Escherichia coli producing SHV-type extended-spectrum β-lactamase is a significant cause of community-acquired infection

Jesús Rodríguez-Baño1, Juan Alcalá2, Jose Miguel Cisneros3, Fabio Grill4, Antonio Oliver5, Juan Pablo Horcajada6, Teresa Tórtola7, Beatriz Mirelis8, Gemma Navarro9, María Cuenca10, María Esteve11, Carmen Peña12, Ana C. Llanos3, Rafael Cantón13 and Alvaro Pascual2,14*

1Sección de Enfermedades Infecciosas, Hospital Universitario Virgen Macarena, Sevilla, Spain; 2Servicio de Microbiología, Hospital Universitario Virgen Macarena, Sevilla, Spain; 3Servicio de Enfermedades Infecciosas, Hospital Universitario Virgen del Rocio, Sevilla, Spain; 4Servicio de Enfermedades Infecciosas, Hospital Universitario Ramón y Cajal, Madrid, Spain; 5Servicio de Microbiología, Hospital Son Dureta, Palma de Mallorca, Spain; 6Servicio de Enfermedades Infecciosas, Hospital Clinic, Barcelona, Spain; 7Servicio de Microbiología, Hospital Vall d’Hebron, Barcelona, Spain; 8Servicio de Microbiología, Hospital Santa Creu i Sant Pau, Barcelona, Spain; 9Unidad de Epidemiología, Corporación Sanitaria Parc Taulí, Sabadell, Spain; 10Servicio de Microbiología, Hospital de la Ribera, Alcira (Valencia), Spain; 11Unidad de Medicina Preventiva, Hospital Universitario Germans Trias i Pujol, Badalona, Spain; 12Servicio de Enfermedades Infecciosas, Hospital Universitario de Bellvitge, Barcelona, Spain; 13Servicio de Microbiología, Hospital Ramón y Cajal, Madrid, Spain; 14Departamento de Microbiología, Facultad de Medicina, Sevilla, Spain

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Objectives: Extended-spectrum β-lactamase (ESBL)-producing Escherichia coli (ESBLEC) is an increasingly significant cause of community-acquired infection worldwide. The epidemiological features of CTX-M- and SHV-producing ESBLEC causing community-acquired infections are compared.

Methods: A multicentre cohort study including all community-acquired infections caused by ESBLEC in four geographical areas of Spain was carried out. ESBL characterization was by isoelectric focusing, PCR and sequencing. Demographics, previous healthcare contact, co-morbidity, use of antimicrobials, invasive procedures and type of infection were collected for all patients. Patients with CTX-M- and SHV-producing isolates were compared using logistic regression.

Results: One hundred and twenty-two cases (95% urinary tract infections) were included. ESBLs were characterized in 112 isolates; 77 isolates (69%) produced CTX-M, 36 (32%) produced SHV and 7 (6%) produced TEM enzymes (8 produced >1 ESBL). Patients with isolates producing CTX-M enzymes only (CTX-M group, n=70) and SHV enzymes only (SHV group, n=31) were compared. There were no differences in terms of underlying disease, previous healthcare contact, invasive procedures, antibiotic use or type of infection. Multivariate analysis including geographical area showed that a Charlson Index score of >2 (OR = 4.0; 95% CI = 1.2–12.6) was associated with SHV isolates, while age >60 (4.7; 1.7–12.5) was associated with CTX-M isolates.

Conclusions: SHV-producing ESBLEC is a significant cause of community-acquired infection in Spain; the clinical epidemiology of such isolates seems very similar to that of CTX-M-producing E. coli.

Keywords: ESBLs, CTX-M, resistance

*Corresponding author. Department of Microbiology, School of Medicine, Av/ Sanchez Pizjuan SN, Sevilla 41009, Spain. Tel: +34-954552863; Fax: +34-954377413; E-mail: apascual@us.es

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Introduction

Community-acquired infections caused by extended-spectrum β-lactamase (ESBL)-producing Escherichia coli (ESBLEC) are increasing worldwide.\(^1\) Urinary tract infections (UTIs) are the most prevalent, although other types of infection, including bacteremia, have been described. In recent years, important shifts have occurred, both in Europe and in other continents, in the prevalence and types of ESBL, with the classic SHV and TEM types of ESBL being surpassed by members of the CTX-M family.\(^2\)

The epidemiology of both ESBLEC and plasmids carrying the bla genes is complex, combining the dissemination of mobile genetic elements and clonal spread. Food, and particularly poultry, has been considered a potential source of these enzymes.\(^3\) In addition, both plasmids and bacterial transmission between closely related humans have also been demonstrated.\(^4\)

Finally, the intercontinental emergence of clonally related isolates harbouring particular ESBLs such as CTX-M-15 has been recently described in many countries.\(^5\)

Among the risk factors for acquiring a community-acquired infection caused by ESBLEC are recent antibiotic therapy, recent hospitalization, surgery and male gender.\(^6\) Most of these studies have included cases caused by E. coli expressing different ESBL families but with a clear preponderance of the CTX-M family. In fact, the dissemination of ESBLs among E. coli within the community has been linked almost exclusively with CTX-M enzymes. This fact might have led to an underestimation of the importance of other ESBLs, including SHV enzymes, in community-acquired infections caused by ESBLEC. In a previous study in Spain, an SHV-type enzyme was involved in 30% of infections caused by ESBLEC.\(^7\) CTX-M-14, followed by SHV-12, was the most frequently found enzyme in non-hospitalized infections caused by ESBLEC. Moreover, the SHV-type ESBL was the most common enzyme detected in E. coli isolates found in some retail raw meat samples in southern Spain.\(^8\) Despite these facts, there is still scarce information about the epidemiology of the SHV-type ESBL in the community. The purpose of this study was to compare the epidemiological features of community-acquired infections caused by CTX-M- and SHV-producing E. coli in a multicentre nationwide study performed in Spain.

Materials and methods

The study sites, subjects, design and definitions of the study were specified previously.\(^6\) In brief, a prospective laboratory-based cohort was conducted between February 2002 and May 2003 in 11 public Spanish hospitals serving a population of >4 million people, located in five Spanish areas including Barcelona (north-east), Madrid (central), Majorca and Valencia (east) and Seville (south). All patients in the participating hospitals with a clinical sample from which an ESBLEC strain had been isolated were considered to be eligible. Patients who had been hospitalized for more than 48 h when the sample was collected, those who had been admitted to a hospital for >48 h during the preceding month and those with a previous isolation of ESBLEC were excluded. Demographic data, predisposing conditions, invasive procedures, previous healthcare contact and infection type were collected. The study protocol was approved by the ethics committees of the participating centres.

Further to the microbiological determinations described in the previous study,\(^6\) the clonal relationship between the isolates was determined by repetitive extragenic palindromic PCR (REP-PCR), as described previously.\(^1\) Isolates that were determined to be clonally related by REP-PCR were also studied by PFGE.\(^2\) β-Lactamase characterization was carried out by isoelectric focusing (Pharmacia PhastSystem) of the sonicated extract, and PCR of the bla genes.\(^9\) Amplicons were further sequenced for the final characterization of the bla gene.\(^9\)

Univariate comparisons were performed using the Mann–Whitney U-test and \(\chi^2\) test (or Fisher’s exact test) for continuous and discrete variables. Multivariate analyses were performed by logistic regression using a backward stepwise process. Interactions between paired variables were investigated. Data were analysed using the SPSS 13.0 software package.

Results

A total of 122 cases was included; the 112 cases in which the ESBLs were characterized were included in this analysis. The CTX-M ESBL was detected in 77 isolates (69%). Of these, 65 belonged to the CTX-M-9 group (40 CTX-M-9 and 25 CTX-M-14), 9 to the CTX-M-1 group (4 CTX-M-32, 2 CTX-M-3, 2 CTX-M-15 and 1 CTX-M-1) and 3 were not sequenced. Thirty-six (32%) belonged to the SHV family (31 SHV-12 and 5 SHV-2) and 7 (6%) belonged to the TEM family (4 TEM-116, 2 TEM-52 and 1 TEM-4). Eight isolates produced more than one ESBL. The percentage of isolates producing SHV enzymes was 22% in the north-east (5/23), 26% in the central area (7/27), 36% in the south (16/44) and 44% in the east (8/18). Clonal relationships were few since only 107 REP-PCR patterns were obtained.

For further analysis, cases of E. coli expressing only the CTX-M and SHV ESBLECs (70 and 31 patients, respectively) were compared. The univariate analysis of microbiological features is shown in Table 1. Among them cefotaxime susceptibility was significantly higher in SHV-producing isolates and ceftazidime susceptibility was significantly higher in CTX-M producers, as expected. No significant differences with other antimicrobial agents were observed.

Predisposing factors by ESBL family were also evaluated. In the univariate analysis, only the severity of the underlying chronic condition was significantly different between both groups; the frequency of a Charlson Index >2 was significantly higher in the SHV group. There were no significant differences observed for the other variables evaluated. More precisely, the percentage of cases with no significant previous healthcare relation (70% for CTX-M cases and 68% for SHV cases) was similar. When a multivariate analysis was performed, the only variables independently associated with SHV-producing isolates were a Charlson Index score of >2 [OR (95% CI) = 4.0 (1.2–12.6), \(P = 0.01\)] and age >60 years as a protective factor [OR (95% CI) = 0.2 (0.08–0.5), \(P = 0.002\)]. No differences were found for type of infection caused by SHV and CTX-M producers; UTIs were the most prevalent infection (97% and 94%, respectively). No patients died.

Discussion

ESBLEC is an increasingly relevant group of community pathogens worldwide. In the last few years, ESBL patterns have been changing rapidly with CTX-M enzymes replacing TEM and
Because of the emergence of infections caused by CTX-M- and SHV-producing isolates, but, more recently, ESBLs in many European countries. Most of these infections were associated with clonally unrelated isolates, but, more recently, countries. In predisposed patients, ESBL-producing E. coli has been recognized as an emerging international clone group expressing specific ESBLs such as CTX-M-15.

A case–control case study was performed in 11 hospitals in Spain to investigate the risk factors for all types of community-acquired infection caused by ESBL-producing E. coli. In predisposed patients, ESBL-producing E. coli was a notable cause of community-acquired infections, particularly UTIs. The estimated population-based incidence of community-acquired infections caused by these microorganisms was 2.2 cases per 100000 population per year. Risk factors associated with infection were: age >60 years, female sex, diabetes mellitus, recurrent UTI, previous invasive procedures of the urinary tract, and previous receipt of antimicrobials. In that study, all ESBL-producing E. coli were considered globally, and the results were not stratified by type of ESBL, since this information was unavailable at the time. In this and other studies developed in Spain, around 30% of infections caused by ESBL-producing E. coli involved an SHV-type enzyme, and the most prevalent was SHV-12.

Because of the emergence of infections caused by CTX-M-producing isolates, the importance of SHV-producing E. coli as a cause of community-acquired infections may be being underestimated. In this study, the epidemiological and clinical data of community-acquired infections caused by CTX-M- and SHV-producing E. coli were compared. No relevant differences were observed between the two groups. Features associated with SHV-producing isolates were age >60 years (protective) and a more severe underlying disease. Of note, previous use of antibiotics, and particularly fluoroquinolones, was similar in both groups, reflecting their similarly high frequency of resistance to these compounds. Since no relevant differences were found between the two groups of isolates, it might be speculated that the risk factors for acquiring infections by CTX-M- and SHV-producing E. coli could also be similar. Food of animal origin has been demonstrated to favour the dissemination of SHV mutants as the predominant ESBLs in many European countries. Most of these infections were associated with clonally unrelated isolates, but, more recently, E. coli O25:H4-ST131 has been recognized as an emerging international clone group expressing specific ESBLs such as CTX-M-15.

A diversity of plasmid incompatibility groups harbouring ESBL genes and substantial variability in the genetic environment have been documented in Spain. Since some of the characterized plasmids carry CTX-M genes belong to the same incompatibility groups as those described for SHV-12, such as IncI1, the possibility of a potential epidemic dissemination of specific plasmids encoding SHV-type ESBLs in E. coli could not be ruled out.

In conclusion, SHV-producing ESBL-producing E. coli is a significant cause of community-acquired infection in Spain; the clinical epidemiology of these isolates seems very similar to that of CTX-M-producing ESBL. The data suggest that SHV-producing E. coli are also spreading in the community.

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**Table 1.** Microbiological data of 101 patients with infections due to ESBL-producing E. coli according to type of enzyme

<table>
<thead>
<tr>
<th></th>
<th>CTX-M group, n = 70</th>
<th>SHV group, n = 31</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Presence of TEM-1</td>
<td>35 (50)</td>
<td>16 (52)</td>
<td>0.1</td>
</tr>
<tr>
<td>Susceptibility to</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>cefotaxime</td>
<td>8 (12)</td>
<td>26 (84)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>ceftazidime</td>
<td>59 (88)</td>
<td>5 (16)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>amoxicillin/clavulanate</td>
<td>47 (70)</td>
<td>24 (77)</td>
<td>0.4</td>
</tr>
<tr>
<td>piperacillin/tazobactam</td>
<td>66 (99)</td>
<td>29 (94)</td>
<td>0.2</td>
</tr>
<tr>
<td>trimethoprim/sulfamethoxazole</td>
<td>28 (42)</td>
<td>8 (26)</td>
<td>0.1</td>
</tr>
<tr>
<td>gentamicin</td>
<td>58 (87)</td>
<td>24 (77)</td>
<td>0.2</td>
</tr>
<tr>
<td>tobramycin</td>
<td>60 (90)</td>
<td>28 (90)</td>
<td>0.9</td>
</tr>
<tr>
<td>amikacin</td>
<td>67 (100)</td>
<td>31 (100)</td>
<td>—</td>
</tr>
<tr>
<td>nalidixic acid</td>
<td>5 (7)</td>
<td>1 (3)</td>
<td>0.6</td>
</tr>
<tr>
<td>ciprofloxacin</td>
<td>23 (34)</td>
<td>10 (32)</td>
<td>0.8</td>
</tr>
<tr>
<td>meropenem</td>
<td>67 (100)</td>
<td>31 (100)</td>
<td>—</td>
</tr>
</tbody>
</table>

Data are expressed as number of isolates (percentage).

aSusceptibility testing was studied in 67 isolates producing CTX-M.

bAccording to CLSI breakpoints.
de Investigaciones Sanitarias; and grants 75/04 and PI0048/2008 from the Junta de Andalucía.

Transparency declarations

None to declare.

References


