Research Article

Investigation of the relationship between Antibiotic resistance pattern and class 1, 2 , 3 Integrons in Acinetobacter baumannii isolated from clinical samples of ICU patients

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ABSTRACT: Acinetobacter baumannii, an opportunistic pathogen with high antibiotic resistance, can cause various infections, especially among hospitalized patients in burn and surgery intensive care units. This study is to investigate the relationship between Antibiotic resistance pattern and class 1, 2 and 3 Integrons in Acinetobacter baumannii isolated from clinical samples of ICU patients of Dr. Mohammad Kermanshahi Hospital in 2019.

Methods: This study was done on 47 Acinetobacter baumannii isolates. Initial identification of the isolates was performed through phenotypic methods and final confirmation based on blaOXA-51 using PCR method. Antibiotic susceptibility of the isolates was determined by Disk – diffusion standard methods. The presence of class 1, 2 and 3 Integrons was investigated using PCR method.

Results: It can be seen that Cefepime showed the highest antibiotic resistance (100%) while Tigecycline (no resistance) and Colistin (8.51%) showed the lowest antibiotic resistance. The frequency of class 1(82.98%), 2(36.17%) and 3(no frequency) Integrons was determined.

Conclusion: We came to this conclusion that Acinetobacter baumannii had a high level of antibiotic resistance. There was a significant relationship between the presence of class 1, 2 and 3 Integrons and antibiotic resistance of the isolates to some kinds of antibiotics

INTRODUCTION

Acinetobacter baumannii is Gram-negative coccobacilli, catalase positive, oxidase negative, aerobic and non-fermentable. Also, it is an opportunistic pathogen with a low virulence. But, it can cause various infections through organs of respiratory system (pneumonia), sepsis, urinary tract, wound infections and contaminated catheters. During the last decade, the rate of hospital – acquired infections due to Acinetobacter baumannii has been increasing. The emergence of this bacterium is serious especially among hospitalized patients in burn and surgery intensive care units (1-3). Acinetobacter baumannii, as one the hospital-acquired infectious agent microorganisms, is an important human pathogenic bacterium which has caused many problems with respect to treatment failure and mortality occurrence of patients due to antibiotic resistance (4).

This bacterium shows a high resistance in environmental conditions so that it can survive in relative humidity of 31% for 11 days and relative humidity of 10% for 4 days. This bacterium can be seen in secretions of human body like sputum, urine, stool as well as vaginal discharges. In fact, the presence of this bacterium has been detected in skins of 25% of people and in pharyngeal regions of 7% of babies and adults (5).

Todays, Antibiotic resistance is one of the biggest problems in treatment of bacterial infections. Acinetobacter baumannii is not exception. According to a report by centers for disease control and prevention (CDC), the emergence of Acinetobacter resistance to Carbapenem has increased from 9% in 1995 to 40% in 2004 and such a resistance can be transferred to other pathogenic bacteria (6-9).

In general, Acinetobactor baumannii shows two types of resistance: extensive drug resistance (XDR), multidrug resistance (MDR) and pandrug resistant (PDR) (10-12). According to some studies, strains of Acinetobactor baumannii show multi drug resistance (13). Mechanisms of antibiotic resistance in these bacteria include purines, efflux pumps, secretion of Beta-lactamase as well as resistance genes acquired from Integrons (14). Integrons are the most effective genetic elements which have a very essential role in capturing and carrying genes, particularly those responsible for antibiotic resistance. By structure, Integrons have three distinct genetic regions of which two are highly conserved the 5′-conserved segment (5′-CS) and the 3′-conserved segment (3′-CS), that flank the central variable region where the gene cassettes are located. Integrase gene (Intl I) which encodes site- specific recombination is in 5′-CS gene and provokes gene cassette insertion through site- specific recombination mechanisms (15-17). Based on differences, present in Integrase gene, more than 9 classes of Integrons have been identified, 4 classes of them are related to clinical isolates. More attention has been paid to class 1 and 2 Integrons among clinical strains (18).

Raising awareness of mechanism and Antibiotic resistance pattern to antibiotics which are used for treatment of infections can accelerate quick and on time treatment of the related infections and decrease emergence of drug resistance and the cost of treatment. This study aimed at Investigation of the relationship between Antibiotic resistance pattern and class 1, 2 and 3 Integrons in Acinetobacter baumannii isolated from clinical samples of ICU patients of Dr. Mohammad Kermanshahi Hospital in 2019.

Material and methods: To study the probability of Acinetobacter baumannii, various clinical samples were isolated from blood, sputum, wound, urine, catheter and other bodily fluids collected from ICU patients of Dr. Mohammad Kermanshahi Hospital in 2019. Then, the collected samples were transported to the laboratory and placed on to culture mediums such as Blood agar and MacConcky agar. Then, they were incubated at 370C and 420C. identification was done using microbiological and biochemical standard methods, including: Gram staining, catalase staining, oxidase staining, glucose oxidation, arginine hydrolase test and Esculin Hydrolysis Test, nitrate resuscitation, Lactose intolerance as well as culture onto citrate mediums, TSI, SIM as well as pigment production (19,20). At the final, 47 Acinetobacter baumannii isolates were identified. Polymerase chain reaction (PCR) method using blaOXA-51 was applied for final confirmation of the isolates (16,21).

Antibiogram test was performed using Disk-diffusion standard method (Kirby Bauer) based on clinical and CLSI laboratory standards institute 2014 regrading 0.5 McFarland standard (22,23). To test antibiotic susceptibility of isolated bacteria, the bacteria were placed onto Mueller-hinton agar medium (Himedia, India) and antibiotic disks (MAST, merseyside, UK) were used, including Amikacin (30 µg), ceftazidime (30 µg), Tobramycin (10 µg), Meropenem (10 µg), Imipenem (10 µg), Piperacillin- Tazobactam (10/100 µg), Gentamicin (10 µg), Doxycycline (30 µg), Cefepime (30 µg),

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Ciprofloxacin (5 µg), Ampicillin-Sulbactam (10/10 µg), Colistin (10 µg) and Tigecycline (15 µg). non-growth zone diameter was measured by a ruler and the results were reported using CLSI 2014 tables. In this study, standard strains of Acinetobacter baumannii ATCC 19606 and E.coli ATCC 25922 were respectively used for positive and negative quality control (22-24). MDR defined as the isolate resistant to at least three classes of antimicrobial agents all penicillins and cephalosporins (including inhibitor combinations), fluroquinolones, and aminoglycosides. XDR Acinetobacter spp shall be the Acinetobacter spp isolate that is resistant to the three classes of antimicrobials described above (MDR) and shall also be resistant to carbapenems. Finally PDR Acinetobacter spp shall be the XDR Acinetobacter spp that is resistant to polymyxins and tigecycline (12). The investigation of the presence of class 1,2 and 3 Integron genes was done using specific primers (table 1) and Polymerase chain reaction (PCR) method.

Each clinical sample was purified by culture and Acinetobacter baumannii was isolated from all of the samples. Then the isolates were stored at-70°C in nutrient broth containing 30% glycerol v/v for further investigation (25). Genomic DNA used for PCR assays was obtained from bacterial suspension grown overnight in Luria broth (LB) with shaking incubator at 37°C. Extraction of genomic DNA from A. baumannii isolates was performed by boiling method. To do so, several purified bacterial colonies were dissolved in sterile water, boiled for 5 minutes, cooled and finally, centrifuged at 7000 r/min for 10 min (25-27). PCR reaction was performed with a final volume of 25µl. PCR compounds was include of 3µl 10X PCR buffer, 5µl of extracted template DNA, 0.5µl dNTPs mix (10mM), 0.8 µl MgCl2 (50 mM), 0.6µl DNA Taq polymerase, 1.5µl (10 pM/µl) of each primer (forward and reverse), and 13.6 µl sterile D/W. PCR reaction was performed to identify genes Int 1, Int 2 and Int 3 in 35 cycles. Cycling conditions were: initial denaturation at 940C for 6 min, denaturation at 940C for 60 s, annealing at 560C for 60 s, extension at 720C for 45 s and final extension at 720C for 6 min (26,28).

The program to identify gene blaOXA-51 included a 35 cycles. initial denaturation at 94°C for 3 min, denaturation at 94°C for 45 s, annealing at 60 °C for 45 s, and extension at 72°C for 60 s, as well as a final extension at 72°C for 5 min (29). PCR products were investigated using electrophoresis on a 1% agarose gel (and 80 V for 40 min) and stained with ethidium bromide (25). At final, the results and sample specifications were analyzed using SPSS software v20.

Statistical test: In the present study, the relationship between Integrons and drug resistance was investigated using SPSS software v20 and chi-square test. The level of significance was less than 0.05 ($P<0.05$).

Results: In this descriptive, cross-sectional study, among clinical samples isolated from blood, sputum, wound, urine, catheter and other bodily fluids collected from ICU patients of Dr. Mohammad Kermanshahi Hospital in 2018, 47 Acinetobacter baumannii isolates were identified using standard methods of identification. Of various understudied samples, 26 samples from males (55.32%) and 21 samples from females (44.68%) were collected. In respect of sampling frequency, 23 samples of blood (48.94%), 4 samples of sputum (8.51%), 3 samples of wound (6.38%), 9 samples of urine (19.15%), 4 samples of catheter (8.51) and 4 samples of trachea were collected. frequency of MDR and XDR isolates was 12 (25.53%) and 8 (17.02%), respectively and none were pan drug resistance.

The frequency of Integrons found in clinical samples showed that the frequencies of class 1 and 2 Integrons were 82.98% and 36.17%, respectively. In 19.14% of samples, class 1 and 2 Integrons were detected simultaneously. Class 3 Integrons were not isolated from the samples.

The final confirmation of the Acinetobacter baumannii with initial identification based on the emergence of blaOXA-51 gene, through PCR has been shown in Figure 1.

Discussion: Acinetobacter baumannii genus members have become resistant to almost all antibiotics and have an endless capacity to acquire antibiotic resistance. Numerous studies have shown that most strains of Acinetobacter baumannii are highly resistant to most clinically available antibiotics and their number of multi drug resistant strains of Acinetobacter is increasing, especially among hospitalized patients. Mobile genetic elements like Plasmids, Transposons as well as Integrons are the most effective elements which play an important role in acquiring and distributing of agents which are resistant to various Gram-negative bacteria, especially Acinetobacter baumannii strains. Various studies show that multi drug resistance in these bacteria is related to the presence of Integrons and gene cassettes (30-32).

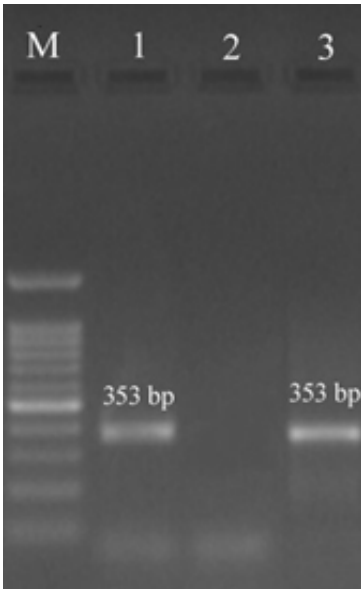

Figure 1) *Agarose gel electrophoresis of PCR products of the amplification of blaOXA -51 DNA in Acinetobacter baumannii isolates. M) 100 bp DNA marker; 1) positive control; 2) negative control; 3) the studied samples containing the 353-bp fragment*

According to the findings of the present study, the most frequent Acinetobacter strains isolated from clinical samples were related to the samples of blood (48.94%) this result was consistent with the results of previous studies done by Kiani and et al (2014, Ahvaz, Iran) 41.79%, Amini and et al (2015, Kermanshah, Iran) 67.2% it showed the role of the circulatory system in multiplication of bacteria but it was not consistent with the result of Izadi`s study (2014, Iran) sputum 60% (19,23,25). In this study 55.32% of the samples were separated from boys and 44.68% from girls, which was consistent with the results of Amini (53.5% men), Izadi (61% men) and Deilem Salehi and et al (2015, Babol, Iran) study 53.2% women are inconsistent (19,20,25).

In this study, the isolates were MDR 25.53% and XDR 17.02% . The frequency of isolates of MDR and XDR has been reported in Izadi (MDR 29%, XDR 21%), Deilem Salehi (MDR 91.4%, XDR 58.3%), Al-Sweih (MDR 85.1%), Koczura and et al (2014, Poland) MDR 65.2% (20,25,35,36). The reason for the differences in the frequency of antibiotic resistance reported in Acinetobacter baumannii isolates in different studies may be due to the presence of multiple clones in the study area and the reasonable rate and level of antibiotic use (12,36).

The results from the investigation of the presence of Int 1 and Int 2 genes in samples containing Acinetobacter baumannii using PCR are shown in Figure 2.

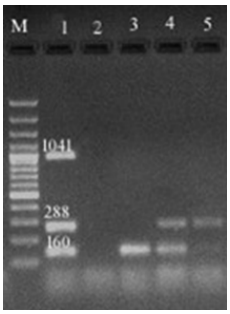

Figure 2) *Agarose gel electrophoresis of PCR products of the amplification of int I, int II and int III DNA in Acinetobacter baumannii isolates. M) 100 bp DNA marker. 1) positive control (160,288 and 1041bp). 2) negative control. 3,4,5) the studied samples containing the 160 and 288-bp fragments*

Based on the results of this study, Acinetobacter baumannii isolates identified by disk diffusion method showed 76.26% antibiotic resistance to the antibiotic groups used in this study like aminoglycosides, carbapenems, penicillins, tetracyclines, cephalosporins (100% resistance to cefepime) and quinolones. However, the lowest amount of antibiotic resistance was observed in glycylcyclins (no resistance to thiocyclines) and polypeptides (colistin). In fact, none of the identified Acinetobacter baumannii isolates were resistant to tigecycline and colistin resistance was found in only 8.51% of the isolates.

Peymani and et al (2012, Tabriz, Iran) reported in their study antibiotic resistance of Acinetobacter baumannii isolates identified from clinical samples to Ceftazidime (92%), Piperacillin/ Tazobactam (88%), Cefepime (85%), Gentamicin (85%), Amikacin (80%), Meropenem (56%), Imipenem (53%) (27). Amini reported antibiotic resistance of Acinetobacter baumannii isolates of their study to Gentamicin (77.6%) (19). but this percent was 86% for Gentamicin and Amikacin (81%) in the study of Aliakbarzadeh and et al (2013, Tabriz, Iran) which is consistent with the result from our study (33). Kiani reported antibiotic resistance of Acinetobacter baumannii isolates to Gentamicin (47.76%) and to Meropenem (2.98%) which was not consistent with the above results (23).

Mirnejad and et al (2010, Tehran, Iran) in their study on the investigation of antibiotic resistance pattern of Acinetobacter baumannii isolates, showed the isolates were resistant to Meropenem (44%), Ampicillin/ Sulbactam (62%), Imipenem (78%), Amikacin (90%), Piperacillin/ Tazobactam (48%), Ceftazidime (100%), Cefepime (100%), Gnetamicin (64%), Ciprofloxacin (92%) which are consistent to the present study (24). From above, it can be seen that antibiotic resistance of Acinetobacter baumannii isolates which are isolated from clinical samples in the above studies is higher than antibiotic resistance of the samples in the present study (specially to Aminoglycosides and Cephalosporins). The only effective antibiotics are Colistin and Tigecycline. In addition, the activation of more toxic antibiotics against this bacterium is a major concern and reminder of the increased attention and precision in the administration of antibiotics in the treatment of infections caused by this bacterium.

In this study, the frequency of Integrons found in clinical samples showed that the frequency of class 1 and 2 Integrons was 82.98 and 36/17%, respectively. In 19.14% of samples, class 1 and 2 Integrons were detected simultaneously. Class 3 Integrons were not isolated from the samples.

Taherikalani and et al (2011, Ilam, Iran) reported the frequency of class 1 Integrons (58%), class 2 Integrons (14%) and class 1 and 2 Integrons (9%). But they did not isolate class 3 Integrons (34). Kinani reported the frequency of class 1 (100%), class 2 (32.8%) and class 3 (4.4%) Integrons. The results from class 1 and 2 are consistent with the results from our study (23). Deylam salehi reported the frequency of class 1 (25.7%), class 2 (88.6%) and class 3(28.6%) Integrons (20). Koczura reported the frequency of class 1 Integrons in Acinetobacter baumannii strains (63.5%) but they did not detect class 2 and 3 Integrons in isolates (35). Lin and et al (2013, Taiwan) reported, in their study, the frequency of class 1 Integrons (72%) but they did not detect class 2 Integrons in isolates (37).

This discrepancy may be due to the ways the researchers used antibiotics (e.g. uncontrolled prescription), differences in geographical regions (e.g. intercountry or intera country), the site of the research or differences in the materials and methods (23). the absence or the lack of class 3 Integrons may indicate that this class has no role in creation or enhancement of resistance of Acinetobacter baumannii strains to antibiotics.

It can be seen from the findings of the study that there was no a significant relationship between the presence of class 1 Integrons in Acinetobacter baumannii isolates and resistance to antibiotics ($p>0.050$) but there was a significant relationship between the presence of class 2 Integrons in Acinetobacter baumannii isolates and resistance to Imipenem, Meropenem, Gentamicin, Ciprofloxacin, Ampicillin/ Sulbactam and Colistin ($p<0.050$). in studies done by Deylam salehi (20), Mirnejad (24) and Peymani (27) a significant relationship between resistance to Ceftazidime, Amikacin, Cefepime, Piperacillin/ Tazobactam, Imipenem, Meropenem, Gentamicin, Ciprofloxacin and Ampicillin/ Sulbactam and class 1 Integrons was reported which was not in accord to our findings. Mirnejad, Koeleman and et al (2001, Amsterdam, Netherlands) and Gaur and et al (2007, Varanasi, India) reported a significant relationship between the presence of class 2 Integrons in Acinetobacter baumannii Isolates and antibiotic resistance to Ceftazidime, Cefepime and Ciprofloxacin (which the Ciprofloxacin was in accord to our findings) (24,26,38). Lin and et al (2013) reported a significant relationship between the presence of Integrons and antibiotic resistance to Cefepime, Amikacin and Ciprofloxacin (37). In the cases when there is no significant relationship between the presence of class 2 Integrons and antibiotic resistance the resultant resistance can occur in different ways like defect in autolytic enzymes in cell wall or due to control plasmids or due to chromosome control (9,26).

Conclusion: The findings of the present study show the presence of the high antibiotic resistance in clinical isolates of Acinetobacter baumannii. In some cases, there was a significant relationship between the presence of class 1, 2 Integrons and antibiotic resistance. Since Integrons can transfer antibiotic resistance factors in to different strains, more attention should be paid to them in control protocols and treatment of infections due to Acinetobacter baumannii and new antibiotic resistances should be prevented by rational use of antibiotic. Colistin and Tigecycline are among effective antibiotic drugs in spite of toxicity.

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Conflict of interest

The authors declare that there is no conflict of interests

REFERENCES

1. Elham B, Fawzia A (2019). Colistin resistance in Acinetobacter baumannii isolated from critically ill patients: clinical characteristics, antimicrobial susceptibility and outcome. Afri Health Sci, 19 (3): 2400-2406.

2. Babay H , Kambal A, Al-Anazy A, Saidu A, Aziz Sh (2003). Acinetobacter Blood Stream Infection in a Teaching Hospital - Riyadh,Saudi Arabia. Kuwait Med J, 35 (3): 196-2011.

3. Hsueh P, Teng L, Chen CH , Chen W, Yu Ch (2002). Pandrug-resistant Acinetobacter baumannii causing nosocomial infections in a university hospital, Taiwan. Emerg Infect Dis, 8 (8): 827-831.

4. Curtis LT (2008). Prevention of hospital-acquired infections: review of non-pharmacological interventions. J Hosp Infect, 69: 204-19.

5. Allen DM, Hartman BJ (2005). Acinetobacter species. In: Principles and Practice of Infectious Disease. Ed, Mandell GL, Bennett JE, Dolin R. Elsevier publishing, Churchill Livingstone, USA, pp. 2632-5.

6. Naas T, Coignard B, Carbonne A, Blanckaert K (2006). VEB-1 Extended-Spectrum β-Lactamase producing Acinetobacter baumannii, France. Emerg Infec Dis, 12 (8): 1214-1220.

7. Schreckenberger PC, Daneshvar MI, Weyant RS, Hollis DG (2007). Acinetobacter, Achromobacter, Chryseobacterium, Moraxella and other nonfermentative gram negative rods: Manual of clinical microbiology. 9th ed. American Society for Microbiology Press. Washington DC.

8. Carey RB, Banerjee SN, Srinivasan A (2006). Multidrug-resistant Acinetobacter infections, 1995-2004. Presented at the 46th Interscience Conference on Antimicrobial Agents and Chemotherapy, San Francisco, pp: 27–30.

9. Fournier PE, Richet H (2006). The epidemiology and control of Acinetobacter baumannii in health care facilities. Clin Infect Dis, 42: 692-99.

10. Jeon BC, Jeong SH, Bae IK, Kwon SB, Lee K, Young D and et al (2005). Investigation of a nosocomial outbreak of imipenem-resistant Acinetobacter baumannii producing the OXA-23 β-lactamase in Korea. J Clin Microbiol,

43(5): 2241-2245.

11. Magiorakos AP, Srinivasan A, Carey R, Carmeli Y, Falagas M, Giske C and et al (2012). Multidrug‐resistant, extensively drug‐resistant and pandrug‐ resistant bacteria: an international expert proposal for interim standard definitions for acquired resistance. Clin Microbiol Infect, 18(3): 268-281.

12. Manchanda V, Sanchaita S, NP S (2010). Multidrug Resistant Acinetobacter. Journal of Global Infectious Diseases, (2)3: 291-304.

13. De Francesco MA, Ravizzola G, Peroni L, Bonfanti C, Manca N (2013). Prevalence ofmultidrug-resistant Acinetobacter baumannii and Pseudomonas aeruginosa in anItalian hospital. J Infect Public Health, 6(3): 179–85.

14. Bonomo RA, Szabo D (2006). Mechanisms of multidrug resistance in Acinetobacter species and Pseudomonas aeruginosa. Clin Infect Dis, 43: 2: 49-56.

15. Yan H, Li L, Zong M, Alam MJ, Shinoda S, Shi L (2010). Occurrence and characteristics of class1 and 2 integrons in clinical bacterial isolate from patients in south China. J Health Sci, 56(4): 442-50.

16. Visca P, Seifert H, Towner KJ (2011). Acinetobacter infection-an emerging threat to human health. IUBMB Life, 63(12): 1048-54.

17. Rowe-Magnus DA, Mazel D (2001). Integrons: natural tools for bacterial genome evolution. Current opinion in microbiology, 4(5): 565-9.

18. Zeighami H, Haghi F, Hajiahmadi F (2014). Integrons and Their role in Antibiotic Resistance. Journal of Laboratory and Diagnosis, 5 (22) :61-71.

19. Amini F, Karimpour HA, Vaziri S, Ghaderi M, Mohamadi S, Azizi M (2017). Prevalence Study of Aminoglycoside-Resistance Genes in Acinetobacter Baumannii Isolated from the Patients in Intensive Care Unit. J Isfahan Med Sch, 34(407): 1381-8.

20. Deylamsalehi M, Ferdosishahandashti E, Yahyapour Y, Khafri S, Pournajaf A, Rajabnia R (2017). Integron-Mediated Antibiotic Resistance in Acinetobacter baumannii Isolated from Intensive Care Unit Patients, Babol, North of Iran. BioMed Research International, 1-8.

21. Turton JF, Gabriel SN, Valderrey C, Kaufmann ME, Pitt TL (2007). Use of sequence-based typing and multiplex PCR to identify clonal lineages of outbreak strains of Acinetobacter baumannii. Clin Microbiol Infect, 13(8): 807-15.

22. Performance standards for antimicrobial susceptibility testing, 24th informational supplement. Clinical and Laboratory Standards Institue 2014; 34: M100-S24: 62-63.

23. Kiani S, Momtaz H, Serajian AA, Tajbakhsh E (2016). Detection of Integrons in Acinetobacter Baumannii Strains Isolated from the Nosocomial Infections of Ahvaz City and their Relation with the Resistance Pattern. Int.J.Med.Lab, 3(1): 50-63.

24. Mirnejad R, Mostofi S, Masjedian F (2013). Antibiotic resistance and carriage class 1 and 2 integrons in clinical isolates of Acinetobacter baumannii from Tehran, Iran. Asian Pac J Trop Biomed, 3(2): 140-5.

25. Izadi B, Souzani R, Farahani A, Mehrabian S, Mohajeri P (2017). Frequency of class 2 integrons in multidrug-resistant Acinetobacter baumannii isolated from patients in West of Iran. Ann Trop Med Public Health, 10: 104-8.

26. Koeleman JGM, Stoof J, Van Der Bijl MW, Vandenbroucke-Grauls CM, Savelkoul PHM (2001). Identification of epidemic strains of Acinetobacter baumannii by integrase gene PCR. J Clin Microbiol, 39(1): 8-13.

27. Peymani A, Farajnia S, Nahaei MR, Sohrabi N, Abbasi L, Ansarin K and et al (2012). Prevalence of class 1 integron among multidrug-resistant Acinetobacter baumannii in Tabriz, northwest of Iran. Pol J Microbiol, 61(1): 57-60.

28. Ploy MC, Denis F, Courvalin P, Lambert T (2000). Molecular Characterization of Integrons in Acinetobacter baumannii: Description of a Hybrid Class 2 Integron. Antimicrob Agents Chemother, 44 (10): 2684-688.

29. Turton JF, Woodford N, Glover J, Yarde S, Kaufmann ME, Pitt TL (2006). Identification of Acinetobacter baumannii by detection of the blaOXA-51 like carbapenemase gene intrinsic to this species. J Clin Microbiol, 44(8): 2974–6.

30. Neonakis IK, Spandidos DA, Petinaki E (2011). Confronting multidrug-resistant Acinetobacter baumannii: a review. Int J Antimicrob Agents, 37(2): 102-9.

31. Garnacho-Montero J, Amaya-Villar R (2010). Multiresistant Acinetobacter baumannii infections: epidemiology and management. Curr Opin Infect Dis, 23(4): 332-9.

32. Cambray G, Guerout AM, Mazel D (2010). Integrons. Annu Rev Genet, 44: 141-66.

33. Aliakbarzade K, Farajnia S, Karimi NA, Zarei F, Tanomand A (2014). Prevalence of Aminoglycoside Resistance Genes in Acinetobacter baumannii Isolates. Jundishapur J Microbiol, 7(10): e11924.

34. Taherikalani M, Maleki A, Sadeghifard N, Mohammadzadeh D, Soroush S, Asadollahi P and et al (2011). Dissemination of class 1, 2 and 3 integrons among different multidrug resistant isolates of Acinetobacter baumannii in Tehran hospitals, Iran. Pol J Microbiol, 60(2): 169-74.

35. Koczura R, Przyszlakowska B, Mokracka J, Kaznowski A (2014). Class 1 integrons and antibiotic resistance of clinical Acinetobacter calcoaceticus-baumannii complex in Poznań, Poland. Curr Microbiol, 69(3): 258-62.

36. Al-Sweih NA, Al-Hubail M, Rotimi VO (2012). Three distinct clones of carbapenem-resistant Acinetobacter baumannii with high diversity of carbapenemases isolated from patients in two hospitals in Kuwait. J Infect Public Health, 5: 102-8.

37. Lin MF, Chang KC, Yang CY, Yang CM, Xiao CC, Kuo HY and et al (2010). Role of integrons in antimicrobial susceptibility patterns of Acinetobacter baumannii. Jpn J Infect Dis, 63(6): 440-3.

38. Gaur A, Prakash P, Anupurba S, Mohapatra TM (2007). Possible role of integrase gene polymerase chain reaction as an epidemiological marker: study of multidrug-resistant Acinetobacter baumannii isolated from nosocomial infections. Int J Antimicrob Agents, 29(4): 446-50.