

## Research Note

# Resistance Phenotypes and Genotypes of *Salmonella enterica* subsp. *enterica* Isolates from Feed, Pigs, and Carcasses in Brazil

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## ABSTRACT

*Salmonella enterica* subsp. *enterica* plays a role as a foodborne pathogen worldwide. The consumption of contaminated pork has been associated with human salmonellosis and the increase in antimicrobial resistance among *Salmonella* from pigs and pork products is a concern. A total of 225 *Salmonella* isolates from feed mills, the lairage environment, and the intestinal contents of pigs and carcasses were investigated for their antimicrobial susceptibility. A MIC for ciprofloxacin was screened by agar dilution, and antimicrobial resistance genes were investigated by PCR assays. Among the tested isolates, 171 (76%) showed resistance to at least one antimicrobial agent, and 91 (40.4%) were multiresistant. Resistance occurred most frequently to tetracycline (54.5%), sulfonamides (39.6%), and streptomycin (33.7%). Thirty-two (94.1%) nalidixic acid-resistant isolates exhibited decreased susceptibility to ciprofloxacin. The resistance genes found were *bla*<sub>TEM</sub> (ampicillin), *tet*(A) (tetracycline), *tet*(B) (tetracycline/minocycline), *sul1*, *sul2*, and *sul3* (sulfonamides), *catA1* (chloramphenicol), *floR* (florfenicol/chloramphenicol), *strA* and *strB* (streptomycin), *aph*(3')-Ia (kanamycin), *aac*(3)-IIa and *aac*(3)-IVa (apramycin/gentamicin), *aadA* variant (streptomycin/spectinomycin), and *dfrA1* (trimethoprim). *Salmonella* isolates from pig feces and carcasses displayed a higher frequency of resistance to most antimicrobials tested than isolates from feed mills. Common resistance gene profiles were found in isolates from the lairage and the intestinal content of pigs and carcasses, demonstrating that resistance genes selected on farms may be found in pork.

*Salmonella enterica* subsp. *enterica* figures among the leading causes of foodborne diseases in humans worldwide. Pigs have been recognized as a source of *Salmonella*, and the consumption of contaminated pork has been associated with a significant number of human cases of salmonellosis (7). Besides carcasses and pork, *Salmonella* isolation has also been reported in feedstuffs and feed mills (7, 21). In Brazil, *Salmonella* infection in swine herds has been demonstrated (30), and a relatively high prevalence of *Salmonella* isolation has been found in pigs at slaughter (17), as well as in carcasses (28).

Antimicrobial resistance has been considered one of the main concerns to human and animal health. The increased frequency of antimicrobial-resistant *Salmonella* isolates, particularly those that are resistant to multiple antimicrobial agents, has been reported in pigs and pork products (5, 6, 12). In southern Brazil, resistance to antimicrobial agents among porcine *Salmonella* has also been reported and characterized (1, 17, 19). Although multiresistant *Salmonella* has been isolated from pigs at slaughter (17, 19), the resistance pattern of isolates originating from feed and carcasses has been less thoroughly investigated in this region.

Thus, this study aimed to assess the antimicrobial resistance frequency in *Salmonella enterica* subsp. *enterica* isolated from different steps of the swine production system (feed, environment, and intestinal content of pigs and carcasses) and to determine the antimicrobial resistance profiles.

## MATERIALS AND METHODS

**Origin of isolates.** During the period of 2008 to 2011, a total of 1,771 samples were collected from different steps of the pig production chain in Brazil for monitoring and surveillance studies (21, 22, 28). Samples were taken from the lairage environment, the intestinal content of pigs, and from carcasses at five slaughterhouses, which were supplied by several finishing farms located in different regions of southern Brazil. In the same period, samples were taken from different steps of the feed manufacturing process at four feed mills. The feed manufacturing facilities belonged to four large swine producing companies, which supplied several large swine companies in the same region. All samples were submitted to a *Salmonella* isolation protocol (21, 22, 28) consisting of nonselective preenrichment, selective enrichment, and plating onto selective solid medium. *Salmonella* isolates were serotyped at the National Reference Centre, Instituto Oswaldo Cruz, Rio de Janeiro, Brazil, according to the Kauffmann-White scheme. A total of 225 *Salmonella* isolates from the aforementioned studies were selected for resistance testing: 54 *Salmonella* isolates from feed mills, 30 from lairage environments, 25 from the intestinal content

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of pigs, and 116 from carcasses. All isolates were kept frozen until testing.

**Antimicrobial susceptibility testing.** *Salmonella* isolates were tested for antimicrobial susceptibility against 12 different antimicrobial agents, including the antimicrobials important for human salmonellosis treatment. The agar disc diffusion method was performed and evaluated according to the specifications of the Clinical and Laboratory Standards Institute, documents VET01-S2 (3) and M100-S23 (4). The following discs (Oxoid, Basingstoke, UK) were used: ampicillin (AMP 10 µg), cefotaxime (CTX, 30 µg), ceftazidime (CAZ, 30 µg), chloramphenicol (CHL, 30 µg), ciprofloxacin (CIP, 5 µg), gentamicin (GEN, 10 µg), kanamycin (KAN, 30 µg), nalidixic acid (NAL, 30 µg), streptomycin (STR, 10 µg), sulfonamides (SUL, 300 µg), tetracycline (TET, 30 µg), and trimethoprim (TMP, 5 µg). *Escherichia coli* ATCC 25922 was used as a reference strain for quality control purposes. Antimicrobial multiresistance was defined as resistance to three or more classes of antimicrobial agents. In multiresistant isolates displaying resistance to NAL, a MIC for CIP was screened by agar dilution following the recommendations of the Clinical and Laboratory Standards Institute document M07-A9 (2).

**Detection of antimicrobial resistance genes.** *Salmonella* isolates that displayed multiresistance profile on the disc diffusion testing were investigated for the presence of several resistance genes previously reported in this genus. Genomic DNA for the PCR assays was prepared by the thermal lysis procedure (single colonies resuspended in 200 µl of double-distilled water were heated for 5 min at 95°C and centrifuged for 10 min at 1,500 × g) or by using the NucleoSpin Tissue Kits (Macherey-Nagel, Düren, Germany). Genes coding class 1 and 2 integrase and resistance to β-lactams, aminoglycosides, TET, SUL, phenicols, and TMP were investigated by PCR assays (Table 1).

**Statistical analysis.** The antimicrobial resistance frequencies of *Salmonella* isolates from different origins were compared by chi-square test ( $\chi^2$ ) using the Stata Data Analysis and Statistical Software, version 12.0 (College Station, TX). A *P* value of <0.05 was considered significant. For the purpose of this study, isolates with intermediate susceptibility were categorized as susceptible for statistical analysis (5).

## RESULTS

**Antimicrobial resistance phenotypes.** Of the 225 *Salmonella* isolates included in this study, 171 (76%) showed resistance to at least one antimicrobial agent (Table 2). Resistance occurred most frequently to TET (54.5%), SUL (39.6%), STR (33.7%), and NAL (33.3%). All isolates were susceptible to CTX or CAZ, and a low frequency (<1%) of resistance to CIP was observed. *Salmonella* isolates from feed mills were significantly less resistant to AMP, CHL, GEN, TET, STR, SUL, TMP, and KAN (*P* < 0.05).

Resistance to three or more classes of antimicrobial agents (40.4%; 91 of 225) was detected in varying combinations (Table 3). Among the feed mill isolates, 23 (42.6%) were resistant to one or two antimicrobial agents, and only one (1.9%) isolate showed multiresistance. On the other hand, among the 30 isolates from the lairage, 22 (73.3%) were resistant to at least one antimicrobial, and 12 (40%) were multiresistant. A similar resistance rate was

observed for intestinal content (18 of 25; 72%) and carcass isolates (10 of 116; 89.7%). A high frequency of multiresistance was observed among intestinal (64%) and carcass isolates (53.4%). A CIP MIC was determined for 34 multiresistant isolates that showed resistance to NAL. Thirty-two (94.1%) isolates exhibited decreased CIP susceptibility (MIC 0.125 to 2 µg·ml<sup>-1</sup>). Among these isolates, 10 (31.2%) exhibited a quite high MIC (1 to 2 µg·ml<sup>-1</sup>) and 18 (56.2%) presented MIC values of 0.5 µg/ml.

Tested isolates belonged to 28 different *Salmonella* serovars; among them *Salmonella* Typhimurium (76 of 225; 33.8%) and *Salmonella* Derby (49 of 225; 21.8%) were the most prevalent. Among the two most common serovars, 46 (60.5%) *Salmonella* Typhimurium and 27 (55.1%) *Salmonella* Derby isolates showed multiresistance. The pattern AMP-NAL-STR-SUL-TET-TMP was the most frequent and was found in 11 *Salmonella* Typhimurium isolates from carcasses. The multiresistance profile STR-SUL-TET was identified in 27 *Salmonella* Derby isolates originated from intestinal content and carcasses (Table 3).

**Antimicrobial resistance genes profiling.** A variety of antimicrobial resistance gene profiles were detected among the 91 multiresistant isolates, and the following resistance genes were detected: *tet(A)* (67.0%); *bla*<sub>TEM</sub> (64.8%); *aadA* variant (60.4%); *sul1* (51.6%); *tet(B)* (32.9%); *catA1* (32.9%); *strA-strB* (29.6%); *aph(3')-Ia* (28.6%); *aac(3)-IIa* (18.7%); *floR* (18.7%); *sul2* (15.4%) and *sul3* (12.1%); and *aac(3)-IVa* (5.5%) (Table 3). The genes *aadB*, *bla*<sub>PSE-1</sub>, *tet(G)*, and all of the TMP resistance genes tested, except *dfrA1*, were not found among the *Salmonella* isolates. Forty-one (45.1%) isolates were positive for the class 1 integrase gene (*int1*), while class 2 integrons were not detected.

## DISCUSSION

In the current study, 76% of the isolates were resistant to at least one antimicrobial agent, and multiresistance was observed in 40.4% of the isolates. The occurrence of resistant *Salmonella* isolates in pigs and pork represents a relevant issue with regard to food safety and consumer protection. Porcine *Salmonella* isolates with a high rate of antimicrobial resistance were reported in several countries (6, 12); however, resistance patterns may differ across geographical boundaries and swine production systems. For this reason, national studies are essential for providing information on the epidemiological status of the region. In Brazil, antimicrobial resistance surveillance targeted at the swine production chain has not yet been conducted. However, the nationwide *Salmonella* surveillance for antimicrobial resistance conducted in chicken (23) demonstrated that all 250 *Salmonella* isolates, derived from 2,710 samples collected at the retail level, presented resistance to at least one antimicrobial tested, and 69.3% were multiresistant. Even considering that the antimicrobials tested and methodologies adopted were different from our study, the results demonstrate in both cases that antimicrobial resistance in *Salmonella* should be monitored in Brazil.



TABLE 1. PCR primers used in this study

Antimicrobial agent class	Gene	Amplicon size (bp)	Sequence (5'-3') <sup>a</sup>	Reference
Aminoglycosides	<i>aac(3)-IIa</i>	369	fw: TGAAACGCTGACGGAGCCTC rv: GTCGAACAGGTAGCACTGAG	26
	<i>aac(3)-IVa</i>	627	fw: GTGTGCTGCTGGTCCACAGC rv: AGTTGACCCAGGGCTGTCCG	26
	<i>aph(3')-Ia</i>	669	fw: AACGTCTTGCTCGAGGCCGCG rv: GGCAAGATCCTGGTATCGGTCTGC	26
	<i>aadB</i>	328	fw: GGGCGCGTCATGGAGGAGTT rv: TATCGCGACCTGAAAGCGGC	26
	<i>aadA</i>	526	fw: GTGGATGGCGGCCTGAAGCC rv: ATTGCCCAGTCGGCAGCG	26
	<i>strA</i>	645	fw: TGACTGGTTGCCTGTCAGAGG rv: CCAGTTGTCTTCGGCGTTAGCA	16
	<i>strB</i>	510	fw: ATCGTCAAGGGATTGAAACC rv: GGATCGTAGAACATATTGGC	18
β-Lactams	<i>bla<sub>TEM</sub></i>	851	fw: ATGAGTATTCAACATTTCCG rv: TTAATCAGTGAGGCACCTAT	11
	<i>bla<sub>PSE-1</sub></i>	419	fw: CGCTTCCCGTTAACAAGTAC rv: CTGGTTCATTTAGATAGCG	27
Phenicol	<i>catA1</i>	551	fw: GGCATTTTCAGTCAGTTG rv: CATTAGCATTCTGCCG	18
	<i>floR</i>	1,291	fw: AGGGTTGATTCGTCATGACCA rv: CGGTTAGACGACTGGCGACT	14
Trimethoprim	<i>dfrA1/dfrA15/dfrA16</i>	414	fw: GATATTCCATGGAGTGCCA rv: ACCCTTTTGCCAGATTTG	9
	<i>dfrA5/dfrA14</i>	379	fw: GATTGGTTGCGCTCCA rv: CTCAAAAACAACCTCGAAGG	9
	<i>dfrA7/dfrA17</i>	345	fw: CAGAAAATGGCGTAATCG rv: TCACCTTCAACCTCAACG	9
	<i>dfrB1/dfrB2/dfrB3</i>	205	fw: CAAAGTAGCGATGAAGCCA rv: CAGGATAAATTTGCACTGAGC	9
Sulphonamides	<i>sul1</i>	839	fw: ATGGTGACGGTGTTCCGGCATCTGA rv: CTAGGCATGATCTAACCCTCGGTCT	10
	<i>sul2</i>	703	fw: ACAGTTTCTCCGATGGAGGCC rv: CTCGTGTGTGCGGATGAAGTC	16
	<i>sul3</i>	803	fw: GAGCAAGATTTTGAATCG rv: CATCTGCAGCTAACCTAGGGCTTTGGA	10
Tetracyclines	<i>tet(A)</i>	953	fw: GTAATTCTGAGCACTGT rv: CCTGGACAACATTGCTT	8
	<i>tet(B)</i>	1,169	fw: ACGTTACTCGATGCCAT rv: AGCACTTGTCTCCTGTT	8
	<i>tet(G)</i>	1,141	fw: CTGCTGATCGTGGGTCT rv: TTGCGAATGGTCTGCGT	8
Class 1 integrase	<i>intI1</i>	871	fw: CGGAATGGCCGAGCAGATC rv: CAAGGTTCTGGACCAGTTGCG	27
Class 2 integrase	<i>intI2</i>	400	fw: ATTAGGCGCGTGGGCAGTAG rv: CGTCATCCTCAGACCATGGGC	27

<sup>a</sup> fw, forward primer; rv, reverse primer.

The risk of antimicrobial resistance in the feed manufacturing process is related to the introduction of resistant strains in pig farms and the potential dissemination of mobile genetic elements, which may be transmitted to other more pathogenic endemic strains (7). Although multiresistant *Salmonella* Senftenberg was found in feed samples collected on farms in Brazil (17), *Salmonella* isolates from feed mills were significantly less resistant to most of the tested antimicrobial agents, and multiresistance

was observed in only one isolate in the current study. Therefore, feed does not seem to play an important role in the introduction of resistance genes on the farm in this region, whereas the selective pressure of antimicrobial use might be a better explanation for the high resistance rates observed in isolates recovered from pigs and the lairage. In Brazil, most of the tested antimicrobial agents are not used as feed additives; however, they are still adopted for prophylactic and therapeutic purposes.



TABLE 2. Antimicrobial resistance in *Salmonella* isolates recovered from feed mills and healthy slaughter pigs

Antimicrobial agent	<i>Salmonella</i> -resistant isolates, no. (%)				Total no. (%) (n = 225)	P value <sup>a</sup>
	Feed mill (n = 54)	Lairage (n = 30)	Intestinal content (n = 25)	Carcass (n = 116)		
Ampicillin	6 (11.1)	11 (36.7)	9 (36.0)	41 (35.3)	67 (29.8)	0.008
Chloramphenicol	1 (1.8)	13 (10.0)	7 (28.0)	21 (18.1)	32 (14.2)	0.006
Gentamicin	1 (1.8)	5 (16.7)	6 (24.0)	12 (10.3)	24 (10.7)	0.017
Nalidixic acid	9 (16.7)	13 (43.3)	3 (12.0)	50 (43.1)	75 (33.3)	<0.001
Tetracycline	2 (3.7)	16 (53.3)	16 (64.0)	88 (75.8)	122 (54.5)	<0.001
Streptomycin	1 (1.8)	8 (26.7)	15 (60.0)	52 (44.8)	76 (33.7)	<0.001
Sulfonamide	12 (22.2)	8 (26.7)	15 (60.0)	54 (46.5)	89 (39.6)	0.001
Trimethoprim	0 (0)	1 (3.3)	1 (4.0)	16 (13.7)	18 (8.0)	0.01
Kanamycin	0 (0)	6 (20.0)	3 (12.0)	24 (20.7)	33 (14.7)	0.004
Cefotaxime	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	—
Ceftazidime	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	—
Ciprofloxacin	0 (0)	1 (3.3)	0 (0)	1 (0.8)	2 (0.9)	0.433

<sup>a</sup> A P value of <0.05 was considered significant. —, analysis not performed.

The highest frequency of resistance was observed against TET (54.5%), SUL (39.6%), and STR (33.7%). In previous studies conducted in Brazil, *Salmonella* isolates from pig and pork displayed resistance against the aforementioned antimicrobials in frequencies ranging from 23 to 90% (1, 17). These drugs have been used for the treatment of respiratory and enteric diseases in swine over the years, justifying the selective pressure observed in the *Salmonella* population. On the contrary, all isolates were susceptible to CTX/CAZ, and a low frequency of resistance to CIP was observed.

Extended spectrum cephalosporins and fluoroquinolones are important categories of antimicrobial agents that are frequently used for treatment of complicated *Salmonella* infections in humans; therefore, special attention has been given to the monitoring of their resistance (5, 6). Particularly, the emergence of extended-spectrum  $\beta$ -lactamases in *Enterobacteriaceae* has been targeted in antimicrobial resistance surveillance worldwide. The reported frequency of extended-spectrum  $\beta$ -lactamase producers among *Salmonella* has, however, been low in studies that surveyed a large number of isolates (25, 31). Therefore, we cannot exclude the presence of extended-spectrum  $\beta$ -lactamase-producing *Salmonella* in Brazil, because we analyzed a relatively low number of isolates.

Regarding the group of quinolones, although only two strains displayed resistance to CIP in our study, 33.3% were resistant against NAL. It has been demonstrated that resistance to NAL is coupled with decreased susceptibility to fluoroquinolones (MIC  $\geq 0.125 \mu\text{g}\cdot\text{ml}^{-1}$ ) in salmonellae (29); therefore, we determined CIP MIC values in multiresistant isolates that displayed resistance against this antimicrobial. Among the 34 isolates tested, 94.1% showed decreased CIP susceptibility (MIC 0.12 to  $2 \mu\text{g}\cdot\text{ml}^{-1}$ ), indicating a tendency toward increase in the number of resistant strains in the future.

A high frequency (53.5%, 62 of 116) of multiresistance in different *Salmonella* serovars isolated from pig carcasses was observed. *Salmonella* strains that contaminate carcasses mainly originate from the lairage and intestinal content (17,

28); therefore, resistant strains selected on farms are expected to be found on the carcass surface. These resistant isolates, in turn, may contaminate pork products and pose a potential risk for consumers. The most prevalent genes, *tet(A)*, *bla*<sub>TEM</sub>, *aadA* variant, and *sulI*, were detected in multiresistant isolates from the lairage and intestinal content of pigs and carcasses, demonstrating that resistance genes selected on farms are present in pork. These genes also predominate in other studies targeting resistance in *Salmonella* (9, 15) and are frequently carried on mobile genetic elements, such as transposons and plasmids (19, 20). The SUL resistance gene *sulI* was mostly detected in isolates carrying the *intl1* gene of class 1 integrons, which are recognized as playing an essential role in the dissemination of multiple antimicrobial resistance genes (20). These genetic elements are widespread in *Salmonella* and usually carry the *sulI* gene in the 3' conserved segment together with *qacA*E1, which encodes a semifunctional derivative of the quaternary ammonium compounds resistance gene *qacE* (20, 27).

Among the various resistance gene clusters associated with class 1 integrons in *Salmonella*, the genomic island associated with the pattern (AMP-CHL-STR/spectinomycin-SUL-TET) of *Salmonella* Typhimurium DT104 is one of the better characterized (9, 20). The backbone of the *Salmonella* genomic island includes the *floR* and *tet(G)* genes flanked by two class 1 integrons containing the *aadA*2 and *bla*<sub>PSE-1</sub> cassettes carried on the chromosome (13). This genotype was not identified among our *Salmonella* isolates, in agreement with previous studies, which indicated that this resistance cluster carried by the phagotype DT104 may have limited epidemiological relevance in Brazil (1, 24).

In our study, the resistance pattern most found among *Salmonella* Typhimurium (AMP-NAL-STR-SUL-TET-TMP) was associated with the *bla*<sub>TEM</sub>, *aadA* variant, *sul3*, and *tet(A)* resistance genes. However, all isolates presenting this resistance pattern were recovered from different carcasses from the same slaughter day (data not shown) and thus may represent a *Salmonella* Typhimurium clone, which has been spread during the holding period,



TABLE 3. The antimicrobial resistance phenotype and genotype patterns of multiresistant *Salmonella* serovars isolates originated from different steps of the swine production chain in southern Brazil

Origin (no. of multiresistant isolates/total)	<i>Salmonella</i> serovar (n)	Resistance phenotype <sup>a</sup>	Resistance genotype
Feed mill (1/53) Lairage (12/30)	Typhimurium (1)	AMP-CHL-STR-SUL-TET	<i>bla</i> <sub>TEM</sub> , <i>catA1</i> , <i>aadA</i> , <i>strA-strB</i> , <i>sul2</i> , <i>tet(B)</i>
	Agona (1)	STR-SUL-TET	<i>aadA</i> , <i>sul1</i> , <i>tet(A)</i> , <i>intI1</i>
	Derby (2)	STR-SUL-TET	<i>aadA</i> , <i>sul1</i> , <i>tet(A)</i> , <i>intI1</i>
	Typhimurium (1)	AMP-GEN-KAN-TET	<i>bla</i> <sub>TEM</sub> , <i>aac(3)-IIa</i> , <i>aph(3')-Ia</i> , <i>tet(A)</i>
	Typhimurium (1)	AMP-NAL-STR-TET-TMP	<i>bla</i> <sub>TEM</sub> , <i>aadA</i> , <i>tet(B)</i>
	Typhimurium (4)	AMP-GEN-KAN-NAL-TET	<i>bla</i> <sub>TEM</sub> , <i>aac(3)-IIa</i> , <i>aph(3')-Ia</i> , <i>tet(A)</i>
	O:6,7 (1)	AMP-CHL-NAL-STR-SUL-TET	<i>bla</i> <sub>TEM</sub> , <i>catA1</i> , <i>floR</i> , <i>strA-strB</i> , <i>sul2</i> , <i>tet(B)</i>
	O:6,7:-:I,w (1)	AMP-CHL-NAL-STR-SUL-TET	<i>bla</i> <sub>TEM</sub> , <i>catA1</i> , <i>floR</i> , <i>strA-strB</i> , <i>sul2</i> , <i>tet(B)</i>
	Panama (1)	AMP-CHL-KAN-NAL-STR-SUL-TET	<i>bla</i> <sub>TEM</sub> , <i>catA1</i> , <i>aph(3')-Ia</i> , <i>strA-strB</i> , <i>sul2</i> , <i>tet(B)</i>
Intestinal content (16/25)	Derby (7)	STR-SUL-TET	<i>aadA</i> , <i>sul1</i> , <i>tet(A)</i> , <i>intI1</i>
	Agona (1)	CHL-STR-SUL-TET	<i>floR</i> , <i>strA-strB</i> , <i>sul2</i> , <i>tet(A)</i>
	Typhimurium (1)	AMP-GEN-KAN-NAL-TET	<i>bla</i> <sub>TEM</sub> , <i>aac(3)-IIa</i> , <i>aph(3')-Ia</i> , <i>tet(A)</i>
	Typhimurium (1)	AMP-GEN-KAN-NAL-STR-TET	<i>bla</i> <sub>TEM</sub> , <i>aac(3)-IIa</i> , <i>aph(3')-Ia</i> , <i>strA-strB</i> , <i>tet(A)</i>
	Senftenberg (1)	AMP-CHL-STR-SUL-TET-TMP	<i>bla</i> <sub>TEM</sub> , <i>floR</i> , <i>strA-strB</i> , <i>sul2</i> , <i>tet(B)</i>
	Typhimurium (1)	AMP-CHL-KAN-STR-SUL-TET	<i>bla</i> <sub>TEM</sub> , <i>catA1</i> , <i>aadA</i> , <i>strA-strB</i> , <i>sul1</i> , <i>tet(B)</i> , <i>intI1</i>
	Typhimurium (4)	AMP-CHL-GEN-STR-SUL-TET	<i>bla</i> <sub>TEM</sub> , <i>catA1</i> , <i>floR</i> , <i>acc(3)-IVa</i> , <i>aadA</i> , <i>sul1</i> , <i>tet(B)</i> , <i>intI1</i>
Carcass (62/116)	Typhimurium (1)	NAL-STR-TET	<i>strA-strB</i> , <i>tet(A)</i>
	Enteritidis (1);	STR-SUL-TET	<i>strA-strB</i> , <i>sul2</i> , <i>tet(B)</i>
	Derby (18)	STR-SUL-TET	<i>aadA</i> , <i>sul1</i> , <i>tet(A)</i> , <i>intI1</i>
	Infantis (1)	AMP-CHL-TET-TMP	<i>bla</i> <sub>TEM</sub> , <i>catA1</i> , <i>floR</i> , <i>tet(A)</i>
	Typhimurium (4)	AMP-GEN-KAN-TET	<i>bla</i> <sub>TEM</sub> , <i>aac(3)-IIa</i> , <i>aph(3')-Ia</i> , <i>tet(A)</i>
	Typhimurium (1)	AMP-NAL-TET-TMP	<i>bla</i> <sub>TEM</sub> , <i>tet(A)</i>
	Typhimurium (1)	CHL-STR-SUL-TET	<i>catA1</i> , <i>floR</i> , <i>aadA</i> , <i>sul1</i> , <i>tet(B)</i> , <i>intI1</i>
	Infantis (1)	AMP-CHL-SUL-TET-TMP	<i>bla</i> <sub>TEM</sub> , <i>catA1</i> , <i>floR</i> , <i>sul1</i> , <i>tet(A)</i>
	Typhimurium (5)	AMP-GEN-KAN-NAL-TET	<i>bla</i> <sub>TEM</sub> , <i>aac(3)-IIa</i> , <i>aph(3')-Ia</i> , <i>tet(A)</i>
	Typhimurium (11)	AMP-NAL-STR-SUL-TET-TMP	<i>bla</i> <sub>TEM</sub> , <i>aadA</i> , <i>sul3</i> , <i>tet(A)</i>
	Typhimurium (1)	AMP-CHL-GEN-STR-SUL-TET	<i>bla</i> <sub>TEM</sub> , <i>catA1</i> , <i>floR</i> , <i>acc(3)-IVa</i> , <i>aadA</i> , <i>sul1</i> , <i>tet(B)</i> , <i>intI1</i>
	O:4,5 (1)	AMP-CHL-KAN-STR-SUL-TET	<i>bla</i> <sub>TEM</sub> , <i>catA1</i> , <i>aadA</i> , <i>strA-strB</i> , <i>sul1</i> , <i>tet(B)</i>
	Panama (4)	AMP-CHL-KAN-STR-SUL-TET	<i>bla</i> <sub>TEM</sub> , <i>catA1</i> , <i>aph(3')-Ia</i> , <i>strA-strB</i> , <i>sul2</i> , <i>tet(B)</i>
	Typhimurium (6)	AMP-CHL-KAN-STR-SUL-TET	<i>bla</i> <sub>TEM</sub> , <i>catA1</i> , <i>aph(3')-Ia</i> , <i>aadA</i> , <i>strA-strB</i> , <i>sul1</i> , <i>tet(B)</i> , <i>intI1</i>
	Ohio (1)	AMP-CHL-NAL-STR-SUL-TET	<i>bla</i> <sub>TEM</sub> , <i>catA1</i> , <i>floR</i> , <i>strA-strB</i> , <i>sul2</i> , <i>tet(B)</i>
	Infantis (1)	AMP-CHL-KAN-NAL-STR-SUL-TET	<i>bla</i> <sub>TEM</sub> , <i>catA1</i> , <i>floR</i> , <i>aph(3')-Ia</i> , <i>strA-strB</i> , <i>sul1</i> , <i>tet(B)</i>
	Panama (2)	AMP-CHL-KAN-NAL-STR-SUL-TET	<i>bla</i> <sub>TEM</sub> , <i>catA1</i> , <i>aph(3')-Ia</i> , <i>strA-strB</i> , <i>sul2</i> , <i>tet(B)</i>
			<i>bla</i> <sub>TEM</sub> , <i>catA1</i> , <i>floR</i> , <i>aph(3')-Ia</i> , <i>strA-strB</i> , <i>sul1</i> , <i>tet(B)</i>
	Typhimurium (1)	AMP-CHL-GEN-NAL-STR-SUL-TET-TMP	<i>bla</i> <sub>TEM</sub> , <i>catA1</i> , <i>floR</i> , <i>strA-strB</i> , <i>sul2</i> , <i>tet(B)</i>
	Typhimurium (1)	AMP-CHL-CIP-GEN-NAL-STR-SUL-TET-TMP	<i>bla</i> <sub>TEM</sub> , <i>catA1</i> , <i>floR</i> , <i>aac(3)-IIa</i> , <i>strA-strB</i> , <i>sul1</i> , <i>tet(A)</i> , <i>dfra1</i>

<sup>a</sup> AMP, ampicillin; CHL, chloramphenicol; STR, streptomycin; SUL, sulphonamide; TET, tetracycline; GEN, gentamicin; KAN, kanamycin; NAL, nalidixic acid; TMP, trimethoprim; CIP, ciprofloxacin.



rather than a disseminated gene cluster. In contrast, the most common resistance pattern (STR-SUL-TET) found in the 27 *Salmonella* Derby isolates that originated from pig feces and carcasses from different slaughterhouses was highly associated with the presence of genes *aadA* variant, *sul1*, *tet(A)*, and *int11*. This genotype was already reported in isolates of *Salmonella* Derby from the same region (19), which may have been circulating in pig farms in the last decade.

Another resistance gene cluster reported in *Salmonella* is the *strA-strB* linked with the *sul2* gene, frequently carried on plasmids (20). In our study, this genotype was detected in isolates belonging to various serovars obtained from feed, lairage, intestinal content, and carcasses, demonstrating that it is widespread in the Brazilian swine production chain. Although we did not determine the location of the genes, they might also be carried on plasmids, such as RSF1010, which proved to be found in many bacteria species, isolated from human, animal, and environmental samples (20, 32).

In conclusion, *Salmonella* isolates from pig feces and carcasses displayed a high frequency of resistance to most of the antimicrobial agents tested, while isolates originating from feed mills presented a much lower frequency of resistance. This result indicates that selective pressure is being exerted on farms. The high rate of antimicrobial resistance and the common resistance gene profiles found in *Salmonella* isolates from pig feces and carcasses highlight the hazard of their transmission through the food chain.

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