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Short Communication

RISK FACTORS THAT DETERMINE LESS FAVOURABLE HOSPITALISATION COURSE AND OUTCOME IN PATIENTS WITH ESBL PRODUCING *ENTEROBACTERIACEAE* INFECTION: PRELIMINARY RESULTS

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Hospitalisation course and outcome for patients with extended-spectrum beta-lactamase (ESBL) producing Enterobacteriaceae infection is less favourable due to extensive antibacterial resistance. This study was conducted to identify possible risk factors that could influence the hospitalisation course and outcome in these patients. The study protocol included demographic, clinical, hospitalisation, bacteriological and plasmid genetic data. The preliminary study results showed that hospitalisation course and outcome was less favourable for internal medicine profile patients with ESBL producing bacteria, TEM gene presence in the bacterial plasmid genome, patient age < 65 years and patients with infectious and musculoskeletal diseases. The study includes preliminary data only and further studies should be carried out to verify the suggested risk factors.

Key words: Enterobacteriaceae*, ESBL, hospitalization, TEM, SHV.*

Extended-spectrum beta-lactamase (ESBL) producing *Enterobacteriaceae* infection reports are increasing due to extensive antibiotic consumption and widespread ESBL bacterial gene — CTX-M, TEM and SHV mutation, determining bacterial, mostly *E. coli* and *K. pneumoniae*, resistance (Kim *et al*., 2002; Spanu *et al*., 2002; Thu Trang *et al*., 2013; Shaikh *et al*., 2015).

Hospitalisation course and outcome in patients with ESBL producing *Enterobacteriaceae* infection is considered less favourable due to extensive antibacterial drug resistance (Kim *et al*., 2002; Peralta *et al*., 2012). In order to choose the best empirical antibiotic therapy, physicians should look for factors that could indicate a less favourable hospitalisation course and outcome. An unfavourable hospitalisation course and outcome in patients with ESBL producing *Enterobacteriaceae* infection previously have been associated with severe concomitant diseases, prior administration of multiple antimicrobial agents, recent surgery, older age and presence of indwelling catheters (Skippen *et al*., 2006; Mehrgan *et al*., 2010).

The aim of this study was to identify additional possible risk factors that could influence the hospitalisation course and outcome in patients with ESBL producing *Enterobacteriaceae* infection. A cross-sectional single-centre study was conducted in Rīga East Clinical University Hospital. All consecutive ESBL producing *Enterobacteriaceae* infection cases were selected during a 6-month period (2014). A total of 136 ESBL producing *Enterobacteriaceae* infection cases were included in the study. Demographic, hospitalisation and clinical data were obtained from medical records on the day patient was discharged. Disease data were encoded according to the International Statistical Classification of Diseases (ICD-10) (Anonymous, 2015). Biomaterial (urine, bronchoalveolar washing, cerebrospinal fluid, wound swab, sputum, blood, abdominal cavity fluid, abscess sample) for bacteriological analysis was collected in transport medium

and transported to the microbiological laboratory within 24 hours. In the laboratory, each sample was cultured. Identification of the bacteria was conducted by a Vitex 2 Compact system (bioMerieux, France) and susceptibility tests by the disk diffusion method (Oxoid, UK; CLSI 2013). ESBL production was confirmed using synergy tests with ESBL and Amp-C inhibiting discs (Rosco, Denmark). Genetic analysis provided information about CTX-M, TEM and SHV bacterial gene presence in the ESBL producing *Enterobacteriaceae* plasmid genome. Bacteria selected from ESBL producing *Enterobacteriaceae* colonies were grown in 2 ml Lysogenic broth media at 37 °C overnight. Plasmid DNA were isolated by manufacturer protocol using a E.Z.N.A. plasmid Mini Kit I (Omega Bio-Tek, USA). CTX-M, SHV and TEM bacterial plasmid gene detection was performed using polymerase chain reaction (PCR) using gene specific primers (Wu *et al*., 2001; Edelstein *et al*., 2003; Anonymous, 2013). CTX-M gene detection was conducted according to Edelstein *et al*., 2003, SHV gene detection according to Nüesch-Inderbinen *et al*. (1996), and TEM gene detection according to Du *et al*. (2013). PCR products were analysed using 2% agarose gel. A study protocol with 106 parameters was completed for each ESBL-producing *Enterobacteriaceae* infection case and used for database development and statistical analysis. All p values were two-tailed and considered significant at the 5% level. The study was conducted according to the Helsinki Declaration, reviewed and approved by the local Ethics Committee.

ESBL producing *Enterobacteriaceae* were isolated from 52 (38.2%) female and 84 (61.8%) male patients at the mean age of 61.35 ± 16.92 (CI 95% 57.76–64.78) years. Mostly ESBL producing *K. pneumoniae* (n = 66, 48.53%) and *E. coli* (n = 36, 26.47%) were isolated (Fig. 1), mainly from wound (n = 44, 32.35%) biomaterial (Fig. 2), in most of the cases (n = 132, 97.06%) containing CTX-M gene (Fig. 3).

In 42 cases (30.9%), ESBL-producing *Enterobacteriaceae* were isolated from internal medicine profile department patients, in 94 cases (69.1%) from surgical profile department patients. Patients with ESBL producing *Enterobacteriaceae* infection spent an average of 60.29 ± 98.93 (CI 95% 30.82–98) days in the hospital. In 82 ESBL producing *Enterobacteriaceae* infection cases (60.29%) patients were admitted to the ICU at least once and time spent there was on average 18.57 ± 21.9 (CI 95% 10.89–26.86) days. In 54 cases (39.71%), patients were never admitted to ICU during their hospitalisation period.

Comparison between hospitalisation course and outcome in patients with ESBL producing *Enterobacteriaceae* infection in internal medicine and surgical departments is presented in Table 1.

TEM gene presence in ESBL producing *Enterobacteriaceae* plasmid genome was associated with increased mortality, compared to cases where the TEM gene was absent (p = 0.018). CTX-M and SHV gene presence in ESBL producing *Enterobacteriacea* plasmid genome was not associated with a statistically significant increase in mortality.

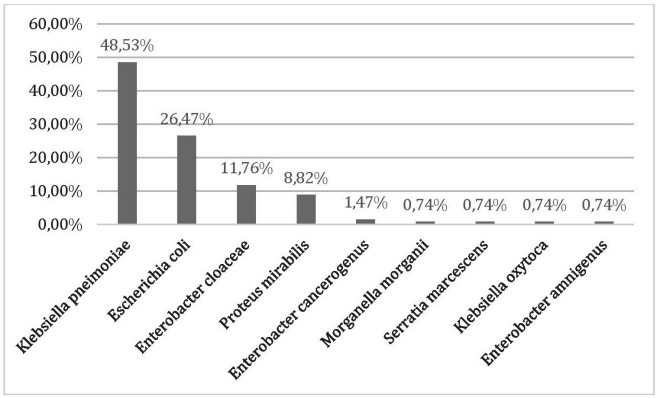

Fig. 1. Isolated ESBL producing *Enterobacteriaceae* strains (%).

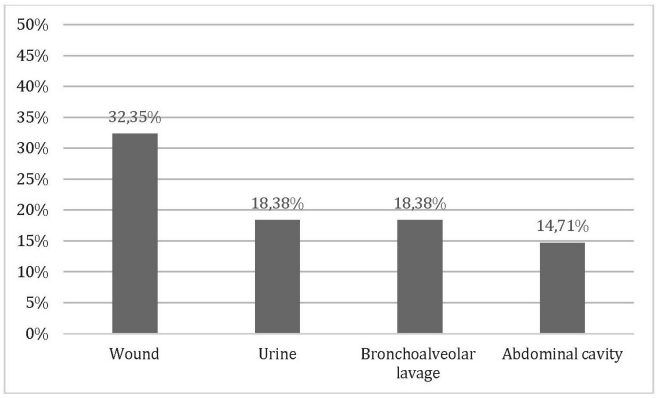

Fig. 2. Biomaterials from which ESBL producing bacteria were isolated (%).

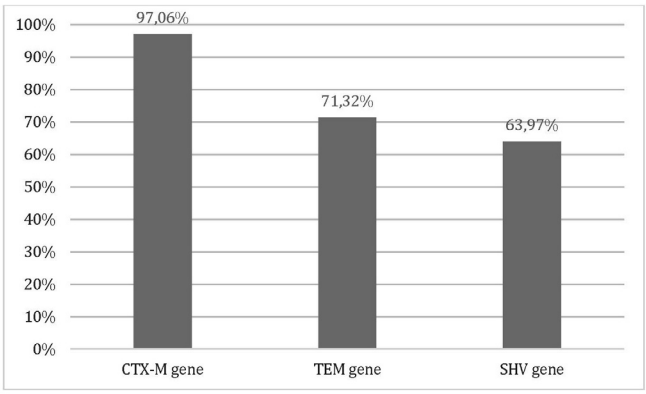

Fig. 3. Detected ESBL producing *Enterobacteriaceae* beta-lactamase plasmid genes (%).

Table 1

COMPARISON BETWEEN HOSPITALISATION COURSE AND OUTCOME IN PATIENTS WITH ESBL PRODUCING *ENTEROBACTERIACEAE* INFECTION IN INTERNAL MEDICINE AND SURGICAL DEPARTMENTS

Internal medicine department	Surgical department
More concomitant diseases ($p < 0.001$)	More cases of sepsis ($p < 0.001$)
More days spent in ICU ($p = 0.043$)	More cases of MODS ($p = 0.001$)
Increased mortality rate ($p = 0.046$)	Death at a younger age ($p = 0.01$)

Patients with ESBL producing *Enterobacteriaceae* infection aged < 65 years ($p = 0.045$) and concomitant infectious diseases ($p = 0.047$) were more frequently admitted to the ICU. Patients with ESBL producing *Enterobacteriaceae* infection admitted to the ICU ($p = 0.02$) and suffering from musculoskeletal diseases ($p = 0.002$) had increased mortality.

A less favourable hospitalisation course and outcome was observed in this study comparing to other literature sources: mean hospital stay was almost three times longer (60 *vs* 24 days) (Wu *et al*., 2014), patients were at least three times more often admitted to the ICU (60% vs 4–23%) (Concepts *et al*., 2010), stayed there almost two times longer (19 vs 12 days) (Abdalla *et al*., 2014) and had a higher mortality rate (21% vs 14%) (Peralta *et al*., 2012). This could be explained by the critically ill and immunocompromised patient predominance in the studied patient profile, including patients with severe abdominal trauma, head trauma, extremity amputations and extensive bedsores, suggesting a more severe clinical condition. Significantly more patients in this study presented with shock (13% vs 6%) (Peralta *et al*., 2012) and were older (70 years) than patients described in the other literature sources (65 years) (Peralta *et al*., 2012).

Musculoskeletal disease presence, hospitalisation in internal medicine profile departments, and TEM bacterial gene presence were associated with increased mortality in this study and could be considered as possible risk factors for less favourable hospitalisation course and outcome for patients with ESBL producing *Enterobacteriaceae* infection. These factors have not been studied as possible risk factors in any studies previously. The CTM-X bacterial gene has been associated with increased mortality in patients with ESBL producing *Enterobacteriaceae* infection in other studies (Mehrgan *et al*., 2010); however, this association was not observed in our study. Concomitant infectious diseases and age less than 65 years predisposed patients with ESBL producing *Enterobacteriaceae* infection for more frequent admission to the ICU in this study. Admission to the ICU has been identified as a risk factor for increased mortality also in other studies (Mayr *et al*., 2006; Du *et al*., 2013). Therefore, infectious diseases and younger age might be considered as risk factors for a less favourable hospitalisation course and outcome for patients with ESBL producing *Enterobacteriaceae* infection.

The study includes preliminary data only and further studies should be carried out to verify the suggested risk factors.

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RISKA FAKTORI, KAS NOSAKA SLIKTĀKU HOSPITALIZĀCIJAS GAITU UN IZNĀKUMU PACIENTIEM AR ESBL PRODUCĒJOŠU *ENTEROBACTERIACEAE* DZIMTAS BAKTĒRIJU INFEKCIJU: SĀKOTNĒJIE REZULTĀTI

Hospitalizācijas gaita un iznākums pacientiem ar paplašināta spektra beta laktamāzi (ESBL) producējošo *Enterobacteriaceae* dzimtas baktēriju infekciju ir nelabvēlīgāks plašās antibakteriālās rezistences dēļ. Šis pētījums tika veikts, lai noskaidrotu riska faktorus, kas varētu ietekmēt hospitalizācijas gaitu un iznākumu šiem pacientiem. Pētījuma protokols ietvēra demogrāfiskos, klīniskos, hospitalizācijas, bakterioloģiskos un plazmīdu ģenētiskos datus. Pētījuma rezultāti parādīja, ka hospitalizācijas gaita un iznākums mazāk labvēlīgs bija terapeitiskā profila pacientiem ar ESBL producējošo baktēriju infekciju, TEM gēna klātbūtni baktēriju plazmīdu genomā, pacientiem vecumā zem 65 gadiem, kā arī pacientiem ar papildus infekcijas un balsta – kustību aparāta slimībām. Pētījums sniedz tikai sākotnējo informāciju — nepieciešami papildus pētījumi konstatēto riska faktoru izvērtēšanai.