

## Antibiotic resistance phenotypes of *Salmonella* isolates of broiler meat and chicken origin

Seran TEMELLI<sup>1</sup>, Serpil KAHYA<sup>2</sup>, Aysegul EYIGOR<sup>1</sup>, Kamil Tayfun CARLI<sup>2</sup>

<sup>1</sup> Department of Food Hygiene and Technology, and <sup>2</sup>Department of Microbiology, Uludag University, Faculty of Veterinary Medicine, Bursa, Turkey.

**Summary:** This study aims to determine the antibiotic resistance phenotypes of *Salmonella* isolates of broiler meat and chicken samples against a total of 23 antimicrobial agents. For this, 64 meat and 79 chicken *Salmonella* isolates, which were obtained from retail meat and farm samples analyzed in our laboratory from February 2006 to January 2009, were used. All *Salmonella* isolates were resistant to at least 4 antimicrobials tested, with the common highest resistance rates to erythromycin (100.0%), bacitracin (99.3%), and rifampicin (98.6%), regardless of the sample type. Additionally, meat isolates were 100.0% resistant to bacitracin, clindamycin, erythromycin, rifampicin, and chicken isolates were 100.0% resistant to erythromycin, spiramycin, penicillin G, doxycycline hydrochloride, and tetracycline. Four to 7 Multi Drug Resistant (MDR) *Salmonella* conformed 82.83% of all meat isolates, and 10, 12, 15, and 17 MDR *Salmonella* conformed 55.68% of all chicken isolates analyzed. The B/DA/E/RD and N/B/CT/E/SP/P/RD/DO/OT/TE resistances were the most common patterns as 35.94% and 13.92% within meat and chicken isolates, respectively. The considerably high resistance rates and MDR types to commonly used antimicrobial agents among the *Salmonella* isolates tested pose a significant risk factor for the treatment of foodborne *Salmonella* infections in humans.

Keywords: *Salmonella*, chicken, broiler meat, antimicrobial resistance.

### Tavuk eti ve tavuk orijinli *Salmonella* izolatlarında antibiyotik dirençlilik fenotipleri

**Özet:** Bu çalışmada tavuk eti ve tavuk orijinli *Salmonella* izolatlarında 23 antimikrobiyal ajana karşı dirençlilik fenotiplerinin belirlenmesi amacıyla perakende satış yerleri ve çiftlik örneklerinden elde edilen 64 adet et ve 79 adet tavuk *Salmonella* izolatı, Şubat 2006 ile Ocak 2009 arasında laboratuvarımızda analiz edildi. Tüm *Salmonella* izolatlarının, örnek tipi göz önünde bulundurulmaksızın, test edilen antimikrobiyaleden en yüksek direnç oranı ile sırası ile eritmisin'e (%100.0), basitrasin'e (%99.3) ve rifampisin'e (%98.6) olmak üzere en az 4 antimikrobiyale dirençli olduğu belirlendi. Ayrıca, et izolatlarının basitrasin, klindamisin, eritmisin, rifampisin'e; tavuk izolatlarının ise eritmisin, spiramisin, penisilin G, doksisiklin hidroklorür ve tetrasiklin'e %100.0 dirençli olduğu bulundu. Tüm et izolatlarının %82.83'ü 4 ile 7 Çoklu İlaç Direnci (ÇİD) gösterirken, tüm tavuk izolatlarının %55.68'i 10, 12, 15 ve 17 antimikrobiyale ÇİD gösterdiği belirlendi. Et izolatlarında %35.94 oranında ve tavuk izolatlarında ise %13.92 oranında olmak üzere en sık rastlanan ÇİD paternleri sırasıyla B/DA/E/RD ve N/B/CT/E/SP/P/RD/DO/OT/TE olarak bulundu. *Salmonella* izolatlarında karşılaşılan bu yüksek dirençlilik oranlarının ve ÇİD paternlerinin insanlardaki *Salmonella* infeksiyonlarının tedavisinde risk oluşturabileceği belirtildi.

Anahtar sözcükler: *Salmonella*, tavuk, broyler eti, antimikrobiyal direnç

### Introduction

*Salmonella enterica* serovars, the etiological agents of foodborne salmonellosis worldwide, are carried by chicken and transmitted to humans mainly by contaminated broiler meat (19). Treatment of these types of infections becomes limited as a consequence of the rising antimicrobial resistance, which appears to be a direct result of acquiring these increasingly resistant strains from farm animals, particularly by the consumption of chicken meat acquiring *Salmonella* (9). Strategies applied in antimicrobial use both for treatment and for prophylactic/growth promoting purposes in chicken production had significant roles in the

emergence, persistence and spread of these antimicrobial resistant strains in the food chain.

In recent years, there are several reports on *Salmonella* isolates that are resistant to several antimicrobial agents (Multi Drug Resistant - MDR) both in the developed and developing countries from chicken meat (4, 15, 17, 18, 21, 22, 23), and from chickens (3, 5, 7, 10, 12). The emergence of these MDR *Salmonella* has significant outcomes for the safety of the food supply, namely chicken meat, directly as a source of MDR *Salmonella*, and indirectly as a reservoir of antimicrobial genetic elements that can be exchanged between intestinal bacteria (2). Also as the human exposure to

these MDR *Salmonella* via the food supply increases, infections can frequently become harder to treat (2). In our country, up to our knowledge, there are only a few studies indicating the presence of antibiotic resistance in *Salmonella* isolated from chicken meat (13) and from chickens (11, 14), showing requirement of more data by further studies.

In this study, antibiotic resistance phenotypes of 143 *Salmonella* isolates of broiler meat and chicken origin were determined against a total of 23 antimicrobials, which are widely used in veterinary and human medicine.

### Materials and Methods

*Salmonella isolates:* Sixty four retail broiler meat, and 79 chicken farm samples (41 cloacal swab, 7 intestine, 22 caecum and 9 gizzard), which were determined as positive for *Salmonella* in our laboratory from February 2006 to January 2009 were the sources of these isolates. Isolates obtained from these sources had been subjected to both biochemical identification by API 20E (Biomerieux, 20100, France), and serological identification using *Salmonella* group-specific antisera (Becton-Dickinson, USA).

*Antimicrobial susceptibility test:* Antimicrobial profile of each *Salmonella* isolate was determined by the disk agar diffusion method according to the guidelines of the Clinical and Laboratory Standards Institute (6) as follows: A loopful of the *Salmonella* stock culture was streaked onto 7% sterile sheep blood added Blood Agar Base No:2 plate (Oxoid, CM0271) and incubated at 37°C for 24 h. One colony per plate was transferred into 5 ml of Tryptone Soya Broth (Oxoid, CM0129) and was incubated at 37°C for 6 h. After turbidity adjustment to 0.5 McFarland by dilution, a sterile swab was dipped into the inoculum and streaked onto Mueller-Hinton Agar (Oxoid, CM0337) plate three times, where the plate was turned 120° each time. Plates were allowed to dry keeping the lid slightly apart for 1 h. The commercial antibiotic discs were placed in such a way that their centers were at least 30 mm apart and at least 10 mm away from the edge of the plate. Plates were incubated at 37°C and examined at 18 and 24 h. Zone diameters of complete inhibition were measured and the interpretation was done following the recommendation of the disc manufacturer. The 23 antimicrobial agents tested and their corresponding concentrations were as follows: Amikacin (AK, 30 µg, Oxoid CT0107B), Amoxycillin/Clavulanic acid (AMC, 30 µg, Oxoid CT0223B), Ampicillin (AMP, 10 µg, Oxoid CT0003B), Apramycin (APR, 15µg, Oxoid CT0545B), Bacitracin (B, 10 units, Oxoid CT0005B), Ceftiofur (EFT, 30 µg, Oxoid CT1751B), Cephalexin (CL, 30 µg, Oxoid CT0007B),

Cephalazolin (KZ, 30 µg, Oxoid CT0011B), Ciprofloxacin (CIP, 1 µg, Oxoid CT0623B), Clindamycin (DA, 10 µg, Oxoid CT0015B), Colistin sulphate (CT, 10 µg, Oxoid CT0017B), Doxycycline hydrochloride (DO, 30 µg, Oxoid CT0018B), Enrofloxacin (ENR, 5 µg, Oxoid CT0639B), Erythromycin (E, 15 µg, Oxoid CT0020B), Gentamicin (CN, 120 µg, Oxoid CT0794B), Neomycin (N, 30 µg, Oxoid CT0033B), Oxytetracycline (OT, 30 µg, Oxoid CT0041B), Penicillin G (P, 10 units, Oxoid CT0043B), Rifampicin (RD, 5 µg, Oxoid CT0207B), Spiramycin (SP, 100 µg, Oxoid CT0232B), Streptomycin (S, 10 µg, Oxoid CT0047B), Sulphamethoxazole/Trimethoprim 19:1 (SXT, 25 µg, Oxoid CT0052B), Tetracycline (TE, 10 µg, Oxoid CT0053B). The reference bacterial strains *E. coli* (ATCC 25922) and *S. aureus* (ATCC 25923) were used as quality control strains following the recommendations of CLSI (6).

### Results

All broiler meat isolates were found 100.0% resistant to bacitracin, clindamycin, erythromycin and rifampicin, regardless of the serogroups tested. All serogroup based antimicrobial resistance rates are summarized in Table 1. Chicken isolates, which all belonged to serogroup D, were found 100.0% resistant to erythromycin, spiramycin, penicillin G, doxycycline hydrochloride, and tetracycline. Details of resistances to other antimicrobials were provided in Table 1.

All broiler meat isolates were found multi resistant from 4 to 13 out of 15 antimicrobials tested. The highest MDR rate (35.94%) belonged to the 4 MDR group, with a sole pattern as B/DA/E/RD, which was also the common pattern within all the broiler meat isolates. Resistances to other antimicrobials over this pattern were given in detail in Table 2. The chicken isolates, which all belonged to serogroup D, showed antimicrobial resistance ranging from 8 to 20 drugs tested. The highest MDR rate (17.72%) in chicken isolates was observed in both 10 MDR and 17 MDR groups, where 11 out of 14 in 10 MDR showed a main pattern as N/B/CT/E/SPP/P/RD/DO/OT/TE, and 9 out of 14 in 17 MDR revealed a pattern as N/CL/KZ/EFT/B/CT/CIP/ENR/E/SP/AMC/AMP/P/RD/DO/OT/TE (Table 3).

### Discussion and Conclusion

The high (100.0%) antimicrobial resistance observed in all of the broiler meat isolates to bacitracin, clindamycin, erythromycin and rifampicin is a unique finding, since in current literature there is no study, which had tested these antimicrobials on this type of isolates. Among these antimicrobials, in our country, bacitracin is the only one used as feed supplement and not used for treatment purposes in poultry. Also, erythromycin,

Table 1. Antimicrobial resistance results of *Salmonella* isolated from broiler meat and chicken samples.  
 Tablo 1. Tavuk etileri ve tavuklardan izole edilen salmonellaların antimikrobiyal dirençlilik sonuçları.

Sero group (n)	Antimicrobials														Rifamycin Sulfonamide			Tetracyclines					
	Aminoglycosides			Cephalosporins			Cyclic polypeptides			Fluoro quinolones			Macrorides			Penicillins			Rifamycin Sulfonamide			Tetracyclines	
AK	APR	CN	N	S	CL	KZ	EFT	B	CT	CIP	ENR	DA	E	SP	AMC	AMP	P	RD	SXT	DO	OT	TE	
B (11)	0	2	0	0	0	0	11	2	1	1	11	11	1	1	11	0	0	0	0	0	0	0	0
C1 (4)	0	4	4	0	0	0	4	4	4	4	4	4	0	0	4	4	4	4	4	4	4	4	3
C2 (7)	1	5	6	5	5	7	2	0	2	7	7	7	5	5	7	1	2	5	5	7	1	2	5
D (27)	0	13	3	0	27	3	5	7	27	27	27	27	0	0	27	0	0	0	0	0	1	1	2
E1 (1)	0	1	0	0	0	1	1	0	0	1	1	1	0	0	0	1	0	0	0	0	0	0	0
E4 (12)	0	3	0	1	12	3	0	3	12	12	12	12	0	0	0	12	0	0	0	0	0	0	0
F (1)	0	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	0
G (1)	0	0	1	0	1	1	0	0	1	1	1	1	1	0	0	0	1	0	0	0	0	0	0
<b>Sub total</b> (64)	1	29	15	7	64	17	11	18	64	64	64	64	7	7	64	6	6	8	10	10	12.5	15.6	15.6
<b>%</b>	<b>1.6</b>	<b>45.3</b>	<b>23.4</b>	<b>10.9</b>	<b>100.0</b>	<b>26.6</b>	<b>17.2</b>	<b>28.1</b>	<b>100.0</b>	<b>100.0</b>	<b>100.0</b>	<b>100.0</b>	<b>10.9</b>	<b>100.0</b>	<b>9.4</b>	<b>12.5</b>	<b>15.6</b>						
Chicken (79)	6	12	0	65	35	38	37	27	78	75	46	43	79	79	33	35	79	77	6	79	76	79	79
<b>%</b>	<b>7.6</b>	<b>15.1</b>	<b>0.0</b>	<b>82.3</b>	<b>44.3</b>	<b>48.1</b>	<b>46.8</b>	<b>34.2</b>	<b>98.7</b>	<b>94.9</b>	<b>58.2</b>	<b>54.4</b>	<b>100.0</b>	<b>100.0</b>	<b>41.8</b>	<b>44.3</b>	<b>100.0</b>	<b>97.5</b>	<b>7.6</b>	<b>100.0</b>	<b>96.2</b>	<b>100.0</b>	<b>100.0</b>
<b>Total</b> (143)	1	94	50	44	142	92	57	61	143	143	143	143	42	42	141	141	141	141	12	87	86	86	86
<b>%</b>	<b>0.7</b>	<b>65.7</b>	<b>34.9</b>	<b>30.8</b>	<b>99.3</b>	<b>64.3</b>	<b>39.9</b>	<b>42.7</b>	<b>100.0</b>	<b>100.0</b>	<b>100.0</b>	<b>100.0</b>	<b>29.4</b>	<b>29.4</b>	<b>98.6</b>	<b>8.4</b>	<b>8.4</b>	<b>60.8</b>	<b>60.1</b>				

AK, Amikacin; APR, Apranyein; CN, Gentamicin; N, Neomycin; S, Streptomycin; CL, Cephalexin; KZ, Cephaezolin; EFT, Ceftiofur; B, Bacitracin; CT, Colistin sulphate; CIP, Ciprofloxacin; ENR, Enrofloxacin; DA, Clindamycin; E, Erythromycin; SP, Spiramycin; AMC, Amoxycillin/Clavulanic acid; AMP, Ampicillin; P, Penicillin G; RD, Rifampicin; SXT, Sulphamethoxazole/Trimethoprim 9:1; DO, Doxycycline hydrochloride; OT, Oxytetracycline; TE, Tetracycline

Table 2. Antimicrobial resistance patterns of broiler meat *Salmonella* isolates  
 Tablo 2. Tavuk eti *Salmonella* izolatlari antimikrobiyal direnciqlik paternleri

No. resistant antimicrobial	No. isolates and serogroup / total (%)	No. isolates with same pattern and serogroup	Antimicrobials										DO	OT
			CN	N	S	KZ	B	CT	CIP	ENR	DA	E	AMP	
4	7B, 9D, 7E4 / 23 (35.94)	23 BDE4					B				DA	E		RD
5	IB, 10D, 2E4 / 13 (20.31)	1B	N				B	CT			DA	E		RD
		7D					B				DA	E		RD
		2D					B			ENR	DA	E		RD
		1D	S				B	CT			DA	E		RD
		1E4					B			ENR	DA	E		RD
6	3B, 1C2, 4D, 1E1, 1E4, 1G / 11 (17.18)	1B	N				B	CT	CIP		DA	E		RD
		1B	N				B		ENR	DA	E	AMP		RD
		1B	N				B	CT		DA	E			RD
		1C2	N				B	CT	CIP		DA	E		RD
		1D	N				B	CT	CIP	ENR	DA	E		RD
		1D	N				B			DA	E			RD
		1D	N				B	CT			DA	E		RD
		1E1	N				B			DA	E			RD
		1E4	N				B			ENR	DA	E		RD
		1G	S				B	CT			DA	E		RD
7	1C2, 3D, 2E4 / 6 (9.40)	1C2	N	S	B		B	CT	CIP		DA	E		RD
		2D	N		B		B	CT		ENR	DA	E		RD
		1D	N		B		B	CT		ENR	DA	E		RD
		1E4	N		B		B	CT		ENR	DA	E		RD
8	2C2 / 2 (3.12)	2C2	S	KZ	B						DA	E	AMP	RD
		1C2	N	KZ	B						DA	E	AMP	RD
9	1C2 / 1 (1.56)	1C1	N	S	B	CT	CIP		ENR	DA	E		RD	OT
		1D	N	S	B	CT	CIP		ENR	DA	E		RD	OT
11	1C1, 1D / 2 (3.12)	3C1	N	S	B	CT	CIP		ENR	DA	E		RD	OT
		1C2	CN	N	KZ	B			ENR	DA	E	AMP	RD	OT
		1C2	N	S	KZ	B			ENR	DA	E	AMP	RD	OT
12	3C1, 2C2 / 5 (7.81)	1F	N	S	KZ	B	CT	CIP	ENR	DA	E	AMP	RD	OT
		1F	N	S	KZ	B	CT	CIP	ENR	DA	E	AMP	RD	OT
13	1F / 1 (1.56)													

CN, Gentamicin; N, Neomycin; S, Streptomycin; KZ, Cephazolin; B, Bacitracin; CT, Colistin sulphate; CIP, Ciprofloxacin; ENR, Enrofloxacin; DA, Clindamycin; E, Erythromycin; AMP, Ampicillin; RD, Rifampicin; SXT, Sulphamethoxazole/Trimethoprim 19:1; DO, Doxycycline hydrochloride; OT, Oxytetracycline

Table 3. Antimicrobial resistance patterns of chicken *Salmonella* isolates  
 Tablo 3. Tavuk *Salmonella* izolatlarının antimikrobiyal dirençlilik paternleri

No. antimicrobial	No. isolates (%)	No. isolates with same pattern	Antimicrobials																				
			AK	APR	CN	N	S	CL	KZ	EFT	B	CIP	ENR	E	SP	AMC	AMP	P	RD	SXT	DO	OT	TE
8	2 (2.53)	1									B	CT		E	SP			P	RD		DO		TE
9	4 (5.06)	3									B	CT		E	SP			P	RD		DO	OT	TE
10	14 (17.72)	11									B	CT		E	SP			P	RD		DO	OT	TE
											B	CT		E	SP			P	RD		DO	OT	TE
											B	CT		E	SP			P	RD		DO	OT	TE
11	6 (7.59)	2									N	S		E	SP			P	RD		DO	OT	TE
12	8 (10.12)	1									N	CL		E	SP			P	RD		DO	OT	TE
											N	S		E	SP			P	RD		DO	OT	TE
											N	CL		E	SP			P	RD		DO	OT	TE
13	5 (6.32)	2									N	S		E	SP			P	RD		DO	OT	TE
											N	S		E	SP			P	RD		DO	OT	TE
14	3 (3.79)	1									N	CL		E	SP			P	RD		DO	OT	TE
											N	CL		E	SP			P	RD		DO	OT	TE
											N	CL		E	SP			P	RD		DO	OT	TE
15	8 (10.12)	1									N	CL		E	SP			P	RD		DO	OT	TE
											N	CL		E	SP			P	RD		DO	OT	TE
16	5 (6.32)	3									N	CL		E	SP			P	RD		DO	OT	TE
											N	CL		E	SP			P	RD		DO	OT	TE
17	14 (17.72)	2									N	CL		E	SP			P	RD		DO	OT	TE
											N	CL		E	SP			P	RD		DO	OT	TE
18	7 (8.86)	1									N	CL		E	SP			P	RD		DO	OT	TE
											N	CL		E	SP			P	RD		DO	OT	TE
19	2 (2.53)	2									N	CL		E	SP			P	RD		DO	OT	TE
20	1 (1.26)	1									N	CL		E	SP			P	RD		DO	OT	TE

AK, Amikacin; APR, Apramycin; CN, Gentamicin; N, Neomycin; S, Streptomycin; CL, Cephalexin; KZ, Cephalozin; EFT, Cefotaxime; CT, Colistin sulphate; CIP, Ciprofloxacin; ENR, Enrofloxacin; E, Erythromycin; SP, Spiramycin; AMC, Amoxycillin/Clavulanic acid; AMP, Ampicillin; P, Penicillin G; RD, Rifampicin; SXT, Sulphamethoxazole/Trimethoprim 19:1; DO, Doxycycline hydrochloride; OT, Oxytetracycline; TE, Tetracycline

which was being used as supplement and for treatment in poultry between 1980s and 1990s, is not an antimicrobial of current use. The isolates were also checked for resistance status against clindamycin and rifampicin, specific drugs particularly used in human medicine, and it was interesting to find out high resistance against these important antimicrobials in all the isolates tested. The intermediate resistance to neomycin, and relatively lower but notable resistances to enrofloxacin, colistin sulphate, streptomycin, ciprofloxacin, oxytetracycline, doxycycline hydrochloride, ampicillin, and sulphamethoxazole/trimethoprim 19:1, all of which are the members of drug classes that have widely been used in poultry medicine and/or production for years were remarkable. Our results are in parallel to the previous studies reporting resistance to these antimicrobials, namely for streptomycin and ampicillin (15, 17, 18, 21, 22, 23), enrofloxacin (4, 21), ciprofloxacin and sulphamethoxazole/trimethoprim 19:1 (4, 17, 22, 23). Also, resistance results to doxycycline hydrochloride and oxytetracycline were similar to the previously reported tetracycline resistance results of others (13, 15, 17, 21, 22, 23).

The high resistance of our serogroup C1 meat isolates particularly to streptomycin, ciprofloxacin, sulphamethoxazole/trimethoprim 19:1 were similar to the findings of Little et al. (16). For the serogroup C2 isolates' resistances, two studies from the USA showed similarity to our results as: by Lestari et al. (15), who found high resistance to ampicillin, oxytetracycline and streptomycin, and by Parveen et al. (18), who reported high resistance to cephalazolin, ampicillin and oxytetracycline in their meat isolates of serovars belonging to serogroup C2. Also, another study from Algeria reported high resistance to tetracycline and streptomycin in their meat isolates belonging to serovar C2 (7). These results indicate that presence of these types of isolates in meat causing human infections would definitely pose risk in treatments, since they show resistance to several important groups of antimicrobials.

The high antimicrobial resistance of the chicken isolates to bacitracin, erythromycin and rifampicin in this study is similar to the findings obtained for the broiler meat isolates. Previously, high resistance to erythromycin in broiler *Salmonella* isolates was reported by Bekele and Ashenafi (3), who similarly had detected high resistance results for penicillin G, doxycycline hydrochloride in parallel to our findings. Also, the resistance rates determined in this study for the chicken isolates were in agreement with previous reports for tetracycline (5, 12), oxytetracycline (10), ciprofloxacin and enrofloxacin (5), enrofloxacin and penicillin G (14), cephalexin and cephalazolin (3), streptomycin and ampicillin (3, 5, 12), amoxicillin/clavulanic acid (3), and ceftiofur (10). The resistance rate observed in the broiler isolates for

sulphamethoxazole/trimethoprim 19:1 was relatively lower than previously reported by Ishihara et al. (10) in chicken isolates.

Among recent reports of antimicrobial resistance in *Salmonella* isolated from chicken meat and chickens, the results vary widely among antimicrobials tested and according to the origin of the isolates. When our isolates' resistance results are compared based on the common antimicrobials used in these studies, relatively lower resistance rates to streptomycin (3, 15, 17, 18, 23), sulphamethoxazole/trimethoprim 19:1 (4, 10, 17, 22, 23) and to oxytetracycline (10) were found in our study to those previously reported.

The resistance rates obtained in this study were higher than earlier reports of Miranda et al. (17) and Zewdu and Cornelius (23) for ciprofloxacin; of Capita et al. (4) and Van et al. (21) for enrofloxacin; of Ishihara et al. (10), Kang et al. (12), Kasimoglu et al. (13), Lestari et al. (15) and Yan et al. (22) for cephalazolin; and of Bekele and Ashenafi (3) for doxycycline hydrochloride. There were almost equivalent number of studies reporting higher (17, 18, 22, 23) and lower (10, 15, 16, 21) resistances compared to our ampicillin resistance rates.

The resistance of the *Salmonella* isolates in this study was substantially higher for the antimicrobials, which are in use for human and veterinary medicine for longer time, with the exceptions of the following drugs previously reported with lower resistance similar to ours: ampicillin (10, 14, 15, 16, 21), sulphamethoxazole/trimethoprim 19:1 (12, 15), and gentamicin (11, 14). The reasons behind these relatively lower resistances in our *Salmonella* isolates for these 3 antimicrobials, which also have been used in the treatment of various poultry diseases for a long time need to be investigated in further studies.

Overall evaluation of the resistance rates of broiler meat and chicken isolates indicated prominent resistance to erythromycin, bacitracin, and rifampicin, the latter two antimicrobials with restricted use in large animal practice (20). Also, results of 11 common antimicrobials tested for these isolates showed that resistance rates of the chicken isolates were almost always higher than those of broiler meat isolates except for sulphamethoxazole/trimethoprim 19:1. All these results imply that the isolates showed resistance to one or more antimicrobial tested, except to gentamicin, to which all but one isolates were found susceptible. This very low overall resistance to gentamicin (with 100% susceptibility in all chicken *Salmonella* isolates) shows similarity to previous data by Kalender and Muz (11), and Kilinc and Aydin (14).

The significantly high antimicrobial resistance rates in our *Salmonella* isolates were probably an indication of their intense use in practice without paying required attention to the following issues: (I) indiscriminate use of antimicrobials with improper administration periods and

doses (8) both in poultry and human medicine (II) self-medication by easy access to antimicrobials without prescription (1) in human medicine, (III) use of subtherapeutic dose of antimicrobials for prophylactic/nutritional purposes in poultry sector; (IV) unintended violations in withdrawal periods of antimicrobials in poultry production. All these practices can cause emergence and dissemination of resistant *Salmonella* in humans, livestock and their products, such as chicken meat (19).

Another very striking outcome of our study is that all of our broiler meat and chicken *Salmonella* isolates were MDR to 4-13 out of 15, and 8-20 out of 22 antimicrobials tested, respectively. The 100% MDR to a minimum of 4 antimicrobials that was detected in our study for the broiler meat isolates surpasses the MDR rates reported in similar recent studies as follows: MDR to  $\geq 1$ : 62.1% (23), 88.9% (21); MDR to  $\geq 2$ : 27.8% (21), 40% (4), 52.4% (15), 68.75% (13), 72.5% (18), 89.6% (22); MDR to  $\geq 3$ : 35.7% (15), 53.4% (18). Within these studies, several had results with MDR *Salmonella* isolates with  $\geq 4$  antimicrobials still with lower rates compared to ours as: 6.25% (13), 52.7% (22), 85.4% (17), and some indicated resistance in some of their isolates up to 10 antimicrobials (15, 23), which is similar to our up to 13 MDR finding. MDR results of our chicken *Salmonella* isolates showed that our 100% MDR rate (minimum to 8 antimicrobials) was higher than previous reports of Elgroud et al. (7) and Ishihara et al. (10) (MDR to  $\geq 2$  antimicrobials: 51.7% and 91.3%, respectively), and of Bekele and Ashenafi (3) (MDR to  $\geq 3$  antimicrobials: 24.6%), who also reported resistance up to 6 antimicrobials in their broiler *Salmonella* isolates, in contrast to our results.

The MDR pattern common to all meat isolates as B/DA/E/RD, and the main pattern of N/B/CT/E/SP/P/RD/DO/OT/TE in most of the chicken isolates showed no one to one similarity/commonality to the previous reports on chicken meat (13, 15, 17, 18, 23) and chicken (10, 12). As one should expect, this could be mainly related to the differences in the specific antimicrobials used in different studies, serovar/serogroup differences of the isolates, country or regional origin of the isolates, and the source or type of the isolate.

In our study, the number of antimicrobials/antimicrobial families that our broiler meat isolates were MDR to were questioned, and it was observed that serogroup B, D and E4 isolates showed relatively lower MDR (4 to 6 antimicrobials in all B; 4 to 7 antimicrobials in all E4; 4 to 7 antimicrobials in all but one D), than C1 and C2 isolates. Additionally, most/all C1 and/or C2 serogroup isolates, with relatively higher MDRs (mostly  $\geq 7$  antimicrobials), were the dominant serogroups in resistance to streptomycin, cephazoline, ciprofloxacin, enrofloxacin, ampicillin, sulphamethoxazole(trimethoprim

19:1, doxycycline hydrochloride and oxytetracycline. This type of a tendency in the studied serogroups is an interesting note open to further genotype based investigations, due to the absence of serovar information of our isolates, and since one serogroup is comprised of many serovars, which can definitely exhibit different antimicrobial resistance phenotypes, and even under a specific serovar, number of resistant antimicrobials can vary in great extent for different isolates as seen in previous reports (13, 21). Also, in the broiler meat versus chicken isolate comparisons, limited to antimicrobials tested in both groups of isolates, revealed that there were substantially more chicken isolates (92.4%) than broiler meat isolates (25.0%) showing MDR with resistance to  $\geq 7$  antimicrobials (data not shown in detail). At this point, this finding should be left as entirely coincidental, since it is restricted to observations in these isolates.

The considerably high resistance rates and MDR types to commonly used antimicrobial agents among the *Salmonella* isolates tested in our study once more indicated strict implementation of pertinent regulations in conjunction with reliable control systems, to establish prudent use of these antimicrobials in veterinary and human medicine.

## References

1. Acha PN, Szyfres B (2001): *Zoonoses and Communicable Diseases Common to Man and Animals*. 3rd edn., Washington DC: Pan American Health Organization, Vol.1, pp. 233-246.
2. Angulo FJ, Nargund VN, Chiller TC (2004): Evidence of an association between use of anti-microbial agents in food animals and anti-microbial resistance among bacteria isolated from humans and the human health consequences of such resistance. J Vet Med B, **51**, 374-379.
3. Bekele B, Ashenafi M (2010): Distribution of drug resistance among enterococci and *Salmonella* from poultry and cattle in Ethiopia. Trop Anim Health Pro, **42**, 857-864.
4. Capita R, Alonso-Calleja C, Prieto M (2007): Prevalence of *Salmonella enterica* serovars and genovars from chicken carcasses in slaughterhouses in Spain. J Appl Microbiol, **103**, 1366-1375.
5. Chiu LH, Chiu CH, Horn YM, Chiou CS, Lee CY, Yeh CM, Yu CY, Wu CP, Chang CC, Chu C (2010): Characterization of 13 multi-drug resistant *Salmonella* serovars from different broiler chickens associated with those of human isolates. BMC Microbiol, **10**, 86.
6. Clinical and Laboratory Standards Institute (CLSI) (2006): *Performance Standards for Antimicrobial Disk Susceptibility Tests*. Approved Standard, 10<sup>th</sup> edn. Documents M02-A10, M7-A8, M100-S19, Vol. 26, No. 1. Pennsylvania, PA, USA.
7. Elgroud R, Zerdoumi F, Benazzouz M, Bouzitouna-Bentchouala C, Granier SA, Frémy S, Brisabois A, Dufour B, Millemann Y (2009): Characteristics of *Salmonella* contamination of broilers and slaughterhouses

- in the region of Constantine (Algeria).* Zoonoses Public Hlth, **56**, 84-93.
8. **Guthrie RK** (1992): *Salmonella*, CRC Press, USA, pp. 23-156.
  9. **Hohmann EL** (2001): *Nontyphoidal salmonellosis*. Clin Infect Dis, **32**, 263-269.
  10. **Ishihara K, Takahashi T, Morioka A, Kojima A, Kijima M, Asai T, Tamura Y** (2009): *National surveillance of Salmonella enterica in food-producing animals in Japan*. Acta Vet Scand, **51**, 35.
  11. **Kalender H, Muz A** (1999): *Typing of Salmonella species isolated from chickens in Elazig region*. Turk J Vet Anim Sci, **23**, 297-303.
  12. **Kang ZW, Jung JH, Kim SH, Lee BK, Lee DY, Kim YJ, Lee JY, Won HK, Kim EH, Hahn TW** (2009): *Genotypic and phenotypic diversity of Salmonella Enteritidis isolated from chickens and humans in Korea*. J Vet Med Sci, **71**, 1433-1438.
  13. **Kasimoglu Dogru A, Ayaz ND, Gencay YE** (2009): *Serotype identification and antimicrobial resistance profiles of Salmonella spp. isolated from chicken carcasses*. Trop Anim Health Pro, **42**, 893-897.
  14. **Kiline U, Aydin F** (2006): *Antibiotic susceptibility of Salmonella spp. isolated chicken from poultry enterprises in Kayseri region*. J Health Sci, **15**, 35-40.
  15. **Lestari SI, Han F, Wang F, Ge B** (2009): *Prevalence and antimicrobial resistance of Salmonella serovars in conventional and organic chickens from Louisiana retail stores*. J Food Prot, **72**, 1165-1172.
  16. **Little CL, Richardson JF, Owen RJ, De Pinna E, Threlfall EJ** (2008): *Prevalence, characterisation and antimicrobial resistance of Campylobacter and Salmonella in raw poultry meat in the UK, 2003-2005*. Int J Environ Heal R, **18**, 403-414.
  17. **Miranda JM, Mondragon AC, Martinez B, Guarddon M, Rodriguez JA** (2009): *Prevalence and antimicrobial resistance patterns of Salmonella from different raw foods in Mexico*. J Food Prot, **72**, 966-971.
  18. **Parveen S, Taabodi M, Schwarz JG, Oscar TP, Harter-Dennis J, White DG** (2007): *Prevalence and antimicrobial resistance of Salmonella recovered from processed poultry*. J Food Prot, **70**, 2466-2472.
  19. **Poppe C** (1999): Epidemiology of *Salmonella enterica* serovar Enteritidis, In: A.M. Saeed, R.K. Gast, P.G. Wall (eds), *Salmonella enterica serovar Enteritidis in Humans and Animals: Epidemiology, Pathogenesis and Control*, Iowa State Univ. Press, USA, pp. 3-18.
  20. **Turkish Food Codex** (2002): *Regulation on the maximum residue limits on veterinary drugs in animal-derived foods*. Legislation No: 2002/30.
  21. **Van TTH, Moutafis G, Istivan T, Tran LT, Coloe PJ** (2007): *Detection of Salmonella spp. in retail raw food samples from Vietnam and characterization of their antibiotic resistance*. Appl Environ Microb, **73**, 6885-6890.
  22. **Yan H, Li L, Alam MJ, Shinoda S, Miyoshi S, Shi L** (2010): *Prevalence and antimicrobial resistance of Salmonella in retail foods in Northern China*. Int J Food Microbiol, **143**, 230-234.
  23. **Zewdu E, Cornelius P** (2009): *Antimicrobial resistance pattern of Salmonella serotypes isolated from food items and personnel in Addis Ababa, Ethiopia*. Trop Anim Health Pro, **41**, 241-249.

Geliş tarihi: 07.09.2011 / Kabul tarihi: 17.11.2011

#### Address for correspondence:

Prof. Dr. Aysegul Eyigor  
 Department of Food Hygiene and Technology  
 Faculty of Veterinary Medicine  
 Uludag University 16059, Görükle Campus  
 Bursa, Turkey  
 Email: aeyigor@uludag.edu.tr  
 Phone: +90.224.2941334  
 Fax: +90.224.294 1202