ORIGINAL ARTICLE

EVALUATION OF ANTIMICROBIAL ACTIVITY OF TIGECYCLINE AGAINST CARBAPENEM RESISTANT ACINETOBACTER SPECIES: STUDY FROM A TERTIARY CARE SETTING

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ABSTRACT

Background: *Acinetobacter* species are a key source of hospital acquired infection in debilitated patients. *Acinetobacter baumannii* being the commonest species implicated in such infections. The clinical significance of Acinetobacter genus is partially due to its capability to develop resistance against broad spectrum antibiotics including beta lactams, cephalosporins, aminoglycosides and quinolones. Resistance to carbapenems is also on the rise. The objective of this study was to evaluate in vitro antimicrobial activity of Tigecycline against carbapenem resistant *Acinetobacter* species (CRAB) isolated from clinical specimens of patients presenting to a Tertiary Care Hospital.

Methods: A total of 230 clinical samples submitted to Microbiology Lab, AIMC were included in this study. All clinical specimens were identified using the standard microbiological protocol. After the confirmation of Gram negative coccobacillary rods as Acinetobacter species, modified Kirby Bauer disc diffusion method was used to detect Carbapenem resistant Acinetobacter species. Antibiotic susceptibility to Tigecycline was checked both by disc diffusion and then MICs were determined with the help of E strips (Epsilometer test).

Results: Out of 230 isolates of Carbapenem resistant *Acinetobacter,* 220(95.6%) were sensitive to Tigecycline, 3(1.3%) were intermediate resistant and 07(3%) were resistant to Tigecycline.

Conclusion: Tigecycline has shown considerable *in vitro* activity against MDR (multi drug resistant) including carbapenem-resistant *Acinetobacter* spp. It seems to be a good treatment option for infections caused by MDR *Acinetobacter*. However, data to support its clinical use is still limited.

Keywords: Acinetobacter, Carbapenem resistant, Multidrug resistant, Tigecycline

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INTRODUCTION

The emergence and spread of antibiotic-resistant bacterial pathogen is a great public health concern throughout the world. *Acinetobacter baumannii*, once considered a low-category pathogen, has emerged as a stubborn infectious agent (Pourhajibagher M *et al*., 2016). The scientific community is paying more attention to this pathogen due to its high prevalence of infections in the hospital setting, and significantly increased rate of community-acquired infections by this organism over the past few years (QiL *et al., 2016*).

Antimicrobial resistance among *Acinetobacter* species has increased substantially in the past decade. The capacity of *Acinetobacter* species for extensive antimicrobial resistance may be due in part to the organism's relatively impermeable outer membrane and its environmental exposure to a large reservoir of

resistance genes (Nemec A *et al.,* 2016). It harbors chromosome-mediated genetic elements on one hand; while on the other hand, it can persist for a prolonged period in harsh environments (walls, surfaces, and medical devices) in the hospital settings (Su, C. H. *et al.*2012).

It is commonly found in infections of the lung, blood stream, urinary tract and wounds in intensive care units, where it especially affects people with immune deficiency. Effective antibiotic treatment of *such* infections is of paramount importance. Carbapenems (i.e., imipenem, meropenem) are recommended agents for the treatment of MDR *A. baumannii* infections. However, a significant rise in carbapenem-resistant *A. baumannii* has been reported globally (Kumarasamy, K. K. *et al.,* 2007).

Regarding treatment options of CRAB, Sulbactam has been successfully used in the treatment of serious *A.baumannii* infections; however, the activity of this agent against carbapenem-resistant isolates is decreasing. Polymyxins show reliable antimicrobial activity against *A.baumannii* isolates. Available clinical reports support their effectiveness and mitigate previous concerns for toxicity (Ilkem Acar Kaya *et al.,* 2017).

FDA-approved Tigecycline, the first identified glycylcycline antibiotic, which mimics the structure of tetracycline but is effective against tetracycline resistant bacteria, has been recommended for the treatment of complicated intra-abdominal infections, skin infections, and community-acquired pneumoniae since 2007(Deng, M. *et al.2014)* .

Chemically, it constitutes the 9-*t*-butylglycylamido derivative of minocycline. Regarding its mechanism of action, tigecycline enters bacterial cells through energy-dependent pathways or with passive diffusion, and reversibly binds to the 30S subunit of the ribosome. It acts by blocking the incorporation of transfer RNA into the A site of the ribosome, thus inhibiting protein synthesis (Akers KS *et al.,* 2010). In comparison with tetracyclines, tigecycline binds to corresponding ribosomal sites with greater affinity, and irrespective of the presence of mutations that confer resistance to tetracyclines. Tgc was recognized as an antibiotic of "last-resort"because of its bacteriostatic activity against MDR *Acinetobacter*. The activities of tigecycline alone and in combination with other antimicrobials are now under consideration for carbapenem resistant *Acinetobacter baumannii* (Garrison MW *et al.,* 2005)

Tigecycline has also shown adequate activity against *Acinetobacter* species of potential clinical significance other than *A. baumannii*. Still, whether tigecycline constitutes a potentially effective treatment option against highly resistant *Acinetobacter* spp. has not been evaluated in a comprehensive manner (Karageorgopoulos DE *et al.,* 2008).

METHODS

This cross-sectional study was conducted at the Microbiology laboratory, Pathology department, Allama Iqbal Medical College, Lahore over a period of 6 months from July 2018 to January 2019, approved by Ethical review board of AIMC, Lahore. A total of 230 clinical samples (i.e pus, wound swabs, urine, sputum) submitted to Microbiology Laboratory, AIMC, Lahore was included in this study. Clinical specimens were processed according to standard protocols. All clinical specimens were cultured on 5% sheep Blood Agar and MacConkey Agar Plate except urine samples which were cultured on CLED Agar. Organism cultured were identified on the basis of colonial morphology (non-lactose fermenter), size of colony, color of colony, Gram staining (coccobacillary rods which stained as gram negative), catalase test, oxidase test and biochemical tests by using API 10S, API 20E or API 20NE.

After the confirmation of Gram negative coccobacillary rods as Acinetobacter species, we further proceeded for the detection of carbapenem resistant Acinetobacter using modified Kirby Bauer disc diffusion method using Clinical and Laboratory Standards Institute (CLSI) 2018 guidelines. Carbapenem resistant *Acinetobacter* species were further checked for their susceptibility to Tigecycline. Zone sizes of Tigecycline were interpreted according to European Committee on Antimicrobial Susceptibility Testing (EUCAST) 2018 guidelines. Zone size of 18mm or greater than 18mm was taken as sensitive, between 15mm-18mm was taken intermediate and less than 15mm were taken as resistant. The Epsilometer test (E test) was used to determine the minimum inhibitory concentrations (MICs) of Tigecycline resistant isolates. Results of E test were interpreted according to EUCAST 2018 guidelines. An MIC equal to or less than 2mg/L was taken as sensitive as and more than 2mg/L was taken as resistant.

RESULTS

Out of 230 *Acinetobacter* species, 1(0.4%) was isolated from Ascitic fluid, 1(0.4%) was isolated from aspirate, 3(1.3%) were isolated from catheter tip, 19(8.3%) were isolated from CVP Tip, 1(0.4%) was isolated from enteric perforation, 4(1.7%) were isolated from ETT Tip, 1(0.4%) was isolated from nasal swab, 3(1.3%) were isolated from pleural fluid, 31(13.5%) were isolated from pus, 30(13.0%) were isolated from sputum, 1(.4%) was isolated from throat swab, 3(1.3%) were isolated from tip, 48(20.9%) were isolated from Tracheal secretions,

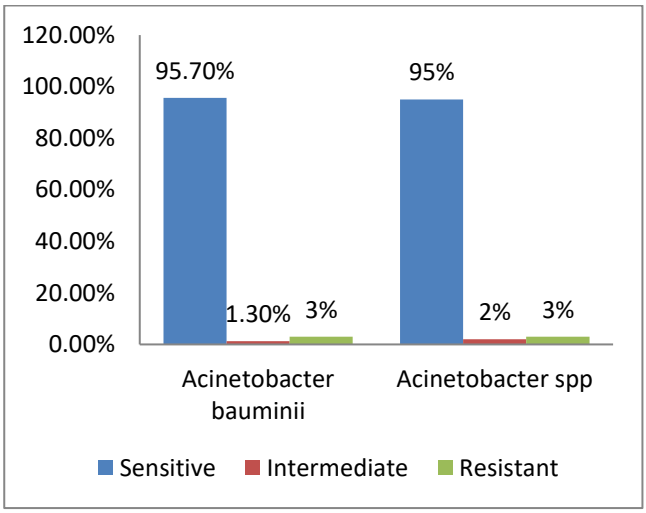
11(4.8%) were isolated from urine and 73(31.7%) were isolated from wound. Out of 230, 157 isolates were identified as *A.baumannii* and 73 isolates were identified as *Acinetobacter spp.* 95.7% *of A.baumannii,* were sensitive to Tigecycline, 1.3% were intermediate resistant and 03% were resistant to Tigecycline. 95% *Acinetobacter spp* were sensitive and 3% were resistant to Tigecycline.

Table: 1 Antibiotic susceptibility of Acinetobacter to various antibiotics

	I		R		S	
	Number	%	Number	%	Number	%
MEM	0	0.0%	230	100.0%	0	0.0%
TZP	0	0.0%	229	99.6%	1	0.4%
CTR	0	0.0%	230	100.0%	0	0.0%
AMK	0	0.0%	200	87.0%	30	13.0%
GEN	0	0.0%	204	88.7%	26	11.3%
CIP	0	0.0%	218	94.8%	12	5.2%
TET	0	0.0%	153	66.5%	77	33.5%
MH	0	0.0%	72	31.3%	158	68.7%
DOX	0	0.0%	112	48.7%	118	51.3%

Figure: 1 Tigecycline Resistant isolates

Figure: 3 Sensitivity of Acinetobacter against Tigecycline



DISCUSSION

A. baumannii infection represents a growing global threat. The resistance rates of *A. baumannii* to antibiotics are highly variable in different parts of the world. The reasons for inclining antibiotic resistance include irrational use of antibiotics, broad-spectrum antibiotic use in intensive care units, crowding, poor hygiene, failure to comply to the infection containment sops and increased worldwide travel.

A.baumannii infections are difficult to treat and it has become quite challenging for the physicians (Fishbian, J *et al.*, 2010).The presence of MDR and XDR *A.baumannii* has become a global threat as more cases of morbidity and mortality are noted *(*Goli, N. M *et al.,* 2017).So today's focus is on Tigecycline, colistin alone or in combination to treat MDR and carbapenem resistant *Acinetobacter*.

In our study out of 230 isolates of Carbapenem resistant *Acinetobacter*, 191(83.0%) were sensitive to Tigecycline, 3(1.3%) were intermediate resistant to

Tigecycline and 36(15.7%) were resistant to Tigecycline with an overall sensitivity of 84 %.

The activity of tigecycline in our isolates is comparable with that of many other national and international studies. Among different studies tigecycline has shown great sensitivity of 92%, 99% and 99.3% (Pachon-Ibanez *et al.*, 2004, Souli, M *et al.*, 2006, Sohail, M *et al.*, 2016). One recent study from Lahore showed 100% susceptibility of XDR *A.baumannii* to trigecycline but due to high mortality rate by using tigecycline as a mono-therapeutic drug in severe bloodstream infections combination therapy with different antibiotics is required (Afshan Z *et al.*, 2019)

A retrospective, hospital record–based, cross-sectional study conducted at a tertiary care hospital in Odisha, India from July 2010 to December 2012, in which a total of 8749 clinical samples were collected from patients. Out of 8749 clinical samples, 4589 (52.5%) yielded significant growth and only 137 (3 %,) *Acinetobacter* spp. were isolated. Out of 137 isolates, 75 (54.7%) were resistant to more than three classes of antibiotics (multidrug resistant) and 8 (5.8%) were resistant to all commonly used antibiotics (pan-drug resistant). Majority of the isolates were sensitive to imipenem, meropenem, and piperacillin/tazobactam, and showed resistance rates of 19%, 22%, and 23%, respectively. All eight pan-drug resistant isolates were 100% sensitive to colistin and Tigecycline (Dash M *et al.*, 2013)

Similarly, a study was performed in Pretoria, South Africa in 2011. A total of 232 carbapenem resistant clinical isolates of *A. baumannii* were collected over six months study period.Out of 232, 169 (75.8%) were fully susceptible, 37 (16.6%) intermediately resistant and only 17 (7.6%) were resistant to Tigecycline. None of the carbapenem resistant isolates were susceptible to ampicillin, amoxicillin/clavullanic acid, piperacillin/tazobactam, cefuroxime, cefuroxime axetil, cefoxitin, cefepime or nitrofurantoin (Ahmed NH *et al.*, 2012)

In 2007, a study was conducted in Italy to determine the *in vitro* activity of tigecycline and comparator agents against carbapenem sensitive and resistant *A.baumannii* clinical isolates. *A. baumannii* showed widespread resistance to ceftazidime, ciprofloxacin and aztreonam in more than 90% of the strains; resistance to imipenem and meropenem was 50 and 59% respectively, amikacin and gentamicin were both active against about 30% of the strains and colistin about 99%, with only one strain resistant. By comparison with tetracyclines, tigecycline and doxycycline showed a higher activity. Among the tetracyclines, the unimodal distribution of susceptibility of tigecycline was comparable with that of doxycycline (93 and 94% susceptible, respectively). The results of antimicrobial activity of tigecycline against A.baumannii in this study are similar to the results of my study (Mezzatesta ML et al., 2008).

Results of our study suggest that Tigecycline has considerable *in vitro* activity against MDR (multi drug resistant) including carbapenem-resistant *Acinetobacter* spp (CRAB). It seems to be a good treatment option for infections caused by MDR and CRAB. So judicial use should be implied to conserve it as an effective tool against waging war of last resort antibiotics.

REFERENCES

1. Akers KS, Chaney C, Barsoumian A, Beckius M, Zera W, Yu X, Guymon C, et al. Aminoglycoside resistance and susceptibility testing errors in Acinetobacter baumannii-calcoaceticus complex. Journal of clinical microbiology. 2010; **48**(4):1132-38.
2. Deng, M. Molecular epidemiology and mechanisms of tigecycline resistance in clinical isolates of *Acinetobacter baumannii* from a Chinese university hospital. *Antimicrob Agents Ch. 2014;* **58,** 297–303.
3. Garrison MW, Neumiller JJ, Setter SM. Tigecycline: An investigational glycylcycline antimicrobialwith activity against resistant gram-positive organisms. Clinical therapeutics. 2005 J Karageorgopoulos DE, Kelesidis T, Kelesidis I, Falagas ME. Tigecycline for the treatment of multidrug-resistant (including carbapenem-resistant) Acinetobacter infections: a review of the scientific evidence. Journal of Antimicrobial Chemotherapy. 2008; **62**(1):45-55.
4. Hsueh PR, Chen ML, Sun CC, Chen WH, Pan HJ, Yang LS, et al. Emergence of antimicrobial drug resistance of major pathogens causing nosocomial infections at a university hospital in Taiwan, 1981-1999.Emerg Infect Dis. 2002;**8**:63–68.
5. Karageorgopoulos DE, Falagas ME. Current control and treatment of multidrug-resistant Acinetobacter baumannii infections. The Lancet infectious diseases. 2008; **8**(12):751-762.
6. 6.Kelesidis T, Karageorgopoulos DE, Kelesidis I, Falagas ME. Tigecycline for the treatment of multidrug-resistant Enterobacteriaceae: a systematic review of the evidence from microbiological and clinical studies. Journal of Antimicrobial Chemotherapy. 2008; **62**(5):895-904.
7. 7.Kumarasamy, K. K. *et al.* Emergence of a new antibiotic resistance mechanism in India, Pakistan, and the UK: a molecular,biological, and epidemiological study. *Lancet Infect Dis*. 2010; **10,** 597–602.
8. 8.Lockhart, S. R., Abramson, M. A., Beekmann, S. E., Gallagher, G., Riedel, S., Diekema, D. J., et al. antimicrobial resistance among Gram-negative bacilli causing infections in intensive care unit patients in the United States between 1993 and 2004. *J Clin Microbiol: 2007;* **45:** 3352-3359.

9. 9.Navon-Venezia S, Leavitt A, Carmeli Y. High tigecycline resistance in multidrug-resistant Acinetobacter baumannii. Journal of Antimicrobial Chemotherapy. 2007 ;**59**(4):772-774.
10. 10.Pachon-Ibanez, M. E., Jimenez-Mejias, M. E., Pichardo, C., Llanos, A. C., & Pachon, J. Activity of Tigecycline (GAR-936) against *Acinetobacter baumannii* Strains, Including Those Resistant to Imipenem. *ANTIMICROB. AGENTS CHEMOTHER:*2004; **48:4479**-4481.
11. 11.Pourhajibagher M, Hashemi FB, Pourakbari B, Aziemzadeh M, Bahador A. Antimicrobial resistance of Acinetobacter baumannii to imipenem in Iran: a systematic review and meta-analysis. *Open Microbiol J.* 2016; **10**:32–42.
12. 12.Qi L, Li H, Zhang C, et al. Relationship between antibiotic resistance,biofilm formation, and biofilm-specific resistance in Acinetobacter baumannii. *Front Microbiol.* 2016; **7:**483.
13. 13.Scheetz MH, Qi C, Warren JR, Postelnick MJ, Zembower T, Obias A, Noskin GA. In vitro activities of various antimicrobials alone and in combination with tigecycline against carbapenem-intermediate or-resistant Acinetobacter baumannii. Antimicrobial agents and chemotherapy. 2007; **51**(5):1621-1626.
14. 14.Slover, C. M., Rodvold, K. A. & Danziger, L. H. Tigecycline: a novel broad-spectrum antimicrobial. *Ann Pharmacother*. 2007; **41,** 965–972.
15. 15.Sohail, M., Rashid, A., Aslam, B., Waseem, M. et al. antimicrobial susceptibility of *Acinetobacter* clinical isolates and emerging antibiogram trends for nosocomial infection management. *Rev Soc Bras Med Trop:* 2016; **49:**300-304.
16. 16.Souli, M., V. Kontopidou, F., Koratzanis, E., Antoniaduo, A., Giannnitsioti, E, et al. In Vitro Activity of Tigecycline against Multiple-Drug-Resistant, Including Pan- Resistant, Gram-Negative and Gram-Positive Clinical Isolates from Greek Hospitals. *Antimicrob. Agents and chemother:* 2006; **50:**3166
17. 17.Su, C. H. *et al.* Increase of carbapenem-resistant *Acinetobacter baumannii* infection in acute care hospitals in Taiwan: association with hospital antimicrobial usage. *PloS one* **7,** e37788 (2012).
18. 18.Wang, J., Pan, Y., Shen, J., & Xu, Y. The efficacy and safety of tigecycline for the treatment of bloodstream infections:a systemic review and meta-analysis. *Ann Clin Microbiol Antimicrob:2017;* **16:** 24.