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Research article

Prevalence of antibiotic resistance genes and genetic relationship of *Escherichia coli* serotype O45, O113, O121, and O157 isolated from cattle in the Mekong Delta, Vietnam

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Abstract

A total of 39 *Escherichia coli* strains serotype O45, O113, O121, and O157 isolated from cattle in the Mekong Delta were examined the antimicrobial susceptibility to 13 antibiotics by the disc-diffusion method. Those strains were also analyzed for the presence of antibiotic resistance genes by PCR assay, and their genetic relationship by ERIC-PCR assay. The results of antimicrobial susceptibility testing showed that those strains were sensitive to most of the examined antibiotics, but were relatively high resistance to ampicillin (64.10%), and colistin (53.85%). Those *E. coli* strains could be resistant against one to eight antibiotics with 22 resistance patterns obtained. Moreover, those *E. coli* strains harbored one to seven antibiotic resistance genes. Gene *tetA* (51.28%) and $bla_{amp}C$ (48.72%) were detected frequently while gene *tetB*, bla_{CMY}, and cat1 were not found in those *E. coli* strains. A total of 21 combined patterns of antibiotic resistance genes were recorded, and the most frequent combined pattern was $bla_{amp}C$+*tetA* (12.82%). ERIC-PCR analysis revealed that each *E. coli* serotype exhibited various genetic patterns with 40%-100% of similarity. The most elevated number of patterns were in *E. coli* O157 (nine patterns), followed by *E. coli* O121 (six patterns). The prevalence of antibiotic resistance genes and diverse genetic characteristics in those *E. coli* strains originated from cattle constitute potential risks to local health in the Mekong Delta.

Keywords: Antibiotic susceptibility, Cattle, *E. coli*, Genes, Genetic relationship

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INTRODUCTION

Shiga toxin-producing *E. coli* (STEC) can cause severe foodborne diseases in humans worldwide. STEC includes various virulent serotypes such as O26, O38, O45, O103, O104, O111, O117, O121, and O157 (Erickson and Doyle, 2007; DebRoy et al., 2011; Lin et al., 2011). STEC is typically transmitted through the fecal-oral route and naturally present in the gastrointestinal tract of ruminants, especially in cattle (Armstrong et al., 1996).

Antibiotic resistance of bacteria has become a major threat to human and animal health. Evidence has showed the spread of resistance genes among bacteria (Carattoli, 2009). Many reports showed the use of antibiotics in animal husbandry is related to the antibiotic-resistant strains isolated in humans (Sorum and L'Abee-Lund, 2002). The gradual increase of antibiotic resistance in *E. coli* has been recorded. There was seen genetic relation between resistance strains isolated from humans and animals, especially among the ESBLs (Extended Spectrum Beta-Lactamase) (Kluytmans et al., 2013; Valentin et al., 2014). Importantly, it has been reported that *E. coli* could harbor multiple antibiotic resistance genes such as fluoroquinolone (qnr), trimethoprim (*dfrA1*), cephalothin (*blaSHV*), tetracycline (tetA, *tetB*), ampicillin (*CITM*), gentamicin (*aac(3)-IV*), sulfonamide (*sulI*, *sulII*), chloramphenicol (*cat1*, *cmlA*), aminoglycosides (*aadA1*), and erythromycin (*ereA*) (Momtaz et al., 2013; Bai et al., 2016; Jaja et al., 2020).

Therefore, the prevalence of antibiotic resistance genes and genetic relationship in *E. coli* isolated from animals was a concerning issue for human public health. However, the in-depth studies on this issue were limited in the Mekong Delta, Vietnam. This study was conducted to determine the antibiotic resistance and genetic relationship of *E. coli* originating from cattle to contribute to the antibiotic using management and safety of public health in this region.

MATERIALS AND METHODS

Bacterial strains

A total of 39 *E. coli* O45 (n = 6), O113 (n = 7), O121 (n = 9), O157 (n=17) isolated from cattle in the Mekong Delta, Vietnam were used in this study. The strains were recovered from healthy cattle from February to December 2021 and kept in the specialized Veterinary Medicine laboratory of the Veterinary Medicine Department, College of Agriculture, Can Tho University, Vietnam.

Antimicrobial susceptibility testing

The *E. coli* strains were examined for their antimicrobial susceptibility following the guidelines of the Kirby-Bauer disk diffusion test (Bauer et al., 1966). The results of the antibiotic resistance zone were determined following the standard of Clinical Laboratory Standards Institute procedure M02-M07 (CLSI, 2020). *Escherichia coli* ATCC 25922 and *E. coli* ATCC 35218 were used as controls.

The selection of antibiotics for testing was based on the results of studies on antibiotics used in domestic animals in the Mekong Delta previously. The antibiotic discs were supplied by Nam Khoa Biotek Ltd. (Vietnam)

including ***ampicillin*** (***Am***, 10 µg), amoxicillin/clavulanic acid (Ac, 20/10 µg), cefuroxime (Cu, 30 µg), ceftazidime (Cz, 30 µg), colistin (Co, 10 µg), gentamycin (Ge, 10 µg), amikacin (Ak, 30 µg), streptomycin (Sm, 10 µg), tetracycline (Te, 30 µg), doxycycline (Dx, 30 µg), chloramphenicol (Cl, 30 µg), levofloxacin (Lv, 5 µg), ofloxacin (Of, 5 µg).

Detection of antibiotic resistance genes by PCR method

DNA of *E. coli* strains were extracted by the heat shocking method following the guideline of Soumet et al. (1994). A total of 13 resistance genes against different groups of antibiotics were examined including aminoglycoside (*aadA1*, *strA*), beta-lactam (*blaTEM, blaCTX-M, bla$_{amp}$C, blaCMY, blaSHV*), chloramphenicol (*cat1*), diaminopyrimidines (dfrA), quinolone (*qnrA*), sulfonamide (*sulII*), tetracycline (tetA, *tetB*).

The PCR mixture contained Master Mix 2X (Promega, USA) (12.5 µL); forward and reverse primers (IDT, USA) at 10 µM (0.5 µL/primer); distilled water (9.5 µL), and DNA template (2.0 µL). The primer sequences and thermocycling PCR reactions were chosen from the previously published literature (Forward et al., 2001; Carattoli et al., 2002; Randall et al., 2004; Weill et al., 2004; Toro et al., 2005; Cattoir et al., 2007; Jouini et al., 2007; Van et al., 2008). In this study, five *E. coli* strains, which were previously isolated from animals in the Mekong Delta and harbored those antibiotic genes, were used as the positive controls. Those strains were kept in the Veterinary Medicine Department, College of Agriculture, Can Tho University, Vietnam.

ERIC-PCR method

ERIC-PCR method was used to investigate genetic relationship among the *E. coli* strains in this study. The reaction and primer sequences were described by Ranjbar et al. (2017). The nucleotide sequence of primers were used as follows: 5'-ATGTAAGCTCCTGGGGATTCAC-3' (forward primer) and 5'-AAGTAAGTGACTGGGGTGAGCG-3' (reverse primer). The picture of electrophoresis gel was inserted into BioNumerics 7.0 software (Applied Math, Belgium) to determine the genetic relationship of those *E. coli* strains via using UPGMA analysis (Unweighted Pair Group Method with Arithmetic Mean).

Statistical analysis

Data were collected and expressed as percentages using Microsoft Excel (Microsoft, USA). Statistical analysis was calculated by using the Statistical Package (the Social Sciences statistical package), version 7.1 (IBM, USA). The statistical significance level was chosen at $P<0.05$.

RESULTS

Antimicrobial susceptibility

The results showed that the *E. coli* strains were susceptible to 9/13 examined antibiotics (82.05-100%), especially to doxycycline (100%), and amikacin (97.44%). However, those strains exhibited resistance to tetracycline (30.77%), streptomycin (33.33%), colistin (53.85%), and ampicillin (64.10%) relatively. The resistance rates against antibiotics of other *E. coli* serotypes were higher than *E. coli* O157 strains were (P<0.05) (Table 1).

Table 1 Antibiotic susceptibility of the *E. coli* strains isolated from cattle in the Mekong Delta.

Antibiotics	*E. coli* serotypes								Total (n = 39)	
	O45 (n = 6)		O113 (n = 7)		O121 (n = 9)		O157* (n = 17)			
	S (%)	R (%)	S (%)	R (%)	S (%)	R (%)	S (%)	R (%)	S (%)	R (%)
Amikacin	6 (100.0)	0 (0.00)	7 (100.0)	0 (0.00)	8 (88.89)	1 (11.11)	17 (100.0)	0 (0.00)	38 (97.44)	1 (2.56)
Ampicillin	3 (50.00)	3 (50.00)	2 (28.57)	5 (71.43)	3 (33.33)	6 (66.67)	6 (35.29)	11 (64.71)	14 (35.09)	25 (64.10)
Amoxicillin/ clavulanic acid	5 (83.33)	1 (16.67)	5 (71.43)	2 (28.57)	8 (88.89)	1 (11.11)	14 (82.35)	3 (17.65)	32 (82.05)	7 (17.95)
Ceftazidime	3 (50.00)	3 (50.00)	5 (71.43)	2 (28.57)	7 (77.78)	2 (22.22)	17 (100.0)	0 (0.00)	32 (82.05)	7 (17.95)
Cefuroxime	5 (83.33)	1 (16.67)	5 (71.43)	2 (28.57)	6 (66.67)	3 (33.33)	16 (94.12)	1 (5.88)	32 (82.05)	7 (17.95)
Chloramphenicol	4 (66.67)	2 (33.33)	5 (71.43)	2 (28.57)	6 (66.67)	3 (33.33)	17 (100.0)	0 (0.00)	33 (84.62)	6 (15.38)
Colistin	3 (50.00)	3 (50.00)	2 (28.57)	5 (71.43)	1 (11.11)	8 (88.89)	12 (70.59)	5 (29.41)	18 (46.15)	21 (53.85)
Doxycycline	6 (100.0)	0 (0.00)	7 (100.0)	0 (0.00)	9 (100.0)	0 (0.00)	17 (100.0)	0 (0.00)	39 (100.0)	0 (0.00)
Gentamycin	5 (83.33)	1 (16.67)	6 (85.71)	1 (14.29)	9 (100.0)	0 (0.00)	17 (100.0)	0 (0.00)	37 (94.87)	2 (5.13)
Levofloxacin	5 (83.33)	1 (16.67)	7 (100.0)	0 (0.00)	8 (88.89)	1 (11.11)	17 (100.0)	0 (0.00)	37 (94.87)	2 (5.13)
Ofloxacin	5 (83.33)	1 (16.67)	7 (100.0)	0 (0.00)	8 (88.89)	1 (11.11)	17 (100.0)	0 (0.00)	37 (94.87)	2 (5.13)
Streptomycin	3 (50.00)	3 (50.00)	3 (42.86)	4 (57.14)	6 (66.67)	3 (33.33)	14 (82.35)	3 (17.65)	26 (66.67)	13 (33.33)
Tetracycline	5 (83.33)	1 (16.67)	3 (42.86)	4 (57.14)	4 (44.44)	5 (55.56)	15 (88.24)	2 (11.76)	27 (69.23)	12 (30.77)

S: No. of sensitive strains; R: No. of resistant strains
**The antibiotic resistance: E. coli O45, O113, O1121 > E. coli O157 (P<0.05)*

Of 39 *E. coli* strains examined, thirty-six strains (92.31%) were resistant from one to eight antibiotics examined, and twenty-two antibiotic resistance patterns were obtained. Multiple antibiotic resistance was detected, and ampicillin (Am) was present in most antibiotic resistance patterns (Table 2).

Table 2 Antibiotic resistance patterns of the studied strains (n = 39).

No. of antibiotics	Antibiotic resistance pattern	No. of patterns	No. of strains	Percentage (%)
1	Am	5	6	15.38
	Co		3	7.69
	Cz		1	2.56
	Sm		2	5.13
	Te		1	2.56
2	Am+Co	3	2	5.13
	Cz+Co		2	5.13
	Sm+Te		2	5.13
3	Am+Ac+Co	2	2	5.13
	Am+Cu+Co		2	5.13
4	Am+Cu+Sm+Te	4	1	2.56
	Am+Ac+Cu+Co		1	2.56
	Am+Ac+Sm+Te		1	2.56
	Am+Co+Te+Cl		2	5.13
5	Am+Ac+Co+Ge+Sm	2	1	2.56
	Am+Cz+Co+Sm+Cl		1	2.56
6	Am+Cu+Co+Sm+Te+Cl	4	1	2.56
	Am+Cz+Co+Sm+Te+Cl		1	2.56
	Am+Cz+Cu+Co+Sm+Cl		1	2.56
	Am+Cz+Cu+Co+Sm+Te		1	2.56
7	Am+Ac+Co+Ak+Te+Lv+Of	1	1	2.56
8	Am+Ac+Ge+Sm+Te+Cl+Lv+Of	1	1	2.56
	Total	22	36	92.31

Ac: amoxicillin/clavulanic acid; Am: ampicillin; Cz: ceftazidime; Cu: cefuroxime; Co: colistin; Ge:gentamicin; Ak: amikacin; Te: tetracycline; Cl: chloramphenicol; Lv: levofloxacin; Dx: doxycycline; Sm: streptomycin; Of: ofloxacin

Prevalence of antibiotic resistance genes

Of 13 examined genes, gene *tetA* was detected at the highest rate (51.28%), followed by bla$_{amp}$C (48.72%), *sulΠ* (28.21%), and *dfrA* (25.64%). Resistance genes *blaCMY*, *tetB*, and *cat1* were not detected. The high prevalence of beta-lactamase genes (10.26-48.72%) seemed to reflect the results of the antimicrobial susceptibility test that ampicillin, ceftazidime, and cefuroxime were highly resistant. blaCTX-M was detected more in *E. coli* O157 strains than in the other serotypes ($P< 0.05$). In other antibiotic groups, the presence of antibiotic resistance genes were determined at a relatively high rate (17.95-28.21%) (Table 3).

Table 3 Prevalence of antibiotic resistance genes.

Antibiotic resistance gene	No. of positive strains (%)				Total No. of posive strains (%) (n = 39)
	E. coli O45 (n = 6)	E. coli O113 (n = 7)	E. coli O121 (n = 9)	E. coli O157 (n =17)	
blaTEM	1 (16.67)	1 (14.29)	2 (22.22)	5 (29.41)	9 (23.08)
*blaCTX-M**	1 (16.67)	0 (0.00)	0 (0.00)	8 (47.06)	9 (23.08)
blaampC	4 (66.67)	2 (28.57)	7 (77.78)	6 (35.29)	19 (48.72)
blaCMY	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
blaSHV	0 (0.00)	0 (0.00)	1 (11.11)	3 (17.65)	4 (10.26)
aadA1	2 (33.33)	1 (14.29)	1 (11.11)	3 (17.65)	7 (17.95)
strA	1 (16.67)	1 (14.29)	3 (33.33)	4 (23.53)	9 (23.08)
cat1	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
tetA	4 (66.67)	5 (71.43)	4 (44.44)	7 (41.18)	20 (51.28)
tetB	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
qnrA	1 (16.67)	1 (14.29)	2 (22.22)	3 (17.65)	7 (17.95)
sulII	2 (33.33)	2 (28.57)	2 (22.22)	5 (29.41)	11 (28.21)
dfrA	2 (33.33)	1 (14.29)	3 (33.33)	4 (23.53)	10 (25.64)

**The prevalence of blaCTX-M in E. coli O157 > E. coli O45, O113, O1121 (P<0.05)*

Most *E. coli* strains (94.87%) harbored from one to seven antibiotic resistance genes, and 21 combined patterns of antibiotic resistance genes were obtained. The most frequent pattern was blaampC+tetA (12.82%) (Table 4).

Table 4 Genotypic antibiotic resistance patterns of the *E. coli* strains (n = 39).

No. of gene	Gene patterns	No. of patterns	No. of strains	Percentage (%)
1	*blaampC*	4	2	5.13
	blaCTX-M		4	10.26
	sulII		3	7.69
	tetA		3	7.69
2	*blaampC+dfrA*	5	2	5.13
	blaampC+tetA		5	12.82
	blaSHV+qrnA		1	2.56
	blaTEM+dfrA		1	2.56
	blaTEM+tetA		1	2.56
3	*blaampC+blaCTX-M+tetA*	3	1	2.56
	tetA+aadA1+qnrA		1	2.56
	tetA+sulII+strA		1	2.56
4	*blaampC+tetA+sulII+dfrA*	2	1	2.56
	blaampC+tetA+sullII+strA		1	2.56
5	*blaampC+tetA+qnrA+sulII+dfrA*	4	1	2.56
	blaampC+tetA+aadA1+qnrA+dfrA		1	2.56
	blaTEM+blaampC+qnrA+strA+dfrA		1	2.56
	blaTEM+blaampC+qnrA+sulII+dfrA		1	2.56
6	*blaTEM+blaampC+aadA1+sulII+strA+dfrA*	2	2	5.13
	blaTEM+blaSHV+blaCTX-M+tetA+aadA1+strA		3	7.69
7	*blaampC+blaCTX-M+blaOXA+tetA+qnrA+sulII+dfrA*	1	1	2.56
	Total	21	37	94.87

Genetic relationship of the studied E. coli strains

ERIC-PCR analyses revealed that *E. coli* O157 strains (n=17) belonged to three groups with a similarity of over 50%. G1 and G2 patterns and G8 and G9 patterns showed the highest homogenous rates (83.5% and 87%, respectively) (Figure 1a).

Nine *E. coli* O121 strains were divided into three groups with a low similarity of 40-50%. The H1 and H2 patterns exhibited the highest similarity of 80% while the two strains of the H6 pattern were 100% homologous (Figure 1b). *E. coli* O113 strains (n=7) displayed a high difference in the genetic relationship with 40-72.5% similarity. The P2 pattern was the most popular (4/7 strains) (Figure 1c). A significant homogeneity among the *E. coli* O45 (n=6) was showed (Figure 1d). In general, the results indicated a relatively high diversity in genetic characteristics of *E. coli* strains isolated from cattle in the Mekong Delta.

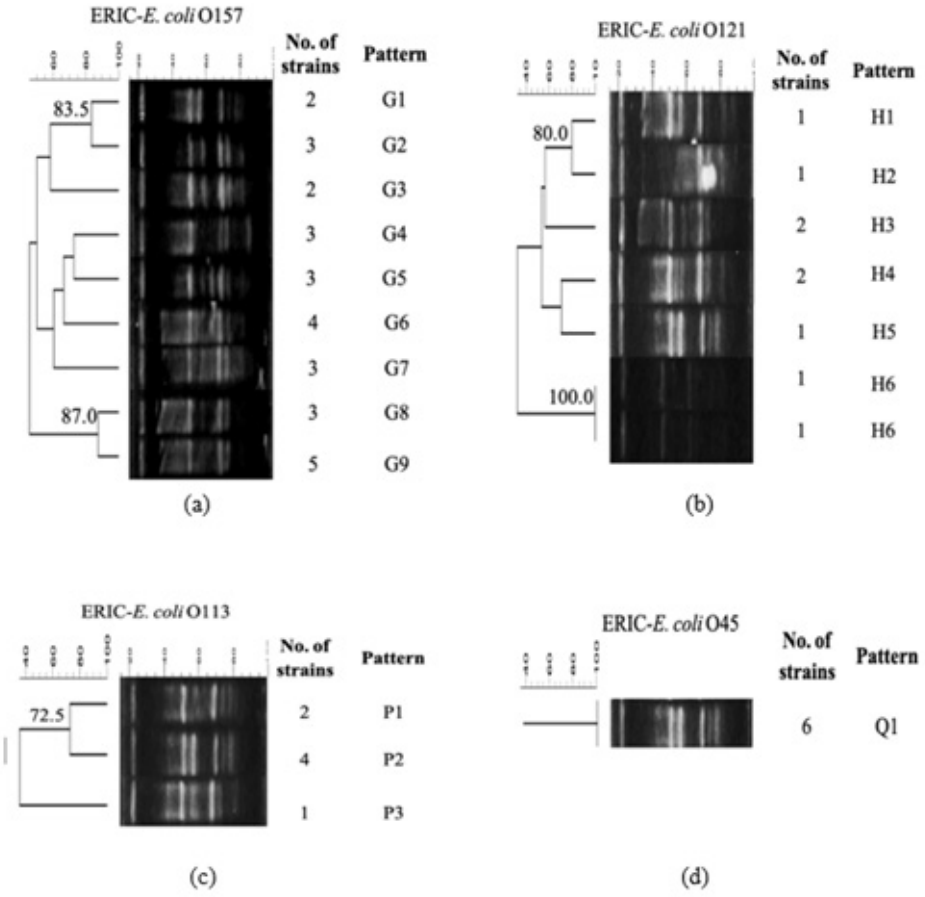

Figure 1 The photographs of the transverse sections of the semitendinosus muscle stained using mATPase from representative newborn piglets after pre-incubation in an alkaline solution at pH 10.4 (A) and pre-incubation in an acidic solution at pH 4.6 (B); magnification: 200x. The yellow and red arrows indicate the primary (P) and secondary (S) fibers, respectively.

DISCUSSION

It was demonstrated that the studied strains of *E. coli* were susceptible to the majority of examined antibiotics. However, the result showed high resistance among the strains to colistin (53.85%), and ampicillin (64.10%), which have been used in animal production in the area for a long time. According to da Costa et al. (2007), long-term use of antibiotics could produce the resistance of bacteria to antibiotics. In a report by Iweriebor et al. (2015), 95 *E. coli* O157 strains isolated from dairy cattle in South Africa exhibited a high resistance to chloramphenicol (89.5%), ampicillin (94.74%), tetracycline (96.84%), oxytetracycline (94.74%), cefuroxime (82%), ceftazidime (32%), streptomycin (94.74%), amoxicillin/clavulanate (84.2%), norfloxacin (10.5%), ciprofloxacin (12.6%), and gentamicin (8.4%). A study in China revealed that *E. coli* strains isolated from dairy cattle were resistant to ampicillin (88.89%), amoxicillin/clavulanic acid (75.00%), chloramphenicol (52.78%), ciprofloxacin (44.44%), gentamicin (72.22%), acid nalidixic (80.56%), tetracycline (83.33%), and trimethoprim/sulfamethoxazole (75%) (Ali et al., 2016).

Otherwise, *E. coli* strains serotype O45, O113, and O121 showed the antibiotic-resistance rates higher than *E. coli* O157 strains did in this study (Table 1). Kennedy et al. (2017) revealed that acquiring antibiotic resistance could provide a competitive advantage, allowing *E. coli* non-O157 strains to enter livestock preferentially than other pathogenic strains do when the antibiotic selection pressure in treatment was present. Buvens et al. (2010) reported that the antibiotic resistance of *E. coli* O26 was higher than *E. coli* O157 such as ampicillin (23.5% vs 5.2%), streptomycin (58% vs 26%), tetracycline (44% vs 15%), sulfonamides (59% vs 22%), and trimethoprim (24% vs 4%) respectively. Cabal et al. (2016) also reported *E. coli* O111, O104, O91, and O26 strains isolated from cattle in Spain were resistant to antibiotics more than *E. coli* O157 was.

In most resistance patterns, ampicillin (Am) was present the most frequent in this study (Table 2). Previous reports in the Mekong Delta recorded that *E. coli* isolated from farm animals demonstrated high resistance to ampicillin, and this antibiotic was usually detected in the resistance patterns (Ly et al., 2009; Bui et al., 2018). In Nigeria, Olatoye (2010) indicated that the misuse of antibiotics in beef cattle could cause an increase of multiple resistance antibiotics of *E. coli*, especially EHEC O157:H7. Thus, the management of using antibiotics should be paid an attention to prevent high prevalence of resistance and spread of *E. coli* strains, which were multiple antibiotic resistances, in husbandry environments as well as human living environments. Of the 13 antibiotic resistance genes examined, tetA was detected at the highest rate (51.28%) (Table 3). However, the results of the antimicrobial susceptibility test showed that those *E. coli* strains were still relatively sensitive to tetracycline (69.23%) and doxycycline (100%) (Table 1). It could be that the tetracycline resistance gene had become a part of the normal genome of *E. coli*, and the absence of a resistance gene in the same group, such as *tetA* and *tetB*, might be due to incompatibility of the gene-coding plasmids (Maynard et al., 2003). Besides, although *E. coli* strains carried antibiotic resistance genes, the expression of resistance could depend on environmental conditions. The diverse presence of antibiotic resistance genes in bacterial populations was not

eliminated even in the absence of using or no resistance expression to antibiotics (Bengtsson-Palme, 2018). Moreover, bacteria could accumulate the antibiotic resistance genes which encrypted on bacterial chromosomes or plasmids (Yamamoto et al., 2014). Besides, mobile genetic elemens and plasmids could play an integral role in antibiotic resistance in *E. coli* (Johnson et al., 2007; Kadlec and Schwarz, 2008; Carattoli, 2009; Frye and Jackson, 2013). The conjugation experiments of Sunde and Norström (2006) showed that 38% of 241 resistant *E. coli* strains were able to transfer resistance genes to susceptible recipient strains. In most cases, multi-resistant strains can transfer all their multi-resistant features to the recipient. Navajas-Benito et al. (2017) examined the molecular characteristics of antibiotic-resistant *E. coli* strains from dairy farms in Spain. The results showed that tetA gene was detected at higher rates than other genes were. Hypotheses have been proposed to explain the difference in the distribution of antibiotic resistance genes in bacteria from various hosts. It included differences in antibiotic use, the nature of certain pathogenic *E. coli* strains, and the epidemiological and ecological relationships of *E. coli* among animal species (Boerlin et al., 2005). Anuradha et al. (2014) investigated the prevalence of antibiotic resistance genes of EHEC strains, which were isolated from cattle manure, fresh meat, and barn floor in India showed that gene *tetB*, strA, and strB were commonly detected. Montso et al. (2019) survey on ESBL-producing *E. coli* strains from cattle manure and raw beef samples in South Africa indicated that genes *blaTEM*, *blaSHV*, and *blaCTX-M* were detected at 85.5%, 69.6%, and 58% respectively.

Among combined antibiotic resistance patterns, the pattern of $bla_{amp}C$+*tetA* was the most prevalent (12.82%) (Table 4). A bacterial strain could carry genes encoding either CTX-M, TEM and SHV or CTX-M and ampC simultaneously, which could alter the multidrug resistance phenotype (Bradford, 2001; Escobar-Páramo et al., 2006). In an investigation of antibiotic resistance of *E. coli* isolated in dairy cows in Bangladesh (Sobur et al., 2019), antibiotic resistance genotypes were present by the combination of genes *ereA*, *tetA*, *tetB*, bla_{SHV}. Algammal et al. (2020) found that 25% of *E. coli* strains isolated from cows in Egypt had the *blaTEM*+*blaCTX* genotype as the most common.

In analyzing the genetic relation, according to Awawdeh (2018), bacterial strains with ERIC-PCR homology ≥ 80% would be considered to have close genetic characteristics. Therefore, the results demonstrated that most of the *E. coli* strains in the present study did not have close genetic relation, and there was found diversity among each serotype. Nine patterns obtained from 17 strains would indicate that *E. coli* O157 strains could have been circulated in the Mekong Delta for a long time and thus harbored diverse genetic characteristics. The reason might be related to the origin of cattle raised in this area, leading to the presence of diverse strains with distinctive genetic characteristics. In previous reports, Worley et al. (2017) recorded genetic diversity of *E. coli* O157 circulating in different herds in California, USA. It was said that this was possibly due to the long-standing circulation of these strains in each region and season. Geue et al. (2009) identified the various genetic diversity of EHEC O26:H11 due to the different viability in host populations, the competition among bacteria belonging to divergent groups in the bovine gut, or potential interactions between EHEC and the host.

Other serotypes, for example *E. coli* O113 and O121, presented variable patterns of 40-70% similarity (Figure. 1b, 1c). Otherwhile, ERIC-PCR of the *E. coli* O45 revealed only one pattern (Figure. 1d). One reason that could affect the results of genetic diversity of the studied strains was the limited number of the strains. Therefore, further research with more isolates should be conducted to clarify the comprehensive genetic relationship of those *E. coli* strains in the Mekong Delta. In addition, various studies have recorded similarities in genetic characteristics of *E. coli* strains originating from cattle, environment, and human patients such as the studies of Badouei et al. (2015), Cobbold and Desmarchelier (2001) in Australia, Habets et al. (2021) in Belgium, Tazari et al. (2021) in Japan. Thus, the prevalence of those *E. coli* serotypes in cattle could be a risk for public health in the Mekong Delta.

CONCLUSIONS

Although the *E. coli* strains isolated from cattle in the Mekong Delta were highly sensitive to several antibiotics, those strains exhibited resistance against ampicillin and colistin, and produced multi-resistant patterns. Gene *tetA* and $bla_{amp}C$ were frequently detected from those *E. coli* strains. On the other hand, *E. coli* strains in each serotype (O45, O113, O121, O157) showed diverse genetic patterns. It indicated that those *E. coli* strains could have penetrated and circulated in cattle populations for a long time in the Mekong Delta. Thus, controlling the prevalence of those antibiotic-resistance *E. coli* strains is essential to prevent transmission between animals and humans.

AUTHOR CONTRIBUTIONS

This work was conducted with contribution of all authors. NKT, NTL and NTPC designed the experimental procedures. NKT, NTL, NTPC and NPK performed the experiments. NKT, NTL, NTPC, NPK, LTLK and TNB interpreted the data and prepared the manuscript. All authors read and approved the final manuscript

CONFLICT OF INTEREST

We have no conflict of interest.

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