



*Original Contribution*

**IN VITRO ACTIVITY OF LEVOFLOXACIN, GEMIFLOXACIN, LINEZOLID, VANCOMYCIN, DALBAVANCIN AND TELAVANCIN AGAINST GRAM-POSITIVE STRAINS, ISOLATED FROM CLINICAL SAMPLES**

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**ABSTRACT**

The activity of six antibacterials / levofloxacin, gemifloxacin, linezolid, vancomycin, dalbavancin and telavancin/ against 55 gram-positive strains was tested using MIC determination. Linezolid, dalbavancin and telavancin were active against all the strains tested, with low MIC's levels. The MIC's range for dalbavancin was 0.012-0.125 µg/ml for all strains and 0.023-0.75 µg/ml – for telavancin respectively. Gemifloxacin could be used with success for staphylococcal / MRSA / strains treatment.

**Key words:** Gram-positive bacteria; Multi-resistance; New antimicrobials

**INTRODUCTION**

Multi-resistant gram-positive bacteria Staphylococci /including MRSA/ and Enterococcus spp are very important pathogens for hospital infectious pathology. According to our data (1) these bacteria – /S. aureus, Enterococcus faecalis and Enterococcus faecium/ are in the scale of the first ten most often isolated microorganisms for 2008 in the clinics of Military Medical Academy in Sofia. Due to the increasing resistance of staphylococci and enterococci to beta lactams, quinolones and glycopeptides (2), physicians have sought to establish the efficacy of other antimicrobial agents against these problem pathogens. Gemifloxacin, (R,S)-7-(3-aminomethyl-4-syn-methoxyimino-1-pyrrolidinyl)-1-cyclopropyl-6-fluoro-1,4-dihydro-4-oxo-1,8-naphthyridine-3-carboxylic acid methanesulfonate, exhibits broad-spectrum antibacterial activity (3). Dalbavancin and telavancin are semisynthetic

lipoglycopeptides that demonstrate promise for the treatment of patients with infections caused by multi-drug-resistant Gram-positive pathogens (4, 5). In many cases the choice of antibacterials is significant problem for effective treatment of such problematic for hospital infectious pathology bacteria.

The aim of this study is to investigate in vitro activity of six antimicrobial drugs with respect gram-positive bacteria, isolated from clinical samples in a multiprofile hospital.

**MATERIALS AND METHODS**

**Hospital**

Military Medical Academy /MMA/ in Sofia, Bulgaria is a community hospital with 800 beds. The hospital is a one of the national centres for trauma, respiratory disease, and liver transplantation patients' treatment. Antibiotic stewardship at Military Medical Academy /MMA/ includes all groups of antibiotics together with carbapenems, quinolones, third and fourth generations of cephalosporins, glycopeptides and others.

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## Bacteria

There was investigated collection of 55 strains, from them 24 MRSA, 5 – *S. epidermidis*, resistant to oxacilin- cefoxitin, 17 strains *Enterococcus faecium* and 9 strains – *Enterococcus faecalis*. The strains were isolated from clinical samples in different clinics at the Military Medical Academy in Sofia, Bulgaria for the period of time – March 2010 to October 2011. Three of the strains were isolated during 2012. All the strains were identified as *Staphylococcus*, *Enterococcus faecalis* or *Enterococcus faecium* according to the results of biochemical profiles obtained by VITEC 2 System, using GP 21342 Card /bioMerieux Vitek Inc., Hazelwood, Mo. (6)

## MIC determination

The MIC of the strains investigated to antimicrobials Levofloxacin, Linezolid and Vancomycin was done by VITEC 2 System using AST 22226 Card /bioMerieux Vitek Inc., Hazelwood, Mo. (6) and estimated according to CLSI 2011 (7). The MIC for Gemifloxacin, Dalbavancin and Telavancin was determined by E-test, obtained from AB Biodisk (Solna, Sweden) (8), because of lack of AST VITEC 2 cards for these antibacterials. All the strains tested with exception of two strains / one MRSA and one *E.faecium*/ were sensitive to vancomycin.

## RESULTS AND DISCUSSION

Emergence of staphylococci that are resistant to methicillin and quinolone and recently to

vancomycin, as well as the isolation of *Enterococcus* spp with their propensity to cause serious systemic infections, necessitates other therapeutic requirements. Gemifloxacin, dalbavancin and telavancin are relatively less used in clinical practice antibacterials in Bulgaria and this study examined the susceptibilities of 55 gram-positive bacteria to gemifloxacin, dalbavancin and telavancin compared with the susceptibilities of levofloxacin, vancomycin and linezolid by MIC analysis.

The susceptibility results, expressed as MIC ranges, MIC<sub>50</sub>s, and MIC<sub>90</sub>s of all 55 strains investigated, MRSA and *Enterococcus* spp respectively are presented in **Table 1**. MICs of all strains, listed in **Table 1** show that all compounds except quinolones /levofloxacin and gemifloxacin/ were active against all strains. Dalbavancin was the most potent agent, with an MIC at which 50% of the strains were inhibited of 0.032 µg/ml and an MIC at which 90% of the strains were inhibited of 0.047 µg/ml. Two strains / one MRSA and one *E.faecium* / showed resistance to vancomycin with MIC > 32 µg/ml. The MIC results for telavancin are were slightly higher to those, obtained for dalbavancin - MIC<sub>50</sub> is 0.125 µg/ml and MIC<sub>90</sub> is 0.25 µg/ml respectively. All the strains were sensitive to linezolid. Because no approved susceptibility breakpoints are available for gemifloxacin, dalbavancin and telavancin the percentages of organisms susceptible to these antimicrobial agents were not recorded. (2, 7).

**Table 1.** Minimum inhibitory concentrations (MIC) of 55 gram-positive strains isolated from clinical samples

Drug	MIC <sub>s</sub> for gram-positive strains (n=55)		MIC <sub>s</sub> for MRSA (n=24)		MIC <sub>s</sub> for <i>Enterococcus</i> (n=26)	
	Range	MIC <sub>50</sub> /MIC <sub>90</sub>	Range	MIC <sub>50</sub> /MIC <sub>90</sub>	Range	MIC <sub>50</sub> /MIC <sub>90</sub>
Levofloxacin	0.12 – 8	4/8	0.12 – 8	4/8	1 – 8	2/8
Gemifloxacin	0.006 – 32	0.5/16	0.006 – 2	0.38/1	0.032 – 32	1.5/32
Linezolid	1 – 4	2/4	1 – 4	2/4	1 – 2	1/2
Vancomycin	0.5 – 32	0.5/1	0.5 – 32	0.5/1	0.5 – 32	0.5/1
Dalbavancin	0.012 – 0.125	0.032/0.047	0.023 – 0.094	0.032/0.047	0.016 – 0.125	0.047/0.125
Telavancin	0.023 – 0.75	0.125/0.25	0.094 – 0.38	0.19/0.38	0.023 – 0.75	0.064/0.38

Results, concerning susceptibility testing for MRSA show, that gemifloxacin was more active than levofloxacin with MIC<sub>90</sub> of 1.0 µg/ml, but less active in comparing with dalbavancin and

telavancin with MIC<sub>90</sub> of 0.047 µg/ml and 0.38 µg/ml respectively. (**Table 1**) According to Gönüllü, N. et al 2001 (9), gemifloxacin, levofloxacin and ofloxacin are the most active

agents against MSSA, with a 100% susceptibility rate for each and gemifloxacin had the lowest MIC<sub>90</sub> ( $\leq 0.06$  mg/L). On the other hand, only 3.8% of the MRSA strains remained susceptible to gemifloxacin. Among the fluoroquinolones tested /ciprofloxacin, gemifloxacin, grepafloxacin, moxifloxacin, ofloxacin, sparfloxacin, trovafloxacin/, gemifloxacin demonstrated the most potent in vitro activity against MSSE, MRSE, *S. haemolyticus*, and *S. hominis* but was not as active as trovafloxacin against *S. aureus* isolates. (3). Lin, G. et al 2005 (4) found that dalbavancin was the most potent agent, with an MIC at which 50% of staphylococci were inhibited of 0.03  $\mu$ g/ml and an MIC at which 90% of staphylococci were inhibited of 0.06  $\mu$ g/ml. The activity of telavancin against 67 methicillin-resistant *Staphylococcus aureus* (MRSA) isolates showed, that all strains investigated were susceptible to telavancin at  $< 1$   $\mu$ g/ml (10). All MRSA strains tested were sensitive to linezolid.

The in vitro activity of gemifloxacin against enterococci is very closed to this presented by levofloxacin - with relatively high levels of MIC<sub>50</sub> - 1.5  $\mu$ g/ml and MIC<sub>90</sub> - 32  $\mu$ g/ml. (**Table 1**). According to Dwight, H. et al 2000 (3) none of the fluoroquinolones tested appears to offer any clinically important activity against penicillin-resistant enterococcal strains. In contrary and also in comparing with the data for vancomycin, the results obtained by us for dalbavancin and telavancin with respect enterococcal strains tested, showed low MIC's levels - MIC<sub>90</sub> of 0.125  $\mu$ g/ml for dalbavancin and MIC<sub>90</sub> of 0.38  $\mu$ g/ml for telavancin. Streit, J. et al 2004 (11) showed also excellent activity of dalbavancin against all gram-positive strains tested except for *vanA* enterococci. Zhanel G. et al 2011 (5) believed, that enterococci exhibiting the VanA phenotype (resistance to both vancomycin and teicoplanin) are resistant to both dalbavancin and telavancin but exhibit activity against VanB vancomycin-resistant enterococci.

The in vitro activity of the relatively new for Bulgarian clinical practice antibacterials gemifloxacin, dalbavancin and telavancin, described in this study, is consistent with previously reported data. (4, 11). The findings

from our studies, show excellent activity of dalbavancin, telavancin and linezolid against all gram-positive microorganisms tested, including MRSA and Enterococcus strains. Gemifloxacin could be used with success for staphylococcal /MRSA/ strains treatment. Results of the current study, will add our data to the experience, obtained by other investigators, needed for the successful treatment of the problematic for hospital infectious pathology bacteria.

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