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Original Article

Clostridioides difficile infection in patients with hematological malignancy: A multicenter study in Taiwan

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Abstract *Background:* Among the individuals with hematological malignancy (HM) complicated with *Clostridioides difficile* infection (CDI), the variables associated with in-hospital mortality and recurrence of CDI were investigated.
Material and methods: Including adults with HM and those without malignancy suffering from CDI from January 2015 to December 2016 in three hospitals in Taiwan.

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Recurrence;
Clostridioides difficile infection

Results: Totally 314 patients including 77 with HM and 237 patients without malignancy were included. HM patients more often had low leukocyte counts (<500 cells/mL: 28.6% *vs.* 2.1%) than those without malignancy and more patients without malignancy had severe CDI than patients with HM (31.6% *vs.* 14.3%, $P = .003$), according to the severity score of IDSA/SHEA. Patients with HM had a higher recurrence rate of CDI (14.3%, 11/77 *vs.* 7.2%, 17/237; $P = .07$) and longer hospital stay (47.2 ± 40.8 days *vs.* 33.3 ± 37.3 days; $P = .006$) than those without malignancy. In the multivariate analyses for those with HM and CDI, the in-hospital mortality was associated with vancomycin-resistant *Enterococcus* (VRE) colonization or infection (odds ratio [OR] 7.72; $P = .01$), and *C. difficile* ribotype 078 complex infection (OR 9.22; $P = .03$). Moreover underlying hematological malignancy (OR 2.74; $P = .04$) and VRE colonization/infection (OR 2.71; $P = .02$) were independently associated with CDI recurrence.
Conclusion: Patients with HM complicated with CDI were often regarded as non-severe infection, but had a similar in-hospital mortality rate as those without malignancy. CDI due to ribotype 078 complex isolates heralded a poor prognosis among HM patients.

Introduction

In the era of antimicrobial therapy, *Clostridioides difficile* may result in disease, ranging from mild diarrhea, pseudomembranous colitis, toxic megacolon, to death with a mortality rate of up to 25%—40%.[1–3] In the clinical guidelines issued in 2017 by the Infectious Diseases Society of America (IDSA) and Society for Healthcare Epidemiology of America (SHEA), a patient with leukocyte count ≥ 15,000 cells/mL or serum creatinine of >1.5 mg/dL was regarded as severe *C. difficile* infection (CDI).[4] Accordingly the guideline recommended oral vancomycin for either non-severe or severe CDI, and metronidazole as an alternative choice for non-severe CDI.[4]

Clinical burden of CDI will vary greatly in different countries, healthcare facilities, and hospital units.[2,3,5–11] It is not surprising that the highest incidence rate of CDI among the wards of internal medicine was present in hematology units of an academic center in Hungary (32.9/1000 admissions).[12] Besides, patients with hematological malignancy (HM) had a high *C. difficile* colonization rate of 9.3%, and 13.3% of colonized patients developed symptomatic disease during hospitalization.[13] In a recent review, the incidence of CDI in hematology-oncology patients ranged from 6% to 33%, with a recurrence rate of up to 40%. The proposed factors related to a significant burden of CDI among these immunocompromised individuals included frequent antimicrobial use, suppressed immune function, increased exposure to healthcare settings, and a high prevalence of *C. difficile* colonization.[14] Although the all-cause 30-day mortality rate in patients with hematological malignancies (25.4%) has been found to be higher than that in patients with solid cancer (15.0%) and without cancer (8.6%).[15]

The utility of leukocyte count and serum creatinine value as the surrogate markers of CDI severity may be problematic for patients with hematological malignancies who often had lower serum creatinine levels and leukocyte counts at the time of CDI diagnosis.[16,17] Cancer patients with CDI had higher treatment failure, mortality, and recurrent rates, but the existence of lower leukocyte count among cancer patients would provoke the classification of non-severe disease according to the IDSA/SHEA guideline, if renal dysfunction was absent.[4,16,17] However, neutropenia had been noted be an independent risk factor for CDI-related mortality in such a population.[17] The variables associated with in-hospital mortality and recurrence of CDI were investigated among the cases of HM were investigated in this study.

Materials and methods

Clinical setting and study design

This study was conducted in three hospitals in Taiwan, including the National Taiwan University Hospital (Hospital A, a medical center in northern Taiwan), Chung Shan Medical University Hospital (Hospital B, a medical center in central Taiwan), and National Cheng Kung University Hospital (Hospital C, a medical center in southern Taiwan), and was approved by the Institutional Review Board in each hospital: Hospitals A (201412190RIND), B (CS14179), and C (B-ER-103-098).

Fecal sample culture and definition of CDAD

From January 2015 to December 2016, patients aged older than 20 years with a hospital stay for at least five days were included. Those with colectomy or intestinal infection due to other enteropathogens were excluded. Those infected with fecal *C. difficile* isolates possessing *tcdB* in the presence of diarrhea without an alternative explanation were diagnosed as having CDI.[18] Diarrhea was defined as the new onset of passage of three or more unformed stools in 24 h. Toxigenic *C. difficile* isolates were detected in the feces by the real-time polymerase-chain-reaction (RT-PCR) test using the GeneXpert *C. difficile* assay (Cepheid, CA, USA) in Hospital A, and BD GeneOhmTM Cdiff assay (BD Diagnostics, San Diego, CA, USA) in two other hospitals. Stools from diarrheal patients in Hospitals A and B were sent for *C. difficile* culture, whereas in Hospital C stool samples would

be send for cultured for *C. difficile*, only if *tcdB* was detected by the RT-PCR test in the feces. Stools would be plated on cycloserine-cefoxitin-fructose selective plates and incubated anaerobically for 24—48 h.

Toxin gene detection in *C. difficile* isolates

C. difficile isolates were sub-cultured on the cycloserine-cefoxitin- fructose and incubated anaerobically for 24—48 h. In *C. difficile* isolates, *tcdA*, *tcdB*, *cdtA*, *cdtB*, and *tcdC* deletion were examined by a multiplex polymerase—chain reaction (PCR), as previously described[19]. After harvesting, genomic DNA was obtained from a Chelex 100-based method and PCR ribotyping was performed on strains with *tcdA* truncation or *tcdC* deletion as previously described.[19] Briefly, after PCR amplification, the samples were concentrated using the Gel/PCR DNA Fragments Extraction Kit (Geneaid, Ltd, Taiwan) and separated with the QIAxcel capillary electrophoresis system (Qiagen, Hilden, Germany) using the "OM500" method and QX Alignment Marker 15-bp/3-kb (Qiagen, Hilden, Germany). All PCR ribotypes were confirmed by the WEBRIBO database (http://webribo.ages.at).

Antimicrobial susceptibility of *C. difficile* isolates

The minimum inhibitory concentration (MIC) was determined by the agar dilution method, according to the Clinical and Laboratory Standards Institute (CLSI) guidelines (M11-A8)[20]. Supplemented *Brucella* agar deeps were obtained from Anaerobe Systems (Morgan Hill, CA). Frozen defibrinated sheep blood (Hema Resources, Inc., Aurora, OR) was thawed to produce laked blood. The tested isolates were applied to the plates using a Steers multipronged inoculator for a final concentration of approximately 10^5 CFU/spot. After 44 h of incubation at 36 °C in an anaerobic chamber incubator, the plates were examined for bacterial growth and the MICs interpreted. The MIC of metronidazole or vancomycin resistance >2 mg/L was regarded to be resistant, according to the breakpoint tables for interpretation MICs of European Committee on Antimicrobial Susceptibility Testing (EUCAST), version 10.0 issued in 2020.

Clinical data analysis

Information about clinical status prior to admission, including comorbid conditions or prior history of CDI was collected. Prior medications, including cytotoxic chemotherapy,[21] antimicrobial agent, proton pump inhibitors, H2-rectpor antagonist, or systemic corticosteroid, within one month before the onset of CDI were obtained from medical records. Chronic kidney disease (CKD) was defined as an estimated glomerular filtration rates (GFRs) < 60 mL/min/1.73 m^2 for at least three months.[22] The antibiotic classes and drugs studied were as follows: cephalosporins - cefazolin, cefuroxime, ceftriaxone, ceftazidime, cefepime; penicillins - penicillin, oxacillin, piperacillin; beta-lactam/beta-lactamase inhibitors - piperacillin/tazobactam, amoxicillin/clavulanic acid, ampicillin/sulbactam; carbapenems - ertapenem, imipenem/cilastatin, meropenem; and glycopeptides — vancomycin, teicoplanin.

Metronidazole treatment failure was defined as the persistence of diarrhea and/or a positive result of *C. difficile* toxin B after six days of treatment, the need for colectomy, or death after five days of therapy, otherwise success.[23] Recurrence was defined as recurrence of *C. difficile* toxin B—positive diarrhea by Day 21 after initial cure.[23] Active surveillance of vancomycin-resistant *Enterococcus* (VRE) by rectal swab or stool sample has been performed merely in the intensive care units (ICUs) in

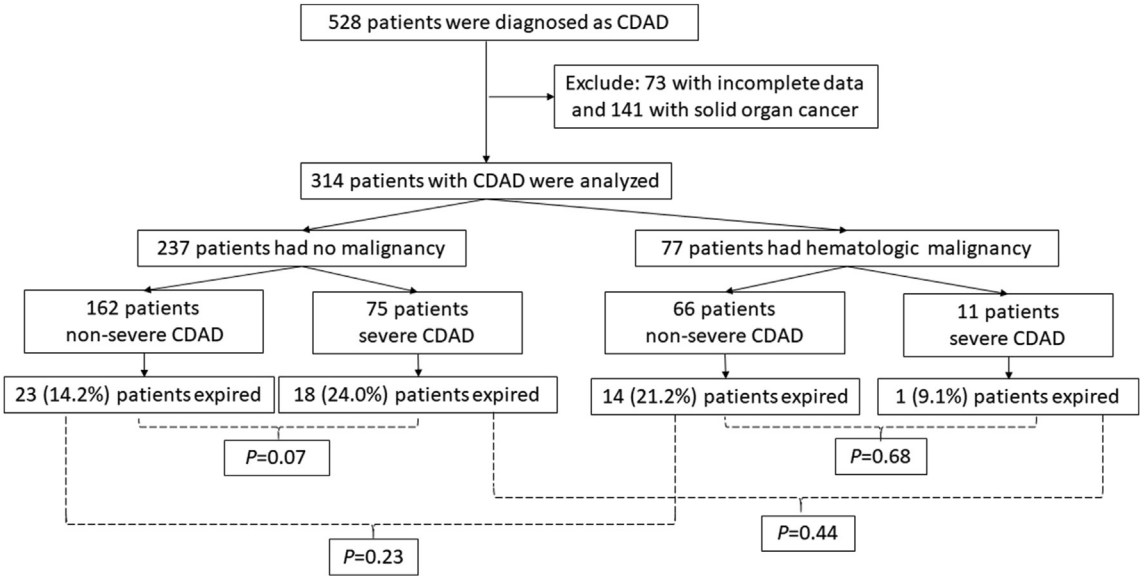

Figure 1. Flow chart of adults with hematological malignancy and without malignancy complicated by *Clostridioides difficile* infection (CDI), stratified by the severity score of IDSA/SHEA.

Table 1 Underlying diseases and prior antibiotic exposure in patients with *Clostridioides difficile* infection (CDI), stratified by the types of hematological malignancy (HM)[a].

Variables (case/isolate number)	Total n = 314	No malignancy n = 237	HM[a] n = 77	*P* value[b]	Acute leukemia n = 45	Chronic leukemia n = 10	Lymphoma n = 22
Age, years	62.1 ± 21.7	66.2 ± 20.9	49.5 ± 19.0	<0.001	44.8 ± 17.3	60.7 ± 17.8	53.9 ± 20.5
Gender, male	171 (54.5)	128 (54.0)	43 (55.8)	0.79	25 (55.6)	5 (50.0)	13 (59.1)
Underlying disease							
Hypertension	125 (39.8)	119 (50.2)	6 (7.8)	<0.001	2 (4.4)	1 (10.0)	3 (13.6)
Diabetes mellitus	81 (25.8)	76 (32.1)	5 (6.5)	<0.001	3 (6.7)	0	2 (9.1)
Chronic kidney disease	62 (19.7)	60 (25.3)	2 (2.6)	<0.001	0	0	2 (9.1)
Old stroke	38 (12.1)	37 (15.6)	1 (1.3)	<0.001	0	1 (10.0)	0
Congestive heart failure	30 (9.6)	29 (12.2)	1 (1.3)	0.003	0	1 (10.0)	0
Coronary artery disease history	24 (7.6)	24 (10.1)	0	0.001	0	0	0
Liver cirrhosis	19 (6.1)	18 (7.6)	1 (1.3)	0.05	1 (2.2)	0	0
Recent medication within one month before CDI onset							
Antimicrobial therapy							
Cephalosporins	123 (39.2)	92 (38.8)	31 (40.3)	0.89	21 (46.7)	2 (20.0)	8 (36.4)
Cefazolin, iv	16 (5.1)	15 (6.3)	1 (1.3)	0.13	1 (2.2)	0	0
Cefuroxime, iv/o	20 (6.4)	17 (7.2)	3 (3.9)	0.42	2 (4.4)	0	1 (4.5)
Ceftazidime or ceftriaxone, iv	64 (20.4)	56 (23.6)	8 (10.4)	0.01	6 (13.3)	0	2 (9.1)
Cefepime, iv	51 (16.2)	24 (10.1)	27 (35.1)	<0.001	17 (37.8)	2 (20.0)	8 (36.4)
Penicillins	80 (25.5)	62 (26.2)	18 (23.4)	0.66	8 (17.8)	1 (10.0)	9 (40.9)
Carbapenem, iv	47 (15.0)	41 (17.3)	6 (7.8)	0.04	3 (6.7)	0	3 (13.6)
Fluoroquinolones, iv/o	36 (11.5)	30 (12.7)	6 (7.8)	0.31	3 (6.7)	0	3 (13.6)
Glycopeptide, iv	23 (7.3)	19 (8.0)	4 (5.2)	0.61	4 (8.9)	0	0
Proton pump inhibitors, iv/o	163 (51.9)	131 (55.3)	32 (41.6)	0.049	18 (40.0)	3 (30.0)	11 (50.0)
H2-receptor antagonists, iv/o	67 (21.3)	60 (25.3)	7 (9.1)	0.002	3 (6.7)	0	4 (18.2)
Steroid, iv/o	52 (16.6)	32 (13.5)	20 (26.0)	0.01	13 (28.9)	2 (20.0)	5 (22.7)

[a] Include acute leukemia, chronic leukemia, and lymphoma.
[b] *P* values for comparing patients without malignancy and those HM.
Data are presented as patient numbers (%) or means ± standard deviations.
iv/o = intravenous/oral.

three study hospitals. VRE infection was defined as the isolation of VRE in initially sterile sites (such as blood or body fluid) and VRE colonization as the detection of VRE from a rectal swab or stool sample.[24] When the clinical significance of VRE isolation was not certain, such a clinical setting was referred as VRE colonization/infection. The date of the first events of VRE colonization/infection before or after the CDI episodes were recorded for the risk factor analyses.

Statistical analysis

Statistical analysis was performed using the statistical software, SPSS, version 13.0. Continuous data were expressed as the means ± standard deviations. The $\chi 2$ test or Fisher's test was used for categorical variables, and the Student *t*-test was used for continuous variables. A two-tailed *P* value of less than 0.05 was considered statistically significant. The parameters with *P* values less than 0.2 in the univariate analysis were entered into a multivariate analysis with a binary logistic regression model. Linear correlation was tested with Cochran-Armitage trend test.

Results

Clinical characters of included patients

During the study period, 528 patients were diagnosed as CDI, and 214 patients were excluded because of incomplete laboratory data (73 patients) and solid organ malignancy (141 patients) (Fig. 1). Finally 314 patients, including 77 with HM and 237 patients without malignancy as the control group, were included for analysis. Of those with HM, 45 patients had acute leukemia, 10 chronic leukemia, and 22 lymphoma.

Control patients were older (66.2 ± 20.9 years) than those with HM (49.5 ± 19.0 years, $P < .001$) (Table 1). Besides, the control group more often had underlying hypertension (50.2% *vs.* 7.8%, $P < .001$), diabetes mellitus (32.1%

Table 2 Clinical and microbiological characters in patients with hematological malignancy (HM) and without malignancy complicated with *Clostridioides difficile* infection (CDI).

Variables	Total, n = 314	No malignancy, n = 237	HM, n = 77	*P* value
Leukocyte count, cells/mL	11.0 ± 14.8	11.9 ± 9.4	8.4 ± 24.9	0.23
≤500	27 (8.6)	5 (2.1)	22 (28.6)	<0.001
500–1000	14 (4.5)	7 (3.0)	7 (9.1)	0.049
1000–5000	50 (15.9)	27 (11.4)	23 (29.9)	<0.001
5000–10,000	86 (27.4)	73 (30.8)	13 (16.9)	0.02
10,000–15,000	69 (22.0)	63 (26.6)	6 (7.8)	<0.001
>15,000	68 (21.7)	62 (26.2)	6 (7.8)	<0.001
Serum creatinine > 1.5 mg/L	23 (7.3)	18 (7.6)	5 (6.5)	1.00
Severe CDI by IDSA/SHEA criteria[a]	86 (27.4)	75 (31.6)	11 (14.3)	0.003
VRE colonization/infection	66 (21.1)	44 (18.6)	22 (28.6)	0.08
VRE before CDI	39 (12.4)	27 (11.4)	12 (15.6)	0.44
Duration from VRE to CDI, days	76.1 ± 103.9	72.7 ± 103.1	84.5 ± 110.3	0.76
VRE after CDI	27 (8.6)	17 (7.2)	10 (13.0)	0.17
Duration from CDI to VRE, days	56.7 ± 97.3	70.6 ± 118.7	33.0 ± 36.7	0.34
C. difficile isolates	n = 301	n = 227	n = 74	
tcdC deletion	27 (9.0)	19 (8.4)	8 (10.8)	0.49
Ribotype 017	15 (5.0)	13 (5.7)	2 (2.7)	0.37
Ribotype 078 complex	22 (7.3)	15 (6.6)	7 (9.5)	0.44
Metronidazole-resistant	1 (0.3)	1 (0.4)	0	1.00
Vancomycin-resistant	6 (2.0)	5 (2.2)	1 (1.4)	1.00

[a] Leukocyte count ≥ 15,000 cells/mL or serum creatinine > 1.5 mg/dL.
Data are presented as patient numbers (%) or means ± standard deviations.
VRE = vancomycin-resistant *Enterococcus*.

vs. 6.5%, $P < .001$), chronic kidney disease (25.3% *vs.* 2.6%, $P < .001$), prior stroke (15.6% *vs.* 1.3%, $P < .001$), coronary artery disease (10.1% *vs.* 0%, $P = .001$), congestive heart failure (12.2% *vs.* 1.3%, $P = .003$), or liver cirrhosis (7.6% *vs.* 1.3%, $P = .05$) than those with HM. Prior exposure to ceftazidime or ceftriaxone (23.6% *vs.* 10.4%, $P = .01$), carbapenem (17.3% *vs.* 7.8%, $P = .04$)), proton pump inhibitors (55.3% *vs.* 41.6%, $P = .049$), or H2-receptor antagonist (25.3% *vs.* 9.1%, $P = .002$) in control patients also were more common than in those with HM. In contrast, patients with HM more often had prior exposure to cefepime (35.1% *vs.* 10.1%, $P < .001$) or steroids (26.0% *vs.* 13.5%, $P = .01$).

Laboratory characters of CDI patients with hematologic malignancy

Regarding the distribution of blood leukocyte counts, more patients with HM were leukopenic, as evidenced by the higher percentages of individuals with a leukocyte count of <500 cells/mL (28.6% *vs.* 2.1%, $P < .001$), 500–1000 cells/mL (9.1% *vs.* 3.0%, $P = .049$), and 1000–5000 cells/mL (29.9% *vs.* 11.4%, $P < .001$). In contrast, control patients more often had leukocyte counts (5000–10,000 cells/mL: 30.8% *vs.* 16.9%, $P = .02$) or leukocytosis (10,000–15,000 cells/mL: 26.6% *vs.* 7.8%, $P < .001$; >15,000 cells/mL: 26.2% *vs.* 7.8%, $P < .001$) than those with HM (Table 2). At the onset of CDI, a similar percentage of renal dysfunction (serum creatinine > 1.5 mg/dL) was noted in those with HM and control patients (6.5% *vs.* 7.6%, $P = 1.00$). According to the IDSA/SHEA severity criteria,[4,20] only 11 (14.3%) of 77 patients with HM had severe CDI, but more control patients (31.6%, 75/237) had severe CDI ($P = .003$).

Microbiological characters of CDI patients

Of the 314 patients with CDI, 301 toxigenic *C. difficile* isolates were obtained from stool culture. Their toxin gene distribution was as follows: *tcdA* (−)/*tcdB* (+) in 18 isolates, and *tcdA* (+)/*tcdB* (+) in 283 isolates, which included 27 isolates with *tcdC* deletion and *cdtA* (+)/*cdtB* (+), and 3 isolates with *cdtA* (+)/*cdtB* (+) but without *tcdC* deletion. Of the 27 isolates with *tcdC* deletion, 3 had an 18-bp deletion (2 isolates were ribotype [RT] 027 and 1 RT 034), and 22 had a 39-bp deletion (1 isolate was RT 033, 2 RT 078, 11 RT 126, and 8 RT 127). Of the 18 isolates with *tcdA* (−)/*tcdB* (+), 15 were identified as RT 017. Since there were published data indicating the predominance of RT 078 ocmplex (including RT 033, 078, 126, and 127) and RT 017 among clinical *C. difficile* isolates in southern Taiwan (Hung et al. 2018), these ribotypes were specifically concerned in this study. The porportion of *tcdC* deletion, RT 017, or RT 078 complex among toxigenic *C. difficile* isolates were similar between two patient groups (Table 2).

Outcomes of CDI patients with hematologic malignancy

Initial antimicrobial therapy for CDI treatment, such as metronidazole or vancomycin, was similar between two

Table 3 Clinical and microbiological characters in 77 patients with hematologic malignancy developing *Clostridioides difficile* infection (CDI), stratified by crude in-hospital mortality.

Variables (case/isolate number)	Survive, n = 62	Expire, n = 15	*P* value	*P* value[a]
Age, years	48.6 ± 19.1	52.8 ± 19.0	0.46	
Leukocyte count, x 1000 cells/mL	7.1 ± 22.5	13.7 ± 33.8	0.48	
Gender, male	36 (58.1)	7 (46.7)	0.56	
Underlying disease				
Hypertension	6 (9.7)	0	0.59	
Diabetes mellitus	4 (6.5)	1 (6.7)	1.00	
Chronic kidney disease	2 (3.2)	0	1.00	
Serum creatinine > 1.5 mg/dL	5 (8.1)	0	0.58	
Severe CDI by IDSA/SHEA criteria	10 (16.1)	1 (6.7)	0.68	
VRE colonization/infection	13 (21.0)	9 (60.0)	0.008	0.01
VRE before CDI	8 (12.9)	4 (26.7)	0.29	
Duration from VRE to CDI, days	64.9 ± 78.8	119.0 ± 160.2	0.46	
VRE after CDI	6 (9.7)	4 (26.7)	0.14	
Duration from CDI to VRE, days	30.8 ± 21.2	36.3 ± 57.2	0.83	
Recent medication within one month before CDI onset				
Cytotoxic chemotherapy	42 (67.7)	5 (33.3)	0.02	0.25
Antimicrobial therapy				
Cephalosporins	26 (41.9)	5 (33.3)	0.77	
Penicillins	15 (24.2)	3 (20.0)	1.00	
Carbapenem, iv	4 (6.5)	2 (13.3)	0.33	
Fluoroquinolones, iv/o	5 (8.1)	1 (6.7)	1.00	
Glycopeptide, iv	3 (4.8)	1 (6.7)	1.00	
Proton pump inhibitors, iv/o	23 (37.1)	9 (60.0)	0.15	0.06
H2-receptor antagonists, iv/o	4 (6.5)	3 (20.0)	0.13	0.21
Steroid, iv/o	18 (29.0)	2 (13.3)	0.32	
Anti-diarrheal drugs	5 (8.1)	2 (13.3)	0.38	
C. difficile isolates	n = 59	n = 15		
tcdC deletion	5 (8.5)	3 (20.0)	0.35	
Ribotype 017	1 (1.7)	1 (6.7)	0.37	
Ribotype 078 complex	4 (6.8)	3 (20.0)	0.14	0.03
Vancomycin-resistant phenotype	1 (1.7)	0	1.00	

[a] Multivariate analysis.
Data are presented as patient numbers (%) or means ± standard deviations.
iv/o = intravenous/oral; VRE = vancomycin-resistant *Enterococcus*.

patient groups (Supplementary Table). The treatment duration for CDI was longer in those with HM than the controls (10.3 ± 6.8 days *vs*. 7.5 ± 7.3 days; $P = .008$). However, the interval between CDI treatment and diarreal resolution (7.6 ± 4.3 days *vs*. 7.6 ± 6.6 days; $P = .98$), antibiotic response rate (41.0% *vs*. 47.2%; $P = .66$), and crude in-hospital mortality rate were similar among those with HM and the controls (19.5% *vs*. 17.3%, $P = .73$). To study the prognostic impact of the disease severity, the in-hospital mortality rate of the two study groups was examined. Among those with HM, severe CDI did not herald a worse prognosis than non-severe CDAD (9.1% *vs*. 21.1%, $P = .68$) (Fig. 1). Of 237 control patients, severe CDI was associated with a higher in-hospital mortality rate than non-severe CDI (24.0% *vs*. 14.2%, $P = .07$), which had a borderline statistical significance, indicative of clinical utility of this severity category in those without malignancy, but not in those with HM. For those with HM, the in-hospital mortality was positively associated with VRE colonization/infection (odds ratio [OR] 7.72, 95% confident interval [CI] 1.50–39.68; $P = .01$), and *C. difficile* RT 078 complex infection (OR 9.22, 95% CI 1.24–68.87; $P = .03$), as shown in Table 3.

Risk factors for CDAD recurrence

Patients with HM had a higher recurrence rate of CDI (14.3%, 11/77 *vs*. 7.2%, 17/237; $P = .07$) and a longer hospital stay (47.2 ± 40.8 days *vs*. 33.3 ± 37.3 days; $P = .006$) than the controls (Table 4). To further analyze the risk factors associated with CDI recurrence in the univariate analyses, those with recurrent CDI more often had HM (39.3% *vs*. 23.1%; $P = .07$), congestive heart failure (21.4% *vs*. 8.4%; $P = .04$), VRE colonization/infection (42.9% *vs*. 18.9%; $P = .0006$), exposure to cefepime (25.0%

Table 4 Clinical and microbiological characters in patients with *Clostridioides difficile* infection (CDI), stratified by recurrence.

Variables (case/isolate number)	No recurrence n = 286	Recurrence n = 28	*P* value	*P* value[b]
Age, years	61.6 ± 21.9	66.8 ± 18.9	0.23	
Gender, male	156 (54.5)	15 (53.6)	1.00	
Underlying disease±				
Hypertension	116 (40.6)	9 (32.1)	0.43	
Diabetes mellitus	76 (26.6)	5 (17.9)	0.37	
Hematological malignancy[a]	66 (23.1)	11 (39.3)	0.07	0.04
Chronic kidney disease	54 (18.9)	8 (28.6)	0.22	
Old stroke	33 (11.5)	5 (17.9)	0.36	
Congestive heart failure	24 (8.4)	6 (21.4)	0.04	0.08
Coronary artery disease history	20 (7.0)	4 (14.3)	0.25	
Liver cirrhosis	17 (5.9)	2 (7.1)	0.68	
Leukocyte count, x 1000 cells/mL	10.5 ± 11.8	16.3 ± 32.3	0.35	
Serum creatinine > 1.5 mg/dL	21 (7.3)	2 (7.1)	1.00	
Severe CDI by IDSA/SHEA criteria	80 (28.0)	6 (21.4)	0.52	
VRE colonization/infection	54 (18.9)	12 (42.9)	0.006	0.02
VRE before CDI	31 (10.8)	8 (28.6)	0.01	
Duration from VRE to CDI, days	73.0 ± 98.8	87.8 ± 127.9	0.73	
VRE after CDI	22 (7.7)	5 (17.9)	0.07	
Duration from CDI to VRE, days	43.8 ± 68.8	73.0 ± 98.8	0.73	
Recent medication within one month before CDI onset				
Antimicrobial therapy				
Cephalosporins	110 (38.5)	13 (46.4)	0.42	
Cefazolin, iv	16 (5.6)	0	0.38	
Cefuroxime, iv/o	19 (6.6)	1 (3.6)	1.00	
Ceftazidime or ceftriaxone, iv	59 (20.6)	5 (17.9)	1.00	
Cefepime, iv	44 (15.4)	7 (25.0)	0.19	0.78
Penicillins	69 (24.1)	11 (39.3)	0.11	0.23
Carbapenem, iv	39 (13.6)	8 (28.6)	0.049	0.05
Fluoroquinolones, iv/o	33 (11.5)	3 (10.7)	1.00	
Glycopeptide, iv	19 (6.6)	4 (14.3)	1.36	
Proton pump inhibitors, iv/o	147 (51.4)	16 (57.1)	0.69	
H2-receptor antagonists, iv/o	62 (21.7)	5 (17.9)	0.81	
Steroid, iv/o	47 (16.4)	5 (17.9)	0.79	
C. difficile isolates	n = 275	n = 26		
tcdC deletion	25 (9.1)	2 (7.7)	1.00	
Ribotype 017	14 (5.1)	1 (3.8)	1.00	
Ribotype 078 complex	20 (7.0)	2 (7.7)	1.00	
Vancomycin-resistant phenotype	6 (2.1)	0	0.53	

[a] Include acute leukemia, chronic leukemia, and lymphoma.
[b] Multivariate analysis.
Data are presented as patient numbers (%) or means ± standard deviations.
iv/o = intravenous/oral.

vs. 15.4%, $P = .19$), penicillin (39.3% *vs.* 24.1%; $P = .11$), or carbapenem (28.6% *vs.* 13.6%, $P = .053$) before the recurrent event (Table 4). For a total of 66 patients with VRE colonization/infection, VRE colonization/infection events were found in 39 (59.1%) patients before the onset of CDI and 27 (40.9%) after the onset of CDI (Table 2). Of note, among 28 patients experiencing recurrent CDI there were 8 (28.6%) patients had VRE colonization/infection before the index CDI epsidoes. In contrast, only 10.8%[30] of 286 patients without recurrence CDI during the study period had prior VRE colonization/infection before the index CDI episodes ($P < .01$, Table 4). In the multivariate analyses, underlying hematological malignancy (OR 2.74, 95% CI 1.05–7.15; $P = .04$), and VRE colonization/infection (OR 2.71, 95% CI 1.16–6.38; $P = .02$) were independently associated with CDI recurrence.

Discussion

The in-hospital crude mortality rate of adults with HM complicating with CDI was 19.5% in our study, which was similar to that of adults without malignancy (16%), but was lower than that (25%) of cancer patients in the study

conducted by Larrainzar-Coghen et al.[15] This discrepancy might be due to younger age in our study than that in Larrainzar-Coghen's study (average age: 49 years *vs.* 64 years). Although a higher recurrence rate (14.3%) of CDI was noted in our patients with HM when the control population was the individuals without malignancy, such a recurrence rate remained far behind the reported data of 40% in both hematology-oncology population and solid organ transplant recipients in a recent review.[14] At least two risk factors for recurrent CDI, including salvage lymphoma chemotherapy and severe CDI, were identified at a medical center of U.S.[25]

It is not surprising that our cases of HM complicated with CDI were less likely to be classified as "severe" CDI, based on the leukocyte counts or elevated serum creatinine suggested by the IDSA/SHEA. Moreover, the crude mortality rate of our severe cases of CDI was not higher than that of those with non-severe CDI. Our finding that the infeasible use of leukocyte count as an indicator of CDI severity for such a specific population was compatible to several published studies.[16,26,27] However, the disease burden of CDI among HM patients is worthy to be reminded. In the United States Nationwide Inpatient Sample database, for leukemic patients the overall CDI incidence between 2005 and 2011 was 3.4% and even higher in those developing neutropenia.[26] Furthermore, longer duration of neutropenia in leukemic patients receiving chemotherapy would have a higher risk of CDI.[27] Thus the development for severity evaluation criteria of CDI for patients with HM or prolonged neutropenia is warranted.

In the present work, VRE colonization/infection was associated with CDI recurrence and heralded a worse outcome among those with HM, irrespective of their timing related to the index CDI events. The correlation between fecal VRE colonization with CDI recurrence among general population had been reported.[28] The association between VRE colonization and increased mortality had been noted among specific populations, including ICU patients,[29] allogeneic hematopoietic stem cell transplant recipients,[30] and acute myelogenous leukemia (AML).[31] Among patients with AML, VRE colonization led to an increased risk of VRE bloodstream infection (BSI), which was associated with increased cost and subsequent mortality.[31] The connection of gastrointestinal VRE colonization to subsequent "true" infection, such as VRE bacteremia, CDI, or CDI recurrence, might be associated with the scenario that VRE colonization signifies loss of colonization resistance within gastrointestinal microbiome, which facilitates the expansion of potential pathogens and increases the risk of subsequent invasive infection and mortality.[29,32]

CDI episodes due to *C. difficile* RT 078 complex were linked to a higher mortality among our patients with HM. *C. difficile* RT 078 complex had been regarded as zoonotic pathogens and had increasing antimicrobial-resistant capacity because of the use of antimicrobials outside the healthcare environment.[33] In our nationwide surveillance in Taiwan, *C. difficile* RT 078 complex was the dominant ribotype among the toxigenic *C. difficile* isolates with *tcdC* deletion.[34] Moreover, the prognostic impact of *C. difficile* clades/sequence types (STs) in England was most significant in clade 5, RT 078/ST 11 strains (the case fatality rate of 25%, 16 of 63), as compared to other clades.[35] In addition, the patients infected by RT 078 and RT 027 strains were reported to have similar rates of severe diarrhea (38.9% *vs.* 40.0%) or attributable mortality (3.8% *vs.* 4.0%).[36] A poor prognosis in the cases of *C. difficile* RT 078 infection was likely to be related to *tcdC* deletion and the presence of binary toxins,[35,36] and both virulent characters were noted in all our isolates of RT 078 complex.

There were some limitations inherent in our study. First, only a small number of patients with HM developing CDI, so the implication of leukocyte counts in the clinical implication of severity score of CDI warrants a large-scale study. Second, the study was conducted in Taiwan, so the application of these results to western countries needs further validation. Third, though leukocytosis was not a suitable parameter for the severity of CDI for patients with HM, we did not find other plausible variables for severity grading. Fourth, the dosage and regimen of cytotoxic chemotherapy and the duration of leukopenia were not analyzed in our study, which might affect the clinical outcome of CDI. Finally, physical conditions of the patients after stroke, including the functional status, use of nasogastric tube or urinary catheterization, were not assessed, and may be associated increased incidence of CDI.[37]

In conclusion, patients with HM complicated with CDI were often regarded as having non-severe infection, but had a similar in-hospital mortality rate as those without malignancy. Of note, the finding that CDI due to ribotype 078 complex isolates heralded a poor prognosis among HM patients suggests the virulence of these indigenous strains. Further elucidation of their potential sources is warranted.

Author contributions

YH, CC, PT, PH, LC, and WK designed the experiments, performed the experiments, analyzed the data, and participated in the writing of the manuscript. YH, CC, PT, YL, HT, HL, HL. JL, BT, PH, LC, and WK read and approved the final version of the manuscript.

Ethical approval

The study was approved by the Institutional Review Board in each hospital: Hospitals A (201412190RIND), B (CS14179), C (10,402—006), D and E (B-ER-103-098).

Declaration of competing interest

All authors report no conflicts of interest relevant to this article.

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Appendix

Supplement table Antimicrobial therapy and clinical outcomes of patients with hematological malignancy (HM) and without malignancy complicated with *Clostridioides difficile* infection (CDI).

Variables	Total n = 314	No malignancy n = 237	HM n = 77	*P* value
Receipt of antibiotics for CDI				0.06
No	92 (29.3)	76 (32.1)	16 (20.8)	
Yes	222 (70.7)	161 (67.9)	61 (79.2)	1.00
Metronidazole	203 (91.4)	147 (91.3)	56 (91.8)	
Vancomycin	19 (8.6)	14 (8.7)	5 (8.2)	
Antibiotic response rate	101/222 (45.5)	76/161 (47.2)	25/61 (41.0)	0.66
Time to diarrheal resolution, days	7.6 ± 6.0	7.6 ± 6.6	7.6 ± 4.3	0.98
Duration of CDI therapy, days	8.3 ± 7.3	7.5 ± 7.3	10.3 ± 6.8	0.008
Hospitalization duration, days	36.7 ± 38.6	33.3 ± 37.3	47.2 ± 40.8	0.006
Crude in-hospital mortality	56 (17.8)	41 (17.3)	15 (19.5)	0.73
Recurrence of CDI	28 (8.9)	17 (7.2)	11 (14.3)	0.07

Data are presented as patient numbers (%) or means ± standard deviations.
*Available case number.