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The investigation of the presence of *Listeria* species in poultry farms and antimicrobial resistance profiles of *Listeria monocytogenes* strains

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ABSTRACT

Listeria species are widespread in nature and found in various environments. In this study, the presence of *Listeria* species in poultry farms environments was investigated. For this purpose, a total of 332 samples including feces, feed, drinking water and nipple swab from 5 broiler flocks reared on the floor and 352 samples including feces, feed, drinking water and eggshell swab from 5 layer flocks reared in cages were obtained. A modified version of the USDA-FSIS MLG 8.13 method was used for the isolation of *Listeria* species from samples. As a result of the study, *Listeria* spp. was isolated from all broiler and layer flocks. Isolation of *Listeria* spp. was carried out from 18 of 190 feces samples (9.4%), 5 of 15 drinking water samples (33.3%), 3 of 102 nipple swab samples (2.9%) in broiler flocks, and 22 of 167 feces samples (13.1%), 2 of 25 feed samples (8%), 3 of 15 drinking water samples (20%), 3 of 145 eggshell swab samples (2%) in layer flocks. Isolates were identified by cultural and biochemical characters, and a total of 56 *Listeria* isolates were identified as 15 *L. monocytogenes,* 3 *L. ivanovii,* 19 *L. innocua,* 13 *L. seeligeri,* 2 *L. welshimeri,* and 4 *L. grayi*. The antibiotic resistance profiles of *L. monocytogenes* isolates to eleven antibiotics were detected by the disc diffusion method. *L. monocytogenes* isolates were found to exhibit the highest resistance to ciprofloxacin (33.3%) among eleven antibiotics, and three isolates (20%) were also multidrug resistant. Consequently, it was determined that *Listeria* species in poultry farms environments are common.

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Introduction

The bacteria of the genus *Listeria* are found in various environments including soil, water, vegetation, sewage, animal feeds and farm environments. The genus *Listeria* currently includes 17 recognized species; *Listeria monocytogenes* (*L. monocytogenes), L. ivanovii*, *L. grayi, L. innocua, L. seeligeri, L. welshimeri*, *L. marthii*, *L. rocourtiae, L. fleischmannii*, *L. weihenstephanensis*, *L. floridensis*, *L. aquatica*, *L. cornellensis*, *L. riparia*, *L. grandensis*, *L. newyorkensis and L. booriae*. Only two of these species, *L. monocytogenes* and *L. ivanovii*, are pathogenic for animals and humans (Orsi and Wiedmann, 2016). However, sporadic human infections due to *L. seeligeri* and *L. innocua* have also been reported (Rocourt et al., 1986; Perrin et al., 2003).

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L. monocytogenes is one of the important foodborne pathogens and threats public health in many countries (Datta and Burall, 2018). Human listeriosis can occur sporadically or epidemically; in both cases, contaminated foods are the primary means of transmission. Direct contact with animals/birds is of less significance in the transmission/spread of *Listeri*a, except for highly susceptible individuals (Dhama et al., 2015). Many different food items, such as dairy products, red meat, poultry meat, seafood, vegetables and ready-to-eat prepared foods have been implicated as vehicles for *L. monocy*togenes transmission (Lunde´n et al., 2004; Şanlıbaba et al., 2018). The severity of listeriosis can range from mild gastroenteritis to severe disease conditions linked to septicemia, encephalitis, meningitis, miscarriage, and may even lead to death. The mortality rate can reach values of around 30%. Some people have a higher risk of listeriosis than others, such as newborns, the elderly, immunocompromised people and pregnant women (Zhu et al., 2017). In the treatment of listeriosis with antibiotics, β-lactam antibiotics (e.g., penicillin or ampicillin) alone or in combination with an aminoglycoside (e.g., gentamicin) is frequently used as the first treatment of choice. The association of trimethoprim and a sulfonamide (e.g. sulfamethoxazole) is considered to be a second-choice therapy, especially for patients allergic to β-lactams. However, vancomycin, erythromycin, rifampicin, chloramphenicol, tetracycline and fluoroquinolones are also used to treat cases of listeriosis (Olaimat et al., 2018). In recent years, *L. monocytogenes* isolated from foods, the environment and human sources have been found to be resistant to commonly used antibiotics (Alonso-Hernando et al., 2012; Soni et al., 2013; Şanlıbaba et al., 2018; Arslan and Baytur, 2019). But, the levels and type of resistance are affected by regional differences and antimicrobial usage in humans and animals. Thus, investigation and monitoring of the antibiotic susceptibility of *Listeria* species isolated from different sources are very important for public health (Dhama et al., 2015).

Listeriosis has been reported in a wide range of species of domestic and wild animals, including birds. In birds, even though outbreaks of listeriosis are infrequent, commonly affected species include chickens, turkey, geese, ducks, pigeons, parrots, wood grouse, snowy owls, eagles and canaries (Seifi, 2012; Dhama et al., 2013). Chickens are thought to be the carriers of *Listeria* and thus contaminate the litter and environment of the poultry production systems. Intestinal colonization and the presence of *L. monocytogenes* in the feces of poultry play a significant role in the spread of listeriosis in domestic animals (Dhama et al., 2015). However, few studies have investigated and characterized *Listeria* species in poultry farm environments (Dahshan et al., 2016; Locatelli et al., 2017). Most studies have focused on investigating the presence of *L. monocytogenes* in final products and in food-processing and retail environments thought to be the main sources of contamination for the final products (Ojeniyi et al., 1996; Oliveira et al., 2018). In a farm-to-fork approach, it is necessary to assess the presence of *L. monocytogenes* throughout the entire poultry production chain, including the farm environment and the primary production stage (Locatelli et al., 2017). In Türkiye, numerous studies have investigated the prevalence of *L. monocytogenes* in chicken carcases or fresh chicken meats (Siriken et al., 2014; Gücükoğlu et al., 2020), but to the best of our knowledge, there is no data on the presence of *Listeria* spp. from poultry farms and environments. This study was aimed to investigate the presence of *Listeria* spp. in broiler and laying hen flocks environment and also determine the antimicrobial resistance profiles of *L. monocytogenes* isolated.

Materials and Methods

Sample collection: A total of 684 samples were taken from 5 broiler flocks reared on the floor and 5 layer flocks reared in cages located in Bandirma, Türkiye. Each of the flocks was determined randomly, and the samples were taken from each flock in one go. Chickens were clinically healthy appearance and there was no record of listeriosis cases in all flocks. From the broiler flocks, a total of 332 samples including feces, feed, drinking water and nipple swabs were collected. Also, a total of 352 samples including feces, feed, drinking water and eggshell swabs were collected from the layer flocks. The details of the samples and the flock's information are given in Table 1. Feces (fresh and not trampled) samples were randomly collected by using sterile swabs from the surface of the litter in broiler houses and from the cages (only one sample from each cage) in layer houses, and fecal samples were placed in sterile screw-top vials. The feed samples (approximately 200 g each) were taken from five different points on the feeding stations in each house. The drinking water samples (1 liter each) from each poultry flock were collected from the water tank in the house (two samples) and from the water tank on the outside of the house (one sample). Swab samples were randomly taken from nipple drinkers in broiler houses. In layer houses, eggs were randomly chosen directly from the collecting conveyor and then

swab samples were taken from eggshell. All samples were taken aseptically and quickly transported to the laboratory under chilled conditions, and stored at 4 °C until analyzed. Samples were analyzed within 3 h. of being taken.

Isolation and identification of *Listeria* spp.: The *Listeria* strains were isolated according to the US Department of Agriculture (USDA/FSIS) method (USDA, 2021) with some modification. Briefly, each of the swab samples was directly inoculated into 100 mL of University of Vermont *Listeria* Enrichment broth (UVM I, Oxoid, CM863, SR142). Twenty-five grams of each feed sample were inoculated into 225 mL of UVM I. The water samples were filtered through 0.45 µm membrane filters (Millipore, GSWG047S1) using a membrane filtration system (Sartorius AG) and the filters were placed in 100 mL of UVM I broth. All enrichments were homogenized and then incubated at 30 °C for 24 h. One mL of primary enrichments was transfered to 9 mL UVM II broth (Oxoid, CM863, SR143) and Fraser broth (Oxoid, CM895, SR156), and incubated at 35 °C for 24 h. Secondary enrichments were streaked onto modified Oxford agar (Oxoid, CM856, SR206) and BrillanceTM *Listeria* agar (Oxoid, CM1080, SR227), and incubated at 37 °C for 48 h. All plates were examined for typical *Listeria* colonies, and colonies that were grayish surrounded with a dark zone on modified Oxford agar, and blue with or without opaque white halos on BrillanceTM *Listeria* agar were accepted as suspects for *Listeria* spp. Five suspected *Listeria* colonies were picked up from each sample and transferred to Tryptone Soya agar (Oxoid, CM131) plates supplemented with 0.6% Yeast extract (Oxoid, L21) for pure culture, and incubated at 37 °C for 24. All the isolates were confirmed to the standard identification and biochemical tests including Gram staining, catalase and oxidase reaction, H2S production, indole test, urease activity, motility at 25°C and 37°C, β-haemolysis, nitrate reduction, methyl-red-Voges Proskauer test, CAMP test with control strains of *Staphylococcus aureus* and *Rhodococcus equi*, and acide production from rhamnose, xylose, mannitol and α-methyl-D-mannopyranoside (Gasanov et al., 2005). All media used were prepared according to the manufacturer's directions (Oxoid). The reference strain *L. monocytogenes* ATCC7644 was used in all biochemical tests.

Antibiotic susceptibility test: All *L. monocytogenes* isolates were tested for antimicrobial susceptibility using the disc diffusion method as described by Clinical and Laboratory Standards Institute (CLSI) (CLSI, 2014). Briefly, a single colony of each isolate was inoculated into 10 mL of tryptic soy broth containing 0.6% yeast extract. After incubation at 30 °C for 24 h., the culture was diluted to a turbidity equivalent to the McFarland 0.5 standard in sterile physiological saline solution. Then, using the sterile swab, inoculation was conducted on Mueller-Hinton agar (Oxoid, CM337) containing 5% sheep blood. Later on, antibiotic discs were placed and the plates were incubated at 35°C for 24 h. After the incubation, the diameters of the inhibition zones were measured with calipers and compared to the breakpoints for *Staphylococcus* spp. as recommended by the CLSI (CLSI, 2014). Currently, there are no resistance criteria for *Listeria* susceptibility testing in the CLSI guidelines, with the exception of susceptibility breakpoints for ampicillin and penicillin. The strains were classified as resistant, intermediate resistant, and susceptible. The eleven antimicrobial agents used and their corresponding concentrations were as follows: penicillin G (10 U), ampicillin (10 µg), gentamycin (10 µg), tetracycline (30 µg), erythromycin (15 µg), streptomycin (10 µg), vancomycin (30 µg), chloramphenicol (30 µg), rifampicin (5 µg), ciprofloxacin (5 µg) and trimethoprim/sulphamethoxazole (1.25/23.75 µg) (Oxoid, UK). *Staphylococcus aureus* ATCC 25923 and *Escherichia coli* 25922 were used for quality control strains and tested in each replicate.

Table 1. The information regarding the samples taken and flocks

Flock No	Broiler flocks					Layer flocks				
	Flock age (day)/ No of chickens	No of samples				Flock age(week)/ No of chickens	No of samples			
		Feces	Feed	Drinking water	Nipple swab		Feces	Feed	Drinking water	Eggshell swab
1	32/9.300	34	5	3	20	23/10.650	30	5	3	25
2	28/11.700	40	5	3	22	45/11.230	32	5	3	27
3	25/13.100	40	5	3	20	53/21.400	40	5	3	35
4	35/11.150	38	5	3	20	42/17.600	35	5	3	33
5	41/12.480	38	5	3	20	36/9.800	30	5	3	25
Total		190	25	15	102		167	25	15	145

Table 2: Prevalence of *Listeria* spp. isolated from broiler and layer flocks

Type of samples	Number of samples	*Listeria* spp. n (%)	*L. monocytogenes* n (%)	*L. İvanovii* n (%)	*L. innocua* n (%)	*L. seeligeri* n (%)	*L. welshimeri* n (%)	*L. grayi* n (%)
Broiler flocks								
Feces	190	18 (9.4)	4 (2.1)	-	7 (3.6)	6 (3.1)	1 (0.5)	-
Feed	25	-	-	-	-	-	-	-
Drinking water	15	5 (33.3)	2 (13.3)	1 (6.6)	-	-	-	2 (13.3)
Nipple swab	102	3 (2.9)	-	-	2 (19.6)	1 (0.9)	-	-
Total (%)	332	26 (7.8)	6 (1.8)	1 (0.3)	9 (2.7)	7 (2.1)	1 (0.3)	2 (0.6)
Layer flocks								
Feces	167	22 (13.1)	8 (4.7)	-	8 (4.7)	3 (1.7)	1 (0.5)	2 (1.1)
Feed	25	2 (8)	-	-	1 (4)	1 (4)	-	-
Drinking water	15	3 (20)	1 (6.6)	2 (13.3)	-	-	-	-
Eggshell swab	145	3 (2)	-	-	1 (0.6)	2 (1.3)	-	-
Total (%)	352	30 (8.5)	9 (2.5)	2 (0.5)	10 (2.8)	6 (1.7)	1 (0.2)	2 (0.5)

Results

Isolation and identification of *Listeria* spp.: In this study, a total of 56 *Listeria* spp. were isolated from 684 poultry farm environment samples. The prevalence of positive samples was 8.1%. Also, the prevalence of *Listeria* spp. in layer flocks as 8.5% (30/352) was higher than that in broiler flocks with the ratio of 7.8% (26/332) (Table 2). In broiler flocks, twenty-six *Listeria* spp. were isolated from feces and drinking water samples of five flocks and nipple swab samples of two flocks, but not isolated from the feed samples. These isolates were identified as: 6 *L. monocytogenes* (4 from feces of three flocks and 2 from drinking water of two flocks), 1 *L. ivanovii* (from drinking water of one flock), 9 *L. innocua* (7 from feces of four flocks and 2 from a nipple swab of two flocks), 7 *L. seeligeri* (6 from feces of five flocks and 1 from a nipple swab of one flock), 1 *L. welshimeri* (from feces of one flock) and 2 *L. grayi* (from drinking water of two flocks). In layer flocks, thirty *Listeria* spp. were isolated from feces samples of five flocks, drinking water and eggshell samples of three flocks, and feed samples of two flocks. These isolates were identified as: 9 *L. monocytogenes* (8 from feces of five flocks and 1 from drinking water of one flock), 2 *L. ivanovii* (from drinking water of two flocks), 10 *L. innocua* (8 from feces of five flocks, 2 from eggshell swab and feed of one flock), 6 *L. seeligeri* (3 from feces of three flocks, 2 from eggshell swab and 1 from feed of one flock), 1 *L. welshimeri* (from feces of one flock) and 2 *L. grayi* (from feces of two flocks). The isolation and identification results are given in Table 2.

Antibiotic susceptibility: Isolates of *L. monocytogenes* recovered from the feces and drinking water samples of the broiler and layer flocks were tested for their antibiotic susceptibility. Among the 15 *L. monocytogenes* isolates, 9 isolates (60%) were susceptible to eleven tested antimicrobial agents. The remaining 6 isolates (40%) were resistant to ciprofloxacin (five isolates, 33.3%, resistance or intermediate), to penicillin G (two isolates, 13.4%), to trimethoprim/sulphamethoxazole (two isolates, 13.4%), to ampicillin (one isolate, 6.7%), to vancomycin (one isolate, 6.7%), to rifampicin (one isolate, 6.7%) and to gentamycin (one isolate, 6.7%, intermediate). These 6 isolates resistant to antimicrobial agents were isolated from feces samples of broiler and layer flocks. The number of antimicrobials to which an isolate was resistant ranged from one to three. However, multidrug-resistance, i.e., resistance to three or more antimicrobial agents, was observed in 3 isolates (20%). Of these multidrug-resistant isolates, one isolate (from broiler feces) was resistant to ampicillin, ciprofloxacin, and trimethoprim/sulphamethoxazole, and the other two isolates (from layer feces) were resistant to ciprofloxacin, rifampicin, trimethoprim/ sulphamethoxazole and ciprofloxacin, penicillin G, vancomycin, respectively. The antimicrobial resistance of *L. monocytogenes* isolates was shown in Table 3.

Discussion

The Bandirma district, where the study was conducted, is located in the Southern Marmara Region of Türkiye, and domestic animal breeding is done quite intensively in this region. Also, the poultry sector is clustered in this region, where poultry farms are very intense, and about 12% of the broiler and 3% of the layer hens in our country are raised in this region. Therefore, this region is located in the front rows of chicken meat and table egg production in Türkiye (Anonymous, 2020). This study was planned due to the absence of a previous study on the subject of the presence of *Listeria* spp. in poultry farms in this region and Türkiye,

Table 3. Antimicrobial resistance profiles of *L. monocytogenes* isolates

Antimicrobial agent	Number of isolates (%) n=15		
	Resistant	Intermediate	Susceptible
Penicillin G	2 (13.4)	-	13 (86.6)
Ampicillin	1 (6.7)	-	14 (93.3)
Gentamycin	0 (0)	1 (6.7)	14 (93.3)
Tetracycline	0 (0)	-	15 (100)
Erythromycin	0 (0)	-	15 (100)
Streptomycin	0 (0)	-	15 (100)
Vancomycin	1 (6.7)	-	14 (93.3)
Chloramphenicol	0 (0)	-	15 (100)
Rifampicin	1 (6.7)	-	14 (93.3)
Ciprofloxacin	4 (26.7)	1 (6.7)	10 (66.6)
Trimethoprim/ Sulphamethoxazole	2 (13.4)	-	13 (86.6)

and has been carried out in broiler and layer flocks. In the study, the presence of *Listeria* spp. was determined at least one of the samples taken from each broiler and layer flock examined, and the overall prevalence of *Listeria* spp. was found as 8.1%, while 7.8% in broiler and 8.5% in layer flocks. In a study conducted by Dahshan et al. (2016) in Egypt, 48.3% of broiler farm samples including litter, feed and water were found to be contaminated with *Listeria* spp. Petersen and Madsen (2000) reported the prevalence of *Listeria* spp. in Danish broiler farm litter and fecal samples as 14%. In a study conducted by Milillo et al. (2012) in pasture-reared poultry demonstrated that environmental samples including cecal, soil and grass collected from the pasture before broiler introduction and samples collected after broiler exposure were found to harbour 5% and 53% *Listeria* spp., respectively. Those reported by the authors were found to be higher than our results. In a study performed by Iida et al. (1991) in Japan, a lower prevalence of *Listeria* spp. (4.7%) than our result was found in four chicken farm samples, including fresh feces. Likewise, Schwaiger et al. (2010) reported that the low *Listeria* spp. prevalence in cloacal swab samples from organic and conventional laying hen flocks was 1.3% and 1.8%, respectively, and only *L. innocua* was isolated. In previous studies, *L. innocua* has been shown to be the predominant species isolated from the poultry farms and environment (Dhama et al., 2013; Dahshan et al., 2016; Locatelli et al., 2017). However, other *Listeria* spp. such as *L. ivanovii*, *L. welshimeri* and *L. seeligeri* were also isolated less frequently (Dahshan et al., 2016), but most studies only focused on *L. monocytogenes* (Chemaly et al., 2008; Esteban et al., 2008, Kanarat et al., 2011). In this study, *L. innocua* was the predominant *Listeria* species and isolated at the highest rates in both production type flocks (2.7% in broilers and 2.8% in layers), and was also more prevalent than *L. monocytogenes* (1.8% in broilers and 2.5% in layers). Petersen and Madsen (2000) reported that non-pathogenic *L. innocua* (13%) was more prevalent than *L. monocytogenes* (3%) in broilers. *L. innocua* was declared to be important because it was closely related to *L. monocytogenes* and commonly used by the food industry as an indicator to identify environmental conditions allowing the presence, growth, and persistence of the relevant human pathogen *L. monocytogenes* (Gwida et al., 2020), and also because it has been reported that it may be an antimicrobial resistance reservoir for *L. monocytogenes* due to its availability in similar habitats (Bertrand et al., 2005). Also, other studies indicated that *L. innocua* was the most common species in chicken meat (Yücel et al., 2005; Fallah et al., 2012; Arslan and Baytur, 2019).

L. monocytogenes has been isolated on poultry farms and environments from litter, dust, feed, water, feces and cecal or cloacal swab samples, with an overall prevalence rate ranging from 0% to 91.5% in those samples (Skovgaard and Morgen, 1988; Petersen and Madsen, 2000; Chemaly et al., 2008; Schwaiger et al., 2010; Kanarat et al., 2011; Dahshan et al., 2016; Ishola et al., 2016; Locatelli et al., 2017; Gwida et al., 2020). In this study, *L. monocytogenes* was the second predominant species, with an average 2.2% prevalence rate, and it was isolated from feces and drinking water samples in both production type flocks. In a study conducted by Ishola et al. (2016) in Nigeria, the overall prevalance of *L. monocytogenes* contamination was found to be 91.5% in cloacal swabs from broilers and layers, and cloacal samples from broilers (98.8%) had significantly higher than those from layers (89.8%). Skovgard and Morgan (1988) reported that 33% harboured *L. monocytogenes* in the fecal samples taken from poultry in a dairy farm and from cages used for transporting poultry to slaughter. In another study, *L. monocytogenes* was detected in 10.5% of the feces samples from laying hen flocks, while in 34.6% of the feces samples when only *L. monocytogenes*-positive flocks were considered (Chemaly et al., 2008). In a study performed by Ojeniyi et al. (1996), *L. monocytogenes* was detected at a rate of 4.7% in caecal samples taken from broiler breeder flocks, but was not detected in 2078 caecal samples taken from 90 randomly selected broiler flocks. However, in some studies, *L. monocytogenes* was not isolated in cloacal or faecal swabs taken at the slaughterhouse but was

detected in chicken meat products (Iida et al., 1991; Cox et al., 1997; Kanarat et al., 2011). Whereas, in a study conducted by Kalender (2003), it was reported that *L. monocytogenes* was isolated from faecal samples of chickens on the slaughter line of abattoir. In our study, we isolated *L. monocytogenes* from 2.1% and 4.7% of feces samples in broiler and layer flocks, respectively. Since no cloacal swabs were taken in this study, cross-contamination of feces with *Listeria* spp. from the environment could not be ruled out. All these results showed that this variation in prevalence of *L. monocytogenes* might be attributed to various factors such as sample type (litter, dust, feces, cecal or cloacal swab), type of production systems and contact with other livestock farms, but also did not clarify whether the feces was a source for *L. monocytogenes* contamination in poultry products.

Drinking water has often been considered a risk factor for *L. monocytogenes* contamination in poultry flocks (Cooper et al., 1992; Aury et al., 2011; Seifi, 2012). In previous studies carried out on broiler chickens, it has been reported the presence of *C. jejuni* in biofilms in the water systems (Zimmer et al., 2003) and the isolation of *C. jejuni* from nipple swab samples, which may cause flock contamination (Cokal et al., 2011). With these, also taking into account the biofilm-forming ability of *L. monocytogenes* isolated from broilers and layers (Osman et al., 2020) and *L. monocytogenes*' ability to form biofilms in tap water (Gião et al., 2014), we collected nipple swab samples in broiler flocks. However, *L. monocytogenes* was not isolated, while *L. innocua* and *L. seeligeri* were isolated in these samples. Aury et al. (2011) reported that the nipples have a protective effect against *L. monocytogenes* contamination in broiler flocks. However, we isolated *L. ivanovii* in drinking water samples in both production type flocks. Similarly, Dahshan et al. (2016) also reported that *L. ivanovii* isolation was performed from water samples as well as litter, feed and raw chicken meat samples from broiler farms. Obviously, isolation from drinking water samples did not surprise us as *Listeria* spp. are highly adapted to soil, water and vegetation. In a study in different environmental ecosystems, it was reported that the most dominant species in soil and water samples were *L. seeligeri*, *L. innocua*, and *L. ivanovii* (Linke et al., 2014). The poultry farms in this study provided their drinking water from groundwater sources. Contamination of groundwater sources with *Listeria* spp. may be possible by prolonged rainfall and flooding (Linke et al., 2014). Further comprehensive studies are needed to provide a better understanding of the presence of *Listeria* spp. in the water systems of poultry farms. Also, in this study, *L. innocua* and *L. seeligeri* were isolated from feed and eggshell swab samples in layer flocks. These findings were comparable to the results reported by other investigators (Farber et al., 1992; Schwaiger et al., 2010; Gwida et al., 2020). It has been reported that feed meal was found to increase the risk of *L. monocytogenes* contamination in laying hen flocks (Aury et al., 2011).

L. monocytogenes is generally susceptible to a wide range of antibiotics (Hof et al., 1997). However, *L. monocytogenes* strains resistant to one or more antibiotics have been isolated from foodstuffs, environmental, animal and human samples. (Bertrand et al., 2005; Olaimat et al., 2018, Şanlıbaba et al., 2018). A few studies examined antibiotic susceptibility patterns of *L. monocytogenes* strains isolated from poultry farms (Dahshan et al., 2016; Ishola et al., 2016). In the present study, we detected that 40% of the *L. monocytogenes* isolates exhibited resistance or intermediate resistance to at least one antibiotic tested. This resistance rate was close to the 47.4% found in the study by Oliveira et al. (2018), higher than the 0.6% found in the study by Walsh et al. (2001) and lower than the 100% found in the study by Gücükoğlu et al. (2020). The highest resistance has been found against ciprofloxacin (33.3%) in this study. Şanlıbaba et al. (2018) reported a resistance rate (35.3%) to this antibiotic close to our finding. Arslan and Baytur (2019) reported a lower resistance rate (3%) to ciprofloxacin than our finding, while Dashan et al. (2016) and Ishola et al. (2016) reported higher resistance rates (50% and 56.2%). On the other hand, Yücel et al. (2005) and Oliveira et al. (2018) indicated that no resistance was detected to ciprofloxacin in their studies. The high resistance rate to ciprofloxacin in this study could be explained by the frequent use of this antibiotic to treat in infection in poultry farms in Türkiye. In the treatment of listeriosis, ampicillin or penicillin G alone or in combination with an aminoglycoside is considered the most effective therapy (Soni et al., 2013). The results of this study indicated that *L. monocytogenes* isolates are resistant to ampicillin (6.7%), penicillin G (13.4%) and gentamycin (6.7%, intermediate). Our levels of resistance to ampicillin, penicillin G and gentamycin were lower than those found by Fallah et al. (2012) at rates of 44.9%, 41.8% and 10.2%, respectively. In their study, Şanlıbaba et al. (2018) reported that all *L. monocytogenes* strains were resistant to ampicillin and penicillin, and susceptible to gentamycin, whereas Wang et al. (2015) reported that all *L. monocytogenes* strains were susceptible to these antibiotics. In this study, 13.4% of *L. monocytogenes*

strains were found to be resistant to trimethoprim/ sulphamethoxazole, which is a second-choice therapy for listeriosis. Similar findings have been reported by other researchers for this antibiotic (Şanlıbaba et al., 2018; Arslan and Baytur, 2019; Gücükoğlu et al., 2020). The percentage of isolates resistant to trimethoprim/ sulphamethoxazole detected by Wang et al. (2015) was 100%, while Fallah et al. (2012) reported that all *L. monocytogenes* strains were susceptible to that antibiotic in their study. In the present study, it has also been detected that *L. monocytogenes* isolates were resistant to vancomycin (6.7%), rifampicin (6.7%). Vancomycin and rifampicin are also used treat human listeriosis (Olaimat et al., 2018; Şanlıbaba et al., 2018). Other studies also reported that *L. monocytogenes* isolates were resistant to vancomycin (Siriken et al., 2014; Gücükoğlu et al., 2020), and rifampicin (Fallah et al., 2012; Şanlıbaba et al., 2018), similar to our results. However, vancomycin resistance of *L. monocytogenes* isolates was not reported in some studies (Fallah et al., 2012; Şanlıbaba et al., 2018; Arslan and Baytur, 2019). In addition, the present study showed that all *L. monocytogenes* isolates were susceptible to erythromycin, streptomycin, tetracycline and chloramphenicol. These results agree with those previously reported by some authors (Yücel et al., 2005; Soni et al., 2013; Siriken et al., 2014; Arslan and Baytur, 2019). Antibiotic susceptibility patterns of *L. monocytogenes* isolates to different antibiotics might be influenced by many factors such as strain variation, sampling sites, geographic origin and factors that trigger gene transfer (Walsh et al., 2001; Olaimat et al., 2018; Osman et al., 2020). Therefore, the present study comprising results that were different from those of some other researchers.

The evidence of the emergence of *L. monocytogenes* strains' multidrug resistance has been documented in recent years (Alonso-Hernando et al., 2012; Soni et al., 2013). In a study conducted by Alonso-Hernando et al. (2012) in Spain, multidrug resistance of *L. monocytogenes* strains from poultry products was reported to be more common in 2006 (84%) as compared to 1993 (18.6%). In India, Dhanashree et al. (2003) reported that all the isolates of *L. monocytogenes* were sensitive to antibiotics tested, while Soni et al. (2013) reported that all *L. monocytogenes* isolates were resistant to multiple antibiotics. In Türkiye, in a study performed by Siriken et al. (2014), it was reported that 1.9% and 5.6% of *L. monocytogenes* isolates from raw chicken meat were resistant to five and six antimicrobial agents, respectively. Şanlıbaba et al. (2018) investigated resistance of 17 *L. monocytogenes* isolates from raw ready-to-eat food to 23 antibiotics and reported that all isolates had multidrug resistant. However, multidrug resistance of *L. monocytogenes* isolates has also been reported in other countries (Fallah et al., 2012; Wang et al., 2015; Dahshan et al., 2016). In this study, we found 3 (20%) of 15 *L. monocytogenes* isolates from poultry farms had multidrug resistance. These results showed that there was multidrug resistance in *L. monocytogenes* strains isolated from various sources around the world, and this could be a potential public health hazard.

Conclusion

Listeria species including *L. monocytogenes* are quite common in poultry farms environments. *L. monocytogenes* is one of the important foodborne pathogens, and sporadic and isolated cases of listeriosis have been attributed to poultry. *L. monocytogenes* is isolated from other stages of the poultry production chain. Therefore, *Listeria* spp. found in the poultry farm environments can be a potential source of contamination for the poultry processing environments. Studies are needed to reveal the relationship between the presence of *Listeria* in poultry farms and *Listeria* contamination of poultry processing environments. Furthermore, the fact that *L. monocytogenes* strains isolated from poultry farms were resistant to antibiotics used in the treatment of listeriosis and that some of these strains were multidrug resistant poses a potential risk to public health. Therefore, taking preventive measures against *Listeria* contamination in poultry farms and continuous surveillance of antibiotic sensitivity are absolutely necessary for future risk assessment and consumer protection, i.e., public health protection.

Ethical approval

The authors declare no ethical approval is required.

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