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Detection of *bla*$_{CTX-M}$, *bla*$_{TEM}$, and *bla*$_{SHV}$ genes in ESBL-producing enterobacterales from poultry farms in the peri-urban area of Ouagadougou, Burkina Faso

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This study investigated the carriage of *bla*$_{CTX-M}$, *bla*$_{TEM}$ and *bla*$_{SHV}$ gene in Extended-spectrum beta-lactamase (ESBL)-producing Enterobacterals from poultry in peri-urban area of Ouagadougou, Burkina Faso. From June to November 2023, fresh fecal samples were collected from 37 consenting poultry farms in four peri-urban areas around Ouagadougou. These samples were cultured to isolate ESBL-producing *Escherichia coli* and *Klebsiella* species. The isolates were then examined for the presence of *bla*$_{CTX-M}$, *bla*$_{TEM}$, and *bla*$_{SHV}$ genes and their antibiotic susceptibility patterns. Out of the 37 poultry samples, 27 ESBL-producing bacteria were recovered; 48.64% (18/37) were ESBL-producing *E. coli* (16 *bla*$_{CTX-M}$ genes, 8 *bla*$_{TEM}$ genes, 2 *bla*$_{SHV}$ genes) and 27.02% (10/37) were *Klebsiella* spp. (10 *bla*$_{CTX-M}$ genes, 8 *bla*$_{SHV}$ genes, 5 *bla*$_{TEM}$ genes). Co-carriage of *bla*$_{CTX-M/TEM}$, *bla*$_{CTX-M/SHV}$, and *bla*$_{TEM/SHV}$, high resistance rates to tetracycline and trimethoprim-sulfamethoxazole, and multi-drug resistance (MDR) phenotypes were observed among the ESBL-producing bacteria isolated in this study. The findings indicate the widespread presence of ESBL-producing bacteria in poultry farms in Burkina Faso, highlighting a potential public health risk.

Key words: Detection of extended-spectrum beta-lactamase (ESBL), enterobacterales, poultry frams, peri-urban, Burkina Faso.

INTRODUCTION

Bacteria acquired resistance to antibiotic by expressing specifics genes that allow them to express several mechanisms of resistance to antibiotics. These include enzymatic inactivation via beta-lactamases, target site

modification, active efflux system, and reducing membrane (Wanda, 2018). Antibiotic resistance is a major global public health challenge, particularly in developing countries. It is exacerbated by factors such as the unauthorized sale of antimicrobials, poor access to diagnostics, and non-humane use of antibiotics (Bhatia, 2019; Tesema and Birhanu, 2024).

Gram-negative bacteria, especially *Escherichia coli* and *Klebsiella* species produce Extended-spectrum beta-lactamases (ESBLs), which confer them resistance to a broad spectrum of beta-lactam antibiotics including penicillin and cephalosporin (Nathisuwan et al., 2001). They pose a significant public health threat within the global context of rising antimicrobial resistance (Wilson and Török, 2018), due to their increasing prevalence and association with higher morbidity, mortality, and healthcare costs (Dhillon and Clark, 2012).

ESBLs bacteria are commonly found in humans, animals, and the environment, with aggregated prevalence rates varying between 19.4 and 56.9% (Ramatla et al., 2023). The global spread of bacteria producing ESBL is associated with various factors, including environmental sources like wastewater, agriculture animals , and international mobility (Chong et al., 2018). In Burkina Faso, poultry production is predominantly characterized by traditional, village-based systems, where chickens are the most commonly raised species (Chalghoumi, 2020). Although 88% of rural households engage in poultry farming, it is largely viewed as a supplementary source of income rather than a primary commercial activity (Chalghoumi, 2020; Hoffmann et al., 2023).

Recent studies on poultry farming in Burkina Faso, particularly in the peri-urban areas of Ouagadougou, highlight a male-dominated sector (79.3%) with a significant proportion of farmers having limited formal education (Tapsoba et al., 2024; Soro et al., 2024). The sector faces various challenges related to health management, nutrition, and inadequate housing (Tapsoba et al., 2024). Concerns about antibiotic use are notable, with 85.65% of farmers lacking adequate knowledge of its proper application, and 31.98% using veterinary medications without prescriptions (Sawadogo et al., 2023). Self-medication is prevalent among farmers (73.6%), and a significant percentage (38%) sources their medications from unregulated suppliers (Soro et al., 2024). Despite these issues, poultry farming plays a vital role in food security and provides a substantial income for smallholder farmers (Hoffmann et al., 2023).

In poultry farms, ESBL-producing *E. coli* and other Enterobacterales are frequently detected, along with their surrounding environments. Significant prevalence rates have been found in poultry feces, soil, water, and air (Sali et al., 2017). In poultry, the most frequently detected ESBL genes included $bla_{CTX-M-1}$, bla_{TEM-52}, and bla_{SHV-12} (Sali et al., 2017). Agricultural practices often involve self-medication and the use of unauthorized drugs, significantly contributing to the spread of antibiotic resistance within food chains (Soro et al., 2024). The risk of transferring ESBL-producing *E. coli* and *Klebsiella* spp. through the food chain has been highlighted in 35.7% of poultry meat vendors (Mwanginde et al., 2021).

In Burkina Faso, studies have identified concerningly high prevalences rates of ESBL-producing Enterobacterals with 41.03% in cattle, 69.60% in pigs, and ranging from 0.8 to 19.1% in poultry , even among animals that have never been treated with antibiotics (Sanou et al., 2019). A substantial proportion of farmers around 85.65% have insufficient knowledge regarding the appropriate use of antibiotics, and 31.98% provided veterinary treatment without professionals prescriptions (Sawadogo et al., 2023).

However, the precise identification of ESBL genes and the detailed characterization of resistance profiles are still areas requiring further exploration in Burkina Faso. To better control and support policies and programs aimed at combating the spread of resistant bacteria through the food chain, it is crucial to assess the prevalence of bacteria that are of public health concern. Additionally, a thorough understanding of resistance mechanisms and the genes that govern them is necessary, especially in livestock farming due to its critical role in the production chain of meat and meat products.

This study aims to address this gap by examining the resistance associated with other classes of antibiotics, the production of AmpC and Carbapenemase, and the distribution of bla_{CTX-M}, bla_{TEM}, and bla_{SHV} genes among ESBL-producing *E. coli* and *Klebsiella* spp. strains collection isolated from poultry farm in the peri-urban areas of Ouagadougou, Burkina Faso.

MATERIALS AND METHODS

Study design

From June to November 2023, a cross-sectional study was conducted on 37 poultry farms located in four peri-urban areas of Ouagadougou: Saaba, Loumbila, Koubri, and Saponé, surrounding Ouagadougou, the capital city of Burkina Faso (Figure 1). Poultry farms were selected using a convenience sampling approach, ensuring a representative distribution from both small-scale and larger commercial farms within the region.

Fecal samples were randomly collected from poultry at the farms and pooled. These samples were processed immediately to isolate Enterobacterales species using standard microbiological

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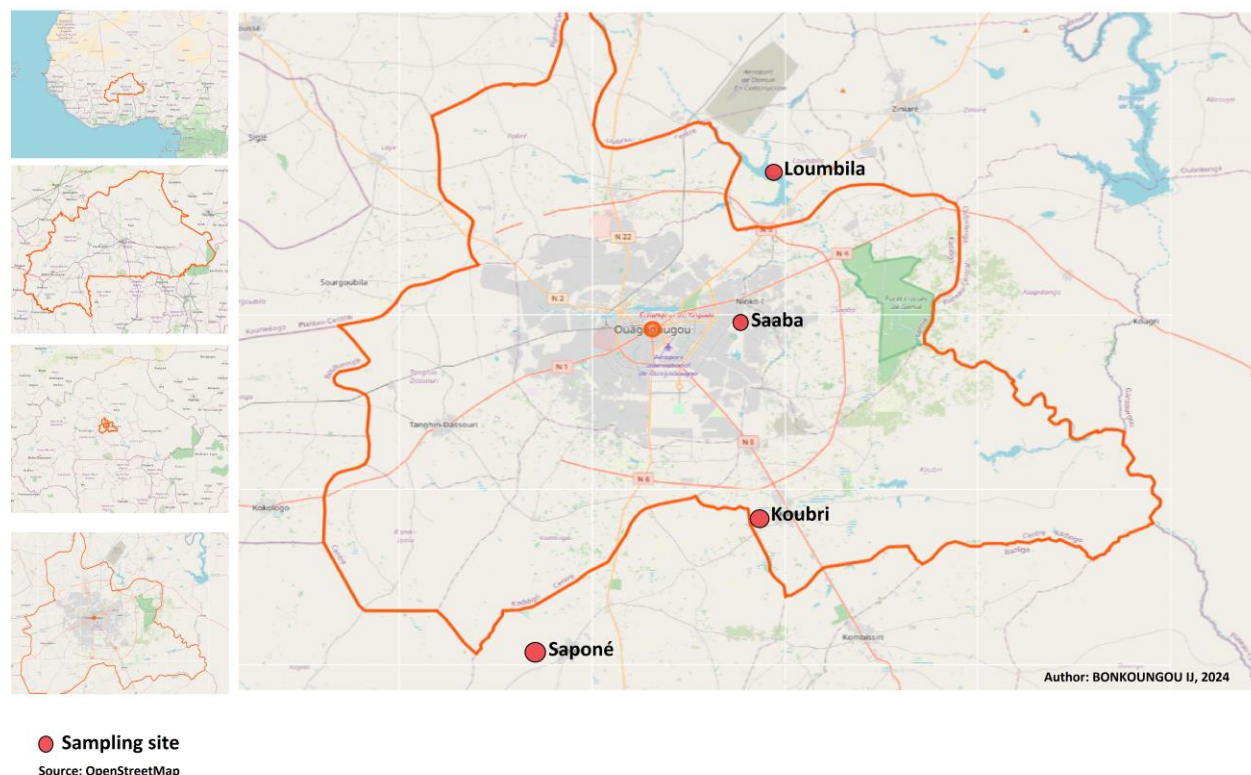
Figure 1. Map showing study sampling sites.

techniques. Molecular detection of the bla_{CTX-M}, bla_{TEM}, and bla_{SHV} genes was performed using PCR-based assays. These genes were targeted due to their significant role in the production of ESBLs, which contribute to antibiotic resistance in Enterobacterales. All samples were analyzed at the "Unité de génomique des Pathogènes One Health (UgenoPath-OH)" at the Joseph KI-ZERBO University, Ouagadougou, Burkina Faso. Ethical approval was obtained from the Health Research Ethics Committee (CERS) of Burkina Faso under the number N° 153-12-2018/CERS, and informed consent was sought from all participants before sample collection.

Sample collection and processing

Fresh fecal samples were collected from henhouses of 37 poultry farm and pooled by farm. A 10 g portion of each pooled fecal sample was suspended in 90 mL of sterile peptone buffer water (PBW), vigorously vortexed, and incubated at 37°C for 24 h for enrichment. From this enriched suspension, 1 mL was plated on MacConkey agar containing 4 µg/mL of cefotaxime. The bacterial isolates obtained were considered as putative ESBL-producers.

Bacterial isolates were identified to the species level using biochemical tests.

ESBL phenotype confirmation and Susceptibility test

Antibiotic susceptibility testing was performed using the Kirby–Bauer disk diffusion method (Bauer, 1959). Phenotypic detection of ESBL production was conducted using the double disc synergy test with Ceftriaxone (30 µg) and Cefepime (30 µg) in combination with Amoxicillin-Clavulanic acid (30 µg), according to EUCAST guidelines 2023 (EUCAST, 2023).

Confirmed ESBL isolates were tested for susceptibility to the following antibiotic disks (Bio-Rad, Marnes-la-Coquette, France): Aztreonam (ATM, 30 µg), Cefixime (CFM, 5 µg), Ceftazidime (CAZ, 10 µg), Cefoxitin (FOX, 30 µg), Cefepime (FEP, 30 µg), Amoxicillin-clavulanic acid (AUG, 10/260 µg), Imipenem (IMI), Ciprofloxacin (CIP, 5 µg), Chloramphenicol (C, 30 µg), Gentamicin (CN, 10 µg) and Trimethoprim-Sulfamethoxazole (SXT, 25 µg), Tetracycline (TE, 30 µg). Isolates resistant to Cefoxitin or Ceftazidime were confirmed as third-generation cephalosporin resistant (CRe).

Isolates with decreased susceptibility (inhibition zone <18 mm) to Cefoxitin (30 µg) were suspected of expressing AmpC AmpC expression was suspected in isolates with reduced susceptibility (inhibition zone < 18 mm) to cefoxitin (30 µg). Presumptive AmpC-βlactamase producers' bacterial isolates were tested for AmpC-β-lactamase production using MH agar supplemented with cloxacillin at 4 µg/L. A bacterial suspension prepared with fresh colonies (McFarland 0.5) was inoculated on to the entire surface of the MH agar supplemented with cloxacillin at 4 µg/L and a disc of cefoxitin was placed on the plate. The test was positive if the inhibition zone diameter around the cefoxitin disc was ≥18 mm.

Isolates with inhibition zone of < 21 mm to imipenem (10 µg) were considered carbapenem resistant (EUCAST, 2023). *Klebsiella pneumoniae* ATCC 700603 and *E. coli* ATCC 25922 were used as quality control strains.

Molecular detection of beta-lactamase producing genes

For each isolate, 10 µL of pure culture on Mueller Hinton agar was suspended in 600 µL Milli-Q® water, heated for 10 min at 100°C,
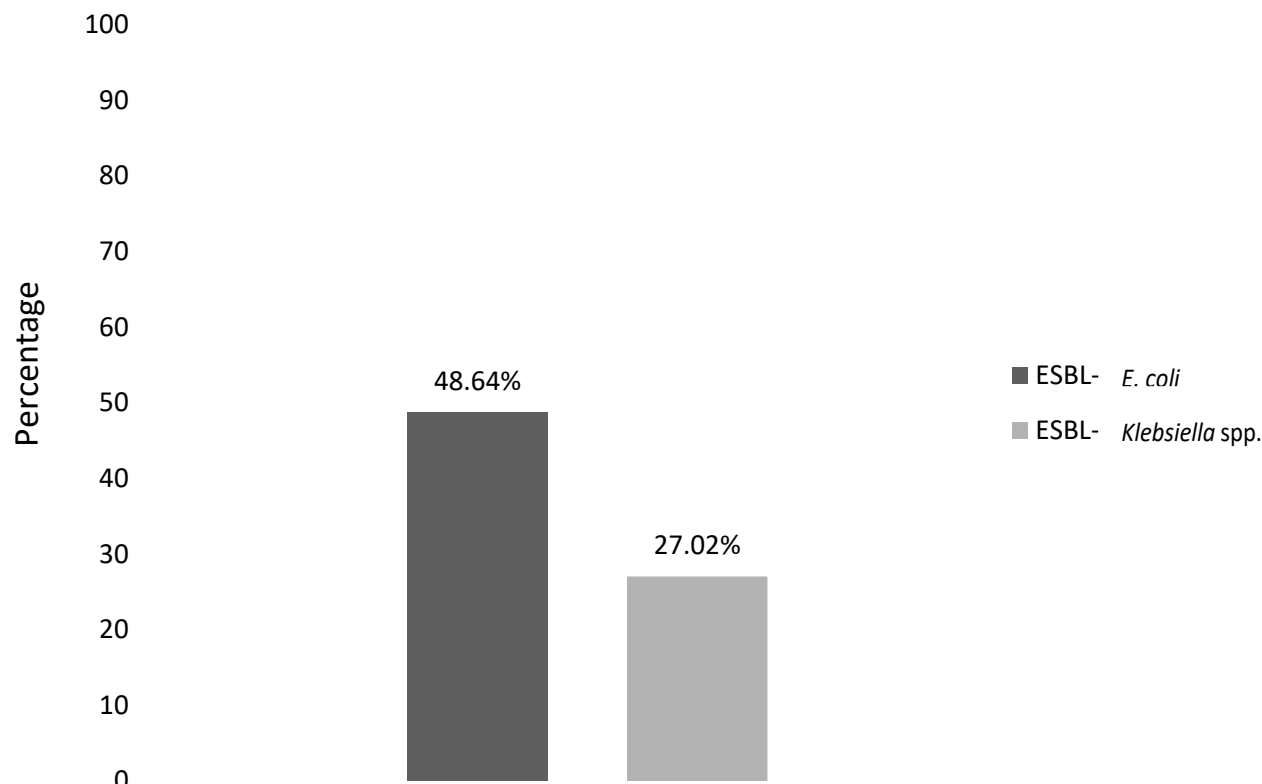
Figure 2. Rate of Extended Spectrum Beta lactamase producing *E. coli* and *Klebsiella* spp.

and subsequently centrifuged for 10 min at 4°C and 18000 rpm. The supernatant DNA lysate was transferred into sterile 1.5 mL Eppendorf® tubes for storage at -5°C. Multipex PCR was performed to amplified *bla*TEM, *bla*SHV, *bla*CTX-M genes.

Data analysis

Data were entered into a Microsoft Excel spreadsheet for analysis by proportions and percentages. Multi-drug-resistant (MDR) isolates were defined as those that were resistant to at least one antibiotic agent in at least three different categories of antimicrobials, including monobactams, carbapenems, aminoglycosides, penicillins, penicillins/β-lactamase inhibitors, antipseudomonal penicillins/β-lactamase inhibitors, cephalosporins, first and second generation cephalosporins, third and fourth generation cephalosporins, and carbapenems.

RESULTS

Prevalence of the ESBLs producing *E. coli* and *Klebsiella* spp. in poultry

A total of 37 pooled poultry fecal samples, based on the number of consenting farms, were collected. Of these, 75.76% (28/37) tested positive for ESBL-producing *E. coli* and *Klebsiella* spp. Specifically, *E. coli* was found in 64.28% (18/28) of the positive samples, while *Klebsiella* spp. was present in 35.71% (10/28) (Figure 2).

Co-resistance associated to the production of ESBLs

In this study, tetracycline and trimethoprim-sulfamethoxazole exhibited the highest rates among the tested antibiotics. For tetracyclin, resistance was observed in 100% (10/10) were noted of *Klebsiella* spp. Isolates and 83.33% (15/18) of for *E. coli* isolates. Similarly, resistance to trimethoprim-sulfamethoxazole was recorded at 100% (10/10) for *Klebsiella* spp. and 72.22% for *E. coli* strains. Conversely, lower resistant rates were noted for aminoglycosides and quinolones. Resistance to ciprofloxacin was identified in 33.33% (6/18) of *E. coli* isolates and 30% (3/10) of *Klebsiella* spp. Isolates. Resistance rate of gentamicin were 16.16% (3/18) in *E. coli* and 20% (2/10) in *Klebsiella* spp; while for amikacin, the rates were16.66% (3/18) and 10% (1/10) for *E. coli* and *Klebsiella* spp.

ESBLs producing genes and antibiotic resistance of *E. coli* isolated from poultry

Among ESBL-producing *E. coli* isolates, *bla*CTX-M gene was detected in 88.88% (16/18), while the *bla*TEM and *bla*SHV gene were identified in 44.44% (8/18), and 11.11% (2/18) of the isolates, respectively. Co-occurrence of the *bla*CTX-M and *bla*TEM gene was observed in 44.44% (8/18)

of isolates, and co-carrying of bla_{CTX-M} and bla_{SHV} gene was found in 11.11% (2/18) of isolates (Table 1.). Additionally, 33.33% (6/18) of isolates harbored only the bla_{CTX-M} gene. A MDR phenotype was identified in 94.44% (17/18) of isolates (Table 1).

ESBLs producing genes and antibiotic resistance of *Klebsiella* spp. isolated from poultry

Among the 10 ESBL-producing *Klebsiella* spp. isolates, AmpC and carbapenemase-producing phenotypes were observed in two different isolates (Table 2), the bla_{CTX-M} gene was detected in 90% (9/10) of isolates, while bla_{SHV} and bla_{TEM} gene were identified in 80% (8/10), and in 50% (5/10) of the isolates, respectively. Co-carrying of the bla_{CTX-M} and bla_{TEM} was found in 30% (3/10) of isolates. Co-occurrence of the bla_{CTX-M} and bla_{SHV} gene was noted in 40% (4/10) of isolates, while the bla_{TEM} bla_{SHV} gene co-occurrence was found in 40% (4/10) of isolates. The carbapenemase-producing isolate harbored bla_{CTX-M}, bla_{TEM}, and bla_{SHV}, while the AmpC-producing isolate harbored bla_{CTX-M} and bla_{TEM} genes. All the strains were multi-drug-resistant bacteria. Of 10 *Klebsiella* spp. isolate, AmpC and Carbapenemase producing phenotype were detected in two different isolates (Table 2). PCR identified bla_{CTX-M} gene in 90% (9/10) of isolates, bla_{SHV} in 80% (8/10) and bla_{TEM} in 50% (5/10). The co-carraige of $bla_{CTX-M/TEM}$ has been observed in 30% (3/10) of isolates, $bla_{CTX-M/SHV}$ has been identified in 40% (4/10) of isolates and $bla_{TEM/SHV}$ has in 40% (4/10) of isolated. The carbapenemase-producing isolate harbored bla_{CTX-M}, bla_{TEM}, and bla_{SHV} genes, while the AmpC-producing isolate harbored bla_{CTX-M} and bla_{TEM} genes. All the strains were multi-drug-resistant bacteria.

DISCUSSION

The characterization of ESBL-producing pathogens with public health interest in poultry remains poorly documented in Burkina Faso, yet it is crucial for the effective management and containment of infections. In this study, a prevalence of 75.76% (28/37) was noted for ESBL-producing bacteria. These results confirm the widespread distribution of antibiotic-resistant bacteria on farms in the peri-urban region of Ouagadougou. ESBL-producing *E. coli* was isolated in 64.28% (18/28) of ESBL-positive samples, underlining its dominant role in the production of ESBL in this environment. The presence of *Klebsiella* spp. was observed in 35.71% (10/28) of ESBL-positive samples, which is also a cause for concern, although less frequently than *E. coli*.

In national context, ESBL producing Enterobacteral were detected in 0.8% of poultry in intensive breeding (02/244) and 19.6% of poultry in extensive breeding (30/153), in study conducted in Bobo Dioulasso (Sanou

et al., 2019). From a global standpoint, high prevalence rates of ESBL-producing *E. coli* in African poultry, ranging from 1 to 100% have been reported from studies (Ayinla and Mateus, 2023). In Asia, the prevalence of ESBL has been also recorded at rates as high as 93% in poultry (Olaru et al., 2023). A global meta-analysis revealed ESBL prevalence in animals of 33.5% for *E. coli* and 19.4% for *Klebsiella* spp. (Ramatla et al., 2023). The results of this study are consistent with these worldwide observations, particularly in Africa and Asia, where the intensive use of antibiotics in intensive livestock farming is leading to increased selection of resistant strains.

This situation reflects the heavy use of antibiotics on farms to stimulate growth or prevent disease, a key factor in the selection of resistant strains. Poor regulation contributes to this problem, particularly in low- and middle-income countries These findings spotlight the concerning situation associated with the presence of beta-lactamase-producing bacteria on livestock farms in Burkina Faso and underscore the pressing necessity for enhanced surveillance, rigorous infection control measures, and implementation of effective antibiotic stewardship in poultry production is essential to mitigate the spread of antimicrobial resistance.

Highest rates of resistance were observed in this study, particularly with tetracycline and trimethoprim-sulfamethoxazole. *Klebsiella* spp. isolates showed alarming resistance rates of 100% (10/10) for both antibiotics, while *E. coli* isolates also showed high resistance rates, reaching 83.33% (15/18) for tetracycline and 72.22% for trimethoprim-sulfamethoxazole. These findings are in line with other studies, carried out high rate of antimicrobial resistance (AMR) in poultry farms, for tetracycline and sulfonamides, which are commonly used antibiotics in poultry farming for the prophylactic and curative treatment of infections.

The resistance rate of 100% noted from *Klebsiella* spp. regarding tetracycline and trimethoprim-sulfamethoxazole, could be due to the increased selective pressure exerted by the inappropriate use of these drugs on poultry farms. Study conducted in Nigerian poultry farms also revealed high resistance rates to tetracycline (81%) and sulfamethoxazole (67%) (Adelowo et al., 2014). These results underline the urgent need for stricter controls on the use of antibiotics in livestock farming, as well as the implementation of more sustainable and responsible farming practices to reduce the risk of antibiotic resistance spreading to the environment and to human populations.

The lowest rates of resistance in this study were observed with aminoglycosides and quinolones, including gentamicin, amikacin and ciprofloxacin. Ciprofloxacin, a widely used fluoroquinolone, showed relatively low resistance rates: 33.33% in *E. coli* and 30% in *Klebsiella* spp. Although these rates are lower than those observed for other antibiotics, they remain worrying because ciprofloxacin is often considered a second-line treatment

Table 1. Extended Spectrum of Beta-lactamase (ESBL) producing genes and antibiotic resistance of *E. coli* isolated from poultry.

Isolate ID	Isolate	Beta-lactamase phenotype	ESBL_producing genes	Antibiotic resistance*												MDR Bacteria
				AUG	FEP	CAZ	TE	CFM	ATM	FOX	SXT	CN	CIP	IMp	AK	
02SV	*E. coli*	ESBL	*bla*CTX-M and *bla*TEM													
04LV	*E. coli*	ESBL	none													
05LV	*E. coli*	ESBL	*bla*CTX-M and *bla*TEM													
12SV	*E. coli*	ESBL	*bla*CTX-M													
13KV	*E. coli*	ESBL	*bla*CTX-M													
14KV	*E. coli*	ESBL	*bla*CTX-M													
15KV	*E. coli*	ESBL	*bla*CTX-M and *bla*TEM													
18SPV	*E. coli*	ESBL	*bla*CTX-M and *bla*TEM													
20KV	*E. coli*	ESBL	*bla*CTX-M and *bla*TEM													
21LV	*E. coli*	ESBL	*bla*CTX-M													
22LV	*E. coli*	ESBL	*bla*CTX-M and *bla*TEM													
23LV	*E. coli*	ESBL	*bla*CTX-M and *bla*TEM													
24LV	*E. coli*	ESBL	*bla*CTX-M and *bla*SHV													
26LV	*E. coli*	ESBL	*bla*CTX-M													
46KV	*E. coli*	ESBL	*bla*CTXM													
50SPV	*E. coli*	ESBL	*bla*CTX-M and *bla*TEM													none
52SPV	*E. coli*	ESBL	*bla*CTX-M and *bla*SHV													
54KV	*E. coli*	ESBL	none													

*As per column headings, dark cells indicate "yes"; white cells indicate "no". AUG: Amoxicillin + clavulanic acid; FOX: cefoxitin; FEP: cefepime; CIP: ciprofloxacin; TE: tetracycline; AK: amikacin; CN: gentamycin; SXT: trimethoprim-sulfamethoxazole; IMP: imipenem; ATM: aztreoname; CAZ: ceftazidime; CFM: cefixime; nd: no detectable; MDR: multi drug resistant.

for serious infections caused by these bacteria (Sharma et al., 2017).

Aminoglycosides, such as gentamicin and amikacin, showed even lower resistance rates, particularly for *E. coli* with 16.16% resistance to gentamicin and 16.66% to amikacin. For *Klebsiella* spp. resistance rates were 20 and 10% respectively for these antibiotics. These results are consistent with those observed thought other studies in Africa, which report that resistance to aminoglycosides, although present, remains relatively rare in isolates of *E. coli* and *Klebsiella* spp. (Ayinla and Mateus, 2023). These low rates of resistance to aminoglycosides and quinolones

could be explained by the more restricted use of these classes of antibiotics in agricultural and veterinary practices, compared with beta-lactam antibiotics or tetracyclines, which are more widely used for prophylaxis and mass treatment (Sanou et al., 2019). These results underline the importance of monitoring the development of resistance to aminoglycosides and quinolones, as they remain crucial therapeutic options for the treatment of serious infections resistant to other antibiotics.

PCR revealed a significant presence of the *bla*CTX-M gene in 88.88% (16/18) of ESBL-producing *E. coli* isolates and 90% (9/10) of

Klebsiella spp. This aligns with the findings of other studies conducted in Africa and elsewhere, particularly in isolates from farm animals such as poultry (Ayinla and Mateus, 2023; Siriphap et al., 2022). The *bla*CTX-M gene is known to confer resistance to third-generation cephalosporins, thus limiting treatment options. The spread of ESBL-producing *E. coli* is facilitated by plasmid transfer and clonal transmission within and between farms (Alonso et al., 2017; Tansawai et al., 2019).

The detection rates for *bla*TEM (44.44%) and *bla*SHV (11.11%) observed for *E. coli* in this study are also in line with other studies which show that

Table 2. Extended Spectrum of Beta-lactamase (ESBL) producing genes and antibiotic resistance of *Klebsiella* spp. isolated from poultry.

Isolate ID	Isolate	Beta-lactamase phenotype	ESBL_resistant genes	Antibiotic Resistance*												MDR
				AUG	FEP	CAZ	TE	CFM	ATM	FOX	SXT	CN	CIP	IMP	AK	
01SV	*Klebsiella* spp.	ESBL	*blaCTX-M, blaTEM* and *blaSHV*	■	■	■	■	■	■		■					■
08LV	*Klebsiella* spp.	Ampc	*blaCTX-M* and *blaTEM*	■	■	■	■	■	■		■					■
11SV	*Klebsiella* spp.	ESBL	*blaCTX-M* and *blaSHV*	■	■	■	■	■	■		■					■
17KV	*Klebsiella* spp.	ESBL	*blaTEM* and *blaSHV*	■	■	■	■	■	■			■	■			■
19KV	*Klebsiella* spp.	ESBL	*blaCTX-M, blaTEM* and *blaSHV*		■	■	■	■	■		■		■			■
25LV	*Klebsiella* spp.	ESBL/Carbapenemase co-producer	*blaCTX-M, blaTEM* and *blaSHV*	■	■	■	■	■	■		■		■	■	■	■
44KV	*Klebsiella* spp.	ESBL	*blaCTX-M* and *blaSHV*	■	■	■	■	■	■			■	■			■
45KV	*Klebsiella* spp.	ESBL	*blaCTX-M* and *blaSHV*	■	■	■	■	■	■		■					■
51SPV	*Klebsiella* spp.	ESBL	*blaCTX-M*	■	■	■	■	■	■		■					■
53SPV	*Klebsiella* spp.	ESBL	*blaCTX-M* and *blaSHV*	■	■	■	■	■	■		■					■

*As per column headings, dark cells indicate "yes"; white cells indicate "no"; AUG=Amoxicillin + Clavulanic acid; FOX: cefoxitin; FEP: cefepime; CIP: ciprofloxacin; TE: tetracycline; AK: amikacin; CN: gentamycin; SXT: trimethoprim-sulfamethoxazole; IMP: imipenem; ATM: aztreoname; CAZ: ceftazidime; CFM: cefixime; nd: no detectable; MDR, multi drug resistant.

although these genes are less frequent than *blaCTX-M*, they continue to play an important role in beta-lactam resistance (Siriphap et al., 2022). Concerning *Klebsiella* spp., 80% (8/10) of isolates carried the *blaSHV* gene and 50% (5/10) of isolate carried *blaTEM* gene, illustrating the diversity of resistance genes present in these isolates. Co-carriage of *blaCTX-M/TEM* and *blaCTX-M/SHV* genes was also observed in 44.44 and 11.11% of ESBL-producing *E. coli* isolates, respectively. These findings have been reported in other studies as well (Gundran et al., 2019; Faridah et al., 2023), highlighting the ability of strains to accumulate multiple resistance genes, which can encode different resistance mechanisms, thereby increasing the complexity of their treatment.

Regarding *Klebsiella* spp., co-carriage has been observed, including *blaCTX-M/TEM* (30%), *blaCTX-M/SHV* (40%), and *blaTEM/SHV* (40%). This highlights the ability of *Klebsiella* spp. isolates to accumulate multiple resistance mechanisms through the carriage of multiple antimicrobial resistance

genes, thus reinforcing their multi-resistance potential and complicating the treatment of infections caused by MDR *Klebsiella* spp.

The observation that 33.33% (6/18) of *E. coli* isolates possessed only the *blaCTX-M* gene further reinforces the importance of this gene as the main beta-lactam resistance factor in avian environments. This could be linked to the intensive use of cephalosporins in veterinary medicine, which exerts a selective pressure favouring the dissemination of the *blaCTX-M* gene.

The observation of a MDR phenotype in 94.44% (17/18) of *E. coli* and 100% (10/10) of *Klebsiella* spp. is alarming, as it indicates that these strains are resistant to several classes of antibiotics. This increases the risk of ineffective treatments and the spread of resistance. These MDR strains pose a serious threat to animal and human health by limiting therapeutic options and increasing the risk of transmitting resistance genes to other pathogens via conjugative plasmids (Ayinla and Mateus, 2023).

The results highlight the presence of AmpC and carbapenemase-producing phenotypes in two distinct isolates of *Klebsiella* spp. These observations are particularly concerning because carbapenemases, such as KPC, NDM, and OXA-48, confer resistance to carbapenems, a class of antibiotics considered to be one of the last lines of defense against multi-resistant *Klebsiella* infections. The detection of the AmpC phenotype also suggests resistance to first to third-generation cephalosporins, further limiting therapeutic options. These findings underscore the widespread presence of ESBL-producing bacteria in livestock in Burkina Faso and the potential risk to public health.

Conclusion

This study provided information on the antibiotic resistance of Enterobacteriaceae (*E. coli* and *Klebsiella* spp. ESBL) isolated from different

poultry farms. ESBL-producing *E. coli* was the most frequently isolated bacterium in the poultry samples. Resistance to tetracycline and trimethoprim-sulfamethoxazole was very high. Nonetheless, sensitivity to cefoxitin and imipenem was noted. The isolates identified had a preponderance of *bla*$_{CTX-M}$ genes, according to molecular characterization of the resistance genes.

The presence of multi-resistant strains of ESBL-producing Enterobacteriaceae in the poultry industry poses a threat to public health, significantly reducing the therapeutic alternatives available for treating infections. Efforts to improve the organization of this sector are crucial to enhancing access to veterinary services, educating poultry producers about the risks associated with antibiotic misuse, and promoting essential hygiene practices to interrupt the transmission of antibiotic-resistant bacteria to humans and the environment.

However, this study has certain limitations. The small sample size and the focus on peri-urban areas of Ouagadougou limit the generalizability of the findings to the broader population of poultry farmers in Burkina Faso, particularly those in rural regions where farming practices may differ.

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CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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