



## Research article

# Antibiotic susceptibility of *Escherichia coli* originated from raw chicken meat in Vinh Long province's retail markets

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## Abstract

One of the bacteria that is frequently found on uncooked poultry meat is *Escherichia coli* (*E. coli*), which commonly results in food poisoning in customers. Therefore, this work was conducted to assess the prevalence and antibiotic susceptibility of *E. coli* recovered from raw chicken meat in the retail markets of Vinh Long Province. The findings revealed that the *E. coli* concentration in raw chicken meat at retail markets exceeds the specified standards for fresh meat. A total of forty-nine *E. coli* strains were obtained from retail chicken samples. On the basis of morphological and biochemical characteristics, 16S rRNA gene sequencing, and phylogenetic analysis, the 3 bacterial strains EP4\_12, ETB\_38, and EBM\_45 were determined to be *E. coli*. The results of antibiotic susceptibility testing demonstrated that the isolated *E. coli* strains were susceptible to ampicillin-sulbactam, sulfamethoxazole-trimethoprim, and ceftazidime at rates of 84%, 47%, and 41%, respectively. In contrast, these bacteria were resistant to ampicillin, ciprofloxacin, amoxicillin, doxycycline, levofloxacin, streptomycin, and tetracycline at percentages of 82%, 73%, 69%, 65%, 49%, 49%, and 55%, respectively. Notably, 94% of the bacterial isolates identified in this study were multiresistant to many antibiotics (multiresistant to 3 to 10 antibiotics), of which multiresistant to six antibiotics accounted for the highest percentage (20%). Moreover, the findings also revealed that *E. coli* isolates were frequently exposed to antibiotics with a multiple antibiotic resistance (MAR) index > 0.2. Consequently, it is important to manage the spread of antibiotic-resistant *E. coli* in fresh chicken retail markets to protect consumer health.

**Keywords:** Antibiotic resistance, Chicken meat, *E. coli*, Food poisoning, Retail market.

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## INTRODUCTION

Chicken meat is one of the most widely used poultry foods in the world as well as in Vietnam because of its high nutritional value, easy digestibility, absorption, and health benefits for consumers (Marangoni et al., 2015; Mottet and Tempio, 2017). However, the consumption of uncooked foods of animal origin is often unsafe because of the potential for bacteria, viruses, parasites, or harmful chemicals (Lehel et al., 2020; Ali and Alsayeqh, 2022). Food poisoning is caused mostly by contaminated food, which kills many people worldwide. The World Health Organization (WHO) estimates that eating tainted food causes illnesses in 600 million people annually, 420,000 of whom pass away, and 33 million years of good life are lost (WHO, 2022). In Vinh Long Province, chicken meat (slaughtered and unprocessed chicken meat) is widely sold in retail markets or supermarkets. In general, most of the chicken meat in traditional markets is supplied from small, manual slaughterhouses with unsanitary conditions, so there is a risk of bacterial contamination from the water source, and the excretion of chicken meat at the slaughterhouse or at the place of sale is very high. Many reports have shown that pathogenic bacteria in humans, including *E. coli* and *Salmonella*, are present in poultry meat (Adeyanju and Ishola, 2014; Rouger et al., 2017; Julqarnain et al., 2022).

*E. coli* is a rod-shaped, gram-negative, anaerobic bacteria that belongs to the family Enterobacteriaceae (Ekici and Dümen, 2019). This is a very dangerous gastrointestinal disease-causing bacteria that is likely related to chicken meat sources that are produced and processed without ensuring food hygiene and safety (Hang et al., 2020). *E. coli* is transmitted mainly through the gastrointestinal tract; it is also capable of growing under both aerobic and anaerobic conditions. As common bacterial flora in the gastrointestinal tracts of people and animals, the majority of *E. coli* strains inadvertently invade them (Kaper et al., 2004). However, some strains have caused infections, such as urinary tract infections and stomach and intestinal infections (Nataro and Kaper, 1998). In particular, *E. coli* isolates are capable of treating diarrheal diseases such as enteroinvasive *E. coli* (EIEC) (Allocati et al., 2013), enterotoxigenic *E. coli* (ETEC) (Gomes et al., 2016), enteropathogenic *E. coli* (EPEC) (Robins-Browne et al., 2016), enterohemorrhagic *E. coli* (EHEC) (Robins-Browne et al., 2016), and enteroaggregative *E. coli* (EAEC) (Allocati et al., 2013). Among them, enterohemorrhagic *E. coli* (EHEC) is a Shiga toxin-producing *E. coli* strain that causes hemolytic uremic syndrome (HUS) and is potentially fatal for humans (Gomes et al., 2016).

Treatment for bacterial infections is being impacted by the growing issue of antibiotic resistance in bacteria. Vietnam is among the countries with the greatest percentage of antibiotic resistance worldwide (Ahmed and Rahman, 2022). Many previous studies have shown that *E. coli* in fresh meat is resistant to many antibiotics (Hleba et al., 2015; Horri et al., 2022; Brätfelan et al., 2023; Hasib et al., 2024; Mortensen et al., 2024). According to Arbab et al. (2022), *E. coli* is one of the reservoirs of antibiotic resistance genes in a community. In *E. coli*, many antibiotic resistance genes are able to spread drug resistance genes among pathogenic bacteria in the environment (Maeusli et al., 2020; Mota-Bravo et al., 2023). To date, many studies on the antibiotic resistance of pathogenic *E. coli* in poultry have been reported worldwide as well as in Vietnam (Vui and Tiep, 2016; Anh and Khai, 2017; Vounba et al., 2019; Bhattarai et al., 2024). However, *E. coli* resistance in chicken meat in retail markets is still limited. Therefore, this work was performed to evaluate the presence and antibiotic susceptibility of *E. coli* strains in chicken meat in Vinh Long Province's retail markets.

## MATERIALS AND METHODS

### Chicken samples

Raw chicken meat samples were obtained at different retail markets (consisting of small, manual slaughterhouses) in Vinh Long Province from 2022-2023. For each selected market (high demand for chicken meat), three fresh chicken meat samples (muscle, skin, and liver) were randomly selected. One hundred sixty-two chicken meat samples (pooled samples) from retail markets were obtained. Sample collection at different locations was performed during the same time of day. The samples were taken to the laboratory to be examined for *E. coli* count determination and isolation after being placed in sterile plastic bags.

### Enumeration of *E. coli* in chicken meat

The bacterial cell density was determined according to [Hoben and Somasegaran \(1982\)](#). First, chicken muscle, skin, and liver samples were combined and homogenized with sterilized distilled water. The sample was shaken on a shaker at 120 rpm for thirty minutes. Next, the sample was serially diluted to a  $10^{-6}$  dilution. A 0.1 mL aliquot of sample at each dilution concentration (repeated 3 times) was spread on a Petri dish containing MacConKey agar medium (Himedia, India). Finally, the quantity of bacterial colonies growing on the medium was recorded after 24 hours, and the *E. coli* count was determined according to the following formula (log CFU/g):  $\text{cfu/g} = \text{level of dilution plated} \times \text{number of colonies counted/volume plated}$  ([Adeyanju and Ishola, 2014](#)).

### Isolation of *E. coli*

The above diluted samples were used to isolate *E. coli*. Similarly, 100  $\mu\text{L}$  of sample was spread on MacConKey agar (Himedia, India) and incubated at 37°C. After 24 hours, typical colonies were selected and subcultured several times on the corresponding media until homogeneous colonies were obtained. The characteristics of the colony morphology, motility, Gram stain, catalase, and oxidase reactions of the isolated bacterial strains were examined ([Cappuccino and Welsh, 2017](#)).

### Antibiotic susceptibility of *E. coli* strains

The antibiotic sensitivity of *E. coli* was determined via the disk diffusion assay method of Kirby-Bauer ([Bauer et al., 1966](#)). *E. coli* strains were tested for antibiograms with twelve antibiotics, including ceftazidime (CEF/30  $\mu\text{g}$ ), streptomycin (STR/10  $\mu\text{g}$ ), kanamycin (KAN/30  $\mu\text{g}$ ), gentamicin (GEN/10  $\mu\text{g}$ ), tetracycline (TET/30  $\mu\text{g}$ ), sulfamethoxazole-trimethoprim (SXT/23.75/1.25  $\mu\text{g}$ ), ciprofloxacin (CIP/5  $\mu\text{g}$ ), levofloxacin (LEV/5  $\mu\text{g}$ ), amoxicillin (AMO/10  $\mu\text{g}$ ), ampicillin (AMP/10  $\mu\text{g}$ ), doxycycline (DOX/30  $\mu\text{g}$ ), and ampicillin-sulbactam (AMS/10/10  $\mu\text{g}$ ). In brief, bacterial colonies of 0.85% NaCl physiological saline sterilized the suspension at a density of  $10^8$  CFU/mL by comparing the turbidity with that of the McFarland standard tube. The bacterial mixture (0.1 mL) was then spread onto Mueller Hinton agar (MHA, Mecrk, Germany). Next, the antibiotic discs were placed in Petri dishes and incubated at 30°C. Finally, the diameter of the sterile zone on the agar surface was measured and assessed for resistance (R), intermediate (I), and susceptible (S) strains on the basis of CLSI standards ([Clinical and Laboratory Standards Institute, 2021](#)) after 24 hours of incubation. Multidrug resistance was defined as resistance to three or more classes of antibiotics ([Davis et al., 2018](#)). In this study, *E. coli* ATCC® 25922 was used as a quality control.

## Multiple antibiotic resistance index (MAR index)

The multiple antibiotic resistance index (MAR index) of each isolated bacterial strain was determined on the basis of the formula of [Jain et al. \(2021\)](#) as follows:  $MAR\ index = a/b$ , where  $a$  indicates the quantity of antibiotics to which the isolate exhibited resistance and  $b$  represents all antibiotics used. Bacterial strains with a MAR index of less than or equal to 0.2 are high-risk sources of contamination where antibiotics are regularly used ([Krumperman et al., 1983](#)).

## Identification of *E. coli* bacteria isolated from chicken meat

### Extraction of bacterial DNA

Bacterial DNA was extracted according to the methods of [Dung \(2011\)](#), with some minor modifications. Briefly, *E. coli* bacterial isolates were enriched in Trypticase Soy Broth (TSB, Himedia, India) and placed on a shaker at 110 rpm at room temperature. Then, 2 mL of the bacterial mixture was centrifuged at 13000 rpm for 5 minutes. Next, the bacterial biomass was dissolved in 1 mL of lysis buffer (1 M Tris-HCl, 0.5 mM EDTA, 5 M NaCl, 0.1% SDS, pH 8.0) and incubated at room temperature for 10 minutes. Ethanol was used to precipitate the DNA-containing supernatant, and 70% ethanol was used to dissolve it again. After extraction, the optical density of the bacterial DNA was measured at wavelengths of 260 and 280 nm to determine its purity and concentration. Finally, the DNA samples were stored in 100  $\mu$ L of 0.1 $\times$  TE mixture (10 mM Tris-HCl and 1 mM EDTA, pH 8.0) at -40°C for PCR.

### PCR

The universal primer pairs 27F: 5'-AGATTTGATCCTGGCTCAG-3' and 1492R: 5'-GGTTACCTTGTTACGACTT-3' ([Heuer et al., 1997](#)) were used to amplify the 16S rRNA gene fragment of the isolated bacterial isolates with an expected amplicon size of 1.500 bp. PCRs were performed in a volume of 25  $\mu$ L containing the following components: distilled water, 1X PCR buffer, 1.5 mM MgCl<sub>2</sub>, dNTPs (200  $\mu$ M), 20 pmol forward primer, 20 pmol reverse primer, 2.5 UI *Taq* DNA polymerase, and the DNA sample. The PCR cycle and conditions included initial denaturation at 94°C for 5 minutes, followed by 30 cycles of denaturation at 94°C for 1 minute, primer annealing at 63°C for 1 minute, extension at 72°C for 2 minutes, and a final extension at 72°C for 10 minutes. After amplification, the PCR products were electrophoresed on a 1.5% agarose gel, photographed via the Analytik Jena gel imaging system, and sent for sequencing at MacroGen Company (Korea) to identify the bacterial species.

### Data analysis

Descriptive statistical methods were used to calculate the average values and the average percentages of resistant, susceptible, and susceptible bacteria. Using the BLASTn tool, the similarity of the isolated bacterial DNA sequence with the sequences of *E. coli* bacterial strains in the National Center for Biotechnology Information (NCBI) database was determined. The bacterial 16S rRNA gene segment sequences were compared via the CLUSTAL W program ([Thompson et al., 1997](#)). A phylogenetic tree showing the genetic relationships between bacterial isolates was built via MEGA X software via the neighbor-joining algorithm ([Saitou and Nei, 1987](#)), with bootstrap values of 1.000 replications ([Tamura et al., 2013](#)).

## RESULTS

### Densities of *E. coli* in chicken meat

The findings indicated that eighty-three out of one hundred sixty-two (51.23%) chicken meat samples from retail markets were contaminated with *E. coli*, with densities ranging from  $4.77 \pm 0.25$  to  $5.05 \pm 0.05$  CFU/g (Table 1). Generally, the concentration of *E. coli* was greater than the allowed standard of  $\leq 10^2$  CFU/g fresh meat (TCVN 7046:2019), but the difference was not statistically significant between retail markets in the sampling districts ( $P > 0.05$ ). The *E. coli* concentrations in chicken meat samples from various retail markets in Vinh Long Province are presented in detail in Table 1.

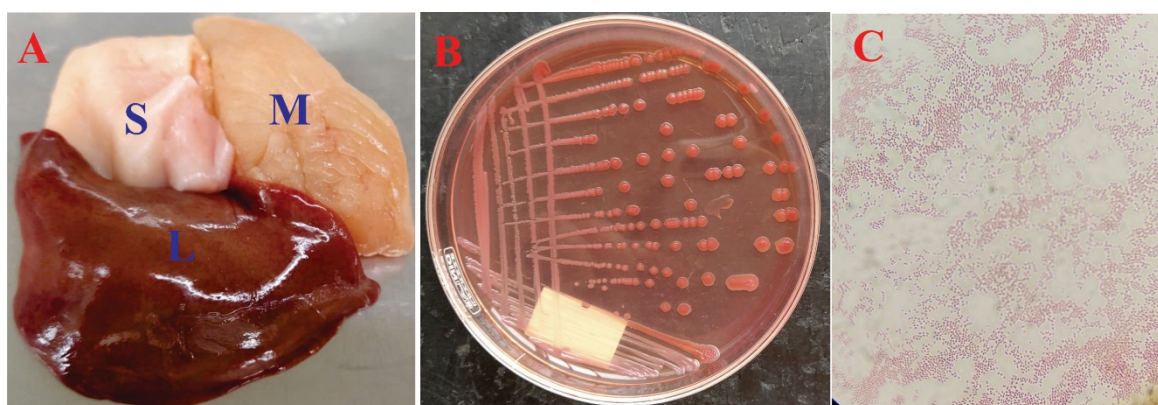
**Table 1** Densities of *E. coli* on chicken meat at retail markets

| Sample collection location | Number of <i>E. coli</i> -contaminated samples | <i>E. coli</i> bacterial densities (log CFU/g) |
|----------------------------|--|--|
| Vinh Long City             | 12   | $4.95 \pm 0.08^a$                              |
| Long Ho district           | 9  | $4.81 \pm 0.12^a$                              |
| Tam Binh district          | 8  | $4.99 \pm 0.04^a$                              |
| Tra On district            | 10   | $4.81 \pm 0.15^a$                              |
| Vung Liem district         | 11   | $4.84 \pm 0.08^a$                              |
| Mang Thit district         | 12   | $4.77 \pm 0.25^a$                              |
| Binh Minh district         | 10   | $4.89 \pm 0.11^a$                              |
| Binh Tan district          | 11   | $5.05 \pm 0.05^a$                              |

\*Note: mean  $\pm$  standard deviation; in the same column, different letters followed by numbers are significantly different ( $P < 0.05$ ).

### Isolation of *E. coli* bacterial strains from chicken meat

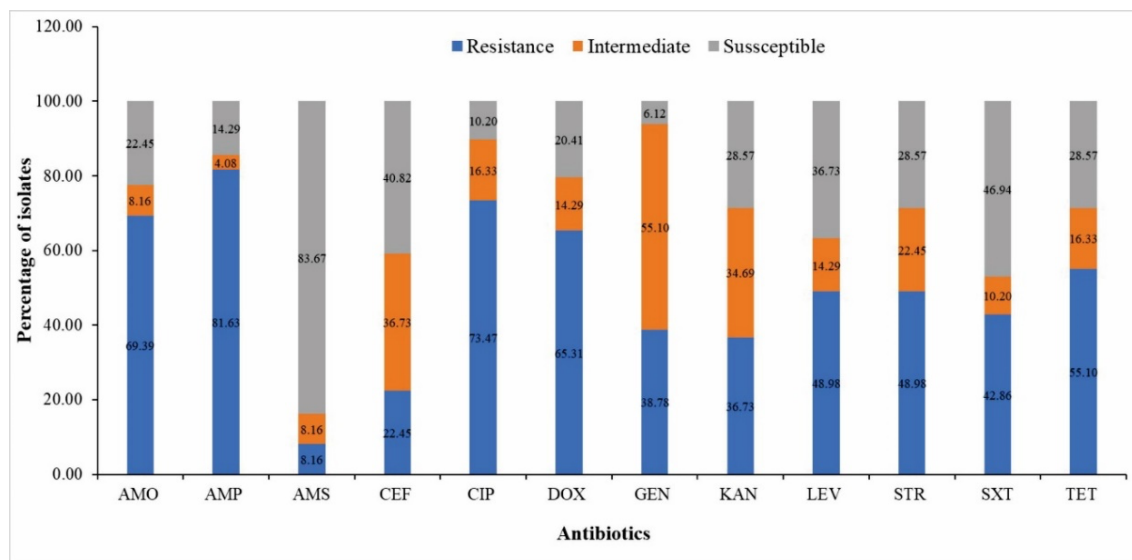
From eighty-three *E. coli*-contaminated chicken meat samples from retail markets, a total of eighty-three strains of *E. coli* were isolated on MacConkey agar media from pooled samples of chicken muscle, liver, and skin on the basis of colony morphology (Figure 1a). The bacterial colonies cultured on MacConkey agar were all reddish pink in color, raised, rough, round, and tiny, with colony diameters ranging from 1–5 mm after 24–48 hours of incubation at 37°C (Figure 1b). All the bacterial strains were motile, gram-negative, and short and rod shaped (Figure 1c), and all were positive for catalase and oxidase reactions.



**Figure 1** *E. coli* strain isolated on MacConkey medium. A. Chicken meat (skin-S, muscle-M, liver-L) for *E. coli* isolation; B. *E. coli* bacterial colonies grow on MacConkey medium; C. Gram staining (X100).

## Antibiotic sensitivity of *E. coli* strains

This investigation revealed that the isolated *E. coli* strains were susceptible to ampicillin-sulbactam (84%), sulfamethoxazole-trimethoprim (47%), and ceftazidime (41%) and had low sensitivity to the antibiotic gentamicin (6%). In contrast, bacteria were resistant to ampicillin (82%), ciprofloxacin (73%), amoxicillin (69%), doxycycline (65%), levofloxacin (49%), streptomycin (49%), tetracycline (55%), and the antibiotic ampicillin-sulbactam (8%). The antibiogram results of the *E. coli* isolates are presented in detail in Figure 2.

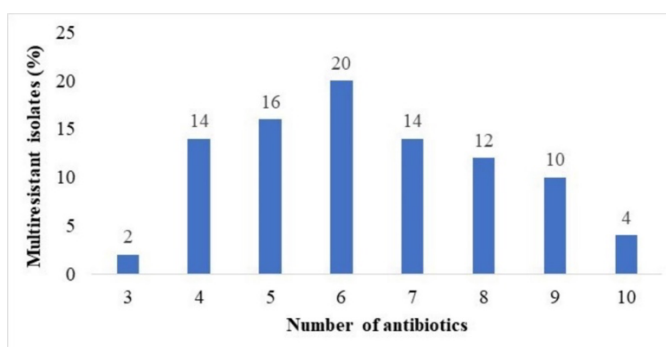


**Figure 2** Proportion of *E. coli* strains susceptibility to antibiotics

\*Note: AMO (amoxicillin), AMP (ampicillin), AMS (ampicillin-sulbactam), CEF (ceftazidime), CIP (ciprofloxacin), DOX (doxycycline), GEN (gentamicin), KAN (kanamycin), LEV (levofloxacin), STR (streptomycin), SXT (sulfamethoxazole-trimethoprim), and TET (tetracycline).

## Multidrug resistance in bacteria

This study revealed that 94% of the *E. coli* isolates in the study (Figure 3) had multidrug resistance or multiresistance (the capacity of certain microbes to withstand the effects of several antimicrobial agents), except for the EP1\_2 bacterial strain, which did not show resistance. Among them, most bacterial isolates were resistant to 10 types of antibiotics (4%), and a minority were resistant to 3 antibiotics (2%). In addition, the isolated bacterial strains were multiresistant to 6 antibiotics, accounting for the highest proportion (20%), followed by those that were multiresistant to 5 antibiotics (16%) and those that were multiresistant to 4 or 7 antibiotics. (14%), multiresistant to 9 antibiotics (10%), and the lowest, multiresistant to 3 antibiotics (2%). The results also revealed that the *E. coli* isolates had forty-seven multiresistant phenotypes out of a total of forty-nine bacterial strains. Among them, two bacterial strains, ELH\_40 and EVL\_47, have the same multiresistant phenotype: AMO-AMP-AMS-CIP-DOX-GEN-STR-SXT (Table 2).



**Figure 3(A)** Multiple drug resistance rate of isolated *E. coli* strains.

### Multiresistance index (MAR) of *E. coli* bacterial strains

The findings revealed that the MAR index of the *E. coli* strains in this study ranged from 0.17–0.83 (Table 2). In general, most bacterial strains (46/48 isolates) had a MAR index > 0.2, of which strain EBM\_45 had the highest MAR index (MAR = 0.83).

**Table 2** Phenotypes and MAR index of *E. coli*

| Bacterial isolates | Multidrug resistance phenotype | MAR index |
|--------------------|--------------------------------|-----------|
| ETB_36             | AMP-LEV                        | 0.17      |
| ELH_39             | AMP-STR                        | 0.17      |
| EP3_8              | AMO-DOX-STR                    | 0.25      |
| EVL_48             | AMP-CEF-SXT                    | 0.25      |
| EP4_11             | AMP-DOX-LEV-STR                | 0.33      |
| ETH_23             | AMO-DOX-KAN-STR                | 0.33      |
| ETN_31             | AMP-AMS-STR-SXT                | 0.33      |
| ETO_32             | AMP-CEF-DOX-KAN                | 0.33      |
| ETB_37             | AMP-AMS-CIP-LEV                | 0.33      |
| EMT_43             | AMO-AMP-DOX-SXT                | 0.33      |
| EVL_49             | AMO-AMS-DOX-SXT                | 0.33      |
| EP3_7              | AMO-AMP-AMS-CEF-DOX            | 0.42      |
| EP3_9              | AMO-AMP-CEF-DOX-STR            | 0.42      |
| EP9_21             | AMP-GEN-LEV-STR-SXT            | 0.42      |
| ETH_25             | AMO-AMP-CIP-STR-SXT            | 0.42      |
| ETA_26             | AMO-AMP-CEF-CIP-STR            | 0.42      |
| ETA_28             | AMP-DOX-LEV-STR-SXT            | 0.42      |
| ETO_33             | AMO-AMP-AMS-CEF-SXT            | 0.42      |
| ETB_34             | AMO-AMP-AMS-CIP-STR            | 0.42      |
| ETB_35             | AMO-AMP-DOX-STR-SXT            | 0.42      |
| EBM_46             | AMO-AMP-KAN-STR-SXT            | 0.42      |
| EP5_14             | AMO-CEF-CIP-GEN-LEV-STR        | 0.50      |
| EP9_20             | AMP-CEF-CIP-GEN-STR-SXT        | 0.50      |
| ETH_24             | AMO-AMP-CIP-DOX-KAN-STR-SXT    | 0.50      |
| ETN_29             | AMO-AMS-CIP-GEN-STR-SXT        | 0.50      |
| ETN_30             | AMO-AMS-CEF-GEN-STR-SXT        | 0.50      |
| EMT_41             | AMO-AMP-DOX-LEV-STR-SXT        | 0.50      |

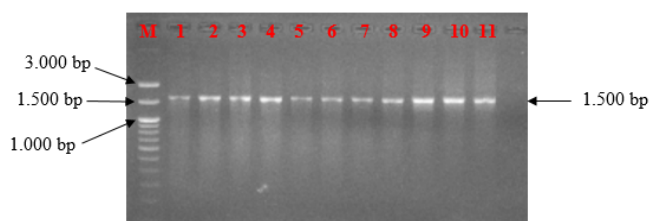
| Bacterial isolates | Multidrug resistance phenotype          | MAR index |
|--------------------|---|-----------|
| EMT_42             | AMO-AMP-DOX-KAN-LEV-SXT                 | 0.50      |
| EP1_1              | AMO-AMP-AMS-CEF-CIP-DOX-SXT             | 0.58      |
| EP2_4              | AMO-AMP-AMS-CIP-DOX-KAN-SXT             | 0.58      |
| EP2_5              | AMO-AMP-AMS-CIP-DOX-GEN-SXT             | 0.58      |
| EP5_13             | AMP-AMS-CIP-GEN-LEV-STR-TET             | 0.58      |
| EP8_18             | AMO-AMP-CEF-GEN-LEV-STR-SXT             | 0.58      |
| ETH_22             | AMO-AMP-AMS-GEN-STR-SUF/TRI-TET         | 0.58      |
| ETA_27             | AMO-AMP-CEF-CIP-GEN-STR-TET             | 0.58      |
| ETB_38             | AMP-CIP-DOX-GEN-LEV-STR-SXT             | 0.58      |
| EP1_3              | AMO-AMP-AMS-CEF-CIP-DOX-GEN-SXT         | 0.67      |
| EP2_6              | AMO-AMP-AMS-CIP-DOX-GEN-KAN-SXT         | 0.67      |
| EP4_10             | AMO-AMP-AMS-CIP-KAN-LEV-STR-SXT         | 0.67      |
| ELH_40             | AMO-AMP-AMS-CIP-DOX-GEN-STR-SXT         | 0.67      |
| EVL_47             | AMO-AMP-AMS-CIP-DOX-GEN-STR-SXT         | 0.67      |
| EP4_12             | AMO-AMS-CEF-CIP-DOX-GEN-LEV-STR-SXT     | 0.75      |
| EP8_15             | AMO-AMS-CEF-CIP-GEN-KAN-LEV-STR-SXT     | 0.75      |
| EP8_16             | AMO-AMP-AMS-CIP-DOX-LEV-STR-SUF/TRI-TET | 0.75      |
| EP8_17             | AMO-AMP-AMS-CEF-DOX-GEN-LEV-STR-SXT     | 0.75      |
| EP9_19             | AMO-AMP-CEF-CIP-DOX-GEN-LEV-STR-SXT     | 0.75      |
| EBM_44             | AMO-AMP-AMS-CIP-DOX-GEN-KAN-LEV-SXT     | 0.75      |
| EBM_45             | AMO-AMP-AMS-CEF-CIP-DOX-GEN-KAN-LEV-SXT | 0.83      |

\*Note: AMO (amoxicillin), AMP (ampicillin), AMS (ampicillin-sulbactam), CEF (ceftazidime), CIP (ciprofloxacin), DOX (doxycycline), GEN (gentamicin), KAN (kanamycin), LEV (levofloxacin), STR (streptomycin), SXT (sulfamethoxazole-trimethoprim), and TET (tetracycline).

## Bacterial identification via PCR

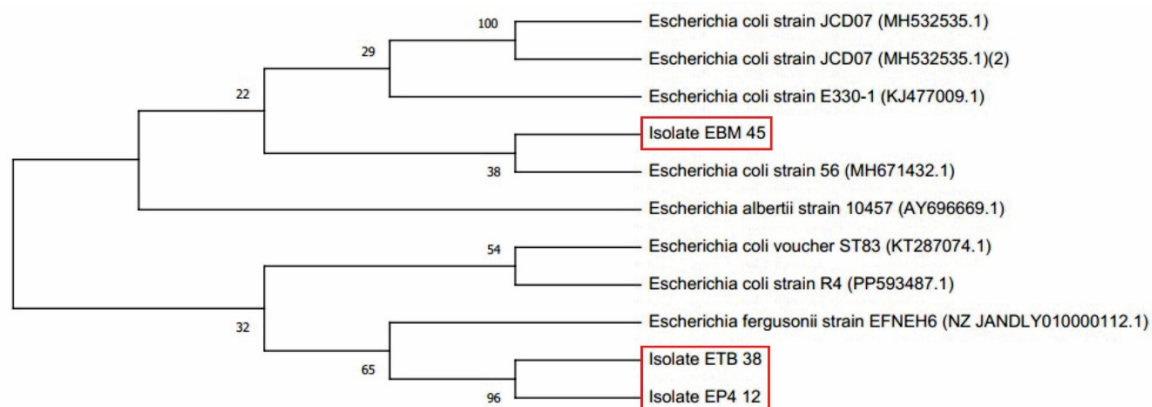
The results demonstrated that the 16S rRNA gene segment of all the isolated bacterial strains was amplified via PCR, with a product size of 1.500 bp (Figure 4).

The sequencing results revealed that the three representative strains EBM\_45, ETB\_38, and EP4\_12 had 97.60%, 99.75%, and 99.88% similarity, respectively, to the bacterial strains *E. coli* voucher ST83 (KT287074.1), *E. coli* strain 56 (MH671432.1), and *E. coli* strain JCD07 (MH532535.1) in GenBank. The phylogenetic tree revealed that the 3 isolated bacterial strains were divided into 2 groups, of which strains ETB\_38 and EP4\_12 had close genetic relationships and were in the same group as the *E. coli* strains in GenBank (Figure 5).



**Figure 4** Amplification of the 16S rRNA gene segment of representative *E. coli* strains by PCR reaction. M. DNA standard ladder 100 bp plus; Lane 1-11: bacterial strains EBM\_45, ETB\_38, EP4\_12, SP9\_9, SP8\_8, SP5\_7, EP3\_7, ETH\_22, ETH\_24, EP2\_6, and EP8\_16; Lane 12. Negative control.





**Figure 5** Phylogenetic tree showing isolated bacterial strains belonging to the same group as *E. coli* on GenBank (*E. albertii* strain 10457 (AY696669.1), and *Escherichia fergusonii* strain EFNEH6 (NZ\_JANDLY010000112.1) was used as an outgroup).

## DISCUSSION

*E. coli* is a mandatory indicator for evaluating food hygiene. This observation revealed that eighty-three chicken meat samples sold at retail markets were contaminated with *E. coli*. Additionally, the *E. coli* concentration in infected chickens in the present study was higher than the allowed standard of  $\leq 10^2$  CFU/g of fresh meat (TCVN 7046:2009). These investigations are similar to those of [Manh et al. \(2016\)](#), who reported that the *E. coli* contamination rate of chicken and duck meat samples was 80.6% and that the amount of *E. coli* was  $5912 \times 10^2$  CFU/g when the level of bacterial contamination of poultry meat at retail markets in Ben Tre City of Ben Tre Province, the Mekong Delta, was surveyed. Similarly, [Trieu et al. \(2022\)](#) demonstrated that fresh chicken meat was contaminated with *E. coli* at traditional markets in Buon Ma Thuot City of Dak Lak Province, Central Highlands, at a rate of 86.70%. Although the percentage of *E. coli* in the above studies was lower than that in chickens sold at retail markets in Vinh Long Province, all the studies revealed that the prevalence of *E. coli* in fresh chickens was high. The high rate and *E. coli* concentration confirmed the unsafe and hygienic conditions of chicken meat sold at retail markets. The cause of bacterial contamination in chicken meat sold at these markets may be due to unsafe hygienic conditions in the chicken production chain from the breeding site, the slaughter location, the storage location, and the transportation process to environmental factors at the place of sale that create conditions for bacterial populations to increase rapidly ([Manh et al., 2016](#)). In Zambia, a study by [Mphund \(2019\)](#) concluded that the high level of bacterial contamination of chicken meat was due to the environmental quality of the slaughterhouses as well as the implementation of hygiene measures, of which the most important factor is the water source used.

In the food sector, reports of *E. coli* antibiotic resistance in animal products have been published in numerous nations. ([Kim et al., 2020](#); [Worku et al., 2022](#); [Lee et al., 2023](#)). The *E. coli* isolates in this study were susceptible to ampicillin-sulbactam, trimethoprim-sulfamethoxazole, and ceftazidime at rates of 84%, 47%, and 41%, respectively. Moreover, the antibiogram results revealed that the isolated *E. coli* strains were resistant to ampicillin, ciprofloxacin, amoxicillin, doxycycline, levofloxacin, streptomycin, and tetracycline at proportions of 82%, 73%, 69%, 65%, 49%, 49%, and 55%, respectively. These findings are consistent with the research of [Noenchat et al. \(2024\)](#), who reported that the majority of *E. coli* O157:H7 isolates from cloacal swabs in Thai Lan were resistant to ampicillin (65.38%), amoxicillin (61.54%), streptomycin (61.54%), and oxytetracycline (65.38%).

Conversely, the majority of *E. coli* O157:H7 isolates found in breast meat were resistant to ampicillin (78.12%), amoxicillin (75%), streptomycin (25%), and oxytetracycline (78.12%). In the Mekong Delta, Vietnam, on the other hand, *E. coli* strains were found to be extremely resistant to amoxicillin (83.54%), ampicillin (78.48%), streptomycin (63.29%), and florfenicol (63.29%) by [Thong et al. \(2025\)](#). These *E. coli* strains, however, were susceptible to amoxicillin+acid clavulanic and amikacin (97.47%), cefaclor (95.45%), cefuroxime (93.67%), and enrofloxacin (92.41%). [Radwan et al. \(2020\)](#) reported that *E. coli* bacterial strains derived from chicken meat in five different provinces in Egypt were highly resistant to amoxicillin (97.5%), sulfamethoxazole (77.5%), and streptomycin (90%). In Qatar, according to [Eltai et al. \(2020\)](#), retail chickens have a significant frequency of *E. coli* that is resistant to antibiotics. Nearly 89% of the strains were resistant to at least one of the eighteen tested antibiotics, of which relatively high resistance was to sulfamethoxazole (62%), tetracycline (59.7%), ampicillin (52.3%), and ciprofloxacin (47.7%). Antibiotic-resistant bacteria have been reported to appear in stages from breeding sites ([Osman et al., 2018](#)), animal feed ([Ngai et al., 2021](#)), broiler and egg-laying chicken farming environments ([Adesiyun et al., 2020](#); [Assoumy et al., 2021](#)), or in processing plants and slaughterhouses, leading to the circulation of resistant bacteria in retail chicken products in traditional markets and posing a threat to consumer health ([Phiri et al., 2020](#)).

In the present study, 94% of the *E. coli* isolates were resistant to 3 to 10 antibiotics. This finding is in line with the work of [Akond et al. \(2009\)](#), who reported that all the strains of *E. coli* isolated from poultry and poultry environments in Bangladesh presented multiple resistances to more than six antibiotics. [Davis et al. \(2018\)](#) reported that 48% of the *E. coli* strains from retail poultry products in the USA, such as turkey meat, were multidrug resistant. In Bangladesh, [Rahman et al. \(2020\)](#) reported that 75.06% of the *E. coli* isolates in raw chicken meat samples collected from poultry shops were multidrug resistant. A similar study by [Parvin et al. \(2020\)](#) surveyed 113 frozen chicken samples purchased from 40 stores of nine branded supermarkets in five major cities in Bangladesh and reported that 76.1% of the samples were positive for *E. coli* and that all the isolates were multidrug resistant. A study by [Eltai et al. \(2020\)](#) revealed that 63.4% of *E. coli* strains isolated from retail chickens in Qatar were multidrug resistant. In Romania, according to [Brăţfelan et al. \(2023\)](#), more than 70% of *E. coli* isolates are multidrug resistant, showing resistance to at least three different classes of antibiotics. In Vietnam, [Trieu et al. \(2022\)](#) reported that 96.05% of *E. coli* originating from several traditional markets in Buon Ma Thuot city presented multidrug resistance. Recently, [Thong et al. \(2025\)](#) reported that 96.2% of the *E. coli* strains from pig slaughterhouses in the Mekong Delta were resistant to one to eight antibiotics.

## CONCLUSIONS

The present findings revealed that *E. coli* contamination was detected in 51.23% of survey samples from chicken meat sold at retail markets in Vinh Long, with concentrations higher than the allowed limit. On the basis of PCR techniques and 16S rRNA gene sequencing, the 3 isolated bacterial strains were identified as *E. coli* strains on the basis of a combination of morphological and biochemical characteristics. This investigation demonstrated that isolated *E. coli* isolates were highly resistant to ampicillin, ciprofloxacin, amoxicillin, doxycycline, levofloxacin, streptomycin, and tetracycline. Notably, most of the bacterial isolates in this work displayed resistance to many antibiotics with different multidrug-resistant phenotypes.

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## REFERENCES

- Adesiyun, A.A., Nkuna, C., Mokgoatheng-Mamogobo, M., Malepe, K., Simanda, L., 2020. Food safety risk posed to consumers of table eggs from layer farms in Gauteng Province, South Africa: Prevalence of *Salmonella* species and *Escherichia coli*, antimicrobial residues, and antimicrobial resistant bacteria. *J. Food. Saf.* 40(3), 1-12.
- Adeyanju, G.T., Ishola, O., 2014. *Salmonella* and *Escherichia coli* contamination of poultry meat from a processing plant and retail markets in Ibadan, Oyo State, Nigeria. *Springerplus.* 12(3), 139.
- Ahmed, M.S., Rahman, M.A., 2022. Antibiogram of *E. coli* and *Salmonella* spp. isolated from chicken meat and frozen milk in Barishal city, Bangladesh. *Bangl. J. Vet. Med.* 20(1), 49-58.
- Akond, M.A., Alam, S., Hassan, S.M.R., Shirin, M., 2009. Antibiotic resistance of *Escherichia coli* isolated from poultry and poultry environment of Bangladesh. *Internet J. food. Saf.* 11, 19-23.
- Ali, S., Alsayeqh, A.F., 2022. Review of major meat-borne zoonotic bacterial pathogens. *Front. Public Health.* 10, 1045599.
- Allocati, N., Masulli, M., Alexeyev, M.F., Di Ilio, C., 2023. *Escherichia coli* in Europe: an overview. *Int. J. Environ. Res. Public Health.* 10(12), 6235-6254.
- Anh, L.V.L., Khai, L.T.L., 2017. The prevalence and antimicrobial susceptibility of *Escherichia coli* on ducks in Can Tho city. *CTU J. Sci.* 50b, 51-58.
- Arbab, S., Ullah, H., Wang, W., Zhang, J., 2022. Antimicrobial drug resistance against *Escherichia coli* and its harmful effect on animal health. *Vet. Med. Sci.* 8, 1780-1786.
- Assoumy, M.A., Bedekelabou, A.P., Teko-Agbo, A., Ossebi, W., Akoda, K., Nimbona, F., Zeba, S.H., Zobo, A.A., Tiecoura, R.C.T., Kallo, V., Dagnogo, K., Bada-Alambédji, R., 2021. Antibiotic resistance of *Escherichia coli* and *Salmonella* spp. strains isolated from healthy poultry farms in the districts of Abidjan and Agnibilékrou (Côte d'Ivoire). *Vet. World.* 14(4), 1020-1027.
- Bauer, A.L., Kirby, W.M.M., Sherris, J.C., Turck, M., 1966. Antibiotic susceptibility testing by a standardized single disk method. *Am. J. Clin. Pathol.* 45(4), 493-496.
- Bhattarai, R.K., Basnet, H.B., Dhakal, I.P., Devkota, B., 2024. Antimicrobial resistance of avian pathogenic *Escherichia coli* isolated from broiler, layer, and breeder chickens. *Vet. World.* 17(2), 480-499.
- Brăţfelan, D.O., Tabaran, A., Colobatiu, L., Mihaiu, R., Mihaiu, M., 2023. Prevalence and antimicrobial resistance of *Escherichia coli* isolates from chicken meat in Romania. *Animals.* 13, 3488.
- Cappuccino, J., Welsh, C., 2017. *Microbiology: a laboratory manual*, 11th edition. Pearson Education, England.
- Clinical and Laboratory Standards Institute, 2021. Performance standards for antimicrobial susceptibility testing for bacteria isolated from aquatic animals. In: CLSI supplement VET04, (3rd edition). CLSI, Wayne, PA.
- Davis, G.S., Waits, K., Nordstrom, L., Grande, H., Weaver, B., Papp, K., Price, L.B., 2018. Antibiotic-resistant *Escherichia coli* from retail poultry meat with different antibiotic use claims. *BMC Microbiology.* 18, 174.
- Ekici, G., Dümen, E., 2019. *Escherichia coli* and food safety. In: Erjavec, M.S. (Ed.), *The universe of Escherichia coli*. Available online: <https://www.intechopen.com/chapters/65498>.

- Eltai, N.O., Yassine, H.M., El-Obeid, T., Al-Hadidi, S.H., Al Thani, A.A., Alali, W.Q., 2020. Prevalence of antibiotic-resistant *Escherichia coli* Isolates from local and imported retail chicken carcasses. *J. Food. Prot.* 83(12), 2200-2208.
- Gomes, T.A.T., Elias, W.P., Scaletsky, I.C.A., Guth, B.E.C., Rodrigues, J.F., Piazza, R.M.F., Ferreira, L.C.S., Martinez, M.B., 2016. Diarrheagenic *Escherichia coli*. *Braz. J. Microbiol.* 47, 3-30.
- Hang, T.T., Khoa, N.N., Ha, D.T., Duc D.H., Diep, N.T.M., 2020. Assessing the extent of infection with *Escherichia coli* and *Salmonella* spp. in pork meat in some key markets of Quy Nhon city, Binh Dinh province. *DTU J. Sci. Technol.* 5(1), 94-98.
- Hasib, F.M.Y., Magouras, I., St-Hilaire, S., Paudel, S., Kamali, M., Lugsomya, K., Lam, H.K., Elsohaby, I., Butaye, P., Nekouei, O., 2024. Prevalence and characterization of antimicrobial resistant *Escherichia coli* in chicken meat from wet markets in Hong Kong. *Front. Vet. Sci.* 11, 1340548.
- Heuer, H., Krsek, M., Baker, P., Smalla, K., Wellington, E.M., 1997. Analysis of actinomycete communities by specific amplification of genes encoding 16S rRNA and gel electrophoretic separation in denaturing gradients. *Appl. Environ. Microbiol.* 63(8), 3233-3241.
- Hleba, L., Petrová, J., Kántor, A., Cubon, J., Kacániová, M., 2015. Antibiotic resistance in Enterobacteriaceae strains isolated from chicken and milk samples. *J. Microbiol. Biotechnol. Food. Sci.* 4, 19.
- Hoben, H.J., Somasegaran, P., 1982. Comparison of the pour, spread, and drop plate methods for enumeration of *Rhizobium* spp. in inoculants made from presterilized peat. *Appl. Environ. Microbiol.* 44, 1246-1247.
- Horri, M., Gholami-Ahangaran, M., 2022. Phenotypic and genotypic characterization of antibiotic resistance in *Escherichia coli* strains isolated from broiler chickens with suspected colibacillosis in Isfahan Province. *Iran. Infect. epidemiol. microbiol.* 8(3), 193-201.
- Jain, P., Bepari, A.K., Sen, P.K., Rafe, T., Imtiaz, R., Hossain, M., Reza, H.M., 2021. High prevalence of multiple antibiotic resistance in clinical *E. Coli* isolates from Bangladesh and prediction of molecular resistance determinants using WGS of an XDR isolate. *Sci. Rep.* 11(1), 22859.
- Julqarnain, S.M., Bose, P., Rahman, Z.M., Khatun, M.M., Islam, M.A., 2022. Bacteriological quality and prevalence of foodborne bacteria in broiler meat sold at live bird markets at Mymensingh City in Bangladesh. *J. Adv. Vet. Anim. Res.* 9(3), 405-411.
- Kaper, J.B., 2005. Pathogenic *Escherichia coli*. *Int. J. Med. Microbiol.* 295(6-7), 355-356.
- Kim, S., Kim, H., Kim, Y., Kim, M., Kwak, H., Ryu, S., 2020. Antimicrobial resistance of *Escherichia coli* from retail poultry meats in Korea. *J. Food. Prot.* 83(10), 1673-1678.
- Krumperman, P.H., 1983. Multiple antibiotic resistance indexing of *Escherichia coli* to identify high-risk sources of fecal contamination of foods. *Appl. Environ. Microbiol.* 46(1), 165-170.
- Lee, K.Y., Lavelle, K., Huang, A., Atwill, E.R., Pitesky, M., Li, X., 2023. Assessment of prevalence and diversity of antimicrobial resistant *Escherichia coli* from retail meats in Southern California. *Antibiotics.* 12(4), 782.
- Lehel, J., Yaucat-Guendi, R., Darnay, L., Palotás, P., Laczay, P., 2020. Possible food safety hazards of ready-to-eat raw fish containing product (sushi, sashimi). *Crit. Rev. Food. Sci. Nutr.* 61(5), 867-888.
- Maesli, M., Lee, B., Miller, S., Reyna, Z., Lu, P., Yan, J., Ulhaq, A., Skandalis, N., Spellberg, B., Luna, B., 2020. Horizontal gene transfer of antibiotic resistance from *Acinetobacter baylyi* to *Escherichia coli* on lettuce and subsequent antibiotic resistance transmission to the gut microbiome. *mSphere.* 5(3), e00329-20.

- Manh, L.H., Dao, T.X., Dung, N.N.X., Minh, B.T.L., 2016. Survey of bacterial infected levels in poultry meat in slaughterhouse and retail markets at Ben Tre city. *CTU J. Sci.* 2016(2), 56-60.
- Marangoni, F., Corsello, G., Cricelli, C., Ferrara, N., Ghiselli, A., Lucchin, L., Poli, A., 2015. Role of poultry meat in a balanced diet aimed at maintaining health and wellbeing: an Italian consensus document. *Food Nutr. Res.* 59, 27606.
- Mortensen, A.M.S., Poulsen, S.J., Berbisá, M.Á.F., Djurhuus, A., 2024. Distribution of antibiotic resistant bacteria and genes in sewage and surrounding environment of Tórshavn, Faroe Islands. *Front. Environ. Sci.* 12, 1336318.
- Mota-Bravo, L., Camps, M., MuñozGutiérrez, I., Tatarenkov, A., Warner, C., Suarez, I., Cortés-Cortés, G., 2023. Detection of horizontal gene transfer mediated by natural conjugative plasmids in *E. coli*. *J. Vis. Exp.* 193, e64523.
- Mottet, A., Tempio, G., 2017. Global poultry production: current state and future outlook and challenges. *J. World. Poult. Sci.* 73(02), 245-256.
- Mpundu, P., Mbewe, A.R., Muma, J.B., Zgambo, J., Munyeme, M., 2019. Evaluation of bacterial contamination in dressed chickens in Lusaka Abattoirs. *Front. Public. Health.* 7, 19.
- Nataro, P.J., Kaper, B.J., 1998. Diarrheagenic *Escherichia coli*. *Clin. Microbiol. Rev.* 11(1), 142-201.
- Ngai, D.G., Nyamache, A.K., Ombori, O., 2021. Prevalence and antimicrobial resistance profiles of *Salmonella* species and *Escherichia coli* isolates from poultry feeds in Ruiru Sub-County, Kenya. *BMC Res. Notes.* 14(1), 41.
- Noenchat, P., Srithong, T., Lertpiriyasakulkit, S., Nhoonoi, C., Saksangawong, C., Somplang, P., 2024. Prevalence and antimicrobial resistance profile of *Escherichia coli* O157: H7 isolated from chickens and chicken meats from local poultry slaughterhouses in Nakhon Ratchasima Province, Thailand. *Vet. Integr. Sci.* 22(3), 769-785.
- Osman, K.M., Kappell, A.D., Elhadidy, M., ElMougy, F., El-Ghany, W.A.A., Orabi, A., Mubarak, A.S., Dawoud, T.M., Hemeg, H.A., Moussa, I.M.I., Hessain, A.M., Yousef, H.M.Y., 2018. Poultry hatcheries as potential reservoirs for antimicrobial-resistant *Escherichia coli*: a risk to public health and food safety. *Sci. Rep.* 8(1), 5859.
- Parvin, M.S., Talukder, S., Ali, M.Y., Chowdhury, E.H., Rahman, M.T., Islam, M.T., 2020. Antimicrobial resistance pattern of *Escherichia coli* isolated from frozen chicken meat in Bangladesh. *Pathogens.* 9(6), 420.
- Phiri, N., Mainda, G., Mukuma, M., Sinyangwe, N.N., Banda, L.J., Kwenda, G., Muligisa-Muonga, E., Flavien, B.N., Mwansa, M., Yamba, K., Munyeme, M., Muma, J.B., 2020. Antibiotic-resistant *Salmonella* species and *Escherichia coli* in broiler chickens from farms, abattoirs and open markets in selected districts of Zambia. *J. Epidemiol. Res.* 6(1), 13-21.
- Radwan, I., El-Halim, A., Abed, A.H., 2020. Molecular characterization of antimicrobial-resistant *Escherichia coli* isolated from broiler chickens. *J. Vet. Med. Res.* 27(2), 128-142.
- Rahman, M.M., Husna, A., Elshabrawy, H.A., Alam, J., Runa, N.Y., Badruzzaman, A.T.M., Ashour, H.M., 2020. Isolation and molecular characterization of multidrug-resistant *Escherichia coli* from chicken meat. *Sci. Rep.* 10(1), 21999.
- Robins-Browne, R.M., Holt, K.E., Ingle, D.J., Hocking, D.M., Yang, J., Tauschek, M., 2016. Are *Escherichia coli* pathotypes still relevant in the era of whole-genome sequencing? *Front. Cell. Infect. Microbiol.* 6, 141.
- Rouger, A., Tresse, O., Zagorec, M., 2017. Bacterial contaminants of poultry meat: sources, species, and dynamics. *Microorganisms.* 5(3), 50.
- Saitou, N., Nei, M., 1987. The neighbor-joining method: a new method for reconstructing phylogenetic trees. *Mol. Biol. Evol.* 4(4), 406-425.
- Sandhu, R., Dahiya, S., Sayal, P., 2016. Evaluation of multiple antibiotic resistance (MAR) index and doxycycline susceptibility of *Acinetobacter* species among inpatients. *Indian J. Microbiol. Res.* 3(3), 299-304.

- Tamura, K., Stecher, G., Peterson, D., Filipinski, A., Kumar, S., 2013. MEGA6: molecular evolutionary genetics analysis version 6.0. *Mol. Biol. Evol.* 30(12), 2725-2729.
- Thompson, J.D., Gibson, T.J., Plewniak, F., Jeanmougin, F., Higgins, D.G., 1997. The CLUSTAL\_X windows interface: Flexible strategies for multiple sequence alignment aided by quality analysis tools. *Nucleic. Acids. Res.* 25(24), 4876-4882.
- Thong, N.V., Thuan, N.K., Minh, L.B.T., 2025. Prevalence of antibiotic resistance genes of *Escherichia coli* at the pig slaughterhouses in the Mekong Delta. *Vet. Integr. Sci.* 23(1), 1-10.
- Trieu, T.Q., Huong, D.T.T., Hue, N.T.K., Thu, T.T., 2022. Survey on infection and antibiotic resistance rates of *Escherichia coli* in pork and chicken meat at some markets in Buon Ma Thuot city in 2021. *Vietnam J. Food. Control.* 5(3), 279-290.
- Vounba, P., Arsenault, J., Bada-Alambe'dji, R., Fairbrother, J.M., 2019. Prevalence of antimicrobial resistance and potential pathogenicity, and possible spread of third generation cephalosporin resistance, in *Escherichia coli* isolated from healthy chicken farms in the region of Dakar, Senegal. *PLoS ONE.* 14(3), e0214304.
- Vui, D.T., Tiep, N.B., 2016. Isolation and characterization of *Escherichia coli* in of Bau and Dom Ducks at Dai Xuyen Duck Breeding and Research Center. *Vietnam J. Agri. Sci.* 14(12), 1894-1902.
- Worku, W., Desta, M., Menjetta, T., 2022. High prevalence and antimicrobial susceptibility pattern of salmonella species and extended-spectrum  $\beta$ -lactamase producing *Escherichia coli* from raw cattle meat at butcher houses in Hawassa city, Sidama regional state, Ethiopia. *PLoS One.* 17(1), e0262308.
- World Health Organization, 2022. Food safety fact sheet. Available online: <https://www.who.int/news-room/fact-sheets/detail/food-safety> (Accessed on April 28, 2024).

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