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Clostridium difficile in a HIV-Infected Cohort: Incidence, Risk Factors, and Clinical Outcomes

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Abstract

Objective—*C. difficile* is the most commonly reported infectious diarrhea in HIV-infected patients in the United States. We set out to determine the incidence, risk factors, and clinical presentation of *C. difficile* infections (CDI) in a cohort of HIV-infected subjects.

Design—We performed a nested, case-control analysis with four non-CDI controls randomly selected for each case.

Methods—We assessed the incidence of CDI in the Johns Hopkins HIV Clinical Cohort between July 1, 2003 and December 31, 2010. Incident cases were defined as first positive *C. difficile* cytotoxin assay or PCR for toxin B gene. We used conditional logistic regression models to assess risk factors for CDI. We abstracted data on the clinical presentation and outcomes from case chart review.

Results—We identified 154 incident CDI cases for an incidence of 8.3 cases/1000 patient years. No unique clinical features of HIV-associated CDI were identified. In multivariate analysis, risk of CDI was independently increased for: CD4 count < 50 cells/mm³ (Adjusted Odds Ratio (AOR) 20.7, 95% CI 2.8–151.4), hospital onset CDI (AOR 26.7 [3.1–231.2]), and use of clindamycin (AOR 27.6 [2.2–339.4]), fluoroquinolones (AOR 4.5 [1.2–17.5]), macrolides (AOR 6.3 [1.8–22.1]), gastric acid suppressants (AOR 3.1 [1.4–6.9]), or immunosuppressive agents (AOR 6.8 [1.2–39.6]).

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Author Contribution: Dr. Haines had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

Study concept and design: Haines, Moore, Bartlett, Sears, Cosgrove, Gebo

Acquisition of data: Moore, Gebo

Analysis and interpretation of data: Haines, Gebo

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Conflicts of Interest

C.F.H., R.D.M., and J.G.B. have no conflicts.

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Conclusions—The incidence of CDI in HIV-infected patients was twice that previously reported. Our data show compromised cellular immunity, as defined by CD4 < 50 cells/mm³ is a risk factor for CDI. Clinicians should be aware of the increased CDI risk, particularly in those with severe CD4 count suppression.

Keywords

case-control; *Clostridium difficile*; HIV; incidence; risk factors

Background

Clostridium difficile is an emerging pathogen that causes antibiotic associated diarrhea, pseudomembranous colitis, toxic megacolon, and death. The incidence of *C. difficile* infection (CDI) and associated morbidity and mortality in the general population have increased over the past decade [1]. Data suggest that immunocompromised patients may be at higher risk of CDI, perhaps because of impaired host immune responses to toxins produced by *C. difficile* strains [2–5]. HIV-infected patients have immunologic defects that may impair the antibody response and thus predispose them to increased incidence of CDI [6, 7].

CDI severity has been increasing in the general population with a near doubling of the U.S. hospitalization and case fatality rates [8, 9]. Over this same time period no published studies have examined CDI in HIV-infected patients. We hypothesize that the current incidence of CDI in HIV-infected patients is higher than previously reported, and that HIV-related immune suppression is a risk factor for CDI independent of antibiotic exposure and healthcare facility exposure. We evaluated the incidence of CDI, identified risk factors for incident CDI, and described the clinical presentation of CDI in this cohort.

Materials and Methods

Study Design

We performed a retrospective cohort analysis of HIV-infected patients receiving longitudinal HIV primary care in the Johns Hopkins Hospital outpatient HIV clinic between July 1, 2003 and December 31, 2010. All patients who initiate care in the clinic for HIV primary care are offered enrollment in the Johns Hopkins HIV Clinical Cohort (98% acceptance rate) [10].

Patients with CDI were identified from hospital electronic records. The medical records of each of these patients were then reviewed manually to confirm the laboratory result, and to collect data on clinical presentation and disease course. Annual CDI incidence was computed using the number of initial cases per calendar year and person-years of follow-up in the cohort. A nested case-control study was performed with four HIV-infected controls with no known CDI matched to each case of CDI. Controls were individually matched on cohort enrollment date within 6 months, duration of follow-up within 6 months, and CD4 count at cohort enrollment within 100 cells/mm³.

Clostridium difficile Tests

To identify both inpatient and outpatient CDI cases, test results were extracted from a hospital electronic database. From July 1, 2003 through May 9, 2004 the stool *C. difficile* cell culture cytotoxicity neutralization assay was used for testing. From May 10, 2004 through June 14, 2009 stool samples were screened with the *C. difficile* glutamate dehydrogenase (GDH) antigen enzyme immunoassay (EIA) and positive samples were tested with the *C. difficile* cytotoxicity assay as previously described [11]. Starting on June

15, 2009 stools screening positive with the *C. difficile* GDH antigen EIA were tested with the BD GeneOhm (BD Diagnostics, Sparks, MD) PCR for *C. difficile* toxin B gene real time PCR assay. From January 1, 2010 through the end of the study, stools were tested exclusively with the *C. difficile* toxin B PCR. The laboratory only accepted unformed stool for testing with the *C. difficile* toxin B PCR, but this requirement was not completely enforced for other CDI tests. No *C. difficile* strain identification data is collected by the laboratory.

Definitions

An incident CDI case was defined as the first positive *C. difficile* test result during the study period in subjects with no known prior CDI. CDI testing results prior to the study period were obtained starting January 1, 1999 in order to exclude subjects with evidence of CDI prior to the study period from the incident case analyses.

Multiple aspects of clinical presentation were examined in the review of case records. Diarrhea was defined as documented subjective patient or physician report of diarrhea. Stools per day were recorded, where available. Bloody stools were defined as documented subjective patient or physician report of bloody stools. The diagnosis of colitis was based on colitis or signs of inflammation consistent with colitis on computerized tomography (CT) scan reports. Pseudomembranous colitis was defined as pseudomembranes reported on colonoscopy or pathology. White blood cell count (WBC) data were extracted from laboratory records as the lab value nearest to and at a maximum of 3 days from the date of the positive stool test result.

Healthcare exposure was defined according to the clinical practice guidelines for *Clostridium difficile* infection in adults [12]. Event date was defined as the date of the positive test result for cases. The same date was used as the event date for matched controls with approximated equivalent follow-up time as the corresponding case. Hospital onset-healthcare facility associated (HO-HCFA) exposure was defined as an event date in cases and matched control occurring from hospital day 3 through discharge. Community onset-healthcare facility associated (CO-HCFA) was defined as any inpatient, outpatient, or skilled nursing facility exposure in the 12 weeks preceding the event date in cases and controls, including the first 3 days of hospitalization. Outpatient exposure was defined as any provider visits, physical therapy, or documented nursing visit where treatment was provided. Healthcare exposure at outside facilities was included when documented in the available records. Community associated CDI (CA) as defined as absence of healthcare facility exposure in the 12 weeks preceding the event date. Recurrent disease was defined as a positive CDI test or documented concern for recurrent CDI by the treating clinician prompting retreatment within 28 days after completion of the initial CDI treatment. Follow-up was defined as documented inpatient or outpatient visit or communication with clinical staff within 28 days after completion of CDI treatment.

We reviewed the outpatient clinic records and inpatient pharmacy order database to determine medications received in the 30 days prior to the date of CDI in cases and corresponding controls. Antibiotics were categorized into 6 groups based on mechanism and antimicrobial spectrum: clindamycin, fluoroquinolones, penicillin derivatives with gram-negative activity, cephalosporins, trimethoprim-sulfamethoxazole (TMP-SMX), and macrolides. Fluoroquinolones (FQ) included ciprofloxacin, moxifloxacin, gatifloxacin, norfloxacin, and levofloxacin. Penicillin derivatives with gram-negative activity category (GN-PCN) included amino-penicillins and anti-pseudomonal penicillins. The cephalosporins category included any cephalosporin use. Macrolide use included azithromycin and erythromycin. Gastric acid suppressants included all histamine H₂ blockers and proton pump inhibitors. Immunosuppressive agents included steroids, methotrexate, mycophenolate,

calcineurin inhibitors, mammalian target of rapamycin (MTOR) inhibitors, monoclonal antibodies, and chemotherapeutic agents.

Statistical Analysis

HIV transmission risk factors were injection drug use (IDU), men who have sex with men (MSM), and heterosexual transmission. Subjects may have multiple HIV risk factors. Laboratory variables ascertained within 90 days before or 30 days after the CDI event included HIV-1 RNA, CD4 count, and serum creatinine. Test results preceding the event were used when available. CD4 cell counts were divided into quartiles of 50, 51–200, 201–350, and >350 cells/mm³. Stages of chronic kidney disease (CKD) were defined according to the National Kidney Foundation/Kidney Disease Outcomes Quality Initiative (NKF/KDOQI) guidelines [13].

Statistical analyses were performed using STATA 12.1 [14]. Two-sided testing was used, with a P value < .05 considered significant. We used the Poisson distribution to calculate the standard error and 95% confidence interval (CI) for the incidence rate. Separate simple conditional logistic regression (SLR) analyses were performed to identify individual variables associated with the development of CDI. Conditional logistic regression models were used to avoid bias resulting from matching. Biologically plausible interactions between pairs of significant variables in the SLR analyses were tested. Variables included in the multiple conditional logistic regression (MLR) model were selected based on clinical relevance and biologic plausibility. These variables were CD4 category, log₁₀ copies/ml HIV-1 RNA, HOHCFA, CO-HCFA, clindamycin use, FQ use, GNPCN use, cephalosporin use, TMP-SMX use, macrolide use, use of gastric acid suppression, age categories, gender, CKD stage. We exponentiated the model coefficients to obtain the adjusted odds ratio (AOR) and corresponding 95% CI. Linear combination of estimates was used to make comparisons between non-referent categories. Conditional logistic regression models estimate AOR, which approximates the relative risk given low CDI incidence and use of incidence density sampling [15]. A Log binomial model was used to estimate the relative risk of CDI recurrence by CD4 category. Three sensitivity analyses of the MLR model were performed: (i) including only subjects with diarrhea documented in the medical record, (ii) including statistically significant and mechanistically plausible interaction terms, and (iii) including imputed missing data for CD4 category and log₁₀ HIV viral load. The multiple imputation utilized chained equations: ordinal logistic regression for CD4 category and linear regression for log₁₀ HIV viral load with MLR model covariates as predictors.

Results

Between July 1, 2003 and December 31, 2010 4,217 subjects were followed for a median of 4.1 years and a total follow-up time of 18,525 person-years. During this interval, we identified 154 incident cases of CDI for an overall incidence of 8.3 cases per 1000 person-years. There was no significant change in incidence noted over the study period. (Figure 1 Figure 1)

Clinical presentation in initial cases of CDI was characterized by diarrhea (81.6%), with a mean of 4.7 stools per day and normal WBC count (Table 1). There were no deaths directly attributable to CDI, but two subjects with pseudomembranous colitis required colectomy, and two additional subjects were diagnosed with toxic megacolon but did not require surgical intervention (Table 1). Colonoscopy was performed on six subjects, two of which had pseudomembranes.

The 152 incident CDI cases were matched with 602 controls. We excluded two cases due to lack of matched controls and two cases had one control. Baseline characteristics are

compared in Table 2. Forty-three cases (28%) had no clindamycin, fluoroquinolone, GN-PCN, cephalosporin exposure, or HO-HCFA exposure. SLR and MLR results are shown in Table 3. There were interactions between HO-HCFA and GN-PCN use ($p=0.05$), CD4 category and TMP-SMX ($p=0.01$), and CD4 category and macrolide use ($p=0.003$). The effect of CD4 category on CDI risk was not modified by either healthcare exposure category or immunosuppressant use. The risk of CDI increased with decreasing CD4 count in the univariate analysis. In the multivariate model there was an increase in CDI risk as CD4 category decreased from 51–200 cells/mm³ to <50 cells/mm³ (Adjusted Odds Ratio (AOR) 5.2, 95% CI 1.3–21.5). There was an increase in CDI risk in HO-HCFA compared to CO-HCFA (AOR 19.9 [3.7–107.7]). Clindamycin, FQ, and macrolide use each independently increased risk for CDI. Any use of clindamycin, FQ, GN PCN, TMP-SMX, or macrolides resulted in a 6.4 fold (95% CI 2.9–14.4) increase in CDI risk (model not shown). In the adjusted model stage 2, 4, and 5 CKD had increased CDI risk compared with stage 1 CKD. Sensitivity analysis using the above model with the 109 cases with recorded diarrhea and 430 corresponding controls was performed. The AOR for CD4 count <50 cells/mm³ was 131.8 (95% CI 5.5–3137.5), for CD4 count 51–200 cells/mm³ AOR 13.1 [0.97–176.7], and for CD4 count 201–350 cells/mm³ AOR 2.8 [0.48–16.2]. The AOR for HO-HCFA was 33.2 [1.6–697.6] and for immunosuppressant use AOR 13.3 [1.2–140.8]. CO-HCFA, TMP-SMX, macrolide, GN-PCN, cephalosporin, clindamycin, quinolone, and acid suppressant use were not significant. A sensitivity analysis including the CD4 count and TMP-SMX and CD4 count and macrolide interactions Abstract

This is a Dummy Abstract in the multivariate model found no significant interactions. Immunosuppressant use was no longer significant in the sensitivity analysis ($p=0.09$). There were no other changes in inference compared to the final model. A sensitivity analysis of imputed missing values of CD4 category and log₁₀ HIV viral load resulted in no change in inference of the effect of CD4 category on the risk of CDI.

Recurrent CDI occurred in 13% of all incident CDI cases and in 17% of cases with documented 28 day follow-up (118 cases with 28 days follow-up). The unadjusted risk for CDI recurrence was 24% higher (95% CI 8%–42%, $p=0.003$) in subjects with a CD4 count <350 cells/mm³ than those with a CD4 count ≥ 350 cells/mm³.

Discussion

This study has several important findings. First, the incidence of CDI was twice that reported in another cohort of HIV-infected patients from 1992–2002 [16]. Impairment of cellular immunity, as measured by CD4 count <50 cells/mm³ was a risk factor for CDI independent of antibiotic use, gastric acid suppression, immunosuppressant use, CKD, and healthcare exposure.

The incidence of CDI in this study cohort could have increased from prior reports for several reasons. First, CDI incidence in the general population is increasing and may contribute to the increased incidence in our study [17]. Additionally, over time CDI testing modalities have changed, and each modality has different sensitivity and specificity [18, 19]. Finally, the most recent cohort study of CDI incidence in an HIV-infected population used the Adult/Adolescent Spectrum of HIV Disease Project (ASD) cohort, which ended in 2002, prior to the emergence of the B1/NAP1/027 epidemic strain. Subsequent reports found evidence of outbreaks of the B1/NAP1/027 strain in several United States cities [20] and this strain was also present at our institution during the study period [21]. Together, the increased CDI incidence nationwide, changes in testing modalities, and the emergence of highly pathogenic *C. difficile* strains likely contributed to the increased CDI incidence in this study.

Clinical presentations of CDI in our cohort were similar to previous reports in HIV seronegative populations, with the exception of lower median leukocyte counts in this study population (approximately 6×10^3 cells/mm³ compared to 12×10^3 cells/mm³) [22], and no mortality was directly attributed to CDI in this study. Previous studies of CDI-associated mortality in HIV-infected populations vary in methodology and have conflicting results [2, 3, 23].

In the multivariable regression, CD4 count < 50 cells/mm³ was an independent risk factor for CDI. In the pre-ART era, this association was demonstrated in one HIV-infected hospitalized population [6]; however, no other study has demonstrated this association in the modern ART era [2, 23, 24]. Our results suggest that severely compromised cellular immunity may confer increased CDI risk in HIV-infected populations. This may occur through impairment of the antibody response to *C. difficile* toxins, which predisposes to CDI [25]. Memory B lymphocytes are reduced in HIV infection, which correlates with diminished vaccine-specific antibody responses [26–29]. Furthermore, two clinical trials found that the antibody responses to conjugate pneumococcal vaccine are diminished in HIV-infected subjects as CD4 counts decreased [30, 31]. These studies suggest that cellular immunity deficits due to HIV infection can affect antibody responses. Hence, it is plausible that severely compromised cellular immunity in advanced HIV may affect the antibody response to *C. difficile* toxins and increase risk of CDI.

Our MLR also demonstrated increased CDI risk with CKD (except stage 3), gastric acid suppressant use, and immunosuppressant use. These covariates were included in the analysis because prior studies had shown an association with CDI [32–34]. Our findings are consistent with those in HIV seronegative populations.

Several interactions were observed. The TMP/SMX and macrolide interactions with CD4 count are to be expected due CD4 count dependent antibiotic prescribing for opportunistic infection (OI) prophylaxis according to HIV treatment guidelines [35]. Not surprisingly, this study has very few cases with low CD4 counts not on OI prophylaxis as our clinic and patients conform to national HIV guidelines. Thus, statistical inference of CDI risk in these subgroups is limited, and the sensitivity analysis supports our exclusion of these interactions from the final multivariable regression model. The interaction of HO-HCFA and GN-PCN was of borderline significance and as such was not included in the model or sensitivity analysis.

Assessing CDI risk in HIV populations by CD4 count is challenging due to confounding by high levels of health care exposure and antibiotic use. We found that 92.8% of subjects with CDI in our study had healthcare exposure in the 12 weeks preceding CDI diagnosis. A recent CDC study found that 94% of CDI cases had healthcare exposure in the 12 weeks prior to CDI testing [36]. As such, routine outpatient clinic visits likely represent a significant CDI risk factor for all patients with HIV in clinical care. However, CO-HCFA exposure was high in both cases and controls and did not affect the risk of CDI in the multivariate model. In contrast, being an inpatient for > 3 days at the time of CDI testing (HO-HCFA) was a strong CDI risk factor in our model. Although we were not fully able to assess healthcare exposure at all other healthcare facilities, this exposure was included where it was clearly indicated in the records. We did not quantify the type and number of healthcare facility exposures. Thus, it is possible residual confounding is present, but there is currently no standardized method for quantification of healthcare exposure in CDI. Antibiotic administration records from outside healthcare facilities were not available and represent another potential source of residual confounding. We thoroughly assessed healthcare exposure and antibiotic use within the limitations of a retrospective study design and demonstrated that a CD4 count < 50 cells/mm³ is a highly significant independent risk

for CDI. Prospective studies are needed to more fully explore the association of healthcare visit type and CDI risk and overcome some of the limitations inherent in retrospective analyses.

We did not observe a decrease in incident CDI despite improvements in HIV clinical care over the study period. The CDI incidence rate was unchanged from 7.5 cases per 1000 person-years in 2003 to 6.8 cases per 1000 person-years in 2010, while ART uptake increased from 70% in 2003 to 84% in 2009 (personal communication, R.D.M., August 2012). In contrast, Sanchez *et al.* demonstrated a non-significant decrease in CDI incidence in the ASD cohort from 4.7 cases per 1000 person-years in 1992 to 2.9 cases per 1000 person-years in 2002 (OR, 0.7 [0.4–1.1]), while ART uptake in the ASD cohort increased from 58.8% in 1995 to 83.5% in 2002 [16, 37]. Additionally, a separate study found CDI incidence dropped from 4.7% in the pre-HAART era to 2.4% in the HAART era (RR=1.94 [1.03–3.68]) [38]. Our seemingly contradictory findings may result from differences in the underlying cohorts, and in particular, our CDI cases had a lower proportion of ART use than controls, consistent with the increased risk identified herein with low CD4 count. The multivariable analysis in this study suggests that subjects with CD4 count ≥ 50 cells/mm³ are at the highest risk for CDI. The percentage of subjects with CD4 count <50 cells/mm³ dropped from approximately 35% in 1994–95 to 15% by 2002–03 in the Michigan ASD sites [39]. Whereas, in our cohort the percent of subjects with CD4 ≥ 50 cells/mm³ dropped from 15% in 2003 to 6% in 2010 (personal communication, R.D.M., August 2012). It is possible that the observed decrease in the percent of subjects with a CD4 count ≥ 50 cells/mm³ in our cohort may not have been sufficiently large to result in a detectable decrease in CDI incidence over the study period. This factor as well as the previously mentioned causes of rising CDI incidence may have obscured any benefit of improved HIV clinical care and treatment on CDI incidence in our cohort over the study period.

Recurrent CDI has not been evaluated previously in HIV-infected patients. We report a lower percentage of recurrent disease than in a prospective study of the general population using a similar recurrence definition [40]. As a retrospective study, our ability to capture recurrences was diminished relative to a clinical trial. However, we only included incident CDI, which may have attenuated the recurrence rate relative to studies including patients with prior CDI. HIV-infected subjects may have lower CDI recurrence rate than the general population. The interpretation of our finding of increased recurrent CDI risk with CD4 <350 cells/mm³ is limited due to small sample size and an unadjusted analysis with risk for confounding. Further study is needed in recurrent CDI in HIV-infected patients.

This study has several limitations. There are wide confidence intervals for some AOR estimates in the MLR, illustrating that our model does not fully explain the observed variation in the data. The source population is comprised of urban HIV-infected subjects who receive care at a hospital-affiliated HIV clinic. Our study only assesses the risk of CDI within our cohort and does not make comparisons to the general population. Furthermore, this study is unable to assess the CDI risk of impaired cellular immunity in other populations. However, the results may be generalizable to similar urban hospital-affiliated HIV clinics. Finally, testing may misclassify asymptomatic colonization with *C. difficile* as CDI. Currently there is no test able to differentiate colonization from CDI. As such we were forced to assume that testing was done only in the appropriate clinical scenario, but this was not always clearly documented on retrospective review of the charts. Of note, once our hospital switched to testing with the Toxin B PCR in June 2009, our laboratory only accepted unformed stool, limiting testing to those who were symptomatic. If we were misclassifying a large portion of asymptomatic colonization as incident CDI we would expect our CDI incidence to decline after introduction of the Toxin B PCR and limiting testing to unformed stool. Such a decline was not observed, but changes in testing modalities

confound direct comparison. Prior retrospective cohort studies found diarrhea in 84–87% of cases, similar to the proportion of subjects with diarrhea in this study [41, 42]. A sensitivity analysis using only subjects with diarrhea remained significant for CD4 count < 50 cells/mm³ and HO-HCFA, suggesting that the inference regarding these covariates is not affected by the inclusion of subjects without recorded diarrhea.

In summary, we present the largest study of CDI in a HIV-infected population in the modern antiretroviral therapy era. The incidence of CDI was more than twice that previously reported in a similar, earlier HIV-infected cohort [16]. This is the first study to show a CD4 count < 50 cells/mm³ results in increased risk of CDI independent of traditional risk factors such as healthcare and antibiotic exposure. Clinicians should be cognizant of the risk of CDI in this population and expose these patients to risks such as hospitalization, antibiotics, immunosuppression, or gastric acid suppression only when necessary.

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References

- Freeman J, Bauer MP, Baines SD, Corver J, Fawley WN, Goorhuis B, et al. The changing epidemiology of *Clostridium difficile* infections. *Clin Microbiol Rev*. 2010; 23:529–549. [PubMed: 20610822]
- Tumbarello M, Tacconelli E, Leone F, Cauda R, Ortona L. *Clostridium difficile*-associated diarrhoea in patients with human immunodeficiency virus infection: a case-control study. *Eur J Gastroenterol Hepatol*. 1995; 7:259–263. [PubMed: 7743309]
- Willingham FF, Ticona Chavez E, Taylor DN, Bowen AB, Crane AR, Gottlieb AL, et al. Diarrhea and *Clostridium difficile* infection in Latin American patients with AIDS. Working Group on AIDS in Peru. *Clin Infect Dis*. 1998; 27:487–493. [PubMed: 9770145]
- Kelly CP, Kyne L. The host immune response to *Clostridium difficile*. *J Med Microbiol*. 2011; 60:1070–1079. [PubMed: 21415200]
- Alonso CD, Treadway SB, Hanna DB, Huff CA, Neofytos D, Carroll KC, et al. Epidemiology and outcomes of *Clostridium difficile* infections in hematopoietic stem cell transplant recipients. *Clin Infect Dis*. 2012; 54:1053–1063. [PubMed: 22412059]
- Barbut F, Meynard JL, Guiguet M, Avesani V, Bochet MV, Meyohas MC, et al. *Clostridium difficile*-associated diarrhea in HIV-infected patients: epidemiology and risk factors. *J Acquir Immune Defic Syndr Hum Retrovirol*. 1997; 16:176–181. [PubMed: 9390569]
- Sivapalasingam S, Blaser MJ. Bacterial diarrhea in HIV-infected patients: why *Clostridium difficile*, and why now? *Clin Infect Dis*. 2005; 41:1628–1630. [PubMed: 16267736]
- McDonald LC, Owings M, Jernigan DB. *Clostridium difficile* infection in patients discharged from US short-stay hospitals, 1996–2003. *Emerg Infect Dis*. 2006; 12:409–415. [PubMed: 16704777]

9. Zilberberg MD, Shorr AF, Kollef MH. Increase in adult *Clostridium difficile*-related hospitalizations and case-fatality rate, United States, 2000–2005. *Emerg Infect Dis.* 2008; 14:929–931. [PubMed: 18507904]
10. Moore RD. Understanding the clinical and economic outcomes of HIV therapy: the Johns Hopkins HIV clinical practice cohort. *J Acquir Immune Defic Syndr Hum Retrovirol.* 1998; 17(Suppl 1):S38–S41. [PubMed: 9586651]
11. Reller ME, Lema CA, Perl TM, Cai M, Ross TL, Speck KA, et al. Yield of stool culture with isolate toxin testing versus a twostep algorithm including stool toxin testing for detection of toxigenic *Clostridium difficile*. *J Clin Microbiol.* 2007; 45:3601–3605. [PubMed: 17804652]
12. Cohen SH, Gerding DN, Johnson S, Kelly CP, Loo VG, McDonald LC, et al. Clinical practice guidelines for *Clostridium difficile* infection in adults, 2010 update by the society for healthcare epidemiology of America (SHEA) and the infectious diseases society of America (IDSA). *Infect Control Hosp Epidemiol.* 2010; 31:431–455. [PubMed: 20307191]
13. KDOQI Clinical Practice Guidelines for Chronic Kidney Disease: Evaluation, Classification, and Stratification. National Kidney Foundation, Inc; 2002.
14. StataCorp. Stata Statistical Software: Release 12. College Station, TX: StataCorp LP; 2011.
15. Greenland S, Thomas DC. On the need for the rare disease assumption in case-control studies. *Am J Epidemiol.* 1982; 116:547–553. [PubMed: 7124721]
16. Sanchez TH, Brooks JT, Sullivan PS, Juhász M, Mintz E, Dworkin MS, et al. Bacterial diarrhea in persons with HIV infection, United States, 1992–2002. *Clin Infect Dis.* 2005; 41:1621–1627. [PubMed: 16267735]
17. Rupnik M, Wilcox MH, Gerding DN. *Clostridium difficile* infection: new developments in epidemiology and pathogenesis. *Nat Rev Microbiol.* 2009; 7:526–536. [PubMed: 19528959]
18. Shanholtzer CJ, Willard KE, Holter JJ, Olson MM, Gerding DN, Peterson LR. Comparison of the VIDAS *Clostridium difficile* toxin A immunoassay with *C. difficile* culture and cytotoxin and latex tests. *J Clin Microbiol.* 1992; 30:1837–1840. [PubMed: 1629341]
19. Tenover FC, Novak-Weekley S, Woods CW, Peterson LR, Davis T, Schreckenberger P, et al. Impact of strain type on detection of toxigenic *Clostridium difficile*: comparison of molecular diagnostic and enzyme immunoassay approaches. *J Clin Microbiol.* 2010; 48:3719–3724. [PubMed: 20702676]
20. McDonald LC, Killgore GE, Thompson A, Owens RC Jr, Kazakova SV, Sambol SP, et al. An epidemic, toxin gene-variant strain of *Clostridium difficile*. *N Engl J Med.* 2005; 353:2433–2441. [PubMed: 16322603]
21. Song X, Bartlett JG, Speck K, Naegeli A, Carroll K, Perl TM. Rising economic impact of *clostridium difficile*-associated disease in adult hospitalized patient population. *Infect Control Hosp Epidemiol.* 2008; 29:823–828. [PubMed: 18643746]
22. Manabe YC, Vinetz JM, Moore RD, Merz C, Charache P, Bartlett JG. *Clostridium difficile* colitis: an efficient clinical approach to diagnosis. *Ann Intern Med.* 1995; 123:835–840. [PubMed: 7486465]
23. Pulvirenti JJ, Mehra T, Hafiz I, DeMarais P, Marsh D, Kocka F, et al. Epidemiology and outcome of *Clostridium difficile* infection and diarrhea in HIV infected inpatients. *Diagn Microbiol Infect Dis.* 2002; 44:325–330. [PubMed: 12543536]
24. Hutin Y, Molina JM, Casin I, Daix V, Sednaoui P, Welker Y, et al. Risk factors for *Clostridium difficile*-associated diarrhoea in HIV-infected patients. *AIDS.* 1993; 7:1441–1447. [PubMed: 8280409]
25. Kyne L, Warny M, Qamar A, Kelly CP. Asymptomatic carriage of *Clostridium difficile* and serum levels of IgG antibody against toxin A. *N Engl J Med.* 2000; 342:390–397. [PubMed: 10666429]
26. De Milito A, Nilsson A, Titanji K, Thorstensson R, Reizenstein E, Narita M, et al. Mechanisms of hypergammaglobulinemia and impaired antigen-specific humoral immunity in HIV-1 infection. *Blood.* 2004; 103:2180–2186. [PubMed: 14604962]
27. Kroon FP, van Dissel JT, de Jong JC, van Furth R. Antibody response to influenza, tetanus and pneumococcal vaccines in HIV-seropositive individuals in relation to the number of CD4R lymphocytes. *AIDS.* 1994; 8:469–476. [PubMed: 7912086]

28. Kroon FP, van Dissel JT, Labadie J, van Loon AM, van Furth R. Antibody response to diphtheria, tetanus, and poliomyelitis vaccines in relation to the number of CD4R T lymphocytes in adults infected with human immunodeficiency virus. *Clin Infect Dis*. 1995; 21:1197–1203. [PubMed: 8589143]
29. Kroon FP, van Dissel JT, Rijkers GT, Labadie J, van Furth R. Antibody response to Haemophilus influenzae type b vaccine in relation to the number of CD4R T lymphocytes in adults infected with human immunodeficiency virus. *Clin Infect Dis*. 1997; 25:600–606. [PubMed: 9314445]
30. Kroon FP, van Dissel JT, Ravensbergen E, Nibbering PH, van Furth R. Enhanced antibody response to pneumococcal polysaccharide vaccine after prior immunization with conjugate pneumococcal vaccine in HIV-infected adults. *Vaccine*. 2000; 19:886–894. [PubMed: 11115712]
31. Miirio G, Kayhty H, Watera C, Tolmie H, Whitworth JA, Gilks CF, et al. Conjugate pneumococcal vaccine in HIV-infected Ugandans and the effect of past receipt of polysaccharide vaccine. *J Infect Dis*. 2005; 192:1801–1805. [PubMed: 16235180]
32. Collini PJ, Bauer M, Kuijper E, Dockrell DH. Clostridium difficile infection in HIV-seropositive individuals and transplant recipients. *J Infect*. 2012; 64:131–147. [PubMed: 22178989]
33. Keddis MT, Khanna S, Noheria A, Baddour LM, Pardi DS, Qian Q. Clostridium difficile infection in patients with chronic kidney disease. *Mayo Clin Proc*. 2012; 87:1046–1053. [PubMed: 23127731]
34. Janarthanan S, Ditah I, Adler DG, Ehrinpreis MN. Clostridium difficile-associated diarrhea and proton pump inhibitor therapy: a meta-analysis. *American Journal of Gastroenterology*. 2012; 107:1001–1010. [PubMed: 22710578]
35. Dybul M, Fauci AS, Bartlett JG, Kaplan JE, Pau AK. Guidelines for using antiretroviral agents among HIV-infected adults and adolescents - The panel on clinical practices for treatment of HIV. *Annals of Internal Medicine*. 2002; 137:381–433. [PubMed: 12617573]
36. Centers for Disease C, Prevention. Vital signs: preventing Clostridium difficile infections. *MMWR Morb Mortal Wkly Rep*. 2012; 61:157–162. [PubMed: 22398844]
37. Hooshyar D, Hanson DL, Wolfe M, Selik RM, Buskin SE, McNaghten AD. Trends in perimortal conditions and mortality rates among HIV-infected patients. *AIDS*. 2007; 21:2093–2100. [PubMed: 17885300]
38. Tacconelli E, Tumbarello M, de Gaetano Donati K, Leone F, Mazzella P, Cauda R. Clostridium difficile-associated diarrhea in human immunodeficiency virus infection—a changing scenario. *Clin Infect Dis*. 1999; 28:936–937. [PubMed: 10825082]
39. Health MDoC. Adult and Adolescent Spectrum of Disease Project in Michigan Summary Report 1990–2003.
40. Louie TJ, Miller MA, Mullane KM, Weiss K, Lentnek A, Golan Y, et al. Fidaxomicin versus vancomycin for Clostridium difficile infection. *N Engl J Med*. 2011; 364:422–431. [PubMed: 21288078]
41. Kaltsas A, Simon M, Unruh LH, Son C, Wroblewski D, Musser KA, et al. Clinical and laboratory characteristics of Clostridium difficile infection in patients with discordant diagnostic test results. *J Clin Microbiol*. 2012; 50:1303–1307. [PubMed: 22238444]
42. Saddi, Vr; Glatt, Ae. Clostridium difficile-associated diarrhea in patients with HIV: a 4-year survey. *J Acquir Immune Defic Syndr*. 2002; 31:542–543. [PubMed: 12473844]

Table 1Clinical Presentation and Complications of First Cases of *C. difficile* Infection.

Presenting Symptoms	Number (%)
Diarrhea, (mean stools/day), (N = 136)	111 (81.6%), (4.7)
Bloody Stools (N = 61)	14 (23.0%)
Studies	
Median WBC (N = 140)	6210 cells/ mm ³ (IQR 3795–10145 cells/ mm ³)
Colitis on CT scan (N = 54)	20 (37.0%)
PMC* on colo/sigmoidoscopy (N = 6)	2 (33.3%)
Complications	
Hospitalized due to CDI (N = 101)	35 (23.7%)
Surgery Consult (N = 154)	10 (6.8%)
Toxic Megacolon (N = 154)	2 (1.4%)
Colectomy (N = 154)	2 (1.4%)
Acute Renal Failure (N = 154)	17 (11.5%)
Altered Mental Status (N = 154)	10 (6.8%)

* PMC, Pseudomembranous colitis.

Table 2Demographic and Clinical Characteristics of Incident *C. difficile* Infection Cases and Matched Controls.

Characteristic	Cases (N=152)	Controls (N=602) number (%)	Total (N=754)
Age			
Mean (years) (Range)	40.5 (20–68)	39.8 (17–76)	39.9 (17–76)
Age 40	83 (54.6)	300 (49.8)	383 (50.8)
Race *			
African-American	131 (86.2)	449 (74.6)	580 (76.9)
White	18 (11.8)	135 (22.4)	153 (20.3)
Hispanic/Other	3 (2.0)	18 (3.0)	21 (2.8)
Gender *			
Male	83 (54.6)	396 (65.8)	479 (63.5)
Female	69 (45.4)	206 (34.2)	275 (36.5)
HIV Risk Factor			
IDU **	74 (48.7)	192 (31.9)	266 (35.3)
MSM *	27 (17.7)	174 (28.9)	201 (26.7)
Heterosexual Transmission	56 (36.8)	203 (33.7)	259 (34.4)
CD4 cells/mm ³ at Event **	N = 145	N = 525	N = 670
<50	45 (31.0)	53 (10.1)	98 (14.6)
50–200	48 (33.1)	103 (19.6)	151 (22.5)
201–350	22 (15.2)	108 (20.6)	130 (19.4)
>350	30 (20.7)	261 (49.7)	291 (43.4)
Log ₁₀ HIV RNA at event **	N = 144	N = 528	N = 672
<4.0	74 (51.4)	416 (78.8)	490 (72.9)
4.0–5.0	31 (21.5)	80 (15.2)	111 (16.5)
>5.0	39 (27.1)	32 (6.1)	71 (10.6)
ART at Event **	98 (64.5)	484 (80.4)	582 (77.2)
Healthcare Exposure at Event **			
CA	11 (7.2)	139 (23.1)	150 (19.9)
CO-HCFA	90 (59.2)	457 (75.9)	547 (72.6)
HO-HCFA	51 (33.6)	6 (1.0)	57 (7.6)
Antibiotic Use			
Clindamycin **	25 (16.4)	2 (0.3)	27 (3.6)
FQ **	47 (30.9)	14 (2.3)	61 (8.1)
GN-PCN **	29 (19.1)	4 (0.7)	33 (4.4)
Cephalosporin **	50 (32.9)	12 (2.0)	62 (8.2)
TMP-SMX **	64 (42.1)	147 (24.4)	211 (28.0)
Macrolide **	79 (52.0)	99 (16.4)	178 (23.6)
Immunosuppressants **	33 (21.7)	8 (1.3)	41 (5.4)

Characteristic	Cases (N=152)	Controls (N=602) number (%)	Total (N=754)
GI Acid suppressors**	77 (50.7)	64 (10.6)	141 (18.7)
KDOQI CKD Stage**			
1	67 (44.1)	407 (67.6)	474 (62.9)
2	36 (23.7)	138 (22.9)	174 (23.1)
3	17 (11.2)	32 (5.3)	49 (6.5)
4	10 (6.6)	3 (0.5)	13 (1.7)
5	22 (14.5)	22 (3.6)	44 (5.8)

* $p < 0.05$ by χ^2 test.

** $p < 0.001$ by χ^2 test.

ART, anti-retroviral therapy; CA, Community associated; CKD, chronic kidney disease; CO-HCFA, community onset healthcare facility associated; HO-HCFA, hospital onset healthcare facility associated; IDU, injection drug use; KDOQI, National Kidney Foundation Disease Outcomes Quality Initiative; MSM, men who have sex with men.

Table 3Univariate and Multivariate Regression Analysis for Factors Associated with Initial *C. difficile* Infection.*

Factor	Univariate Odds Ratio (95% CI)	P value	Multivariate (N = 633) Adjusted Odds Ratio (95% CI)	P value
CD4 count (cells/mm ³)				
50	24.46 (10.91 – 54.86)	<0.001	20.67 (2.82 – 151.45)	0.003
51–200	9.70 (4.80 – 19.58)	<0.001	3.97 (0.84 – 18.81)	0.082
201–350	2.43 (1.24 – 4.74)	0.010	1.93 (0.62 – 6.06)	0.259
>350	Ref		Ref	
Log ₁₀ HIV RNA	1.79 (1.54 – 2.09)	<0.001	1.09 (0.73 – 1.62)	0.677
Health Care Exposure at Event				
CA	Ref		Ref	
HO-HCFA	135.45 (36.37 – 504.41)	<0.001	26.67 (3.08 – 231.18)	0.003
CO-HCFA	2.42 (1.21 – 4.84)	0.012	1.34 (0.31 – 5.86)	0.698
Antibiotic Use				
Clindamycin	97.54 (13.21 – 720.26)	<0.001	27.59 (2.24 – 339.40)	0.010
FQ	19.44 (9.50 – 39.77)	<0.001	4.51 (1.16 – 17.54)	0.030
GN-PCN	29.00 (10.20 – 82.49)	<0.001	2.14 (0.32 – 14.22)	0.429
Cephalosporin	23.47 (11.10 – 49.59)	<0.001	3.15 (0.76 – 13.03)	0.112
TMP-SMX	2.76 (1.81 – 4.21)	<0.001	0.81 (0.31 – 2.10)	0.658
Macrolide	9.64 (5.79 – 16.03)	<0.001	6.25 (1.76 – 22.15)	0.005
Immunosuppressants	21.22 (8.88 – 50.72)	<0.001	6.76 (1.15 – 39.60)	0.034
GI Acid suppressors	7.25 (4.81 – 10.93)	<0.001	3.12 (1.42 – 6.86)	0.005
KDOQI CKD Stage				
1	Ref		Ref	
2	1.60 (1.01 – 2.54)	0.047	3.21 (1.23 – 8.40)	0.017
3	3.47 (1.77 – 6.80)	<0.001	0.80 (0.18 – 3.63)	0.773
4	17.78 (4.77 – 66.26)	<0.001	30.33 (2.08 – 442.86)	0.013
5	6.41 (3.22 – 12.76)	<0.001	4.53 (1.08 – 19.03)	0.039

* Adjusted for age category and gender.

CA, Community associated; CKD, chronic kidney disease; CO-HCFA, community onset healthcare facility associated; FQ, fluoroquinolone; GN-PCN, includes: aminopenicillins and anti-pseudomonal penicillins; HO-HCFA, hospital onset healthcare facility associated; KDOQI, National Kidney Foundation Disease Outcomes Quality Initiative; TMP-SMX, trimethoprim-sulfamethoxazole