

Salmonella spp. and antibiotic-resistant strains in wild mammals and birds in north-western Italy from 2002 to 2010

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Summary

Salmonella is an important zoonotic pathogen of economic importance. In Europe, salmonellosis is the second food-borne infection, in Italy, *Salmonella* is still the major cause of food-borne outbreaks. In Europe, there are many *Salmonella* surveillance plans on farmed animals, while *Salmonella* survey of wild animals is occasionally performed. The aim of this study was to investigate the presence of *Salmonella* including the antibiotic-resistant strains in wild animals. Between 2002 and 2010, 2,713 wild animals (canids, mustelids, birds, rodents, ungulates), were collected in north-western Italy and tested for *Salmonella* by classical microbiological culture method followed by serological and biochemical typing. One hundred and seventeen wild animals (63 canids, 25 mustelids, 24 birds, 5 ungulates) were found positive for *Salmonella* (4.3%). One hundred and thirty strains, belonging to several serotypes were isolated, and *S. Typhimurium* was the most common serotype found. Antibiotic susceptibility was tested by disk-diffusion test on 88 strains. Almost all the analyzed strains (97.7%) showed resistance/intermediate resistance to at least one class of antibiotics and the highest resistance values were observed for the tetracycline class. In conclusion, zoonotic and antibiotic-resistant serotypes were found in many species of wildlife.

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Introduction

Salmonella is a zoonotic bacterium of public health significance, with considerable economic impact (19). Salmonellosis is one of the most common and widely distributed food-borne diseases. It is noteworthy that in Europe, Salmonellosis is the second food-borne disease after campylobacteriosis.

In 2010, the European Food Safety Authority (EFSA) reported 99,020 cases of human salmonellosis from 27 EU Members States including 2,730 cases in Italy. In our country *Salmonella* is still the major cause of food-borne outbreaks (10). The genus *Salmonella* consists of two species: *S. enterica* and *S. bongori* (22). The first one includes 6 subspecies (subsp.): *enterica*, *salamae*, *arizonae*, *diarizonae*, *houtenae*, and *indica*. There are over 2,500 serotypes of zoonotic *Salmonella*, most belonging to the subspecies *enterica*.

Salmonella resides in a variety of different hosts: in farmed and domestic animals but also in wild and exotic fauna, including reptiles. Several European Countries endorse *Salmonella* surveillance plans on

farmed animals. In this respect it is noteworthy that in 2009 the great majority of strains was collected from poultry (45.7%), swine (16.02%), and turkey (4.14%) (3). Conversely, *Salmonella* survey in wild animals is not regulated by national monitoring plans and it is only occasionally performed for research purpose. Several studies have been performed on wild birds (17, 27), however few reports are available on the presence of *Salmonella* in wild mammals. This study aimed to investigate the presence of *Salmonella* including antibiotic-resistant strains in wild animals (canids, mustelids, wild birds, rodents, and ungulates) in north-western Italy.

Material and methods

Sample collection

Between January 2002 and December 2010, 2,713 wild animals (1,222 canids, 221 mustelids, 1,101 wild birds, 100 rodents, 69 ungulates) were collected in north-western Italy (Valle d'Aosta,

Table I. List of sampled animals.

	Family	Genus/species (common name)	
Canids	Canidae	<i>Vulpes vulpes</i> (red fox)	
Mustelids	Mustelidae	<i>Meles meles</i> (badger)	
		<i>Martes martes</i> (stone marten)	
		<i>Martes foina</i> (marten)	
		<i>Mustela putorius</i> (polecat)	
Wild birds	Accipitridae	<i>Accipiter gentilis</i> (goshawk)	
		<i>Accipiter nisus</i> (sparrowhawk)	
		<i>Aquila chrysaetos</i> (golden eagle)	
		<i>Buteo buteo</i> (common buzzard)	
		<i>Circus gallicus</i> (short-toed snake-eagle)	
		Ardeidae	<i>Nycticorax nycticorax</i> (black-crowned night heron)
			<i>Columba livia</i> (common pigeon)
	Corvidae	<i>Garrulus glandarius</i> (eurasian jay)	
		<i>Corvus corone</i> (carrion crow)	
		<i>Pica pica</i> (magpie)	
	Cuculidae	<i>Cuculus canorus</i> (cuckoo)	
	Falconidae	<i>Falco peregrinus</i> (peregrine falcon)	
		<i>Falco</i> spp. (hawk)	
		<i>Falco tinnunculus</i> (common kestrel)	
		<i>Pernis apivorus</i> (honey buzzard)	
	Fringillidae	<i>Fringilla coelebs</i> (chaffinch)	
	Laridae	<i>Larus</i> spp. (seagull)	
	Passeridae	<i>Passer domesticus</i> (house sparrow)	
	Picidae	<i>Picus viridis</i> (green woodpecker)	
	Scolopacidae	<i>Scolopax rusticola</i> (woodcock)	
	Strigidae	<i>Asio otus</i> (long-eared owl)	
		<i>Bubo bubo</i> (eagle-owl)	
		<i>Otus scops</i> (scops owl)	
		<i>Strix aluco</i> (tawny owl)	
	Sturnidae	<i>Sturnus vulgaris</i> (european starling)	
	Tetraonidae	<i>Tetrao tetrix</i> (black grouse)	
	Turdidae	<i>Turdus merula</i> (blackbird)	
Tytonidae	<i>Tyto alba</i> (barn owl)		
Rodents	Cricetidae	<i>Clethrionomys glareolus</i> (bank vole)	
	Myocastoridae	<i>Myocastor coypus</i> (nutria)	
	Muridae	<i>Apodemus</i> spp. (field mouse)	
	Soricidae	<i>Sorex</i> spp. (shrew)	
Ungulates	Cervidae	<i>Cervus elaphus</i> (red deer)	
	Suidae	<i>Sus scrofa</i> (wild boar)	

Piemonte and Liguria regions) and tested for *Salmonella* spp. (Table I). The animals were mostly found dead (usually for a trauma or following a car accident); some of them were hunted for provincial or regional control plans; in fewer cases, the animals were admitted to wildlife rehabilitation centres.

Dead animals, delivered to our laboratory, were submitted to a complete necropsy and the sampling

of mesenteric lymph nodes, faeces and viscera was performed for a total of 3,862 biological samples. In particular 927 red foxes (*Vulpes vulpes*) and 146 mustelids were sampled both for faeces and lymph nodes. A cloacal swab was generally collected from live animals.

Isolation and identification of *Salmonella*

All samples were homogenized in a Stomacher blender. After incubation in Buffered Peptone Water for 18 hours, three enrichment broths - Selenite Cystine, Rappaport-Vassiliadis, and Mueller Kauffman Tetrathionate - were inoculated and incubated for 24 hours; 10 µl of each broth were inoculated onto 2 plates of selective media, Brilliant Green Agar and Xylose Lysine Deoxycholate agar. The plates were incubated for 24 hours at 37°C. Suspected colonies were transferred on Triple Sugar Iron for a first identification of *Enterobacteriaceae*, based on sugar fermentation and H₂S production.

The presumptive *Salmonellae* were confirmed with biochemical (API 20E®) and serological tests. Monovalent antisera have been deployed for the identification of the serotypes. *S. Typhimurium* and *S. Enteritidis* were phage typed by the Centro di Referenza Nazionale per le Salmonellosi, Italy, according to standard methods.

Antimicrobial susceptibility testing

The antibiotic susceptibility was tested using disk-diffusion test (Kirby-Bauer Method), performed on Mueller-Hinton agar from a bacterial suspension of turbidity equal to McFarland 0.5. The interpretation was made according to the criteria provided by the Clinical Laboratory Standard Institute (4).

Antibiotics tested in this study are representative of different classes: β-lactams, tetracyclines, quinolones, aminoglycosides, sulphonamides, polypeptides, and phenicols. These molecules were selected for their relevance to public health and taking into account the EFSA guidance (11), the network Enter-net Italy (5), and the available literature. The antibiotics tested and their concentrations are shown in Table II.

Results

Strains of *Salmonella*

One hundred and seventeen wild animals (63 canids, 25 mustelids, 24 wild birds, 5 ungulates) were found positive for *Salmonella* (4.3%). None of them showed clinical symptoms or pathological lesions related to salmonellosis. The positivity frequency was: 5.2%

Table II. Antibiotic susceptibility (Kirby-Bauer Method), number and percentage of resistant/intermediate resistant strains.

Antimicrobial class	Active molecule	Symbol	Dose (μg)	Antibiotic R/IR Strains Fr % (no)
Aminoglycosides	Amikacin	AK	30	0% (n=0)
	Streptomycin	S	10	46.6% (n=41)
	Neomycin	N	30	7.9% (n=7)
	Gentamicin	CN	10	0% (n=0)
	Kanamycin	K	30	4.5% (n=4)
β -lactams	Ampicillin	AM	10	13.6% (n=12)
	Amoxicillin + clavulanic acid	AMC	30 (20&10)	7.9% (n=7)
	Cefotaxim	CTX	30	0% (n=0)
	Cefalotin	KF	30	1.0% (n=1)
Phenicol	Chloramphenicol	C	30	1.0% (n=1)
Polypeptides	Colistin	CT	10	1.0% (n=1)
Quinolones	Nalidixic acid	NA	30	3.4% (n=3)
	Enrofloxacin	ENR	5	3.4% (n=3)
	Ciprofloxacin	CIP	5	0% (n=0)
Sulphonamides	Trimethoprim + sulfamethoxazole	SXT	25 (23.75&1.25)	1.0% (n=1)
Tetracyclines	Oxytetracycline	T	30	72.3% (n=68)
	Tetracycline	TE	30	81.8% (n=72)

Fr = frequency; R = resistant strain; IR = intermediate resistant strain.

Table III. Antibiotic susceptibility (Kirby-Bauer Method), number and percentage of resistant/intermediate resistant strains.

Year	Canids		Mustelids		Wild birds		Rodents		Ungulates		Animals	
	Analyzed (no)	Positive Fr (no)	Analyzed (no)	Positive Fr (no)	Analyzed (no)	Positive Fr (no)	Analyzed (no)	Positive Fr (no)	Analyzed (no)	Positive Fr (no)	Analyzed (no)	Positive Fr (no)
2002	48	6.3% (n=3)	19	15.8% (n=3)	25	-	2	-	-	-	94	6.4% (n=6)
2003	56	7.1% (n=4)	27	22.2% (n=6)	126	1.6% (n=2)	-	-	4	-	213	5.6% (n=12)
2004	142	9.9% (n=14)	57	7.0% (n=4)	545	0.6% (n=3)	65	-	2	-	811	2.6% (n=21)
2005	263	6.8% (n=18)	35	5.7% (n=2)	84	3.5% (n=3)	30	-	26	3.8% (n=1)	438	5.5% (n=24)
2006	178	6.2% (n=12)	33	21.2% (n=7)	135	4.4% (n=6)	-	-	8	-	354	7.0% (n=25)
2007	175	5.1% (n=9)	17	5.9% (n=1)	131	3.1% (n=4)	1	-	-	-	324	4.3% (n=14)
2008	296	0.7% (n=2)	19	-	14	-	2	-	28	14.3% (n=4)	359	1.7% (n=6)
2009	46	-	5	20.0% (n=1)	36	13.9% (n=5)	-	-	1	-	88	6.8% (n=6)
2010	18	5.6% (n=1)	9	11.1% (n=1)	5	20.0% (n=1)	-	-	-	-	32	9.4% (n=3)
Total	1,222	5.2% (n=63)	221	11.3% (n=25)	1,101	2.2% (n=24)	100	-	69	7.2% (n=5)	2,713	4.3% (n=117)

Fr = frequency.

(63/1,222) in canids, 11.3% (25/221) in mustelids, 2.2% (24/1,101) in wild birds, and 7.2% (5/69) in ungulates. No rodent was positive for *Salmonella* (Table III).

A total of 130 *Salmonella* strains was isolated:

2 animals showed 2 subspecies of *Salmonellae* in the same matrix and 11 animals, sampled both for faeces and for lymph nodes [1 marten (*Martes foina*), 2 badgers (*Meles meles*), and 8 red foxes], were positive for both matrixes (Tables IV and V).

The identified *Salmonella* subspecies were as follows: *S. enterica* subsp. *enterica* (n=91), *S. enterica* subsp. *houtenae* (n=13), *S. enterica* subsp. *diarizonae* (n=15), *S. enterica* subsp. *arizonae* (n=5), *S. enterica* subsp. *salamae* (n=3). We also found *Salmonella* spp. indicated as 'new serotype' (n=2) and 'untypifiable serotype' (n=1) by the Food Safety Department (Istituto Zooprofilattico Sperimentale del Piemonte, Liguria e Valle d'Aosta, Italy).

S. Typhimurium was the most common identified serotype and it was documented in 7 red foxes, 4 badgers, 2 common pigeons (*Columba livia*), 1 eurasian jay (*Garrulus glandarius*), 1 house sparrow (*Passer domesticus*), 1 common buzzard (*Buteo buteo*), 1 long-eared owl (*Asio otus*), and 1 barn owl (*Tyto alba*).

The phage typing was made for 10 *S. Typhimurium* and the definitive bacteriophage types (DT) were DT 104, DT 12, DT 193, DT 302 and only 1 'untypifiable phage'. *S. Enteritidis* was found in 5 animals [2 red foxes, 1 short-toed snake-eagle (*Circaetus gallicus*), 1 tawny owl (*Strix aluco*), 1 seagull (*Larus* spp.)] from all sampling areas; in 3 cases the phage type was

Table IV. Coinfection by different *Salmonella* strains or phage types in the same animals (4 cases).

Animal species	Faeces	Mesenteric lymph node
Red fox	<i>S. Typhimurium</i> DT12	<i>S. Typhimurium</i> DT104
Red fox	<i>S. Veneziana</i>	<i>S. Kottbus</i>
Red fox	<i>S. enterica</i> subsp. <i>salamae</i> AND <i>S. Kimuenza</i>	Not tested
Short-toed snake-eagle	<i>S. enterica</i> subsp. <i>arizonae</i> gr. Z:50:z4,z23:-: AND <i>S. Enteritidis</i> gr. D1-1,9,12:g,m:-: PT4	Not tested

Table V. *Salmonella* presence in faeces and lymph nodes of canids and mustelids sampled for both matrixes.

Year	<i>Salmonella</i> in lymph node and faeces		<i>Salmonella</i> in lymph node		<i>Salmonella</i> in faeces	
	Mustelids	Canids	Mustelids	Canids	Mustelids	Canids
2002	-	-	3	2	-	-
2003	2	2	1	1	-	1
2004	-	1	1	6	-	5
2005	2	-	1	12	-	1
2006	1	1	5	8	-	1
2007	-	2	-	2	-	4
2008	-	-	-	1	-	1
2009	-	-	-	-	1	-
2010	-	-	1	-	-	1
Total	5	6	12	32	1	14

PT4. The other serotypes of *S. enterica* subspecies *enterica* are described in Table VI.

Antimicrobial resistance

Eighty-eight strains were tested for antibiotic susceptibility. Almost all the analyzed strains (97.7%) showed resistance (R) / intermediate resistance (IR) to at least one class of antibiotics, with the highest resistance values observed for the tetracycline class. The percentage of resistance to individual active molecules is shown in Table II. Only 2 *S. Enteritidis* strains were fully antimicrobial susceptible (S). Thirty three strains were Multi Drug Resistant (MDR) showing R/IR towards 2 or more classes of antibiotics (Table VII).

Table VI. Serotypes of *Salmonella enterica subspecies enterica*.

<i>Salmonella</i> serotype	No	Species
Alfort	1	Red fox
Bonariensis	3	Red fox, marten
Bredeney	1	Red fox
Braenderup	1	Red fox
Brancaster	1	Common kestrel
Coeln	5	Red fox, badger, marten
Corvallis	1	Badger
Djugu	1	Red fox
Farsta	1	Common pigeon
Galiema	1	Wild boar
Heidelberg	3	Red fox
Hessarek	1	Red fox
Hiduddify	1	Red fox
Infantis	4	Red fox, marten, carrion crow, red deer
Kibi	2	Red fox, marten
Kimuenza	1	Red fox
Kottbus	5	Red fox, wild boar
Livingstone	2	Red fox, scops owl
Loanda	1	Scops owl
Massenya	1	Badger
Mikavasima	1	Red fox
Muenchen	3	Red fox, badger
Napoli	2	Badger
Nordufer	2	Badger
Ohio	1	Golden eagle
Strourbridge	1	Black grouse
Suberu	1	Tawny owl
Thompson	2	Wild boar
Tsevie	1	Common pigeon
Tshiongwe	2	Red fox
Veneziana	8	Red fox, short-toed snake-eagle, peregrine falcon
Wil	1	Red fox

Table VII. Pattern of Multi Drug Resistant strains.

Serotype	Animal species	Pattern of MDR
S. Typhimurium DT 104	Red fox	AM-C-S-T-TE
S. Typhimurium DT 193	Long-eared owl	AM-AMC-N-S-T-TE
S. Typhimurium	Common buzzard	AM-AMC-S-T-TE
	Red fox	AM-NA-ENR-S-TE
	Red fox	AM-NA-ENR-S-T-TE
	Red fox	S-T-TE
S.Brancaster	Common kestrel	AM-AMC-NA-N-S-K-T-TE-SXT
S. Heidelberg	Red fox	AM-AMC-T-TE
S. Bredeney	Red fox	N-S-T-TE
S. Ohio	Eagle	AM-AMC-S-T-TE
New serotype	Red fox	AM-S-T-TE
Untypeable	Marten	AM-AMC-KF-ENR-T-TE

Discussion

This study analyzes the presence of *Salmonella* in wildlife from north-western Italy (canids, mustelids, ungulates, and wild birds).

To our knowledge this is one of the few studies analyzing several wild animal classes for the presence of *Salmonella* in Italy. A similar research, performed in Basque Country (20) for a shorter period of time (2001-2002), and on a lower number of animals, showed that the prevalence of *Salmonella* in wild mammals is comparable to that of wild birds. Positive animals showed no signs of salmonellosis as reported in our study. However, the frequencies of positivity found in our survey in mammals and birds were lower than those reported from Millán *et al.* (5.8% vs 7.2% and 2.2% vs 8.5% respectively) (20).

Salmonella infection in canids was tested only in one study performed on red foxes in Norway. Red foxes were infected with the same serovar of *S. Typhimurium* that caused outbreaks in *Passeriformes* (14). In contrast, in our work there is no correlation among strains of *S. Typhimurium* isolated from different animal species and there is no evidence of salmonellosis outbreaks in wild birds. Nonetheless, some serotypes of *Salmonella* (*S. Infantis*, *S. Kottbus*, *S. Livingstone*, *S. Veneziana*) were detected in different food chain related animals {red fox, marten, raptors [short-toed snake-eagle, scops owl (*Otus scops*), peregrine falcon (*Falco peregrinus*)] and wild boar (*Sus scrofa*}.

The behaviour and the feeding habits of wild animals certainly influence the likelihood to be infected with *Salmonella* (9). In particular, badgers, red foxes, raptors and wild boars could acquire *Salmonella* by scavenging on contaminated carcasses or on different human leftover (20).

A survey on nontyphoidal *Salmonellae* was performed on badger social groups in the United Kingdom, revealing a high prevalence of the

bacteria (72%) (29). These results, even if apparently different, are comparable to our data, which show a high prevalence despite the low sample size. This frequency can be explained by the biology of badgers: they are social animals that share the same setts and latrines and they have an omnivorous diet that includes a variety of food as insects, small mammals and birds.

In Italy a study (18) was conducted on wild boar, roe deer (*Capreolus capreolus*), red deer (*Cervus elaphus*), and red fox in the areas of Lombardia and Emilia Romagna in 2005-2009. *Salmonella* was isolated from wild boars with various prevalences (from 3.9% to 26%) depending on the geographical area of sampling. Only occasionally, the bacterium was detected in roe deer, red deer and red foxes. Similarly to our results, the serotyping revealed the presence of *S. Typhimurium*, *S. Napoli*, *S. Veneziana*, *S. Mishmarhaeme* and, in particular, *S. Thompson* in wild boars.

Several studies were performed on the presence of *Salmonella* in wild birds. In Spain, Reche *et al.* (24) quoted a prevalence of 4% in raptors, mainly *S. Typhimurium* DT 104. In another study, a higher *Salmonella* percentage (10%) is reported in wild raptors (19). In this study, a lower *Salmonella* positivity (2.2%) was reported in all wild birds, even if half of the isolations occurred in raptors (n=12/24).

As reported by Vieira-Pinto *et al.* (28) and Hilbert *et al.* (16), the game animals - in our study wild boars, red deer and black grouse (*Tetrao tetrix*) - could represent an infection source for humans, when shots in the abdomen or an incorrect evisceration might cause faecal contamination of meat intended for consumption. Another source of human infection can be represented by the incorrect manipulation of live wild animals near rehabilitation centres, especially wild birds - in our study golden eagle (*Aquila chrysaetos*), scops owl, common kestrel (*Falco tinnunculus*), tawny owl, common pigeon, long-eared owl (*Asio otis*), and sparrow.

All the animals sampled in this study can be considered healthy carriers of *Salmonella* because of the absence of pathological lesions related to salmonellosis (haemorrhagic enteritis, glaucomatous hepatitis, etc.). The majority of positive red foxes and mustelids, analyzed for two matrixes, was positive only for lymph nodes and not for faeces (n=44). Therefore these animals are *Salmonella* carriers but not excretory at the time of sampling. It is likely that stress factors (adverse climate conditions, nutritional deficiencies, other infectious diseases) facilitate the transfer of *Salmonella* into faeces, so making the animal an excretory carrier. Wildlife may be a rich reservoir of a great diversity of serotypes, most of which are not considered pathogenic. However, some genes

encoding for virulence factors, located on plasmids, can be transferred from one strain to another and they can cause an increase in the pathogenicity of serotypes (1).

Different serotypes can be present in different animal species: some of them are considered species-specific, while others are ubiquitous (13). The presence of virulence plasmids in host-adapted serovars suggests that horizontal virulence acquisition can have expanded the host range of *Salmonella* (25).

Among the *Salmonella* strains isolated in our study, 32% (n=41/130) is also listed in the 2002-2009 Enter-net Italy reports as responsible for human infections: *S. Enteritidis*, *S. Typhimurium*, *S. Heidelberg*, *S. Muenchen*, *S. Thompson*, *S. Bredeney*, *S. Napoli*, and *S. Infantis* (7, 8). In particular, *S. Napoli* is an emerging serovar in Italy. A recent study (12) suggests that *S. Napoli* is mostly present in the environment whence it can spill over to animals and humans, even if Graziani *et al.* (12) do not point out a specific source of exposure for humans. Our survey indicates that badgers may be a substantial source of *S. Napoli*.

S. Thompson has been associated with a salmonellosis outbreak caused by rocket lettuce massively contaminated by irrigation with non-drinkable water (21). Wild animals, in particular wild birds, might cause the contamination of vegetables crops (15) either directly with faecal material, or indirectly, with pollution of irrigation water.

In a previous study (6), performed in Valle d'Aosta region, we observed that the estimated spatial distribution of carnivores and humans infected by *Salmonella* are broadly overlapping. In fact, *S. Typhimurium*, *S. Heidelberg* and *S. Infantis* were isolated from both of them. The overlapping can be explained by the sinantropic behaviour of wild species that live close to residential areas and near houses, farmland, and waste dumps. The generalist diet of wild boars, badgers and foxes increases the risk to acquire *Salmonella* and it justifies the high diversity of serotypes (9).

The data analysis revealed the presence of widespread antimicrobial-resistant *Salmonella* in north-western Italy. The diffusion of zoonotic bacteria resistant to antibiotics is an important concern for the treatment of human infections, because it can compromise the effectiveness of the therapy. All EU Member States collect and analyze a variety of animal and food samples to monitor the presence of antimicrobial-resistant zoonotic bacteria: commonly *Salmonella*, *Campylobacter*, *Escherichia coli*, and *Enterococcus*. However, a comparison of our results with previous research is not possible as the only available data about

antimicrobial resistance in wildlife are those reported by EFSA.

The antimicrobial resistance rate in wildlife, reported in the present study, was higher (97.7%) than the one indicated by the EFSA report concerning livestock (from 0.2% to 75%) (11); however, EFSA data are referred to a higher number of samples.

In our study, the highest resistance values were recorded for tetracyclines. This can be explained by the fact that this antibiotic is used in two thirds of the therapeutic regimens applied in veterinary medicine (26). In addition, in contrast with European data, our results show a complete sensitivity of strains to cefotaxime and ciprofloxacin.

Among the MDR *Salmonellae*, a significant case is represented by *S. Brancaster* isolated by a common kestrel that showed R/IR towards ampicillin, amoxicillin, clavulanic acid, nalidixic acid, neomycin, streptomycin, kanamycin, oxytetracycline, tetracycline, trimethoprim and, sulfamethoxazole. Epidemiological information concerning this serotype is rather limited. The literature indicates two sporadic isolates: *S. Brancaster* in Tuscany in 1985 and 1991 from human samples (23), but without antimicrobial susceptibility data.

S. Typhimurium DT 104, considered an emerging pathogen, shows a higher number of antibiotic-resistance than the other phage types. So far, it is not known whether the isolates of DT 104 possess greater virulence, or whether the virulence is associated with multiple genes. If the genes for virulence were associated with the antibiotic-resistance ones, the virulence could be selected by the use of antibiotics (2).

Massive treatments in breeding animals for therapy and prophylaxis of bacterial infections, inaccurate posology and inadequate treatment times are probable causes promoting the selection of antibiotic-resistant strains. Even the use of food supplemented with antibiotics in sub therapeutic conditions, as growth promoters, can produce bacterial resistance towards molecules already in use and those structurally and pharmacologically related (cross-resistance) (13).

Conclusions

In conclusion a constant supervision of *Salmonella* in wildlife and its antibiotic-susceptibility is necessary to the early identification of zoonotic strains and the emergence of new resistance profiles, and to evaluate the control measures. Our findings in wildlife and the widespread *Salmonellae* outbreaks underline the need to better coordinate investigations between human and veterinary health and food safety organisations and networks.

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