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Original Article

Occurrence and Antibiotic Susceptibility of *Listeria* Species and *Staphylococcus aureus* in Cattle Slaughterhouses of Kerala, South India

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Abstract

A total of 765 samples were collected from beef carcasses, knives, cutting table surfaces, beef, hands, air, and water from four cattle slaughterhouses of Kerala, South India, to determine the occurrence and antibiotic susceptibility of *Listeria* species and *Staphylococcus aureus*. *Listeria* spp. were isolated from beef carcasses (2.0%), knives (3.7%), cutting table surfaces (1.9%), beef (0.7%), and water (1.3%). The identified species were *Listeria monocytogenes* (0.1%), *Listeria innocua* (0.9%), and *Listeria ivanovii* (0.4%). Most of the *Listeria* spp. were susceptible to majority of the antibiotics tested. The virulence genes were not detected in *Listeria* spp. However, all the *L. innocua* isolates were found to harbor the *iap* gene. The overall occurrence of *S. aureus* in slaughterhouses was 50.8%. The highest occurrence was observed on hands of abattoir workers (79.6%) and beef carcasses (59.9%). The isolates were commonly resistant to penicillin (38.0%), followed by ceftriaxone (31.9%), ampicillin (29.0%), amoxicillin (28.8%), tetracycline (24.4%), and chloramphenicol (23.9%). Overall, 53.0% of *S. aureus* isolates were resistant to three or more antibiotics. Vancomycin and methicillin resistance were observed in 8.5% and 5.4% of *S. aureus* isolates, respectively. Eight methicillin-resistant *S. aureus* isolates were found to harbor the *mec*A gene. In conclusion, *Listeria* spp. was only rarely found in the slaughterhouse environment and on beef. Nevertheless, the recovery of *L. monocytogenes* from a water reservoir containing sea water that was used to wash carcasses indicates the potential risk of contamination of the carcasses with *L. monocytogenes* when using sea water. *S. aureus* was frequently isolated from abattoir workers and beef carcasses, and the occurrence of *S. aureus* differed significantly between slaughterhouses. The high occurrence of *S. aureus*, which were often resistant toward different antibiotics, represents a significant public health concern.

Keywords: cattle slaughterhouses, *Listeria* spp., *Staphylococcus aureus*, antibiotic susceptibility

Introduction

Listeria species are ubiquitous bacteria and are widely distributed in environment, animals, and vegetables (Pesavento *et al.*, 2010; Dhama *et al.*, 2015). Most *Listeria* spp., are regarded as nonpathogenic, although *Listeria monocytogenes* causes severe infections (listeriosis) in humans and primarily affects young, old, pregnant, and immunocompromised people (Pesavento *et al.*, 2010). The ubiquitous nature of the bacteria and the ability to survive or grow under adverse conditions (such as a low pH and high salt concentrations) have made *Listeria* spp. an important concern to public health and the meat industry (Aznar and Alarcon, 2003).

Staphylococcus aureus is commonly found on the skin and nasal passages of most humans and animals (Jackson *et al.*, 2013). Many *S. aureus* strains produce enterotoxins and cause food intoxications or infections such as bacteremia and pneumonia (Jackson *et al.*, 2013). Infected animals and humans are a major source of contamination in the slaughterhouse, from where the bacteria may spread to processing facilities (Alarcon *et al.*, 2006). The organism can grow in a wide range of temperatures, pH, and salt concentrations, and it can adapt, survive, and colonize even on potentially dry and stressful environments, which may favor the growth of the organism in a slaughterhouse environment and in meat (Alarcon *et al.*, 2006; Jackson *et al.*, 2013).

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The increase in the incidence of antibiotic-resistant strains isolated from meat is an important public health concern worldwide (Dan *et al.*, 2015). For instance, methicillin-resistant *S. aureus* (MRSA) has emerged as a health concern and has been found in several meat-producing animal species, including cattle, pigs, and chickens (Bhargava *et al.*, 2011; Waters *et al.*, 2011; Jackson *et al.*, 2013). In developing countries like India, antimicrobial agents are extensively used for prophylaxis and to treat clinical and subclinical infections in animals, which has resulted in the emergence of resistant strains in food-producing animals.

Meat may get contaminated by a variety of sources, including the animal, dirty equipment, and meat handlers (Bhandare *et al.*, 2007). Despite several studies on the occurrence of these pathogens in meat, milk, and the environment (Dhanashree *et al.*, 2003; Kalorey *et al.*, 2005; Thaker *et al.*, 2013), there are no data on the occurrence of these pathogens in Indian cattle slaughterhouses. Since India is the largest exporter of beef, exporting over 2 million metric tons every year (Agricultural and processed food products export development authority [APEDA], 2015), more data regarding the occurrence and antibiotic resistance of *Listeria* spp. and *S. aureus* are needed to assess the potential risk to public health. Therefore, this study determined the occurrence and antibiotic susceptibility of *Listeria* spp. and *S. aureus* in cattle slaughterhouses of Kerala, South India.

Materials and Methods

Experimental design

This study was performed from August 2013 to January 2014, in three unorganized (I, II, and III) and one organized (IV) cattle slaughterhouse(s) located at different districts of Kerala state, South India. Unorganized slaughterhouses are traditional slaughterhouses with only basic slaughter equipment and without refrigeration units. All procedures (bleeding, dehiding, evisceration, and pluck removal) were carried out manually on the floor. Organized slaughterhouses are mechanized and automated slaughterhouses with proper infrastructure and trained manpower. The capacity of the organized slaughterhouse was 50 animals per day. Bleeding was carried out on the floor. Dehiding, evisceration, pluck removal, splitting, and washing of the carcasses were carried out hanging on the slaughter line. In all the slaughterhouses, all animals were culled cattle (the primary reasons for culling these cattle were reproduction failure, mastitis, and low production). Sampling was carried out on three sampling days per slaughterhouse.

Sample collection and preparation

Beef carcasses, knives, and cutting table surfaces were swabbed (100 cm^2 area) using sterile cotton swabs. For beef carcasses, 500 cm^2 area was swabbed (neck, brisket, loin, flank, and outer round region; 100 cm^2 area each) after evisceration. All swabs were transferred into 100 mL of 0.1% peptone water.

For beef samples, around 250 g of meat was collected from the neck, loin, brisket, outer round, and flank regions of the carcass after evisceration (slaughterhouse I, II, and III) or after deboning (slaughterhouse IV). A representative subsample of 25 g was used for further analysis. Hand samples were collected from randomly selected individuals involved in beef slaughter or deboning operations by collecting the rinse after washing with 100 mL of 0.1% peptone water. For air samples, duplicate Petri dishes containing Polymyxin-Acriflavin-Lithium chloride-Ceftazidime-Aesculin-Mannitol (PALCAM, Himedia, India) and Baird-Parker (BP, Himedia, India) agar were directly exposed in different processing rooms for 15 min (Evancho *et al.*, 2001). Water samples (250 mL) were collected from the main water tank that was used as processing water in the slaughterhouses. In slaughterhouse, I, II, and IV, fresh water was used as processing water, whereas slaughterhouse III used sea water. All the samples were transported to the laboratory under refrigerated conditions and analyzed within 2 h of collection.

Swab and water samples were thoroughly agitated with a cyclomixer (Remi labworld, India) at 8000 rpm for 3 min. Beef samples were homogenized in 225 mL of 0.1% peptone water using a stomacher blender (AES Chemiuex, France) for 2 min.

Isolation and identification of Listeria *spp.*

Twenty-five milliliter of homogenate was inoculated in 225 mL of University of Vermont broth (UVM I, Himedia, India) and incubated at 37°C for 24 h. One milliliter of the primary enrichment was transferred to 9 mL of UVM II and incubated at 37°C for 48 h (Ryser and Donelly, 2001). A loopful of enriched inoculum from UVM II was streaked on PALCAM agar, which was incubated at 37°C for 24 h. Typical colonies were confirmed by biochemical tests (Barrow and Feltham, 1993).

Isolation and identification of S. aureus

For the isolation of *S. aureus*, the remaining homogenate (75 mL for hand samples and 200 mL for beef samples) was incubated at 37°C for 24 h. After incubation, a loopful of enriched inoculum was streaked on BP agar and incubated at 37°C for 24–48 h. Typical colonies were identified up to species level using biochemical tests (Lancette and Benett, 2001).

Antibiotic susceptibility

Antimicrobial susceptibility was performed by the Kirby-Bauer disk diffusion method on Mueller-Hinton agar, incubated at 37°C for 24 h. After incubation, results were interpreted according to the guidelines of Clinical and Laboratory Standard Institute (CLSI, 2010).

Polymerase chain reaction assays

Single polymerase chain reaction assays were used to detect the presence of *plc*A, *prf*A, *act*A, *hly*A, and *iap* genes in *Listeria* spp. and the *mec*A gene in MRSA strains using specific primers adapted from Rawool *et al.* (2007) and Frey *et al.* (2013). Amplification reactions were performed in a 25-μL mixture, containing primers at a concentration of 40 pmol/μL for *plc*A and *prf*A, 30 pmol/μL for *act*A, 10 pmol/μL for *hly*A, 20 pmol/μL for *iap*, and 10 pmol/μL for *mec*A gene, 1 U of *Taq* polymerase, 1 × reaction buffer, 0.2 mM dNTP mix, 2 mM $MgCl_2$, and 2.5 μL of cell lysate.

The following conditions were used: initial denaturation at 95°C for 2 min, denaturation at 95°C for 15 s, primer

Table 1. Occurrence of *Listeria* spp. in Cattle Slaughterhouses of Kerala, South India

	Slaughterhouse I				*Slaughterhouse II*				*Slaughterhouse III*				*Slaughterhouse IV*			
Samples	N	*Spp. 1*	*Spp. 2*	*Spp. 3*	n	*Spp.1*	*Spp. 2*	*Spp. 3*	n	*Spp. 1*	*Spp. 2*	*Spp. 3*	n	*Spp. 1*	*Spp. 2*	*Spp. 3*
Beef carcass	31	0	0	0	30	0	1	0	33	0	1	0	58	0	1	0
Knife	20	0	1	0	30	0	2	0	24	0	0	0	35	0	0	1
Cutting table surface	22	0	1	1	30	0	0	0	26	0	0	0	26	0	0	0
Beef	31	0	0	1	30	0	0	0	33	0	0	0	58	0	0	0
Hands	30	0	0	0	25	0	0	0	21	0	0	0	32	0	0	0
Air	15	0	0	0	15	0	0	0	15	0	0	0	15	0	0	0
Water	22	0	0	0	25	0	0	0	18	1	0	0	15	0	0	0
Total	171	0	2	2	185	0	3	0	170	1	1	0	239	0	1	1

n, number of samples; Spp. 1, *Listeria monocytogenes*; Spp. 2, *Listeria innocua*; Spp. 3, *Listeria ivanovii.*

annealing at 60°C for 30 s, and extension at 72°C for 1 min (35 cycles of amplification), and a final extension at 72°C for 10 min. For *mec*A, primer annealing was done at 55°C. The amplified products were analyzed by gel electrophoresis with 2% agarose gel using ethidium bromide staining.

Statistical analysis

The occurrence of organisms between slaughterhouses was compared using logistic regression analyses, including the sampling visit as random effect when necessary. Bonferroni corrections were applied for multiple testing. To compare differences in resistance, the proportion of resistant isolates was compared between MRSA and methicillin-sensitive *S. aureus* (MSSA) strains for each of the antibiotics tested using a two-sample test of portions. All analyses were carried out in STATA 11 (StataCorp, 2011).

Results

Occurrence and antimicrobial susceptibilities of Listeria *spp.*

Overall, 1.4% (11/765) of the samples were positive for *Listeria* spp. (Table 1). *L. monocytogenes* was only isolated from one water sample. *Listeria innocua* was detected in all slaughterhouses, and was isolated from beef carcasses, knives, and cutting tables (Table 1). *Listeria ivanovii* was isolated from a knife, cutting table, and beef sample. All the *L. innocua* isolates harbored the *iap* gene, but other virulence genes were not detected. No virulence genes were detected in *L. monocytogenes* and *L. ivanovii*.

The resistance of *Listeria* spp. toward commonly used antibiotics is shown in Table 2. The *L. monocytogenes* isolate was susceptible to majority of the antibiotics, although was resistant to clindamycin and gentamicin. All *L. innocua* isolates ($n=7$) were sensitive to streptomycin and erythromycin. Resistance was observed toward ampicillin, amoxicillin, ceftazidime, and doxycycline. Most of the *L. ivanovii* isolates were susceptible to the antibiotics tested.

Occurrence and antimicrobial susceptibilities of S. aureus

The overall occurrence of *S. aureus* in slaughterhouses was 51% (389/765). The percentage of samples that was positive for *S. aureus* was higher in slaughterhouses I (70%) and III (62%) compared to slaughterhouses II (39%) and IV (38%) ($p<0.001$). Among the samples, the highest occurrence was

Table 2. Antibiotic Resistance of *Listeria* spp. Isolated from Cattle Slaughterhouses of Kerala, South India

	No. of isolates (%)								
	Listeria monocytogenes *(n=1)*			Listeria innocua *(n=7)*			Listeria ivanovii *(n=3)*		
Antibiotics	*S**	*I**	*R**	*S**	*I**	*R**	*S**	*I**	*R**
Amoxicillin	1 (100)	0	0	6 (86)	0	1 (14)	3 (100)	0	0
Ampicillin	1 (100)	0	0	4 (57)	2 (29)	1 (14)	2 (67)	0	1 (33)
Ceftazidime	1 (100)	0	0	5 (71)	1 (14)	1 (14)	2 (67)	0	0
Cefuroxime	1 (100)	0	0	5 (71)	2 (29)	0	3 (100)	0	0
Chloramphenicol	1 (100)	0	0	6 (86)	1 (14)	0	3 (100)	0	0
Clindamycin	0	0	1 (100)	5 (71)	2 (29)	0	3 (100)	0	0
Cotrimoxazole	0	1 (100)	0	3 (43)	4 (57)	0	2 (67)	1 (33)	0
Doxycycline	1 (100)	0	0	5 (71)	1 (14)	1 (14)	3 (100)	0	0
Erythromycin	0	1 (100)	0	7 (100)	0	0	3 (100)	0	0
Gentamicin	0	0	1 (100)	6 (86)	1 (14)	0	3 (100)	0	0
Kanamycin	0	1 (100)	0	5 (71)	2 (29)	0	2 (67)	1 (33)	0
Streptomycin	0	1 (100)	0	7 (100)	0	0	3 (100)	0	0

R*, resistant; I*, intermediate; S*, sensitive.

Table 3. Occurrence of *Staphylococcus aureus* in Cattle Slaughterhouses of Kerala, South India

	Slaughterhouse I				*Slaughterhouse II*				*Slaughterhouse III*				*Slaughterhouse IV*			
Sample	n	*MRSA*	*MSSA*	*Total*	n	*MRSA*	*MSSA*	*Total*	n	*MRSA*	*MSSA*	*Total*	n	*MRSA*	*MSSA*	*Total*
Beef carcasses	31	5	23	28	30	3	10	13	33	2	24	26	58	0	24	24
Knives	20	0	11	11	30	1	7	8	24	1	18	19	35	0	18	18
Cutting table surfaces	22	0	13	13	30	0	7	7	26	1	15	16	26	0	16	16
Beef	31	2	14	16	30	1	8	9	33	0	18	18	58	0	13	13
Hands	30	2	28	30	25	2	20	22	21	1	16	17	32	0	17	17
Air	15	0	5	5	15	0	2	2	15	0	2	2	15	0	2	2
Water	22	0	16	16	25	0	11	11	18	0	8	8	15	0	2	2
Total	171	9	110	119	185	7	65	72	170	5	101	106	239	0	92	92

n, number of samples; MRSA, methicillin-resistant *S. aureus*; MSSA, methicillin-susceptible *S. Aureus.*

found on hands (80%) and beef carcasses (60%) (Table 3). The proportion of *S. aureus*-positive beef carcasses was significantly higher in slaughterhouses I and III than in slaughterhouses II and IV ($p < 0.05$). *S. aureus* was less frequently isolated from cutting tables in slaughterhouse II (23%) than in slaughterhouses III and IV (62%; $p = 0.030$). In slaughterhouse I, *S. aureus* was isolated from 73% of water samples, which was significantly higher than the 13% of water samples in slaughterhouse IV ($p = 0.009$). *S. aureus* was isolated from all hand samples in slaughterhouse I, whereas in slaughterhouse IV, only 53.1% of the hands were *S. aureus* positive.

Among the 389 *S. aureus* isolates, 81.5% was sensitive to methicillin, followed by vancomycin (77.6%) and oxacillin (76.6%). The isolates exhibited resistance to penicillin (38.0%), ceftriaxone (31.9%), ampicillin (29.0%), amoxicillin (28.8%), tetracycline (24.4%), and chloramphenicol (23.9%) (Table 4). None of the isolates was simultaneously resistant to all the antibiotics. Overall, 53.0% of *S. aureus* strains were resistant to three or more of the antibiotics tested.

Out of the 389 *S. aureus* isolates, 21 (5.4%) were resistant to all β-lactam antibiotics, including methicillin and oxacillin (MRSA). The *mec*A gene was detected in eight of these MRSA isolates (38.1%). Most MRSA were recovered from beef carcasses (6.6%) and hands (4.6%). Out of the 91 and 86 *S. aureus* isolates that were recovered from beef carcasses and hands, 11.0% and 5.8% were MRSA, respectively. Although *S. aureus* was frequently detected in water samples, none of the isolates was MRSA. In slaughterhouse IV, only MSSA was recovered. Among the MRSA isolates, 28.6% and 19.0% were resistant to erythromycin and cotrimoxazole compared to 14.1% and 11.7% in MSSA isolates (Table 4; $p = 0.071$ and $p = 0.314$, respectively). For chloramphenicol, 9.5% of MRSA compared to 24.7% of MSSA strains showed resistance ($p = 0.112$). Resistance toward tetracycline, neomycin, and vancomycin was very similar in MRSA and MSSA strains ($p > 0.5$).

Discussion

The observed occurrence of *Listeria* spp. in this study was low (1.4%). This is in agreement with AjayKumar (2014) who reported a similar occurrence of *L. monocytogenes*, *L. innocua*, and *L. ivanovii* in cattle (0.6%), soil (0.6%), and water (0.4%) in the same region. In other parts of the world, a higher prevalence of these *Listeria* species in beef

Table 4. Antibiotic Resistance of *Staphylococcus aureus* Isolated from Cattle Slaughterhouses of Kerala, South India

	No. of isolates (%)								
	Methicillin-resistant S. aureus *(21 isolates)*			*Methicillin-susceptible* S. aureus *(368 isolates)*			*Total (389 isolates)*		
Antimicrobial agents	*Sensitive*	*Intermediate*	*Resistant*	*Sensitive*	*Intermediate*	*Resistant*	*Sensitive*	*Intermediate*	*Resistant*
Amoxicillin	0	0	21 (100)	176 (47.8)	101 (27.4)	91 (24.7)	176 (45.2)	101 (26.0)	112 (28.8)
Ampicillin	0	0	21 (100)	152 (41.3)	124 (33.7)	92 (25.0)	152 (39.1)	124 (31.9)	113 (29.0)
Ceftriaxone	0	0	21 (100)	191 (51.9)	74 (20.1)	103 (28.0)	191 (49.1)	74 (19.0)	124 (31.9)
Chloramphenicol	13 (62)	6 (29)	2 (10)	205 (55.7)	72 (19.6)	91 (24.7)	218 (56.0)	78 (20.1)	93 (23.9)
Cotrimoxazole	9 (43)	8 (38)	4 (19)	275 (74.7)	50 (13.6)	43 (11.7)	284 (73.0)	58 (14.9)	47 (12.1)
Erythromycin	6 (29)	9 (43)	6 (29)	235 (63.9)	81 (22.0)	52 (14.1)	241 (62.00)	90 (23.1)	58 (14.9)
Methicillin	0	0	21 (100)	317 (86.1)	51 (13.9)	0	317 (81.5)	51 (13.1)	21 (5.4)
Neomycin	4 (19)	11 (52)	6 (29)	191 (51.9)	91 (24.7)	86 (23.4)	195 (50.1)	102 (26.2)	92 (23.7)
Oxacillin	0	0	21 (100)	298 (81.0)	47 (12.8)	23 (6.3)	298 (76.6)	47 (12.1)	44 (11.3)
Penicillin	0	0	21 (100)	148 (40.2)	93 (25.3)	127 (34.5)	148 (38.0)	93 (23.9)	148 (38.0)
Tetracycline	5 (24)	11 (52)	5 (24)	174 (47.3)	104 (28.3)	90 (24.5)	179 (46.0)	115 (29.6)	95 (24.4)
Vancomycin	14 (67)	5 (24)	2 (10)	288 (78.3)	49 (13.3)	31 (8.4)	302 (77.6)	54 (13.9)	33 (8.5)

slaughterhouses and meat have been reported, in prevalences up to 78.4% (Barros *et al.*, 2007; Pesavento *et al.*, 2010). Besides the variation in prevalence rates due to the difference in sampling and isolation methods, management practices, and environmental factors (Nightingale *et al.*, 2004; Zhu *et al.*, 2012), the lower occurrence of *Listeria* spp. in this study and other studies in the same region may be related to the high ambient temperature, which might not have provided an opportunity for *L. monocytogenes* to compete with other bacteria (Wu *et al.*, 2015). Nevertheless, the isolation of *L. monocytogenes* from the water tank in slaughterhouse III indicates the potential risk of cross contamination of carcasses when using sea water. *L. monocytogenes* may contaminate sea water due to sewage and farming effluents and may survive in sea water due to its halophilic nature.

This investigation revealed a high occurrence of *S. aureus* in cattle slaughterhouses of Kerala, South India. This may be due to poor hygiene and sanitation and excessive handling of the carcasses. In unorganized slaughterhouses, animals were slaughtered on the floor and all the process were carried out on the floor. The position of workers was changing in these slaughterhouses and a single person was carrying out all the procedures. The organized slaughterhouse was automated and mechanized. However, the slaughter line was not automated and carcasses were moved by hand. In slaughterhouse IV, *S. aureus* was observed on 41.4% of beef carcasses, which indicates that the hygienic conditions maintained in slaughterhouse IV were also unsatisfactory Other studies have shown a much lower occurrence of *S. aureus* on beef carcasses as Phillips *et al.* (2001) and Schlegelova *et al.* (2004) reported only 24.3% and 7.5% *S. aureus*-positive beef carcasses in Australia and the Czech Republic, respectively. In slaughterhouse I, II, and III, dressing was carried out on the floor and standard hygienic practices were not performed. With over 90% of carcasses being *S. aureus* positive, the occurrence of *S. aureus* was significantly higher in slaughterhouse I compared to the other slaughterhouses, which may be related to the high proportion of *S. aureus*-positive water samples (73%). Slaughterhouses I, II, and IV were supplied with water from a nearby river, which might have been contaminated due to animal or human activities (Lechevallier and Seidler, 1980). This water may thus represent an important source of contamination of meat and the slaughterhouse environment as it is used to wash carcasses, knives, hands, and the slaughterhouse environment.

The lower occurrence of *S. aureus* on hands of the abattoir workers in the organized slaughterhouse (IV) may be related to better training and resulting hand hygiene practices compared to other slaughterhouses.

In this study, cutting table surfaces and knives represented an important source of (cross) contamination since over 50% of knife and table samples in slaughterhouses I, III, and IV were *S. aureus* positive. The relatively low occurrence of *S. aureus* on knives and cutting tables in slaughterhouse II could be related to the better cleaning and disinfection practices after termination of the slaughtering activities compared to other slaughterhouses, and warrant a thorough cleaning of slaughterhouses and equipment after completion of the operations. In slaughterhouse IV, sanitation of knives was carried out by steam sterilization. However, changing of knives after manipulation of each carcass was not strictly followed. This might have affected the contamination process during slaughtering.

Indiscriminate use of antibiotics in cattle results in the development of resistant strains and dissemination of resistance genes within the bacterial population (Lipsitch *et al.*, 2002). In this investigation, *L. monocytogenes* showed sensitivity to most of the antibiotics. However, intermediate resistance was noticed toward cotrimoxazole, which is of concern for public health because of its second-line use in case of allergy to β-lactams (Caplan *et al.*, 2014). Although *Listeria* spp. are generally reported as susceptible to antibiotics active against Gram-positive bacteria (Charpentier and Courvalin, 1999), *L. innocua* isolates showed variable susceptibility patterns, which is in line with a general worldwide pattern of an increasing prevalence of antibiotic resistance.

In this study, none of the antibiotics had shown 100% sensitivity against all the *S. aureus* isolates and 53% of *S. aureus* isolates were resistant to three or more antibiotics tested. This is in close agreement with Alian *et al.* (2012) who reported 82.6% of *S. aureus* isolates resistant to one or more antimicrobial agents in raw meat in Iran. Contrarily, a low occurrence (4%) of multidrug-resistant *S. aureus* was reported in beef products in Georgia (Jackson *et al.*, 2013). The World Health Organization (WHO) has recommended the prohibition of chloramphenicol in all food-producing animals, particularly in lactating cows (WHO, 2012). The resistance of *S. aureus* isolates toward chloramphenicol in this study (23.9%) was much higher than the 0.5% reported from retail ground meats in the United States (Kelman *et al.*, 2011). The proportion of resistance toward erythromycin was also higher in this study, whereas resistance toward penicillin was very similar in both studies. In contrast, the proportion of isolates that were resistant toward tetracycline was much lower in this study (69% compared to 24%) (Kelman *et al.*, 2011). The variation in the resistance pattern may be expected considering the geographical location and differences in restrictions on the use of antibiotics in food animals.

Conclusion

The results of this study indicate that *Listeria* spp. is not common in cattle slaughterhouses, whereas *S. aureus* is widespread in cattle slaughterhouses in Kerala, South India. There were some differences in the rate of bacterial contamination between the slaughterhouses, with a lower occurrence of *S. aureus* on beef carcasses and beef in slaughterhouse II and IV compared to other slaughterhouses. The high occurrence of *S. aureus* in processing water in slaughterhouses indicates that water may be an important source of contamination of meat and the slaughter environment. Most MRSA were recovered from beef carcasses and hands of the abattoir workers. Transmission of MRSA might have occurred between animals and humans, which addresses the importance of avoiding cross contamination in the slaughterhouse. The isolates displayed varying degrees of resistance to the antibiotics tested and none of the antibiotics showed 100% sensitivity against all the *S. aureus* isolates. Vancomycin, methicillin, and multiple-drug resistant *S. aureus* were also noticed and warrant a regular monitoring of animals and abattoir workers for the occurrence of drug-resistant bacteria. These results provide useful information to assess the possible

risk associated with the consumption of contaminated beef produced in these slaughterhouses.

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Disclosure Statement

No competing financial interests exist.

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