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Research Article

Prevalence and Phenotypic and Genotypic Patterns of Antibiotic Resistance of *Acinetobacter baumannii* Strains Isolated from Fish, Shrimp, and Lobster Samples

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Acinetobacter baumannii has emerged as a significant hospital pathogen, quickly becoming resistant to commonly prescribed antimicrobials. The present survey was done to evaluate the prevalence, antibiotic resistance pattern, and distribution of antibiotic resistance genes amongst the *A. baumannii* strains isolated from fish, shrimp, and lobster samples. Four-hundred and fifty seafood samples (100 g each) were collected from Shiraz, Iran. *Acinetobacter baumannii* was determined using culture and biochemical tests. Pattern of antibiotic resistance and distribution of antibiotic resistance genes were determined using the disk diffusion and polymerase chain reaction, respectively. *A. baumannii* contamination rate amongst the examined seafood samples was 4.44%, with the higher contamination rate of fish samples (7.85%). *A. baumannii* isolates harbored the maximum resistance rate against tetracycline (85%), ampicillin (85%), gentamicin (70%), and erythromycin (60%). Resistance rates toward trimethoprim-sulfamethoxazole, ciprofloxacin, ceftazidime, and azithromycin were 55%, 45%, 35%, and 30%, respectively. The minimum rates of resistance were obtained against imipenem (10%) and chloramphenicol (15%). The most commonly detected antibiotic resistance genes were bla_{CITM} (75%), bla_{SHV} (70%), *tetA* (70%), *qnrA* (55%), bla_{VIM} (50%), and *aac(3)-IV* (50%). *aadA1*, *sul1*, *dfrA1*, *qnr*, bla_{VIM}, bla_{SIM}, $bla_{\text{OXA-51}}$, $bla_{\text{OXA-23}}$, and $bla_{\text{OXA-58}}$ genes were detected in 40%, 30%, 45%, 50%, 35%, 25%, 30%, and 20% of isolates, respectively. The role of seafood samples as a potential reservoirs of antibiotic-resistant *A. baumannii* strains was determined. However, further investigations are required to identify additional epidemiological features of *A. baumannii* in seafood samples.

1. Introduction

Seafoods are noteworthy nutrient, marketing, and economic foodstuffs worldwide. Seafoods are rich sources of different nutrient molecules, including proteins, minerals, fatty acids, and even vitamins [1]. Their routine consumption will decrease the risk of diverse metabolic and nutrition diseases [2]. However, seafood human manipulation at the ports and also their own filter feeding manner may increase the risk of microbial contamination and subsequent foodborne diseases [3].

Acinetobacter baumannii (*A. baumannii*) is a Gram-negative, rod-shaped, aerobic, nonfermentative, catalase-positive, and oxidase-negative bacterium with ubiquitous and saprophytic nature [4, 5]. *A. baumannii* have appeared as an imperative nosocomial pathogen because of its high survival rate in the environment [6]. In the hospital cases, *A. baumannii* is responsible for the occurrence of diverse infections, particularly meningitis, endocarditis, pneumonia, peritonitis, skin, burn and wound, urinary and respiratory tract infections, and bacteremia [7]. Rendering its high resistance to environmental conditions, *A. baumannii* can exist in water and foodstuffs [8]. *A. baumannii* strains have also been rarely isolated from meat [9], vegetable [10], and milk [11] samples. Some Acinetobacter species are also responsible for different

types of infections in marine animals and cause economic burden to the fish, shrimp, and lobster farming [12, 13]. However, information about the *A. baumannii* clonality in food is scarce, which may bold the role of foods in transmission of this pathogen into the human population.

Many studies showed an emergence of antibiotic resistance amongst the *A. baumannii* strains of different sources [14]. In this regard, high resistance rate of *A. baumannii* strains against diverse antibiotic groups, including tetracycline, penicillins, aminoglycosides, quinolones, macrolides, carbapenems, and phenicols, has been reported several times [15]. Many studies showed that the genes that encode resistance toward streptomycin (*aadA1*), gentamicin (*aac(3)-IV*), tetracyclines (*tetA* and *tetB*), sulfonamide (*sul1*), beta-lactams (bla_{CITM} and bla_{SHV}), chloramphenicol (*cmlA* and *cat1*), trimethoprim (*dfrA1*), quinolones (*qnrA*), carbapenems (bla_{VIM}, bla_{IMP}, and bla_{SIM}), and oxacillins (OXA-51-like, OXA-23-like, OXA-24-like, and OXA-58-like) are mainly responsible for the occurrence of antibiotic resistance [16, 17]. Determination of the antibiotic resistance pattern of *A. baumannii* strains may help a lot in finding the best therapeutic approaches and changing the infection epidemiology.

Contamination of various type of foods by *A. baumannii* is largely reported [18–20]. *A. baumannii* was recovered from various raw meat including sheep, goat, cow, and camel [21–23]. Recently, Ababneh et al. also reported that *A. baumannii* was recovered from fresh products including vegetables and fruits [24]. However, few studies report the presence of *A. baumannii* in seafood. Isolated studies have reported the presence of *A. baumannii* carrying resistance genes (Oxa-23) in some seafood species, but their prevalence has not been determined [25, 26]. In Iran, only one study reported the presence of *A. baumannii* in seafood, and a low prevalence of 5.6% was found [27]. Regarding the clinical significance of *A. baumannii* as an emerging foodborne pathogen, thus, the present study was aimed at assessing the prevalence and phenotypic (disk diffusion) and genotypic (detection of antibiotic resistance genes) patterns of antibiotic resistance of *A. baumannii* strains isolated from fish, shrimp, and lobster samples.

2. Materials and Methods

2.1. Samples. From May to September 2020, 450 seafood samples including shrimp ($n = 130$), fish ($n = 140$), and lobster ($n = 180$) samples were randomly collected from fish market centers at Shiraz, Iran. Each sample (100 g from the dorsal muscle) was collected separately in highly hygienic condition using sterile tissue forceps in laboratory tubes containing peptone water solution (Merck, Germany). Samples were healthy and fresh and all were caught from the Persian Gulf, Iran. All samples were transferred to laboratory using cool boxes at 4°C.

2.2. Isolation and Identification of A. baumannii. The *A. baumannii* strains were isolated by microbial culture and identified using different biochemical tests and polymerase chain reaction (PCR) method. For this purpose, 10 g of seafood meat samples was homogenized in 90 mL of nutrient broth using a stomacher (Stomacher 400 Circulator, Seward, Norfolk, UK) for about 1 min. All media were incubated at 37°C overnight with agitation. Then, 10 μL of enriched media was inoculated onto selective ChromID ESBL agar (bioMérieux, France) and incubated for 24 h at 37°C. White colonies in selective ChromID ESBL agar were considered presumptive *A. baumannii* and transferred onto tryptic soy agar (TSA, Merck, Germany) plates supplemented with sheep blood (5%, Merck, Germany). Media were incubated for 24 h at 37°C. *A. baumannii* colonies were further identified using the biochemical tests, including Gram staining, citrate, catalase, oxidase, urease, malonate consumption, sugar oxidation and fermentation, indole production, and motility [28]. Species identification was done using gelatin liquefaction, glucose oxidation, arginine hydrolysis, hemolysis on blood agar, growth at 37°C and 42°C, and chloramphenicol susceptibility test [28]. Final confirmation of *A. baumannii* isolates was done using the PCR (targeted the *16S-23S* ribosomal DNA of *A. baumannii*) [29]. For this aim, *16S-23S* ribosomal DNA was targeted using the forward: 5′-CATTATCACGGTAATTAGTG-3′ and reverse: 5′-AGAGCACTGTGCACTTAAG-3′ (208 bp) primers [29].

2.3. Antibiotic Resistance Examination. A. baumannii antibiotic resistance pattern was assessed by the simple disk diffusion. The Kirby-Bauer disk diffusion method on the Mueller-Hinton agar (MHA, Merck, Germany) rendering the Clinical and Laboratory Standard Institute (CLSI) guidelines was applied [30]. *A. baumannii* resistance was examined against tetracycline (30 μg/disk) (T30), erythromycin (15 μg/disk) (E15), azithromycin (15 μg/disk) (Az15), ceftazidime (30 μg/disk) (Cft30), gentamicin (10 μg/disk) (G10), ciprofloxacin (5 μg/disk) (Cip5), trimethoprim/sulfamethoxazole (25 μg/disk) (Tr-Sul), chloramphenicol (30 μg/disk) (C30), imipenem (30 μg/disk) (I30), and ampicillin (10 μg/disk) (A10) (Oxoid, UK). After superficial culture of *A. baumannii* on the MHA plates, antibiotic discs were placed in plates with significant distance from each other. Media containing antibiotic discs were incubated for 24 h at 37°C. Then, the *A. baumannii*'s diameter of the growth inhibition zones surrounding the discs was measured and compared with the CLSI instructions [30]. *A. baumannii* ATCC 19606 and *Escherichia coli* (*E. coli*) ATCC 25922 were used as controls.

2.4. PCR-Based Detection of Antibiotic Resistance Genes. At first, all *A. baumannii* isolates were prepared for DNA extraction. For this purpose, *A. baumannii* were subcultured on tryptic soy broth (TSB) media and incubated for 48 h at 37°C. Then, the DNA was extracted from colonies using the kit of DNA extraction (Thermo Fisher Scientific, St. Leon-Rot, Germany). Then, the quality (by electrophoresis on a 2% agarose gel) and quantity (by the spectrophotometer (A260/A280)) of extracted DNA were checked [31, 32].

Table 1 shows the conditions used for the PCR-based detection of antibiotic resistance genes in the *A. baumannii* isolates [33–36]. A programmable DNA thermocycler (Eppendorf Mastercycler 5330, Eppendorf-Netheler-Hinz GmbH, Hamburg, Germany) was used in all PCR reactions. All ingredients were purchased (Thermo Fisher Scientific, St. Leon-Rot,

Table 1: Conditions used for the PCR-based detection of antibiotic resistance genes in the *A. baumannii* isolates [23–26].

Target gene	Primer (5′-3′)	Size (bp)	PCR cycles	PCR volume
aadA1	(F) TATCCAGCTAAGCGCGAACT (R) ATTTGCCGACTACCTTGGTC	447	1 cycle: 94°C for 6 min 33 cycles: 95°C for 70 s, 55°C for 65 s, and 72°C for 90 s 1 cycle: 72°C for 8 min	5 μL of PCR buffer 10x 2 mM of Mgcl2 150 μM of dNTP 1 μM of each primer (F and R) 1 U of Taq DNA polymerase 3 μL of DNA template
aac(3)–IV	(F) CTTCAGGATGGCAAGTTGGT (R) TCATCTCGTTCTCCGCTCAT	286		
sul1	(F) TTCGGCATTCTGAATCTCAC (R) ATGATCTAACCCTCGGTCTC	822		
bla_{SHV}	(F) TCGCCTGTGTATTATCTCCC (R) CGCAGATAAATCACCACAATG	768		
bla_{CITM}	(F) TGGCCAGAACTGACAGGCAAA (R) TTTCTCCTGAACGTGGCTGGC	462		
cat1	(F) AGTTGCTCAATGTACCTATAACC (R) TTGTAATTCATTAAGCATTCTGCC	547		
cmlA	(F) CCGCCACGGTGTTGTTGTTATC (R) CACCTTGCCTGCCCATCATTAG	698		
tetA	(F) GGTTCACTCGAACGACGTCA (R) CTGTCCGACAAGTTGCATGA	577		
tetB	(F) CCTCAGCTTCTCAACGCGTG (R) GCACCTTGCTGATGACTCTT	634		
dfrA1	(F) GGAGTGCCAAAGGTGAACAGC (R) GAGGCGAAGTCTTGGGTAAAAAC	367		
qnrA	(F) GGGTATGGATATTATTGATAAAG (R) CTAATCCGGCAGCACTATTTA	670		
bla_{IMP}	(F) GAATAGAATGGTTAACTCTC (R) CCAAACCACTAGGTTATC	188	1 cycle: 95°C for 4 min 30 cycles: 95°C for 45 s, 58°C for 60 s, and 72°C for 40 s 1 cycle: 72°C for 5 min	5 μL of PCR buffer 10x 2 mM of Mgcl2 150 μM of dNTP 1 μM of each primer (F and R) 1 U of Taq DNA polymerase 3 μL of DNA template
bla_{VIM}	(F) GTTTGGTCGCATATCGCAAC (R) AATGCGCAGCACCAGGATAG	382		
bla_{SIM}	(F) GTACAAGGGATTCGGCATCG (R) GTACAAGGGATTCGGCATCG	569		
$bla_{Oxa\text{-}51\text{-like}}$	(F) TAATGCTTTGATCGGCCTTG (R) TGGATTGCACTTCATCTTGG	353	1 cycle: 94°C for 5 min 32 cycles: 95°C for 50 s, 60°C for 60 s, and 72°C for 70 s 1 cycle: 72°C for 10 min	5 μL of PCR buffer 10x 2 mM of Mgcl2 150 μM of dNTP 1 μM of each primer (F and R) 1 U of Taq DNA polymerase 3 μL of DNA template
$bla_{Oxa\text{-}23\text{-like}}$	(F) GATCGGATTGGAGAACCAGA (R) ATTTCTGACCGCATTTCCAT	501		
$bla_{Oxa\text{-}24\text{-like}}$	(F) GGTTAGTTGGCCCCCTTAAA (R) AGTTGAGCGAAAAGGGGATT	246		
$bla_{Oxa\text{-}58\text{-like}}$	(F) AAGTATTGGGGCTTGTGCTG (R) CCCCTCTGCGCTCTACATAC	599		

Germany). In addition, amplified samples were analyzed by electrophoresis (120 V/208 mA) in a 2.5% agarose gel stained with 0.1% ethidium bromide (0.4 μg/mL) [36]. Besides, UVI doc gel documentation systems (Grade GB004, Jencons PLC, London, UK) were used to analyze images [37–39].

2.5. Data Analysis. SPSS software was applied to assess any statistical analysis of extracted data. For this aim, chi-square and Fisher's tests were used. Statistical differences between sample types and *A. baumannii* prevalence and antibiotic resistance were examined. P value < 0.05 was considered as significant level.

3. Results

3.1. Prevalence of A. baumannii. Figure 1 shows a sample of gel electrophoresis of PCR products for *A. baumannii* detection. Table 2 shows the *A. baumannii*'s prevalence amongst the seafood samples. Twenty out of 450 (4.44%) seafood samples were contaminated with *A. baumannii*. Amongst the examined samples, fish harbored the highest (7.85%) contamination rate with the *A. baumannii*, while lobster harbored the lowest (1.66%).

3.2. Phenotypic Pattern of Antibiotic Resistance. Table 3 shows the *A. baumannii*'s phenotypic pattern of antibiotic resistance. *A. baumannii* isolates harbored the maximum resistance rate against tetracycline ($n = 17$; 85%), ampicillin ($n = 17$; 85%), gentamicin ($n = 14$; 70%), and erythromycin ($n = 12$; 60%). Resistance rates toward trimethoprim-sulfamethoxazole, ciprofloxacin, ceftazidime, and azithromycin were 55% ($n = 11$), 45% ($n = 9$), 35% ($n = 7$), and 30% ($n = 6$), respectively. The minimum rates of resistance were

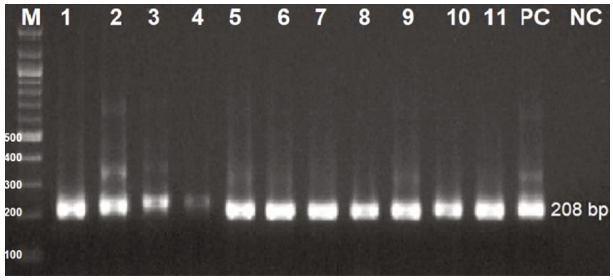

Figure 1: Gel electrophoresis of *A. baumannii 16S-23S* ribosomal DNA in PCR reaction. M: ladder (100 bp)' PC: positive control (*A. baumannii* ATCC 19606); NC: negative control (water (PCR-grade)); 1-11: positive samples for the *16S-23S* ribosomal DNA gene (208 bp).

Table 2: *A. baumannii*'s prevalence amongst the seafood samples.

Samples	No. of collected samples	*A. baumannii* prevalence (%)
Lobster	180	3 (1.66)
Fish	140	11 (7.85)
Shrimp	130	6 (4.61)
Total	450	20 (4.44)

obtained against imipenem ($n = 2$; 10%) and chloramphenicol ($n = 3$; 15%).

3.3. Genotypic Pattern of Antibiotic Resistance. Figure 2 shows the samples of gel electrophoresis of PCR products for antibiotic resistance gene detection. Table 4 shows the *A. baumannii*'s genotypic pattern of antibiotic resistance. Amongst the examined antibiotic resistance genes, bla_{CITM} (75%), bla_{SHV} (70%), *tetA* (70%), *qnr* (55%), *bla*VIM (50%), and *aac(3)-IV* (50%) had the highest distribution rate. The *aadA1*, *sul1*, *dfrA1*, *qnrA*, bla_{VIM}, bla_{SIM}, $bla_{\text{OXA-51}}$, $bla_{\text{OXA-23}}$, and $bla_{\text{OXA-58}}$ were detected in 40%, 30%, 45%, 50%, 35%, 25%, 30%, and 20% of strains, respectively. The lowest rates were observed for *cmlA* (5%), *cat1* (10%), bla_{IMP} (10%), *tetB* (15%), and $bla_{\text{OXA-24}}$ (15%) genes.

4. Discussion

Cumulative consumption of fresh, raw, and undercooked food is not only measured to be the most significant foodborne disease leading cause [40], but also related to frequent bacterial pathogens eruptions. In this regard, *A. baumannii* is considered an important risk of foodborne diseases in contaminated food samples in rare studies [18]. In this survey, *A. baumannii* prevalence amongst the lobster, fish, and shrimp samples was 1.66%, 7.85%, and 4.61%, respectively. The higher catch rate of fish compared to shrimp and lobster and the transfer of contamination between caught fishes can be a possible reason for the higher prevalence rate of *A. baumannii* in fish samples. Filter-feeding nature of shrimp and lobster which accumulate the pathogens may face them in a higher risk of contamination. Additionally, the role of hand manipulation of seafoods in ports may increase the risk of contamination. Scarce studies have been aimed at assessing the *A. baumannii* prevalence in seafood samples. In our previous survey [27], *A. baumannii* prevalence amongst the fish, shrimp, and lobster samples was 10%, 5.30%, and 2.50%, respectively. Hasiri et al. found *A. baumannii* in 5.6% seafood in Iran. This low prevalence in Iran indicates that *A. baumannii* contamination of seafood is emerging. In this study, samples were collected only from seafood markets in central Shiraz, so this low prevalence is not representative of the whole province, and different results could be found in other seafood markets [27]. In the United States [41], *Acinetobacter* was detected in 9.54% of retail seafood samples. In India, a virulent *A. baumannii* associated with mortality of farmed Indian major carp Labeo rohita was found in 2017 [42]. Another study performed in India reported multi-drug-resistant *Acinetobacter baumannii* associated with snakehead Channa striatus eye infection [43]. Antibiotic-resistant *Acinetobacter johnsonii* and *Acinetobacter lwoffii* were isolated from fish cultured in Poland [44]. Other food-based research focused on other food samples. Askari et al. [45] reported that the *A. baumannii* prevalence amongst the raw meat samples collected from Iran was 20.10%. According to the findings of Askari et al. [21], *A. baumannii* prevalence amongst the retail camel, sheep, and goat meat samples was 2.26%, 41.12%, and 11.72%, respectively. In Egypt [19], *A. baumannii* prevalence amongst the camel, cow, sheep, and goat meat samples was 9.68%, 15%, 46.55%, and 32.50%, respectively. In another Iranian survey, Tavakol et al. [46] reported that the *A. baumannii* prevalence amongst the raw chicken, bovine, camel, and ovine meat samples was 45.45%, 18.18%, 13.64%, and 9.10%, respectively. Such a large variation across countries in *A. baumannii* prevalence rate from food samples may show real regional differences or may be affected by the use of various detection techniques. Additional developments in the *A. baumannii* detection techniques in foods are desirable.

Acinetobacter baumannii strains isolated from seafood samples harbored a high resistance rate toward some of the basic therapeutic options mainly used in veterinary and medicine, particularly tetracycline, ampicillin, gentamicin, and erythromycin. Unauthorized and improper antibiotic administration, antibiotic and disinfectant overuse, and self-medication with antibiotics can be conceivable reasons for the high prevalence of antibiotic resistance. Contact of the seafood surface with the port environment, equipment used for their sorting, and contaminated staff can cause the transfer of antibiotic-resistant strains to them. Kim et al. [47] stated the high *A. baumannii* prevalence (27.80%) amongst the raw milk samples which is supported by the high antibiotic resistance of isolates against ceftriaxone (4.4%), tetracycline (30.8%), gentamicin (2.9%), and cefotaxime (12.5%). Similarly, in surveys conducted on Iran [48], Brazil [49], Jordan [50], United States [51], Korea [52], and Africa [53], high *A. baumannii* resistance rates against tetracycline, ampicillin, gentamicin, trimethoprim, ciprofloxacin, and erythromycin were reported. Askari et al. [21] stated that the *A. baumannii* strains isolated from raw meat of animal species harbored the highest resistance rate against tetracycline (82.35%), gentamycin (74.50%), streptomycin (54.90%), cotrimoxazole (70.58%), and trimethoprim (62.74%). In another study, Askari et al. [45] mentioned that

Table 3: *A. baumannii*'s phenotypic pattern of antibiotic resistance.

Samples (no. of positive *A. baumannii*)	Phenotypic pattern of antibiotic resistance (%)									
	T30	E15	Az15	Cft30	G10	Cip5	Tr-Sul	C30	I30	A10
Lobster (3)	2 (66.66)	2 (66.66)	1 (33.33)	1 (33.33)	2 (66.66)	1 (33.33)	2 (66.66)	0 (0)	0 (0)	2 (66.66)
Fish (11)	10 (90.90)	7 (63.63)	3 (27.27)	4 (36.36)	8 (72.72)	5 (45.45)	6 (54.54)	2 (18.18)	1 (9.09)	10 (90.90)
Shrimp (6)	5 (83.33)	3 (50)	2 (33.33)	2 (33.33)	4 (66.66)	3 (50)	3 (50)	1 (16.66)	1 (16.66)	5 (83.33)
Total (20)	17 (85)	12 (60)	6 (30)	7 (35)	14 (70)	9 (45)	11 (55)	3 (15)	2 (10)	17 (85)

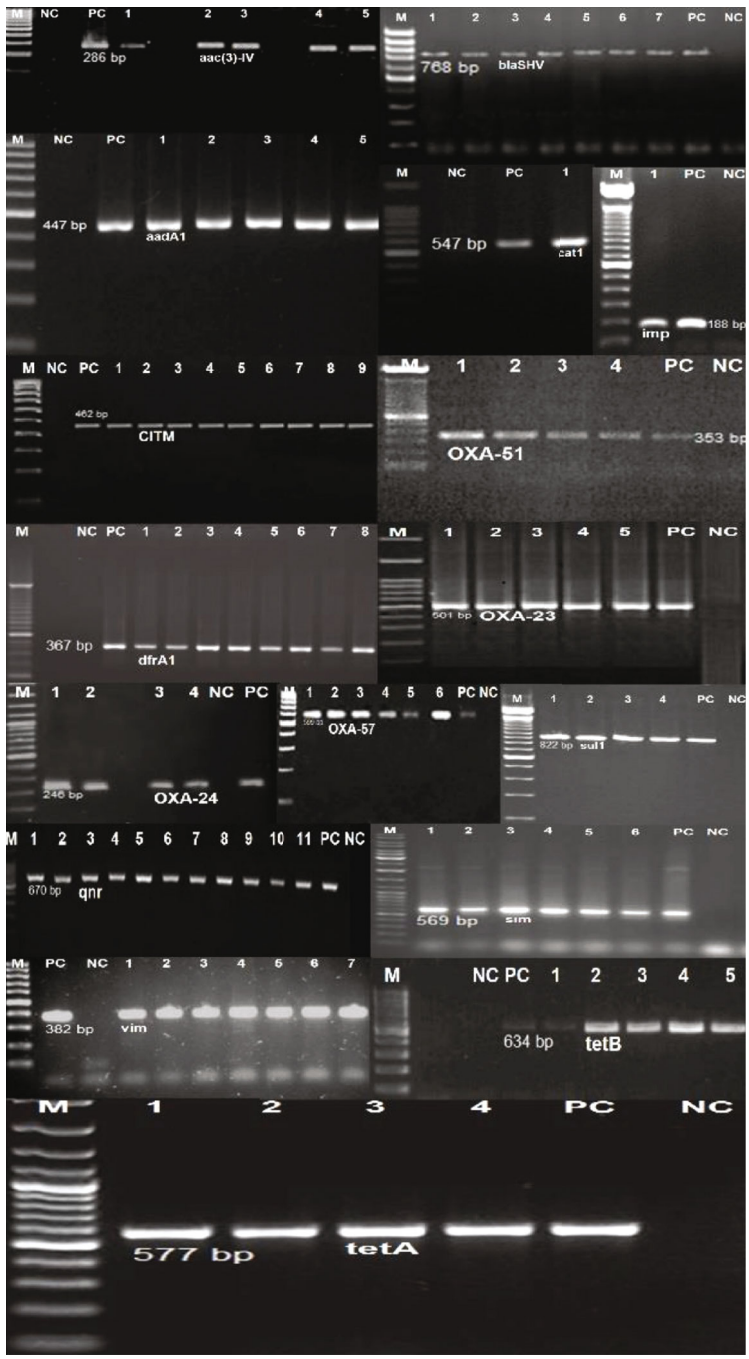

Figure 2: Gel electrophoresis of *A. baumannii* antibiotic resistance genes in PCR reactions. In all figures, M: ladder (100 bp); PC: positive control; NC: negative control; numbers: positive samples for antibiotic resistance genes.

Table 4: *A. baumannii*'s genotypic pattern of antibiotic resistance.

Samples (no. of positive *A. baumannii*)	Genotypic pattern of antibiotic resistance (%)																	
	aadA1	*aac(3)-IV*	*sul1*	bla_{SHV}	bla_{CITM}	*cat1*	*cmlA*	*tetA*	*tetB*	*dfrA1*	*qnrA*	bla_{IMP}	bla_{VIM}	bla_{SIM}	$bla_{Oxa\text{-}51}$	$bla_{Oxa\text{-}23}$	$bla_{Oxa\text{-}24}$	$bla_{Oxa\text{-}58}$
Lobster (3)	1 (33.33)	1 (33.33)	1 (33.33)	2 (66.66)	2 (66.66)	—	—	2 (66.66)	—	1 (33.33)	2 (66.66)	—	2 (66.66)	1 (33.33)	1 (33.33)	1 (33.33)	—	1 (33.33)
Fish (11)	5 (45.45)	6 (54.54)	3 (27.27)	8 (72.72)	9 (81.81)	1 (9.09)	1 (9.09)	8 (72.72)	2 (18.18)	5 (45.45)	6 (54.54)	1 (9.09)	7 (63.63)	4 (36.36)	3 (27.27)	4 (36.36)	1 (9.09)	2 (18.18)
Shrimp (6)	2 (33.33)	3 (50)	2 (33.33)	4 (66.66)	4 (66.66)	1 (16.66)	—	4 (66.66)	1 (16.66)	3 (50)	3 (50)	1 (16.66)	1 (16.66)	2 (33.33)	1 (16.66)	1 (16.66)	2 (33.33)	1 (16.66)
Total (20)	8 (40)	10 (50)	6 (30)	14 (70)	15 (75)	2 (10)	1 (5)	14 (70)	3 (15)	9 (45)	11 (55)	2 (10)	10 (50)	7 (35)	5 (25)	6 (30)	3 (15)	4 (20)

the resistance rate of *A. baumannii* strains isolated from raw meat samples toward gentamicin, tetracycline, erythromycin, azithromycin, ciprofloxacin, trimethoprim-sulfamethoxazole, and rifampin was 87.17%, 79.48%, 74.35%, 66.66%, 58.97%, 56.41%, and 51.28%, respectively. Ahmad et al. [54] reported that the prevalence of resistance of *A. baumannii* isolated from meat samples against ampicillin, ceftriaxone, imipenem, gentamicin, kanamycin, tetracycline, chloramphenicol, trimethoprim, sulfamethoxazole, and norfloxacin was 100%, 20.80%, 33.30%, 16.60%, 54.10%, 79.10%, 66.60%, 100%, 8.30%, and 16.60%, respectively. An Egyptian survey [19] showed the highest resistance rate of *A. baumannii* strains against amoxicillin/clavulanic acid (89.10%), gentamicin (74.55%), tetracycline (72.73%), ampicillin (65.45%), and tobramycin (52.73%). The differences reported in the *A. baumannii* phenotypic pattern of antibiotic resistance in various studies are probably due to the availability or nonavailability of antibiotics, the level of strict rules in prescribing antibiotics, and the opinion of physicians and veterinarians on prescribing antibiotics. The prevalence of resistance to imipenem (10%) and chloramphenicol (15%) was lower than that of other antibiotics. Imipenem, a human-prescribed antibiotic in the hospital, is not used in veterinary medicine and also for the treatment of marine animals (in fish, shrimp, and lobster cultures). Thus, it is not surprising that *A. baumannii* strains harbored a low resistance rate against imipenem. Chloramphenicol is also an illicit drug with a limited prescription. The use of this antibiotic illegally is done only in poultry farms in Iran. Thus, it should have a low resistance rate. The final part of the study is aimed at assessing the genotypic pattern of antibiotic resistance amongst the *A. baumannii* strains. Findings showed that bla_{CITM} (encodes resistance against beta-lactams), bla_{SHV} (encodes resistance against beta-lactams), *tetA* (encodes resistance against tetracyclines), *qnrA* (encodes resistance against quinolones), bla_{VIM} (encodes resistance against carbenicillin), and *aac(3)-IV* (encodes resistance against gentamicin) were the most commonly detected antibiotic resistance genes. The high distribution of the genes that encode resistance against diverse classes of antibiotics in the *A. baumannii* strains in this survey revealed the critical role of seafood samples as a possible vehicle for the community-wide dissemination of antibiotic-resistant *A. baumannii* strains. Similarly, Ghazaei [55] reported that the distribution of bla_{SHV}, bla_{TEM}, and bla_{PER} antibiotic resistance genes amongst the *A. baumannii* isolates of foodstuffs was 29.62%, 18.51%, and 14.81%, respectively. Tavakol et al. [46] also identified *aadA1*, *aac(3)-IV*, bla_{SHV}, bla_{CITM}, *cat1*, *cmlA*, *tetA*, *tetB*, *sul1*, *dfrA1*, bla_{IMP}, bla_{SIM}, bla_{VIM}, and *qnrA* antibiotic resistance genes amongst the *A. baumannii* strains of raw meat. In a German study [56], OXA-23, OXA-51, and OXA-58 oxacillinase genes were detected in *A. baumannii* strains isolated from milk powder.

5. Conclusion

In conclusion, *A. baumannii* strains were detected in 4.44% of seafood samples, with the higher prevalence of bacteria amongst the fish samples (7.85%). The considerable prevalence of *A. baumannii* strains was accompanied by the high rate of bacterial resistance toward commonly used antibiotic agents, particularly tetracycline, ampicillin, gentamicin, and erythromycin. The findings may show the high antibiotic resistance of *A. baumannii* and the potential role of seafood samples in its transmission to the human population. Some strains harbored different antibiotic resistance genes, particularly bla_{CITM}, bla_{SHV}, *tetA*, *qnrA*, bla_{VIM}, and *aac(3)-IV*. These findings may show the role of seafood samples as a source of antibiotic resistance genes. It seems that the consumption of contaminated seafood with the resistant *A. baumannii* may cause severe food-borne diseases that resist antibiotic therapy. However, the role of contaminated seafood as a hazard of foodborne infection has not been determined yet. Thus, several studies should perform to assess the role of seafood samples in the transmission of virulent and resistant *A. baumannii* foodborne diseases.

Data Availability

The data that support the findings of this study are available on request from the corresponding author.

Conflicts of Interest

The authors declare no potential conflict of interest.

Authors' Contributions

All authors read and approved the final manuscript.

References

[1] E. Reames, *Nutritional benefits of seafood*, Southern Regional Aquaculture Center, 2012.

[2] D. Jamioł-Milc, J. Biernawska, M. Liput, L. Stachowska, and Z. Domiszewski, "Seafood intake as a method of noncommunicable diseases (NCD) prevention in adults," *Nutrients*, vol. 13, no. 5, p. 1422, 2021.

[3] A. Ali, A. Parisi, M. Conversano et al., "Food-borne bacteria associated with seafoods: a brief review," *Journal of food quality and hazards control*, vol. 7, no. 1, pp. 4–10, 2020.

[4] C. Ayoub Moubareck and H. D. Hammoudi, "Insights into *Acinetobacter baumannii*: a review of microbiological, virulence, and resistance traits in a threatening nosocomial pathogen," *Antibiotics*, vol. 9, no. 3, p. 119, 2020.

[5] S. Poursina, M. Ahmadi, F. Fazeli, and P. Ariaii, "Assessment of virulence factors and antimicrobial resistance among the Pseudomonas aeruginosa strains isolated from animal meat and carcass samples," *Veterinary medicine and science*, vol. 9, no. 1, pp. 315–325, 2023.

[6] C. R. Lee, J. H. Lee, M. Park et al., "Biology of *Acinetobacter baumannii*: pathogenesis, antibiotic resistance mechanisms, and prospective treatment options," *Frontiers in Cellular and Infection Microbiology*, vol. 7, no. 7, p. 55, 2017.

[7] J. Garnacho-Montero and J. F. Timsit, "Managing *Acinetobacter baumannii* infections," *Current Opinion in Infectious Diseases*, vol. 32, no. 1, pp. 69–76, 2019.

[8] A. Carvalheira, J. Silva, and P. Teixeira, "Acinetobacter spp. in food and drinking water-a review," *Food Microbiology*, vol. 95, p. 103675, 2021.

[9] M. Marí-Almirall, C. Cosgaya, M. J. Pons et al., "Pathogenic Acinetobacter species including the novel Acinetobacter dijkshoorniae recovered from market meat in Peru," *International Journal of Food Microbiology*, vol. 305, no. 305, article 108248, 2019.

[10] A. M. Soliman, H. Nariya, D. Tanaka et al., "Vegetable-derived carbapenemase-producing high-risk *Klebsiella pneumoniae* ST15 and *Acinetobacter baumannii* ST2 clones in Japan: coexistence of $bla_{\text{NDM-1}}$, $bla_{\text{OXA-66}}$, $bla_{\text{OXA-72}}$, and an *AbaR4-like* resistance island in the same sample," *Applied and Environmental Microbiology*, vol. 87, no. 9, p. e02166, 2021.

[11] R. Rafei, M. Hamze, H. Pailhoriès et al., "Extrahuman epidemiology of *Acinetobacter baumannii* in Lebanon," *Applied and Environmental Microbiology*, vol. 81, no. 7, pp. 2359–2367, 2015.

[12] S. Dekić, G. Klobučar, T. Ivanković et al., "Emerging human pathogen *Acinetobacter baumannii* in the natural aquatic environment: a public health risk?," *International Journal of Environmental Health Research*, vol. 28, no. 3, pp. 315–322, 2018.

[13] X. Wang, J. Li, X. Cao, W. Wang, and Y. Luo, "Isolation, identification and characterisation of an emerging fish pathogen, *Acinetobacter pittii*, from diseased loach (*Misgurnus anguillicaudatus*) in China," *Antonie Van Leeuwenhoek*, vol. 113, no. 1, pp. 21–32, 2020.

[14] J. Moradi, F. B. Hashemi, and A. Bahador, "Antibiotic resistance of Acinetobacter baumannii in Iran: a systemic review of the published literature," *Osong Public Health and Research Perspectives*, vol. 6, no. 2, pp. 79–86, 2015.

[15] I. Kyriakidis, E. Vasileiou, Z. D. Pana, and A. Tragiannidis, "*Acinetobacter baumannii* antibiotic resistance mechanisms," *Pathogens.*, vol. 10, no. 3, p. 373, 2021.

[16] M. A. Rizk and N. T. Abou El-Khier, "Aminoglycoside resistance genes in *Acinetobacter baumannii* clinical isolates," *Clinical Laboratory*, vol. 65, 2019.

[17] S. Cheng, P. Lu, and Q. Y. Feng, "Distribution of antibiotic resistance genes and microbial communities in a fishery reclamation mining subsidence area," *Huan Jing Ke Xue*, vol. 42, no. 5, pp. 2541–2549, 2021, Chinese.

[18] A. M. Amorim and J. D. Nascimento, "*Acinetobacter*: an underrated foodborne pathogen?," *Journal of Infection in Developing Countries*, vol. 11, no. 2, pp. 111–114, 2017.

[19] A. Elbehiry, E. Marzouk, I. M. Moussa et al., "Acinetobacter baumannii as a community foodborne pathogen: peptide mass fingerprinting analysis, genotypic of biofilm formation and phenotypic pattern of antimicrobial resistance," *Journal of Biological Sciences*, vol. 28, no. 1, pp. 1158–1166, 2021.

[20] R. C. Malta, G. L. Ramos, and J. dos Santos Nascimento, "From food to hospital: we need to talk about *Acinetobacter* spp.," *Germs*, vol. 10, no. 3, p. 210, 2020.

[21] N. Askari, H. Momtaz, and E. Tajbakhsh, "*Acinetobacter baumannii* in sheep, goat, and camel raw meat: virulence and antibiotic resistance pattern," *AIMS microbiology*, vol. 5, no. 3, pp. 272–284, 2019.

[22] A. Lupo, D. Vogt, S. N. Seiffert, A. Endimiani, and V. Perreten, "Antibiotic resistance and phylogenetic characterization of Acinetobacter baumannii strains isolated from commercial raw meat in Switzerland," *Journal of Food Protection*, vol. 77, no. 11, pp. 1976–1981, 2014.

[23] R. Zhang, W. Huo, W. Zhu, and S. Mao, "Characterization of bacterial community of raw milk from dairy cows during subacute ruminal acidosis challenge by high-throughput sequencing," *Journal of the Science of Food and Agriculture*, vol. 95, no. 5, pp. 1072–1079, 2015.

[24] Q. Ababneh, E. Al-Rousan, and Z. Jaradat, "Fresh produce as a potential vehicle for transmission of *Acinetobacter baumannii*," *International Journal of Food Contamination*, vol. 9, no. 1, p. 5, 2022.

[25] S. Brahmi, A. Touati, A. Cadière et al., "First description of two sequence type 2 *Acinetobacter baumannii* isolates carrying OXA-23 carbapenemase in *Pagellus acarne* fished from the Mediterranean Sea near Bejaia, Algeria," *Antimicrobial Agents and Chemotherapy*, vol. 60, no. 4, pp. 2513–2515, 2016.

[26] Y. Mani, W. Mansour, A. Lupo et al., "Spread of $bla_{\text{CTX-M15}}$-producing *Enterobacteriaceae* and OXA-23-producing *Acinetobacter baumannii* sequence type 2 in Tunisian seafood," *Antimicrobial Agents and Chemotherapy*, vol. 62, no. 9, p. e00727, 2018.

[27] Z. Hasiri, E. Rahimi, A. Shakerian, and H. Momtaz, "The first report of the antimicrobial resistance of *Acinetobacter baumannii* isolates from seafood samples," *Egyptian Journal of Veterinary Science*, vol. 52, no. 2, pp. 175–183, 2021.

[28] J. G. Cappuccino and C. T. Welsh, *Microbiology: A Laboratory Manual*, Pearson Education, London, 11th edition, 2016.

[29] M. C. Chiang, S. C. Kuo, Y. C. Chen, Y. T. Lee, T. L. Chen, and C. P. Fung, "Polymerase chain reaction assay for the detection of Acinetobacter baumannii in endotracheal aspirates from patients in the intensive care unit," *Journal of Microbiology, Immunology and Infection*, vol. 44, no. 2, pp. 106–110, 2011.

[30] CLSI, *Performance Standards for Antimicrobial Susceptibility Testing; Twenty-Fifth Informational Supplement*, Clinical and Laboratory Standards Institute, Wayne, 2017.

[31] F. Safarpoor Dehkordi, H. Gandomi, A. A. Basti, A. Misaghi, and E. Rahimi, "Phenotypic and genotypic characterization of antibiotic resistance of methicillin-resistant *Staphylococcus aureus* isolated from hospital food," *Antimicrobial Resistance and Infection Control*, vol. 6, no. 1, p. 104, 2017.

[32] F. Safarpoor Dehkordi, B. Tavakoli-Far, S. Jafariaskari et al., "Uropathogenic Escherichia coli in the high vaginal swab samples of fertile and infertile women: virulence factors, O-serogroups, and phenotyping and genotyping characterization of antibiotic resistance," *New Microbes and New Infections*, vol. 38, no. 38, article 100824, 2020.

[33] C. S. Toro, M. Farfán, I. Contreras, O. Flores, N. Navarro, G. C. Mora et al., "Genetic analysis of antibiotic-resistance determinants in multidrug-resistant Shigella strains isolated from Chilean children," *Epidemiology and Infection*, vol. 133, no. 1, pp. 81–86, 2005.

[34] N. Woodford, M. J. Ellington, J. M. Coelho, J. F. Turton, M. E. Ward, S. Brown et al., "Multiplex PCR for genes encoding prevalent OXA carbapenemases in Acinetobacter spp.," *International Journal of Antimicrobial Agents*, vol. 27, no. 4, pp. 351–353, 2006.

[35] H. Mammeri, M. Van De Loo, L. Poirel, L. Martinez-Martinez, and P. Nordmann, "Emergence of plasmid-mediated quinolone resistance in *Escherichia coli* in Europe," *Antimicrobial Agents and Chemotherapy*, vol. 49, no. 1, pp. 71–76, 2005.

[36] R. E. Mendes, K. A. Kiyota, J. Monteiro et al., "Rapid detection and identification of metallo-β-lactamase-encoding genes by multiplex real-time PCR assay and melt curve analysis," *Journal of Clinical Microbiology*, vol. 45, no. 2, pp. 544–547, 2007.

[37] R. Ranjbar, F. Yadollahi Farsani, and D. F. Safarpoor, "Antimicrobial resistance and genotyping of *vacA*, *cagA*, and *iceA* alleles of the Helicobacter pylori strains isolated from traditional dairy products," *Journal of Food Safety*, vol. 39, no. 2, article e12594, 2019.

[38] S. Nejat, H. Momtaz, M. Yadegari, S. Nejat, F. Safarpour Dehkordi, and F. Khamesipour, "Seasonal, geographical, age and breed distributions of equine viral arteritis in Iran," *Kafkas Üniversitesi Veteriner Fakültesi dergisi*, vol. 21, no. 1, pp. 111–116, 2015.

[39] E. Kabiri and Z. Mashak, "Antimicrobial resistance properties and enterotoxigenic gene profile of methicillin-resistant and methicillin-susceptible *Staphylococcus aureus* isolates from raw milk," *Journal of Positive School Psychology*, vol. 6, no. 8, pp. 1244–1252, 2022.

[40] F. S. Dehkordi, M. R. Borujeni, E. Rahimi, and R. Abdizadeh, "Detection of *Toxoplasma gondii* in raw caprine, ovine, buffalo, bovine, and camel milk using cell cultivation, cat bioassay, capture ELISA, and PCR methods in Iran," *Foodborne Pathogens and Disease*, vol. 10, no. 2, pp. 120–125, 2013.

[41] H. Tate, S. Ayers, E. Nyirabahizi et al., "Prevalence of antimicrobial resistance in select bacteria from retail seafood-United States, 2019," *Frontiers in Microbiology*, vol. 13, no. 13, article 928509, 2022.

[42] B. K. Behera, P. Paria, A. Das, S. Bhowmick, A. K. Sahoo, and B. K. Das, "Molecular characterization and pathogenicity of a virulent Acinetobacter baumannii associated with mortality of farmed Indian major carp Labeo rohita (Hamilton 1822)," *Aquaculture*, vol. 471, pp. 157–162, 2017.

[43] P. R. Rauta, K. Kumar, and P. K. Sahoo, "Emerging new multidrug resistant bacterial pathogen, *Acinetobacter baumannii* associated with snakehead *Channa striatus* eye infection," *Current Science*, vol. 101, pp. 548–553, 2011.

[44] A. Kozińska, E. Paździor, A. Pękala, and W. Niemczuk, "*Acinetobacter johnsonii* and *Acinetobacter lwoffii*-the emerging fish pathogens," *Journal of Veterinary Research*, vol. 58, no. 2, pp. 193–199, 2014.

[45] N. Askari, H. Momtaz, and E. Tajbakhsh, "Prevalence and phenotypic pattern of antibiotic resistance of Acinetobacter baumannii isolated from different types of raw meat samples in Isfahan, Iran," *Veterinary Medicine and Science*, vol. 6, no. 1, pp. 147–153, 2020.

[46] M. Tavakol, H. Momtaz, P. Mohajeri, L. Shokoohizadeh, and E. Tajbakhsh, "Genotyping and distribution of putative virulence factors and antibiotic resistance genes of *Acinetobacter baumannii* strains isolated from raw meat," *Antimicrobial Resistance and Infection Control*, vol. 7, no. 1, p. 120, 2018.

[47] H. Kim, Y. Hong, J. Jo et al., "Raw ready-to-eat seafood safety: microbiological quality of the various seafood species available in fishery, hyper and online markets," *Letters in Applied Microbiology*, vol. 64, no. 1, pp. 27–34, 2017.

[48] S. Armin, A. Karimi, F. Fallah et al., "Antimicrobial resistance patterns of *Acinetobacter baumannii*, *Pseudomonas aeruginosa* and *Staphylococcus aureus* isolated from patients with nosocomial infections admitted to Tehran hospitals," *Archives of Pediatric Infectious Diseases*, vol. 3, no. 4, article e32554, 2015.

[49] E. A. Ribeiro, A. C. Gales, A. P. Oliveira et al., "Molecular epidemiology and drug resistance of *Acinetobacter baumannii* isolated from a regional hospital in the Brazilian Amazon region," *Revista da Sociedade Brasileira de Medicina Tropical*, vol. 54, p. e20200087, 2021.

[50] A. Batarseh, A. Al-Sarhan, M. Maayteh, S. Al-Khatirei, and M. Alarmouti, "Antibiogram of multidrug resistant *Acinetobacter baumannii* isolated from clinical specimens at King Hussein Medical Centre, Jordan: a retrospective analysis," *Eastern Mediterranean Health Journal*, vol. 21, no. 11, pp. 828–834, 2015.

[51] M. D. Zilberberg, M. H. Kollef, and A. F. Shorr, "Secular trends in *Acinetobacter baumannii* resistance in respiratory and blood stream specimens in the United States, 2003 to 2012: a survey study," *Journal of Hospital Medicine*, vol. 11, no. 1, pp. 21–26, 2016.

[52] K. Ko, J. Suh, K. Kwon et al., "High rates of resistance to colistin and polymyxin B in subgroups of *Acinetobacter baumannii* isolates from Korea," *Journal of Antimicrobial Chemotherapy*, vol. 60, no. 5, pp. 1163–1167, 2007.

[53] N. Nogbou, D. Phofa, M. Nchabeleng, and A. Musyoki, *Genetic determinants of multi-drug resistance in Acinetobacter baumannii at an academic hospital in Pretoria, South Africa*, bioRxiv, 2019.

[54] A. Ahmad, F. Akhtar, A. A. Sheikh, M. Y. Tipu, and M. Nasar, "Comparative antibiotic resistance profile of *Acinetobacter* species, isolated from fish, chicken and beef meat," *Journal of Microbiology and Antimicrobials*, vol. 2, no. 1, p. 1, 2018.

[55] C. Ghazaei, "Characterization, Prevalence and antibiotic resistance of *Acinetobacter baumannii* Infecting Foodstuff," *Research in Molecular Medicine*, vol. 7, no. 4, pp. 25–32, 2019.

[56] G. S. Cho, B. Li, A. Rostalsky et al., "Diversity and antibiotic susceptibility of *Acinetobacter* strains from milk powder produced in Germany," *Frontiers in Microbiology*, vol. 9, no. 9, p. 536, 2018.