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#### SUPPLEMENTARY MATERIAL

To view supplementary material for this article, please visit <https://doi.org/10.1017/ice.2017.285>

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## Perirectal Screening for Carbapenem-Resistant Enterobacteriaceae Obtained From 100 Consecutive Healthy Pregnant Women in Labor at a Brooklyn Hospital: Results and Risk Factors

*To the Editor*—Carbapenem-resistant Enterobacteriaceae (CRE) have emerged as a worldwide problem; they have been

associated with antibiotic use, long-term and acute-care hospitalization; and they have spread to endemic areas.<sup>1</sup> The spread of CRE in communities is a public health threat because CRE infections have limited treatment options and increased mortality.<sup>2</sup>

In our hospital in 2016, a premature baby developed sepsis on day of life 29 and died within 24 hours. A blood culture grew CRE *Klebsiella pneumoniae*. Subsequent investigations into the source of the organism in the neonatal intensive care unit (NICU) did not find any CRE isolates from multiple environmental surface cultures, including isolettes, monitors, weighing scales, sinks, etc. We therefore decided to investigate the prevalence of CRE rectal carriage in our maternity population. Our hypothesis was that the baby was colonized at birth from exposure to maternal colonization with the organism.

We screened 100 consecutive pregnant women delivering babies at The Brooklyn Hospital Center (TBHC) to investigate whether CRE had become a significant clinical issue in this population.

#### METHODS

Pregnant women admitted in labor were approached and requested to have a perirectal sample taken for surveillance purposes. A convenience sample of 100 sequential perirectal specimens was taken. The study plan was submitted to the TBHC Institutional Review Board as a quality assurance/quality improvement study and was given waived status. The study was anonymous and required only verbal consent. We also administered an epidemiology questionnaire consisting of 15 questions related to travel history, hospitalizations, surgery, and antibiotics during pregnancy. Perirectal swabs were refrigerated and processed within 24 hours of collection. We used the Centers for Disease Control and Prevention (CDC) laboratory protocol for detection of CRE from rectal swabs.<sup>3</sup> Samples that screened positive for CRE were identified using the Vitek system (bioMérieux, Marcy-l'Étoile, France). Carbapenem minimum inhibitory concentrations (MICs) for these isolates were determined using the Etest method (bioMérieux). Carbapenem-resistant isolates were screened by polymerase chain reaction (PCR) for *bla*<sub>KPC</sub>, *bla*<sub>NDM</sub> and *bla*<sub>OXA48</sub> as previously described.<sup>4</sup>

#### RESULTS

We identified 2 specimens that grew CRE organisms (both *Klebsiella pneumoniae*), for a prevalence of CRE colonization in 2% of the population with a confidence interval of 0.2%–7.0% using the Clopper-Pearson method. The MICs of ertapenem and imipenem were >32 µg/mL for both isolates. In addition, PCR testing revealed the presence of *bla*<sub>KPC</sub> in both isolates; other carbapenemase genes were not detected. The 2 women colonized with CRE had no history of travel or antibiotics during pregnancy, but 1 of them had been hospitalized in the previous 6 months (Table 1).

TABLE 1. Carbapenem-Resistant Enterobacteriaceae Prevalence Among Pregnant Women in a Brooklyn, New York, Hospital

Variable	CRE Positive (n = 2), No. (%)	CRE Negative (n = 98), No. (%)
Travel outside the United States during pregnancy	0	9 (9)
Hospitalized in the United States in the previous 6 months	1 (50)	8 (8)
Any antibiotics during pregnancy	0	33 (34)

NOTE. CRE, carbapenem-resistant Enterobacteriaceae.

## DISCUSSION

The rate of asymptomatic CRE colonization in this cohort of pregnant women is concerning. New York State has been collecting data on the prevalence of CRE isolates in New York hospitals since 2013. While nearly all hospitals in New York State have had at least 1 CRE isolate, the majority of CRE isolates are found in New York City, particularly the borough of Brooklyn. Publicly reported data from our hospital in 2014 revealed 27 CRE community-onset isolates from all sites in 99,800 patient days and an unadjusted community onset CRE rate of 1.43 per 1,000 admissions.<sup>5</sup> These isolates are from clinical cultures and not the result of routine screening of the population. The rate of asymptomatic colonization could be considerably higher, which could be consistent with the higher rate of carriage in our maternal population.

Around the world, prevalence rates of CRE colonization have a wide range. A recent study in the United States in 7 different communities found a range from 0.82 (Oregon) to 4.80 (Georgia) per 100,000 population in 2013.<sup>6</sup> Comparison data on the prevalence of CRE around the world are not readily available for most countries. However, CRE have been identified in Europe, India and Pakistan, Israel, and the Middle East, South and Central America, China, and Africa.<sup>7</sup>

Many studies describe CRE prevalence and CRE outbreaks in hospitals. Risk factors for CRE carriage include healthcare exposures such as short-stay and long-term acute-care hospitalization, long-term care facilities, surgical procedures, indwelling devices, and travel to endemic areas. Recent publications have identified CRE carriage and outbreaks in NICUs, particularly in China.<sup>8</sup> The prevalence of CRE carriage in a study in Turkey was 2.6% among NICU patients and 3.6% among PICU patients.<sup>9</sup> Subsequent infection with CRE organisms occurred in 18% of colonized NICU patients and 39% of pediatric intensive care units (PICU) patients, with a 16.5% mortality rate.

Although much of the literature describes CRE carriage in association with exposure to health care, antibiotics, and comorbid conditions. Once CRE have been introduced into a hospital population, they may subsequently spread to the general population. A laboratory-based surveillance study in Colorado<sup>10</sup> found that 6 of 10 patients identified with CRE

between 2014 and 2016 were community associated and not healthcare associated. One of these patients was a pregnant woman.

Although a CRE prevalence rate of 2% in pregnant women may not seem significant, this information has already changed our empiric antibiotic therapy for very sick neonates with suspected gram-negative infections. In addition, the question has arisen of whether we should routinely screen women or a NICU babies for CRE carriage. Identifying CRE carriage would enable us to apply infection prevention isolation techniques to prevent hospital spread. However, CRE screening of all women would be expensive, and contact isolation of exposed babies would be burdensome for the busy staff.

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## Hypervirulent *Clostridium difficile* Strain Has Arrived in Brazil

To the Editor—*Clostridium difficile* is an important cause of diarrhea in hospitals all over the world. It is among the most common organisms related to healthcare-associated infections in the United States and represents a serious public health concern.<sup>1</sup> Clinical manifestations of *C. difficile* infection (CDI) range from asymptomatic carriage, mild or moderate diarrhea, to fulminant colitis.<sup>2</sup>

In the last 20 years, hypervirulent isolates of *C. difficile* have been increasingly reported, mostly in North America and Europe.<sup>1</sup> The main hypervirulent strain was named ribotype 027 (North American BI, NAP1/027). This epidemic strain was also reported in Asia, providing evidence of worldwide spread.<sup>3</sup> Patients infected with the 027 strain are 3 times more likely to have severe disease than those infected with non-027 strains.<sup>4,5</sup> This finding was linked to increased production of A, B, and binary toxins, in association with a mutation in the gene regulating of the expression of these toxins, leading to overproduction. However, few studies have investigated the presence of hypervirulent strains of *C. difficile* in developing countries, particularly in Latin America.<sup>6,7</sup> Here, we make the first report of the detection of the hypervirulent *C. difficile* strain in Brazil.

## METHODS

### Setting and Patients

The hypervirulent *C. difficile* strain was noticed during the conduction of a point-prevalence multicenter study in Brazil. Fecal samples were tested with a commercial real-time PCR kit (Xpert *C. difficile* test; Cepheid, Sunnyvale, CA) in accordance with the manufacturer's instructions. In addition, samples were also submitted to *C. difficile* culture, using absolute alcohol at room temperature, subcultured in CM0601 *C. difficile* agar (Oxoid), enriched with 7% blood horse, D-cycloserine, and cefoxitin for 48 hours under an anaerobic atmosphere. Species identification of suspected colonies was confirmed by matrix-assisted laser desorption/ionization (Brucker Daltonics, Bremen, Germany).

Presumptive diagnosis of infection with the 027 strain was conducted with GeneXpert and confirmed with in-house polymerase chain reaction (PCR), designed to amplify the *tcdA* and *tcdB* toxin genes and *tcdC* negative regulator toxin gene.<sup>10</sup> The PCR products were purified using the enzyme Exo-SAP-IT (Thermo Fischer, Waltham, MA) and sequenced using the BigDye Terminator version 3.1 Cycle Sequencing Kit (Life Technology, Carlsbad, CA). *Clostridium difficile* ATCC 9689 was used as a control. Sequence type was determined by multilocus sequencing typing,<sup>10</sup> and an online database was used to assign sequence typing (<http://pubmlst.org/cdifficile>).

## RESULTS

The index case with a GeneXpert 027 positive result was recovered from a 68-year-old man who had a 2-month history of diarrhea and was heavily exposed to antimicrobial drugs (ie, vancomycin, metronidazole, amikacin and meropenem). He was admitted to a university hospital in Porto Alegre, Southern Brazil. At the time of sample collection, the patient had a C-reactive protein level of 109.7 mg/L and a leukocyte count of 8,000 cells/mL. The isolate harbored *tcdA* and *tcdB* toxin genes and the *tcdC* deletion (frameshift deletion, 18 nucleotides) that indicates toxin overproduction (Figure 1). The isolate was classified as ST67, belonging to clade 2. This clade contains a high diversity of sequence typing that also includes ST1 (NAP1/027).

In another hospital located in the same city 1 month later, we identified another case of ribotype 027 using GeneXpert. Unfortunately, this case occurred in an outpatient, and we were unable to recover any sample for DNA sequencing. The patient had been using the following antibiotics >1 month for a difficult-to-treat otitis: amoxicillin, amoxicillin/clavulanate, and cefuroxime. She arrived in the emergency room with watery diarrhea lasting for 7 days. In the 3 days before admission, she reported abdominal pain, nausea, and malaise. Blood tests revealed 15,580 leucocytes/mL (81.4% neutrophils) and 349,000 platelets/mL. Abdominal ultrasound showed signs of colitis. The patient was discharged with metronidazole and was lost to follow-up.