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Research article

Occurrence of extended-spectrum beta-lactamase producing *E. coli* in broiler farm workers and the farm environment in Chiang Mai-Lamphun, Thailand

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Abstract

Antimicrobial resistance is a major global public health threat. For example, extended-spectrum beta-lactamase (ESBL) producing *Escherichia coli*'s emergence has resulted in treatment failure and increase mortality. The following study was conducted to determine the occurrence of ESBL producing *E. coli* in broiler farm workers and the broiler farm environment in Chiang Mai-Lamphun, Thailand. Twenty-nine (60.4%) of 48 broiler farms had evidence of ESBL producing *E. coli*. ESBL producing *E. coli* was recovered from 43.8% of boot swabs, 12.5% of feed and 2.1% of water. Fifteen (55.7%) of 27 farm workers had ESBL producing *E. coli* recovered from rectal swab samples. All isolates were susceptible to imipenem but resistant to ampicillin. The ESBL producing *E. coli* isolate were highly resistant to streptomycin (94.3%), gentamicin (86.8%), tetracycline (77.4%), chloramphenicol (66.0%), nalidixic acid (58.5%), and sulfamethoxazole/trimethoprim (56.6%). A large percentage (96.2%) of isolates was classified as multidrug resistance (MDR). Thirty-five antimicrobial resistance profiles were identified with AMP-GEN-SXT-NAL-TET-CHL-STR, AMP-GEN-SXT-TET-CHL-STR (14.3%) as the 2 most prevalent antimicrobial resistant profiles. Common resistance profiles were observed between farm workers and farm environment. These findings suggest possible transmission between poultry flock and humans on broiler farms, likely from contact with birds or their environment. It is important to increase awareness of hygiene practices on broiler farms and control antimicrobial usage to limit the emergence and spread of ESBL producing *E. coli*.

Keywords: Antimicrobial resistance, Broiler, *E. coli*, ESBL, Farm worker

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INTRODUCTION

Antimicrobial-resistant infections in humans and food animals is a global public health problem (Pormohammad et al., 2019). Estimates that antimicrobial resistance will cause 10 million deaths annually in 2050 (O'Neill, 2014). The inappropriate use of antibiotics in human and animal health is likely the major leading cause for the increase of antimicrobial resistance bacteria (Falgenhauer et al., 2019). Among the antimicrobial resistance pathogens, *Escherichia coli* is the predominant normal flora in the intestinal tract of humans and animals and can cause opportunistic infections (Costa et al., 2013). *E. coli* can transfer resistance determinants and induce antimicrobial resistance in other bacteria through horizontal gene transfer. Extended-spectrum beta-lactamase (ESBL) producing *E. coli* is an important pathogen and increasing in prevalence worldwide (Dandachi et al., 2018). ESBL producing pathogens are resistant to numerous antimicrobials and are associated with longer hospital stays and other negative clinical outcomes in humans (Lautenbach et al., 2001). These bacteria produce enzymes that confer resistance to most beta-lactam antibiotics, including 3rd and 4th generation cephalosporins (Le et al., 2015; Gundran et al., 2019).

A recent study reported high prevalence of ESBL-producing *E. coli* in samples from healthy adults, healthy food producing animals, and in water samples collected from selected areas in Thailand (Boonyasiri et al., 2014). Food-producing animals, including poultry, may serve as a reservoir for transmission of ESBL producing *E. coli* to humans. The bacteria have potential to be transmitted to humans via direct contact with colonized animals, consumption of contaminated meat products and contact with contaminated environments (Falgenhauer et al., 2019). Limited work has been done to identify ESBL organisms from broiler farms. This is important to characterize to better inform evidence-based policies to mitigate antimicrobial resistance. Chiang Mai and Lamphun provinces were considered as a major broiler production sites of northern Thailand since the two provinces are very close to each other, similar geographical characters and sharing the same source of materials for broiler production such as feed and day-old-chicks. The objective of this study was to determine the occurrence and antimicrobial resistance profiles of ESBL producing *E. coli* from broiler farm workers and the farm environment in Chiang Mai-Lamphun, Thailand

MATERIALS and METHODS

Sample collection

A total of 171 samples were collected from 48 broiler farms in Chiang Mai and Lamphun provinces from March-December 2016. One sample from each sample types were collected including boot swab (n=48), rectal swabs from farm workers (n=27), feed (n=48) and water (n=48). Farm visit for sample collection were carried out within 7 day before sending birds to slaughterhouse. Broiler fecal samples were collected using a sterile boot swab, by walking through out the raising area. Farm worker's rectal swab samples were self-collected by the willing farm workers following the guid-

ance and instructions provided. Pooled feed and water samples were collected from different spots inside the barn to obtained 500 g and 500 ml, respectively. Samples were kept under 4 ºC and transported to the laboratory within 5 hours of collection.

Ethical statement

Ethical approval for collecting human samples was approved by the ethics committee, Faculty of Medicine, Chiang Mai University (Code: NONE-2558-03215).

Bacterial isolation and identification

Boot swab samples and 25 g of feed sample were mixed with 225 ml of Luria-Bertani Broth (Difco®, USA) while 50 ml water sample were mixed with 50 ml Luria-Bertani Broth (Difco®, USA). Sample were incubated at 37°C for 18-24 hours before plated onto MacConkey agar (Merck®, Germany) with 1 mg/L cefotaxime (MAC-CTX) and incubated at 37°C for 18-24 hours. *E. coli* ATCC 25922 were used as negative control ESBL. Two suspected colonies from each sample were subcultured on nutrient agar (Merck®, Germany) for further confirmation of *E. coli* species using Matrix-Assisted Laser Desorption/ Ionization Time-of-Flight Mass Spectrometry (MALDI-TOF MS) (Biotyper®, Bruker Daltonik GmbH, Germany).

ESBL phenotyping

Combination disk test were used for confirmation of ESBL producing *E. coli* following Clinical and Laboratory Standards Institute guidelines (CLSI, 2014). Briefly, 0.5 McFarland bacterial suspension of suspected ESBL producing *E. coli* were spread onto Mueller Hinton agar (Merck®, Germany) using sterilized cotton swab. Then, antibiotic disk containing cefotaxime (30 µg) and ceftazidime (30 µg) with and without clavulanic acid (10 µg) were placed on the agar before incubated at 37°C for 16-18 hours. The inhibition zone diameter of each disk were measured, if the differences of inhibition zone diameter of the disk with clavulanate was larger than that of the disk without clavulanate at least by 5 mm, the suspected colony was interpreted as an ESBL producing *E. coli*. *E. coli* ATCC 25922 and *K. pneumoniae* ATCC 700603 were used for quality control.

Antimicrobial susceptibility testing

Antimicrobial susceptibility against 12 antimicrobial agents belonging to 11 antimicrobial classes was evaluated using disk diffusion method; penicillins (ampicillin 10 µg [AMP]), beta-lactam/beta lactamase inhibitor (amoxicillin-clavulanic acid 20/10 µg [AMC]), cephems (cefoxitin 30 µg [FOX]), fluoroquinolone (ciprofloxacin 5 µg [CIP]), tetracycline (tetracycline 30 µg [TET], quinolone (nalidixic acid 30 µg [NAL]), folate pathway inhibitor (trimethoprim/sulfamethoxazole 1.25/23.75 ug [SXT]), aminoglycosides (streptomycin 10 µg [STR], Gentamicin 10 µg [GEN]), phenicol (chloramphenicol 30 µg [CHL]), polymyxin (colistin 10 µg [CL]) and monobactams (Imipenem 10 µg [IPM]). The Muller Hinton agar (Merck®, Germany) with confirmed ESBL producing *E. coli* were prepared as previous described. The antimicrobial disks were placed onto the agar using Multiple

Disk Dispenser (Oxoid®, Thermo Fisher Scientific, USA) before incubated at 37°C for 16-18 hours. The inhibition zone diameter was measured and interpreted following CLSI standard (CLSI, 2014). *E. coli* ATCC® 25922 was used for quality control. The isolates were classified as multidrug resistance (MDR) when resistance to at least one agent in three or more antimicrobial classes.

Statistical analysis

Prevalence and antimicrobials resistance profiles were described using descriptive statistics. Statistical analyses were performed by using SAS (2018). To compare differences between percentage, chi-squared tests were used considering statistical significance if $P < 0.05$.

RESULTS

Prevalence of ESBL producing *E. coli* from broiler farms and farm workers

Twenty-nine (60.4%) of 48 broiler farms had evidence of ESBL producing *E. coli*. ESBL producing *E. coli* was recovered from 43.8% of boot swabs, 12.5% of feed and 2.1% of water. Fifteen (55.7%) of 27 farm workers had ESBL *E. coli* recovered from rectal swab samples (Table 1). There was no significant different between the prevalence of ESBL producing *E. coli* in boot swabs and farmers (P=0.7036, chi-squares). Likewise, there was also no significant relationship between boot swab samples and farm worker rectal swab samples with odds ratio equal to 1.6 (95% CI; 0.3372-7.5922). A total of 53 ESBL producing *E. coli* were isolated from 171 samples (Table 1).

Antimicrobial resistance

Antimicrobial resistance of ESBL producing *E. coli* isolated by each sample type are shown in Table 2. All of ESBL producing *E. coli* isolates were resistant to ampicillin whereas none of the isolates were resistant to imipenem. High resistance rates were observed for streptomycin (94.3%), gentamicin (86.8%), tetracycline (77.4%), chloramphenicol (66.0%), nalidixic acid (58.5%), and sulfamethoxazole/trimethoprim (56.6%).

Antimicrobial resistance profiles

Among 53 ESBL producing *E. coli*, a total of 35 antimicrobial resistance profiles were observed (Table3). The three most common profile patterns shared between human and farm environment were AMP-GEN-SXT-NAL-TET-CHL-STR (5/53), AMP-GEN-SXT-TET-CHL-STR (5/53) and AMP-GEN-NAL-CIP-TET-STR (2/53), with the first two profiles being the most frequently observed profile. Almost all the ESBL producing *E. coli* (96.2%, 51/53) in this study were considered as multidrug resistant, except only 2 isolates that resistant to two classes of antimicrobial agents. In addition, resistance up to 9 antimicrobial classes was observed and accounted for 7.5% of the isolates (4/53).

Table 1 Prevalence of ESBL producing E. coli from broiler farms in Chiang Mai and Lamphun provinces

Samples	n	No. of positive ESBL-*E. coli*	Prevalence (%)	95% Confidence interval		Total no. of ESBL- *E. coli* isolates
				Lower	Upper	
Broiler farm	48	29	60.4	46.6	74.3	
Farm worker rectal swabs	27	15	55.7	36.8	74.3	19
Environment						
-Boot swabs	48	21	43.8	29.7	57.8	27
-Feed	48	6	12.5	3.1	21.9	6
-Water	48	1	2.1	0.0	6.1	1
Total	171	43	25.1	18.6	31.7	53

Table 2 Antimicrobial resistance of ESBL producing E. coli isolated from broiler farms and farm workers

Antimicrobial agents	No. of isolates (%)									
	Boot swabs (n=27)		Farm worker rectal swabs (n=19)		Feed (n=6)		Water (n=1)		Total (n=53)	
	R	I	R	I	R	I	R	I	R	I
AMP	27 (100)	0 (0)	19 (100)	0 (0)	6 (100)	0 (0)	1 (100)	0 (0)	53 (100)	0 (0)
AMC	8 (29.6)	13 (48.1)	0 (0)	9 (47.4)	0 (0)	3 (50.0)	0 (0)	0 (0)	8 (15.1)	25 (47.2)
FOX	7 (25.9)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	7 (13.2)	0 (0)
GEN	26 (96.3)	0 (0)	13 (68.4)	0 (0)	6 (100)	0 (0)	1 (100)	0 (0)	46 (86.8)	0 (0)
SXT	15 (55.6)	1 (3.7)	13 (68.4)	1 (5.3)	1 (16.7)	0 (0)	1 (100)	0 (0)	30 (56.6)	2 (3.8)
CL	5 (18.5)	0 (0)	2 (10.5)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	7 (13.2)	0 (0)
NAL	20 (74.1)	5 (18.5)	9 (47.4)	7 (36.8)	1 (16.7)	1 (16.7)	1 (100)	0 (0)	31 (58.5)	13 (24.5)
CIP	9 (33.3)	6 (22.2)	3 (15.8)	4 (21.1)	0 (0)	0 (0)	1 (100)	0 (0)	13 (24.5)	10 (18.9)
TET	20 (74.1)	0 (0)	16 (84.2)	0 (0)	4 (66.7)	0 (0)	1 (100)	0 (0)	41 (77.4)	0 (0)
IPM	0 (0)	3 (11.1)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	3 (5.7)
CHL	16 (59.3)	1 (3.7)	15 (79.0)	0 (0)	3 (50.0)	0 (0)	1 (100)	0 (0)	35 (66.0)	1 (1.9)
STR	27 (100)	0 (0)	16 (84.2)	3 (15.8)	6 (100)	0 (0)	1 (100)	0 (0)	50 (94.3)	3 (5.7)

R = resistance, I = intermediate, AMP = Ampicillin, AMC = Amoxicillin/clavulanic acid, FOX = Cefoxitin, GEN = Gentamicin, SXT = Sulfamethoxazole/trimethoprim, CL = Colistin, NAL = Nalidixic acid, CIP = Ciprofloxacin, TET = Tetracycline, IPM = Imipenem, CHL = Chloramphenicol, STR = Streptomycin

Table 3 Antimicrobial resistance patterns of ESBL producing E. coli isolated from broiler farms

No.	NR	Antimicrobial resistance patterns	NI	No. of isolates (%)			
				Boot swab (n=27)	Rectal swab (n=19)	Feed (n=6)	Water (n=1)
1	9	AMP-AMC-FOX-GEN-SXT-CL-NAL-CIP-CHL-STR	1	1 (3.7)			
2	9	AMP-AMC-FOX-GEN-SXT-CL-NAL-TET-CHL-STR	1	1 (3.7)			
3	9	AMP-AMC-FOX-GEN-SXT-NAL-CIP-TET-CHL-STR	1	1 (3.7)			
4	9	AMP-FOX-GEN-SXT-CL-NAL-CIP-TET-CHL-STR	1	1 (3.7)			
5	8	AMP-AMC-FOX-GEN-NAL-CIP-TET-CHL-STR	1	1 (3.7)			
6	8	AMP-GEN-SXT-CL-NAL-CIP-TET-CHL-STR	1		1 (5.3)		
7	7	AMP-AMC-FOX-GEN-SXT-CL-TET-STR	1	1 (3.7)			
8	7	AMP-GEN-SXT-NAL-CIP-TET-CHL-STR	3	2 (7.4)			1 (100)
9	6	AMP-AMC-FOX-GEN-TET-CHL-STR	1	1 (3.7)			
10	6	AMP-AMC-GEN-NAL-CIP-TET-STR	1	1 (3.7)			
11	6	AMP-GEN-SXT-NAL-CIP-TET-STR	1	1 (3.7)			
12	6	AMP-GEN-SXT-NAL-TET-CHL-STR*	5	2 (7.4)	2 (10.5)	1 (16.6)	
13	6	AMP-GEN-CL-NAL-TET-CHL	1		1 (5.3)		
14	6	AMP-SXT-CL-TET-CHL-STR	1	1 (3.7)			
15	6	AMP-SXT-NAL-CIP-TET-STR	1		1 (5.3)		
16	6	AMP-SXT-NAL-TET-CHL-STR	1		1 (5.3)		
17	5	AMP-GEN-NAL-CIP-TET-STR*	2	1 (3.7)	1 (5.3)		
18	5	AMP-GEN-NAL-TET-CHL-STR	3	3 (11.1)			
19	5	AMP-GEN-SXT-NAL-TET-STR	1	1 (3.7)			
20	5	AMP-GEN-SXT-TET-CHL-STR*	5	1 (3.7)	4 (21.1)		
21	5	AMP-GEN-NAL-TET-CHL	1		1 (5.3)		
22	5	AMP-GEN-SXT-TET-CHL	1		1 (5.3)		

Table 3 Antimicrobial resistance patterns of ESBL producing E. coli isolated from broiler farms (Cont.)

No.	NR	Antimicrobial resistance patterns	NI	No. of isolates (%)			
				Boot swab (n=27)	Rectal swab (n=19)	Feed (n=6)	Water (n=1)
23	5	AMP-SXT-TET-CHL-STR	2		2 (10.5)		
24	4	AMP-AMC-GEN-NAL-STR	1	1 (3.7)			
25	4	AMP-GEN-NAL-CHL-STR	1	1 (3.7)			
26	4	AMP-GEN-SXT-CHL-STR	1		1 (5.3)		
27	4	AMP-GEN-SXT-TET-STR	1	1 (3.7)			
28	4	AMP-GEN-TET-CHL-STR	1		1 (5.3)		
29	3	AMP-GEN-CHL-STR	2			3 (33.3)	
30	3	AMP-GEN-NAL-STR	2	2 (7.4)			
31	3	AMP-GEN-SXT-STR	1	1 (3.7)			
32	3	AMP-GEN-TET-STR	3			3 (50.0)	
33	3	AMP-NAL-STR	1		1 (5.3)		
34	2	AMP-GEN-STR	1	1 (3.7)			
35	2	AMP-STR	1		1 (5.3)		

NR = Number of antimicrobial resistance classes, NI = Number of isolates, AMP = Ampicillin, AMC = Amoxicillin/clavulanic acid, FOX = Cefoxitin, GEN = Gentamicin, SXT = Sulfamethoxazole/trimethoprim, CL = Colistin, NAL = Nalidixic acid, CIP = Ciprofloxacin, TET = Tetracycline, IPM = Imipenem, CHL = Chloramphenicol, STR = Streptomycin
* = patterns observed in both boot swab and rectal swab sample

DISCUSSION

Emergence of ESBL producing *E. coli* in livestock animals has become a public health concern over the last decade. Infections in human can lead to treatment failure, prolonged hospitalization, and increased mortality especially in patients with septicemia and urinary tract infection (Anunnatsiri et al., 2012). Our study found a high prevalence of ESBL producing *E. coli* in both broiler farmers (55.7%) and broiler flocks (43.8%) in Chiang Mai and Lamphun provinces. The results are similar to the study by Boonyasiri et al. (2014) who reported the prevalence of ESBL producing *E. coli* isolated from 30 healthy animal farm workers (56.7%) and 80 individual broilers (40.0%) in northern and eastern provinces of Thailand.

There is great variation in the prevalence of ESBL producing *E. coli* in broiler farms from different countries. For example, ESBL farm prevalence was reported in 28.21% of boot swab samples and, 60.26% by cloacal swab samples in the Philippines, 45 % from Belgium, 100% from Netherlands, 6.3% from Iran, and 42.1% from Portugal (Costa et al., 2009; Dierikx et al., 2013; Doregiraee et al., 2018; Gundran et al., 2019; Huijbers et al., 2014; Smet et al., 2008). These differences may be due to differences in locations, time, sampling strategy, and laboratory techniques (Li et al., 2016; Gundran et al., 2019). A study by Dahms et al. (2015) suggested that boot swabs seem more appropriate for identifying ESBL-positive barns than cloacal swabs. Similar discrepancies have been noted in humans by Pornsinchai et al. (2015), who suggested that lower prevalence of ESBL producing *E. coli* in humans could be due to lower recovery rates from rectal swabs as compared to stool samples.

There appears to be variability in ESBL recovery from humans in Thailand. Fecal carrier rate of ESBL producing *E. coli* in humans was reported as 58% in healthy volunteers in Kanchanaburi (Sasaki et al., 2010), 16.5% in patients admitted to the emergency room of Ramathibodi Hospital (Pornsinchai et al., 2015), 38.2% in hospitalized patients in Chonburi Hospital (Waiwarawooth et al., 2006), and 6% in patients with community-acquired urinary tract infection at Songklanagarind Hospital (Tunyapanit and Pruekprasert, 2006). In Chiang Mai, 13% of clinical isolates collected from patients admitted to Maharaj Nakorn Chiang Mai Hospital were identified as ESBL producing *E. coli* (Tharavichitkul et al., 2005). Huijbers et al. (2014) reported the carriage of ESBL producing *E. coli* from individuals with minimal and frequent contact with healthy broilers between 14.3% and 27.1%, respectively.

ESBL producing *E. coli* contamination of meat has been suggested as source of human infection. Nahar et al. (2018) reported the prevalence of ESBL producing *E. coli* in domestic (77%) and imported chicken meats (52%) in Japan, while zero of six samples from Thailand were positive. Tansawai et al. (2018) reported 69.2% of ESBL producing *E. coli* in poultry meat samples in Phitsanulok province, Northern Thailand. Genetic element studies of ESBL from the Netherlands suggested that ESBL genes, plasmids and *E. coli* isolates were most likely transmitted from poultry to humans via the food chain and also coincide with the recent emergence of ESBL producing *E. coli* infections in human (Kluytmans et al., 2013; Leverstein-van Hall et al., 2011). Our study highlighting the high prevalence of ESBL producing *E. coli* from broiler farms documents the need for continued monitoring and control of spreading ESBL

bacteria in the broiler production chain in Thailand in order to reduce the potential risk of food-borne transmission to consumers. Our study also found ESBL producing *E. coli* from boot swab samples (43.8%), feed (12.5%) and water (2.1%). This likely demonstrates the broad contamination that occurs in the farm environment. This contamination could serve to spread antimicrobial resistance over the entire flock and subsequently increase the risk of introducing the bacterium on poultry meat.

World Health Organization (2019) has classified quinolones, third and fourth generation cephalosporins and polymyxins as critically important antimicrobials, since these antimicrobials are used as a last-resort or are one of few available treatment for serious infections caused by multidrug resistance gram negative bacteria in humans. In our study, a high percentage of nalidixic acid resistance was observed in 74.1% of broiler flocks and 47.4% of farmers. Whereas, the overall resistance to ciprofloxacin (24.5%) was less. The resistance rates of ciprofloxacin were lower than reported by Gundran et al. (2019) at 88.4% in Philippines broiler farms. However, we did observe that 24.5% and 18.9% of all isolates had intermediate resistance to nalidixic acid and ciprofloxacin, respectively. Therefore, control and monitoring quinolone use in poultry production is necessary. In addition, colistin had been widely used in livestock production including in poultry production in Thailand. Recent strict controls were announced by the Thai Department of Livestock Development in 2017. In this present study, resistance to colistin was detected in both broiler flocks (18.5%) and farmers (10.5%). This resistance rate was higher than a study in the Philippines which reported 8.7% (Gundran et al., 2019) among broiler farms.

Other findings include the high level of resistance to other antimicrobials. Our findings are consistent to other ESBL producing *E. coli* studies in the Philippines and Japan (Gundran et al., 2019; Nahar et al., 2018) with 100% resistance reported to ampicillin. Additionally, we also observed resistance to streptomycin (94.3%), gentamicin (86.8%), tetracycline (77.4%), chloramphenicol (66.0%) and sulfamethoxazole/trimethoprim (56.6%). Gundran et al. (2019) reported the high resistance to ciprofloxacin (88.4%) and sulfamethoxazole/trimethoprim (72.5%) among ESBL producing *E. coli* isolated from broiler farms in the Philippines. Also 15.1% of the ESBL producing *E. coli* isolates were resistant to amoxicillin/clavulanic acid and another 47.2% had intermediate resistance. Amoxicillin/clavulanic acid is important in both human and animal medicine. Again, this reflects a need to monitor and control unnecessary use in both humans and animals.

Another concerning observation was that most of ESBL producing *E. coli* isolated from the broiler farms were multidrug resistant (MDR) (96.2%). In fact, some MDR isolates were resistant to nine different antimicrobial classes. There were two farms that the same antimicrobial resistance profile of AMP-GEN-SXT-NAL-TET-CHL-STR was observed from different sample type; (Farm A) boot swab and feed sample, (Farm B) boot swab and farm worker rectal swab sample. And this profile also presented as one of the most frequent profiles in this study. A common profile was identified across a number of farms and sample types suggesting possible transmission of ESBL producing *E. coli* between broiler farm environment and farmers. The resistance profile of AMP-GEN-SXT-TET-CHL-STR was the most frequent observed profile among farm worker rectal samples. This profile also observed in boot swab sample from one

farm. The direction of transmission is unclear, but broiler is likely a common source of ESBL producing *E. coli*. This demonstrates the need for further molecular characterization of isolates.

A number of resistance profiles were observed some in producers and some from the poultry environment. For example, there was one farm had a common resistance profile from a boot swab and water sample. This positive farm used surface water as a water source for broiler production. Therefore, it is likely that contaminated water can be a potential source of ESBL producing *E. coli* in broiler farms. Also, we cannot exclude that humans could introduce some resistant strains onto the farm.

CONCLUSION

This study documents the frequency of resistant ESBL producing *E. coli* from broiler farm workers and farm environment. The high level of resistance observed likely reflects the use of antibiotics on broiler farms in Chiang Mai and Lamphun provinces. Prior to recent regulations, amoxycillin, enrofloxacin, lincomycin, spectinomycin, erythromycin and colistin were used in broiler farms in the area. With current antimicrobial control measures, the use of antimicrobials on poultry operations is likely changing. It is important to assess the impact of these new regulations and guidelines. This study provides a baseline to observe these changes.

CONFLICT of INTEREST

All authors declare no conflict of interest.

AUTHOR CONTRIBUTIONS

Conceived and designed the experiments: NA
Ethical process: NA, AK
Sample collection: NA, JR
Performed the laboratory analysis: NA, MI, JR
Analyzed the data: NA, MI
Contributed reagents/materials/analysis tools: NA, KK
Wrote the paper: NA, MI, JB

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