insufficient to differentiate closely related *Listeria* species whereas molecular tools provide higher specificity sensitivity and epidemiological resolution (Jadhav *et al.*, 2012; Law *et al.*, 2015). The distinct PCR banding patterns and species specific phylogenetic clustering observed in this study therefore reinforce the value of molecular diagnostics in routine food safety surveillance.

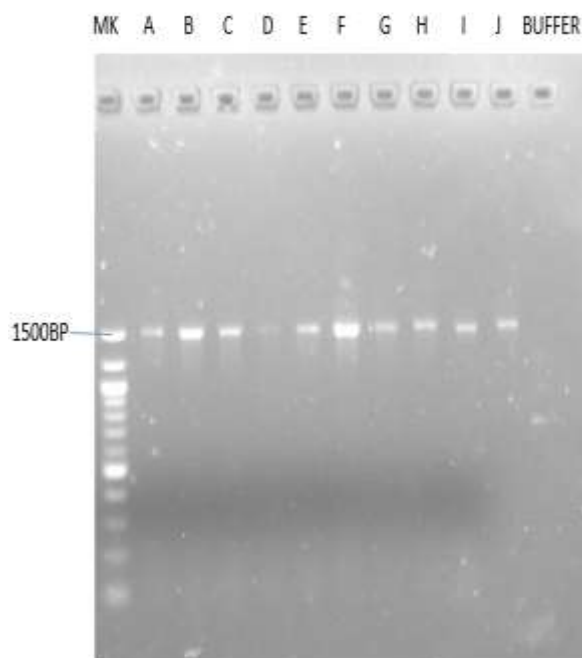


Plate 1: agarose gel electrophoresis of the PCR products of 16S rRNA gene amplified from *Listeria* isolates. (Band size approximately 1500bp). Mk = molecular size marker. A-J represent different isolates of *Listeria* as presented in Table 3

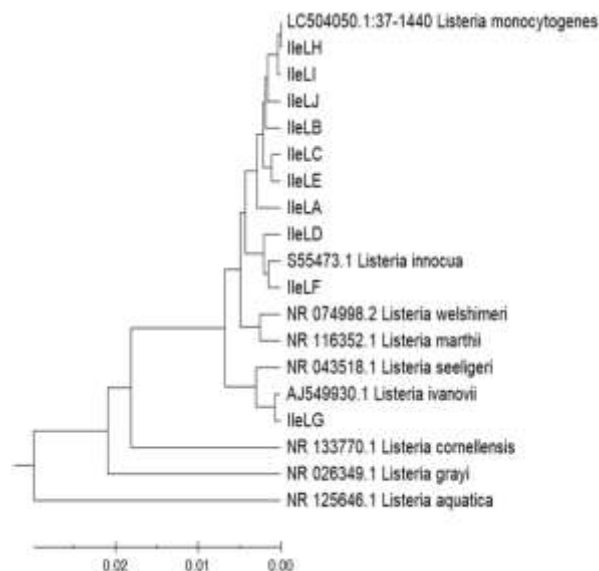


Fig. 1: phylogenetic tree analysis of *Listeria* isolates

Table 4 outlines the distribution of *Listeria* species isolated from lettuce samples obtained from the five markets. Out of the 118 lettuce samples analyzed, 21 (17.80%) were positive for various *Listeria* species.

Ikotun and Mile 12 showed relatively higher contamination rates, at 16.67% and 21.74% respectively, while Sabo-Yaba recorded the lowest prevalence at 12.50%. Multiple *Listeria* species were detected, including *Listeria monocytogenes*, *L.*

innocua, and *L. ivanovii*. Notably, *L. monocytogenes*, the primary pathogenic species associated with listeriosis, was detected in several samples across different markets, highlighting a significant public health concern. The distribution of *Listeria* species across lettuce carrot and cabbage samples provides important insight into the epidemiology of *Listeria* contamination in fresh vegetables sold in open markets and reinforces concerns regarding the microbiological safety of raw produce. The detection of *Listeria* species in all three vegetables across multiple markets confirms that contamination is widespread and not limited to a single produce type or location. The overall prevalence values ranging from 17.80 percent to 19.49 percent are consistent with reports from similar market based studies in low and middle income settings where fresh vegetables are frequently exposed to contaminated environments during harvesting transportation and retail display (Alegbeleye *et al.* 2018; Uyttendaele *et al.* 2015). In lettuce samples the detection of *Listeria* species in nearly one fifth of the analyzed samples highlights the vulnerability of *Lactuca sativa* to microbial contamination. Lettuce possesses a broad uneven leaf surface that readily retains moisture soil particles and microorganisms which facilitates bacterial attachment and persistence (Gil *et al.* 2015).

Table 2: cultural, morphological and biochemical characteristics of *Listeria* isolates

Parameters	A	B	C	D	E	F	G	H	I	J
Cultural										
Shape	Circular	Circular	Circular	Circular	Circular	Circular	Circular	Circular	Circular	Circular
Colour	Grey-green	Grey-green	Grey-green	Grey-green	Grey-green	Grey-green	Grey-green	Grey-green	Grey-green	Grey-green
Size	Small	Small	Small	Small	Small	Small	Small	Small	Small	Small
Elevation	Raised	Raised	Raised	Raised	Raised	Raised	Raised	Raised	Raised	Raised
Morphological										
Gram Stain	+	+	+	+	+	+	+	+	+	+
Cell Type	Rod	Rod	Rod	Rod	Rod	Rod	Rod	Rod	Rod	Rod
Spore Staining	-	-	-	+	+	-	-	-	-	-
Motility	+	+	+	+	+	+	+	+	+	+
Biochemical										
Urease	-	-	-	-	-	-	-	-	-	-
Indole	-	-	-	-	-	-	-	-	-	-
Citrate	-	-	+	-	+	+	+	+	-	-
Catalase	+	+	+	+	+	+	+	+	+	+
Nitrate reduction	-	-	-	-	-	-	-	-	-	-
Methyl-red	+	+	+	+	+	+	+	+	+	+
Esculin hydrolysis	+	+	+	+	+	+	+	+	+	+
Oxidase	-	-	-	-	-	-	-	-	-	-
H ₂ S	-	-	-	-	-	-	-	-	-	-
Oxidative fermentation test										
Glucose	+	+	+	+	+	+	+	+	+	+
Sucrose	-	+	+	+	+	+	+	+	+	+
Lactose	-	+	+	+	+	+	+	-	+	+
Rhamnose	+	+	+	+	+	+	+	+	+	+
Xylose	+	-	-	-	-	-	-	+	-	-
Probable isolates	<i>Listeria</i> sp.	<i>Listeria</i> sp.	<i>Listeria</i> sp.	<i>Listeria</i> sp.	<i>Listeria</i> sp.	<i>Listeria</i> sp.	<i>Listeria</i> sp.	<i>Listeria</i> sp.	<i>Listeria</i> sp.	<i>Listeria</i> sp.

Table 3: identity of *Listeria* species

Sample Id	Scientific Name	Max Score	Total Score	Query Cover	E value	Per. Ident	Accession
A	<i>Listeria monocytogenes</i>	2518	15080	100%	0	99.64%	PX070064
B	<i>Listeria monocytogenes</i>	2531	15158	99%	0	99.93%	PX070065
C	<i>Listeria monocytogenes</i>	2529	15147	100%	0	99.85%	PX070066
D	<i>Listeria innocua</i>	2521	2521	100%	0	99.71%	PX070067
E	<i>Listeria monocytogenes</i>	2536	15158	100%	0	99.93%	PX070068
F	<i>Listeria innocua</i>	2532	2532	100%	0	99.93%	PX070069
G	<i>Listeria ivanovii</i>	2512	2512	100%	0	99.35%	PX070070
H	<i>Listeria monocytogenes</i>	2549	15269	100%	0	100.00%	PX070071
I	<i>Listeria monocytogenes</i>	2543	15236	100%	0	99.93%	PX070072
J	<i>Listeria monocytogenes</i>	2532	15169	99%	0	99.93%	PX070073

Table 4: distribution of *Listeria* in *Lactuca sativa* obtained from different markets

<i>Listeria</i>	Markets					
	Ikotun (24)	Ajangbadi (24)	Mile12 (23)	SaboYaba (24)	Idumota (23)	Total (118)
<i>Listeria monocytogenes</i>	1(4.17)	1 (4.17)	0(0.0)	1(4.17)	0(0.0)	3(2.54)
<i>Listeria monocytogenes</i>	0(0.0)	1(4.17)	1(4.35)	0(0.0)	0(0.0)	2(1.69)
<i>Listeria monocytogenes</i>	0(0.0)	0(0.0)	1(4.35)	0(0.0)	1(4.35)	2(1.69)
<i>Listeria monocytogenes</i>	0(0.0)	1(4.17)	0(0.0)	1(4.17)	0(0.0)	2(1.69)
<i>Listeria monocytogenes</i>	1(4.17)	0(0.0)	0(0.0)	0(0.0)	1(4.35)	2(1.69)
<i>Listeria monocytogenes</i>	0(0.0)	0(0.0)	0(0.0)	0(0.0)	1(4.35)	1(0.85)
<i>Listeria monocytogenes</i>	1(4.17)	0(0.0)	1(4.35)	0(0.0)	1(4.35)	3(2.54)
<i>Listeria innocua</i>	0(0.0)	0(0.0)	1(4.35)	1(4.17)	0(0.0)	2(1.69)
<i>Listeria innocua</i>	1(4.17)	0(0.0)	0(0.0)	0(0.0)	1(4.35)	2(1.69)
<i>Listeria ivanovii</i>	0(0.0)	1(4.17)	1(4.35)	0(0.0)	0(0.0)	2(1.69)
Total	4(16.67)	44(16.67)	5 (21.74)	3 (12.50)	55 (21.74)	21(17.80)

The relatively higher prevalence observed in Ikotun and Mile 12 compared with Sabo Yaba suggests that factors such as water quality used for washing frequency of handling and sanitation of display surfaces may vary substantially between markets. Several studies have demonstrated that the use of contaminated irrigation or wash water is a major route through which *Listeria* is introduced into leafy vegetables and subsequently disseminated at retail points (Uyttendaele *et al.* 2015).

The isolation of *Listeria monocytogenes* from lettuce is particularly concerning because this vegetable is commonly consumed raw and therefore bypasses any thermal processing that could reduce microbial load. Previous outbreaks of listeriosis have been directly linked to the consumption of contaminated leafy greens underscoring the public health significance of these findings (Carstens *et al.* 2019).

Table 5 shows the occurrence of *Listeria* species in carrot samples collected from the five markets. Carrots showed an overall *Listeria* prevalence of 17.80% (21 out of 118 samples), closely matching the pattern observed in lettuce.

Sabo-Yaba recorded the highest contamination rate at 21.74%, followed by Idumota at 20.83%. Ikotun, Ajangbadi, and Mile 12 exhibited moderate but comparable prevalence values ranging between

12.50% and 17.39%. Similar to lettuce, several isolates of *L. monocytogenes* were detected in carrot samples. The presence of *L. innocua* and *L. ivanovii* further reflects a diverse *Listeria* population contaminating produce in these markets. Carrot samples exhibited a prevalence pattern similar to that observed in lettuce which indicates that root vegetables are also important reservoirs for *Listeria* species when hygienic controls are inadequate.

Although carrots are often peeled or washed before consumption, contamination can persist on their surfaces and within surface irregularities particularly when they are grown in contaminated soil or washed with untreated water (Beuchat 2021). The highest prevalence recorded in Sabo Yaba suggests intense environmental exposure likely due to poor sanitation and handling practices within the market.

The detection of *Listeria monocytogenes* in carrots corroborates earlier findings that root vegetables can serve as vehicles for pathogenic *Listeria* especially when consumed raw or lightly processed as in salads or juices (Oliveira *et al.* 2015).

The presence of *Listeria innocua* and *Listeria ivanovii* further reflects environmental contamination and indicates that the market ecosystem supports the survival of multiple *Listeria* species.

Table 5: distribution of *Listeria* in *Daucus carota* obtained from different markets

<i>Listeria</i>	Market					Total
	Ikotun (24)	Ajangbadi (23)	Mile12 (24)	SaboYaba (23)	Idumota (24)	118
<i>Listeria monocytogenes</i>	1(4.17)	0(0.0)	1(4.17)	0(0.0)	0(0.0)	2(1.69)
<i>Listeria monocytogenes</i>	0(0.0)	1(4.35)	0(0.0)	1(4.35)	0(0.0)	2(1.69)
<i>Listeria monocytogenes</i>	0(0.0)	0(0.0)	1(4.17)	0(0.0)	1(4.17)	2(1.69)
<i>Listeria monocytogenes</i>	0(0.0)	0(0.0)	1(4.17)	1(4.35)	0(0.0)	2(1.69)
<i>Listeria monocytogenes</i>	0(0.0)	0(0.0)	0(0.0)	1(4.35)	0(0.0)	1(0.85)
<i>Listeria monocytogenes</i>	1(4.17)	1(4.35)	0(0.0)	0(0.0)	1(4.17)	3(2.54)
<i>Listeria monocytogenes</i>	0(0.0)	1(4.35)	0(0.0)	0(0.0)	1(4.17)	2(1.69)
<i>Listeria innocua</i>	0(0.0)	1(4.35)	0(0.0)	1(4.35)	0(0.0)	2(1.69)
<i>Listeria innocua</i>	1(4.17)	0(0.0)	1(4.17)	0(0.0)	1(4.17)	3(2.54)
<i>Listeria ivanovii</i>	0(0.0)	0(0.0)	0(0.0)	1(4.35)	1(4.17)	2(1.69)
	3(12.50)	4 (17.39)	4(16.67)	5(21.74)	5(20.83)	21 (17.80)

Table 6: distribution of *Listeria* in *Brassica oleracea* obtained from different markets

<i>Listeria</i>	Markets					Total
	Ikotun (23)	Ajangbadi (24)	Mile12 (24)	SaboYaba (24)	Idumota (23)	118
<i>Listeria monocytogenes</i>	2	0(0.0)	0(0.0)	1(4.17)	0(0.0)	3(2.54)
<i>Listeria monocytogenes</i>	1(4.35)	1(4.17)	1(4.17)	0(0.0)	0(0.0)	3(2.54)
<i>Listeria monocytogenes</i>	1(4.35)	0(0.0)	0(0.0)	1(4.17)	0(0.0)	2(1.69)
<i>Listeria monocytogenes</i>	0(0.0)	1(4.17)	0(0.0)	0(0.0)	1(4.35)	2(1.69)
<i>Listeria monocytogenes</i>	1(4.35)	1(4.17)	0(0.0)	0(0.0)	1(4.35)	3(2.54)
<i>Listeria monocytogenes</i>	1(4.35)	0(0.0)	1(4.17)	0(0.0)	0(0.0)	2(1.69)
<i>Listeria monocytogenes</i>	0(0.0)	1(4.17)	0(0.0)	1(4.17)	0(0.0)	2(1.69)
<i>Listeria innocua</i>	0(0.0)	0(0.0)	1(4.17)	0(0.0)	1(4.35)	2(1.69)
<i>Listeria innocua</i>	0(0.0)	0(0.0)	1(4.17)	0(0.0)	0(0.0)	1(0.85)
<i>Listeria ivanovii</i>	1(4.35)	0(0.0)	1(4.17)	1(4.17)	0(0.0)	3(2.54)
Total	7 (30.43)	4(16.67)	5 (20.83)	4(16.67)	3 (13.04)	23 (19.49)

Table 6 details the distribution of *Listeria* species in cabbage samples from the five markets. Cabbage recorded the highest overall prevalence of *Listeria* contamination among the three vegetables, with 23 positive samples out of 118 (19.49%). Ikotun demonstrated the highest contamination level, with 30.43% of its cabbage samples testing positive, representing the single highest value across all vegetables and markets. Conversely, Idumota had the lowest contamination rate at 13.04%. As with the other vegetables, *L. monocytogenes* was frequently isolated, alongside *L. innocua* and *L. ivanovii*. Cabbage demonstrated the highest overall prevalence of *Listeria* contamination among the three vegetables which may be attributed to its structural characteristics. The compact layered leaves of *Brassica oleracea* can trap dirt, organic matter and microorganisms and can protect bacteria from removal during routine washing (Gil *et al.* 2015). The exceptionally high prevalence recorded in Ikotun suggests substantial contamination pressure possibly arising from prolonged storage, unsanitary display conditions and repeated handling. Studies have shown that *Listeria* species can persist in the folds of cabbage leaves for extended periods and may even multiply under favorable moisture conditions making this vegetable a particularly high risk commodity (Alegbeleye *et al.* 2018). As observed in lettuce and carrots the frequent isolation of *Listeria monocytogenes* in cabbage samples raises significant

food safety concerns due to the severity of listeriosis and its high mortality rate among vulnerable populations.

Across all vegetables the coexistence of pathogenic and non-pathogenic *Listeria* species suggests continuous environmental seeding of produce with bacteria from soil water and handling surfaces. The presence of *Listeria innocua* is often regarded as an indicator of environmental contamination and poor hygiene and its detection alongside *Listeria monocytogenes* supports the notion of shared contamination routes (Uyttendaele *et al.* 2015). The variability in prevalence among markets further reinforces the role of market specific hygiene practices rather than vegetable type alone in shaping contamination patterns. Markets with inadequate waste disposal, limited access to clean water, and poor infrastructure are more likely to support the survival and transmission of *Listeria* species.

Conclusion: This study demonstrates that fresh vegetables sold in open markets in Lagos State harbour a considerable burden of *Listeria* species, including the pathogenic *Listeria monocytogenes*. The predominance of *L. monocytogenes* raises significant food safety and public health concerns, particularly given the common consumption of these vegetables in raw or minimally processed forms. The coexistence of pathogenic and non-pathogenic

Listeria species further suggests continuous environmental contamination and inadequate hygienic controls along the vegetable supply chain. The findings emphasize the urgent need for improved sanitation practices at points of sale, enforcement of food safety regulations, and education of vegetable handlers on hygienic handling practices. Routine microbiological monitoring incorporating molecular diagnostics is strongly recommended to mitigate the risk of listeriosis associated with fresh vegetable consumption and to protect vulnerable populations.

DECLARATION

Ethical Approval: This study did not involve human participants or animal experimentation and therefore did not require formal ethical approval. All procedures involving data handling and analysis were conducted responsibly and in accordance with standard academic and institutional guidelines.

Disclaimer (Artificial Intelligence): Author(s) hereby declare that a generative AI technology was used to arrange references. Detail of the AI usage is: ChatGPT

Declaration of Conflict of Interest: The authors declare that no known conflict of interest or personal relationships that could have influence the work reported in this paper

Declaration of Data Availability Statement: Data are available upon reasonable request from the corresponding author.

REFERENCES

- Abakari, G; Cobbina, SJ; Yeleliere, E. (2018). Microbial quality of ready-to-eat vegetables sold in selected markets in Ghana. *Apl. Water Sci.* 8(3), 1–8. <https://doi.org/10.1007/s13201-018-0719-2>
- Ajayeoba, TA; Atanda, OO; Obadina, AO; Bankole, MO; Adelowo, OO; Adeyemi, IA (2015). Prevalence of *Listeria monocytogenes* in ready-to-eat vegetables sold in South Western Nigeria. *J. Food Saf.*, 35(2), 144–152. <https://doi.org/10.1111/jfs.12162>
- Alegbeleye, OO.; Singleton, I; Sant'Ana, AS (2018). Sources and contamination routes of microbial pathogens to fresh produce during field cultivation. *Food Microbiol.*, 73, 177–208. <https://doi.org/10.1016/j.fm.2018.01.003>
- Alegbeleye, OO; Singleton, I; Sant'Ana, AS (2018). Sources and contamination routes of microbial pathogens to fresh produce during field cultivation. *Food Microbiol.*, 73, 177–208. <https://doi.org/10.1016/j.fm.2018.01.003>
- Amajoud, N; Leclercq, A; Soriano, JM; Bracq Dieye, H; El Maadoudi, M; Senhaji, NS; Lecuit, M (2018). Prevalence of *Listeria monocytogenes* in food products in Morocco and characterization of isolates by serotyping and pulsed field gel electrophoresis. *Food Control*, 84, 1–8. <https://doi.org/10.1016/j.foodcont.2017.07.018>
- Becker, B; Henningsen, A; Scholderer, J; Teuber, R (2019). Consumer preferences for ready-to-eat foods and food safety concerns. *Food Control*, 96, 72–79. <https://doi.org/10.1016/j.foodcont.2018.08.019>
- Beuchat, LR (2021). Surface decontamination of fruits and vegetables eaten raw. *Food Saf.*, 9(1), 1–16. <https://doi.org/10.14252/foodsafetyfscj.D-20-00008>
- Bolocan, AS; Nicolau, AI; Álvarez Ordóñez, A; Borda, D; Oniciuc, EA; Stessl, B; Gurgu, L; Wagner, M; Jordan, K (2016). Dynamics of *Listeria monocytogenes* colonisation in food processing environments. *Food Control*, 66, 75–85. <https://doi.org/10.1016/j.foodcont.2016.01.012>
- Buchanan, RL; Gorris, LGM; Hayman, MM; Jackson, TC; Whiting, RC (2017). A review of *Listeria monocytogenes* an update on outbreaks virulence dose response ecology and risk assessments. *Food Control*, 75, 1–13. <https://doi.org/10.1016/j.foodcont.2016.12.016>
- Bucur, FI; Grigore Gurdas, M; Crauwels, P; Riedel, CU; Nicolau, AI; Wagner, M (2018). Resistance of *Listeria monocytogenes* to stress conditions encountered in food and food processing environments. *Front. Microbiol.*, 9, 2700. <https://doi.org/10.3389/fmicb.2018.02700>
- Bustamante, F; Maury, MM; Aubry, C; Pichon, M; Bracq Dieye, H; Dominguez, M; Vignaud, ML; Lecuit, M (2020). *Listeria monocytogenes* in Chile a genomic epidemiology study of human clinical isolates. *Clin. Microbiol. Infect.*, 26(9), 1136–1143. <https://doi.org/10.1016/j.cmi.2019.12.017>
- Carpentier, B; Cerf, O (2011). Review persistence of *Listeria monocytogenes* in food industry equipment and premises. *Int. J. Food Microbiol.*,

- 145(1), 1–8.
<https://doi.org/10.1016/j.ijfoodmicro.2011.01.005>
- Carstens, CK; Salazar, JK; Darkoh, C (2019). Multistate outbreaks of foodborne illness in the United States associated with fresh produce from 2010 to 2017. *Front. Microbiol.*, 10, 2667.
<https://doi.org/10.3389/fmicb.2019.02667>
- European Centre for Disease Prevention and Control (2018). Listeriosis annual epidemiological report for 2016. ECDC Stockholm.
<https://www.ecdc.europa.eu/en/publications-data/listeriosis-annual-epidemiological-report-2016>
- Falavina dos Reis, CM; Barbieri, E; Oliveira, AM; Wiedmann, M (2011). Molecular characterization and virulence gene profiles of *Listeria monocytogenes* isolates from food products. *Foodborne Pathog. Dis.*, 8(6), 699–707.
<https://doi.org/10.1089/fpd.2010.0785>
- Ferreira, V; Wiedmann, M; Teixeira, P; Stasiewicz, MJ (2014). *Listeria monocytogenes* persistence in food associated environments a critical review. *Crit. Rev. Food Sci. Nutr.*, 54(7), 1–17.
<https://doi.org/10.1080/10408398.2012.706667>
- Gandhi, M; Chikindas, ML (2007). *Listeria* a foodborne pathogen that knows how to survive. *Int. J. Food Microbiol.*, 113(1), 1–15.
<https://doi.org/10.1016/j.ijfoodmicro.2006.07.008>
- Gil, MI; Selma, MV; Suslow, T; Jacxsens, L; Uyttendaele, M; Allende, A (2015). Pre and postharvest preventive measures and intervention strategies to control microbial food safety hazards of fresh leafy vegetables. *Crit. Rev. Food Sci. Nutr.*, 55(4), 453–468.
<https://doi.org/10.1080/10408398.2012.657808>
- Guillet, C; Join Lambert, O; Monnier, AL; Leclercq, A; Mechai, F; Mamzer Brugère, C; Lecuit, M (2010). Human listeriosis caused by *Listeria ivanovii*. *Emerg. Infect. Dis.*, 16(1), 136–138.
<https://doi.org/10.3201/eid1601.091155>
- Ieren, II (2013). Occurrence of *Listeria monocytogenes* in salad vegetables sold in Zaria Nigeria. *Afr. J. Microbiol. Res.*, 7(18), 1912–1917. <https://doi.org/10.5897/AJMR2013.5759>
- Jadhav, S; Bhawe, M; Palombo, EA (2012). Methods used for the detection and subtyping of *Listeria monocytogenes*. *J. Microbiol. Methods*, 88(3), 327–341.
<https://doi.org/10.1016/j.mimet.2012.01.002>
- Kurpas, M; Wieczorek, K; Osek, J (2018). Ready-to-eat meat products as a source of *Listeria monocytogenes*. *J. Vet. Res.*, 62(1), 49–55.
<https://doi.org/10.2478/jvetres-2018-0008>
- Kwon, HJ; Chen, Z; Glover, JNM (2020). Structural and functional insights into virulence regulation in *Listeria monocytogenes*. *Trends Microbiol.*, 28(5), 373–386.
<https://doi.org/10.1016/j.tim.2019.11.006>
- Law, JWF; Ab Mutalib, NS; Chan, KG; Lee, LH (2015). Rapid methods for the detection of foodborne bacterial pathogens principles applications advantages and limitations. *Front. Microbiol.*, 5, 770.
<https://doi.org/10.3389/fmicb.2014.00770>
- Lee, S; Ward, TJ; Graves, LM; Wolf, LA; Sperry, K; Siletzky, RM; Kathariou, S (2018). Atypical genetic features of *Listeria monocytogenes* serotype 4b strains associated with clinical cases. *Appl. Environ. Microbiol.*, 84(4), e02011-17.
<https://doi.org/10.1128/AEM.02011-17>
- Louwen, R; Staals, RHJ; Endtz, HP; van Baarlen, P; van der Oost, J (2014). The role of CRISPR Cas systems in virulence of pathogenic bacteria. *Microbiol. Mol. Biol. Rev.*, 78(1), 74–88.
<https://doi.org/10.1128/MMBR.00039-13>
- Maertens de Noordhout, C; Devleeschauwer, B; Angulo, FJ; Verbeke, G; Haagsma, J; Kirk, M; Havelaar, A; Speybroeck, N (2017). The global burden of listeriosis a systematic review and meta-analysis. *Lancet Infect. Dis.*, 14(11), 1073–1082.
[https://doi.org/10.1016/S1473-3099\(14\)70870-9](https://doi.org/10.1016/S1473-3099(14)70870-9)
- Maury, MM; Tsai, YH; Charlier, C; Touchon, M; Chenal Francisque, V; Leclercq, A; Lecuit, M (2016). Uncovering *Listeria monocytogenes* hypervirulence by harnessing its biodiversity. *Nat. Genet.*, 48(3), 308–313.
<https://doi.org/10.1038/ng.3501>
- Maury, MM; Tsai, YH; Charlier, C; Touchon, M; Chenal Francisque, V; Leclercq, A; Criscuolo, A; Gaultier, C; Roussel, S; Brisabois, A; Lecuit, M; Brisse, S (2016). Uncovering *Listeria monocytogenes* hypervirulence by harnessing its biodiversity. *Nat. Genet.*, 48(3), 308–313.
<https://doi.org/10.1038/ng.3501>

- McCollum, JT; Cronquist, AB; Silk, BJ; Jackson, KA; O'Connor, KA; Cosgrove, S; Mahon, BE (2013). Multistate outbreak of listeriosis associated with cantaloupe. *N. Engl. J. Med.*, 369(10), 944–953. <https://doi.org/10.1056/NEJMoa1215837>
- Monteiro, CA (2010). The big issue is ultra-processing. *World Nutr.*, 1(6), 237–269. <https://www.wphna.org/htdocs/downloads/November2010/10-11%20WN%20Comm%20Food%20processing.pdf>
- Mumuney, K. T., Abdullateef, A., & Bala, A. (2021). Occurrences and antibiogram pattern of *Listeria monocytogenes* in vegetables sold within Sokoto metropolis, Nigeria. *Journal of Advances in Microbiology*, 21(4), 83–90. <https://doi.org/10.9734/jamb/2021/v21i430344>
- Olaimat, AN; Al Hadidi, S; Al Homsy, R; Holley, RA (2018). Occurrence of *Listeria monocytogenes* in fresh produce risk factors and mitigation strategies. *Food Control*, 85, 8–19. <https://doi.org/10.1016/j.foodcont.2017.09.019>
- Olaimat, AN; Al Hadidi, S; Al Homsy, R; Holley, RA (2018). Occurrence of *Listeria monocytogenes* in fresh produce risk factors and mitigation strategies. *Food Control*, 85, 8–19. <https://doi.org/10.1016/j.foodcont.2017.09.019>
- Oliveira, M; Abadias, M; Usall, J; Torres, R; Teixidó, N; Viñas, I (2015). Application of modified atmosphere packaging as a safety approach to fresh cut fruits and vegetables. *Trends Food Sci. Technol.*, 46(1), 13–26. <https://doi.org/10.1016/j.tifs.2015.07.017>
- Oliveira, M; Abadias, M; Usall, J; Torres, R; Teixidó, N; Viñas, I (2015). Application of modified atmosphere packaging as a safety approach to fresh cut fruits and vegetables. *Trends Food Sci. Technol.*, 46(1), 13–26. <https://doi.org/10.1016/j.tifs.2015.07.017>
- Oliveira, M; Viñas, I; Colàs, P; Anguera, M; Abadias, M (2018). Effectiveness of sanitizers in reducing *Listeria monocytogenes* on fresh cut lettuce. *Food Microbiol.*, 70, 71–78. <https://doi.org/10.1016/j.fm.2017.09.012>
- Orsi, RH; Wiedmann, M (2016). Characteristics and distribution of *Listeria* species in nature. *Appl. Microbiol. Biotechnol.*, 100(12), 5273–5287. <https://doi.org/10.1007/s00253-016-7451-2>
- Orsi, RH; den Bakker, HC; Wiedmann, M (2011). *Listeria monocytogenes* lineages population structure evolution and virulence. *Annu. Rev. Microbiol.*, 65, 287–310. <https://doi.org/10.1146/annurev-micro-090110-102732>
- Pouillot, R; Klontz, KC; Chen, Y; Burall, LS; Macarasin, D; Doyle, M; Bally, KM; Strain, E; Datta, AR; Hammack, TS; Van Doren, JM (2016). Infectious dose of *Listeria monocytogenes* in outbreak linked ready-to-eat foods. *Risk Anal.*, 36(8), 1445–1456. <https://doi.org/10.1111/risa.12569>
- Ragon, M; Wirth, T; Hollandt, F; Lavenir, R; Lecuit, M; Le Monnier, A; Brisse, S (2008). A new perspective on *Listeria monocytogenes* evolution. *PLoS Pathog.*, 4(9), e1000146. <https://doi.org/10.1371/journal.ppat.1000146>
- Ryser, ET (2021). Public health concerns. In Ryser, ET; Marth, EH (Eds.), *Listeria listeriosis and food safety* (4th ed., pp. 3–38). CRC Press Taylor and Francis Group.
- Sant Ana, AS; Barbosa, MS; Destro, MT; Landgraf, M; Franco, BDGM (2017). Growth potential of *Listeria monocytogenes* in ready-to-eat leafy vegetables. *Food Control*, 78, 155–161. <https://doi.org/10.1016/j.foodcont.2017.02.038>
- Schlech, WF (2019). Epidemiology and clinical manifestations of *Listeria monocytogenes* infection. *Microbiol. Spectr.*, 7(3), 1–14. <https://doi.org/10.1128/microbiolspec.GPP3-0014-2018>
- Self, JL; Conrad, A; Stroika, S; Jackson, A; Burnworth, L; Beal, J; Silk, BJ (2019). Multistate outbreak of listeriosis associated with packaged leafy green salads. *Clin. Infect. Dis.*, 68(4), 635–641. <https://doi.org/10.1093/cid/ciy600>
- Townsend, DE; Cary, JW; Whiting, RC (2021). Biofilm formation by *Listeria monocytogenes* on plant associated surfaces and implications for produce safety. *Food Microbiol.*, 95, 103694. <https://doi.org/10.1016/j.fm.2020.103694>
- Truong, HN; Garmyn, D; Gal, L; Fournier, C; Sevellec, Y; Guillier, L (2021). Persistence of *Listeria monocytogenes* in soil and association

- with plant roots. *Front. Microbiol.*, 12, 635686. <https://doi.org/10.3389/fmicb.2021.635686>
- Uyttendaele, M; Jaykus, LA; Amoah, P; Chiodini, A; Cunliffe, D; Jacxsens, L; Rose, J (2015). Microbial hazards in irrigation water standards guidelines and mitigation strategies. *Food Microbiol.*, 43, 193–203. <https://doi.org/10.1016/j.fm.2014.07.007>
- Uyttendaele, M; Jaykus, LA; Amoah, P; Chiodini, A; Cunliffe, D; Jacxsens, L; Rose, J (2015). Microbial hazards in irrigation water standards guidelines and mitigation strategies. *Food Microbiol.*, 43, 193–203. <https://doi.org/10.1016/j.fm.2014.07.007>
- Wilson, K (2001). Preparation of genomic DNA from bacteria. In Ausubel, FM et al. (Eds.), *Curr. Protoc. Mol. Biol.* (pp. 2.4.1–2.4.5). John Wiley and Sons.