Shigellosisis a major source of gastroenteritis throughout the world (14). Extended-spectrum β-lactamases (ESBLs), including cephalosporinases (CTX-M), confer resistance to extended-spectrum cephalosporins and significantly compromise the treatment options for shigellosis. Numerous ESBLs have been described among Enterobacteriaceae (2, 8, 13); however, only a single CTX-M-producing Shigella isolate has been reported in the United States (10).

From 1999 to 2007, 3,880 Shigella isolates were screened for antimicrobial susceptibility to 14 to 17 antimicrobials by broth microdilution (Sensititre; Trek Diagnostics, Westlake, OH). Six isolates displayed decreased susceptibility (MIC ≥ 2 mg/liter) to ceftriaxone (Table 1). The six case-patients included three males and two females (gender information was unavailable for one patient), and the median age was 3 years (range, 1 to 8 years). Additional details were available for five patients. Three of the five (60%) were hospitalized, and one was admitted twice. One patient had an adopted sibling from Russia but had not traveled herself. The second patient traveled to a neighboring state prior to illness onset, and the third reported no travel. Of the nonhospitalized patients, one was an asymptomatic adoptee from China and the second reported no travel. Two patients received antimicrobial therapy: ceftriaxone, and trimethoprim-sulfamethoxazole for one patient, azithromycin for the other patient.

PCR analysis was used to screen the six isolates for 13 different classes or groups of β-lactam genes, and PCR results were confirmed by DNA sequencing (1, 5, 11, 12, 16, 18–21). Four isolates were positive for the bla<sub>CTX-M-15</sub> gene, while two were positive for the bla<sub>CTX-M-14</sub> gene (Table 1). All four bla<sub>CTX-M-15</sub> isolates were PCR positive for non-ESBL bla<sub>TEM-1</sub> genes. Both bla<sub>CTX-M-14</sub> isolates were PCR positive for non-ESBL bla<sub>OXA-1</sub> genes, and a single isolate was positive for both bla<sub>TEM-1</sub> and bla<sub>OXA-1</sub>. By pulsed-field gel electrophoresis (PFGE) analysis, all three S. sonnei and all three S. flexneri isolates demonstrated distinct patterns (data not shown) (15).

All six bla<sub>CTX-M</sub> genes were determined to be plasmid encoded (6). The non-ESBL β-lactamases (OXA-1, TEM-1) did not transfer and were not encoded on the same CTX-M plasmids. All three S. sonnei plasmids and two of the S. flexneri plasmids harbored only the CTX-M-associated resistance. The remaining S. flexneri plasmid contained additional determinants conferring resistance to trimethoprim-sulfamethoxazole and gentamicin.

All three S. sonnei plasmids were incompatibility type Inc1I and approximately 90 kb in size (plasmid pulsed-field gel electrophoresis) (Table 1) (4). Plasmid multilocus sequence typing (pMLST) identified them as novel sequence types designated as ST31 complex. The plasmid from AM22451 contained several point mutations in one allele, necessitating the ST32 designation within the ST31 clonal complex (http://pubmlst.org/plasmid) (7). Of the three S. flexneri plasmids, the bla<sub>CTX-M-15+</sub> positive plasmid was a 165-kb IncA/C plasmid, while the two bla<sub>CTX-M-14+</sub> positive plasmids were identical 75-kb IncFII plasmids. CTX-M-14 and CTX-M-15 are the most common types of cefotaximases identified among Shigella isolates (9, 17, 22), and Inc1I plas-

---

**TABLE 1.** Characterization of CTX-M-positive Shigella isolates, transformants, and CTX-M-encoding plasmids<br>

<table>
<thead>
<tr>
<th>Isolate no.</th>
<th>Shigella species</th>
<th>State, yr isolated</th>
<th>MIC (µg/ml)</th>
<th>Additional resistance profile</th>
<th>β-Lactamase</th>
<th>Plasmid size (kb)</th>
<th>Plasmid incompatibility type (sequence type)</th>
</tr>
</thead>
<tbody>
<tr>
<td>DH10B</td>
<td>flexneri</td>
<td>MA, 2002</td>
<td>≤0.06</td>
<td>STR, TEM-1, OXA-1</td>
<td>CTX-M-15, TEM-1, OXA-1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>AM13291</td>
<td>flexneri</td>
<td>MA, 2002</td>
<td>16</td>
<td>AMP, AUG, COT, GEN, TIO</td>
<td>CTX-M-15</td>
<td>165</td>
<td>A/C</td>
</tr>
<tr>
<td>DH-13291</td>
<td>sonnei</td>
<td>WI, 2003</td>
<td>32</td>
<td>OXA-1</td>
<td>CTX-M-14, TEM-1, OXA-1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>AM19035</td>
<td>flexneri</td>
<td>WI, 2003</td>
<td>32</td>
<td>AMP, CHL, COT, FIS, TIO</td>
<td>CTX-M-14</td>
<td>165</td>
<td>A/C</td>
</tr>
<tr>
<td>AM20369</td>
<td>sonnei</td>
<td>WI, 2004</td>
<td>32</td>
<td>AMP, COT, FIS, NAL, TIO</td>
<td>CTX-M-14, TEM-1, OXA-1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>AM20369</td>
<td>sonnei</td>
<td>WI, 2004</td>
<td>&gt;64</td>
<td>AMP, STR, TEM-1</td>
<td>CTX-M-15</td>
<td>165</td>
<td>A/C</td>
</tr>
<tr>
<td>AM22451</td>
<td>sonnei</td>
<td>NH, 2005</td>
<td>&gt;64</td>
<td>AMP, COT, FIS, NAL, TIO</td>
<td>CTX-M-15, TEM-1, OXA-1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>AM22451</td>
<td>sonnei</td>
<td>NC, 2005</td>
<td>&gt;64</td>
<td>AMP, COT, FIS, NAL, TIO</td>
<td>CTX-M-15</td>
<td>165</td>
<td>A/C</td>
</tr>
<tr>
<td>DH-22855</td>
<td>flexneri</td>
<td>CA, 2006</td>
<td>&gt;64</td>
<td>AMP, COT, FIS, NAL, TIO</td>
<td>CTX-M-15, TEM-1, OXA-1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>AM26336</td>
<td>flexneri</td>
<td>NE, 2006</td>
<td>16</td>
<td>AMP, CHL, COT, FIS, GEN, TIO</td>
<td>CTX-M-14, TEM-1, OXA-1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>DH-26336</td>
<td>—</td>
<td>—</td>
<td>32</td>
<td>AMP, STR, TEM-1</td>
<td>CTX-M-14</td>
<td>165</td>
<td>A/C</td>
</tr>
</tbody>
</table>

---

**Notes:**
- AMP, ampicillin; AUG, amoxicillin-clavulanic acid; CHL, chloramphenicol; CAZ, cefazidime; COT, trimethoprim-sulfamethoxazole; CRO, ceftriaxone; CTX, cefotaxime; FEP, cefepime; FIS, sulfisoxazole; GEN, gentamicin; KAN, kanamycin; NAL, nalidixic acid; STR, streptomycin; TET, tetracycline; TIO, cefotiofur.
- Additional drugs tested: AMI, amikacin; CIP, ciprofloxacin; FOX, cefoxitin.
- * — not applicable.

---

**Identification and Characterization of CTX-M-Producing Shigella Isolates in the United States**

---

**Downloaded from** [http://aac.asm.org/](http://aac.asm.org/) **on May 18, 2021 by guest**
We thank the NARMS participating public health laboratories for submitting the isolates, Evangeline Sowers for confirming the Shigella species, Anne Whitney for DNA sequencing, Lisa Theobald and the rest of the PulseNet team, and Rebecca Howe for their assistance. This work was supported by an interagency agreement between the CDC and the FDA Center for Veterinary Medicine.

REFERENCES

Jason P. Folster* Gary Pecic Amy Krueger Regan Rickert

*Phone: (404) 639-4948 Fax: (404) 639-4290

Karen Burger

Wake Forest School of Medicine Winston-Salem, North Carolina

Alessandra Carattoli

Department of Infectious, Parasitic and Immune-Mediated Diseases Istituto Superiore di Sanita Rome, Italy

Jean M. Whichard

Division of Foodborne, Bacterial and Mycotic Diseases Centers for Disease Control and Prevention

Atlanta, Georgia 30333

Karen Burger

Wake Forest School of Medicine Winston-Salem, North Carolina

Alessandra Carattoli

Department of Infectious, Parasitic and Immune-Mediated Diseases Istituto Superiore di Sanita Rome, Italy

Jean M. Whichard

Division of Foodborne, Bacterial and Mycotic Diseases Centers for Disease Control and Prevention

Atlanta, Georgia

*Phone: (404) 639-4948 Fax: (404) 639-4290 E-mail: guru@cdc.gov

Published ahead of print on 8 March 2010.