MAJOR ARTICLE

Epidemiology and Outcomes of *Clostridium difficile* Infections in Hematopoietic Stem Cell Transplant Recipients

Carolyn D. Alonso,[1,2] Suzanne B. Treadway,[1] David B. Hanna,[3] Carol Ann Huff,[4] Dionissios Neofytos,[1] Karen C. Carroll,[5] and Kieren A. Marr[1,4]

[1]Department of Medicine, The Johns Hopkins Hospital, Baltimore, Maryland; [2]Department of Medicine, Beth Israel Deaconess Medical Center, Boston, Massachusetts; [3]Department of Epidemiology, The Johns Hopkins Bloomberg School of Public Health, [4]Department of Oncology, and [5]Department of Pathology, The Johns Hopkins Hospital, Baltimore, Maryland

Background. *Clostridium difficile* is the leading cause of infectious diarrhea among hospitalized patients and is a major concern for patients undergoing hematopoietic stem cell transplantation (HSCT). Risk factors and the natural history of *C. difficile* infection (CDI) are poorly understood in this population.

Methods. We performed a retrospective nested case-control study to describe the epidemiology, timing, and risk factors for CDI among adult patients who received HSCTs at our center from January 2003 through December 2008.

Results. The overall 1-year incidence of CDI was 9.2% among HSCTs performed (n = 999). The median time to diagnosis of CDI was short among both autologous and allogeneic HSCT recipients (6.5 days and 33 days, respectively). Risk factors for CDI in allogeneic HSCT recipients included receipt of chemotherapy prior to conditioning for HSCT, broad-spectrum antimicrobial use, and acute graft-versus-host disease (GVHD; adjusted odds ratio [AOR], 4.45; 95% confidence interval [CI], 1.54–12.84; $P = .006$). There was a strong relationship between early CDI and subsequent development of gastrointestinal tract GVHD in the year following allogeneic HSCT ($P < .001$). Gastrointestinal GVHD was also strongly associated with an increased risk for recurrent CDI (AOR, 4.23 [95% CI, 1.20–14.86]; $P = .02$).

Conclusions. These results highlight the high incidence and early timing of CDI after HSCT. Early timing, coupled with the noted risk of pretransplant chemotherapy, suggests that the natural history of disease in some patients may involve colonization prior to HSCT. A potentially important interplay between CDI and GVHD involving the gastrointestinal tract was observed.

Clostridium difficile is the leading cause of infectious diarrhea among hospitalized patients and is a concern for patients who are immunosuppressed due to hematopoietic stem cell transplantation (HSCT) [1–5]. Emergence of a hypertoxigenic strain of *C. difficile* known as North American Pulsed Field Type 1 (NAP1) has been associated with a globally increased incidence and severity of disease [6].

Patients who receive HSCT may be at particularly high risk for *C. difficile* infection (CDI) because of long hospitalizations, receipt of broad-spectrum antibiotics, and chemotherapy-related disruption of enteric mucosal barriers [7, 8]. Prior reports suggested that the proportion of diarrheal episodes attributable to CDI following HSCT is approximately 5% [9–11]. However, much of these data come from the pre-NAP1 era, include few patients, and did not evaluate detailed host risks. In this study, we evaluated the timing and risk factors for CDI within a large longitudinal cohort of HSCT recipients at The Johns Hopkins Hospital in Baltimore, MD, from 2003 through 2008 (n = 999 HSCT recipients). Results demonstrate that CDI is

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Correspondence: Kieren A. Marr, MD, Johns Hopkins University School of Medicine, Division of Infectious Diseases, 720 Rutland Ave, Ross-1064, Baltimore, MD 21205 (kmarr4@jhmi.edu).

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a common early complication after HSCT. Risk factor analyses provide insight into the natural history of disease and suggest a link between CDI and gastrointestinal (GI) tract graft-versus-host disease (GVHD).

MATERIALS AND METHODS

Patient Population

This study was approved by the Johns Hopkins University institutional review board. From 1 January 2003 through 31 December 2008, 999 adult allogeneic and autologous HSCTs were performed. Patients who received high-dose chemotherapy with cyclophosphamide without stem cell infusion were classified as having received an autologous HSCT [12].

Study Design

We performed a retrospective, case-control study nested within the cohort of HSCT recipients, stratified by graft type. An additional cohort analysis limited to patients with CDI was performed to assess risk factors associated with recurrent infection. Patients with CDI were identified by review of electronic microbiology and infection control databases and confirmed by medical record review. CDI was recorded from day −7 through day 365 of HSCT. To minimize confounding, controls were matched 2:1 to cases by HSCT type and date by means of cumulative incidence sampling. Among 274 patients available for analysis, 4 cases had only 1 control available for each, and in 2 cases, there were 3 controls used for each. Data were collected and managed using REDCap electronic data capture tools [13]. The Social Security Death Index was used to confirm and identify death dates.

Definitions

CDI was defined as a clinical history compatible with CDI, diarrheal stool (unformed stool conforming to the shape of a specimen container), and a positive test result for toxin-producing *C. difficile*. Because the frequency and severity of stool output is difficult to determine retrospectively from physician and nursing notes, it was not included in our case definition. Recurrent CDI was defined as that occurring after the completion of a course of metronidazole or vancomycin for an initial episode [14].

Patient follow-up included the period from January 2003 through December 2009. From October 2002 through May 2004, *C. difficile* toxin was tested using the cell culture cytotoxin neutralization assay [15]. From June 2004 through June 2009, an initial screen was performed for *C. difficile* common antigen (glutamate dehydrogenase) by means of a commercial antigen immunoassay (C. DIFF CHEK-60; TechLab), and positive tests were confirmed with a cell culture cytotoxin neutralization assay. From June 2009 through December 2009, an initial screen for common antigen was performed, followed by confirmatory testing for toxin using quantitative polymerase chain reaction (PCR) for the toxin B gene (BD GeneOhm Cdiff assay; BD Diagnostics). The NAP1 strain was detected in 21% and 43% of convenience samples among randomly chosen hospital isolates in 2005 and 2008, respectively [16].

Infections were defined as bacterial, viral, or fungal on the basis of symptoms, microbiologic data, and response to specific treatment. Fungal infections were defined as possible, probable, or proven based on established guidelines [17]. GVHD was graded according to the Keystone criteria [18] and National Institutes of Health guidelines [19]. Fever was defined as any temperature of ≥38.0°C at the time of CDI. Neutropenia, lymphopenia, and monocytopenia were defined as <500, <300, and <300 cells/mL, respectively, on 3 consecutive days concurrent with CDI. Hypoalbuminemia was defined as a serum albumin level of <2.5 mg/dL. Acute renal failure was defined as a creatinine level >50% of the baseline level. Antipseudomonal penicillins, fourth-generation cephalosporins, carbapenems, absorbable fluoroquinolones, and clindamycin were considered high-risk antibiotics [20, 21]. Failure to engraft was defined as <5% donor cells on day 60. Transplant-related mucositis [22] was defined according to established guidelines.

Statistical Analysis

Among the overall population of HSCT recipients from 2003 through 2008, incidence rates were calculated by dividing the number of patients who developed CDI within 1 year of transplant by the total number of transplants performed in the cohort year. For patients who received >1 HSCT in a calendar year, the first transplant was used. A test for linear trend was used to compare differences in the rates of infection by transplant year. Two-sided P values of ≤.05 were considered to be significant.

In the case-control analysis, patients were stratified by graft type using a matched study design. Because of few autologous HSCT recipients, multivariate risk analysis was performed among allogeneic HSCT recipients only; no significant risks were noted in univariate analysis of autologous HSCTs. Conditional logistic regression was used to account for the matched study design. A multivariable model was built in a stepwise fashion using variables from univariate analyses ($P < .1$), considering biologic plausibility and potential confounders. Time to GI GVHD after HSCT was estimated with Kaplan-Meier curves, and rates of GVHD by infection status were compared using the log-rank test.

All patients with CDI were included in a cohort analysis for recurrent disease. Univariate analyses were performed

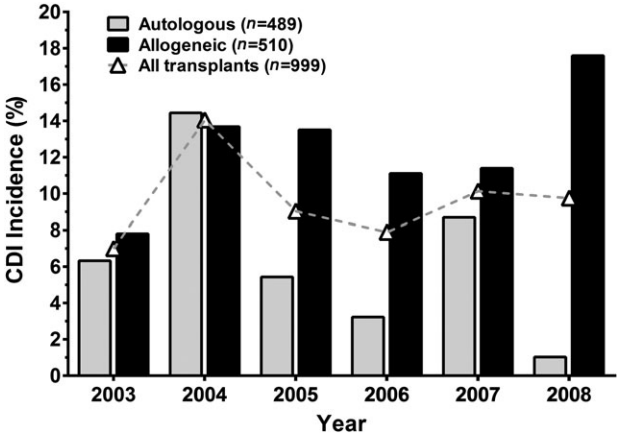

Figure 1. One-year incidence of *Clostridium difficile* infection (CDI), stratified by transplant type. Overall CDI incidence did not vary significantly from 2003 through 2008. Infections were recorded from 7 days before transplant through 1 year after transplant. Patients having received >1 transplant were included once per calendar year.

using χ^2 tests and Fisher exact tests. A multivariate unconditional logistic regression model was built in a stepwise fashion. Kaplan-Meier survival curves were generated for allogeneic HSCT recipients to compare 1-year all-cause mortality between patients who developed CDI and those who did not. The log-rank test was used to compare survival between the 2 groups.

RESULTS

Study Population and CDI Rates

From 2003 through 2008, 999 adult HSCTs were performed, including 489 autologous and 510 allogeneic HSCTs. CDI was observed among 92 patients in the cohort (9.2%). The overall incidence of CDI was 6.5% (30 of 489 patients) in autologous HSCT recipients and 12.5% (62 of 510 patients) in allogeneic HSCT recipients. All cases were diagnosed before the institutional switch to PCR-based testing in June 2009.

The overall CDI incidence did not vary significantly during the study period ($P = .36$) despite institutional increases in the prevalence of NAP1 strains (Figure 1). The observed increased rates among allogeneic transplants in 2004 and 2008 did not meet statistical significance ($P = .39$ and $P = .27$, respectively). A marginally significant decrease in disease incidence was observed among autologous transplants in 2008 ($P = .06$).

Among autologous HSCT recipients, 26 cases (86.7%) occurred within the first month after HSCT (median time, 6.5 days; interquartile range [IQR], day −1 to day 21). Among allogeneic HSCT recipients, the median time to infection was 33 days (IQR, 5–70 days; Figure 2*A* and 2*B*).

Case-Control Study for Risks

Ninety-two people who developed CDI (cases) were matched to 182 controls (Table 1). Among patients with CDI (n = 92), there were few who had a prior history of documented CDI (3 people [3.3%]). Development of other infections appeared more frequently during the first 40 days among the patients who developed CDI compared with controls (72.6% vs 58.5%; $P = .04$).

Risk factors for CDI among allogeneic HSCT recipients (n = 185) were evaluated (Table 2). Univariate analysis initially revealed that acute myelogenous leukemia/myelodysplastic syndrome, receipt of myeloablative conditioning, receipt of high-risk C. *difficile* antibiotics, acute GVHD (aGVHD; all grades), GVHD treatment, and vancomycin-resistant *Enterococci* (VRE) colonization were associated with increased odds of CDI (all $P < .05$). The strongest association was observed between CDI and aGVHD involving the GI tract (odds ratio [OR], 3.38; 95% confidence interval [CI], 1.49–7.65; $P = .004$).

In the multivariable model, receipt of chemotherapy prior to HSCT conditioning (adjusted OR [AOR], 6.39; 95% CI, 1.00–40.74; $P = .049$), receipt of high-risk antibiotics after transplant (AOR, 4.66; 95% CI, 1.23–17.66; $P = .02$), aGVHD (AOR, 3.82; 95% CI, 1.39–10.50; $P = .009$), and VRE colonization (AOR, 6.27; 95% CI, 2.07–19.01; $P = .001$) remained independently associated with CDI. Receipt of a proton pump inhibitor (PPI) demonstrated a protective effect on CDI in the univariate analysis and remained significant in the multivariate analysis (AOR, 0.29; 95% CI, 0.11–0.78; $P = .01$). Treatment of GVHD and early infectious complications were omitted from the model because of association with aGVHD and receipt of antibiotics, respectively.

Relationship Between CDI and GVHD

We evaluated the relationship between CDI and GVHD in more depth. This analysis focused specifically on GI tract GVHD because of similarities in clinical presentation compared with infectious diarrhea. There were 15 case patients and 7 control patients who developed Grade ≥2 GVHD involving the GI tract (median time to GI GVHD, 61 days; IQR, 34–165 days). Patients who developed CDI were more likely to develop GI GVHD compared with patients who never developed CDI (log-rank test; $P < .001$; Figure 3). The median time to diagnosis of GI GVHD among case patients with CDI was also shorter compared with those who never developed CDI (58.5 days vs 77 days), but the difference was not statistically significant ($P = .82$).

The timing of CDI and GVHD was evaluated more closely, including only patients who had a biopsy-confirmed diagnosis of GI tract GVHD with an available date (14 cases).

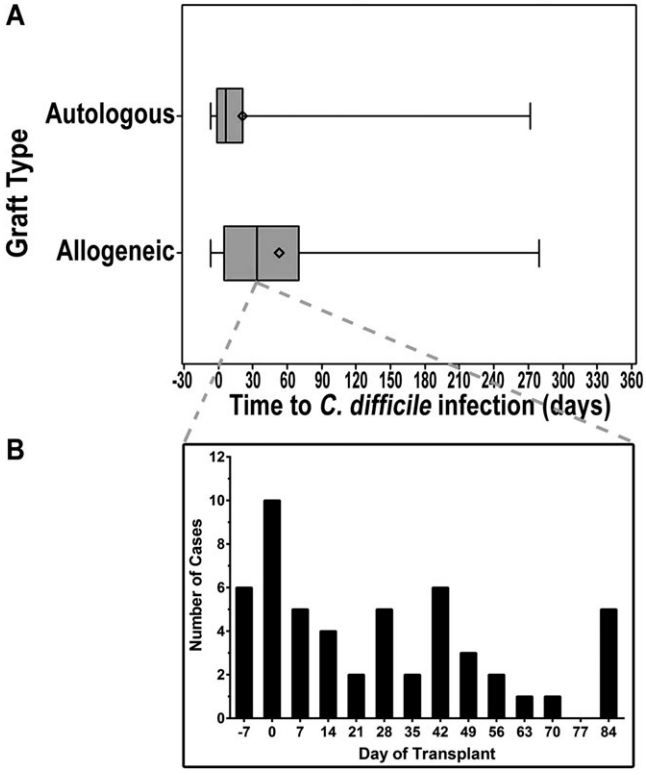

Figure 2. Timing of *Clostridium difficile* infection (CDI) by transplant type. Among autologous hematopoietic stem cell transplant (HSCT) recipients with CDI (n = 30), the median time to infection was 6.5 days compared with a median time to infection of 33 days among allogeneic HSCT recipients (n = 62). The inset demonstrates the timing of infection among allogeneic HSCT recipients. An additional 5 patients who received allogeneic transplants had disease after day 180 (not shown).

The diagnosis of CDI preceded the diagnosis of GI GVHD in the majority of subjects (12 [85.7%] of 14 patients). Among the 12 patients who developed GI GVHD following CDI, GI GVHD diagnosis occurred at a median of 21.5 days after CDI (IQR, 12–49.5 days).

Clinical Characteristics and Outcomes of CDI

Fever at the time of CDI was observed in 29.3% of patients (n = 27) for a mean duration of 2 days. Cytopenia was common, and traditional markers of severe CDI were infrequent; 2 people had white blood cell counts of >20 000 cells/mm^3 (2.2%), 5 had hypoalbuminemia (5.4%), and 10 had acute renal failure (10.9%). Most people had normal serum albumin levels (median level, 3.5 g/dL; IQR, 3.2–3.9 g/dL) and leukopenia (median white blood cell count, 3870 cells/mm^3; IQR, 610–6470 cells/mm^3).

Most patients (n = 88 [95.7%]) received *C. difficile*–targeted antimicrobial therapy with oral metronidazole alone (n = 69 [75%]). Others received oral vancomycin (n = 2), oral vancomycin plus oral metronidazole (n = 6), intravenous metronidazole (n = 2), oral plus intravenous metronidazole (n = 6), or all 3 agents (n = 3).

Recurrent CDI was observed in 20 patients (21.7% [median time, 69 days after initial CDI; IQR, 34–148 days; Table 3). CDI recurrence was more frequent in patients who received therapy with metronidazole only (23.4%) compared with patients who received vancomycin-containing regimens (18.2%), but the finding did not reach statistical significance (OR, 1.98; 95% CI, 0.41–9.62; P = .40). Recurrent CDI was strongly associated with GI GVHD (any grade; AOR, 4.23; 95% CI, 1.20–14.86; P = .02). When only biopsy-proven severe GVHD (grade ≥2) was considered as a risk, there was only a marginally significant difference between cases and controls (AOR, 2.82; 95% CI, 0.82–9.77; P = .10).

Colonoscopies or flexible sigmoidoscopies were performed in 12 patients (13%). Pseudomembranes were observed in 3 patients (3.3%). Few patients (n = 11 [23%]) had radiographic imaging. Ascites were the most common finding (n = 4 [36.4%]). There were no colectomies or episodes of toxic megacolon among cases. One patient required mechanical ventilation, which was attributed to an unrelated cryptogenic organizing pneumonia.

Of the patients with CDI, 28 (30.4%) died in the 1-year follow-up period. Kaplan-Meier survival curves among allogeneic HSCT recipients (n = 485) showed no difference in all-cause mortality at 1 year for case patients compared with the uninfected allogeneic HSCT recipients (log-rank test; P = .80).

DISCUSSION

This study represents the largest and most comprehensive examination of the epidemiology, timing, and outcomes of CDI in the HSCT population, spanning a period during which epidemic toxigenic strains increased in the hospital. Data underscore the early timing of CDI in HSCT recipients and outline the role of both host variables (receipt of chemotherapy prior to HSCT) and antimicrobial use as conferring risks for infection. Data indicate that CDI enhances risks for GVHD involving the GI tract, suggesting the potential involvement of microbial antigenicity or damage response in the pathogenesis of GVHD.

In this study, we found that CDI is a major complication of HSCT, affecting 9.2% of all HSCT recipients. The overall incidence of CDI was 6.5% in autologous HSCT recipients and 12.5% in allogeneic HSCT recipients. Higher rates among allogeneic HSCT recipients may be explained by numerous differences, including longer hospitalizations, increased antibiotic exposures, and longer periods of impaired host immunity.

Risk factors for CDI in the general hospitalized population include increased age, antibiotic exposure, and duration of hospitalization [23]. Host risks are not well understood in

Table 1. Demographic and Clinical Characteristics of 274 Hematopoietic Stem Cell Transplant Recipients at The Johns Hopkins Hospital, 2003–2008

Characteristic	Control Patients (n = 182)	Case Patients With CDI (n = 92)	All Patients (n = 274)
Age in years at transplant, median (IQR)	50 (43–59)	50 (40–60)	50 (41–59)
Male sex	94 (51.6)	60 (65.2)	154 (56.2)
Race or ethnicity			
White	142 (78.0)	71 (77.2)	213 (77.7)
African American	31 (17.0)	13 (14.1)	44 (16.1)
Other or unknown	9 (4.9)	8 (8.7)	17 (6.2)
Underlying condition			
Acute myelogenous leukemia or myelodysplastic syndrome	44 (24.2)	34 (37.0)	78 (28.5)
Acute lymphoblastic leukemia	9 (4.9)	3 (3.3)	12 (4.4)
Chronic myelogenous leukemia	6 (3.3)	7 (7.6)	13 (4.7)
Chronic lymphocytic leukemia	14 (7.7)	3 (3.3)	17 (6.2)
Multiple myeloma	16 (8.8)	6 (6.5)	22 (8.0)
Lymphoma	78 (42.9)	28 (30.4)	106 (38.7)
Other malignancy[a]	11 (6.0)	7 (7.6)	18 (6.6)
Other condition[b]	4 (2.2)	4 (4.3)	8 (2.9)
Prior stem cell transplantation	13 (7.1)	4 (4.3)	17 (6.2)
Myeloablative conditioning	131 (72.0)	77 (83.7)	208 (75.9)
Receipt of total body irradiation	48 (26.4)	19 (20.7)	67 (24.5)
Transplant type			
Autologous	45 (24.7)	22 (23.9)	67 (24.5)
Allogeneic	123 (67.6)	62 (67.4)	185 (67.5)
Matched related[c]	61 (49.6)	34 (54.8)	95 (51.4)
Matched unrelated[c]	21 (17.1)	17 (27.4)	38 (20.5)
Mismatched related[c]	40 (32.5)	11 (17.7)	51 (27.6)
Mismatched donor[c]	1 (0.8)	0 (0.0)	1 (0.5)
High-dose cyclophosphamide	14 (7.7)	8 (8.7)	22 (8.0)
Source of stem cell			
Bone marrow	125 (74.4)	59 (70.2)	184 (73.0)
Peripheral blood	42 (25.0)	23 (27.4)	65 (25.8)
Cord blood	1 (0.6)	2 (2.4)	3 (1.2)
Stem cell manipulation			
CD34 selected[d]	21 (12.5)	12 (14.3)	33 (13.1)
T-cell depleted[c]	5 (3.0)	0 (0.0)	5 (2.0)
CMV serostatus[c]			
D−/R−	37 (20.3)	12 (13.0)	49 (17.9)
D+/R−	12 (6.6)	7 (7.6)	19 (6.9)
D−/R+	24 (13.2)	9 (9.8)	33 (12.0)
D+/R+	29 (15.9)	16 (17.4)	45 (16.4)
Unknown	21 (11.5)	18 (18.5)	38 (13.9)

Data are no. (%) of subjects, unless otherwise indicated. Patients were matched by graft type and calendar time.

Abbreviations: CDI, C. difficile infection; CMV, cytomegalovirus; D/R, donor/recipient relationship; IQR, interquartile range.

[a] The most common other malignancies are acute and chronic myelomonocytic leukemia and promyelomonocytic leukemia.

[b] The most common other conditions are myasthenia gravis, systemic lupus erythematosis, and multiple sclerosis.

[c] Among allogeneic transplants only.

[d] Among autologous transplants only.

HSCT recipients. Here, we found that receipt of chemotherapy prior to HSCT, broad-spectrum antimicrobial use, and VRE colonization are the strongest predictors for CDI. VRE colonization may represent a surrogate marker for severity of illness or may correlate with prior antimicrobial exposure and disrupted gut microbiota. Results of univariate analysis also

Table 2. Risk Factor Analysis for *Clostridium difficile* Infection Among Allogeneic Hematopoietic Stem Cell Transplant Recipients and Matched Controls at The Johns Hopkins Hospital, 2003–2008

Characteristic	No. (%) of Patients			Crude OR (95% CI)	*P*	AOR (95% CI)	*P*	AOR[a] (95% CI)	*P*
	Control (n = 123)	Case (n = 62)	Total (n = 185)						
Recipient-related factors									
Age in years, median (IQR)	50 (42–59)	48.5 (38–56)	50 (41–59)	…	…	…	…	…	…
Male sex	61 (49.6)	39 (62.9)	100 (54.1)	1.63 (0.88–3.02)	.12	…	…	…	…
Hematologic malignancy									
AML or MDS	41 (33.3)	32 (51.6)	73 (39.5)	1.97 (1.08–3.58)	.03	…	…	…	…
ALL	9 (7.3)	3 (4.8)	12 (6.5)	0.67 (0.18–2.46)	.54	…	…	…	…
CML	6 (4.9)	7 (11.3)	13 (7.0)	2.86 (0.81–10.05)	.1	…	…	…	…
CLL	13 (10.6)	3 (4.8)	16 (8.6)	0.45 (0.13–1.64)	.23	…	…	…	…
MM	6 (4.9)	3 (4.8)	9 (4.9)	1.00 (0.16–4.68)	.99	…	…	…	…
Lymphoma	43 (35.0)	8 (12.9)	51 (27.6)	0.26 (0.11–0.61)	.0021	…	…	…	…
Other malignancy	5 (4.1)	6 (9.7)	11 (5.9)	2.35 (0.71–7.75)	.16	…	…	…	…
Prior chemotherapy regimens									
None	14 (11.4)	2 (3.2)	16 (8.6)	1.0 (Ref)	>.999 (Ref)	1.0 (Ref)	…	…	…
1–3 regimens	90 (73.2)	50 (80.6)	140 (75.7)	4.53 (0.95–21.47)	.06	6.39 (1.00–40.74)	.049	6.63 (1.04–42.22)	.045
>3 regimens	19 (15.4)	10 (16.1)	29 (15.7)	4.45 (0.79–25.15)	.09	6.32 (0.83–48.11)	.08	5.92 (0.79–44.18)	.08
Antibiotics within ≤1 month of HSCT	59 (48.0)	21 (33.9)	80 (43.2)	0.53 (0.26–1.05)	.07	0.53 (0.21–1.33)	.18	0.53 (0.21–1.36)	.19
Transplant-related characteristics									
Myeloablative conditioning	73 (59.3)	47 (75.8)	120 (64.9)	2.08 (1.06–4.11)	.03	0.37 (0.03–5.51)	.47	0.26 (0.02–4.19)	.35
Source of stem cell									
Bone marrow	122 (99.2)	56 (90.3)	178 (96.2)	1.0 (Ref)	…	…	…	…	…
Peripheral blood	0 (0)	4 (6.5)	4 (2.2)	Inf. (NA)	.99	…	…	…	…
Cord blood	1 (0.8)	2 (3.2)	3 (1.6)	3.77 (0.32–43.72)	.29	…	…	…	…
Transplant type[b]									
Matched related	61 (49.6)	34 (54.8)	95 (51.4)	1.0 (Ref)	…	…	…	…	…
Matched unrelated	21 (17.1)	17 (27.4)	38 (20.5)	1.36 (0.62–2.96)	.44	…	…	…	…
Unmatched related	40 (32.5)	11 (17.7)	51 (27.6)	0.52 (0.23–1.16)	.11	…	…	…	…
Receipt of steroids (day 0–40)	68 (55.3)	42 (67.7)	110 (59.5)	1.65 (0.89–3.08)	.12	…	…	…	…
Receipt of PPI (day 0–40)	98 (79.7)	40 (64.5)	138 (74.6)	0.42 (0.20–0.88)	.02	0.29 (0.11–0.78)	.01	0.30 (0.11–0.81)	.02
Hospital stay in days for HSCT, median (IQR)	25 (0–29)	26.5 (3–30)	25 (0–30)	…	…	…	…	…	…
<20 days	50 (40.7)	16 (25.8)	66 (35.7)	1.0 (Ref)	>.999 (Ref)	…	…	…	…
≥20 days	73 (59.3)	46 (74.2)	119 (64.3)	1.88 (0.98–3.61)	.06	4.00 (0.31–52.33)	.29	4.96 (0.35–69.72)	.23
High-risk *C. difficile* antibiotic use[c]	95 (77.2)	58 (93.5)	153 (82.7)	3.91 (1.34–11.38)	.01	4.66 (1.23–17.66)	.02	4.76 (1.20–18.88)	.03

Table 2 continued.

Characteristic	No. (%) of Patients			Crude OR (95% CI)	*P*	AOR (95% CI)	*P*	AOR[a] (95% CI)	*P*
	Control (n = 123)	Case (n = 62)	Total (n = 185)						
Transplant-related complications									
Mucositis[d]	49 (39.8)	26 (41.9)	75 (40.5)	1.17 (0.60–2.26)	.65	…	…	…	…
Grades 1–2	39 (79.6)	22 (84.6)	61 (81.3)	…	…	…	…	…	…
Grade ≥3	7 (14.3)	3 (11.5)	10 (13.3)	…	…	…	…	…	…
Failure to engraft	12 (9.8)	1 (1.6)	13 (7.0)	0.17 (0.02–1.32)	.09	0.29 (0.03–2.53)	.26	0.32 (0.04–2.94)	.31
GVHD, acute (all)	53 (43.1)	39 (62.9)	92 (49.7)	2.40 (1.22–4.74)	.01	3.82 (1.39–10.50)	.009	…	…
More than 1 site with grade ≥2 GVHD	44 (83.0)	38 (97.4)	82 (89.1)	2.90 (1.46–5.77)	.002	4.45 (1.54–12.84)	.006	…	…
Location of acute GVHD									
GI tract (all)	11 (8.9)	17 (27.4)	28 (15.1)	3.38 (1.49–7.65)	.004	…	…	…	…
Grade ≥2[e]	7 (5.7)	15 (24.2)	22 (11.9)	4.47 (1.72–11.63)	.002	…	…	…	…
Skin (all)	51 (41.5)	34 (54.8)	85 (46.0)	1.73 (0.92–3.28)	.09	…	…	…	…
Grade ≥2	44 (35.8)	31 (50.0)	75 (40.5)	1.86 (0.96–3.60)	.07	…	…	…	…
Liver (all)	7 (5.7)	5 (8.1)	12 (6.5)	1.35 (0.43–4.27)	.61	…	…	…	…
Grade ≥2	4 (3.3)	6 (9.7)	10 (5.4)	2.58 (0.71–9.30)	.15	…	…	…	…
GVHD, chronic	14 (11.4)	7 (11.3)	21 (11.4)	0.99 (0.37–2.62)	.97	…	…	…	…
Treatment for GVHD	50 (40.7)	39 (62.9)	89 (48.1)	2.4 (1.27–4.54)	.007	…	…	…	…
Early infection (any), day 0–40	72 (58.5)	45 (72.6)	117 (63.2)	2.2 (1.05–4.62)	.04	…	…	…	…
VRE colonization	24 (19.5)	27 (43.5)	51 (27.6)	4.04 (1.84–8.85)	.0005	6.27 (2.07–19.01)	.001	5.87 (1.97–17.47)	.002

Prior stem cell transplantation, receipt of a T-cell–depleted graft, and rituximab use during transplant were evaluated but did not reach statistical significance.

Abbreviations: ALL, acute lymphoblastic leukemia; AML, acute myelogenous leukemia; AOR, adjusted odds ratio; CI, confidence interval; CLL, chronic lymphocytic leukemia; CML, chronic myelogenous leukemia; GI, gastrointestinal; GVHD, graft-versus-host disease; HSCT, hematopoietic stem cell transplant; IQR, interquartile range; MDS, myelodysplastic syndrome; MM, multiple myeloma; OR, odds ratio; PPI, proton pump inhibitor; Ref, reference value; VRE, vancomycin-resistant *Enterococci*.

[a] Analysis performed for acute GVHD grade 2 or higher only.

[b] One control patient received a transplant from a mismatched donor.

[c] High-risk antibiotic use included receipt of antipseudomonal penicillin, fourth-generation cephalosporin, carbapenem, absorbable fluoroquinolone, or clindamycin.

[d] Three control patients and 1 case patient had an unknown grade of mucositis.

[e] Two control patients and 1 case patient were excluded from the time to GI tract GVHD analysis due to missing date of biopsy or development of GVHD >1 year after HSCT.

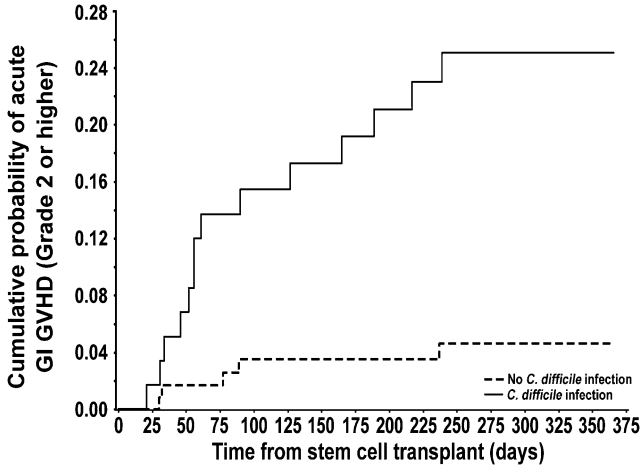

Figure 3. Comparison of acute graft-versus-host disease (GVHD) following transplantation among allogeneic transplant recipients with and without *Clostridium difficile* infection (CDI; n = 180). The 1-year probability of developing grade 2 or higher gastrointestinal (GI) GVHD was 25% in case patients and 4.6% in control patients (log-rank test; $P = .0001$).

suggested that underlying disease is an important determinant of posttransplant CDI risk. However, assessing the contribution of underlying disease in multivariable models presents complications given collinearity between underlying disease and transplant type. Prospective studies will need to be performed to account for clinical variables within subgroups.

CDI typically occurs early in the transplant course, although risks appear to be more protracted in allogeneic HSCT recipients [24, 25]. The very early development of disease, which is within 1 week in autologous HSCT recipients, suggests that a proportion of people may be colonized with toxigenic *C. difficile* prior to receipt of conditioning therapy rather than acquiring infection after hospitalization. This hypothesis is supported by additional observations that receipt of chemotherapy prior to conditioning for allogeneic HSCT predicts risk for CDI after transplant. Prospective studies are warranted to define the natural history of CDI after HSCT, as early identification of high-risk patients may allow for development of preventative strategies.

In this analysis, patients who received PPIs after HSCT had a reduction in risk for primary CDI. This finding contradicts some published reports suggesting that PPI use may increase the risk for CDI, although the published data in this field appear to be conflicting [26, 27]. PPI use may represent a marker of severity of illness, rather than a causative factor for CDI.

Results of small studies have suggested a possible link between GVHD and development of CDI in allogeneic HSCT recipients [28, 29]. Our study evaluated this potentially important interplay in greater depth, as GVHD is such an important factor associated with overall transplant outcomes. Specifically, we closely evaluated timing, included conservative definitions of GVHD, and evaluated the relationship of CDI specifically to GVHD involving the GI tract. Data demonstrate that GVHD is associated with a high risk for CDI. Because more of these patients have diarrhea, this association may reflect a considerable testing bias. However, the most intriguing observation is that the diagnosis of CDI occurred *before* that of GI tract GVHD in the bulk of patients. This raises the likelihood that CDI is actually serving to increase the biologic risk for GVHD involving the GI tract, implicating a component of microbial antigenicity or response to mucosal damage that serves to drive or enhance the development of GI GVHD. Our current understanding of the pathophysiology of aGVHD is that there are multiple steps involved, with an initial insult to the GI tract serving to amplify an immune response [30]. Initially, components of preparative regimens, such as total body irradiation or other agents administered as part of conditioning, serve to damage tissues. Proinflammatory cytokines promote a milieu in which antigen-presenting cells activate donor T cells, driving a cascade of cytotoxicity against recipient tissues. It is likely that local infection in the gut may further propagate this cycle by destroying epithelial integrity in the beginning and/or by augmenting responses during a cycle of inflammation [31].

We observed that HSCT recipients with CDI are less likely to have the traditional markers of severe disease such as fever, hypoalbuminemia, and leukocytosis [32–34] and that the majority of patients with CDI responded to standard treatment courses with oral metronidazole. This observation raises 2 possibilities. First, the definitions historically applied to severe CDI come from data from nonimmunocompromised hosts and may not be applicable to the population of HSCT recipients. It is biologically plausible that these patients may have an attenuated colonic inflammatory response to *C. difficile* toxin as a consequence of transplantation. Observational data suggest that pseudomembranes may not be present in patients after HSCT [35]; hence, the pathophysiology of CDI in this population may be different.

We also wanted to test whether these patients have higher rates of recurrent CDI, as predicted by antibody deficiency or functional immune impairment. In total, 21.7% of HSCT patients with CDI had a CDI recurrence, which is consistent with recurrence rates in the general hospitalized population [36]. In allogeneic HSCT recipients, GI GVHD conferred nearly 5-fold higher odds of CDI recurrence. This finding remained statistically significant after controlling for variables considered to be important in risk for recurrent disease (including steroid use) [37]. It is not clear whether this is associated with impaired systemic immunity or a local GI tract phenomenon.

Table 3. Risk Factor Analysis for Recurrent *Clostridium difficile* Infection (N = 92)

Characteristic	No. (%) of Patients			P^b	Crude OR (95% CI)	P	AOR[c] (95% CI)	P
	Single-Episode CDI (n = 72)	Recurrent CDI[a] (n = 20)	Total (n = 92)					
Recipient-related factors[d]								
Age in years, median (IQR)	50.5 (40.5–59.5)	52 (38–59.5)	50.5 (39.5–59.5)	.49	…	…	…	…
18–49	35 (48.6)	9 (45.0)	44 (47.8)	…	1.00 (Ref)	…	1.00 (Ref)	…
≥50	37 (51.4)	11 (55.0)	48 (52.2)	.81	1.16 (0.43–3.13)	.78	1.09 (0.36–3.26)	.88
Myeloablative conditioning	61 (84.7)	16 (80.0)	77 (83.7)	.73	0.72 (0.20–2.57)	.61	…	…
Number of prior chemotherapy regimens				.25				
None[e]	7 (9.7)	0 (0.0)	7 (7.6)	…	…	…	…	…
1–3 regimens	56 (77.8)	16 (80.0)	72 (78.3)	…	1.00 (Ref)	…	1.00 (Ref)	…
>3 regimens	9 (12.5)	4 (20.0)	13 (14.1)	…	1.75 (0.48–6.42)	.40	1.04 (0.23–4.58)	.96
Rituximab use during transplant	19 (26.4)	4 (20.0)	23 (25.0)	.77	0.70 (0.21–2.35)	.56	…	…
Transplant-related factors								
Receipt of steroids (day 0–40)	55 (76.4)	11 (55.0)	66 (71.7)	.09	0.38 (0.13–1.06)	.07	0.38 (0.12–1.18)	.09
Receipt of PPI	47 (65.3)	13 (65.0)	60 (65.2)	>.99	0.99 (0.35–2.79)	.98	…	…
Hospital stay in days for HSCT, median (IQR)	21 (3–28)	25.5 (2–32.5)	21.5 (3–28)	.41	…	…	…	…
Antimicrobial utilization								
Antibiotics within ≤1 month of HSCT	22 (30.6)	5 (25.0)	27 (29.3)	.78	0.76 (0.25–2.34)	.63	…	…
High-risk *C. difficile* antibiotic use	62 (86.1)	18 (90.0)	80 (87.0)	>.99	1.45 (0.29–7.23)	.65	…	…
Transplant-related complications								
GVHD, acute (all)	31 (43.1)	12 (60.0)	43 (46.7)	…	…	…	…	…
More than 1 site with grade ≥2 GVHD	30 (41.7)	11 (55.0)	41 (44.6)	.32	1.71 (0.63–4.64)	.29	…	…
Location of acute GVHD								
GI tract (all)	9 (12.5)	8 (40.0)	17 (18.5)	.009	4.67 (1.50–14.52)	.008	4.23 (1.20–14.86)	.02
Grade ≥2	9 (12.5)	6 (31.6)	15 (16.7)	.08	3.18 (0.96–10.49)	.06	…	…
Skin (all)	27 (37.5)	10 (50.0)	37 (40.2)	.44	1.67 (0.61–4.52)	.32	…	…
Grade ≥2	27 (37.5)	7 (35.0)	34 (37.0)	>.99	0.90 (0.32–2.53)	.84	…	…
Liver (all)	3 (4.2)	2 (10.0)	5 (5.4)	.30	2.56 (0.40–16.46)	.32	…	…
Grade ≥2	3 (4.2)	3 (15.8)	6 (6.7)	.11	4.19 (0.77–22.71)	.10	…	…
CDI treatment modality[f]								
Regimens containing metronidazole only	59 (81.9)	18 (90.0)	77 (83.7)	.51	1.98 (0.41–9.62)	.40	…	…
Metronidazole, oral	52 (72.2)	17 (85.0)	69 (75.0)	…	…	…	…	…
Metronidazole, intravenous	2 (2.8)	0 (0.0)	2 (2.2)	…	…	…	…	…
Metronidazole, oral and intravenous	5 (6.9)	1 (5.0)	6 (6.5)	…	…	…	…	…
Vancomycin-containing regimens	9 (12.5)	2 (10.0)	11 (12.0)	>.99	0.78 (0.15–3.93)	.76	…	…
Vancomycin, oral; and metronidazole, oral	5 (2.8)	1 (5.0)	6 (6.5)	…	…	…	…	…
Vancomycin, oral	2 (6.9)	0 (0.0)	2 (2.2)	…	…	…	…	…
Vancomycin, oral; and metronidazole, oral and intravenous	2 (2.8)	1 (5.0)	3 (3.3)	…	…	…	…	…
CDI treatment duration in days, median (IQR)	15 (11–16)	15.5 (13–19.5)	15 (11–17)	.23	…	…	…	…

Abbreviations: AOR, adjusted odds ratio; CDI, *C. difficile* infection; CI, confidence interval; GI, gastrointestinal; GVHD, graft-versus-host-disease; HSCT, hematopoietic stem cell transplant; IQR, interquartile range; OR, odds ratio; PPI, proton pump inhibitors; Ref, reference value.

[a] Relapse patients included 17 allogeneic HSCT recipients (85%), 2 autologous HSCT recipients (10%), and 1 recipient of high-dose cyclophosphamide (5%). More than 1 relapse was observed in 3 patients.

[b] Fisher exact test for categorical variables, trend test for ordinal variables, Wilcoxon test for continuous variables.

[c] When analysis was limited to GVHD grade 2 or higher, the AORs included the following: age of >50 years, 1.15 (95% CI, 0.39–3.37; $P = .8$); receipt of >3 prior chemotherapy agents, 1.26 (95% CI, 0.30–5.38; $P = .75$); receipt of steroids (D0-40), 0.39 (95% CI, 0.13–1.20; $P = .10$); and GI GVHD, 2.82 (95% CI, 0.82–9.77; $P = .10$).

[d] No differences were observed on the basis of sex, race, underlying disease, or history of prior HSCT.

[e] Categories for 0 and 1–3 were combined because no patients initiating chemotherapy for the first time (ie, 0 regimens) had recurrent disease.

[f] All patients who did not receive CDI-specific therapy (n = 4) had single episodes of CDI.

There are limitations to this study. This was a heterogeneous population of patients undergoing HSCT. In the study period, multiple HSCT protocols were used, limiting detailed evaluation of protocol-specific risks. Inclusion of all cases testing positive for toxin may overestimate rates of CDI in this study and may underestimate associated risk factors. Finally, there are disparities in patient follow-up (eg, closer monitoring of allogeneic HSCT recipients and patients with GVHD) and more frequent CDI testing that may explain some of the differences in CDI rates among HSCT types.

In conclusion, this single-study experience from 2003 through 2008 demonstrates that CDI is a frequent, early complication after HSCT. Infections are related to underlying host risks at time of HSCT in addition to transplant-related complications, suggesting that some people may have predetermined high risks prior to conditioning therapy. In addition, there is a potentially important interplay between CDI and GVHD involving the GI tract. Future prospective studies would be valuable, especially to define the natural history of early disease, determine mechanisms of interaction between infectious colitis and GVHD, and define preventative strategies.

Notes

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