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Prevalence of Extended-Spectrum *β*-Lactamase-producing *Enterobacterales* and Distribution of *bla*$_{ESBL}$ Genes from Patients who Underwent Abdominal Surgery

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ABSTRACT

The proliferation of extended-spectrum *β*-lactamase-producing *Enterobacterales* (ESBL-PE) is a serious global health problem. Screening for ESBL-PE from rectal swabs of patients who underwent abdominal surgery and determination of *bla*$_{ESBL}$ genes were performed. *E. coli* producing the extended-spectrum *β*-lactamase enzyme (ESBL) (77.41%) and ESBL-producing *K. pneumoniae* (12.9%) were found in 31 out of 104 patients. The most prevalent *bla*$_{ESBL}$ in both *E. coli* and *K. pneumoniae* were *bla*$_{TEM}$ and *bla*$_{OXA-2}$, whereas *bla*$_{SHV}$ was most prevalent only among *K. pneumoniae*. In contrast, *bla*$_{CTX-M}$ and *bla*$_{OXA-10}$ were the least detected in *E. coli*

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and *K. pneumoniae.* The fecal ESBL producers isolated from swabs taken prior to and after abdominal surgery illustrated persistence of colonization in patients' guts. Multiple-drug resistant bacterial infections found among ESBL carriages appeared to be associated with prolonged hospitalization and underlying complex diseases. It is essential that healthcare professionals are cognizant of the importance of monitoring for the presence of these ESBL producers, and reducing both intra-abdominal surgery site infections and the spread of resistance genes in prolonged fecal ESBL carriages in high-risk patients.

Keywords: Abdominal surgery; bla_{ESBL} genes; Colonization; ESBL producing *E. coli*; ESBL producing *K. pneumoniae*; Extended-spectrum β-lactamase (ESBL)-producing *Enterobacterales*; Rectal swabs

1. Introduction

Extended-spectrum β-lactamase-producing *Enterobacterales* (ESBL-PE) spp. have been a serious global health concern for transmission of multidrug resistance, particularly in Gram-negative bacteria such as *Escherichia coli* and *Klebsiella pneumoniae.* They have been responsible for hospital-acquired infections, including surgical site infections (SSIs), and are associated with considerable morbidity and mortality [1, 2]. β-lactamase resistance genes are generally plasmid mediated and rapidly evolving, which leads to extended spectra of various β-lactam antibiotics. ESBL-producing *E. coli* (EPEC) was first associated with hospital-acquired infections and, in reports from the 2000s, it was noted as an important cause of community-acquired infections [3-5]. The community sources of bacterial transmission were associated with hospital and healthcare settings, and colonization of food and water [6, 7]. SSIs are a common form of hospital acquired infection caused by *Staphylococcus aureus*, *E. coli*, *Enterococcu*s spp., *Pseudomonas aeruginosa*, and *Enterobacter* spp. Their presence has been a major concern due to the consequent increased mortality and longer hospitalization, which also leads to higher cost of treatment [8, 9]. ESBL-producing gram negative bacteria, especially *Enterobacterales* and *P. aeruginosa,* are able to hydrolyze extended spectrum cephalosporins including ceftazidime, ceftriaxone, cefotaxime, and oxyimino-monobactam; however, β-lactam inhibitors such as clavulanic acid are still available for treatment of ESBL infections [10-12].

ESBL-producing *Enterobacterales*, particularly *E. coli* and *Klebsiella* spp., have been involved in SSIs from colonization of the throat, nose, and rectum [13-15]. This occurrence called attention to the need for careful monitoring, effective safety measures, and communicating warnings of ESBL carriage. Infection control measures, prevention of ESBL transmission, and appropriate use of antibiotics are essential for reduction of infection risk [16].

ESBL genes are horizontally transmitted by mobile genetic elements among the same, as well as different, species [5]. The use of new generations of β-lactams for treatment helped to enable selection pressure to generate the diversification of different antibiotic resistance mechanisms [17].

ESBL carriage had not previously been screened in patients who underwent abdominal surgery at the 750-bed tertiary-care hospital where we conducted this study. The distribution of ESBL genes was explored among the isolated strains. Detailed information regarding present strains is essential for health care professionals in the monitoring and reducing of intra-abdominal surgery site infection and the spread of resistance genes in high-risk cases of prolonged ESBL carriage.

2. Materials and Methods

2.1 Study setting and patients

Rectal swabs from 104 patients who underwent abdominal surgery at a 750-bed tertiary-care hospital between July 2018 and March 2019 were collected twice, at 1 day before and 1-3 days after abdominal surgery.

ESBL-PE detected in patients 1 day prior to abdominal surgery was considered to be colonization. The isolates were considered to be persistent when detected at both 1 day prior to and 1-3 days after surgery.

This study was approved by the Human Research Ethics Committee No. 1, Faculty of Medicine, Thammasat University, current name: Human Ethics Committee of Thammasat University (Medicine), Thailand (MTU-EC-DS-2-014/61;083/2561). All patients gave informed consent prior to the study.

2.2 Screening for ESBL-producing *Enterobacterales*

Rectal swabs (in Amies transport medium) were collected from 104 patients who underwent abdominal surgery, at 1 day prior to and 1-3 days after surgery, to screen for ESBL-PE. Selective media included CHROM agar ESBL (Merck, USA) and MacConkey agar (Becton Dickinson, USA), which were used for isolation of ESBL-PE. The isolates were identified by standard biochemical tests. Combination disk diffusion test (CDT) was performed for phenotypic confirmation of the presence of ESBLs in *E. coli* and *K. pneumoniae*. Extended spectrum cephalosporin disks (Becton Dickinson, USA) were used as follows: a) a disk of 30 µg ceftazidime (CAZ) alone and a disk of CAZ with 10 µg clavulanic acid (CAZ/CLA); b) a disk of 30 µg cefotaxime (CTX) alone and a disk of CTX with 10 µg clavulanic acid (CTX/CLA). The disks mentioned above were placed on Mueller-Hinton agar (Becton Dickinson, USA) plates after inoculating the bacterial suspension (0.5 McFarland turbidity standard) and incubating at 37°C for 16-18 hours. An inhibition zone diameter ≥ 5 mm on a combination disk and not on the corresponding CAZ or CTX only disk was interpreted as positive confirmation of ESBL production [18]. *E. coli* and *K. pnuemoniae* strains which were resistant to either cefotaxime (inhibition zone ≤ 27 mm) or ceftazidime (inhibition zone ≤ 22 mm) with no increment of inhibition zones on either CAZ/CLA or CTX/CLA disks were interpreted as resistant strains. *K. pneumoniae* ATCC 700603, a positive control of ESBL producing bacteria, and *E. coli* ATCC 25922 were included as negative controls.

2.3 Detection of *bla*$_{\text{ESBL}}$ genes

The isolates were further examined for *bla*$_{\text{ESBL}}$ genes including *bla*$_{\text{TEM}}$, *bla*$_{\text{SHV}}$, *bla*$_{\text{CTX-M}}$, *bla*$_{\text{OXA-2}}$, and *bla*$_{\text{OXA-10}}$ by polymerase chain reaction (PCR). Primers used are as follows: TEM-F TCCGCTCATGAGACAAT AACC and TEM-R TTGGTCTGACAGTTACC AATGC for *bla*$_{\text{TEM}}$ (931bp) [19], SHV-F TGGTTATGCGTTATATTCG CC and SHV-R GGTTA GCGTTGCCAGTGCT for *bla*$_{\text{SHV}}$ (868bp) [20], CTX-F TCTTCCAGAATAAGGAATCCC and CTX-R CCGTTTCCGCTATTACAAAC for *bla*$_{\text{CTX-M}}$ (909bp) [19], OXA-2-F AAGAAACGCTA CTCGCCTGC and OXA-2-R CCACTCAAC CCATCCTACCC for *bla*$_{\text{OXA-2}}$ group (478bp)[21], OXA-10-F GTCTTTCGAGTACGGCATTA and OXA-10-R ATTTTCTTAGCGGCAA CTTAC for *bla*$_{\text{OXA-10}}$ group (720 bp) [22]. The primers and PCR conditions used are the same as described in previous reports with some modifications by using multiplex PCR. The annealing temperatures in this study were 58ºC for *bla*$_{\text{TEM}}$, *bla*$_{\text{SHV}}$, and *bla*$_{\text{OXA-10}}$ and 55ºC for *bla*$_{\text{CTX-M}}$ and *bla*$_{\text{OXA-2}}$. Positive controls of all genes were included. Negative results for *bla*$_{\text{ESBL}}$ genes were repeated for accuracy.

3. Results and Discussion

3.1 Isolation of *E. coli* and *K. pneumoniae*

EPEC (*n*=39), ESBL-producing *K. pneumoniae* (EPKP, n=8), resistant strains of *E. coli* (EC^R, *n*=5), and *K. pneumoniae* (KP^R, *n*=14) were detected in 36 of the 104 patients (34. 6%) who underwent abdominal surgery. A total of 31 of the 104 patients (29.8%) were found to have EPEC (23 patients) and EPKP (8 patients) strains present.

In the early 2000s, ESBL carriage dramatically increased by more than 50% in Southeast Asia, including Thailand [23]. In contrast, reports in Europe showed the lowest carriage rate at about 10% [24]. This study showed that 29.8% of the patients had EPEC and EPKP present. Similarly, ESBL producers were isolated from stool in 30.8% of healthy adults in Thailand [25].

EPEC and KP^R strains isolated at both pre- and post- abdominal surgery were found from 12 and 3 patients, respectively (Table 1).

Table 1. Detection of EPEC, EPKP, EC^R, and KP^R strains isolated from rectal swab pre- and post-abdominal surgery.

Pre-surgery	Post- surgery	Patient no.	Number of strains
EPEC	EPEC	3, 4, 9, 13, 14,17, 18, 20, 22, 23, 24, 27	12
EPEC	Not found	6, 10, 12, 19, 25, 34	6
Not found	EPEC	1, 11, 15, 16, 28, 35	6
EC^R	Not found	3, 5	2
Not found	EC^R	2, 35, 36	3
EPKP	Not found	7, 26, 36	3
Not found	EPKP	8, 20, 28, 30, 33	5
KP^R	KP^R	5, 20, 35	3
KP^R	Not found	6, 32	2
Not found	KP^R	30	1

3.2 Laboratory data

Seventeen patients provided bacterial culture from specimens including bile, percutaneous transhepatic biliary drainage, percutaneous catheter drainage, abdominal fluid, urine, and blood. Multidrug resistant (MDR) bacterial infections were reported in ESBL carriages (8 out of 17, 47.06%). One ESBL strain isolated from patient no. 4 appeared to be MDR. Other carriages were reported with no significant infections and no bacterial growth (*n*=7).

Length of hospital stay of all patients ranged from 2 to 232 days. All of the infected patients with MDR bacteria had prolonged length of hospitalization. Four of the 8 patients who had MDR bacterial infections died. All patients who died in-hospital were hospitalized longer than one and a half months. In addition, these patients had underlying complex diseases, such as malignant neoplasm of retroperitoneum, peritoneum, oesophagus, gallbladder, or rectum, and acute renal failure, as illustrated in Table 2.

Table 2. ESBL producers and multiple drug resistant (MDR) bacteria in ESBL carriages.

Patient No.	Age	Date of isolation	Rectal swab culture		Date of isolation	Specimen*	Isolated bacteria	Length of stay (status of treatment)	Diagnosis
			Before surgery	After surgery					
4	36	14/12/2018	SK4 EPEC	SK6 EPEC	1/1/2018 26/7/2019	PCD PCD (fluid) PTBD (fluid) Blood	KP MDR -EPEC & MDR-EC, KP, X. *maltophilia*, *Aeromonas* spp. EPEC, MDR-EC, EC, KP MDR-EC, MDR-KP, EC	21 (Survive)	Cholangitis, Obstruction of bile duct
12	31	5/2/2019	SK56 EPEC	Not found	5/2/2019 - 17/2/2019	Urine Abdominal fluid	MDR-EC MDR-EC	60 (death)	Acute renal failure, of uncertain Neoplasm or unknown behavior ~ peritoneum
14	49	13/2/2019	SK82 EPEC	Not found	27/2/2019 - 12/3/2019	Urine	MDR-EC CRE-KP	81 (death)	Malignant neoplasm of oesophagus, unspecified
16	71	1/3/2019	SK87 MDR_KP	SK85 EPECCC	12/3/2019	Bile Urine	MDR-*P. aeruginosa* KP	52 (transferred to other hospital)	Cholangitis, Intrahepatic bile duct carcinoma
17	61	7/3/2019	SK88 EPEC	Not found	14/2/2019 - 19/6/2019	Bile PTBD (fluid) Sputum	MDR-EC MDR-EC, CRE-ECL MDR-EC	232 (death)	Secondary malignant neoplasm of retroperitoneum and peritoneum, Acute cholecystitis
19	71	8/3/2019	SK93 EPEC	Not found	18/9/2019 6/10/2020	Urine Blood	*E. coli* MDR -*Acinetobacter indicus*, *Serratia marcescens*	42 (survive)	Calculus of bile duct without cholangitis or cholecystitis, Urinary tract infection, site not specified
23	76	18/3/2019	SK103 EPEC	SK104 EPEC	22/7/2019	Urine	MDR-KP	33 (Survive)	Obstruction of bile duct, Malignant neoplasm of head of pancreas Acute renal failure, unspecified

25	73	12/7/2018	SK107 EPEC	Not found	9/10/2019	Bile	MDR-EC	15 (death)	Malignant neoplasm of gallbladder, Malignant neoplasm of rectum

Note: * PCD, percutaneous catheter drainage; PTBD, percutaneous transhepatic biliary drainage

As shown in Table 2, prolonged hospitalization and complex underlying diseases of ESBL carriage seemed to be associated with increased risk of MDR bacterial infection and mortality. Moreover, the persistence of ESBL-PEs in patients' guts could put them at risk of infection and cause spread of infection in hospital and community settings in the future, as has been suggested in previous reports [26, 27].

Since ESBL genes are transmissible and result in increased spread of antimicrobial resistance by natural selection [28-30], horizontal transfer of certain plasmid-mediated resistance genes is almost certain to take place [5, 31, 32]. However, further detailed exploration of ESBL gene transmission is needed. It is crucial to investigate if ESBL-PE strains cause infections in community or healthcare settings to reinforce antimicrobial stewardship programs, infection prevention, and control measures.

3.3 Distribution of blaESBL genes

Regarding the results of ESBL gene detection, SHV, TEM, CTX-M, OXA-2, and OXA-10 type ESBLs were disseminated among EPEC, EPKP, and KPR strains, except for CTX-M type ESBL, which was not detected in ECR, as indicated in Table 3.

The EPEC and ECR isolates mainly possessed blaTEM and blaOXA-2, while EPKP and KPR possessed blaSHV and blaOXA-2. Strikingly, CTX-M and OXA-10 type ESBLs were the least detected in this study. OXA-10 type ESBL was found in EPKP and KPR more than it was in EPEC and EPR (Table 3).

Table 3. Distribution of *bla*$_{ESBL}$ genes (%) in *E. coli* and *K. pneumoniae* isolated from rectal swab.

Strains	*bla*$_{SHV}$	*bla*$_{TEM}$	*bla*$_{CTX-M}$	*bla*$_{OXA-2}$	*bla*$_{OXA10}$
EPEC (*n*= 39)	30.77	87.18	43.59	89.74	15.38
EC^{R} (*n*=5)	40.00	80.00	0	100.0	40.00
EPKP (*n*=8)	81.82	87.50	14.29	71.43	57.14
KP^{R} (*n*=14)	78.57	42.86	14.29	78.57	50.00

TEM and OXA-2 type ESBLs were the most common ESBL genes found in this study while CTX-M and OXA-10 were the least found in this study. In contrast, *bla*$_{CTX-M}$ showed a significant increase in many countries, including Thailand [23, 30, 33-35]. However, the potential threat of plasmid-mediated transmission cannot be ignored, since it may lead to rapid spread and infections in cases of high-risk ESBL carriage.

From data of *bla*$_{ESBL}$ gene detection, designated resistance gene patterns of EPEC/EC^{R} and EPKP/KP^{R} are presented in Table 4 and Table 5, respectively. The results indicated that the EPEC isolates from patients no. 3 and 4 had the same resistance gene pattern (9a). An ECR strain was also isolated from patient no. 3, harboring *bla*$_{TEM}$ and *bla*$_{OXA-2}$, as was found in the EPEC strain (Table 4).

Two other patients, no.17 and no. 22, had bacteria with resistance gene patterns 3a

and 5a, respectively. The resistance genes, including *bla*$_{SHV}$, *bla*$_{TEM}$, and *bla*$_{OXA-2}$ were present in pattern 3a but *bla*$_{SHV}$ was absent in pattern 9a. Pattern 5a strains carried *bla*$_{TEM}$, *bla*$_{CTX-M}$, and *bla*$_{OXA-2}$. It is remarkable that the most common pattern found among the isolates was pattern 9a (Table 4).

Table 4. Designated resistance gene patterns of EPEC and EC^{R} strains.

Patient No.	Strain code	Strain	Pre (0)/ post (1) surgery	*bla*$_{SHV}$	*bla*$_{TEM}$	*bla*$_{CTX-M}$	*bla*$_{OXA-2}$	*bla*$_{OXA-10}$	Pattern
1	SK1	EPEC	1	+	+	+	+	-	1a
2	SK2	EC^{R}	1	+	+	-	+	-	3a
3*	SK3	EPEC	0	+	+	-	+	-	3a
	SK76	**EPEC**	**1**	**-**	**+**	**-**	**+**	**-**	**9a**
	SK77	EPEC	1	+	+	+	+	-	1a
	SK86	**EC^{R}**	**0**	**-**	**+**	**-**	**+**	**-**	**9a**
4*	**SK4**	**EPEC**	**0**	**-**	**+**	**-**	**+**	**-**	**9a**
	SK6	**EPEC**	**1**	**-**	**+**	**-**	**+**	**-**	**9a**
5	SK8	EC^{R}	0	+	+	-	+	-	3a
6	SK11	EPEC	0	-	+	-	+	-	9a
9	SK20	EPEC	0	-	+	+	+	-	5a
	SK23	EPEC	1	+	+	+	+	-	1a
10	SK45	EPEC	0	-	+	+	+	-	5a
11	SK79	EPEC	1	-	+	-	+	-	9a
12	SK56	EPEC	0	-	+	+	+	-	5a
13	SK80	EPEC	0	-	+	-	+	+	7a
	SK81	EPEC	1	-	+	+	-	-	8a
14	SK82	EPEC	0	-	+	-	-	-	11a
	SK83	EPEC	1	-	+	-	+	-	9a
15	SK84	EPEC	1	-	+	-	+	-	9a
16	SK85	EPEC	1	-	+	-	+	-	9a
17*	**SK88**	**EPEC**	**0**	**+**	**+**	**-**	**+**	**-**	**3a**
	SK89	**EPEC**	**1**	**+**	**+**	**-**	**+**	**-**	**3a**
18	SK91	EPEC	0	+	+	-	+	-	3a
	SK92	EPEC	1	-	+	-	+	-	9a
19	SK93	EPEC	0	-	-	-	+	-	11a
20	SK96	EPEC	0	+	-	-	+	-	4a
	SK100	EPEC	1	-	+	+	+	-	5a
22*	**SK101**	**EPEC**	**0**	**-**	**+**	**+**	**+**	**-**	**5a**

	SK102	**EPEC**	**1**	**-**	**+**	**+**	**+**	**-**	**5a**
23	SK103	EPEC	0	-	-	+	+	+	10a
	SK104	EPEC	1	-	+	-	+	+	7a
24	SK105	EPEC	0	+	+	+	-	+	2a
	SK106	EPEC	1	-	+	+	+	-	5a
25	SK107	EPEC	0	-	+	-	+	-	9a
27	SK109	EPEC	0	+	+	+	+	-	1a
	SK110	EPEC	0	+	-	+	+	-	13a
	SK111	EPEC	1	+	-	-	+	-	4a
	SK112	EPEC	1	-	+	-	+	-	9a
28	SK114	EPEC	1	-	+	+	-	+	14a
34	SK137	EPEC	0	-	+	+	+	-	5a
35	SK129	EC^R	1	-	-	-	+	+	12a
	SK130	EPEC	1	-	+	-	+	+	7a
36	SK133	EC^R	1	-	+	-	+	-	9a

Note: * The same designated resistance patterns found both pre- and post- surgery are indicated in bold.

Table 5. Designated resistance gene patterns found among EPEC, EPKP, ECR, and KPR strains.

Designated pattern	*bla*$_{SHV}$	*bla*$_{TEM}$	*bla*$_{CTX-M}$	*bla*$_{OXA-2}$	*bla*$_{OXA-10}$	Number of isolates		
						EPEC	**EPKP**	**EC^R/KP^R**
1b	+	+	+	+	+	0	1	0
1a*	**+**	**+**	**+**	**+**	**-**	**4**	**0**	**1_KP^R**
2a	+	+	+	-	+	1	0	0
1c	+	+	+	-	-	0	0	1_KP^R
2b	+	+	-	+	+	0	3	0
3a*	**+**	**+**	**-**	**+**	**-**	**4**	**1**	**4_KP^R** **2_EC^R**
3b	+	+	-	-	-	0	1	0
13a	+	-	+	+	-	1	0	0
4b	+	-	-	+	+	0	0	4_KP^R
4a*	**+**	**-**	**-**	**+**	**-**	**2**	**0**	**1_KP^R**
5b	+	-	-	-	+	0	1	0
5a	-	+	+	+	-	8	0	0
14a	-	+	+	-	+	1	0	0
8a	-	+	+	-	-	1	0	0
7a	-	+	-	+	+	1	0	0
9a*	**-**	**+**	**-**	**+**	**-**	**11**	**1**	**2_EC^R**
10a	-	-	+	+	+	1	0	0
11a*	**-**	**-**	**-**	**+**	**-**	**2**	**0**	**1_KP^R**
12a*	**-**	**-**	**-**	**+**	**+**	**1**	**0**	**2_KP^R**
2c	-	-	-	-	+	0	0	1_KP^R

Note: * Different genera belonging to the same designated patterns are indicated in bold.

Interestingly, different genera appeared to have the same gene patterns, including patterns 1a, 3a, 4a, 9a, 11a, and 12a (Table 5). It is noted that patterns 3a and 9a were detected in all strains. KP^R strains (patients no. 5 and 6) and EC^R strains (patients no. 2 and 5), belonging to pattern 3a, harbored the same gene pattern as detected in EPEC (patients no. 3, 17, and 18) and EPKP (patients no. 20 and 33). In addition, pattern 9a was also detected in EPEC (patients no. 3, 4, 6, 11, 14,

15, 16, 18, 25, and 27), EC^R (patients no. 3 and 36), and KP^R (patient no. 7).

Other patterns including 1a, 4a, and 11 were found in EPEC and KP^R. Pattern 1a (patients no. 1, 3, 9, and 27), 4a (patients no. 20 and 27) and 11a (patients no. 4 and19) in EPEC strains were found in KP^R strains which carried the resistance gene pattern 1a (patient no. 16), 4a (patient no. 35), and 11a (patient no. 35). Pattern 12a was detected in EC^R (patient no. 35) and KP^R (patient no. 32).

EPKP strains harbored to two distinct patterns, patterns 1c and 2c. We hypothesized that these prototypes most likely originated from different sources. Specifically, the prototype 1c consisted of bla_{SHV}, bla_{TEM}, and $bla_{CTX\text{-}M}$ belonging to class A as designated in the Ambler classification. This ESBL genotype is predominant as demonstrated in previous reports [35-37]. The bla_{SHV}, bla_{TEM}, and $bla_{CTX\text{-}M}$ containing strains were potentially transferable reservoirs. The other prototype (2c) contained only $bla_{OXA\text{-}10}$, belonging to class D β-lactamases, which is also transferable. Emerging infection of these variants could hinder infection control measures and cause infection to spread more widely. It is encouraged to have active surveillance and infection control measures to reduce the risk of infections and to prevent transmission.

Rapid spread of ESBL-PEs and inappropriate antibiotic treatment for ESBL carriage pose a significant threat globally. For instance, enteric colonization of ESBL-PE was associated with a higher risk of deep SSI among patients who underwent colorectal surgery [38]. As a consequence of having a high risk of infection, new antibiotic treatments, transmission control, and decolonization have been studied to eradicate ESBL-PE colonization, including in SSIs, and to reduce the risk of transmission and infections. As the problem of ESBL-PE fecal colonization has been reported globally, screening for ESBL-PE carriage in those who will undergo colorectal surgery was proposed for preoperative prophylaxis but not recommended for other abdominal surgery which might have different risks for SSIs. However, there are several factors to consider before performing fecal screening for ESBL-PE colonization [39, 40]. It was recommended to screen for ESBL-PE colonization in patients who traveled in regions with a high prevalence of ESBL-Pes or other potential risk factors such as prolonged colonization and travel-related antibiotic use. Moreover, the prevalence of ESBL-PE fecal colonization in healthy subjects and the risk factors of ESBL-PE colonization previously reported have revealed that resistance in gram negative bacteria has significantly increased [41]. Therefore, we suggest screening for fecal ESBL carriages in those who are at high risk of infections to ensure appropriate treatment for MDR bacterial infections and to prevent transmission of resistance genes.

4. Conclusion

The distribution of ESBL genes seen in this study has provided detailed information which has aided recognition of rectal colonization with ESBL-producing *E. coli* and *K. pneumoniae*. The persistent colonization of ESBL producing *E. coli* and *K. pneumoniae* in the fecal flora of patients who underwent abdominal surgery could lead to transmission of resistance genes as indicated in the carriages of those who were at high risk of MDR bacterial infections. It is therefore necessary to establish and reinforce antibiotic stewardship programmes for more rigorous infection control. This awareness highlights the urgency of effective surveillance, prevention, infection control measures, and contact precaution for disseminating alerts of prolonged fecal ESBL carriages to reduce the risk of intra-abdominal SSIs from MDR ESBL-producers. As ESBL genes can spread by plasmid-mediated transmission, screening is important in order to prevent further emergence and spread, and to provide appropriate treatment. In addition, it is essential that health care professionals are cognizant of the importance of monitoring and reducing intra-

abdominal SSIs and the spread of resistance genes in cases of prolonged fecal ESBL carriages at high risk of infection.

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