Brazilian Journal of Poultry Science
Revista Brasileira de Ciência Avícola

ISSN 1516-635X 2023 / v.25 / n.1 / 001-006

http://dx.doi.org/10.1590/1806-9061-2022-1628

Original Article

High Genetic Similarity Among Salmonella Heidelberg Isolated from Poultry Farms, Wild Animals, Beef, Poultry and Pork Meat, and Humans in Brazil

■Author(s)

Santos AFM[I] https://orcid.org/0000-0002-7067-2852
Machado SCA[II] https://orcid.org/0000-0002-4617-4239
Dias TS[II] https://orcid.org/0000-0001-8815-3316
Rodrigues DP[I] https://orcid.org/0000-0003-1101-5518
Pereira VLA[II] https://orcid.org/0000-0003-2197-8916

[I] Oswaldo Cruz Institute, Brasil Av., 4365, 21040-900, Rio de Janeiro, Brazil.
[II] Faculty of Veterinary Medicine, Federal Fluminense University, Vital Brasil Filho Street, 64, 24230-340, Niterói, Brazil.

■Mail Address

Corresponding author e-mail address
André Felipe Santos
Oswaldo Cruz Institute, Brasil Av., 4365, 21040-900, Rio de Janeiro, RJ, Brazil.
Phone: +55 021-2562-1651
Email: nanet@ioc.fiocruz.br

■Keywords

Salmonella Heidelberg, pulsed-field gel electrophoresis, public health, foodborne pathogens, one health.

Submitted: 31/January/2022
Approved: 14/September/2022

ABSTRACT

Salmonellosis is an important gastrointestinal infection in humans and cause of foodborne outbreaks in the world. In this context, molecular characterization is essential to understand how the strains circulate. The aim of this study was to evaluate the genotypic distribution of *S.* Heidelberg according to the source of isolation. The genetic relatedness of the *S.* Heidelberg isolates was determined by pulsed-field gel electrophoresis (PFGE). The most prevalent pulsotypes of cluster A were BRJF6X01.006 (27/95 = 28,42%) related between 1995 and 2011 in broilers, poultry meat and poultry farms, meat product and human, and BRJF6X01.001 (21/95 = 22,10%) related between 2011 and 2017 in wild animals, broilers, poultry meat, poultry farms, meat product, animal feed, and pork meat. The pulsotype BRJF6X01.001 shows a high distribution in the environmental and productive chain. The degree of similarity between pulsotypes BRJF6X01.006 and BRJF6X01.001 is 88%. To ensure the safety of human and animal health, holistic approaches, including surveillance of *Salmonella* throughout the environment and in the production chain, together with control measures, are critical. As transmission of *Salmonella* from food producing animals to wildlife and to the environment is considered potential public health problem, information on the survival and persistence of *Salmonella* in the environment and in potential reservoirs is of considerable importance.

INTRODUCTION

Salmonellosis is the second most reported gastrointestinal infection in humans after Campylobacteriosis, and an important cause of foodborne outbreaks in the world (CDC, 2019b; European Food Safety *et al.*, 2021). *Salmonella* can be classified into typhoid and non-typhoid regarding their ability to develop specific pathologies in humans and animals. Typhoid serovars are a subcategory of serovars capable of infecting and colonizing only a very narrow range of hosts and highly adapted to humans, presenting only higher primates and humans as reservoirs; in contrast, the non-typhoid serovars are capable of triggering infections in both humans and animals (Knodler & Elfenbein, 2019). Symptoms caused by non-typhoid serovars are usually limited to diarrhea, and there is no need for the use of antibiotics, but severe illness can occur. Such cases may include extra-intestinal infections such as septicemia and myocarditis, and can result in death, especially in children, pregnant women, and the elderly, as infection can be complicated by resistance to third generation cephalosporins – one of the antimicrobial classes for treating severe or invasive Salmonellosis (Parisi *et al.*, 2018; Collineau *et al.*, 2020).

The most common non-typhoid *Salmonella* reservoir is the intestinal tract of a wide range of domestic and wild animals and a

variety of food matrices that can serve as a vehicle for transmission of *Salmonella* spp. to humans through fecal contamination (European Food Safety, 2021). *Salmonella* Heidelberg is a serotype widely distributed and frequently associated with human diseases, being more frequent in North America than in other parts of the world (Ferrari *et al.*, 2019; Collineau *et al.*, 2020; Melo *et al.*, 2021). Since 2013, *Salmonella* Heidelberg joined the group of 10 serovars with the highest incidence in Brazil, in which food sources were the largest contributor with 65%, followed by environmental samples, animals, raw materials, and human beings (Santos *et al.*, 2022).

Despite the lower costs of genome sequencing techniques, they are still expensive for laboratories in developing countries, (Voss-Rech *et al.*, 2019) and PFGE still can be considered the golden standard for genotyping of *Salmonella* due to the stability of the generated profiles, the discriminatory power and reproducibility of the results (Ferrari *et al.*, 2017).

Considering the relevant frequency of isolation Heidelberg in the last 25 years in Brazil, the aim of this study was to evaluate the genotypic distribution of *S.* Heidelberg according to the source of isolation and the relevance to public health.

MATERIAL AND METHODS

Isolates were received in The National Reference Laboratory of Cholera and Enteric Diseases of the Oswaldo Cruz Institute Foundation - FIOCRUZ, Rio de Janeiro, Brazil (NRL) inoculated in nutrient agar and on arrival, confirmed the identification as *S. enterica* by serotyping according to the Kauffman-White scheme (Issenhuth-Jeanjean *et al.*, 2014). NRL receives *Salmonella* strains from Brazilian meat industries, health services laboratories and universities for serotyping and genotyping.

Between 1995 and 2017, the NRL serotyped 11932 isolates as *S.* Heidelberg; we took a sample of 124 strains preserved on stock agar for genotyping and comparison of their pattern of similarity by PFGE. Strains isolated from the 1990s to 2017 obtained from different sources and regional origins were included. The strains were from poultry (42), broilers (16), environmental (16), humans (14), wild animals (13), poultry farms (9), vegetables (5), meat products (3), animal feed (3), pork (2) and cattle (1).

Genotyping protocol was applied according to the Center of Disease Control of the PulseNet Network (CDC, 2017). The isolates were genotyped by DNA macrorestriction analysis, using 40 U of the enzyme XbaI (New England Biolabs, Beverly, MA, USA) followed by PFGE, as previously described (Ribot *et al.*, 2006). *Salmonella* Braendrup H9812 was used as size standard. Restriction fragments were electrophoresed in certificated 1.2% PFGE agarose gels (Bio-Rad, Hercules, CA, USA) in tris-borate buffer (TBE; tris-borate 0.045 M, EDTA 0.001M) at 14ºC, using the CHEF DR III system (Bio-Rad), with an initial switch time of 2.2s and a final switch of 63.8s at 6V/s for 18 h. Gels were stained in ethidium bromide (1µlg/mL) and visualized under UV light. Images were captured using a digital camera, and macrorestriction patterns were compared using BioNumerics 7.6 software (AppliedMaths, Sint-Martens-Latem, Belgium). Similarity was calculated by the Dice coefficient with 1.7% tolerance. A dendrogram was generated by cluster analysis using the unweighted pair of group method with arithmetic mean (UPGMA). Strains sharing the same number and position of DNA macrorestriction fragments were considered to belong to the same genotype.

The interpretation of PFGE profile was evaluated according to Tenover *et al.* (1995) and Barrett *et al.* (2006), considering the difference from one another. The pulsotypes should be considered closely related when the difference is up to three bands, and profiles that differ by up to six bands should be considered possibly related. The rationale behind this recommendation is that a single genetic event (a point mutation in a local restriction, a deletion or insertion) would result in up to three bands of difference. Thus, a three-band difference would be the result of one genetic event and a six-band difference the result of two genetic events.

RESULTS AND DISCUSSION

In the 124 strains obtained from poultry (42), broilers (16), environmental (16), humans (14), wild animals (13), poultry farms (9), vegetables (5), meat products (3), animal feed (3), pork (2) and cattle (1), 18 clusters (Table 1 and Figure 1) of *S.* Heidelberg were found with the cut-off value of 85%, and the Cluster A was the most prevalent, having been detected in strains isolated from all the sources between 1995 and 2017 (Table 1).

Cluster A was composed of 27 pulsotypes (Table 1), and the pulsotype BRJF6X01.006 (26/95= 27.34%) was the most prevalent one, detected between 1995 and 2011 in broilers, poultry meat and poultry farms, meat products and humans. The pulsotype BRJF6X01.001 (21/95 = 22.10%) was the second most prevalent one, detected between 2011 and

2017 in wild animals, broilers, poultry meat, poultry farms, meat product, animal feed, and pork meat. The pulsotype BRJF6X01.001 shows a high distribution in the environmental and food chain. The pulsotypes BRJF6X01.010 (7/95 = 7.3%) and BRJF6X01.021(6/95= 6.3%) showed a high degree of similarity with approximately 91%. The pulsotype BRJF6X01.010 isolated in poultry meat (4) and farms (3) during 2014 to 2016 showed a high degree of similarity with approximately 96% of BRJF6X01.006, and BRJF6X01.027 (9/95 = 9.5%) pulsotype isolated from poultry meat, poultry farms and wild ducks between 2014 and 2016 showed a high degree of similarity with approximately 94% with BRJF6X01.006 and 92% with BRJF6X01.001 pulsotype. The human isolates were related between 2009 and 2011 and pulsotype

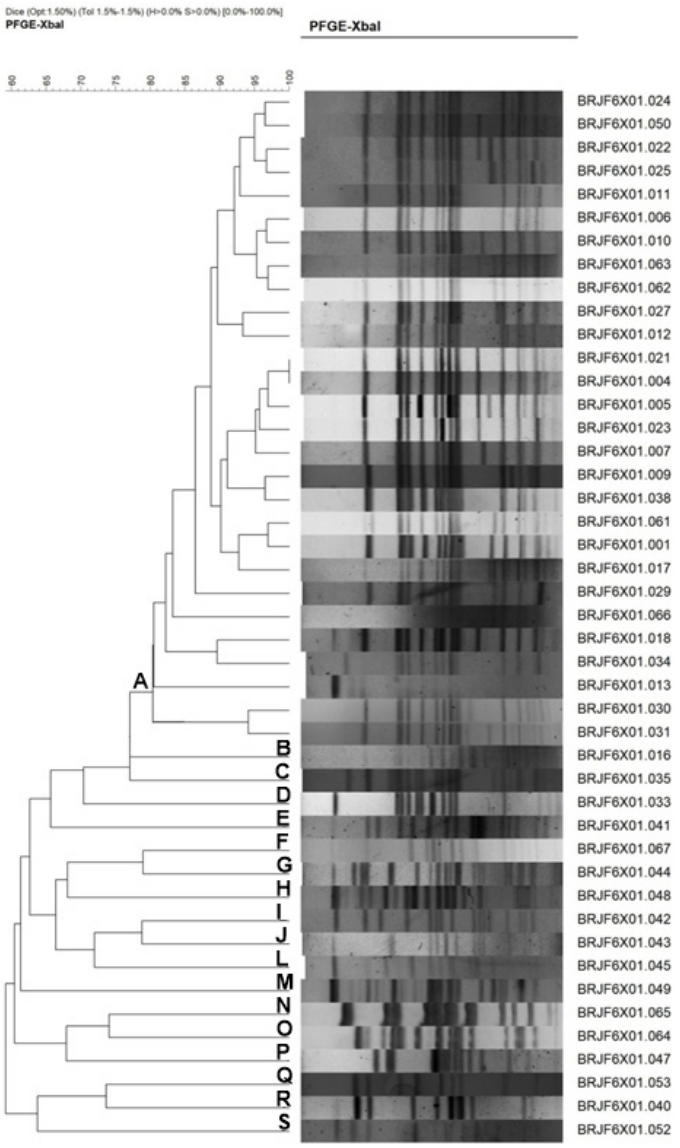

Figure 1 – Dendrogram of pulsotypes detected in *Salmonella* Heidelberg from wild animals, broilers, poultry meat, poultry farms, beef, pork meat, humans, meat products and animal feed between 1995 to 2017, Brazil.

BRFJ6X01.006 was identified in 8/9 (88.88%) of strains isolated. A single human pulsotype BRJF6X01.062 was isolated in 2010 and 100% of non-human sources of this pulsotype was found in isolates from poultry environment and poultry products, isolated between 1995 to 2011 (Table 1).

PFGE is a method based on genomic DNA restriction well established for *Salmonella* genotyping (Cosby *et al.*, 2015), and can be used to trace routes of strains dispersion and elucidate outbreaks. Voss-Rech *et al.* (2019) detected undifferentiated *Salmonella* Heidelberg genotypes by PFGE in poultry environment of two companies supporting the clonal dispersing of this serovar. *S.* Heidelberg emerged in commercial poultry farms and derived products from Brazil (Voss-Rech *et al.*, 2019; Rodrigues *et al.*, 2020), making this serovar a concern to public health. The persistence in poultry chain is probably due to the capacity of *S.* Heidelberg to resist to different litter treatments (Voss-Rech *et al.*, 2017) and to form biofilms under different temperatures and the tolerance to biocidal agents (Melo *et al.*, 2021). A study using multi-locus sequence typing supported the clonality of this serovar in poultry chain (Campos *et al.*, 2018), and in another study, using whole genome sequence, the strains were clustered together with isolates from poultry and swine in Brazil and poultry products from other countries (Kipper *et al.,* 2021). The high level of genomic similarity in *S.* Heidelberg indicated a probable clonal origin. The detection of this cluster could be explained by the dissemination among livestock industry worldwide as reported in other bacteria (Peirano & Pitout, 2010); or because Brazil is one of the largest chicken meat exporters in the world, our products are available all over the globe for researchers (Campos *et al.*, 2018; Van Den Berg *et al.*, 2019).

The National Program of Poultry Health (PNSA) and The National Program of Pathogens Control performs regular monitoring in the poultry production chain to check the prevention and control *Salmonella* programs of industries, which could explain the higher frequency of isolates from poultry and poultry products. Since the beginning of these programs in 1994 and 2003, there has been a decrease in the frequency of *Salmonella* Enteritidis and Typhimurium in poultry products and by-products, especially after the introduction of vaccine in the primary production. These serovars have been replaced by other serovars such as Heidelberg and Minnesota (Voss-Rech *et al.*, 2017; Voss-Rech *et al.*, 2019; Rodrigues *et al.*, 2020), however *S.* Enteritidis and *S.* Typhimurium remain the most common serovars

Table 1 – Cluster, pulsotype, source and year of isolation of 124 *Salmonella* Heidelberg strains.

Cluster	Pulsotype	Year	Region	State	Source	N*
A	BRJF6X01.001	2011	South	SC	Meat products	1
		2012	South	PR	Pork	1
					Poultry farms	1
		2014	South	PR	Poultry farms	2
				SP	Broiler	3
					Environmental	1
			Southeast	RJ	Broiler	2
		2015	South	RS	Poultry farms	1
				SC	Environmental	1
					Animal feed	1
			Southeast	RJ	Wild animal	5
		2016	South	SC	Environmental	2
		2017	South	SP	Wild animal	2
	BRJF6X01.004	2015	South	SC	Environmental	1
			Southeast	RJ	Animal	1
	BRJF6X01.005	2014	South	PR	Poultry meat	1
		2016	South	SC	Environmental	1
					Poultry meat	2
	BRJF6X01.006	1995	South	SC	Poultry farms	1
		1999	South	RS	Poultry farms	1
		2005	South	SP	Poultry farms	6
		2009	South	RS	Human	5
					Poultry farms	3
		2010	Midwest	DF	Human	2
			South	PR	Meat products	1
				RS	Poultry farms	3
				SC	Poultry farms	1
				SP	Broiler	1
		2011	South	SC	Environmental	1
			Southeast	MG	Human	1
	BRJF6X01.007	2015	Southeast	RJ	Wild animal	1
	BRJF6X01.009	2017	South	SP	Wild animal	1
	BRJF6X01.010	2014	South	RS	Broiler	1
					Environmental	1
			Southeast	RJ	Broiler	1
		2016	South	SC	Poultry meat	4
	BRJF6X01.011	1997	South	SP	Poultry meat	1
		2002	South	RS	Broiler	1
		2012	South	SC	Poultry meat	1
		2014	South	SC	Poultry meat	1
	BRJF6X01.012	2014	South	SP	Environmental	1
	BRJF6X01.013	1997	South	SP	Poultry meat	1
	BRJF6X01.017	2016	South	SC	Environmental	1
	BRJF6X01.018	2016	South	SP	Environmental	1
	BRJF6X01.021	2014	South	PR	Poultry meat	1
				RS	Pork	1
				SC	Poultry meat	1
		2016	South	SC	Environmental	1
					Poultry meat	2
	BRJF6X01.022	2014	South	SC	Poultry meat	1
	BRJF6X01.023	2014	South	PR	Poultry meat	1
	BRJF6X01.024	2017	South	RS	Poultry farms	1
		2017	South	RS	Poultry meat	1
	BRJF6X01.025	2014	South	RS	Environmental	1
	BRJF6X01.027	2014	Southeast	RJ	Broiler	5
		2015	Southeast	RJ	Wild animal	3
		2016	South	SC	Poultry meat	1
	BRJF6X01.029	2014	Southeast	RJ	Broiler	1
	BRJF6X01.030	2002	Southeast	SC	Poultry farms	1
	BRJF6X01.031	2005	South	RS	Poultry farms	1
	BRJF6X01.038	2010	South	PR	Poultry farms	1
		2015	Midwest	GO	Human	1
	BRJF6X01.050	2017	South	RS	Poultry farms	1
	BRJF6X01.061	2010	South	PR	Poultry farms	1
	BRJF6X01.062	2010	Midwest	DF	Human	1
		2011	South	SC	Poultry farms	1
		2016	South	SC	Meat products	1
	BRJF6X01.063	2016	South	SC	Bovine	2
	BRJF6X01.066	2015	South	SC	Animal feed	1
B	BRJF6X01.016	2016	South	SC	Environmental	1
C	BRJF6X01.035	2014	Southeast	RJ	Broiler	1
D	BRJF6X01.033	2015	South	SP	Vegetable	1
E	BRJF6X01.041	2015	South	SP	Vegetable	1
F	BRJF6X01.067	2008	South	RS	Human	1
G	BRJF6X01.044	2015	South	SC	Poultry meat	1
H	BRJF6X01.048	2016	South	SP	Environmental	1
I	BRJF6X01.042	2013	South	SC	Poultry meat	1
J	BRJF6X01.043	2015	South	SC	Poultry meat	1
L	BRJF6X01.045	1995	South	SC	Environmental	1
M	BRJF6X01.049	2016	South	SP	Animal feed	1
N	BRJF6X01.065	2015	Midwest	DF	Human	1
O	BRJF6X01.064	2015	Midwest	DF	Human	1
P	BRJF6X01.047	2015	South	SP	Vegetable	1
Q	BRJF6X01.053	2017	South	RS	Human	1
R	BRJF6X01.040	2015	South	SP	Vegetable	1
S	BRJF6X01.052	2017	North	PA	Poultry meat	1

*n- number of strains.

related to human cases (Santos *et al.*, 2022). This fact reinforces the importance of these programs to the control of *Salmonella* serovars all over the food chain, considering that the strategy of control should be based on good practices, hazards and risks analysis to support a farm to fork strategy.

The degree of similarity between pulsotypes BRJF6X01.006 and BRJF6X01.001 was 88% and the timeline shows that the pulsotype BRJF6X01.006 was mostly present in poultry chain sources between 1995 to 2011 and was detected in human cases between 2009 to 2011; the pulsotype BRJF6X01.001 appears since 2011, showing a high distribution in the environmental and food chain, but was not more isolated from human cases. Due to the high degree of similarity between the BRJF6X01.006 and BRJF6X01.001 pulsotypes and the fact that BRJF6X01.006 was no longer detected after the appearance of BRJF6X01.001 in 2011, we believe that the BRJF6X01.006 pulsotype would be the precursor of BRJF6X01.001.

Humans can be asymptomatic or can develop enteric disease, which can include bloody diarrhea and fever, and in some cases trigger severe systemic disease (Knodler & Elfenbein, 2019). *S.* Heidelberg has been associated with invasive human infections and mortality rates (Folster *et al.*, 2012; Gieraltowski *et al.*, 2016; Palmeira *et al.*, 2016). In most cases, salmonellosis does not require antibiotic therapy, but in patients with comorbidities or invasive infection the antibiotic treatment can be performed (CDC, 2019b). In 2019, 87,923 cases of human salmonellosis were reported in European Union with a rate of 20.0 cases per 100,000 population (European Food Safety, 2021). In the United States, CDC estimates that *Salmonella* causes around 1.35 million infections, 26500 hospitalizations, and 420 deaths in the United States every year and food is the main source of illness (CDC, 2019a). The annual cost associated with the major foodborne pathogens in the US has been estimated to be approximately $14 billion, and non-typhoid *Salmonella* alone accounts for $3.3 billion of the total (Hoffmann *et al.*, 2012). Regarding Brazilian status about salmonellosis, as in other developing countries, outbreak information is frequently incomplete because health authorities lack the capabilities or resources for detection, or because diarrheal diseases are widespread and outbreaks may be less common or clear than in developed countries (Panzenhagen *et al.*, 2015).

The phylogenetic comparison between the strains from food, poultry, humans, and wild animals suggest a probable common source since they presented, approximately, 90% of genetic similarity. The most common non-typhoid *Salmonella* reservoir is the intestinal tract of a wide range of domestic and wild animals and a variety of food matrices which can serve as a vehicle for transmission of *Salmonella* spp. to humans through fecal contamination. Failure in biosecurity measures in animal production can allow free-living animals to invade the livestock environment, allowing for the exchange of strains between farm and wild animals. The transfer frequently occurs when theses microorganisms are introduced into food preparation areas, with subsequent proliferation in food items through improper storage temperature, inadequate cooking, and/or cross contamination, as well as through direct contact with infected animals and humans (European Food Safety, 2021).

The analyzed *S*. Heildelberg isolates confirm to us an example of connection between humans, poultry farms, beef, poultry, pork and wild animals. Strains of *Salmonella* were also frequently found in farm animals, which could be due to contamination from different sources such as wildlife or the environment.

There are several limitations in this study that must be considered in the prevalence values. Furthermore, when flour made from dairy, bovine and pork products are mixed with poultry, they appeared more likely to have *Salmonella* strains of public health importance, such as *S*. Heidelberg. This finding suggests that foraging animals not normally associated with these important human pathogens can be exposed, colonized, and become exterminators in mixed avian operations. Although our data are insufficient to assess whether certain host species are important for human infection, the distribution of different pulsotypes may arise from a combination of factors, including variation in host range and sampling structure for different species, which suggest that companion animals and wild animals deserve more attention.

CONCLUSION

The results from this study lend support to the hypothesis that *S.* Heidelberg circulates between several sources and some cases could be infections carried by wild animals. As transmission of *Salmonella* from food producing animals to wildlife and to the environment is considered a potential public health problem, information on the survival and persistence of *Salmonella* in the environment and in potential reservoirs is of considerable importance. Genetic surveillance of *Salmonella* from different sources is necessary to understand the complex epidemiology

of this bacterium, due to its relevance for human and animal health.

ACKNOWLEDGMENTS

The authors would like to thank Fundação de Amparo à Pesquisa do Estado do Rio de Janeiro – FAPERJ through Process: E-26/010.001349/2019 Ref.210.607/2029 and Coordenação de Aperfeiçoamento de Pessoal de Nível Superior – CAPES and for their financial support.

REFERENCES

Barrett TJ, Gerner-Smidt P, Swaminathan B. Interpretation of pulsed-field gel electrophoresis patterns in foodborne disease investigations and surveillance. Foodborne Pathogens and Disease 2006;3(1):20-31.

Campos J, Mourão J, Silveira L, Saraiva M, Correia CB, Maçãs AP, et al. Imported poultry meat as a source of extended-spectrum cephalosporin-resistant CMY-2-producing *Salmonella* Heidelberg and *Salmonella* Minnesota in the European Union, 2014-2015. International Journal of Antimicrobial Agents 2018;51(1):151-4.

CDC - Center of Disease Control. Standard operating procedure for PulseNet PFGE of Escherichia coli O157:H7, Escherichia coli non-O157 (STEC), *Salmonella* serotypes, Shigella sonnei and Shigella flexneri. Protocols PNL05; 2017. p.13. Available from: https://www.cdc.gov/pulsenet/pdf/ecoli-shigella-*Salmonella*-pfge-protocol-508c.pdf.

CDC - Center of Disease Control. Antibiotic resistance threats in the United States; Atlanta; 2019. Available from: http://dx.doi.org/10.15620/cdc:82532 2019a.

CDC - Center of Disease Control. Surveillance for foodborne disease outbreaks united states [annual report 2017]. 2019b.

Collineau L, Chapman B, Bao X, Sivapathasundaram B, Carson CA, Fazil A, et al. A farm-to-fork quantitative risk assessment model for *Salmonella* Heidelberg resistant to third-generation cephalosporins in broiler chickens in Canada. International Journal of Food Microbiology 2020;330:108559.

Cosby DE, Cox NA, Harrison MA, Wilson JL, Buhr RJ, Fedorka-Cray PJ. *Salmonella* and antimicrobial resistance in broilers: a review. Journal of Applied Poultry Research 2015;24(3):408-26.

EFSA - European Food Safety A. The European Union One Health 2019 Zoonoses Report. EFSA Journal 2021;19(2):e06406.

Ferrari RG, Panzenhagen PHN, Conte-Junior CA. Phenotypic and genotypic eligible methods for *Salmonella* Typhimurium source tracking. Frontiers in Microbiology 2017;8:2587.

Ferrari RG, Rosario DKA, Cunha-Neto A, Mano SB, Figueiredo EES, Conte-Junior CA. Worldwide epidemiology of *Salmonella* serovars in animal-based foods: a meta-analysis. Applied and Environmental Microbiology 2019;85(14): e00591-19.

Folster JP, Pecic G, Rickert R, Taylor J, Zhao S, Fedorka-Cray PJ, et al. Characterization of multidrug-resistant *Salmonella enterica* serovar Heidelberg from a ground turkey-associated outbreak in the United States in 2011. Antimicrobial Agents and Chemotherapy 2012;56(6):3465-6.

Gieraltowski L, Higa J, Peralta V, Green A, Schwensohn C, Rosen H, et al. National outbreak of multidrug resistant *Salmonella* Heidelberg infections linked to a single poultry company. PLoS One 2016;11(9):e0162369.

Hoffmann S, Batz MB, Morris Jr JG. Annual cost of illness and quality-adjusted life year losses in the United States due to 14 foodborne pathogens. Journal of Food Protection 2012;75(7):1292-302.

Issenhuth-Jeanjean S, Roggentin P, Mikoleit M, Guibourdenche M, Pinna E de, Nair S, et al. Supplement 2008-2010 (no. 48) to the White-Kauffmann-Le Minor scheme. Research in Microbiology 2014;165(7):526-30.

Kipper D, Orsi RH, Carroll LM, Mascitti AK, Streck AF, Fonseca ASK, et al. Recent evolution and genomic profile of *Salmonella enterica* serovar heidelberg isolates from poultry flocks in Brazil. Applied and Environmental Microbiology 2021;87(21):e0103621.

Knodler L, Elfenbein J. *Salmonella enterica*. Trends in Microbiology 2019;27(11):964-5

Melo RT, Galvão NN, Guidotti-Takeuchi M, Peres P, Fonseca BB, Profeta R, et al. Molecular characterization and survive abilities of *Salmonella* Heidelberg strains of poultry origin in Brazil. Frontiers in Microbiology 2021;12:674147.

Palmeira A, Santos LR, Borsoi A, Rodrigues LB, Calasans M, Nascimento VP. Serovars and antimicrobial resistance of *Salmonella* spp. Isolated from turkey and broiler carcasses in southern Brazil between 2004 and 2006. Revista do Instituto Medicina Tropical de São Paulo 2016;58:19.

Panzenhagen P, Aguiar W, Frasao B, Pereira V, Abreu D, Rodrigues D, et al. Prevalence and fluoroquinolones resistance of Campylobacter and *Salmonella* isolates from poultry carcasses in Rio de Janeiro, Brazil. Food Control 2015;61:243-7.

Parisi A, Crump JA, Glass K, Howden BP, Furuya-Kanamori L, Vilkins S, et al. Health outcomes from multidrug-resistant *Salmonella* infections in high-income countries: a systematic review and meta-analysis. Foodborne Pathogens and Disease 2018;15(7):428-36.

Peirano G, Pitout JD. Molecular epidemiology of Escherichia coli producing CTX-M beta-lactamases: the worldwide emergence of clone ST131 O25:H4. International Journal of Antimicrobial Agents 2010;35(4):316-21.

Ribot EM, Fair MA, Gautom R, Cameron DN, Hunter SB, Swaminathan B, et al. Standardization of pulsed-field gel electrophoresis protocols for the subtyping of Escherichia coli O157:H7, *Salmonella*, and Shigella for PulseNet. Foodborne Pathogens and Disease 2006;3(1):59-67.

Rodrigues I, Silva R, Menezes J, Machado S, Rodrigues DP, Pomba C, et al. High Prevalence of multidrug-resistant nontyphoidal *Salmonella* recovered from broiler chickens and chicken carcasses in Brazil. Brazilian Journal of Poultry Science 2020;22(1).

Santos AFM, Dias TS, Machado SCA, Rodrigues DP, Pereira VLP. Top 10 *Salmonella* serovars associated with human salmonellosis in Brazil (2013-2020). Research, Society and Development 2022;11(8):e28011830533.

Tenover FC, Arbeit RD, Goering RV, Mickelsen PA, Murray BE, Persing DH, et al. Interpreting chromosomal DNA restriction patterns produced by pulsed-field gel electrophoresis: criteria for bacterial strain typing. Journal of Clinical Microbiology 1995;33 (9):2233-9.

Van den Berg RR, Dissel S, Rapallini M, Van der Weijden CC, Wit B, Heymans R. Characterization and whole genome sequencing of closely related multidrug-resistant *Salmonella enterica* serovar Heidelberg isolates from imported poultry meat in the Netherlands. PLoS One 2019;14(7):e0219795.

Voss-Rech D, Kramer B, Silva VS, Rebelatto R, Abreu PG, Coldebella A, et al. Longitudinal study reveals persistent environmental *Salmonella* Heidelberg in Brazilian broiler farms. Veterinary Microbiology 2019;233:118-23.

Voss-Rech D, Trevisol IM, Brentano L, Silva VS, Rebelatto R, Jaenisch FRF, et al. Impact of treatments for recycled broiler litter on the viability and infectivity of microorganisms. Veterinary Microbiology 2017;203:308-14.