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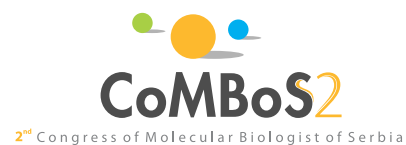
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AdeABC EFFLUX PUMP-MEDIATED RESISTANCE TO TIGECYCLINE IN *ACINETOBACTER BAUMANNII* ISOLATES FROM BALKAN HOSPITALS

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Introduction: Multidrug-resistant (MDR) *Acinetobacter baumannii* has been recognized as one of the most serious healthcare challenges worldwide. Although tigecycline represents one of the last resort therapies for MDR *A. baumannii*, resistance to this antibiotic has been reported and mostly is mediated by AdeABC efflux pump. The aim of our study was to investigate the molecular mechanism responsible for tigecycline resistance of thirty-seven *A. baumannii* isolates from Balkan medical settings (Serbia, Bosnia and Herzegovina and Montenegro) gathered in 2016 and 2022.

Methods: Minimal inhibitory concentration (MIC) values for tigecycline were determined using microdilution method according to EUCAST guidelines. Inhibition of the efflux of tigecycline was tested by the same method using a combination of antibiotic and efflux pump inhibitor (CCCP). Amino acid alterations within AdeS and AdeR proteins were detected by comparing to sequences of referent isolates ATCC19606 and ATCC17978. Expression of the *adeB* gene of selected isolates was monitored by RT-qPCR.

Results: All tested isolates were resistant to tigecycline and showed significant decrease in tigecycline MIC values in presence of CCCP (≥ 16 -fold reduction) indicating that antibiotic efflux is responsible for tigecycline resistance. The analysis of two-component system AdeRS, regulatory system of RND efflux pump AdeABC, revealed that most of the isolates have G186V and N268H alternations in AdeS (n=32), while most common changes in AdeR were V120I and A136V (n=29). In addition, RT-qPCR showed that selected isolates upregulate expression of the *adeB* gene (from 1,13- to 3-fold).

Conclusion: This study revealed that AdeABC overexpression is the main mechanism of tigecycline resistance in *A. baumannii* isolated in Balkan hospitals.

Key words: *Acinetobacter baumannii*; tigecycline resistance; efflux pump; AdeABC; AdeRS

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