



ORIGINAL ARTICLE

Prevalence of Antimicrobial Resistance in *Escherichia coli* Strains Isolated from Fishery Workers

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Abstract

Objectives: This study aimed to characterize the prevalence of antibiotic resistance in *Escherichia coli* isolates from the fecal samples of fishery workers who work in fish farms and often use antibiotics for the feeding fishes.

Methods: Seventy-three *E. coli* strains isolated from the fecal samples of fishery workers and 180 isolates from a control group of restaurant workers were tested for antibiotic resistance by agar disk diffusion with 16 antimicrobial agents.

Results: About 30% of isolates from each group showed antimicrobial resistance to ampicillin, and 60% of isolates from fishery workers and 41% from restaurant workers were resistant to tetracycline. The isolates showed higher resistance to cephalothin and cefoxitin than to other cephem antibiotics and to gentamicin than to other aminoglycosides. Our data indicated that fecal *E. coli* isolates from fishery workers showed higher antibiotic resistance than those of non-fishery workers (restaurant workers), especially to cephalothin, tetracycline, and trimethoprim–sulfamethoxazole ($p < 0.05$). However, rates of multidrug resistance were similar among the fishery workers and restaurant workers.

Conclusion: Frequent use of antibiotics may cause increased antibiotic resistance in the human microbiome.

1. Introduction

Microorganisms with increased antibiotic resistance are a significant health problem that may be worsened by frequent use of antibiotics in hospital [1,2] or in animal feed [3–6]. However, the antibiotic resistance of

microorganisms isolated from humans who add antibiotics to animal feed is not well known. Therefore, we have studied the antibiotic resistance of commensal *Escherichia coli* in volunteers over several years and have demonstrated that higher resistance to the antibiotics most frequently prescribed for diarrhea was found

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in the isolates of patients with diarrhea and apparently healthy individuals [7] and in livestock workers who often use antibiotics [8].

Here, we report the prevalence of antibiotic resistance in fecal *E. coli* isolates from healthy fishery workers who often use antibiotics for the feeding fishes and compare these data with isolates obtained from a control group of healthy individuals from the same region.

2. Materials and Methods

2.1. Fecal samples and *E. coli* isolated from samples

We collected fecal samples from 73 fishery workers and 180 healthy restaurant workers in the same region for comparison (Table 1). The fishery workers were 30–50 years of age and the restaurant workers were 20–50 years of age. The samples were plated on to MacConkey agar directly or occasionally after enrichment in tryptic soy broth (TSB) containing vancomycin (Sigma Chemical Co., St. Louis, MO, USA). Bacteria found on MacConkey agar were assayed using the API20E system (bioMérieux, Marcy l'Étoile, France). For individual samples, one or two *E. coli* isolates were selected randomly for the purpose of determining susceptibility.

2.2. Antimicrobial susceptibility test

Antibiotic susceptibility was determined by disk diffusion according to the guidelines of the Clinical and Laboratory Standards Institute (CLSI) (formerly NCCLS) [9]. Antimicrobial susceptibility was determined by agar disk diffusion (Kirby-Bauer method) using MuellerHinton agar (Difco, MI, USA). The following antibiotics (Oxoid, Hampshire, UK) were tested: ampicillin–sulbactam, ampicillin, tetracycline, aztreonam, cefotetan, cefepime, cefoxitin, cefotaxime, tobramycin, trimethoprim/sulfamethoxazole, cephalothin, imipenem, gentamicin, amikacin, piperacillin/tazobactam, and netilamicin. *E. coli* ATCC 25922 and *E. coli* ATCC 35218 were used as quality controls.

Table 1. Fecal samples collected for this study

Age groups (years)	Fishery workers (n = 73)		Healthy individuals (n = 180)	
	Male	Female	Male	Female
1–10	0	0	0	0
11–20	0	0	0	0
21–30	2	0	4	6
31–40	16	0	9	35
41–50	23	2	13	62
51–60	18	3	10	32
61+	8	1	2	7
Total	67	6	38	142

2.3. Statistical analysis

The antimicrobial susceptibility data are expressed as percentages or frequencies of the avian or human isolates. A one-way analysis of variance or χ^2 test was used to estimate overall difference between the percentages or frequencies of resistance between avian and human *E. coli* isolates. In all cases, $p < 0.05$ was regarded as statistically significant.

3. Results

A total of 258 isolates derived from 73 fishery workers and 180 isolates from restaurant workers were used to examine the antibiotic resistance of commensal *E. coli* strains. Among the fishery workers, more samples were collected from men than from women, while among the restaurant workers, more samples were collected from women than from men (Table 1).

About 30% of isolates from each group showed antimicrobial resistance to ampicillin and 60% of isolates from fishery workers and 41% from restaurant workers were resistant to tetracycline. The isolates of

Table 2. Antibiotic resistance rates of the isolates in each group

Antimicrobial agents	Antibiotic resistances (%) of isolates	
	Fishery workers	Control groups
β-Lactams		
Ampicillin	29	34
β-Lactam/β-lactamase inhibitor combinations		
Ampicillin–sulbactam	14	23
Piperacillin–tazobactam	0	0
Cephems		
Cephalothin*	36	0
Cefepime	0	0
Cefotetan	1	0
Cefotaxime	0	1
Cefoxitin	5	13
Carbapenems		
Imipenem	0	0
Aminoglycosides		
Amikacin	0	0
Gentamicin	15	9
Tobramycin	1	5
Netilamicin	1	0
Tetracyclines		
Tetracycline*	60	41
Monobactams		
Aztreonam	0	2
Folate pathway inhibitors		
Trimethoprim–sulfamethoxazole*	29	1

* $p < 0.05$.

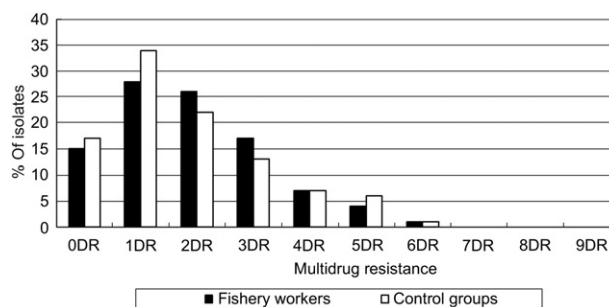


Figure 1. Antibiotic multi-resistance patterns of *Escherichia coli* strains isolated from fishery workers and restaurant workers. DR = drug resistance.

fishery workers showed higher resistance to cephalothin than to other cephem antibiotics (36%). However, the isolates of restaurant workers showed higher resistance to cefoxitin than to other antibiotics (13%). High resistance to trimethoprim/sulfamethoxazole was found in the isolates of fishery workers but not in the isolates of restaurant workers. Among aminoglycosides, the highest resistance was to gentamicin. There was a trend towards higher resistance among fishery workers than restaurant workers, especially to cephalothin, tetracycline, and trimethoprim–sulfamethoxazole ($p < 0.05$) (Table 2).

As shown in Figure 1, the multiple resistance patterns in the *E. coli* isolates of fishery workers are similar to those of restaurant workers. Overall, 55% of fishery workers and 49% of restaurant workers showed resistance to two or more antibiotics. Approximately 12% of the isolates showed resistance to four or more antibiotics. The most frequently observed resistance patterns in the fishery workers were to tetracycline/sulfamethoxazole (6 isolates) and to tetracycline/sulfamethoxazole/cephalothin (6 isolates). In the restaurant workers, the most frequently observed resistance pattern was to cefoxitin/tetracycline (10 isolates). The samples with resistance to the most antibiotics were found in a fishery worker isolate (resistance to 6 antibiotics: ampicillin–sulbactam, ampicillin, tetracycline, sulfamethoxazole, cefoxitin, and gentamicin) and in the isolates of four restaurant workers (resistance to 6 antibiotics: ampicillin, tetracycline, tobramycin, sulfamethoxazole, cefoxitin, and gentamicin).

4. Discussion

Antimicrobial resistance of intestinal bacteria isolated from food animals due to antibiotic usage is an increasing global problem in livestock environments [6,10,11]. The fishery industry is a significant economic force in Korea but there is little information regarding the antibiotic resistance of fecal *E. coli* isolates from fishery workers. Therefore, in this study, the prevalence of antibiotic resistance in fecal *E. coli* isolates from healthy fishery workers was examined.

As shown, fecal *E. coli* isolates from fishery workers showed higher antibiotic resistance than those of non-fishery workers (restaurant workers), especially to cephalothin, tetracycline, and trimethoprim-sulfamethoxazole ($p < 0.05$). This may be partly due to frequent feeding of antibiotics to fish by the fishery workers. Tetracycline is a common first-line antibiotic for many domestic animals and is used as a growth promoter and infection control agent, and is often used before the antibiotic resistance profile of a pathogen has been determined [12–14]. This result agrees with the findings of other research [8,15]. In this study, we found that fishery workers often use neomycin, destomycin A, and hygromycin B with feeds. The finding of a higher prevalence of resistance in fishery worker isolates suggests that antimicrobial use in fish may be a factor in the emergence of antimicrobial resistance in the human fecal *E. coli* isolates.

Other studies by us have reported that there is higher multidrug resistance in the *E. coli* isolates of patients with diarrhea [7] and in livestock workers who often use antibiotics for the feeding animals [8]. However, in this study, the multiple resistance patterns in the *E. coli* isolates of fishery workers are similar to those of restaurant workers.

In conclusion, the higher resistance in fishery workers may be explained by the greater exposure of these workers to antibiotics. The information gathered may help us manage the evolution of antimicrobial resistance in the future.

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