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ORIGINAL PAPERS

Antibiotic resistance of *Escherichia coli* O157:H7 isolated from cattle and sheep

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Abstract A total of 102 *Escherichia coli* O157:H7 colonies recovered from 11 cattle and 14 sheep were collected and tested for their antibiotic resistance profiles using a disc diffusion method, according to the Clinical and Laboratory Standards Institute. Four (36.36 %) of the 11 cattle *E. coli* O157:H7 isolates were resistant to cephalothin, one (9.09 %) isolate was resistant to streptomycin, and one (9.09 %) to nalidixic acid. Two (14.28 %) of the 14 sheep *E. coli* O157:H7 isolates were resistant to sulphamethoxazole, one (7.14 %) isolate was resistant to sulphonamide compounds, and one (7.14 %) to streptomycin. All cattle and sheep isolates were found to be susceptible to cephazolin, gentamicin, ciprofloxacin, imipenem, trimethoprim/ sulphamethoxazole, chloramphenicol, trimethoprim, and ceftiofur. Six cattle isolates were susceptible at a ratio of 54.54 %, and 11 (78.57 %) isolates from sheep were susceptible to all 20 antibiotics tested. As an overall result, 68 % of the *E. coli* O157:H7 isolates belonging to cattle and sheep were susceptible to all antibiotics tested. On the other hand, most of the *E. coli* O157:H7 isolates were intermediately resistant to streptomycin, cephalothin, sulphamethoxazole, ampicillin, and kanamycin.

Keywords *E. coli* O157:H7 · Antibiotic resistance · Cattle · Sheep

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Introduction

Escherichia coli O157:H7 is considered one of the most important food-borne pathogens among shiga toxin-producing *E. coli* (STEC) strains. It causes diarrhea that may result in life-threatening conditions ranging from hemorrhagic colitis (HC) to hemolytic-uremic syndrome (HUS) (Mead et al. 1999; Meng et al. 2001). Gastrointestinal tracts of ruminants especially cattle and sheep have been shown to act as a reservoir of *E. coli* O157:H7 (Kudva et al. 1996, 1997; Shere et al. 1998). Epidemiological investigations have clearly associated *E. coli* O157:H7 human infections to the consumption of contaminated raw or undercooked ground beef and products with feces during slaughterhouse processing (Beutin et al. 1993; Paiba et al. 2002).

Antimicrobial resistance of food-borne bacteria should not necessarily be considered distinct from that in isolates from humans, food animals, or other niches. When food animals, as carriers of asymptomatic *E. coli* O157:H7, are exposed to antimicrobial agents, they may become the reservoir of this antimicrobial-resistant bacteria. So it becomes important to determine whether the bacteria develop resistance to antimicrobials during food animal production. Also, recent studies have reported that there has been a rise in the antimicrobial resistance patterns of *E. coli* O157:H7 (Galland et al. 2001; Schroeder et al. 2002, 2004). It is controversial to use antibiotic treatment in humans to prevent HUS due to lysis of the bacteria and increased releasing of the expression of the shiga toxins in the intestinal tract (Takahashi et al. 1997; Wong et al. 2000). However, it is has been reported that using some antimicrobials in the early stage of infection may be protective against HUS progression (Fukushima et al. 1999; Ikeda et al. 1999).

The objective of this study was to evaluate the antibiotic resistance profile to 20 different antibiotics of *E. coli* O157:H7 isolates collected from cattle and sheep at slaughter for at least a 1-year period. As this is the first comprehensive study to evaluate the antibiotic resistance profile of *E. coli* O157:H7 in Turkey, it was possible to acquire epidemiological data for the future analysis.

Materials and methods

Bacterial strains

Escherichia coli ATCC 25922 and *Staphylococcus aureus* ATCC 25923 were used for antibiotic susceptibility testing. *E. coli* O157:H7 ATCC 43895 was used as positive controls for the detection of $fliC_{h7}$ gene in PCR assay.

Isolates

A total of 102 *E. coli* O157:H7 isolates recovered from 25 animals including 11 cattle and 14 sheep were analyzed. The 41 isolates from the 11 cattle were obtained from 282 cattle (207 beef and 75 dairy cattle) feces and/or colon tissue samples between April 2002 and December 2003, and 61 isolates from 14 sheep were obtained from 218 sheep feces and/or colon tissue samples between November 2007 and November 2008 (with a total of 500 animals) (Table 1).

IMS based cultural technique for the isolation of *E. coli* O157:H7

Immunomagnetic separation (IMS)-based selective enrichment technique was used for the isolation of *E. coli* O157 (Byrne et al. 2003). Briefly, 10 g of feces and/or colon tissue samples of cattle and sheep were weighed in a sterile bag and enriched with 90 ml EC broth (Oxoid CM0853, Hampshire, UK) containing novobiocin (20 µg/l; Sigma N-1628, St. Louis, MO, USA) and incubated at 37°C at 100 rpm for 18 h in a shaking incubator (Bellco Shel Lab Shaking Incubator S16R, Oregon, USA). After the incubation period, IMS was performed according to the manufacturer's protocol. Then, 20 µl of magnetic beads coated with specific antibody against *E. coli* O157 (Dynabeads anti *E. coli* O157, Prod. No. 710.04; Dynal, Oslo, Norway) were used. Following the IMS procedure, 100 µl of PBS-Tween 20 was added to resuspend the bead and *E. coli* O157 complex.

Resuspended IMS mixture was plated on Sorbitol MacConkey Agar (Oxoid CM0813) plates containing Cefixime-tellurite supplement (Oxoid SR0172). After overnight incubation at 42°C, sorbitol negative colonies were tested for the O157 antigen by latex agglutination (Oxoid DR0620), and up to five agglutination positive colonies were taken and stored at −80°C (Sanyo MDF-U5186S, Japan) in cryovials for PCR analysis and antibiotic resistance determination.

DNA extraction and PCR analysis for detection of $fliC_{h7}$ gene

DNA extraction was performed using Chelex-100 (Bio-Rad, Hercules, CA, USA) resin-based technique as reported previously (Goncuoglu et al. 2010).

The PCR was performed with FLICH7-F and FLICH7-R primers in a final volume of 50 µl containing 1X Reaction Buffer (Promega, Madison, WI, USA), 1.5 mM $MgCl_2$ (Promega), 400 µM each of the four deoxynucleoside triphosphates (Promega), 2.5 U *Taq* DNA polymerase (Promega), 0.50 µM of primers (Promega) and 10 µl DNA. Thermal cycling (Biometra Personel Cycler, Göttingen, Germany) was carried out with the initial denaturation at 94°C for 2 min and then 35 cycles of denaturation at 94°C for 20 s, annealing at 54°C for 1 min, and extension at 72°C for 1 min, with a final extension for 10 min at 72°C (Fratamico et al. 2000). A 10-µl aliquot of each PCR product was subjected to 1.5 % agarose gel (SeaKem® LE Agarose, Rockland, ME, USA) electrophoresis containing 0.1 µg/ml ethidium bromide for 1 h at 100 V. Amplicon visualization and documentation was performed using gel documentation and analysis system (Syngene Ingenius, Cambridge, UK).

Antibiotic susceptibility testing

The antibiotic susceptibility test of *E. coli* O157:H7 isolates was performed with the disc diffusion method as recommended by the Clinical and Laboratory Standards Institute (CLSI) (Anon 2006) in Mueller-Hinton agar (Oxoid CM0337) with 20 different antibiotics and antibiotic combinations at given concentrations (Oxoid) as shown in Table 2.

Escherichia coli O157:H7 isolates were transferred to BHI broth (Oxoid CM0225) and incubated at 37°C for 24 h. A loopfull of growth was transferred into 5 ml containing Tryptone Soya Broth (Oxoid CM0129). The culture was incubated at 37°C until it achieved the turbidity of the 0.5 McFarland standard. The correct density of the turbidity standard was measured by NanoDrop Spectrophotometer (NanoDrop ND-100, Delaware, USA). The absorbance of the broth cultures at 625 nm was fixed to 0.08–0.10. The suspension was inoculated uniformly to Mueller Hinton agar (uniform depth of 4 mm) with a sterile cotton swab. Then, antibiotic discs were placed onto the plate and incubated at 37°C for 18 h. According to the sizes of the inhibition zones, interpreta-

Table 1 Sampling design and antibiotic resistance profiles of *E. coli* O157:H7 isolates from cattle and sheep samples

Sampling date	Source of isolates	No. of isolates	Code of isolates	Resistance to:
16.05.2002	Cattle (male/enteritis)	5	1001–1005	KF[a]
01.10.2002	Catlle (female/healthy)	5	1005–1010	S[a]
01.10.2002	Catlle (female/healthy)	5	1011–1015	
01.10.2002	Catlle (female/healthy)	4	1016–1019	KF[a]
01.10.2002	Catlle (female/healthy)	2	1020–1021	
24.07.2003	Cattle (male/healthy)	5	1022–1026	
24.07.2003	Catlle (male/healthy-colon tissue)	5	1027–1031	
24.07.2003	Cattle (male/healthy)	5	1032–1036	
24.07.2003	Cattle (male/healthy)	1	1037	
24.07.2003	Cattle (male/healthy)	1	1038	KF[a], NA[a]
24.07.2003	Cattle (male/healthy)	3	1039–1041	KF[a]
10.12.2007	Sheep (healthy)	5	1042–1046	
18.04.2008	Sheep (healthy)	5	1047–1051	
18.04.2008	Sheep (healthy)	5	1052–1056	
16.05.2008	Sheep (healthy)	5	1057–1061	
17.07.2008	Sheep (healthy)	2	1062–1063	
17.07.2008	Sheep (healthy)	4	1064–1067	RL[a]
23.07.2008	Sheep (healthy)	5	1068–1072	
29.08.2008	Sheep (healthy)	5	1073–1077	
29.08.2008	Sheep (healthy)	4	1078–1081	
29.08.2008	Sheep (healthy)	4	1082–1085	S3[a]
29.08.2008	Sheep (healthy)	4	1086–1089	
29.08.2008	Sheep (healthy)	5	1090–1094	
29.08.2008	Sheep (healthy)	5	1095–1099	S[a], RL[a]
29.08.2008	Sheep (healthy)	3	1100–1102	

[a] *KF* Cephalothin, *S3* Sulphonamide compounds, *S* Streptomycin, *NA* Nalidixic acid, *RL* Sulphamethoxazole

tion of the strains as susceptible, intermediate, or resistant was made according to the CLSI (Anon 2006).

Results and discussion

All the *E. coli* O157:H7 colonies (up to five colonies belonging to a positive sample) isolated from the same positive animals showed the same antibiotic resistance/susceptible profiles (Tables 1 and 2). Therefore, the results are given according to the number of the positive animals, e.g., 11 cattle and 14 sheep, instead of the total number (102) of the isolates tested in this study (Table 2). All isolates except one were recovered from healthy animals. Four of the 11 cattle isolates were recovered from cows. Nevertheless, no antibiotic resistance profile differences were seen between male and female host samples, one of the animals even showed enteritis.

Overall, among the cattle isolates, 36.36 % (4/11) of them were resistant to cephalothin, 9.09 % (1/11) were resistant to streptomycin and 9.09 % (1/11) were resistant to nalidixic acid. In one (9.09 %) *E. coli* O157:H7 cattle isolate, multiple drug resistance was shown to cephalothin and nalidixic acid. Also, ten isolates (90.90 %) were intermediately resistant to streptomycin, eight (72.72 %) were intermediately resistant to kanamycin, seven (63.63 %) were intermediately resistant to sulphamethoxazole, cephalothin, and ampicillin. In addition, five (45.45 %) isolates were intermediately resistant to amikacin, three (27.27 %) were intermediately resistant to sulphonamide compounds, and two different isolates (9.09 %) were intermediately resistant to amoxicillin/clavulanic acid and cefoxitin, respectively.

Among those isolates recovered from sheep, two (14.28 %) were resistant to sulphamethoxazole, one (7.14 %) to streptomycin, and one (7.14 %) to sulphonomide compounds. In one (7.14 %) *E. coli* O157:H7 sheep isolate, multiple drug resistance was shown to streptomycin and sulphonomide compounds. From total sheep isolates, eleven (78.57 %) of them were intermediately resistant to streptomycin, ten (71.42 %) were intermediately resistant to cephalothin, seven (50.00 %) were intermediately resistant to sulphamethoxazole, and five (35.71 %) were intermediately resistant to ampicillin. For all that, three (21.42 %) isolates were intermediately resistant to tetracycline and nalidixic acid, respectively. In addition, two (14.28 %) isolates were intermediately resistant to amikacin and kanamycin, respectively, and one (7.14 %) was intermediately resistant to ceftriaxone.

Table 2 Antibiotic resistance profiles of *E. coli* O157:H7 isolates from cattle and sheep feces and/or colon tissue samples

Number of *E. coli* O157:H7 isolates (%)

	Cattle (n = 11)			Sheep (n = 14)		
Antibiotic (μg disc^{-1})	R[a]	I[a]	S[a]	R[a]	I[a]	S[a]
Ampicillin (AMP-10)		7 (63.63)	4 (36.36)		5 (35.71)	9 (64.28)
Cephazolin (KZ-30)			11 (100)			14 (100)
Cephalothin (KF-30)	4 (36.36)	7 (63.63)			10 (71.42)	4 (28.57)
Gentamicin (CN-120)			11(100)			14 (100)
Amikacin (AK-30)		5 (45.45)	6 (54.54)		2 (14.28)	12 (85.71)
Amoxicillin/clavulanic acid (AMC-30)		1 (9.09)	10 (90.90)			14 (100)
Cefoxitin (FOX-30)		1 (9.09)	10 (90.90)			14 (100)
Ceftriaxone (CRO-30)			11 (100)		1 (7.14)	13 (92.85)
Ciprofloxacin (CIP-5)			11 (100)			14 (100)
Imipenem (IPM-10)			11 (100)			14 (100)
Trimethoprim/sulphamethoxazole (SXT-25)			11 (100)			14 (100)
Chloramphenicol (C-30)			11 (100)			14 (100)
Kanamycin (K-30)		8 (72.72)	3 (27.27)		2 (14.28)	12 (85.71)
Tetracycline (TE-30)			11 (100)		3 (21.42)	11 (78.57)
Trimethoprim (W-5)			11 (100)			14 (100)
Sulphonamide compounds (S3-300)		3 (27.27)	8 (72.72)	1 (7.14)		13 (92.85)
Ceftiofur (EFT-30)			11 (100)			14 (100)
Streptomycin (S-10)	1 (9.09)	10 (90.90)		1 (7.14)	11 (78.57)	2 (14.28)
Nalidixic acid (NA-30)	1 (9.09)		10 (90.90)		3 (21.42)	11 (78.57)
Sulphamethoxazole (RL-25)		7 (63.63)	4 (36.36)	2 (14.28)	7 (50.00)	5 (35.71)

[a] *R* Resistant, *I* Intermediately resistant, *S* Susceptible

All isolates from cattle and sheep were susceptible to cephazolin, gentamicin, ciprofloxacin, imipenem, trimethoprim/sulphamethoxazole, chloramphenicol, trimethoprim, and ceftiofur. Among those isolates recovered from cattle, six (54.54 %) were susceptible to all 20 antibiotics tested. On the other hand, these susceptible isolates were all intermediately resistant to cephalothin and streptomycin. Regarding sheep isolates, eleven (78.57 %) isolates were susceptible to all tested antibiotics. Similar to the cattle isolates, most of the susceptible sheep isolates (ten, 71.42 %) were intermediately resistant to streptomycin.

Similar to the previous studies, in this study, susceptibility was amore common character instead of showing resistance to tested antibiotics (Meng et al. 1998; Zhao et al. 2001; Schroeder et al. 2002; Walsh et al. 2006). In fact, no multiresistant character was observed in all the isolates tested but, in agreement with Vali et al. (2004), a low prevalence of resistance to cephalothin, sulphamethoxazole, streptomycin, sulphonamide compounds, and nalidixic acid, was detected. Also, cephalothin and nalidixic acid resistance ratios were similar to Khan et al.'s (2002) findings. All isolates were susceptible to tetracycline used commonly on cattle and sheep feedlots as well as to chloramphenicol. Scott et al. (2006) showed that *E. coli* O157:H7 strains isolated from cattle feedlots were susceptible to 13 tested antibiotics. In particular, different studies (Kim et al. 1994; Galland et al. 2001; Schroeder et al. 2002; Wilkerson et al. 2004) have evidenced that only from 6.63 to 38.46% of *E. coli* O157:H7 isolates from bovines were resistant to tetracycline. Various countries including Turkey have banned the usage of chloramphenicol for food-producing animals, so this result is good data for chloramphenicol resistance. Sheep isolates were more susceptible to antibiotics than cattle isolates. This difference may be explained by the slaughtering age of the animals that reach up to 1 year old for sheep and more than 1 year old for cattle. Moreover, since intensive sheep breeding does not occur generally in Turkey, the antibiotics are not widely used on farms for animal prophylaxis, particularly for sheep.

Although ciprofloxacin, aminoglycosides and trimethoprim/sulphamethoxazole are used as therapeutics in human medicine, our isolates showed high susceptibility rates, except streptomycin and kanamycin with high intermediate resistant patterns. This variation in results may be due to attaining resistance from other organisms by genetic

linkage of genetic factors, or by being from different origin, or these resistance patterns are the natural characteristic of the bacteria.

Although some of the isolates were recovered on the same slaughter day, the antibiotic resistance patterns of the isolates from different animals were quite distinct (Table 1). The antibiotic resistance profiles of four cattle isolates, which were coded as 1005 to 1021, were all different from each other. One of the isolates was resistant to streptomycin and another to cephalothin. On the other hand, two isolates showed no resistance to any antibiotic tested; also, all the four isolates were intermediately resistant to different antibiotics. Similarly, among the sheep isolates coded as 1073–1102, the antibiotic resistance profiles did not have the same pattern. Also, the intermediate resistance profiles of the isolates were all different from each other; in isolates 1078–1081, the only intermediate resistance was to streptomycin, but isolates 1100 to 1102 were intermediately resistant to nine different antibiotics.

According to the previous studies conducted worldwide on this subject, most isolates that are resistant to antibiotics are widely used on farms in food animals, more commonly used as prophylactics and therapeutics, but our results were unforeseeable. In the present study, most of the isolates showed an intermediate resistant profile which is a clue for potential resistance risk that may occur in the future. Therefore, attention has to be paid to high intermediate resistant profiles of these antibiotics.

This is the first report of antibiotic resistance patterns of *E. coli* O157:H7 strains isolated from cattle and sheep in Turkey. It is concluded that the overall prevalence of antibiotic resistance of *E. coli* O157:H7 isolates recovered from cattle and sheep tested in this study is very low. However, longitudinal studies should be performed to monitor and detect any changing in antibiotic resistance profiles of this bacterium in the future.

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References

Anon (2006). Clinical and Laboratory Standards Institute. Performance standards for antimicrobial susceptibility testing; sixteenth informational supplement. M100-S16 Vol.26 No.3.

Beutin L, Geier D, Steinruck H, Zimmermann S, Scheuts F (1993) Prevalence and some properties of verotoxin (shiga-like toxin)-producing *Escherichia coli* in seven different species of healthy domestic animals. J Clin Microbiol 31:2483–2488

Byrne C, Erol I, Call J, Buege D, Kaspar CW, Hiemke C, Fedorka-Cray P, Hermosillo J, Ball T, Wallace M, Handy M, Luchansky JB (2003) Characterization of *Escherichia coli* O157:H7 from downer and healthy dairy cattle in the upper Midwest region of the United States. Appl Environ Microbiol 69:4683–4688

Fratamico PM, Bagi LK, Pepe T (2000) A multiplex polymerase chain reaction assay for rapid detection and identification of *Escherichia coli* O157:H7 in foods and bovine feces. J Food Prot 63:1032–1037

Fukushima H, Hashizume T, Morita Y, Tanaka J, Azuma K, Mizumoto Y, Kaneno M, Matsuura M, Konma K, Kitani T (1999) Clinical experiences in Sakai City Hospital during the massive outbreak of enterohemorrhagic *Escherichia coli* O157 infections in Sakai City. Pediatr Int 41:213–217

Galland JC, Hyatt DR, Crupper SS, Acheson DW (2001) Prevalence, antibiotic susceptibility and diversity of *Escherichia coli* O157:H7 isolates from a longitudinal study of beef cattle feedlots. Appl Environ Microbiol 67:1619–1627

Goncuoglu M, Erol I, Ayaz ND, Bilir Ormanci FS, Kaspar CW (2010) Isolation and genomic characterization of *Escherichia coli* O157:H7 in bile of cattle. Ann Microbiol 60:293–297

Ikeda K, Ida O, Kimoto K, Takatorige T, Nakanishi N, Tatara K (1999) Effect of early fosfomycin treatment on prevention of hemolytic uremic syndrome accompanying *Escherichia coli* O157:H7 infection. Clin Nephrol 52:357–362

Khan A, Das SC, Ramamurthy T, Sikdar A, Khanam J, Yamasaki S, Takeda Y, Nair GB (2002) Antibiotic resistance, virulence gene, and molecular profiles of shiga toxin-producing *Escherichia coli* isolates from diverse sources in Calcutta. India J Clin Microbiol 40:2009–2015

Kim H, Samadpour M, Grimm L, Clausen C, Besser T, Baylor M, Kobayashi J, Neill ML, Schoenknecht F, Tarr P (1994) Characteristics of antibiotic-resistance *Escherichia coli* O157:H7 in Washington State, 1984–1991. J Infect Dis 170:1606–1609

Kudva IT, Hatfield PG, Hovde CJ (1996) *Escherichia coli* O157:H7 in microbial flora of sheep. J Clin Microbiol 34:431–433

Kudva IT, Hatfield PG, Hovde CJ (1997) Characterization of *Escherichia coli* O157:H7 and other shiga-toxin producing *E. coli* serotypes isolated from sheep. J Clin Microbiol 35:892–899

Mead PS, Slutsker L, Dietz V, McCaig LF, Bresee JS, Shapiro C, Griffin PM, Tauxe RV (1999) Food-related illness and death in the United States. Emerg Infect Dis 5:607–625

Meng J, Zhao S, Doyle MP, Joseph SW (1998) Antibiotic resistance of *Escherichia coli* O157:H7 and O157:NM isolated from animals, food, and humans. J Food Prot 61:1511–514

Meng J, Doyle MP, Zhao T, Zhao S (2001) Enterohemorrhagic *Escherichia coli* O157:H7. In: Doyle MP, Beuchat LR, Montville TJ (eds) Food microbiology: fundamentals and frontiers, 2nd edn. ASM Press, Washington, D.C., pp 193–213

Paiba GA, Gibbens JC, Pascoe SJS, Wilesmith JW, Kidd SA, Byrne C, Ryan JBM, Smith RP, McLaren IM, Futter RJ, Kay ACS, Jones YE, Chappel SA, Willshaw GA, Cheasty T (2002) Faecal carriage of verocytotoxin-producing *Escherichia coli* O157:H7 in cattle and sheep at slaughter in Great Britain. Vet Rec 150:593–598

Scott L, McGee P, Minihan D, Sheridan JJ, Earley B, Leonard N (2006) The characterization of *E. coli* O157:H7 isolates from cattle faeces and feedlot environment using PFGE. Vet Microbiol 114:331–336

Schroeder CM, Zhao C, DebRoy C, Torcolini J, Zhao S, White DG, Wagner DD, McDermott PF, Walker RD, Meng J (2002) Antimicrobial resistance of *Escherichia coli* O157 isolated from humans, cattle, swine and food. Appl Environ Microbiol 68:576–581

Schroeder CM, White DG, Meng J (2004) Retail meat and poultry as a reservoir of antimicrobial-resistant *Escherichia coli*. Food Microbiol 21:249–255

Shere JA, Barlett KJ, Kaspar CW (1998) Longitudinal study of *Escherichia coli* O157:H7 dissemination on four dairy farms in Wisconsin. Appl Environ Microbiol 64:1390–1399

Takahashi K, Narita K, Kato Y, Sugiyama T, Koide N, Yoshida T, Yokochi T (1997) Low-level release of Shiga-like toxin (Verocytotoxin) and endotoxin from enterohemorrhagic *E. coli* treated with imipenem. Antimicrob Agents Chemother 41:2295–2296

Vali L, Wisely KA, Pearce MC, Turner EJ, Knight HI, Smith AW, Amyes SGB (2004) High-level genotypic variation and antibiotic sensitivity among *Escherichia coli* O157 strains isolated from two Scottish beef cattle farms. Appl Environ Microbiol 70:5947–5954

Walsh C, Duffy G, Mahony RO, Fanning S, Blair IS, McDowell DA (2006) Antimicrobial resistance in Irish isolates of verocytotoxigenic *Escherichia coli* (*E. coli*)-VTEC. Int J Food Microbiol 109:173–178

Wilkerson C, Samadpour M, van Kirk N, Roberts MC (2004) Antibiotic resistance and distribution of tetracycline resistance genes in *Escherichia coli* O157:H7 isolates from humans and bovines. Antimicrob Agents Chemother 48:1066–1067

Wong CS, Jelacic S, Habeeb RL, Watkins SL, Tarr PI (2000) The risk of hemolytic-uremic syndrome after antibiotic treatment of *Escherichia coli* O157:H7 infections. N Eng J Med 342:1930–1936

Zhao S, White DG, Ge B, Ayers S, Freidman 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