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**Paper**



# Genotyping and antibiotic susceptibility of *Salmonella* strains collected from sheep and cattle samples in Algeria

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## Abstract

The present work investigates the genetic relatedness and antibiotic susceptibility of *Salmonella* strains collected from the red meat supply chain, highlighting the public health significance of these pathogens. Pulsed-field gel electrophoresis (PFGE-*Xba*I) was applied to genotype a collection of 84 *Salmonella* strains isolated from slaughterhouses. The antibiotic susceptibility of these strains to fourteen antimicrobial agents was determined using the minimum inhibitory concentration (MIC) method. The isolates were classified into 22 fingerprints, with two strains being non-typable. The predominant PFGE types identified were Mu1 (n=18), I2 (n=10), and K2 (n=8), indicating a high level of genetic similarity among isolates (>80%). All *Salmonella* strains exhibited resistance to at least two antimicrobials, with approximately 34.5% displaying resistance to three or more classes of antibiotics. Twelve distinct resistant patterns were identified, and notably, only one colistin-resistant *Salmonella* strain was detected. These findings underscore the need for ongoing surveillance and control measures in the red meat industry.

## Keywords

Meat, *Salmonella*, genetic profiling, antibiotic resistance, public health

## Introduction

Public awareness about food safety has significantly surged due to the rising number and severity of foodborne illnesses on a global scale. *Salmonella* is one of the main pathogens responsible for foodborne illnesses. Each year, a total of 93.8 million cases of gastroenteritis caused by *Salmonella* species worldwide, resulting in 155,000 deaths (Rincón-Gamboa *et al.*, 2021). *Salmonella*'s ability to cause disease is linked to various virulence genes located in its chromosome or large virulence-related plasmids. These genes encode products that facilitate interactions with host organisms during key stages such as colonisation, invasion, intracellular replication, and tissue damage (Nouichiet *et al.*, 2023). The primary sources of human *Salmonella* infections are often linked to contaminated animal-derived food products like meat, poultry, eggs, and dairy products (Fatima *et al.*, 2023). Meat can be contaminated at multiple stages of the supply chain, particularly in abattoirs during evisceration and intestinal content removal, mainly due to cross-contamination from equipment, utensils, and personnel (Cetin *et al.*, 2020; Rincón-Gamboa *et al.*, 2021). In Algeria, various studies have indicated that *Salmonella* is present in meat products, leading to greater exposure of the population to this pathogenic bacterium (Djeffal *et al.*, 2018; Nouichi *et al.*, 2018; Mezali *et al.*, 2019). This necessitates a thorough investigation into the genetic diversity of these strains.

Typing of foodborne pathogens like *Salmonella* through molecular genotyping is essential for identifying specific

strains responsible for outbreaks and detecting emerging health threats (Ed-Dra *et al.*, 2018). Understanding the genetic relatedness among *Salmonella* isolates can provide insights into transmission pathways and outbreak dynamics. Although Whole Genome Sequencing (WGS) has recently emerged as a more advanced and precise technique for genomic characterization, Pulsed-Field Gel Electrophoresis (PFGE) remains a valuable and widely used tool, particularly in settings where WGS is not feasible due to resource constraints. PFGE continues to serve as a standard method for genotyping, especially in outbreak investigations and source tracking by public health authorities (Turky *et al.*, 2014). In addition, the rising resistance of *Salmonella* to one or more antimicrobial agents, driven by the uncontrolled use of these substances in production systems for preventing and treating infectious diseases, presents a significant public health challenge (Rincón-Gamboa *et al.*, 2021). Many strains have shown resistance to commonly used antibiotics, including tetracyclines, sulphonamides, and ampicillin, which complicates treatment options for infected individuals (Mthembu *et al.*, 2019; Cetin *et al.*, 2020). The emergence of multidrug-resistant *Salmonella* strains, particularly those associated with food products, raises concerns about the effectiveness of existing therapeutic strategies (Fatima *et al.*, 2023) and underscores the need for continuous monitoring of resistance patterns.

The objective of this study was to evaluate the antimicrobial susceptibility and genetic diversity of 84 *Salmonella enterica* strains isolated from red meats in Algeria, focusing on determining the minimum inhibitory concentrations (MIC) of antibiotics and using pulsed-field gel electrophoresis (PFGE) with the restriction enzyme *Xba* I for genotyping.

## Materials and methods

### Bacterial isolates

The 84 analysed *Salmonella* strains were recovered during a previous study (Nouichi *et al.*, 2018) from 826 samples (350 from cattle and 476 from sheep) collected from red meat abattoirs in Algiers. These strains were originally isolated from cattle carcasses (n= 39), sheep carcasses (n= 32), cattle faeces (n= 11) and sheep faeces (n=2). All *Salmonella* strains were performed according to the International Organisation of Standardisation method (ISO 6579), serotyped by slide agglutination as specified by the White- Kauffmann-Le Minor scheme, and confirmed by PCR technique using the *Salmonella*-specific *invA* gene (284 bp).

### Antimicrobial Susceptibility Testing

All *Salmonella* isolates were further characterised for antimicrobial susceptibility using the Sensititre broth microdilution assay with EUVSEC plates (Thermo Scientific, West Palm Beach, USA). The minimum inhibitory concentration (MIC) was determined for each of the following 14 antimicrobials with tested concentration range (µg/ml) in brackets: ampicillin (1–64), azithromycin (2–64), cefotaxime (0.25–4), ceftazidime (0.5–8), chloramphenicol (8–128), ciprofloxacin (0.015–8), colistin (1–16), gentamicin (0.5–32), meropenem (0.03–16), nalidixic acid (4–128), sulfamethoxazole (8–1024), tetracycline (2–64), tigecycline (0.25–8) and trimethoprim (0.25–32). The tests were performed according to the manufacturer guidelines. *Escherichia coli* ATCC 25922 was used as the quality control strain. Clinical breakpoint values from the European Committee on Antimicrobial Susceptibility Testing (EUCAST, 2023) were used for antimicrobial susceptibility testing. For each antibiotic, the MIC<sub>50</sub> and MIC<sub>90</sub> were calculated as the minimum concentrations that inhibited the growth of the isolates by 50% and 90% respectively.

### Pulsed Field Gel Electrophoresis (PFGE) Analyses

Clonal relatedness among the strains was carried out using PFGE according to the standardized CDC PulseNet protocol. Briefly, bacterial suspensions of *Salmonella* isolates were prepared from fresh overnight cultures and fixed in agarose plugs. Then, agarose-embedded genomic DNA was subjected to digestion with application of *Xba*I restriction enzyme (Promega, Madison, WI, USA). The generated fragments were separated in 1% agarose using Chef Mapper (CHEF DR III PFGE, Bio-Rad, Hercules, CA, USA). *Salmonella enterica* serovar Braenderup H9812 DNA was used as a molecular size marker. A dendrogram was developed according to the Dice similarity coefficient by employing the unweighted pair group method with arithmetic means (UPGMA) using GelCompar II software v. 6.6 (Applied Maths, Sint-Martens-Latem, Belgium) with a position tolerance of 1.4%.

## Results

Antimicrobial resistance rates within each *Salmonella* serovar against the 14 antimicrobial agents tested are shown in Table I and the MIC distributions for the different antibiotics are shown in Table II. All the *Salmonella* isolates tested were found to be susceptible to meropenem, azithromycin, cefotaxime, tigecycline and ceftazidime. The highest resistance was observed in sulphonamides (100%) with minimum inhibitory concentrations (MIC) reaching 1024 µg/mL for all isolates, indicating a total ineffectiveness of this antibiotic. Similarly, widespread resistance was noted for diaminopyrimidines, with MIC values of 32 µg/mL.

Regarding quinolones, including nalidixic acid and ciprofloxacin, there is a wide variation in minimum inhibitory concentrations (MIC) across strains, with some displaying resistance levels of up to 8 µg/mL. Overall, the resistance rates for these quinolones range from 28.6% to 30.9%, it is particularly troubling that *S. Kentucky* shows 100% resistance to both antibiotics. Resistance rates were 21% for gentamicin, and 20% for tetracycline with MIC values reaching 32 µg/mL and 64 µg/mL, respectively. In contrast, our isolates exhibited low levels of resistance to colistin (1.2%) and chloramphenicol (3.6%).

All of the *Salmonella* isolates exhibited resistance to two or more antimicrobial agents used. Twenty-nine isolates (34.5%) belonging to 8 serovars were multidrug-resistant, being resistant to at least three different classes of antimicrobials. Eighty strains (21.4%) were resistant to 7 antibiotics. All multidrug resistant (MDR) strains were originated from carcasses without regard to the animal species and the slaughterhouses. Furthermore, twelve resistant phenotypes, i.e. RI–RXII, were defined (Table III). The two most dominant resistant phenotypes were RI and RXI, represented by 55 and 17 strains, respectively. The remaining phenotypes were represented by one or two strains.

Sulfamethoxazole showed the highest MIC<sub>50</sub> and MIC<sub>90</sub> values (>1024 µg/ml). Trimethoprim yielded MIC<sub>50</sub> and MIC<sub>90</sub> values >32 µg/ml. High values of MIC<sub>90</sub> were found with tetracycline, Nalidixic acid, ampicillin and gentamicin. The MIC<sub>50</sub> and MIC<sub>90</sub> values were less than the resistance breakpoint of azithromycin, cefotaxime, chloramphenicol and tigecycline.

PFGE analysis produced multiple patterns consisting of several fragments ranging in size from 40 to 1100 kb. PFGE of *Xba* I-digested genomic DNA from 82 *Salmonella* isolates showed 22 different macro-restriction profiles or clusters (Figure 1), while the remaining 2 isolates were unclustered.

The distribution of different genotypes according to the sampling source in each of the two slaughterhouses is detailed in Table IV.

Family	ATB	Number (%) of resistant isolates										TOTAL
		S.Muenster	S.Anatum	S.Infantis	S.Kentucky	S.Havana	S.Richmond	S.Typhimurium	S.Monteideo	S.Virginia	S.Braenderup	
Sulfamids	<b>SMX</b>	33(100)	11(100)	12(100)	13(100)	3(100)	4(100)	3(100)	3(100)	1(100)	1(100)	84(100)
Diaminopyrimidines	<b>TMP</b>	33(100)	11(100)	12(100)	13(100)	3(100)	4(100)	3(100)	3(100)	1(100)	1(100)	84(100)
Quinolones	<b>CIP</b>	4(12.1)	2(18.2)	3(25)	13(100)	1(33.3)	0	2(66.7)	0	1(100)	0	26(30.9)
	<b>NAL</b>	3(9.1)	2(18.2)	3(25)	13(100)	1(33.3)	0	2(66.7)	0	0	0	24(28.6)
Macrolides	<b>AZI</b>	0	0	0	0	0	0	0	0	0	0	0
Bêta-lactamines	<b>MERO</b>	0	0	0	0	0	0	0	0	0	0	0
	<b>FOT</b>	0	0	0	0	0	0	0	0	0	0	0
	<b>TAZ</b>	0	0	0	0	0	0	0	0	0	0	0
	<b>AMP</b>	4(12.1)	3(27.3)	2(16.7)	13(100)	1(33.3)	0	2(66.7)	0	0	0	25(29.8)
Phenicol	<b>CHL</b>	0	0	1(8.3)	0	0	0	2(66.7)	0	0	0	3(3.6)
Tetracyclines	<b>TGC</b>	0	0	0	0	0	0	0	0	0	0	0
	<b>TET</b>	2(6.1)	1(9.1)	2(16.7)	11(84.6)	1(33.3)	0	3(100)	0	0	0	20(23.8)
Polymyxines	<b>COL</b>	0	0	0	0	0	1(25)	0	0	0	0	1(1.2)
Aminoglycosides	<b>GEN</b>	4(12.1)	2(18.2)	2(16.7)	12(92.3)	1(33.3)	0	0	0	0	0	21(25)

**Table I.** Antibiotic resistance rates of *Salmonella* strains by serovar: SMX: sulfamethoxazole; TMP: trimethoprim; CIP: ciprofloxacin; TET: tetracycline; MERO: meropenem; AZI: azithromycin; NAL: nalidixic acid; FOT: cefotaxime; CHL: chloramphenicol; TGC: tigecycline; TAZ: ceftazidime; COL: colistin; AMP: ampicillin; GEN: gentamicin.

Antimicrobial	Number of <i>Salmonella</i> isolates with minimal inhibitory concentrations (µg/mL)																MIC <sub>50</sub>	MIC <sub>90</sub>
	0.02	0.03	0.06	0.12	0.25	0.5	1	2	4	8	16	32	64	128	256	512	1024	
SMX																84	1024	1024
TMP												84					32	32
CIP	50	8		1	3				7	15							0.02	8
TET								59	5	3	4	13					2	64
MERO		83	1														0.03	0.03
AZI								83	1								2	2
NAL								59	1	1	23						4	128
FOT				84													0.25	0.25
CHL									80	1	3						8	8
TGC					82	2											0.25	0.25
TAZ					84												0.5	0.5
COL							53	30	1								1	2
AMP							58	1			25						1	64
GEN						55	8			6	5	10					0.25	32

**Table II.** Distribution of the minimal inhibitory concentration (MIC) values for the 84 tested *Salmonella* isolates: SMX: sulfamethoxazole; TMP: trimethoprim; CIP: ciprofloxacin; TET: tetracycline; MERO: meropenem; AZI: azithromycin; NAL: nalidixic acid; FOT: cefotaxime; CHL: chloramphenicol; TGC: tigecycline; TAZ: ceftazidime; COL: colistin; AMP: ampicillin; GEN: gentamicin.

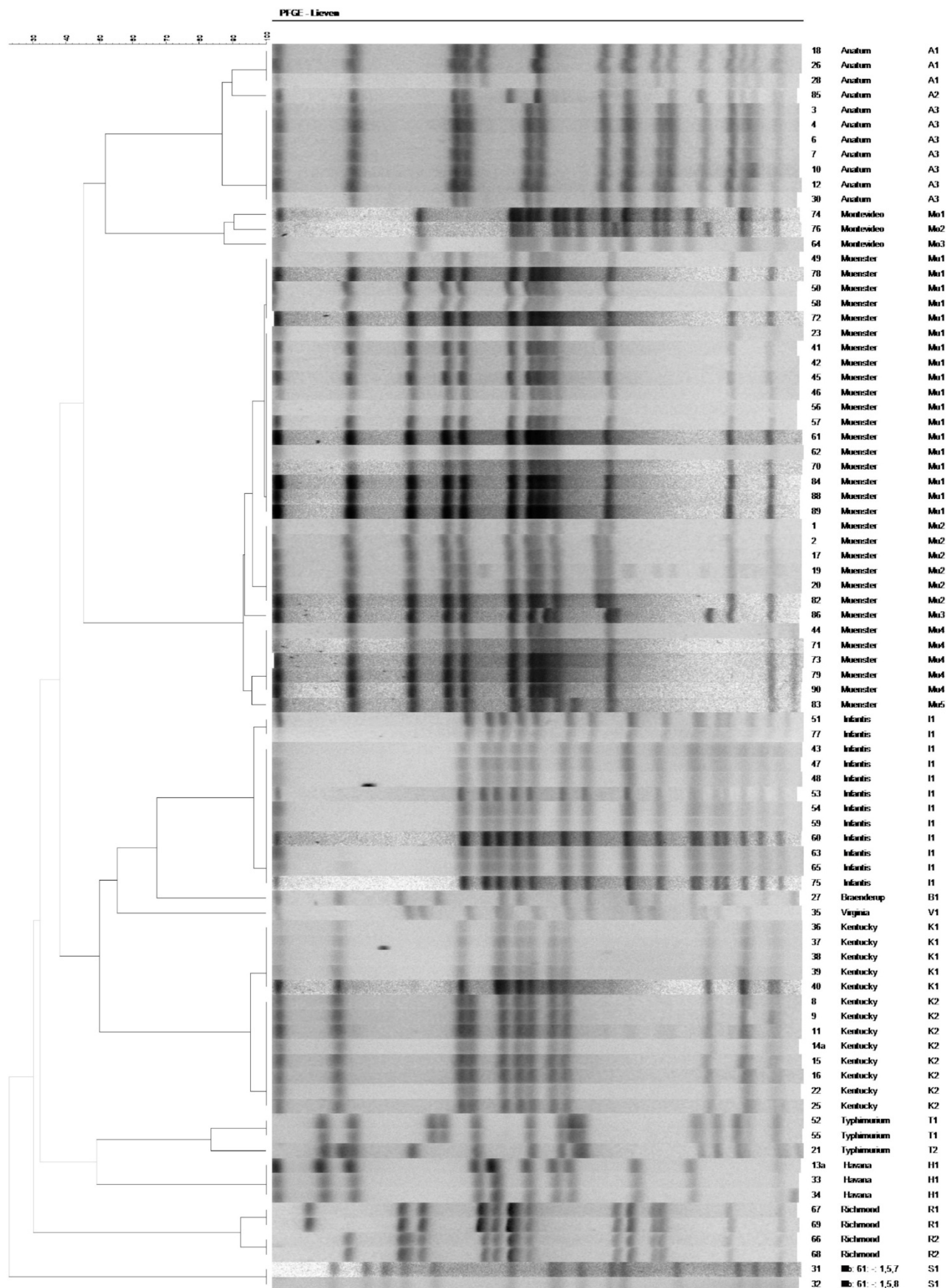
Resistance phenotype	Resistance pattern	Number of strains	Serovar	Origin of isolate
RI	SMX, TMP	55	29 Muenster	13 Beef C, 5 Beef F, 10 Sheep C, 1 Sheep F
			9 Infantis	1 Beef C, 2 Beef F, 6 Sheep C
			8 Anatum	2 Beef C, 1 Beef F, 4 Sheep C, 1 Sheep F
				1 Beef C, 2 Beef F
				Beef Carcass
				Beef C
			3 Montevideo	Beef F
			3 Richmond	
			2 Havana	
			1 Braenderup	
RII	SMX,TMP,AMP	1	Anatum	Sheep C
RIII	SMX,TMP,CIP	1	Virginia	Sheep C
RIV	SMX,TMP,COL	1	Richmond	Beef C
RV	SMX,TMP,AMP,GEN	1	Muenster	Beef C
RVI	SMX,TMP,CIP,NAL,AMP	1	Kentucky	Beef C
RVII	SMX,TMP,TET,NAL,AMP	1	Typhimurium	Sheep
RVIII	SMX,TMP,NAL,AMP,GEN	1	Muenster	Beef carcass
RIX	SMX,TMP,CIP,NAL,AMP,GEN	2	Kentucky	Sheep carcass
			Infantis	Beef carcass
RX	SMX,TMP,CIP,TET,NAL,CHL	2	Typhimurium	Sheep carcass
			Infantis	Beef carcass
RXI	SMX,TMP,CIP,TET,NAL, AMP,GEN	17	2 Anatum	Sheep c
			1 Havana	Beef
			2 Muenster	(1 Beef, 1 sheep)
			1 Infantis	Beef
			11 Kentucky	8 Beef, 3 sheep
RXII	SMX,TMP,CIP,TET,NAL,CHL,AMP	1	Typhimurium	Sheep

**Table III.** Resistance pattern profiles of the studied *Salmonella* strains: C: carcasses; F: Faeces; SMX: sulfamethoxazole; TMP: trimethoprim; CIP: ciprofloxacin; TET: tetracycline; MERO: meropenem; AZI: azithromycin; NAL: nalidixic acid; FOT: cefotaxime; CHL: chloramphenicol; TGC: tigecycline; TAZ: ceftazidime; COL: colistin; AMP: ampicillin; GEN: gentamicin.



Serovar	Pattern	Number of strains								Total
		El Harrach slaughterhouse				Hussein Dey slaughterhouse				
		Beef		Sheep		Beef		Sheep		
		Carcasses	Faeces	Carcasses	faeces	Carcasses	faeces	Carcasses	faeces	
Muenster	Mu-1	9	2	6				1		18
	Mu-2	1				2		3		6
	Mu-3	1								1
	Mu-4	1	3	1						5
	Mu-5	1								1
Kentucky	K-1			2						5
	K-2	3				6		2		8
Infantis	I-1		1	1						2
	I-2	4	1	5						10
Anatum	A-1							2		3
	A-2	1					1			1
	A-3					1		5	1	7
Richmond	R-1	2								2
	R-2	2								2
Havana	H-1	2				1				3
Typhimurium	T-1			2						2
	T-2							1		1
Montevideo	Mo-1		1							1
	Mo-2		1							1
	Mo-3	1								1
Virginia	V-1			1						1
Braenderup	B-1						1			1

**Table IV.** Genotyping results of *Salmonella* strains via PFGE according to the sample source.



**Figure 1.** PFGE patterns and dendrogram analysis of XbaI digested genomic DNA from of the 84 studied *Salmonella* strains.

## Similarity coefficient

Dice

Optimization: 0 %

Tolerance: 1 %

Tolerance change: 0 %

Minimum height: 0 %

Minimum surface: 0 %

Uncertain bands: Ignore

Relaxed doublet matching: No

Fuzzy logic: No

Area sensitive: No

### Cluster analysis

Clustering method: UPGMA

Use advanced clustering: No

Branch quality: Cophenetic correlation

## Discussion

The increasing prevalence of *Salmonella* spp. strains that have developed resistance to multiple antibiotic families can be attributed to the excessive and improper utilisation of antibiotics in human and veterinary healthcare for therapeutic and prophylactic purposes. Additionally, the systematic inclusion of antibiotics in animal feed to promote growth and enhance yield further contributes to this phenomenon. This overuse and misuse of antibiotics have resulted in the transmission of drug-resistant *Salmonella* to humans through the food chain, posing a potential threat to human health (Fatima *et al.*, 2023).

None of the *Salmonella* strains were sensitive to all the antibiotics tested in this study. Resistance to sulfamethoxazole and trimethoprim was common (100%), while 20% of the isolates were found to be resistant to tetracyclines with especially high rates in *S. Kentucky*. These findings contrast with those of Elki *et al.* (2019), who reported complete sensitivity of *Salmonella* isolates from meat in Ghana to the same antibiotics.

These molecules are older first-line molecules widely used in livestock farming. They are the most accessible classes of antibiotics, making them attractive to developing countries with limited healthcare budgets. Resistance to these drugs is generally due to a plasmid gene that can be easily acquired by bacteria.

Regarding chloramphenicol, resistance rates of 3.6% were found. This result could be explained by the moderate use of these drugs due to their withdrawal from the Algerian nomenclature.

A quarter (25%) of the isolated strains exhibited resistance to gentamicin, a prominent antibiotic used in urinary tract infections treatment in humans.

Although colistin demonstrates a low resistance rate (1.2%), the crucial role of polymyxins as a last resort for treating multidrug-resistant infections, particularly in the context of limited viable therapeutic alternatives (Marchant *et al.*, 2024), makes this finding significant. Our previous study (Nouichi *et al.*, 2018) did not identify any resistance to this molecule. This prior research was based on the disk diffusion test, which is not appropriate for detecting colistin resistance (Bertelloni *et al.*, 2022). Furthermore, until 2015, colistin resistance mechanisms were known to be encoded in the chromosome. However, the subsequent identification of a colistin resistance gene (*mcr-1*) in a conjugative plasmid in *Escherichia coli* isolates of animal origin raised significant concern within the scientific community (Lima *et al.*, 2019). Supporting these concerns, Gutema *et al.* (2021) also reported colistin resistance in two strains of *Salmonella* isolated from cattle in slaughterhouses in Ethiopia. Notably, the resistance was observed in the Dublin serovar, while in our study, it was detected in the Richmond serovar. This underscores the need for continued surveillance and stringent antimicrobial stewardship.

Conversely, resistance to  $\beta$ -lactams was limited to ampicillin (29.8%), previous studies from different countries have



reported higher resistance rates (Sallam *et al.*, 2014; Moawad *et al.*, 2017; Pławińska-Czarnak *et al.*, 2022; Tadesse *et al.*, 2024). On the other hand, the absence of resistance to third-generation cephalosporins is a positive aspect, as they are clinically essential in the treatment of invasive salmonellosis in humans. Moreover, tigecycline, a new class of glycylcyclines, showed good activity against *Salmonella*. It is known to exhibit broad-spectrum activity against most *Enterobacteriaceae* bacteria. Tigecycline's effectiveness against *Salmonella* is further supported by Gutema *et al.* (2021), who found no resistance to the drug in *Salmonella* strains isolated from cattle in Ethiopia.

However, all *S. Kentucky* and *S. Typhimurium* isolates were found to be resistant to ciprofloxacin. This could be attributed to the uncontrolled use of these expensive drugs in livestock farming in Algeria. *Salmonella* that are resistant to fluoroquinolones are included in the World Health Organization's high-priority list (Mthembu *et al.*, 2019). This poses a significant concern due to the rapid development of bacterial resistance to newly discovered antibiotics. Resistance to ciprofloxacin has been reported in *Salmonella* isolates from food across several African countries, including Tunisia (Oueslati *et al.*, 2021; Hassena *et al.*, 2022), Morocco (El Hanafi *et al.*, 2023; Sabri *et al.*, 2023), Egypt (Abd-Elghany *et al.*, 2022), Nigeria (Beshiru *et al.*, 2019), Ghana (Adzitey *et al.*, 2015) and Ethiopia (Wabeto *et al.*, 2017; Eguale, 2018; Mustefa *et al.*, 2018). A different study, however, reported no resistance to ciprofloxacin in Kentucky and Typhimurium serovars isolated from raw chicken and beef meat in northern Egypt (Moawad *et al.*, 2017).

In addition to ciprofloxacin, resistance of these strains to quinolones also involved the nalidixic acid. Complete resistance to quinolones is achieved only when two or more mutations in the *gyr A* gene, which encodes the targets of these drugs, are present simultaneously (Mthembu *et al.*, 2019). It appears that the isolates tested in this study have undergone this type of mutation, as they are resistant to nalidixic acid and other quinolone molecules such as ciprofloxacin.

In this study, 12 distinct multidrug resistance patterns were recorded. The two serovars commonly involved in collective foodborne illnesses, *S. Kentucky* and *S. Typhimurium*, had the highest number of multidrug-resistant phenotypes. This aligns with multiple recent studies that have documented the global occurrence of MDR *Salmonella* in raw meat worldwide (Moawad *et al.*, 2017; Cetin *et al.*, 2020; Gutema *et al.*, 2021; Rincón-Gamboa *et al.*, 2021; Fatima *et al.*, 2023). Multidrug resistance defined as resistance to at least three classes of antimicrobial agents to antibiotics, may result from the transfer of resistance genes and random chromosomal mutations (Mthembu *et al.*, 2019). Multidrug-resistant strains are associated with a high risk of invasive infections and mortality compared to sensitive strains (Mouttoutu *et al.*, 2017).

However, it is important to note that the *Salmonella* strains obtained from faeces exhibited resistance solely to trimethoprim and sulphonamides. This observation can be attributed to the possibility that the elevated quantity of *Salmonella* found in carcasses during this study is not predominantly linked to the presence of the bacteria in faeces.

In this study, the PFGE analysis of the 84 *Salmonella* strains revealed 22 distinct genotypes. Two non-typable strains belonging to serovar *S. Muenster* were observed, possibly due to DNA degradation during the addition of endonuclease buffer as reported by Turkey *et al.* (2014). The high clonality observed, above 80%, suggests limited genetic variation between the different patterns.

The study revealed the prevalence of genotype Mu1, representing 21.42% of the isolated *Salmonella* strains. These strains were mainly recovered from El-Harrach slaughterhouse and exhibited similar antibiotic resistance profiles. Genotype I1, representing 14.28% of the strains, was also isolated from El-Harrach slaughterhouse. These two clones persisted for a long period, suggesting adaptation to the environment of livestock in that slaughterhouse. Some clones were specific to certain slaughterhouses, such as clone K1 in El-Harrach slaughterhouse and K2 in Hussein-Dey slaughterhouse. This could be attributed to the presence of persistent clones in each slaughterhouse. Some genetic profiles were present in both slaughterhouses, which could be attributed to the movement of animals through commercial circuits.

Three strains of *S. Havana* were isolated from both slaughterhouses, suggesting possible clone persistence in the wilaya of Algiers or a common origin of the slaughtered animals. Some genetic profiles were ubiquitous on carcasses and animal faeces, indicating cross-contamination through equipment, operator hands, and the environment. Certain serovars, such as *S. Infantis*, showed high genetic similarity, suggesting clonal persistence in the environment of El-Harrach slaughterhouse. This is consistent with the study by Ed-Draet *et al.* (2018), which showed that all 21 strains of *S. Infantis* isolated from beef belonged to a single fingerprint. Other studies have also reported high similarity of *S. Infantis* in animal and human isolates (Hauser *et al.*, 2012; Rahmani *et al.*, 2013; Velhner *et al.*, 2014; Franco *et al.*, 2015). The *Salmonella* strains exhibited evident genetic diversity, with frequent changes in clones during different sampling visits. Each group of animals introduced new clonal types of *Salmonella*, indicating that each batch of animals could potentially contaminate other carcasses.

*Salmonella* Muenster was the most diverse serovar, with five distinct genotypes. The genetic variability among strains could be due to the presence of linear plasmids and transposons (Karatuğ *et al.*, 2018). The study did not show a seasonal effect for certain *Salmonella* genotypes. The reasons for the high genetic similarity of the strains require further research. Nationwide sample collection would provide a broader understanding of *Salmonella* serovars in ovine and bovine farms.

## Conclusion

In summary, these results demonstrate the clonality of some serotypes, confirming the spread and persistence of the same clone in the studied establishments. Conversely, the heterogeneity of the profiles of other serotypes suggests a possible diversity of sources of contamination in these two slaughterhouses.

On the other hand, the *Salmonella* strains displayed resistance to multiple antibiotics. This alarming situation is further compounded by the fact that all of our strains were obtained from food samples. It highlights the combination of irrational antibiotic usage, inadequate surveillance, and insufficient facilities for detecting multidrug-resistant strains, emphasising the urgency of the issue.

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