African Journal of Microbiology Research Vol. 7(4), pp. 309-316, 22 January, 2013
Available online at http://www.academicjournals.org/AJMR
DOI: 10.5897/AJMR12.930
ISSN 1996 0808 ©2013 Academic Journals

Full Length Research Paper

Frequency of antibiotic resistant *Salmonella*, *Escherichia coli, Enterococcus,* and *Staphylococcus aureus* in meat in Saudi Arabia

Kay Greeson[1], Gamaleldin M. Suliman[2], Ahmed Sami[2], Abdullah Alowaimer[2] and Mohammad Koohmaraie[1,2]*

[1]IEH Laboratories and Consulting Group, 15300 Bothell Way NE, Lake Forest Park, Seattle, WA 98155, USA.
[2]College of Food and Agricultural Sciences, King Saud University, P. O. Box 2460, Riyadh 11451Saudi Arabia.

Accepted 24 January, 2013

This study was undertaken to estimate the prevalence of antibiotic resistance in *Salmonella* spp., *Escherichia coli*, *Enterococcus* spp. and *Staphylococcus aureus* in meat in Saudi Arabia. Samples of domestic and imported meat (beef, camel, lamb) and poultry were purchased from local retail outlets in Riyadh area. There was some contamination from each of the bacteria in all types of meat analyzed, with *E. coli* being the most prevalent overall at 72.2%, *Enterococcus* prevalence was 26.2%, *S. aureus* prevalence was 24.6% and *Salmonella* prevalence was 10.7%. Additionally, these bacteria were resistant to a number of antibiotics and some were multidrug resistant. *S. aureus* and *Enterococcus* were both either resistant or intermediate to Erythromycin (79 and 86%, respectively). *E. coli* was resistant to Ampicillin (44%). *Salmonella* was resistant to Ceftiofur (67%). Bacterial contamination of meat is a multi-country problem and consideration should be made to improve methods of decontaminating food animals and work surfaces during meat processing to reduce the levels of bacteria that are transferred to the finished product. This will also help to decrease the growing crisis of bacterial antibiotic resistance.

Key words: Foodborne pathogens, antibiotic resistance, food animals, meat.

INTRODUCTION

Increasingly, reports of bacterial contamination in meat destined for human consumption are appearing worldwide, and frequently this contamination is responsible for illness. Noteworthy foodborne outbreaks have occurred from *Salmonella* spp. (Roels et al., 1997; Urfer et al., 2000.), *Escherichia coli* (Barrett et al., 1994; CDC, 2002) and *Staphylococcus aureus* (Breckinridge and Bergdoll, 1971; CDC, 1986), resulting in serious disease in many cases. *Enterococcus* spp. is a commensal bacteria found in the intestinal tracts of humans and other mammals that is responsible for food spoilage (Bell and De Lacy, 1984), and is a common cause of nosocomial infections and outbreaks (Borgmann et al., 2004; Montecalvo et al., 1994). *Enterococcus* spp. are also known to transfer resistance traits to and from other bacteria (Simjee and Gill, 1997; Valdivia et al., 1996). A wide range of animals serve as a reservoir for these bacteria including cattle, sheep, pigs, and chickens. Meat can become contaminated at any of several stages of processing. In ruminants, bacterial transfer from hide to carcass is perhaps the most significant step in the process whereby pathogens are transferred from exposed surfaces to an otherwise sterile carcass during harvest (Koohmaraie et al., 2005). For poultry, overcrowding and contaminated food and water pose a risk of transfer of pathogens. Previous reports have demonstrated the presence of bacteria not only on the food animal (Hussein et al., 2003; Smith et al., 2009), but also associated with the finished product available

*Corresponding author. E-mail: mk@iehinc.com. Tel: 206-940-3334. Fax: 206-260-7922.

commercially (Bosilevac et al., 2009; Samadpour et al., 1994).

Moreover, many of these bacteria have become resistant to antimicrobials while some are multi-drug resistant (MDR) (Dechet et al., 2006; Graham et al., 2009; Kunze et al., 2008; Smith et al., 2009). The development of resistance to drugs could be due to general use of antibiotics for therapeutic and subtherapeutic treatments of food animals, as well as in humans. Additionally, antibiotic resistance can be passed from one bacterial strain to another by gene transfer (Simjee and Gill, 1997). It follows that antibiotic resistance could be conferred from commensals to pathogenic bacteria when both types are present on food animals. Reports suggest that infections with MDR bacteria are responsible for more serious disease than susceptible bacteria(Holmberg et al., 1984; Martin et al., 2004).

In this study, a preliminary survey of several types of meat, both locally produced and imported available for sale were undertaken in the Riyadh area of Saudi Arabia. The samples were assayed for the presence of four genera of bacteria that can be pathogenic for humans. We then performed antimicrobial susceptibility testing on the isolated bacteria.

MATERIALS AND METHODS

Meat collection

Between September 2009 and January 2010, a total number of 288 unprocessed meat samples of four different types (beef, camel, lamb and poultry) were purchased to be used in this study. They were divided into domestic chilled (144) and imported frozen (144) meat samples. They were collected from four different retail outlets including local markets and large international hypermarkets based in Riyadh to ensure representation of different producing companies. The retail outlets are located in different areas of Riyadh city; North, East, South and West. The selection of samples was performed twice a week. With the exception of camel meat (all produced locally), the same number of samples of each type of meat was either produced locally or was imported from Brazil and Pakistan (beef), Pakistan and New Zealand (lamb) or France (chicken).

Initial culturing and enrichment

One hundred to two hundred grams of various cuts of beef, camel or lamb meat, and 700 to 900 g of whole chicken were rinsed in sterile Whirl-Pak stomacher-400 bags (Nasco, Modesto, CA, USA) with Lactose broth (Accumedia, Lansing, MI, USA), massaged and then the entire contents of the bag was incubated at 36±1°C for 18 to 24 h as a pre-enrichment. Then, several techniques were used to isolate individual bacteria as follows:

Salmonella

One ml of the cultured Lactose broth was inoculated into Selenite Cysteine broth (Becton Dickinson, Sparks, MD, USA) and incubated for 18 to 24 h at 36±°C as enrichment. Isolation of colonies from the Selenite broth was completed separately on Brilliant Green Bile agar (Accumedia, Lansing, MI, USA), Salmonella-Shigella agar (Becton Dickinson, Sparks, MD, USA) and Xylose Lysine Deoxycholate agar (Accumedia, Lansing, MI, USA) by subculturing 13 mm loopful of broth onto the plates. Typical isolated colonies from each were then inoculated and streaked for isolation onto Triple Sugar Iron slanted agar (Accumedia, Lansing, MI, USA) at 36±1°C for 18 to 24 h. Tubes with a red slant and a black or yellow butt were kept as presumptive *Salmonella*.

Fecal coliforms

One ml of cultured Lactose broth was inoculated into Brilliant Green Bile broth (Accumedia, Lansing, MI, USA) containing inverted Durham tubes and incubated in a 44.5°C water bath for 18 to 24 h. Eosin Methylene Blue agar (Oxoid Ltd, Basingstoke, Hampshire, England) was inoculated with 13 mm loopful of broth for isolation of small metallic colonies, which were kept as presumptive coliforms.

Enterococcus

Ten microliters of cultured Lactose broth was inoculated onto plates containing Bile Aesculin agar (Oxoid Ltd., Basingstoke, Hampshire, England) and incubated as above. Typical colonies were inoculated onto Trypticase Soy slanted agar (Accumedia, Lansing, MI, USA) and kept as presumptive enterococci.

Staphylococcus

Ten microliters of cultured lactose broth was inoculated onto Baird Parker agar (Difco, Detroit, MI, USA) and black colonies surrounded by a zone of clearing were kept as presumptive staphylococci.

Confirmation

Following initial isolation confirmation of each type of bacteria was done as follows:

Salmonella

The API 20E system (bioMérieux, Marcy L'Etoile, France) was used for confirmation following manufacturers instructions.

Fecal coliforms

Colonies were inoculated onto EC medium (Accumedia, Lansing, MI, USA) and incubated for 24 hours in a 44.5°C water bath.

Enterococcus

Gram stain and catalase test were used to confirm, using protocols from *Clinical Microbiology Procedures Handbook*, edited by Henry D. Isenberg, 2nd edition, 2004.

Staphylococcus

Coagulase test, following the protocol of Vanderzant and Splittstoesser (Vanderzant, 2005), was used to confirm. Briefly, suspected *S. aureus* colonies were transferred to small tubes

Table 1. Number of confirmed positive isolates for each type of bacteria from various meat sources (n%).

Meat type	*Salmonella* spp.	*E. coli*	*Enterococcus* spp.	*S. aureus*
Camel	11 (30.6)	21 (58.3)	10 (27.8)	2 (5.6)
Beef				
local	3 (8.3)	21 (58.3)	3 (8.3)	8 (22.2)
imported	2 (5.6)	32 (88.9)	8 (22.2)	4 (11.1)
Lamb				
local	2 (5.6)	32 (88.9)	5 (13.9)	15 (41.7)
imported	2 (5.6)	26 (72.2)	18 (50.0)	3 (8.3)
Poultry				
local	5 (13.9)	24 (66.7)	9 (25.0)	13 (36.1)
imported	2 (5.6)	26 (72.2)	13 (36.1)	17 (47.2)
Total	27 (10.7)	182 (72.2)	66 (26.2)	62 (24.6)

containing 2 ml Brain Heart Infusion broth (Becton Dickinson, Sparks, MD, USA) These were thoroughly emulsified and then 13 mm loopful of the culture suspension was transferred to Tryptic Soy broth or agar slants (Accumedia, Lansing, MI, USA) and incubated for 18 to 24 h at 35°C. The slant cultures were kept at room temperature for ancillary or repeat testing in case the coagulase test results were questionable. Then 0.5 ml Coagulase Plasma with EDTA (Becton Dickinson BBL, Sparks, MD, USA) was added to 0.2 ml of each broth culture in a 10 × 75 mm tube and mixed thoroughly. These were incubated at 35 to 37°C and examined periodically during a 6 h interval for clot formation.

Antibiotic susceptibility testing

The Kirby-Bauer method was used to identify antibiotic resistance of bacteria. Cultures of confirmed isolates were spread onto Mueller Hinton agar and antibiotic disks (Oxoid Ltd. Basingstoke, Hampshire, England) were placed on the agar. For the Gram negative bacteria (*Salmonella* spp. and *E. coli*) susceptibility was assessed for Amoxicillin, Chloramphenicol, Kanamycin, Ampicillin, Cefoxitin and Ceftiofur. For the Gram positive bacteria (*Enterococcus* spp. and *S. aureus*) susceptibility was assessed for Chloramphenicol, Ciprofloxacin, Linezolid, Erythromycin and Nitrofurantoin. Also, *Enterococcus* was tested for Vancomycin susceptibility. Zones of inhibition were measured and then susceptible, intermediate or resistant was reported for each based on comparison with standard values (CLSI M100-S20: Performance Standards for Antimicrobial Susceptibility Testing).

RESULTS

Samples of meat produced locally in Saudi Arabia or imported were purchased and tested for the presence of bacteria by confirmatory culturing and specific biochemical tests. Thirty six samples of meat from each type of animal were included in this study. The numbers and proportions of bacteria found in each type of meat are shown in Table 1. *E. coli* was the most frequent contaminant of all types of meat from 58.3% (for camel and local beef) to 88.9% (for imported beef and local lamb). *Enterococcus* and *S. aureus* had similar levels of contamination at 26.2 and 24.6% overall, respectively. Interestingly, imported beef, lamb and poultry more frequently harbored *Enterococcus* than did their locally produced versions. Conversely, local beef (27.2%) and lamb (41.7%) contained more *S. aureus* than the imported versions (11.1 and 8.3%, respectively). The prevalence of *Salmonella* was relatively low, 10.7%, overall, but 30.6% of the camel meat was positive for this pathogen. Imported poultry harbored more *E. coli*, *Enterococcus* and *S. aureus* (72.2, 47.2 and 47.2%, respectively) than locally produced poultry (66.7, 25.0 and 36.1%, respectively) but less *Salmonella* (5.6 versus 13.9%).

We performed disk diffusion assays for identification of antibiotic resistance on the bacteria isolated from the meat samples and found that among the four bacteria analyzed, there was some level of resistance to every antibiotic screened (Tables 2 through 5). The level of resistance to a particular antibiotic generally did not vary much by the type of meat in which it was found. However, the different types of bacteria were more or less resistant to certain antibiotics across meat types. *S. aureus* and *Enterococcus* were both either resistant or intermediate to Ciprofloxacin (58% for both) and Erythromycin (79 and 86%, respectively). *E. coli* had a high level of resistance to Ampicillin (44%); however this percentage was much lower in imported beef (9%). *Salmonella* was frequently resistant to Ceftiofur (67%).

DISCUSSION

Previous reports from Saudi Arabia indicate contamination

Table 2. Antibiotic resistance profiles of *Salmonella* spp. Isolated from meat sold in Riyadh and imported.

Source	AMC30mcg	C30mcg	K30mcg	AMP25 mcg	FOX 30 mcg	CXM 30mcg	Resistant to 2 or more
Camel	3 (2)/11	3 (1)/11	4 (5)/11	4/11	3 (1)/11	8 (3)/11	5/11 (45%)
Beef							
Local	1/3	0/3	(3)/3	0/3	0/3	2 (1)/3	0/3 (0%)
Imported	1 (1)/2	(1)/2	(1)/2	2/2	0/2	(2)/2	1/2 (50%)
Lamb							
Local	0/2	0/2	(1)/2	1/2	0/2	1 (1)/2	1/2 (50%)
Imported	1/2	0/2	0/2	0/2	(1)/2	1/2	1/2 (50%)
Poultry							
Local	1 (1)/5	(1)/5	(2)/5	1 (1)/5	0/5	4 (1)/5	2/5 (40%)
Imported	(1)/2	0/2	(1)/2	1/2	0/2	2/2	1/2 (50%)
Total resistant (intermediate)	7 (5)/27	3 (3)/27	4 (13)/27	9 (1)/27	3 (2)/27	18 (8)/27	11/27 (41%)
Percent resistant (intermediate)	26 (18)	11 (11)	15 (48)	33 (4)	11 (7)	67 (30)	

Number of resistant (intermediate) divided by total number of bacteria isolated. AMC, Amoxicillin; C, Chloramphenicol; K, Kanamycin; AMP, Ampicillin; FOX, Cefoxitin; CXM, Ceftiofur.

Table 3. Antibiotic resistance profiles of *Escherichia coli* Isolated from meat sold in Riyadh and imported.

Source	AMC30mcg	C30mcg	K30mcg	AMP25 mcg	FOX30 mcg	CXM30mcg	Resistant to 2 or more
Camel	3 (2)/21	1/21	3 (1)/21	14/21	2 (1)/21	2 (3)/21	5/21 (24%)
Beef							
Local	0 (1)/21	1 (1)/21	12/21	9/21	3/21	0 (5)/21	8/21 (38%)
Imported	1/32	4 (2)/32	6/32	3/32	1/32	1 (2)/32	4/32 (12%)
Lamb							
Local	6/32	2 (2)/32	5/32	16/32	3 (1)/32	0 (5)/32	9/32 (28%)
Imported	1/26	0/26	6 (1)/26	6 (1)/26	1/26	0(4)/26	5/26 (19%)

Table 3. Continued.

Poultry							
Local	2 (2)/24	2 (2)/24	17/24	17/24	4 (3)/24	4 (5)/24	7/24 (29%)
Imported	0 (1)/26	1/26	15 (1)/26	15 (1)/26	0 (1)/26	3 (1)/26	4/26 (15%)
Total resistant (intermediate)	13 (6)/182	11 (7)/182	64 (3)/182	80 (2)/182	14 (6)/182	10 (25)/182	42/182 (23%)
Percent resistant (intermediate)	**7 (3)**	**6 (4)**	**35 (2)**	**44 (1)**	**8 (3)**	**5 (14)**	

Number of resistant (intermediate) divided by total number of bacteria isolated. AMC, Amoxicillin; C, Chloramphenicol; K, Kanamycin; AMP, Ampicillin; FOX, Cefoxitin; CXM, Ceftiofur.

Table 4. Antibiotic resistance profiles of *Enterococcus* spp. Isolated from meat sold in Riyadh and imported.

Source	C30mcg	CIP5mcg	LZD30mcg	VA30 mcg	E15 mcg	F300mcg	Resistant to 2 or more
Camel	3 (2)/10	2 (6)/10	1 (2)/10	0 (2)/10	7 (2)/10	0(2)/10	4/10 (40%)
Beef							
Local	0/3	0/3	2/3	2 (1)/3	1 (3)/3	1/3	2/3 (67%)
Imported	0 (1)/8	2 (1)/8	2/8	1 (3)/8	1 (4)/8	0 (1)/8	2/8 (25%)
Lamb							
Local	0/5	0 (3)/5	3/5	2/5	2 (2)/5	0 (2)/5	2/5 (40%)
Imported	3 (5)/18	4 (7)/18	12 (1)/18	5 (4)/18	11 (6)/18	4 (1)/18	13/18 (72%)
Poultry							
Local	1 (2)/9	4 (1)/9	4 (1)/9	4 (1)/9	4 (5)/9	1/9	6/9 (67%)
Imported	0 (4)/13	3 (5)/13	6/13	5 (3)/13	5 (5)/13	1 (2)/13	6/13 (46%)
Total resistant (intermediate)	7 (14)/66	15 (23)/66	30 (4)/66	19 (14)/66	31 (27)/66	7 (8)/66	36/66 (54%)
Percent resistant (intermediate)	10 (21)	23 (35)	45 (6)	29 (21)	47 (41)	11 (12)	

Number of resistant (intermediate) divided by total number of bacteria isolated. C, Chloramphenicol; CIP,Ciprofloxacin; LZD, Linezolid; VA, Vancomycin; E, Erythromycin; F, Nitrofurantoin.

by various bacteria, fungi and parasites of meat and environments where food and animals are raised (Bin Saeed et al., 2005; Nabbut et al., 1982). Additionally, bacterial resistance to multiple antimicrobials is adding to the problem of meat contamination from animal hides, feces, environments

Table 5. Antibiotic resistance profiles of *Staphylococcus aureus* Isolated from meat sold in Riyadh and imported.

Source	C 30mcg	CIP 5mcg	LZD 30mcg	E 15 mcg	F 300mcg	Resistant to 2 or more
Camel	1/2	2/2	1 (1)/2	2/2	1 (1)/2	2/2 (100%)
Beef						
Local	2 (2)/8	4 (1)/8	3 (1)/8	2 (5)/8	0 (6)/8	4/8 (50%)
Imported	0 (1)/4	1 (1)/4	0 (2)/4	1 (3)/4	1 (2)/4	1/4 (25%)
Lamb						
Local	1 (1)/15	15 (3)/15	1 (3)/15	1 (13)/15	3 (10)/15	6/15 (40%)
Imported	0/3	0 (1)/3	0 (1)/3	0 (2)/3	0/3	0/3 (0%)
Poultry						
Local	2 (1)/13	2 (9)/13	1 (2)/13	3 (8)/13	3 (5)/13	4/13 (31%)
Imported	1 (2)/17	2 (5)/17	1 (2)/17	3 (7)/17	2 (1)/17	4/17 (24%)
Total resistant (intermediate)	7 (7)/62	16 (20)/62	7 (12)/62	12 (36)/62	10 (25)/62	21/62 (34%)
Percent resistant (intermediate)	11 (11)	26 (32)	11 (19)	19 (58)	16 (40)	

Number of resistant (intermediate) divided by total number of bacteria isolated. C, Chloramphenicol; CIP,Ciprofloxacin; LZD, Linezolid; E, Erythromycin; F, Nitrofurantoin.

(Alexander et al., 2008; Graham et al., 2009; Smith et al., 2009), and resistance is occurring against the newer antibiotics. The purpose of this study was to determine the prevalence of four types of bacteria in meat commercially available in Riyadh, the largest city and capital of Saudi Arabia, and the level of antibiotic resistance associated with each bacteria.

In our study, *E. coli* prevalence was 72.2% of all meat sources. Although we did not specifically test pathogenic strains of *E. coli*, the fact that the prevalence was so high, leads to the supposition that some of these could be of the serotypes containing toxin producing genes, such as those for Shiga toxin and intimin. A previous report demonstrated that 28% of cattle used for food in the US shed *E. coli* of the O157 serotype responsible for most of the enterohemorrhagic illness outbreaks there (Elder et al., 2000). Nevertheless, the presence of any *E. coli*, as well as *Enterococcus*, in meat indicates fecal contamination of the meat. Many types of *E. coli* and *Enterococcus* are normal flora in the intestines of animals, and can contaminate the hides and carcasses of food animals during the slaughtering process. These bacteria can then be transferred to the meat. In Saudi Arabia, Nassar and coworkers found that treatment with radiation reduced the numbers of *Salmonella* on chicken carcasses (Nassar et al., 1997). Our data suggest that these methods may not be implemented on a large scale. Additionally, some contamination can occur through dirty equipment or improper meat handling. Interestingly we found a higher prevalence of *Enterococcus* spp. on imported meat of any kind than from locally produced meat. Conversely, prevalence of *S. aureus* was higher for locally produced beef and lamb.

Contaminated meat with antibiotic resistant bacteria were identified and in some cases more than one type of bacteria was present in a single sample. The level of resistance was high and many bacteria were resistant to more than one antibiotic. Although at least some of these bacteria may not be pathogenic strains themselves, they are a cause of concern because they can extend antibiotic resistance to other bacteria. *Enterococcus* spp. are environmental and commensal gut bacteria that, although not known to cause illness through food, are a common cause of nosocomial infections and can be very difficult to treat if they are antibiotic resistant (Sood et al., 2008). Moreover, they are proficient at transferring resistance traits to and from other bacteria (Simjee and Gill, 1997; Valdivia et al., 1996), and their co-presence in meat with other bacteria gives them a great opportunity to do that. Previous studies have demonstrated *in vitro* (Weigel et al., 2003) and *in vivo* (Jacobsen et al. 1999; Schwalbe et al., 1990) transfer of antibiotic resistance from one bacteria to another, as well as from an *Enterococcus* to a *Staphylococcus* (Noble et al., 1992). *Salmonella* (Doublet et al., 2005), *E. coli* (Zhao et al., 2001) and *S. aureus* (Maiques et al., 2006) are also capable of this type of genetic transfer.

Alexander and colleagues found an initial 40% tetracycline resistant *E. coli* population in cattle that increased following subtherapeutic treatments (Alexander et al., 2008). da Costa and coworkers, likewise, saw an increase in antibiotic resistance in *E. coli* and *Enterococcus* spp. from chickens after treatment versus those that did not receive antibiotics (da Costa et al., 2009).

When these bacteria are present in the food supply they have the potential to infect humans. Antibiotic resistant, but not susceptible *E. coli* from human infections in the United States were found to be indistinguishable from those in chickens suggesting a directional transmission of resistant bacteria from a food source (Weese et al., 2010). Additionally, Methicillin resistant *S. aureus* was found in pigs and the people who work with them (Smith et al., 2009). Reports suggest antibiotics are also being used inappropriately. For instance in Saudi Arabia, tetracycline residue was found in chickens and eggs for human consumption exceeding permissible limits (Al-Ghamdi et al., 2000). Bacterial antibiotic resistance can be selected by other means, such as human misuse of antibiotics. Antibiotic resistant *Salmonella* was identified in food poisoning patients in Saudi Arabia, and molecular characterization of the resistance determinants suggests inappropriate use of antibiotics (Halawani and Shohayeb, 2008).

A study characterized resistance patterns in *Campylobacter* from humans and meat and determined sources other than meat could also be responsible for the human infections (Thakur et al., 2010). Recently, MDR *E. coli* was found in drinking water in Pakistan that could have been mixed with sewage from both human and animal sources (Patoli et al., 2010). Once these bacteria enter the human gastrointestinal tract they could transfer their antibiotic resistance traits to both normal flora and other pathogenic bacteria that also happen to be present. Jacobsen et al. (1999) demonstrated transfer of antibiotic resistance between strains of *E. faecium* in gnotobiotic rats. In another study, Schwalbe et al. (1990) discovered identical resistance patterns in *Salmonella typhi*, *E. coli* and *Klebsiella pneumoniae* that recovered from a human stool sample following therapeutic antibiotic treatment.

The current work was a preliminary screening study of the presence and antibiotic resistance profile of four types of bacteria in various types of meat sold commercially in Riyadh. Our study identified four types of bacteria contained in packaged retail meat and our data demonstrate widespread resistance of *Enterococcus* spp. and *S. aureus* to Erythromycin, and *E. coli* to Ampicillin. Many of the isolated bacteria displayed multidrug resistance. This may translate into increased resistance in bacteria infecting humans, which in turn could transfer resistance genes to both commensals and pathogenic bacteria in the human gut, and then potentially be passed along by person to person or fomite contact. It is recommende that future studies be more precisely focused on whether these bacteria found in meat are pathogenic, by serotyping or identifying toxins in the *E. coli*, *Salmonella* and *S. aureus*, and speciating the *Enterococcus*. Also the finding of lower prevalence of *Enterococcus* and higher prevalence of *S. aureus* in locally produced lamb can be further explored by increasing sample size and perhaps an examination of the butchering process and environments. Additionally, antibiotic resistance patterns can be examined to determine whether there has been gene transfer between bacteria present on the same food source.

Conclusion

A proportion of packaged meat sold commercially contained potentially pathogenic bacteria and many of these bacteria were antibiotic resistant. This contamination came from both domestically produced and imported meat of various kinds. More care should be taken to clean and disinfect meat during all stages of production, as these procedures are shown to be effective at reducing the levels of bacteria on the finished meat product, thereby reducing the risk of infections with antibiotic resistant bacteria. Also consideration should be given to reducing or eliminating general therapeutic and subtherapeutic antibiotic treatments in food animals in order to decrease resistance in bacteria present in these animals. This will reduce the spread of antibiotic resistance from normal flora to pathogenic bacteria when they are present together.

REFERENCES

Alexander TW, Yanke LJ, Topp E, Olson ME, Read RR, Morck DW, McAllister TA (2008). Effect of subtherapeutic administration of antibiotics on the prevalence of antibiotic-resistant *Escherichia coli* bacteria in feedlot cattle. Appl. Environ. Microbiol. 74:4405-4416.

Al-Ghamdi M S, Al-Mustafa Z H, El-Morsy F, Al-Faky A, Haider I, Essa H (2000). Residues of tetracycline compounds in poultry products in the eastern province of Saudi Arabia. Public Health 114:300-304.

Barrett TJ, Lior H, Green JH, Khakhria RJ, Wells JG, Bell BP, Greene KD, Lewis J, Griffin PM (1994). Laboratory investigation of a multistate food-borne outbreak of *Escherichia coli* O157:H7 by using pulsed-field gel electrophoresis and phage typing. J. Clin. Microbiol. 32:3013-3017.

Bell RG, De Lacy KM (1984). Heat injury and recovery of *Streptococcus faecium* associated with the souring of chub-packed luncheon meat. J. Appl. Bacteriol. 57:229-236.

Bin Saeed AA, Al-Hamdan NA, Fontaine RE (2005). Plague from eating raw camel liver. Emerg. Infect. Dis. 11:1456-1457.

Borgmann S, Niklas DM, Klare I, Zabel LT, Buchenau P, Autenrieth IB, Heeg P (2004). Two episodes of vancomycin-resistant *Enterococcus faecium* outbreaks caused by two genetically different clones in a newborn intensive care unit. Int. J. Hyg. Environ. Health 207:386-389.

Bosilevac JM, Guerini MN, Kalchayanand N, Koohmaraie M (2009). Prevalence and characterization of salmonellae in commercial ground beef in the United States. Appl. Environ. Microbiol. 75:1892-1900.

Breckinridge JC, Bergdoll MS (1971). Outbreak of food-borne gastroenteritis due to a coagulase-negative enterotoxin-producing staphylococcus. N. Engl. J Med. 284:541-543.

CDC (1986). Staphylococcal food poisoning from turkey at a country club buffet--New Mexico. MMWR Morb. Mortal Wkly. Rep. 35:715-716, 721-722.

CDC (2002). Multistate outbreak of *Escherichia coli* O157:H7 infections associated with eating ground beef--United States, June-July 2002. MMWR Morb. Mortal Wkly. Rep. 51:637-639.

da Costa PM, Belo A, Goncalves J, Bernardo F (2009). Field trial

evaluating changes in prevalence and patterns of antimicrobial resistance among *Escherichia coli* and *Enterococcus* spp. isolated from growing broilers medicated with enrofloxacin, apramycin and amoxicillin. Vet. Microbiol. 139:284-292.
Dechet AM, Scallan E, Gensheimer K, Hoekstra R, Gunderman-King J, Lockett J, Wrigley D, Chege W, Sobel J (2006). Outbreak of multidrug-resistant *Salmonella enterica* serotype Typhimurium Definitive Type 104 infection linked to commercial ground beef, northeastern United States, 2003-2004. Clin. Infect. Dis. 42:747-752.
Doublet B, Boyd D, Mulvey MR, Cloeckaert A (2005). The Salmonella genomic island 1 is an integrative mobilizable element. Mol. Microbiol. 55:1911-1924.
Elder RO, Keen JE, Siragusa GR, Barkocy-Gallagher G A, Koohmaraie M, Laegreid WW (2000). Correlation of enterohemorrhagic *Escherichia coli* O157 prevalence in feces, hides, and carcasses of beef cattle during processing. Proc. Natl. Acad. Sci. USA. 97:2999-3003.
Graham JP, Price LB, Evans SL, Graczyk TK, Silbergeld EK (2009). Antibiotic resistant enterococci and staphylococci isolated from flies collected near confined poultry feeding operations. Sci. Total Environ. 407:2701-2710.
Halawani E, Shohyeb M (2008). Molecular Characterization of Multiple Antibiotic Resistance in *Salmonella enterica* Serovar Typhimurium and Enteritidis Isolated in Saudi Arabia. World J. Med. Sci. 3:65-70.
Holmberg SD, Wells JG, Cohen ML (1984). Animal-to-man transmission of antimicrobial-resistant Salmonella: investigations of U.S. outbreaks, 1971-1983. Science 225:833-835.
Hussein HS, Thran BH, Hall MR, Kvasnicka WG, Torell RC (2003). Verotoxin-producing Escherichia coli in culled beef cows grazing rangeland forages. Exp. Biol. Med. (Maywood). 228:352-357.
Jacobsen BL, Skou M, Hammerum AM, Jensen LB (1999). Horizontal Transfer of the satA Gene Encoding Streptogramin A Resistance Between Isogenic *Enterococcus faecium* Strains in the Gastrointestinal Tract of Gnotobiotic Rats. Microb. Ecol. Health Dis. 11:241-247.
Koohmaraie M, Arthur TM, Bosilevac JM, Guerini M, Shackelford SD, Wheeler TL (2005). Post-harvest interventions to reduce/eliminate pathogens in beef. Meat Sci. 71:79-91.
Kunze DJ, Loneragan GH, Platt TM, Miller MF, Besser TE, Koohmaraie M, Stephens T, Brashears MM (2008). *Salmonella enterica* burden in harvest-ready cattle populations from the southern high plains of the United States. Appl. Environ. Microbiol. 74:345-351.
Maiques E, Ubeda C, Campoy S, Salvador N, Lasa I, Novick R P, Barbe J, Penades J R (2006). beta-lactam antibiotics induce the SOS response and horizontal transfer of virulence factors in *Staphylococcus aureus.* J. Bacteriol. 188:2726-2729.
Martin LJ, Fyfe M, Dore K, Buxton JA, Pollari F, Henry B, Middleton D, Ahmed R, Jamieson F, Ciebin B, McEwen SA, Wilson JB (2004). Increased burden of illness associated with antimicrobial-resistant *Salmonella enterica* serotype typhimurium infections. J. Infect. Dis. 189:377-384.
Montecalvo MA, Horowitz H, Gedris C, Carbonaro C, Tenover FC, Issah A, Cook P, Wormser GP (1994). Outbreak of vancomycin-, ampicillin-, and aminoglycoside-resistant *Enterococcus faecium* bacteremia in an adult oncology unit. Antimicrob. Agents Chemother. 38:1363-1367.
Nabbut NH, Barbour E K, Al-Nakhli HM (1982). Salmonella species and serotypes isolated from farm animals, animal feed, sewage, and sludge in Saudi Arabia. Bull. World Health Organ. 60:803-807.
Nassar TJ, Al-Mashhadi AS, Fawal AK, Shalhat AF (1997). Decontamination of chicken carcasses artificially contaminated with Salmonella. Rev. Sci. Tech. 16:891-897.
Noble WC, Virani Z, Cree RG (1992). Co-transfer of vancomycin and other resistance genes from *Enterococcus faecalis* NCTC 12201 to *Staphylococcus aureus.* FEMS Microbiol. Lett. 72:195-198.
Roels TH, Frazak PA, Kazmierczak JJ, Mackenzie WR, Proctor ME, Kurzynski TA, Davis JP (1997). Incomplete sanitation of a meat grinder and ingestion of raw ground beef: contributing factors to a large outbreak of *Salmonella typhimurium* infection. Epidemiol. Infect. 119:127-134.
Samadpour M, Ongerth JE, Liston J, Tran N, Nguyen D, Whittam TS, Wilson RA, Tarr PI (1994). Occurrence of Shiga-like toxin-producing *Escherichia coli* in retail fresh seafood, beef, lamb, pork, and poultry from grocery stores in Seattle, Washington. Appl. Environ. Microbiol. 60:1038-1040.
Schwalbe RS, Hoge CW, Morris JG, O'Hanlon PN, Crawford RA, Gilligan PH (1990). *In vivo* selection for transmissible drug resistance in Salmonella typhi during antimicrobial therapy. Antimicrob. Agents Chemother. 34:161-163.
Simjee S, Gill MJ (1997). Gene transfer, gentamicin resistance and enterococci. J. Hosp. Infect. 36:249-259.
Smith TC, Male MJ, Harper AL, Kroeger JS, Tinkler GP, Moritz ED, Capuano AW, Herwaldt LA, Diekema DJ (2009). Methicillin-resistant *Staphylococcus aureus* (MRSA) strain ST398 is present in midwestern U.S. swine and swine workers. PLoS One 4: e4258.
Sood S, Malhotra M, Das BK, Kapil A (2008). Enterococcal infections and antimicrobial resistance. Indian J. Med. Res. 128:111-121.
Thakur S, Zhao S, McDermott PF, Harbottle H, Abbott J, English L, Gebreyes WA, White DG (2010). Antimicrobial resistance, virulence, and genotypic profile comparison of *Campylobacter jejuni* and *Campylobacter coli* isolated from humans and retail meats. Foodborne Pathog. Dis. 7:835-844.
Urfer E, Rossier P, Mean F, Krending MJ, Burnens A, Bille J, Francioli P, Zwahlen A (2000). Outbreak of *Salmonella braenderup* gastroenteritis due to contaminated meat pies: clinical and molecular epidemiology. Clin. Microbiol. Infect. 6:536-542.
Valdivia E, Martin-Sanchez I, Quirantes R, Martinez-Bueno M, Galvez A, Maqueda M (1996). Incidence of antibiotic resistance and sex pheromone response among enterococci isolated from clinical human samples and from municipal waste water. J. Appl. Bacteriol. 81:538-544.
Vanderzant C (2005). Compendium of Methods for the Microbiological Examination of Foods, ed. A.T.C.o.M.M.f. Foods, American Public Health Association.
Weese JS, Reid-Smith R, Rousseau J, Avery B (2010). Methicillin-resistant Staphylococcus aureus (MRSA) contamination of retail pork. Can. Vet. J. 51:749-752.
Weigel LM, Clewell DB, Gill SR, Clark NC, McDougal LK, Flannagan SE, Kolonay JF, Shetty J, Killgore GE, Tenover FC (2003). Genetic analysis of a high-level vancomycin-resistant isolate of *Staphylococcus aureus.* Science 302:1569-1571.
Zhao S, White DG, Ge B, Ayers S, Friedman S, English L, Wagner D, Gaines S, Meng J (2001). Identification and characterization of integron-mediated antibiotic resistance among Shiga toxin-producing *Escherichia coli* isolates. Appl. Environ. Microbiol. 67:1558-1564.