DERLEME REVIEW

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Overview of *Listeria Monocytogenes* as a Foodborne Pathogen: Traditional Review

Gıda Kaynaklı Bir Patojen Olarak *Listeria Monocytogenes'e* Genel Bakış: Geleneksel Derleme

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ABSTRACT There are more than 200 diseases caused by microorganisms such as bacteria, viruses and parasites, which are taken with foods and cause 600 million people to fall ill and about 420 thousand of them lose their lives. These factors arise due to the failure of the heat treatment of the ready to eat foods. One of them, L. *monocytogenes*, known for many years, is a biofilm-capable psychrophic bacteria with a wide range of physiological response mechanisms that can adapt to different stress conditions. It draws attention with its high mortality rate compared to many other bacteria, especially in pregnant women, the elderly and children. Despite this, it still has not created sufficient awareness among consumers. In addition, it plays an important role in increasing antibiotic resistance in antibiotics used in the poultry industry. This review aims to give information about general features of L. *monocytogenes*, virulence factors, biofilm resistance and inactivation, prevalence, antibiotic resistance and disease status in humans.

Keywords: Biofilm; listeriosis; prevalance; virulence

ÖZET Gıdalar ile alınarak, her yıl 600 milyon kişinin hastalanmasına ve bu kişilerin yaklaşık 420 bininin ölümüne neden olan bakteri, virüs ve parazit gibi mikroorganizmalardan kaynaklı 200'den fazla hastalık etkeni bulunmaktadır. Özellikle tüketime hazır gıdaların ısı işlemi uygulanmamasına bağlı olarak bu etkenler ortaya çıkmaktadır. Bunlardan biri olan ve yıllardan beri bilinen Listeria *monocytogenes,* farklı stres koşullarına karşı adaptasyon sağlayabilen çok çeşitli fizyolojik yanıt mekanizmalarına sahip, biyofilm oluşturabilen psikrotrof özellikte bir bakteridir. Diğer birçok bakteriye göre özellikle hamile kadınlar, yaşlılar ve çocuklar üzerindeki ölüm oranının yüksekliğiyle dikkat çekmektedir. Buna rağmen hâlen tüketiciler arasında yeterli farkındalığı sağlayamamıştır. Bunun yanında kanatlı sektöründe kullanılan antibiyotiklerde, antibiyotik direncinin artmasında önemli rol oynamaktadır. Bu derlemede, L. *monocytogenes*'in genel özellikleri, virulens faktörleri, biyofilm direnci ve inaktivasyonu, prevalansı, antibiyotik direnci ve insanlarda oluşturduğu hastalık durumuyla ilgili bilgiler verilmesi amaçlanmıştır.

Anahtar Kelimeler: Biyofilm; listeriosis; prevalans; virulens

Nutrition and food safety are integral parts of a whole. Unsafe foods particularly affect infants, young children and the elderly. There are more than 200 diseases known to be transmitted by food. Every year, as a result of contaminated food, an estimated 600 million people get sick and 420 thousand of them lose thier lives.[1] There are various agents that cause foodborne illnesses such as bacteria, viruses and parasites. Among them, bacterial agents are more common. Consumption of animal derived foods such as dairy or meat and also ready to eat products are considered as an important source of foodborne agents. Amid them, *Listeria monocytogenes* is seen as a significant cause of serious illnesses in humans and animals.[2] Although the disease manifests itself with a mild fever in most cases, it can also present as systemic listeriosis with more severe symptoms, high hospitalization and mortality rates. Generally, the frequency of listeriosis in population remains low, despite the wide distribution of the microorganism in the environment and the relatively high frequency of isolation in foods. The occurence of systemic

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listeriosis is larger in vulnerable groups such as gravid women, the elderly, and immunocompromised individuals.[3] Since *L. monocytogenes* has a vast diversity of physiological response mechanisms that adapt to different stress conditions, it can survive and therefore reproduce in different environmental conditions. Cooling, low pH and high salt concentration cause the pathogen to overcome food protection and safety barriers and become a potential threat for human health.[4] *L. monocytogenes* isolated from food and processing facilities can develop resistance to many antibiotics. In addition, the outgrowth and spread of resistance amid foodborne hazards to disinfectants used by the food industry is also a concern. Regardless of the used disinfectant, standard procedures such as controlling this microorganism, cleaning, degreasing, and disinfection must be strictly followed.[5] *Listeria* in food production facilities requires a constant focus on risk management. Hence, a better understanding of the microorganism's features, environmental influence, and the interactions of virulence factors with host susceptibility is needed by establishing better control measures to reduce the incidence of listeriosis.[3]

GENERAL CHARACTERISTICS OF *L. MONOCYTOGENES*

L. monocytogenes is a gram positive bacterium whose metabolic activity can be either aerobic or facultative anaerobe. It is a pathogenic bacterium in the form of short rods or coccobacilli, 0.4-0.5 µm wide and 1-2 µm long, that does not form capsule or spore form. It is catalase positive, oxidase negative, esculin and sodium hypurate hydrolysis. When cultured in rhamnose broth, acid is formed but does not form acid in mannitol and xylose. Although the optimal breeding temperatures are in the range of 30-37 ºC, it can continue to grow between <0-45 ºC. While the movement is provided by peritric flagella between 20-25 ºC, it loses this feature at 37 ºC. It can multiply in foods with high water activity (A_{su}>0.92). This bacterium exhibits a psychrotrophic feature. It tolerates $NaNO_2$ and NaCl, it can grow at 10% salt, 200 ppm $NaNO_2$ concentrations, and maintain its vital activities at 20-30% salt concentrations. It maintains its metabolic activities in a wide pH range (4.3-9.6) that can vary according to temperature and acidity regulators. The application of 2-4 kGy irradiatiation can cause a reduction of 6-7 log of the total population.[6-8] A summary of the general characteristics of *L. monocytogenes* is given in Table 1.

SUBTYPES OF L. *MONOCYTOGENES*

L. monocytogenes is a member of the *Firmicutes* phylum, *Bacilli* class, *Bacillales* order, *Listeriaceae* family, and the genus *Listeria*.[9] *Listeria* species are classified as serotypes based on the serological reactions of somatic O and flagellar H antigens with specific antisera. *L. monocytogenes* includes serotypes 1/2a, 1/2b, 1/2c, 3a, 3b, 3c, 4a, 4b, 4c, 4d, 4e and serotype 7. More than 98% of isolates of the clinical cases of human listeriosis include serotypes 4b, 1/2a, 1/2b, 1/2c.[8] *L. monocytogenes* serotypes were first divided into three lineages as lineage I, II and III according to their genetic structure. In lineage III; it is composed of three subgroups as IIIA, IIIB and IIIC. Ward et al. also reported lineage IIIB as lineage.[10] Lineage I; 1/2b, 3b, 4b, lineage II; 1/2a, 1/2c, 3c, lineage III and IV; include serotypes 4a, 4c and atypical 4b.[11] The lineage and characteristics of *L. monocytogenes* are given in Table 2.

TABLE 1: General characteristics of L. *monocytogenes*.

Characteristics	Properties
Metabolic activity	Aerob or facultative anaerob
Cell morphology	Short rods or coccobacilli, 0.4-0.5 µm wide and 1-2 µm long, that does not form capsule or spore form
Gram staining	(+)
Catalase	(+)
Oxidase	(-)
Esculin	(+)
Sodium hippurate hydrolysis	(+)
Motility	Provided by peritrich flagella between 20-25 ºC
Water activity	>0.92
pH	4.3-9.6
Temperature	<0-45 ºC
Irradiation (2-4 kGy)	Reduction of 6-7 log of the total population
$NaNO_2$	Grow until 200 ppm
NaCl	Grow until 10%, maintain its vital activities at 20-30%

TABLE 2: Brief of *L. monocytogenes* lineages.[11]

Lineage	Initial identification	Serotypes	Genetic characteristics	Distribution
I	First described in an MLEE research by Piffaretti et al. (1989).	1/2b, 3b, 3c, 4b	Bottommost diversity amid the lineages; lowest levels of recombination among the lineages.	Extensively isolated from various sources; overrepresented among human isolates.
II	Initially exhibited in an MLEE study by Piffaretti et al. (1989).	1/2a, 1/2c, 3a	Most diverse, highest recombination levels.	Commonly isolated from diverse sources; overrepresented among food and food-related as well as natural environments.
III	Originally described using partial sequence data analyses by Rasmussen et al. (1995).	4a, 4b, 4c	Very diverse; recombination levels within those for lineage I and lineage II.	Great quantity of isolates obtained fromruminants.
IV	First described as IIIB using partial sequence data analyses by Roberts et al. (2006); first reported as lineage IV by Ward et al. (2008).	4a, 4b, 4c	Not too many isolates analyzed to date.	The greater part of isolates obtained out of ruminants.

VIRULENCE FACTORS

Virulence is identified as the capability of a bacteria to induce disease. Intracellular parasitism is caused by the expression of a series of genes that are physically collected in a 9 kb gene cluster called *Listeria* pathogenicity island 1, referred to as virulence genes due to the impact on the host. These genes encode various enzymes responsible for infection.[12,13] Some of these virulence factors are: *Listeriolysin O* (LLO), a pore-forming toxin; *ActA*, a factor responsible for the polymerization of the host actin; *InlA* and *InlB* are a group of internalin proteins that play an important role in entry into host cells; two phospholipases (*PlcA* and *PlcB*); a metalloprotease (*Mpl*); a system for sugar uptake (*UhpT*).[14] *UhpT* promotes proliferation by helping bacteria take glucose-6-phosphate into the cell.[12] These virulence factors are carried out under the direct control of *PrfA*, a transcriptional factor.[14] The virulence-related genes of *L. monocytogenes* are shown in Table 3.

TABLE 3: Characteristics of several virulence-affliated genes.[15]

Gene	Property
hly	Encodes the cholesterol-binding, pore-forming LLO required for bacterial escape from phagosomes.
plcA	Encodes the PlcA enzyme. Together with LLO, plays a role in breaking down the first phagosome vacuole of cells.
plcB	Encodes the PlcB enzyme. It plays an important part in the breakdown of the two-membrane phagocytic vacuole that surrounds the bacteria during transfer with LLO.
mpl	Mediates the maturation of the PLC.
actA	Actin-based encodes the ActA protein, which is essential for intra and intercellular motility.
inlA	Facilitates the entry into the host cell via a receptor called E-cadherin.
inlB	Contributes in the entering the cell through the c-Met receptor.
inlC (irpA)	It blocks the activation of NF-κB, thus slowing down the innate immune system.
prfA	Transcriptional activator of hly, plcA, plcB, actA, inlA, inlB and mpl.

hly: Hemolysin; LLO: Listerolysin O; plcA: Phospholipase A; plcB: Phospholipase B; mpl: Metalloprotease; PLC: Phospholipase; actA: Actin polymerization protein, İnlA: İnternalin A; inlB: İnternalin B; inlC: İnternalin C; irpA: İnternalin-related protein A; NF-κB: Nuclear factor kappa B; prfA: Positive regulatory factor A.

BIOFILM RESISTANCE AND INACTIVATION

Normally, the biofilm form of the bacteria is more resistant to disinfection than its free form. Therefore, the resistance of bacteria to antimicrobials changes due to biofilm formation and adhesion. Biofilm forming and adhesion properties differ, even among strains. Biofilm forms of *Listeria* vary according to the physical and chemical properties of the surface, bactericidal agents, temperature, growth phase of the bacteria, cell characteristics, presence of food residues and other microorganisms in the environment. There is no effective strategy and antimicrobial selection standardization for the control of biofilm formation.[16]

Chemical disinfectants such as chlorine, ammonium quartener compounds, hydrogen peroxide and sodium hypochlorite are among the most popular methods for removing biofilms. The disadvantage of chemical disinfectants such as hypochlorides, ammonium quartener compounds and iodophors is that their effectiveness decreases at low temperatures.[17] The use of disinfectants can trigger resistant phenotypes and raise concerns about their environmental impact. Therefore, the emergence of disinfectants that do not create dangerous by-products has become inevitable. Ozone or acidic electrolyzed water, which does not leave any chemical residues, are considered environmentally friendly biocides.[18] Enzymes can also be used to improve disinfection processes. These can increase the effectiveness of classical disinfectants by dissolving the biofilm matrix. Depending on the nature of the biofilm composition, different enzyme types such as protease, cellulease, DNAase, alginate lyases, polysaccharide depolymerases and dispersin B are preferred.[19] Natural or engineered bacteriophages have also been found to be effective against foodborne pathogens. However, their effectiveness is more effective on those removed from the surface than in biofilm formation.[20] Functional carbohydrates acting as lectin inhibitors can also be used to coat materials and inhibit the production of adhesins.[21] Natural or chemical alternative coating molecules have also been found to be effective in preventing bacterial growth and adhesion on surfaces.[17] Superficial modifications using nanomaterials such as silver, cobalt, and iron mixed oxides have also been studied.[18] Another promising method to prevent biofilm formation is to use *Quorum Sensing* antagonistic molecules.[19] These molecules prevent biofilm application and maturation as they inhibit bacterial communication. Compounds such as brominated furanones have been shown to effectively reduce biofilm formation.[22]

RESISTANCE AGAINST STRESS FACTORS

Bacteria are exposed to many food-related stress factors, external factors or host's own defense mechanisms that affect their survival and activities throughout the food chain. *L. monocytogenes* is known to be a food pathogen that causes epidemics with a high mortality rate of 20-30%.[23] In this respect, it exceeds the fatality rates of other common foodborne pathogens such as *Salmonella Enteritidis* (0.38%), *Campylobacter* spp. (0.02-0.1%) and *Vibrio* spp. (0.005-0.01%).[24] Its ability to survive under various stress conditions increases the risk of contamination and colonization. Among the stress factors that bacteria are exposed are pH, salt, bacteriocins and some food preservative additives. In addition, it should not be forgotten that there are many stresses such as thermal processes at various process stages involved in this stress, high pressure, radiation, cooling processes during storage, and adjustments in atmospheric composition. Cross-protection of different stresses against each other also poses an important problem. For example, storage at low temperatures increases resistance to high salt concentration. Along with that, osmotic stress in *L. monocytogenes* can lead to cross defence opposing other stressors, including heat, ethanol, high levels of acidity, alkalinity and oxidative stress. In *L. monocytogenes* and other gram positive bacteria, sigma factor (σ^B) has been defined as a general alternative stress response. Adequate resistance mechanisms are triggered by the activation of promoters linked to σ^B, and σ^B is activated after exposure to various environmental factors. Thus, osmotic stress, cold stress, acid stress oxidative stress contribute to maintain its viability under high hydrostatic pressure.[23] Some response proteins formed by *L. monocytogenes* under stress conditions are presented in Table 4.

TABLE 4: Proteomic and transcriptional analysis of the stress response profiles of *L. monocytogenes*.[25]

Stress	Response protein
Cold	Fri, Ctc, GroEL, Dnak, Csp1-Csp4, CspA, CspD
Heat	Hsps, DnaK, GroES
Salt	Ctc, DnaK, GbuA, AppA
Acid	ASP, GbuA, GroEL, ClpP
Alkaline	DdlA, GroEL, Dnak
High hydrostatic pressure	Fri, GroES, PepF, PepT

ANTIBIOTIC RESISTANCE

The first antimicrobial resistant *L. monocytogenes* strains were identified in 1988. Since then, there has been an increasing resistance to a single or multiple antibiotics in strains isolated from human, animal, food businesses and dairy farms. Although *L. monocytogenes* is thought to be sensitive to a wide variety of antibiotics, intrinsic antibiotic resistance has been described against first generation quinolones, fosfomycin, monobactams and broad spectrum cephalosporins. In addition, the widespread use of antibiotics in the treatment of listeriosis has also contributed to the emergence of resistant strains for antibiotics. Beta lactams such as penicillin and ampicillin with or without gentamicin constitute the main antimicrobials used for the therapy of listeriosis. Vancomycin and trimethoprim/sulfamethoxazole can be used as an alternative treatment in penicillin-sensitive individuals.[26] The use of antimicrobials in the prophylaxis, disease treatment and as growth promoters in food producing animals is also important in the emergence of resistant strains. It has been reported that antibiotic resistance is formed against antibiotics such as sulfisoxazole, amoxacillin/clavulanic acid, ampicillin, spectinomycin, tetracycline, chloramphenicol, florphenicol, streptomycin, cotrimoxazole, 12 carbadox and trimethoprim, which are extensively used in poultry farming.[27,28] Antibiotic resistance, and especially multiple resistance, remains a central concern for public health. Thus, changes in *L. monocytogenes* antibiotic resistance need to be monitored as a consequence of the continuous emergence of resistant strains, remarkably through the integration of phenotypic and genotypic techniques.[26]

PREVALANCE IN FOOD

L. monocytogenes is a ubiquitous microorganism that can often be isolated from soil, aquatic environments, animal feces and food. To date, this pathogen has been identified in a variety of ready-to-eat foods such as meat and dairy products, appetizers and salads. In the recent decades, cases of foodborne listeriosis are related to the rised frequency in the uptake of ready-to-eat foods, especially in western countries.[17]

READY TO EAT MEAT PRODUCTS

It is believed that raw red meat and poultry will not be a source of listeriosis as long as they are cooked adequately and are not cross contaminated. Nevertheless, raw and poorly cooked meat is the main source of post-processing contamination. It is stated that hot dogs, delicatessen meat and poultry products are responsible for major epidemics in the USA.[29] In Turkey, İşleyici et al. analyzed 290 ready-made meatball samples. They stated that 32 (11.04%) of these samples were contaminated with *L. monocytogenes*.[30] Di Pinto et al. detected *L. monocytogenes* in 23 (20.5%) of 112 vacuum packaged sliced salami samples collected from supermarkets in Italy.[31] The prevalence is thought to depend on the type of the ready-to-eat meat products, whether it is heat-treated or fermented, its composition and internal characteristics. *L. monocytogenes* contamination was reported to be higher in meat with high pH and water activity and low nitrite concentration. However, *L. monocytogenes* has been found to be less frequently in fermented meat products.[29] Büyükünal et al. determined the presence of *L. monocytogenes* in 132 sausage and 66 bacon samples collected from retail sales points and producers in Istanbul, Adapazarı, Afyon and Kayseri. In this study, the prevalence of *L. monocytogenes* was found to be 2.02%.[32] However, Angelidis and Koutsoumanis in Greece found close values in fermented (8.2%) and heat-treated meats (8.1%) in their study on pre-cut and packaged meat selled in supermarkets.[33]

SEAFOODS

It is stated that the cases of *L. monocytogenes* related to seafood are sporadic or in the form of small epidemics. Listeriosis outbreaks are often reported to be associated with consumption of cold-smoked rainbow trout.[29] Vitas et al. found that the prevalence of smoked salmon samples collected from supermarkets in Spain was higher than other ready-made food samples.[34] Acargil detected *L. monocytogenes* in 4 out of 60 hot-smoked rainbow trout fillets and found its prevalence to be 6.6%.[35] Although cold smoked fish products have higher contamination than hot smoked fish products, post-

processing contamination can cause the pathogen to multiply rapidly depending on the storage temperature. Loncarevic et al. detected the highest *L. monocytogenes* concentration in hot smoked rainbow trout samples among fish samples collected from markets in Sweden.[36] Contamination situations related to the type of packaging may vary. Dominguez et al. collected cold smoked salmon samples in a study they conducted in Spain and reported that the prevalence of the pathogen was higher in open samples (28.5%) than vacuum-packed samples (17.7%).[37] However, in a study conducted in the USA between 2000-2001, they found that the prevalence of seafood salads packaged by producers (1.4%) was lower than those packaged at the point of sale (6.9%).[38] In addition, a study found the prevalence to be 0.2% in 500 haddock and red mullet fish samples caught from the Black Sea.[39]

MILK AND MILK PRODUCTS

The presence of *L. monocytogenes* related to listeriosis outbreaks has been reported in various milk and dairy products, especially pasteurized milk, chocolate milk, soft cheese and butter. If raw milk is used unpasteurized, it is a major concern in products such as cheese. As a matter of fact, Altun found that 4 out of 88 raw goat milk samples from *Listeria* spp. (4.54%), and identified 2 of them (2.27%) as virulent *L. monocytogenes*.[40] In another study conducted on 110 raw milk and dairy products in Ankara, the presence of *L. monocytogenes* was found to be 4.55% (5).[41] Application of normal pasteurization processes is sufficient to eliminate the appereance of *L. monocytogenes* in milk. Its presence in fully pasteurized products is due to post-processing contamination. It has been reported that the currency of *L. monocytogenes* is lower in dairy products than in other seafood and processed ready-to-eat foods.[29] In a study conducted in the USA, only 1 (0.018%) of 5,519 milk samples taken from retail outlets were found to be pathogen-positive.[42] However, cheeses, especially soft cheeses, have been found to be more contaminated than other types of cheese due to their high pH and water activity and low salt concentration.[29]

FRUITS AND VEGETABLES

It seems difficult to determine the source of contamination of fruits and vegetables in the production chain. The soil is seen as the main reservoir for fruits and vegetables. Especially pre-harvest and improper agricultural practices can contribute to the contamination of the pathogen in fresh produce. In addition, they can serve this contribution at various stages in humans and animals. Owing to the fact that fruits and vegetables are generally consumed raw, their prevalence in production is not clear compared to other convenience foods.[29] In Spain, Soriano et al. found the incidence equal when comparing raw and prepared lettuce.[43] Kara et al. detected only 1 (1.43%) of *L. monocytogenes* in 70 fresh lettuce samples in Afyon province.[44] Mena et al. found the contamination rates between 14.8% and 22.6% in their research on frozen vegetables in Portugal. Chen et al. evaluated the prevalence and contamination degree of *L. monocytogenes* in 72 vegetable samples.[45,46] Approximately 22% of the samples were found to be positive for *L. monocytogenes*. Lee et al., found the occurence of *L. monocytogenes* in 3 of the frozen pepper samples examined in Bursa.[47] Although such products are considered to be less risky than ready-to-use ones, the necessary preparation conditions must be clearly stated on the label before consumption. In addition, melon and chopped celery have been shown to be the cause of major listeriosis outbreaks.[17]

PREVALANCE OF *L. MONOCYTOGENES*

The World Health Organization (WHO) reports that annually, 600 million people are infected with foodborne diseases. Foodborne diseases affect the socio-economic development by straining the health system, as well as harming the country's economy, tourism and trade. *L. monocytogenes*, the etiological agent of listeriosis, is known to cause sporadic cases in which a particular food source is rarely identified. In the view of the fact that, listeriosis has a long and inconsistent incubation time (3 to 70 days), it is difficult to investigate the origin of an outbreak.[48]

UNITED STATES OF AMERICA (USA)

The US Center for Disease Control and Prevention reported that every year, listeriosis is responsible for

approximately 1,600 diseases and 260 deaths in the US. The annual incidence in 2013 was 0.26 cases per 100,000 people, with a total of 123 cases, 112 hospitalizations and 24 deaths reported.[49,50]

EUROPE

According to 2017 case reports in Europe, a rate of 0.42 cases per 100,000 people was stated. This rate was the same in 2016. The highest rate was 1.7 cases per 100,000 people over the age of 64.[51,52] The number of cases in the European Union for the years 2013-2017 is given in Table 5.

AUSTRALIA

Listeriosis as a consequence of the ingestion of contaminated food is common in Australia. In Australia, the five-year average of cases between 2011 and 2015 was 0.3 per 100,000 population, and the average annual number of cases was 78.[54]

AFRICA

In South Africa, the world's massive foodborne listeriosis epidemic occurred in a period from January 2017 to mid-July 2018.[55] In addition, the number of cases until October 2013-2019 is shown in Figure 1.

ASIA

Unlike the USA and Europe, where the health surveillance system is efficient, data collection for listeriosis has not been properly done in Asia. In addition, cases of listeriosis have been divulged in some Asian countries.[26] Feng et al. examined the patients reported for listeriosis in China between 1964 and 2010 and stated that there were 147 sporadic cases and 82 epidemic cases. They announced that mortality rates were 26% in total listeriosis and 46% in neonates listeriosis.[57] By systematically evaluating the articles on listeriosis published between 2011 and 2017 in China, the clinical and epidemiological features of listeriosis were reviewed. Out of 562 reported patients, excluding one patient, the others were infected with *L. monocytogenes*. These cases were associated with various factors, and it was reported that only 17 patients consumed raw/cold food, mostly barbecue, or a contaminated food diet. Three other patients were infected due to consumption of beef bought in supermarkets.[58] In a study conducted in Japan, cases of listeriosis were lower compared to Europe and the USA. The reason for the similarity of isolations in foods, but the low incidence of listeriosis cases has been attributed to other factors.[59]

TABLE 5: Human cases of listeriosis in the European Union during 2013-2017.[53]

	2013	2014	2015	2016	2017
Number of confirmed cases	1,883	2,217	2,183	2,509	2,480

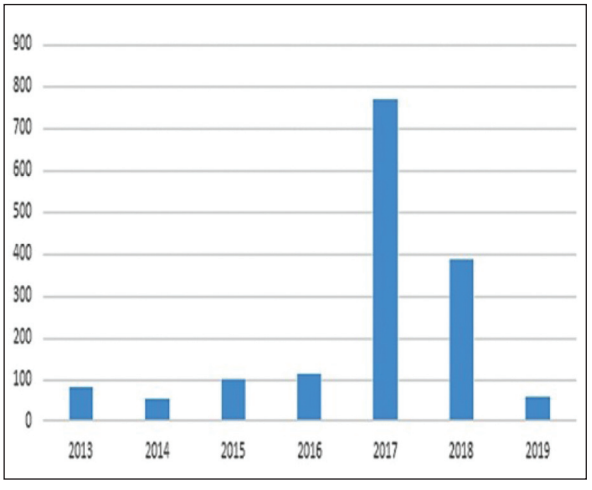

FIGURE 1: Listeriosis cases in South Africa 2013- October 2019.[56]

TURKEY

Reported listeriosis cases in Turkey has been in limited numbers. As a matter of fact, Özgenç et al. found 24 cases in separate cases in the last 20 years.[60] Doğanay, in his study by evaluating the listeriosis cases reported in Turkish journals between 1987 and 2001, reported that there were 32 cases in total, 6 of which were not published. In both of these studies, no information was provided about the connection of listeriosis cases with food.[61] Contrastingly, investigations have declared the ubiquity of *L. monocytogenes* in many foods. Especially grilled chicken, kokoretsi, grilled fish, fried mussels, frozen raw İnegöl (40° 4' 41" North, 29° 30' 48" East) meatballs were among the risky foods due to their *L. monocytogenes* content.[62] Listeriosis cases in some regions in the world and the foods responsible for listeriosis are given in Table 6.

TABLE 6: Listeriosis cases and responsible foods by region.

Country	Year	City	Case number (person)	Food	Death (person or %)	Reference
Africa						
	2017-2018	Multistate	1060	Polony	216	55
Asia						
	2001	Japan	86	Cheese		70
Australia						
	2003		60-70		%20-30	
	2009	Multijurisdictional	36	Chicken wrap	4	
	2010	Multijurisdictional	71	Melon	15	73
	2014	%56 of New South Wales and Victoria	80			
	2018	New South Wales	22	Rockmelon	7	
Europa						
	2012-2016	Czechia	26	Turkey meat	3	69
	2015-2018	Finlad Sweden United Kingdom Denmark Austria	32		2	68
	2016-2018	Austria Denmark Finland Sweden	32	Frozen corn	6	67
	2015-2018	Denmark Germany Poland	12	Cold smoked salmon	6	66
	2019	Denmark, Estonia, France, Sweden, Finland	22	Cold smoked fish products	5	65
	2019	Spain	222	Chilled roasted pork	3	64
USA						
	2011	Multistate	147	Cantaloupes	33	
	2012	Multistate	22	Ricotto Salata cheese	4	
	2013	Illinois, Indiana, Minnesota, Ohio, Texas	6	Cheese	1	
	2014	Multistate	8	Dairy products	1	
	2015	Multistate	30	Soft cheese	3	
	2015	Arizona, Kansas, Oklahoma, Texas	10	İce cream	3	
	2016	California Connecticut Maryland Washington	9	Frozen vegetables	3	
	2016	California Florida	2	Raw milk	1	63
	2016	Multistate	19	Packaged salads	1	
	2017	Connecticut Florida New York Vermont	8	Soft raw milk cheese	2	
	2018	North Carolina Virginia	4	Deli ham	1	
	2018	Louisiana Michigan Tennessee Texas	4	Pork products	0	
	2019	Multistate	8	Hard boiled eggs	1	
	2019	Multistate	10	Deli-sliced meats and cheeses	1	
	2020	Multistate	36	Enoki mushrooms	4	

LISTERIOSIS CAUSED BY *L. MONOCYTOGENES*

Listeriosis is a serious disease that can be mild or severe, generalized and localized in humans and animals. The disease primarily affects pregnant women, infants, people with chronic diseases and the seniors. Healthful youngsters and children have occasional listeriosis, but the infection rarely causes serious distress in these individuals. Most cases in humans are sporadic and often unreported, making its incidence difficult to be determined. Hospitalization rates in Europe and the USA are over 95%.[71] It is estimated that 80-90% of the listeriosis cases are caused by the uptake of tainted foods, and the rest are caused by maternal transmission from pregnant women.[7]

Outbreaks in humans usually occur in urban environments during the summer months, and outbreaks can occur with animal-derived or faecally contaminated vegetable products, post-processing contamination of meats or proliferation of *L. monocytogenes* in refrigerated foods. Ready-to-eat foods and the gastrointestinal presence of bacteria are also among the important risk factors. Although the average incubation period is 8 days, it can go up to 70 days. This has been reported as 27.5 days on average in pregnant women.[71] There is a 10-fold increased risk in pregnant women, especially due to low immunity in the last trimester of pregnancy.[72] The dose of infection is not known exactly, and this contaminated food can go up to 10^6 cfu/g. This may vary depending on the strain virulence, host sensitivity, and the nature of the food matrix.[71] 20% to 33% of gravidness-related infections result in stillbirth or miscarriage. Neonatal infection occurs in two forms. Early sepsis syndrome occurs mostly within the first 48 hours after birth, while late meningitis syndrome occurs at the age of 2 weeks. Bacteremia without typical symptoms is the most frequent listeriosis infection after newly-born infection. Clinical findings typically present with fever and myalgia, nausea and diarrhea. Meningitis cases caused by *L. monocytogenes* are alike to many other meningitis cases. Listerial endocarditis adds up to about 6% to 7.5% of adult listerial infections. The aforementioned, includes cases that result in local infections, conjunctivitis, skin infection, and lymphadenitis. Fever gastroenteritis can occur as a precursor symptom in invasive patients.[72]

LISTERIOSIS AWARENESS OF CONSUMERS

Despite various educational campaign and media attention following deadly and massive food outbreaks, awareness of listeriosis among endangered personages appears to be low-lying. It is believed that even if awareness of listeriosis increases, people will not be able to take precautions more precisely, and the number of cases will not decrease.[73] This is supported by studies published in the recent past by Xu et al.[74] Of the 78.9% of pregnant women who reported being aware of *L. monocytogenes*, only 28.9% reported not eating high-risk food. Consumers' behavior are largely unconscious towards preparing safe food in their daily routines and tend to have a strong self-assurance in their abilities.[73]

Middle-aged people do not perceive the key listeriosis risk factors and do not think that their individual food practices have the potential to increase the risk of listeriosis. They are confident in their abilities, and that confidence generally increases with age. The same scenarios are seen in immunocompromised individuals.[73] In a case-control study, Preuβel et al. reported that 80% of listeriosis patients not linked with pregnancy in Germany had no knowledge of listeriosis before they got sick. On the other hand, only 6% of the immunodeficient control population stated that they did not know that listeriosis was caused by food.[75]

CONCLUSION

L. monocytogenes can be found ubiquitously and can be isolated from fresh and frozen meats, including soil, vegetation, sewage, water, animal feed, poultry, slaughterhouse waste, and feces of healthy animals and humans. Because *L. monocytogenes* can survive and grow in low temperatures, this pathogen adapts to an extensive variety of environmental conditions due to the resistance mechanisms it uses. Contamination of food can occur at any stage: after manufacturing, processing and cooking. Consumption of food

contaminated with *L. monocytogenes* causes listeriosis among fragile groups of people such as pregnant women and fetuses, the neonate, the elderly and people with weakened immune systems. Therefore, the rules to be followed particularly for this group of people but not only are listed below:

- Food should be thoroughly cooked (70ºC and above), notably meat, poultry, eggs and seafood
- Keep food at safe temperatures
- Keep raw meat apart from fresh products and different ready-to-eat foods
- Do not leave prepared food at ambient temperature for more than 2 hours
- Cooked and perishable food should be immediately cooled below 5°C
- Do not store food for too long, even in the fridge
- By no means, defrost frozen food at room temperature
- Must utilize safe water for raw materials
- Prefer safe processed foods such as pasteurized milk
- Fruits and vegetables should be washed prior to consumption
- Kitchen equipment such as knives, cutting boards and graters should be washed pre and post preparing ready-made foods.

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Conflict of Interest

No conflicts of interest between the authors and / or family members of the scientific and medical committee members or members of the potential conflicts of interest, counseling, expertise, working conditions, share holding and similar situations in any firm.

Authorship Contributions

Idea/Concept: *Fulya Taşçı, Zeki Erol;* ***Design:*** *Zeki Erol;* ***Control/Supervision:*** *Fulya Taşçı;* ***Data Collection and/or Processing:*** *Zeki Erol;* ***Analysis and/or Interpretation:*** *Fulya Taşçı, Zeki Erol;* ***Literature Review:*** *Zeki Erol;* ***Writing the Article:*** *Zeki Erol;* ***Critical Review:*** *Fulya Taşçı;* ***References and Fundings:*** *Zeki Erol;* ***Materials:****Zeki Erol.*

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