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Original article

Clostridium difficile outbreak caused by NAP1/BI/027 strain and non-027 strains in a Mexican hospital

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ABSTRACT

Background: *Clostridium difficile* infections caused by the NAP1/B1/027 strain are more severe, difficult to treat, and frequently associated with relapses.
Methods: A case–control study was designed to examine a *C. difficile* infection (CDI) outbreak over a 12-month period in a Mexican hospital. The diagnosis of toxigenic CDI was confirmed by real-time polymerase chain reaction, PCR (Cepheid Xpert *C. difficile*/Epi).
Results: During the study period, 288 adult patients were evaluated and 79 (27.4%) patients had confirmed CDI (PCR positive). *C. difficile* strain NAP1/B1/027 was identified in 31 (39%) of the patients with confirmed CDI (240 controls were included). Significant risk factors for CDI included any underlying disease ($p < 0.001$), prior hospitalization ($p < 0.001$), and antibiotic ($p < 0.050$) or steroid ($p < 0.001$) use. Laboratory abnormalities included leukocytosis ($p < 0.001$) and low serum albumin levels ($p < 0.002$). Attributable mortality was 5%. Relapses occurred in 10% of patients. Risk factors for *C. difficile* NAP1/B1/027 strain infections included prior use of quinolones ($p < 0.03$).

Risk factors for CDI caused by non-027 strains included chronic cardiac disease ($p < 0.05$), chronic renal disease ($p < 0.009$), and elevated serum creatinine levels ($p < 0.003$). Deaths and relapses were most frequent in the 027 group (10% and 19%, respectively).
Conclusions: *C. difficile* NAP1/BI/027 strain and non-027 strains are established pathogens in our hospital. Accordingly, surveillance of *C. difficile* infections is now part of our nosocomial prevention program.

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Introduction

Clostridium difficile infections (CDI) are the leading worldwide cause of healthcare-associated diarrhea and in some countries CDI surpass all other healthcare-associated infections (HCAI).[1] A recent prevalence survey of HCAI conducted across 183 hospitals determined that *C. difficile* was the most frequently reported infectious agent, responsible for 12.1% of all HCAI.[1]

In the United States of America (USA) during 2011, 15,461 CDI cases were reported with 24.2% of cases having an onset during hospitalization. Incident CDI cases were estimated to be >450,000 with an estimated >29,000 deaths.[2] However, the emergence of the *C. difficile* NAP1/B1/027 strain in 2000 changed the morbidity and mortality rates associated with CDI.[3,4]

Since 2004, the role of other emergent *C. difficile* strains causing human disease has expanded. These strains are derived from 39 different ribotypes and some *C. difficile* strains have been found to be toxin A-negative but toxin B-positive,[5] and 027 strain was the second most common isolate responsible for CDI.[6] Ribotype 078 was reported to have an increased prevalence,[7] and ribotype 244 seems to cause more severe disease with higher mortality rates than rates associated with ribotype 027.[8,9] The prevalence of other ribotypes now appears to surpass that of 027, including ribotypes 037, 018, and 078.[10–12]

Epidemiologic research of CDI resulting from infections with diverse *C. difficile* strains, including strain NAP1/B1/027 in developing countries, is expanding and includes data regarding hospital epidemiology, clonal spread, and dissemination across the respective countries.[13–16]

The present study reports on a 12-month evaluation of a CDI outbreak caused by different *C. difficile* strains including the NAP1/BI/027 strain.

Methods

Setting, study design, and study population

The outbreak described in this report occurred at the Hospital Civil de Guadalajara Fray Antonio Alcalde, an 899-bed tertiary care teaching hospital located in the city of Guadalajara, the second largest city in Mexico.

This was a case–control study of adult patients with hospital-onset CDI presenting between December 2013 and December 2014. During the study period 288 adult patients were evaluated and all patients had diarrhea defined as the passage of ≥3 unformed stools (Bristol scale type 5–7) within 24 or 48 h after admission.[17] Case patients were defined as those with a first episode of nosocomial CDI.

Clostridium difficile toxin identification

Starting in April 2014, all stool samples were tested for *C. difficile* toxins using real-time polymerase chain reaction (PCR) (Cepheid Xpert *C. difficile*/Epi, Cepheid, Sunnyvale CA) to identify toxin-producing *C. difficile* strains, including strain NAP1/B1/027. Prior to the availability of PCR-based diagnostic approaches all diarrhea specimens were tested by enzyme immunoassay (Meridian Bioscience, Cincinnati, OH, USA). All positive specimens were saved for future testing. All stool specimens were stored at 4° C for five days, and then frozen at −70° C. After PCR retesting, only positive samples were included in the final analysis.

Control patients

Patients without diarrhea or a positive CDI test were selected at the same time and ward that CDI patients were identified. Control patients were randomly selected across the study period. Control patients were matched to case patients at a 3:1 ratio.

Definitions

Previous hospitalization was defined as a hospital stay six weeks prior to the onset of diarrhea. Recent antibiotic therapy and steroid use were defined as exposure to these medicines six weeks prior to diarrhea onset.

Clinical severity score assessment and outcome

Patients were clinically evaluated for disease severity using the SHEA/IDSA definitions of mild, moderate, or severe disease. Serum creatinine levels were included in the definition of severe disease.[18] In addition, age >60 years, fever >38.3 °C, and a WBC count >15,000 were used to further define clinically severe disease.[19] Patients with >2 findings were considered to have severe disease.

A poor outcome was defined as death within 14 days after CDI diagnosis. Favorable outcome was defined by survival 14 days after CDI diagnosis. Relapse was defined as a second episode of diarrhea after adequate response to therapy.

Therapy and follow-up

Therapy for CDI was administered for 10 days after an adequate response to treatment was achieved (defined as a 50% reduction of loose stools after 24 h of therapy, continuous reduction after 48 h of treatment, and no diarrhea after 72 h of treatment). All patients discharged where followed via telephone every 30 days.

Statistical analysis

The data generated were coded, entered, validated, and analyzed using the Statistical Package for Social Science (SPSS), version 22.0. Univariate analyses were used to describe significant variables among cases and controls and among individuals infected with strain 027 and individuals infected with non-027 strains. *P*-values were calculated using the Chi-squared test or the Fisher's exact test for categorical variables and the Student's *t*-test or Wilcoxon rank-sum test for continuous variables. A p-value ≤ 0.05 was considered statistically significant. Multivariate analysis: logistic regression analysis was carried out considering CDI as dependent variable and clinical and demographic data as independent variables.