Faecal carriage of
ESBL-producing Enterobacteriaceae
in the community

Liège - Belgium

Cécile Meex
Medical Microbiology (Prof. De Mol)
CHU Liège - Belgium
Introduction

- Infections due to ESBL-producing Enterobacteriaceae (ESBL-E)
  - acute-care hospitals
  - other healthcare facilities (nursing homes...)
  - community

- Global programme related to appropriate use of antibiotics in the community in Belgium
  → Prevalence of ESBL-E colonizing the digestive tract
Introduction

Aim of the study:

- To determine the carriage rate of ESBL-E in community patients’ faecal flora.
- To characterize the detected ESBLs
Materials and Methods
Samples

- 6 general practitioners from 10 independant surgeries
- 284 patients chosen at random without relation to the reason of their consultation

→ Collection of faecal specimens and filling of a case report form.
Inoculation

- Faecal suspension in 1 ml sterile saline
- 50 µl onto 3 different selective culture media:
  - ChromID ESBL agar (bioMérieux)
  - Bi-plate media (AES Chemunex): MacConkey agar with ceftazidime and Drigalski agar with cefotaxime
Identification and detection of ESBL-E

- Identification of all the Enterobacteriaceae performed by Vitek2 (bioMérieux)
- ESBL production screened by the combined double disk synergy method
- Antibiotic susceptibility testings of the ESBL-E performed by Vitek2 (bioMérieux)
Genotypic characterization (1)

- DNA extraction for each ESBL-E with the QIAamp DNA mini kit (Qiagen)

- Molecular detection of \( \text{blaTEM} \), \( \text{blaSHV} \) and \( \text{blaCTX-M} \) but also of beta-lactamase of type BEL, VEB, GES or OXA 1/2/10
Genotypic characterization (2)

- When a PCR was positive:
  - Purification of the amplified DNA
  - Sequencing
  - Analysis of the nucleotid sequence
Genotypic characterization (3)

- Deduced amino-acid sequence compared to that present in public database or in the Lahey website (www.lahey.org/studies).

- Identification of the beta-lactamase
Results
Phenotypic results

- 284 faecal samples
- 53 Enterobacteriaceae isolated from 46 samples
- 25 of these: phenotypically characterized as ESBL producers

The 25 ESBL-E originated from 20 patients (7.04%)
Genotypic results (1)

Results of PCR and sequencing:

<table>
<thead>
<tr>
<th>Species (Number of isolates)</th>
<th>Type of beta-lactamase</th>
<th>TEM</th>
<th>CTX-M</th>
<th>TEM and CTX-M</th>
<th>SHV, BEL, VEB, GES or OXA 1/2/10</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>E.coli (19)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>TEM-1 (7)</td>
<td></td>
<td>CTX-M-1 (4)</td>
<td>TEM-1 and CTX-M-1 (1)</td>
<td>/</td>
</tr>
<tr>
<td></td>
<td>TEM-19 (1)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>TEM-52 (4)</td>
<td></td>
<td>CTX-M-15 (2)</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>E.aerogenes (1)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>TEM-52 (1)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>P.mirabilis (3)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>TEM-24 (3)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>S. fonticola (1)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Genotypic results (2)

- Distribution of the ESBLs among the 25 ESBL-E

- 8 *E. coli*
- 1 *S. fonticola*

![Pie chart showing distribution of ESBLs among 25 ESBL-E strains.]

- TEM-19: 3
- TEM-24: 5
- TEM-52: 5
- CTX-M-1: 2
- CTX-M-15: 1
- No TEM, SHV, CTX-M, BEL, VEB, GES or OXA 1/2/10 ESBL: 9
## Antibiotic susceptibility profiles

<table>
<thead>
<tr>
<th></th>
<th>Resistance to:</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Trimethoprim-sulfamethoxazole</td>
<td>Quinolones</td>
<td>Aminoglycosides</td>
</tr>
<tr>
<td><strong>E. coli (20)</strong></td>
<td>15 (75%)</td>
<td>6 (30%)</td>
<td>5 (25%)</td>
</tr>
<tr>
<td><strong>P. mirabilis (3)</strong></td>
<td>3 (100%)</td>
<td>3 (100%)</td>
<td>0 (0%)</td>
</tr>
<tr>
<td><strong>E. aerogenes (1)</strong></td>
<td>0 (0%)</td>
<td>0 (0%)</td>
<td>0 (0%)</td>
</tr>
<tr>
<td><strong>S. fonticola (1)</strong></td>
<td>0 (0%)</td>
<td>0 (0%)</td>
<td>0 (0%)</td>
</tr>
</tbody>
</table>
## Expected risk factors for ESBL-E carriage

<table>
<thead>
<tr>
<th>Expected risk factors for ESBL-E carriage</th>
<th>Among ESBL-E carriers (n=20)</th>
<th>Among ESBL-E negative carriers (n=264)</th>
<th>Fischer test</th>
</tr>
</thead>
<tbody>
<tr>
<td>Recent consumption of antibiotics</td>
<td>4</td>
<td>74</td>
<td>p &gt; 0.05</td>
</tr>
<tr>
<td>Recent hospitalization</td>
<td>2</td>
<td>15</td>
<td>p &gt; 0.05</td>
</tr>
<tr>
<td>Recent trip abroad</td>
<td>1</td>
<td>43</td>
<td>p &gt; 0.05</td>
</tr>
<tr>
<td>Pets at home</td>
<td>11</td>
<td>135</td>
<td>p &gt; 0.05</td>
</tr>
<tr>
<td>Mean age</td>
<td>57</td>
<td>50</td>
<td></td>
</tr>
</tbody>
</table>
Discussion and conclusions
Prevalence rate (1)

- Liège, Belgium (2007):
  Unrelated outpatients: 6.7%

- Rodriguez-Baño et al., Spain (2008):
  Unrelated nonhospitalized persons: 7.4%

- Tian et al., China (2008): elderly people in community settings: 7%

- Valverde et al., Spain (2003): Outpatients: 5.5%
Prevalence rate (2)

- Kinshasa, Congo (2006):
  - Hospitalized patients: 33.1%
  - Non hospitalized persons: 13.1%

Predominant isolated ESBL: CTX-M-15
Isolated ESBL-E

- *E. coli* accounted for the majority of ESBL-E isolates

- Various ESBL genes were identified
  - TEM- and CTX-M-derived enzymes predominant

- 36% of the phenotypically characterized ESBL-E did not possess any ESBL of type:
  - TEM, SHV, CTX-M, BEL, VEB, GES or OXA 1/2/10
Antibiotic susceptibility testings

- High level of sulfamethoxazole-trimethoprim resistance among the isolated ESBL-E (72%)
  - All the CTX-M producing Enterobacteriaceae were sulfamethoxazole-trimethoprim resistant.

- Co-resistance with quinolones observed for 9 Enterobacteriaceae (36%)
Conclusions

- ESBL-related antimicrobial resistance mechanism(s) among Enterobacteriaceae in the community is a reality.

- High prevalence of ESBL-E faecal carriage among the non hospitalized population should be taken into account in treatment recommendations in ambulatory medicine.
  - Modification of the empiric antimicrobial therapy?
  - Patients screening before hospitalization?

- Efforts of vigilance should be made to identify and control spread from these community reservoirs.