Original Article | Iran J Med Microbiol. 2022; 16(6): 566-572

10.30699/ijmm.16.6.566

Iranian Journal of Medical Microbiology | ISSN:2345-4342

Prevalence of the *per*, *tem*, *veb*, *shv* genes in *Acinetobacter baumannii* Isolated from Educational Hospital of Zahedan, Iran

Ramin Ordoni[1], Yousef Amin[1,2], Morad Ali Ranayi[1], Zoleikha Avestan[3], Amir Hossein Mohagheghi-Fard[1], Mehdi Zandhaghighi[1], Zakaria Bameri[1,2], Ebrahim Kord[1,2], Shahram Shahraki-Zahedani[1,2*], Aslam Dehvari[1]

1. *Department of Microbiology, School of Medicine, Zahedan University of Medical Sciences, Zahedan, Iran*
2. *Infectious Diseases and Tropical Medicine Research Center, Resistant Tuberculosis Institute, Zahedan University of Medical Sciences, Zahedan, Iran*
3. *Community Nursing Research Center, Zahedan University of Medical Sciences, Zahedan, Iran*

ABSTRACT

Background and Aim: *Acinetobacter baumannii* is one of the most important pathogens found in ICUs as it is responsible for nosocomial infection with a high mortality rate and a rising level of resistance to most antibiotics. One of the main mechanisms of resistance is the production of extended-spectrum beta-lactamase (ESBL) genes such as *veb*, *per*, *tem*, and *shv*. The current study aimed to determine the antibiotic resistance pattern and the frequency of *per*, *tem*, *veb*, and *shv* genes in *A. baumannii* strains isolated from Zahedan, Iran.

Materials and Methods: The current study was conducted on 150 strains of *A. baumannii* isolated from different clinical samples from January to September 2018. The antibiotic resistance pattern was determined by the disk diffusion method for 18 antibiotics and the minimum inhibitory concentration for colistin. Later, the bacterial genome was extracted, and PCR detected ESBL genes.

Results: Out of the 150 studied isolates, 141 were *A. baumannii* and only nine isolates were *A. nosocomialis*. *A. baumannii* strains were strongly resistant to many selected antibiotics such as CAZ (99.3%), CTX (97.9), PTZ (97.2%), SXT (97.2%), IMI (97.2%), and LEV (96.5%); while this value was low for a few antibiotics including MN (69.5%), DXT (52.5%), SAM (21.3), and TN (16.7). The *eb*, *per*, *tem*, and *shv* genes were detected in 20 (13.3%), 15 (10%), 23 (15.4%), and 11 (7.3%) isolates, respectively.

Conclusion: The isolates were highly resistant to ceftazidime, ceftriaxone, piperacillin-tazobactam, cotrimoxazole, imipenem, and levofloxacin; in addition, the frequency of *per, veb, tem*, and genes was lower in the current study than those reported in other regions.

Keywords: *Acinetobacter baumannii*, Extended-spectrum beta-lactamase, Antibiotic resistance

Received: 2021/12/12; **Accepted**: 2022/07/07; **Published Online:** 2022/09/09

Corresponding Information: **Shahram Shahraki-zahedani**, *Department of Microbiology, School of Medicine, Zahedan University of Medical Sciences, Zahedan, Iran* Email: shahram17ir@yahoo.com

Use your device to scan and read the article online

Ordoni R, Amini Y, Ranayi M A, Avestan Z, Mohagheghi-Fard A H, et al. Prevalence of the *per, tem, veb, shv* genes in *Acinetobacter baumannii* isolated from educational hospital of Zahedanm Iran. Iran J Med Microbiol. 2022; 16 (6):566-72.

Download citation: BibTeX | RIS | EndNote | Medlars | ProCite | Reference Manager | RefWorks

Send citation to: Mendeley Zotero RefWorks

1. Introduction

Acinetobacter spp. complex includes species with varying levels of clinical importance, of which *Acinetobacter baumannii* and *A. nosocomialis* are of particular significance **(1)**. However, they have different associations with human health **(2)**. *A. baumannii* is one of the most important nosocomial pathogens due to its longevity in the hospital environment and resistance to various antimicrobial agents, giving it the ability to colonize susceptible patients treated with broad-spectrum antibiotics **(3, 4)**. *Acinetobacter baumannii* is involved in different infections such as bacteremia, urinary tract infection,

endocarditis, pneumonia, secondary meningitis, surgical wound infection, skin and soft tissue infection, and septicemia **(5-7)**. As of today, *A. baumannii* has developed the ability to withstand different classes of antibiotic components such as beta-lactam, carbapenem, and aminoglycosides **(8-10).** Different types of beta-lactamases have been identified based on their characteristics and activities **(11)**. Extended-spectrum beta-lactamases (ESBLs) are a group of enzymes that can hydrolyze beta-lactam antibiotics, including penicillin, expanded-spectrum cephal-osporins, fourth generation cephalosporins (cefepime), and aztreonam **(12)**. The most important enzymes initially identified in 1980 were *shv* and *tem* (beta-lactamase genes). In addition to the main families, new families of ESBLs, including *per, veb, bel, tla, ges,* and *bes* have emerged globally. In fact, the rapid transmission and dissemination of these genes has increased the prevalence of nosocomial infections **(13, 14)**. The present study aimed to investigate the prevalence of *per, veb, tem, and shv* genes and their relationship with antimicrobial resistance among *A. baumannii* strains isolated from patients referred to educational hospitals in Zahedan, Iran.

2. Materials and Methods

2.1 Bacterial Isolation and Identification:

A total of 150 non-duplicate isolates of *A. baumannii* were collected from two hospitals (Ali-ibn-Abi-Talib and Khatam) in Zahedan, southeast of Iran, from January to September 2018. The sources of the bacterial isolates were human urine, blood, wound, and bronchoalveolar lavage fluid. Medical microbiological tests including Gram staining, growth at 44°C, oxidase test, O/F test and conventional biochemical methods were used to identify *A. baumannii* isolates **(15)**. Moreover, Molecular differentiation of ABC complex was investigated by presence of *bla*oxa-51-like family and gyrB multiplex PCR.

2.2 Antimicrobial Susceptibility

Kirby Bauer disk diffusion method was used to find susceptibility for Amikacin (30µg), Gentamycin (10µg), Minocycline (30µg), Doxycycline (30µg), Tetracycline (25µg), Ampicillin-sulbactam (20µg), Piperacillin (100µg), Piperacillin beta-lactamase (100/10µg), Cefepime (30µg), Levofloxacin (5µg), Cotrimoxazole (25µg), Imipenem (10µg), Meropenem (10µg), Tobramycin (10µg), Cefotaxime (30µg), Cefazolin (30µg), and Ciprofloxacin (5µg). According to the guidelines (CLSI, 2018), the diameter of inhibition zones was determined to show resistance, intermediate, and sensitivity of each isolate against antibiotics. *Escherichia coli* ATCC25922 and *Pseudomonas aeruginosa* ATCC27853 were used as the control strains.

2.3 Determination of the Minimum Inhibitory Concentration (MIC)

The MIC of Colistin was determined for all *A. baumannii* species isolates using the disk diffusion test and broth microdilution method according to the guidelines of the CLSI **(16)**. Escherichia coli ATCC 25922 was used as the reference strain.

2.4 DNA Extraction and PCR Amplification:

As previously described, the boiling method was used to extract genomic DNA **(17)**. DNA extraction concentration and quality were checked with 1.5% agarose gel and Nanodrop. The presence of *oxa-51*, *spf-2*, *spf-4*, *per*, *veb*, *tem*, and *shv* genes in *A. baumannii* strains were detected by PCR. The PCR was performed in a final volume of 20 µL containing 1 µL of isolate genomic DNA (~70 ng/mL), 10 µL of master mix (Ampliqon Taq 2x Master Mix, Denmark), 1.0 µL of each primer 10 ng/mL (Table 1)**,** and 6 µL of distilled water (SinaClon Bio-Science Co., Tehran, Iran), The PCR products were analyzed by electrophoresed on agarose gel and then observed with a transilluminator under UV light.

2.5 statistical Analyses

Statistical analyses were performed using SPSS software version 22.0 (SPSS, Chicago, IL, USA).

Table 1. The primers used in this study

Genes	Primers	Sequence (5′ to 3′)	fragments	Annealing Temp	Reference
oxa-51	F	TAATGCTTTGATCGGCCTTG	352bp	52	**(17)**
	R	TGGATTGCACTTCATCTTGG			
spf-2	F	GTTCCTGATCCGAAATTCTCG	294bp	60	**(18)**
	R	AACGGAGCTTGTCAGGGTTA			
spf-4	F	CACGCCGTAAGAGTGCATTA	490bp	60	**(18)**
	R	AACGGAGCTTGTCAGGGTTA			
veb	F	CGACTTCCATTTCCCGATGC	643bp	58	**(19)**
	R	GGACTCTGCAACAAATACGC			
per	F	GCAACTGCTGCAATACTCGG	340bp	56	**(19)**
	R	ATGTGCGACCACAGTACCAG			
tem	F	TTTCGTGTCGCCCTTATTCC	404bp	57	**(20)**
	R	ATCGTTGTCAGAAGTAAGTTGG			
shv	F	CGCCTGTGTATTATCTCCCT	290bp	57	**(20)**
	R	CGAGTAGTCCACCAGATCCT			

3. Results

3.1 Patients and Bacterial Isolation

The isolates were used in the previous study **(15)**. As reported previously, 141 *A. baumannii* strains were isolated from bronchoalveolar lavage fluid (50.4%), blood (39.7%), urine (2.1%), and wound (7.8%) samples. The samples were obtained from patients with a mean age of 40.77±23.49 years, of whom 86 (61%) were male and 55 (39%) female.

3.2 Molecular Identification of Acinetobacter species

Biochemical tests showed that all isolates belonged to *A. baumannii* species. As reported previously, *A. baumannii* species was confirmed by positive for *oxa-51* gene *PCR*. Moreover, *A. baumannii* and *A. nosocomial* were differentiated by *spf-2* and *spf-4* amplifications. Among all positive isolates, 100% of isolates were positive for *spf-2,* whereas 141 out of 150 isolates were positive for another *spf* (*spf-4*) showing there are 141 *A. baumannii* and only nine *A. nosocomial* **(15)**.

3.3 Antibiotic susceptibility testing

As reported previously, the highest levels of resistance were observed against cefazolin (100%), aztreonam and ceftazidime (98.6%), piperacillin, and cefotaxime (97.9%). Moreover, the frequency of MDR and extensively drug-resistant (XDR) isolates was 97.2 and 41.1%), respectively. Since all isolates were susceptible to colistin (MIC ≤ 2 µg/mL), we did not have pan-drug-resistant (PDR) isolates **(15)**.

3.4 ESBLs Genotypes

Molecular analysis showed that 15.4% and 7.3% of isolates harbored *tem* and *shv,* and 13.3% and 10% of them carried *veb* and *per* genes, respectively (Figure.1). Combination (Com) analyses between *veb* and *per* genes showed that three isolates were positive for both genes (Com^{-1}, *veb*$^{+}$, *per*$^{+}$) and 118 isolates lacked them (Com^{-4}, *veb*$^{-}$, *per*$^{-}$). The current study results showed that 17 and 12 samples were only positive for *veb* (Com^{-2}, *veb*$^{+}$, *per*$^{-}$) and *per* (Com^{-3}, *veb*$^{-}$, *per*$^{+}$), respectively. Analyses between *tem* and *shv* genes showed that four isolates were positive for both genes (Com^{-5}, *shv*$^{+}$, *tem*$^{+}$) and 120 isolates lacked them (Com^{-4}, *shv*$^{-}$, *tem*$^{-}$). Molecular analysis showed that 7 and 19 samples were positive only for *shv* (Com^{-6}, *shv*$^{+}$, *tem*) and *tem* (Com^{-7}, *shv*$^{-}$, *tem*$^{+}$), respectively. Moreover, the relation between gene frequency and antimicrobial agents was investigated. There was no significant correlation between resistance genes and Antibiotic agents ($p > 0.05$; Table.2).

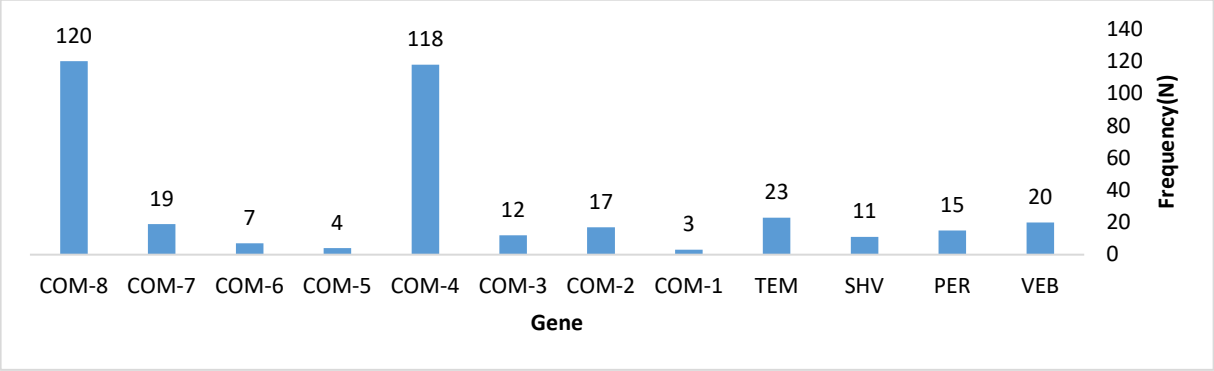

Figure 1. Distribution of *per*, *veb*,*tem* ,*shv* and their combinations genes in *A. baumannii* species isolates.(COM-1: *veb*$^{+}$, *per*$^{+}$, COM-2: *veb*$^{+}$, *per*$^{-}$, COM-3: *veb* $^{-}$, *per*$^{+}$, COM-4: *veb*$^{-}$, *per*$^{-}$, COM-5:shv^{+}, tem^{+}, COM-6: shv^{+}, tem^{-} , COM-7: shv^{-}, tem^{+}, COM-8: shv^{-}, tem^{-})

Table 2. Relation between gene frequency and antimicrobial agents in *A. baumannii* species isolates (R= Resistance, I= Intermediate, S= Sensitive, P= Positive, N= Negative

Antimicrobial agents		Mn(30µg)			DXT(30µg)			T(25µg)			SAM(20 µg			PRL(100µg)			PTZ(100 µg)			CPM(30 µg)		
		R	I	S	R	I	S	R	I	S	R	I	S	R	I	S	R	I	S	R	I	S
veb	P	5	3	12	10	0	10	15	0	5	13	3	4	19	1	0	19	0	1	18	1	1
	N	19	12	99	53	12	65	90	22	18	74	34	22	125	3	2	126	1	3	117	4	9
per	P	3	2	10	8	1	6	12	1	2	10	3	2	14	1	0	15	0	0	14	0	1
	N	21	13	101	55	11	69	93	21	12	77	34	24	130	3	2	130	1	4	121	5	9
shv	P	3	1	7	6	1	4	8	1	2	7	3	1	10	1	0	11	0	0	10	0	1
	N	15	6	118	62	9	68	98	18	23	81	32	26	132	4	1	133	1	5	125	5	9
tem	P	5	4	14	13	2	8	12	4	7	12	5	6	16	7	0	21	0	2	20	0	3
	N	18	11	98	53	9	66	82	20	25	72	32	23	121	4	2	118	2	7	117	3	7

Antimicrobial agents		Lev(5 µg)			SXT(25 µg)			IMI(10 µg)			MEM(10 µg)			TN(10 µg)			CTX(30 µg)			CAZ(30 µg)			CP(5 µg)		
		R	I	S	R	I	S	R	I	S	R	I	S	R	I	S	R	I	S	R	I	S	R	I	S
veb	P	19	0	1	20	0	0	19	0	1	20	0	0	17	1	2	20	0	0	20	0	0	19	0	1
	N	124	3	3	123	2	5	117	0	13	115	2	13	101	6	23	130	0	0	128	2	0	118	0	12
per	P	15	0	0	15	0	0	13	0	2	14	1	0	11	2	2	15	0	0	15	0	0	14	0	1
	N	128	3	4	128	2	5	123	0	12	121	1	13	107	5	23	135	0	0	133	0	2	113	0	10
shv	P	11	0	0	11	0	0	10	0	1	9	2	0	8	2	1	10	1	0	11	0	0	8	0	3
	N	133	8	9	134	7	9	135	0	15	132	5	13	118	5	27	136	2	12	139	7	4	128	5	17
tem	P	23	0	0	21	0	2	21	0	2	17	1	5	16	3	4	22	1	0	23	0	0	17	3	3
	N	131	8	11	135	9	6	138	0	12	133	3	14	117	8	25	141	5	4	144	0	6	135	2	13

4. Discussion

Acinetobacter baumannii is considered one of the seriously health-threatening pathogens, which causes many infections such as pneumonia, urinary tract infections, wound infections, bacteremia, and meningitis **(6, 21, 22)**. One of the main concerns in treating A. baumannii is its resistance to a wide range of antibiotics, including the latest treatment options Beta-lactamase and carbapenems, caused by the overuse of these drugs. Detection of ESBL genes involved in resistance is important for treatment and epidemiology studies **(22)**. Molecular methods such as PCR determine which ESBL gene exists in the isolate **(12)**. In this study, the highest number of isolates was obtained from ICU. Moreover, *A. baumannii* was mostly found in trachea samples. Almost similar results were observed in a study by Sana Islahi et al. in India **(23).** In the current study, almost 93.6% of the *Acinetobacter* strains isolated were shown to be *A. baumannii*, whereas *A. nosocomialis* only constituted 6.3% (nine out of 150 samples). *A. baumannii* appears to be an important cause of infections **(24)**. According to the study results by Ardebili et al. 81.3% and 18.7% of the collected isolates were *A. baumannii* and *A. nosocomial*, respectively **(25)**. In this study, all 141 *A. baumannii* isolates were susceptible to colistin (MIC ≤2 µg/ml), and only 20 isolates (14.2%) were resistant to Minocycline. Our results also suggest that colistin can be a proper choice against *A. baumannii* isolates *in vitro*, which is in line with the findings of other studies **(21, 26, 27),** which report colistin and tigecycline as the most effective antibiotics against *A. baumannii* isolates **(4)**. In another study in Iran, Sharif et al. concluded that more than 90% of *A. baumannii* isolates were resistant to Ceftadizim, Ceftriaxone, and Cefepime. Similar to our study, the resistance to imipenem was determined to be 95%, with no reported colistin resistance **(28)**. In the present study, the resistance rate against imipenem is in line with the findings presented by Farajnia et al. in 2013 (>80%) **(26)** and is in contrast with Hujer et al. in 2006 (20%)- **(29)**. The determined resistance rate of meropenem (92%) is in contrast with the findings (25%) of Hujer et al. **(29)**. The congruent findings noted above may be due to similar research processes, while the conflicting results can be due to using different types of samples, the type of antibiotic disks used and variation in performing the antibiotic susceptibility test. Molecular analysis showed that 15.4 %, 7.3 %, 13.3%, and 10% of the isolates harbored *tem, shv, veb*, and per genes, respectively (Figure.1). In our study, the highest frequency of ESBLs gene isolates belonged to the ICU and internal ward, respectively. Interestingly, the samples isolated from ETT (Endotracheal) showed the highest rate of the studied gene, followed by and blood culture. One of the reasons for this observation could be the higher use of β-lactam antibiotics to treat infections and the consequent increased expression of β-lactam resistant genes in the strains isolated from these samples. In a study in Iran Fallah et al. showed that out of 91 *A. baumannii* isolates, 78.03% and 39.5% were positive for per and *veb* genes, respectively. Their results showed remarkable differences from those of the current study, which may be due to different regions of studies **(20)**. In another study performed in the Northwest of Iran, Farajnia et al. **(26)** found that only 10% of the 100 isolates of *A. baumannii* were positive for *veb*, while 51% were positive for per. Furthermore, the antimicrobial resistance pattern showed that A. baumannii strains were highly resistant to cefepime (88%), gentamicin (78%), ciprofloxacin (78%), levofloxacin (76%), meropenem (63%), imipenem (62%), and ampicillin-sulbactam (55%) **(30)**. In a study in Korea, Yang et al. detected 53 per+ *A. baumannii* strains out of 97 isolates examined **(27)**. Dai et al., reported that among 39 *A. baumannii* isolates, two carried *shv* gene, while 15 strains harbored *tem* **(30)**. Interestingly, the frequency of *shv* in the current study was significantly different from that reported by Taherikalani et al., which did not detect *shv* in any of the studied isolates **(31)**.

The present study showed high antimicrobial resistance among isolated *A. baumannii* in Zahedan, Southeast Iran. Although the prevalence of per, *veb*, *tem*, and *shv* genes were less than those reported from other regions, it is necessary to review the prescription pattern of antibiotic agents.

5. Conclusion

These results indicate that ESBL genes play a special role in creating antimicrobial resistance among the studied strains of *A. baumannii*. These findings underscore the need for antimicrobial monitoring to control the spread of resistance.

Acknowledgment

The authors would like to thank the University of Medical Sciences, Zahedan, Iran, for supporting this project.

Funding

This study was supported by the Research Deputy of Zahedan University of Medical Sciences (Grant No. 1397.276 IR.ZAUMS.REC).

Conflict of Interest

Non-declared.

Reference

1. Wang X, Chen T, Yu R, Lü X, Zong Z. Acinetobacter pittii and Acinetobacter nosocomialis among clinical isolates of the Acinetobacter calcoaceticus-baumannii complex in Sichuan, China. Diagn Microbiol Infect Dis. 2013;76(3): 392-5. [DOI:10.1016/j.diagmicrobio.2013.03.020] [PMID]
2. Molina J, Cisneros JM, Fernández-Cuenca F, Rodríguez-Baño J, Ribera A, Beceiro A, et al. Clinical features of infections and colonization by Acinetobacter genospecies 3. J Clin Microbiol. 2010;48(12):4623-6. [DOI:10.1128/JCM.01216-10] [PMID] [PMCID]
3. Boo TW, Walsh F, Crowley B. Molecular characterization of carbapenem-resistant Acinetobacter species in an Irish university hospital: predominance of Acinetobacter genomic species 3. J Med Microbiol. 2009;58(2): 209-16. [DOI:10.1099/jmm.0.004911-0] [PMID]
4. Meshkat Z, Salimizand H, Amini Y, Khakshoor M, Mansouri D, Farsiani H, et al. Molecular characterization and genetic relatedness of clinically Acinetobacter baumanii isolates conferring increased resistance to the first and second generations of tetracyclines in Iran. Ann Clin Microbiol Antimicrob. 2017;16(1):1-6. [DOI:10.1186/s12941-017-0226-9] [PMID] [PMCID]
5. Turton JF, Woodford N, Glover J, Yarde S, Kaufmann ME, Pitt TL. Identification of Acinetobacter baumannii by Detection of the blaOXA-51-like Carbapenemase Gene Intrinsic to This Species. J Clin Microbiol. 2006;44(8):2974-6. [DOI:10.1128/JCM.01021-06] [PMID] [PMCID]
6. Safari M, Nejad ASM, Bahador A, Jafari R, Alikhani MY. Prevalence of ESBL and MBL encoding genes in Acinetobacter baumannii strains isolated from patients of intensive care units (ICU). Saudi J Biol Sci. 2015;22(4):424-9. [DOI:10.1016/j.sjbs.2015.01.004] [PMID] [PMCID]
7. Abdi HA, Hormozi B, Najimi M, Noorzehi N. High prevalence of iron acquisition genes among Acinetobacter baumannii strains isolated from patients with urinary tract infections in southeast of Iran. Avicenna J Clin Microbiol Infect. 2015;2(4). [DOI:10.17795/ajcmi-30656]
8. Rezai M, Rafiei A, Ahangarkani F, Bagheri-Nesami M, Nikkhah A, Shafahi K. Emergence of extensively drug resistant acinetobacter baumannii-encoding integrons and extended-spectrum beta-lactamase genes isolated from ventilator-associated pneumonia patients. Jundishapur J Microbiol. 2017;10(7). [DOI:10.5812/jjm.14377]
9. Salimizand H, Zomorodi AR, Mansury D, Khakshoor M, Azizi O, Khodaparast S, et al. Diversity of aminoglycoside modifying enzymes and 16S rRNA methylases in Acinetobacter baumannii and Acinetobacter nosocomialis species in Iran; wide distribution of aadA1 and armA. Infect Genet Evol. 2018;66:195-9. [DOI:10.1016/j.meegid.2018.09.028] [PMID]
10. Meshkat Z, Amini Y, Sadeghian H, Salimizand H. ISAba1/bla OXA-23-like family is the predominant cause of carbapenem resistance in Acinetobacter baumannii and Acinetobacter nosocomialis in Iran. Infect Genet Evol. 2019;71: 60-6. [DOI:10.1016/j.meegid.2019.03.009] [PMID]
11. Thirapanmethee K. Extended spectrum β-lactamases: critical tools of bacterial resistance. Mahidol Univ J Pharm Sci. 2012;39(1):1-8.
12. Naas T, Poirel L, Nordmann P. Minor extended-spectrum β-lactamases. Clin Microbiol Infect. 2008;14(1):42-52. [PMID] [DOI:10.1111/j.1469-0691.2007.01861.x]
13. Higgins PG, Lehmann M, Wisplinghoff H, Seifert H. gyrB multiplex PCR to differentiate between

Acinetobacter calcoaceticus and Acinetobacter genomic species 3. J Clin Microbiol. 2010;48(12): 4592-4. [DOI:10.1128/JCM.01765-10] [PMID] [PMCID]

14. Higgins PG, Wisplinghoff H, Krut O, Seifert H. A PCR-based method to differentiate between Acinetobacter baumannii and Acinetobacter genomic species 13TU. Clin Microbiol Infect. 2007;13(12):1199-201. [DOI:10.1111/j.1469-0691.2007.01819.x] [PMID]
15. Ranaei MA, Shahraki-Zahedan S, Mohagheghi-Fard AH, Salimizand H, Ordoni R, Amini Y. Prevalence of the blaCTX-M and antibiotic resistance pattern among clinical isolates of Acinetobacter baumannii isolated from Zahedan, Southeast Iran. Gene Reports. 2020;19:100626. [DOI:10.1016/j.genrep.2020.100626]
16. Cockerill FR. Performance standards for antimicrobial susceptibility testing: twenty-second informational supplement. Clin Labo Standards Institute. 2010:M100-S22.
17. Yan Z-Q, Shen D-X, Cao J-R, Chen R, Wei X, Liu L-P, et al. Susceptibility patterns and molecular epidemiology of multidrug-resistant Acinetobacter baumannii strains from three military hospitals in China. Int J Antimicrob Agents. 2010;35(3):269-73. [DOI:10.1016/j.ijantimicag.2009.10.016] [PMID]
18. Woodford N, Ellington MJ, Coelho JM, Turton JF, Ward ME, Brown S, et al. Multiplex PCR for genes encoding prevalent OXA carbapenemases in Acinetobacter spp. Int J Antimicrob Agents. 2006;27(4):351-3. [DOI:10.1016/j.ijantimicag.2006.01.004] [PMID]
19. Higgins PG, Wisplinghoff H, Krut O, Seifert H. A PCR-based method to differentiate between Acinetobacter baumannii and Acinetobacter genomic species 13TU. Clin Microbiol Infect. 2007;13(12):1199-201. [DOI:10.1111/j.1469-0691.2007.01819.x] [PMID]
20. Fallah F, Noori M, Hashemi A, Goudarzi H, Karimi A, Erfanimanesh S, et al. Prevalence of blaNDM, blaPER, blaVEB, blaIMP, and blaVIM genes among Acinetobacter baumannii isolated from two hospitals of Tehran, Iran. Scientifica (Cairo). 2014;2014:245162. [DOI:10.1155/2014/245162] [PMID] [PMCID]
21. Bali E, Sultan N, Açık L. Phenotypic and molecular characterization of SHV, TEM, CTX-M and extended-spectrum β-lactamase produced by Escherichia coli, Acinobacter baumannii and Klebsiella isolates in a Turkish hospital. Afr J Microbiol Res. 2010;4(8):650-4.
22. Visca P, Seifert H, Towner KJ. Acinetobacter infection-an emerging threat to human health. IUBMB life. 2011;63(12):1048-54. [DOI:10.1002/iub.534] [PMID]
23. Islahi S, Ahmad F, Khare V, Mishra N, Yaqoob S, Shukla P, et al. Prevalence and resistance pattern of Acinetobacter species in hospitalized patients in a tertiary care centre. J Evol Med Dent Sci. 2014; 3(17):4629-36. [DOI:10.14260/jemds/2014/2487]
24. Tripathi PC, Gajbhiye SR, Agrawal GN. Clinical and antimicrobial profile of Acinetobacter spp: An emerging nosocomial superbug. Adv Biomed Res.2014;3(13). [PMID] [PMCID] [DOI:10.4103/2277-9175.124642]
25. Ardebili A, Azimi L, Mohammadi-Barzelighi H, Beheshti M, Talebi M, Jabbari M, et al. Determination of resistance pattern of isolated Acinetobacter baumannii from hospitalized burned patients in Motahari Hospital, Tehran. J Adv Med Biomed Res. 2012;20(83):112-9.
26. Farajnia S, Azhari F, Alikhani MY, Hosseini MK, Peymani A, Sohrabi N. Prevalence of PER and VEB type extended spectrum betalactamases among multidrug resistant Acinetobacter baumannii isolates in North-West of Iran. Iran J Basic Med Sci. 2013;16(6):751-5.
27. Yong D, Shin JH, Kim S, Lim Y, Yum JH, Lee K, et al. High prevalence of PER-1 extended-spectrum β-lactamase-producing Acinetobacter spp. in Korea. Antimicrob Agents Chemother. 2003;47 (5):1749-51. [PMID] [PMCID] [DOI:10.1128/AAC.47.5.1749-1751.2003]
28. Sharif M, Mirnejad R, Amirmozafari N. Molecular identification of TEM and SHV extended spectrum βlactamase in clinical isolates of Acinetobacter baumannii from Tehran hospitals. J Gen Microb Immun. 2014:1-9. [DOI:10.5899/2014/jgmi-00020]
29. Hujer KM, Hujer AM, Hulten EA, Bajaksouzian S, Adams JM, Donskey CJ, et al. Analysis of Antibiotic Resistance Genes in Multidrug-Resistant Acinetobacter sp. Isolates from Military and Civilian Patients Treated at the Walter Reed Army Medical Center. Antimicrob Agents Chemother. 2006;50(12):4114-23. [DOI:10.1128/AAC.00778-06] [PMID] [PMCID]
30. Dai N, Li D-z, Chen J-c, Chen Y-s, Geng R, Hu Y-h, et al. Drug-resistant genes carried by Acinetobacter baumanii isolated from patients with lower respiratory tract infection. Chin Med J. 2010;123(18):2571-5.
31. Taherikalani M, Sekawi Z, Azizi-Jalilian F, Keshavarz B, Soroush S, Akbari M, et al.

Distribution of extended spectrum beta-lactamase resistance genes among nosocomial imipenem resistant A. Baumannii strains harboring BLAoxa-23 carbapenemases isolated from Ilam and Tehran. J Biol Regul Homeost Agents. 2013;27(3):883-9.