

Antimicrobial resistance and subtyping of *Salmonella enterica* subspecies *enterica* serovar Enteritidis isolated from human outbreaks and poultry in southern Brazil

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ABSTRACT To investigate antimicrobial resistance, 96 *Salmonella enterica* subspecies *enterica* serovar Enteritidis strains isolated from salmonellosis outbreaks and poultry-related products obtained in southern Brazil were analyzed. Macrorestriction patterns, obtained by pulsed-field gel electrophoresis and phage types, were assessed. Although 43.75% of samples were sensitive to all drugs tested, resistance to sulfonamide (34.37%), trimethoprim-sulfamethoxazole (25.00%), nalidixic acid (14.58%), streptomycin (2.08%), gentamicin, and tetracycline (1.04%) was identified. Furthermore, 89.60% of

strains belonged to phage type 4, and a predominant pulsed-field gel electrophoresis genotype represented by 82.29% of the strains was identified, suggesting that a clonal group was distributed in poultry, food, and human isolates. Although it was not possible to associate strains from different sources, the occurrence of antimicrobial-resistant *Salmonella* Enteritidis strains supports the need to establish monitoring programs to identify the emergence of potential resistance patterns and to direct policies for use of these drugs in food-producing animals.

Key words: *Salmonella* Enteritidis, antimicrobial resistance, pulsed-field gel electrophoresis, phage type, poultry

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INTRODUCTION

The global incidence of human infection by *Salmonella enterica* subspecies *enterica* serovar Enteritidis (*Salmonella* Enteritidis) has increased since 1990 (Rabsch et al., 2001). The high prevalence of *Salmonella* Enteritidis in fowls (Kanashiro et al., 2005) suggests that poultry is an important epidemiological reservoir (Coyle et al., 1988; Kimura et al., 2004). In Brazil, *Salmonella* Enteritidis has been associated with human foodborne infections caused by the ingestion of contaminated foods of animal origin, mainly undercooked poultry meat and eggs (Tavechio et al., 1996; Peresi et al., 1998), and has emerged as the most frequent serovar isolated from human illness (Tavechio et al., 1996; Fernandes et al., 2006). Different countries, including Brazil, have reported the same *Salmonella* Enteritidis phage type (PT) 4 (dos Santos et al., 2003; Nunes et al., 2003; Fisher, 2004), which is associated with poultry (Coyle et al., 1988; Rabsch et al., 2001).

On the other hand, the prevalence of antimicrobial-resistant bacteria has increased worldwide in the last decades. The incidence of multidrug-resistant *Salmonella* Typhimurium definitive PT (DT) 104 has increased widely (Gebreyes et al., 2006; Ghilardi et al., 2006). Selection pressure exerted by the use of antimicrobial drugs to promote growth or disease prophylaxis in food-producing animals is suggested as the most important selective pressure to induce the emergence of resistant bacteria (Schwarz et al., 2001). Because these animals may act as reservoirs of resistant bacteria, resistant strains may be spread from animals to humans through foods of animal origin. Furthermore, many antimicrobial agents used in animals belong to the same classes with applications in human medicine (Schwarz et al., 2001; Phillips et al., 2004). Thus, the emergence of resistant pathogens from animals represents an important public health concern. Human gastroenteritis caused by salmonellosis is usually self-limiting. However, antimicrobial therapy may be necessary in invasive infections, especially in immunocompromised patients. In these cases, antimicrobial-resistant strains can play a critical role in the success or failure of treatment.

Subtyping of antimicrobial-resistant *Salmonella* Enteritidis is crucial in epidemiological studies and might

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help to provide the relatedness of strains. Because *Salmonella* Enteritidis is known as a clonal pathogen, molecular and phenotypic methods have been associated for strain characterization to improve discriminatory power. A range of molecular fingerprinting approaches is available for *Salmonella* characterization, as amplified fragment length polymorphism (Torpdahl et al., 2005), repetitive sequence-based PCR (Gebreyes et al., 2006), multilocus sequence typing (Torpdahl et al., 2005), and pulsed-field gel electrophoresis (PFGE; Torpdahl et al., 2005; Gebreyes et al., 2006; Valdezate et al., 2007). Pulsed-field gel electrophoresis is a well-standardized fingerprinting method for bacterial genome analysis, which might provide *Salmonella* Enteritidis strain subdivision. Because it has been reported as a reproducible approach, macrorestriction analysis has been widely used in epidemiological surveillance of antimicrobial-resistant *Salmonella* strains (Bakeri et al., 2003; Cardinale et al., 2005).

Until recently, *Salmonella* Enteritidis strains have been reported to be sensitive to most of the antimicrobial agents tested (Antunes et al., 2003). Overall, studies have shown low or moderate levels of antimicrobial resistance in *Salmonella* Enteritidis isolated in Brazil (Tavechio et al., 1996; de Castro et al., 2002; Fernandes et al., 2003; de Oliveira et al., 2006), although high levels have also been reported (Dias de Oliveira et al., 2005). The poultry industry in southern Brazil is an important segment due to poultry meat exports to many countries, which lend increasing importance to antibiotic-resistance surveillance. The aim of this study was to investigate the antimicrobial resistance, PT, and PFGE patterns in *Salmonella* Enteritidis isolated from humans and food samples involved in salmonellosis outbreaks and also from poultry-derived products of southern Brazil.

MATERIALS AND METHODS

Salmonella Enteritidis Strains

A total of 96 *Salmonella* Enteritidis isolates were selected so as to represent the years from 1995 up to 2003 in southern Brazil (state of Rio Grande do Sul), a period in which chloramphenicol, penicillins, tetracyclines, and sulfonamides (SF) were banned as growth promoters in food-producing animals. Fifty-three strains were obtained from poultry-related products noninvolved in salmonellosis outbreaks (poultry carcass, raw poultry meat, viscera, and environmental swab) and were isolated in Faculdade de Veterinária (Universidade Federal do Rio Grande do Sul). They were also selected with a view to representing different producing companies and regions of the state. Moreover, 43 strains, which were available from salmonellosis outbreaks in southern Brazil, were isolated by the human health surveillance service in Rio Grande do Sul (Fundação Estadual de Produção e Pesquisa em Saúde, Instituto de Pesquisas Biológicas, Laboratório Central do Estado do Rio

Grande do Sul, Brazil) using its current *Salmonella* isolation protocol. These strains were selected to represent different outbreaks and included 29 isolates from different foods, either related or nonrelated to poultry meat or eggs, as well as 14 strains from humans (food handlers and food-poisoning hospitalized patients).

For *Salmonella* isolation from poultry-related products noninvolved in salmonellosis outbreaks, 1 g of viscera was inoculated into 10 mL of brain heart infusion broth (Merck, Darmstadt, Germany) and tetrathionate broth (Merck), and 0.1 g was inoculated into 10 mL of Rappaport-Vassiliadis broth (Merck). Brain heart infusion broth was incubated at 37°C for 18 to 24 h and selective broth at 37 and 42°C for 18 to 24 h. Environmental swabs and meat meal were inoculated into 1% buffered peptone water and incubated at 37°C for 18 to 24 h, after which 100 µL of each sample was transferred to 10 mL of Rappaport-Vassiliadis broth and 1 mL into 10 mL of tetrathionate broth and incubated at 37 and 42°C for 18 to 24 h. Aliquots of selective broth were streaked onto selective plating media Rambach agar (Merck) and brilliant-green phenol-red lactose sucrose with novobiocin agar (Merck). After 24 h of incubation at 37°C, presumptive *Salmonella* colonies were characterized by biochemical assays and serotyped using slide agglutination tests to establish the somatic antigen. All *Salmonella* Enteritidis strains were confirmed by Laboratório de Referência Nacional para Cólera e Doenças Entéricas, Instituto Oswaldo Cruz, FIOCRUZ (Rio de Janeiro, Brazil).

Antimicrobial Susceptibility Testing

Salmonella Enteritidis strains were investigated for susceptibility to 11 different antimicrobials, representing some of the classes commonly used in human therapy. The agar disk diffusion method was carried out as described in the document M2-A8 of the Clinical and Laboratory Standards Institute (CLSI, 2005). All strains were tested with the following antimicrobial agents (Cefar, São Paulo, Brazil): ampicillin (10 µg), cefaclor (30 µg), ceftazidime (30 µg), nalidixic acid (NX, 30 µg), ciprofloxacin (5 µg), chloramphenicol (30 µg), gentamicin (G, 10 µg), streptomycin (STR, 10 µg), tetracycline (T, 30 µg), SF (300 µg), and trimethoprim-sulfamethoxazole (STX, 1.25-23.75 µg). *Escherichia coli* ATCC 25922 and *Enterococcus faecalis* ATCC 29212 were used as reference strains for disk control.

Phage Typing

Phage typing was performed at the Laboratório de Referência Nacional para Cólera e Doenças Entéricas, Instituto Oswaldo Cruz, FIOCRUZ, using the phage typing scheme standard (Ward et al., 1987). The phage set was provided by the World Health Organization Reference Center for Phage Typing (Colindale, London, UK). The serological typing was performed by the slide agglutination method using antisera prepared

by Laboratório de Referência Nacional para Cólera e Doenças Entéricas.

DNA Macrorestriction Analysis

Whole-cell DNA for PFGE was obtained as described previously (Olsen et al., 1994). The DNA in agarose plugs was incubated for 12 h with 20 U of *Xba*I (Fermentas Life Sciences, Burlington, Canada). Pulsed-field gel electrophoresis was performed according to a previously described protocol (Peters et al., 2007). The DNA fragments were electrophoresed in a CHEF-DR II system (BioRad, Hercules, CA) on a PFGE-certified 1% agarose gel (BioRad) with $0.5 \times$ Tris-borate-EDTA as running buffer for 22 h at 6 V/cm. The pulse time was increased from 2 to 64 s. *Salmonella* Typhimurium LT2 DNA (Liu et al., 1993) digested with *Xba*I was used as size standard. Agarose gel was stained by ethidium bromide (1 μ g/mL, Invitrogen, Carlsbad, CA) and registered by digital capture (UltraLum, Paramount, CA). Macrorestriction patterns were compared by GelCompar software package (Applied Maths, Sint-Martens-Latem, Belgium). Similarity was calculated by the Dice coefficient, whereas a dendrogram was obtained by cluster analysis using the unweighted pair group method with arithmetic averages.

Statistical Analysis

Statistical analyses were performed using the WINPEPI software (PEPI Computer Programs for Epidemiologists, Salt Lake City, UT). Multiple pairwise comparisons were performed using the Tukey procedure, assisted by the Describe module version 1.55 (PEPI Computer Programs for Epidemiologists). Comparison between groups was performed by Compare2 module version 1.45 (PEPI Computer Programs for Epidemiologists), using the χ^2 test or Fisher's exact test when appropriate.

RESULTS

Antimicrobial Susceptibility

Of the 96 *Salmonella* Enteritidis strains analyzed, 42 (43.75%) were sensitive to all antimicrobial agents tested. All strains were susceptible to ampicillin, cefaclor, ceftazidime, ciprofloxacin, and chloramphenicol. In contrast, resistance to 1, 2, or 3 antimicrobial drugs was detected among *Salmonella* Enteritidis isolates from poultry-derived products (69.81%), food samples involved in salmonellosis outbreaks (44.83%), and patients (28.57%). The poultry strains had higher resistance rates than the human strains ($P < 0.05$). The highest percentage of resistance was identified against SF, STX, and NX; in turn, a lower percentage of resistance was observed against STR, G, and T (Table 1).

Multiresistance, which was defined as resistance to 3 antimicrobials displayed by a tested isolate, was detect-

ed in 3 *Salmonella* Enteritidis strains (Table 2), with 2 strains being isolated from poultry viscera and 1 strain from a food-poisoning patient (NXSFSTX, STRGSF, and TSFSTX, respectively).

The distribution of antimicrobial resistance in terms of the years considered showed resistance to SF and STX in strains isolated in all years, except 1997 and 2001, respectively. Resistance to NX was observed in strains isolated from 1997 until 2003. Furthermore, *Salmonella* Enteritidis strains resistant to STR and G were isolated in 1999, whereas resistance to T was found in a strain isolated in 1997 (Table 1).

A total of 11 antimicrobial resistance patterns were identified (Table 2), the predominant pattern (SF) and also STX were observed in strains isolated from poultry, humans, and food sources. Isolates from food samples obtained from salmonellosis outbreaks had higher rates ($P < 0.01$) of resistance to STX than isolates from poultry-derived products nonrelated to outbreaks. Resistance to T was identified only in 1 strain isolated from a food-poisoning patient, which showed the TSF-STX resistance pattern. Furthermore, the remaining antimicrobial profiles were primarily associated with strains isolated from poultry-related products. Thus, resistance against NX, G, and STR was observed only in poultry-derived products nonrelated to outbreaks. A higher number of strains isolated from food was resistant to NX than the strains isolated from poultry ($P < 0.001$), but only a marginal difference was noted between strains from humans and poultry ($P < 0.05$).

PT

Of the 96 *Salmonella* Enteritidis strains included in this study, 86 isolates (89.60%) were assigned to PT4 and 5 isolates (5.21%) belonged to PT1. The type PT6a was identified in 2 (2.08%) strains from foods involved in salmonellosis outbreaks, which were both related and nonrelated to poultry meat or eggs, whereas PT4a was found in 1 (1.04%) strain also isolated from food involved in salmonellosis outbreak. *Salmonella* Enteritidis PT4 was distributed between strains isolated from poultry products, food, and humans (Table 2).

PFGE

Electrophoresis of *Xba*I-digested DNA from the 96 *Salmonella* Enteritidis strains displayed 8 closely related macrorestriction genotypes, called X1 to X8 (Figure 1). The predominant macrorestriction pattern (X1) was shared by 79 (82.29%) strains isolated from poultry products, humans, and foods (Table 2). Such genotype was present in samples collected in all of the years considered. The X2 (6.25%) pattern was identified only in isolates from poultry products and foods, and every strain had a different antimicrobial resistance profile. The X8 (5.26%) pattern was detected in poultry strains nonrelated to outbreaks, which were isolated in 2003 from nearby cities and that also displayed 2 antimicro-

Table 1. Number of *Salmonella* Enteritidis strains displaying antimicrobial resistance according to the source and year of isolation

Source (no. of strains analyzed/yr)	Resistance to antimicrobial, ¹ no. (%)					
	SF	STX	NX	STR	G	T
Food related to poultry meat or eggs (9/1996)	2 (22.22)	0	0	0	0	0
Food nonrelated to poultry meat or eggs products (20/1995, 1996)	11 (55.0)	2 (10.0)	0	0	0	0
Food-poisoning patients (10/1995, 1996)	3 (30.0)	2 (20.0)	0	0	0	1 (10.0)
Food handlers (4/1995)	1 (25.0)	1 (25.0)	0	0	0	0
Poultry carcass (20/1995, 1996, 1999)	6 (30.0)	5 (25.0)	1 (5.0)	0	0	0
Raw poultry meat (11/1995, 1996, 1997, 2003)	3 (27.27)	6 (54.54)	4 (36.36)	0	0	0
Poultry viscera (19/1997, 1998, 1999, 2001, 2003)	6 (31.57)	8 (42.10)	7 (36.84)	2 (10.52)	1 (5.26)	0
Environmental swab (3/1998, 1999)	1 (33.33)	0	2 (66.66)	0	0	0
Total	33 (34.37)	24 (25.0)	14 (14.58)	2 (2.08)	1 (1.04)	1 (1.04)

¹SF = sulfonamide; STX = trimethoprim-sulfamethoxazole; NX = nalidixic acid; STR = streptomycin; G = gentamicin; T = tetracycline.

bial phenotypes (NXSF and NXSTX). The X5 (2.08%) pattern was identified in 2 strains isolated from foods nonrelated to poultry meat or eggs obtained from dif-

ferent cities in 1996, which shared the same antimicrobial pattern. The remaining patterns (X3, X4, X6, and X7) were represented by single strains, isolated from

Table 2. Distribution of antimicrobial resistance, phage types, and pulsed-field gel electrophoresis (PFGE) patterns from *Salmonella* Enteritidis strains isolated from poultry, human, and food

Antimicrobial resistance pattern ¹ (%)	Phage type ²	PFGE pattern ³	Poultry-derived products noninvolved in salmonellosis outbreaks				Humans involved in salmonellosis outbreaks		Foods involved in salmonellosis outbreaks	
			Poultry carcass	Raw poultry meat	Viscera	Environmental swab	Human	Food handler	Related to poultry meat or eggs	Nonrelated to poultry meat or eggs
Fully sensitive (43.75%)	PT4	X1	6	3	3	1	7	1	4	8
	PT4	X2	1		1				1	
	PT4	X6								1
	PT4	X7							1	
	PT1	X1						2		
SF (19.79%)	PT6a	X1							1	
	ND	X1	1							
	PT4	X1	6		1		1		2	6
SFSTX (6.25%)	PT4	X5								2
	PT1	X2								1
	PT4	X1		2			1			1
STX (12.50%)	PT4a	X1						1	1	
	PT6a	X1								1
	PT4	X1	4	2	4					
TSFSTX (1.04%)	PT4	X3	1							
	PT4	X4			1					
	PT4	X1					1			
NXSF (5.21%)	PT4	X1			2	1				
	PT4	X8		1						
	ND	X8			1					
NXSTX (3.12%)	PT4	X8		1						
	PT1	X8		1	1					
NXSFSTX (1.04%)	PT4	X1			1					
	PT4	X1	1	1	2	1				
STRGSF (1.04%)	PT4	X2			1					
	PT4	X2			1					
STRSF (1.04%)	PT4	X2			1					
	PT4	X2			1					

¹SF = sulfonamide; STX = trimethoprim-sulfamethoxazole; T = tetracycline; NX = nalidixic acid; STR = streptomycin; G = gentamicin.

²PT = phage type; ND = phage typing was not performed.

³Pulsed-field gel electrophoresis patterns obtained with *Xba*I.

poultry carcass, poultry viscera, food nonrelated to poultry, and food that included poultry components, respectively.

DISCUSSION

In the present study, antimicrobial resistance, PT, and PFGE patterns were evaluated in *Salmonella* Enteritidis isolated from poultry-derived products and also from humans and foods involved in salmonellosis outbreaks in southern Brazil. All strains were susceptible to ampicillin, cefaclor, ceftazidime, ciprofloxacin, and chloramphenicol, which have been some of the antimicrobials of choice for human therapy in *Salmonella* infections over the years. This result is in accordance with previous observations that *Salmonella* Enteritidis is more susceptible to antimicrobials when compared with other *Salmonella* serovars (Antunes et al., 2003). In spite of that, the judicious use of antimicrobial drugs in veterinary medicine has been consistently discussed to prevent the increase of antimicrobial resistance in pathogenic and commensal bacteria. For these reasons, some antimicrobial agents such as chloramphenicol and nitrofurans have been banned from use in food-producing animals worldwide (Schwarz et al., 2001).

Sulfonamide (34.37%) showed the highest resistance rate (Table 1). Sulfonamide resistance has been reported in *Salmonella* Enteritidis worldwide. A low percentage of SF resistance in strains from humans, foods, and animals was identified in Italy (Busani et al., 2004), whereas a higher frequency was identified in strains isolated from a Spanish poultry slaughterhouse (99.0%; Carramiñana et al., 2004) and in Brazil (75.8%; Dias de Oliveira et al., 2005). In Brazil, penicillin, T, SF, and chloramphenicol were banned as animal growth promoters in 1998, although their use is still allowed for therapeutic purposes in food-producing animals. It should be remembered that SF was widely used for

growth promotion in Brazilian poultry until 1998, when such antimicrobials were banned for this purpose. However, the former adoption of this drug for continued use may still be the cause of resistance observed in strains isolated from poultry in years after 1998. On the other hand, resistance phenotypes detected less frequently in this study, such as aminoglycoside resistance, were not observed in strains isolated after the growth promoter banning and may reflect that a low selection pressure was exerted.

Salmonella Enteritidis resistant to NX (14.58%) was identified only in strains isolated from poultry. Nalidixic acid is commonly used in poultry therapy in Brazil. Thus, a higher NX resistance level was expected in isolates from poultry-related products. Indeed, there was a difference ($P < 0.001$) in NX resistance between strains isolated from poultry nonrelated to salmonellosis outbreaks and *Salmonella* Enteritidis isolated from foods; similarly, a difference ($P < 0.05$) was also observed between *Salmonella* Enteritidis isolated from poultry nonrelated to outbreaks and humans. High rates of NX resistance have been observed in *Salmonella* Enteritidis worldwide (Busani et al., 2004; de Oliveira et al., 2006; Soler et al., 2006; Zaidi et al., 2006; Valdezate et al., 2007). It was suggested that an increase in the incidence of quinolone-resistant strains from animals was linked to the licensed utilization of NX in food-producing animals (Malorny et al., 1999). Fluoroquinolones, especially ciprofloxacin, pose a particular concern for human therapy (Mølbak et al., 2002; Threlfall et al., 2006). Although resistance to ciprofloxacin was not identified here nor in previous studies conducted in Brazil (Peresi et al., 1998; Dias de Oliveira et al., 2005), we must bear in mind that the emergence of resistance to narrow-spectrum quinolones, such as NX (Mølbak et al., 2002), and decreased susceptibility to fluoroquinolones have increased among *Salmonella* spp. from food animals and human infections (Malorny et al., 1999).

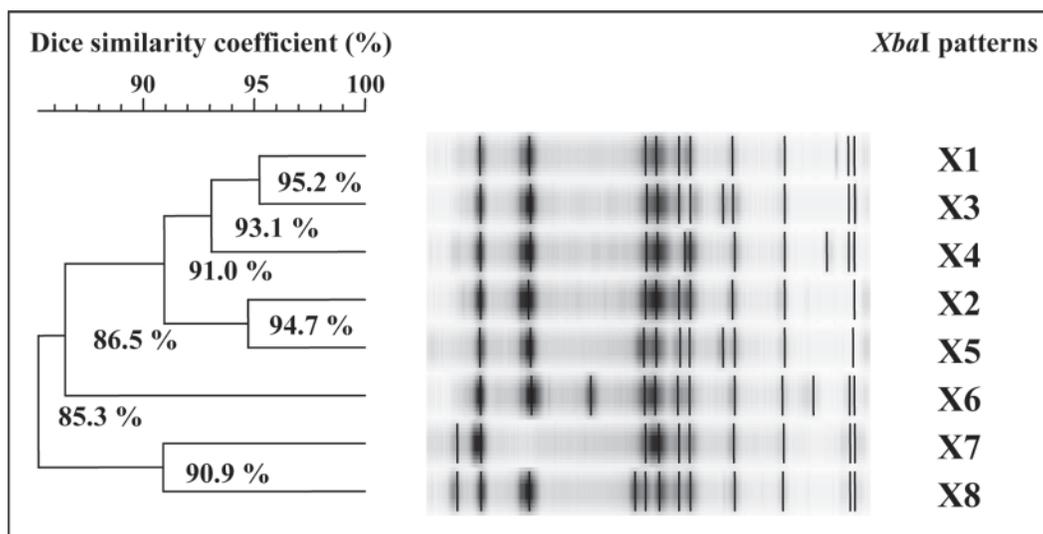


Figure 1. Macroestriction patterns of *Salmonella* Enteritidis isolates obtained with *Xba*I. Lanes 1 to 8 = representative X1 to X8 pulsed-field gel electrophoresis patterns. The dendrogram displays the relationships of the *Salmonella* Enteritidis *Xba*I patterns. Similarity analysis was performed using the Dice coefficient and the clustering was generated by the unweighted pair group method with arithmetic averages.

The multiresistance pattern, identified in only 3 strains, was less frequent than that reported in a previous study conducted with *Salmonella* Enteritidis strains from the same region over the period 1995 to 1997 (Dias de Oliveira et al., 2005). This finding may also reflect the control of antimicrobial prescription implemented a decade ago, and since then, the occurrence of multiresistant *Salmonella* Enteritidis has been more often associated with hospitalized patients (Fernandes et al., 2003). The percentage of *Salmonella* Enteritidis resistant to 3 or more drugs is also low in European countries (Threlfall et al., 2003; Valdezate et al., 2007), whereas multiple resistance has been more commonly observed in *Salmonella* Typhimurium (Busani et al., 2004; Ghilardi et al., 2006; Valdezate et al., 2007), specially *Salmonella* Typhimurium DT104 clones (Gebreyes et al., 2006). Moreover, resistance in *Salmonella* Enteritidis seems consistently responsive to antimicrobial selection pressure, whereas *Salmonella* Typhimurium DT104 resistance is not correlated with antimicrobial use in food-producing animals (Rabsch et al., 2001).

Most of the *Salmonella* Enteritidis strains isolated from poultry and also salmonellosis outbreaks were assigned as PT4 (89.60%). Previous studies have already shown PT4 to be the most prevalent PT in strains from different sources in Brazil (dos Santos et al., 2003; Nunes et al., 2003) and in European countries (Peters et al., 2007), except in Spain, where PT1 was the most prevalent (Valdezate et al., 2007). *Salmonella* Enteritidis PT4 has been associated worldwide with poultry (Coyle et al., 1988; Rabsch et al., 2001; Peters et al., 2007) and may have had a single common source dispersed through international trade of breeders (Rabsch et al., 2001). Other PT identified in the present study were also previously reported in Brazil (dos Santos et al., 2003; Nunes et al., 2003), and PT1, the second most prevalent PT in this study, was identified in strains from poultry and also human outbreaks. The emergence of other PT has been reported in Western Europe (Fisher, 2004), demonstrating that the phage profile can change over time.

Similarly, the majority of the poultry, food, and human *Salmonella* Enteritidis strains (82.29%) shared a common PFGE pattern (X1). The predominant X1 pattern was observed in different PT (Table 2), and a same PT presented different PFGE pattern. In a previous study, *Xba*I patterns were likewise not limited to a single PT, although a strong association of PT4 with a single PFGE pattern was observed (Peters et al., 2007). *Salmonella* Enteritidis strains usually display a genetic homogeneity, which is evidenced by a prevalent clone (Bakeri et al., 2003; Valdezate et al., 2007). Although PFGE was able to discriminate some of the PT4 isolates, the *Salmonella* Enteritidis strains analyzed in the present study had only minor pattern differences. The association of other fingerprinting approaches, as multilocus sequence typing or amplified fragment length polymorphism, might have improved the discrimination between the *Salmonella* Enteritidis strains analyzed.

However, these more expensive and time-consuming techniques have showed similar or lower discriminatory power when compared with PFGE for *Salmonella* subtyping (Torpdahl et al., 2005; Gebreyes et al., 2006) and were not available in our study.

The predominant patterns X1 and X2 were both observed in resistant and sensitive strains isolated from poultry products, foods, and humans. Because some strains shared identical PT and PFGE genotype but different antimicrobial resistance patterns, it is possible that such differences in antimicrobial susceptibility were associated to recent genetic changes, such as plasmid acquisition, which were insufficient to change the PFGE profile. As previously reported, this association is difficult to find because of the complexity of transmission routes from food production animals to humans and of temporal changes in the occurrence of resistance to antimicrobials (Phillips et al., 2004).

The occurrence of a dominant PT and PFGE patterns distributed between strains from poultry-related products and also strains involved in salmonellosis outbreaks in southern Brazil suggests that common clonal groups are present in poultry and in human salmonellosis outbreaks. Because antimicrobial resistance in *Salmonella* Enteritidis strains from poultry-related products was significantly higher than the resistance observed in strains from humans involved in salmonellosis outbreaks, continued antimicrobial resistance surveillance should be taken into consideration. The early identification of potential resistance patterns emerging by monitoring programs might be a factor to be considered in the policies for use of antimicrobial agents in food-producing animals in Brazil.

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