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Eleven-Year Experience of Clostridial Bacteremia at a Tertiary Care Hospital in South Korea

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Background: Clostridial bacteremia (CB) is the second most frequent anaerobic bacteremia, and CB patients show high mortality without prompt antimicrobial therapy. We retrospectively reviewed 11 years of CB cases in a tertiary care hospital to describe the clinical and microbiological characteristics of CB and to define the risk factors of fatal CB.
Methods: All patients with CB from January 2002 to December 2012 were included in the study. Age, sex, underlying diseases, antibiotic use, and clinical outcome were reviewed. Antibiotic therapy was classified as either 'appropriate' or 'inappropriate' based on the activity against *Clostridium* species.
Results: A total of 118 *Clostridium* isolates (0.79% of all blood culture isolates) were recovered from the blood cultures of 114 patients. The underlying conditions of patients with CB were neoplasm in 87 cases (76.3%), gastrointestinal symptoms in 84 cases (73.7%), diabetes in 17 cases (14.9%), and hemodialysis in six cases (5.3%). Of the 118 *Clostridium* isolates, *C. perfringens* was the most frequent species (42 isolates, 35.6%). Thirty-two patients (28.1%) showed polymicrobial bacteremia, which was most commonly combined with *Escherichia coli*. Two patients harbored more than two *Clostridium* species. 'Appropriate' antibiotics were given to 97 (85.1%) patients. The mortality rate of CB at days 2, 8, and 30 was 7.9% (9/114), 14.0% (16/114), and 26.3% (30/114), respectively.
Conclusion: Neoplasm, especially in the gastrointestinal tract or of hematologic origin, and hemodialysis were considered to be risk factors of blood stream clostridial infection. Early appropriate antibiotic coverage of CB was not definitely associated with lower mortality in our study. **(Ann Clin Microbiol 2015;18:126-132)**

Key Words: Anaerobic bacteria, Bacteremia, *Clostridium*, Sepsis

INTRODUCTION

Anaerobic bacteremia is uncommon. The frequency of anaerobic bacterial isolation in hospitals accounts for 0.5-9% of all positive blood cultures [1]. *Clostridium* is a genus of anaerobic, Gram-positive, spore-forming bacteria and is a clinically important pathogen causing anaerobic infections in humans [2]. *Clostridium* species account for the second most frequent number of anaerobes isolated from blood, with *Clostridium perfringens* the most common species [2,3].

Among hospitalized patients, clostridial bacteremia (CB) represents less than 2% of positive blood cultures and is predominantly found as part of polymicrobial bacteremia cases [4]. CB patients show high mortality [5] and the disease is usually rapidly fatal without prompt and appropriate antimicrobial therapy [6,7]. However, treatment may not always be required when *Clostridium* species are isolated from blood cultures, as the detection of clostridial species may be transient or may even be due to specimen contamination [8]. In one study, the clinical relevance of *Clostridium* isolation from blood cultures was doubtful in one-third of the cases, but in two-thirds of the cases, clostridial isolation were considered to be of clinical significance [9].

CB has a primary association with intra-abdominal sepsis af-

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ter trauma or surgery, or it may occur in patients suffering from malignancies or diabetes mellitus [10]. Gas gangrene, botulism, tetanus, and fulminant intravascular hemolysis are the most notable histocytotoxic infections caused by *Clostridium* species; however, they could be the causative pathogen for a wide range of clinical diseases [11].

Herein, we retrospectively reviewed CB cases for 11 years in a tertiary care hospital. The aims of this study were to describe the characteristics of CB cases and define the risk factors of fatal CB cases relative to non-fatal CB cases.

MATERIALS AND METHODS

1. Study population and methods

All patients with positive blood culture results from January 2002 to December 2012 were reviewed. Results with *Clostridium* species isolates were culled from this data and patients with CB were included in the study. Clinical information for these patients was reviewed for age, sex, underlying diseases, antibiotic use, and clinical outcome. Non-CB cases during the study period were used as controls and characteristics of patients with CB were compared with the non-CB patients using the Pearson's Chi-square test. A P-value <0.05 was considered statistically significant. Statistical Package for the Social Sciences for Windows, version 21 (IBM Inc., Armonk, NY, USA) was used for statistical analysis. The Vitek system (bioMérieux, Inc., Durham, NC, USA) was used to identify *Clostridium* isolates from January 2002 to April 2009 along with the Vitek ANI card, and The Vitek 2 system (bioMérieux, Inc.) was used from May 2009 to December 2012 along with the Vitek 2 ANC card. This study was approved by the institutional review board of our institution (SMC 2015-10-001).

Antibiotic therapy was classified as either 'appropriate' or 'inappropriate' based on the activity against *Clostridium* species. 'Appropriate' therapy for CB was defined as when the antibiotic regimen included penicillin G, ampicillin-sulbactam, piperacillin-tazobactam, cefoxitin, clindamycin, imipenem-cilastatin, and metronidazole [12,13]. Carbapenems were considered as imipenem equivalent based on the data from Appendix D of the 2014 CLSI guideline M100-S24 [14]. Other antibiotic regimens were considered 'inappropriate' due to their lack of anti-clostridial activity.

2. Definitions

1) Community-acquired infection: Blood cultures were obtained within 48 h of admission, the patient had not been discharged from a hospital within the preceding week, and the bacteremia was not related to a procedure performed in the hospital [9].

2) Healthcare-associated infection: When the first sign of infection appeared >48 h following admission, or the infection was related to an in-hospital procedure even within the first 48 h of admission, or the patient had been hospitalized within the previous week [9].

3) Polymicrobial infection: One or more additional bacterial species isolated from blood concurrently with *Clostridium* species.

RESULTS

The 114 patients who had at least one positive blood culture episode for *Clostridium* species were reviewed. A total 118 *Clostridium* isolates were found from these patients. Identification of the same isolates from the same patient within one month was considered a single isolate. Of all positive blood cultures during the 11-years study period at our hospital, *Clostridium* species were found in 0.79% (118/14,905) of all blood culture isolates.

Of the 118 *Clostridium* isolates, *C. perfringens* was the most frequent species (42 isolates, 35.6%), followed by *C. clostridioforme* (20, 16.9%), *Clostridium* species-not-definitely identified (14, 11.9%), *C. bifermentans* (10, 8.5%), *C. histolyticum* (7, 5.9%), *C. difficile* (6, 5.1%), and *C. septicum* (5, 4.2%). The details are described in Table 1 and Fig. 1.

The mean age of the CB patients was 58 years (SD=15.2) and 78 patients (68.4%) were male. The underlying conditions of patients with CB included neoplasms in 87 cases (76.3%), gastrointestinal symptoms in 84 cases (73.7%), diabetes in 17 cases (14.9% in all patients, 15.5% in patients over 30 years), and hemodialysis with renal disease in six cases (5.3%). Of the 87 patients with neoplasms, 60 (69.0%) had gastrointestinal disease including colorectal cancer (19 cases, 21.8%), malignancy on liver (15 cases, 17.2%) and stomach cancer (13 cases, 14.9%), followed by 14 (16.1%) with hematologic malignancy, nine (10.3%) with genitourinary neoplasms, and four (4.6%) with lung cancer.

Among the 10,356 patients who had positive blood cultures during the 11-years study period at our hospital, CB was found in 0.54% (27/5,032) of patient without neoplasms and in 1.48% (87/5,324) of patient with neoplasms ($P<0.001$), respectively.

Table 1. The frequency of clinical condition and 2-, 8-, 30-day mortality in each *Clostridium* species

Type of *Clostridium* species (Total number)	DM	GI Symptom	HD	Neoplasms	Polymicrobial bacteremia	Healthcare-associated	Community-acquired	Mortality (2 day)	Mortality (8 day)	Mortality (30 day)
Clostridium perfringens (42)	3 (7.1%)	25 (59.5%)	1 (2.4%)	31 (73.8%)	16 (38.1%)	18 (42.9%)	24 (57.1%)	7 (16.7%)	9 (21.4%)	15 (35.7%)
Clostridium clostridioforme (20)	3 (15%)	16 (80%)	2 (10%)	16 (80%)	3 (15%)	7 (35%)	13 (65%)	0 (0%)	5 (25%)	7 (35%)
Clostridium species* (14)	4 (28.6%)	10 (71.4%)	0 (0%)	12 (85.7%)	4 (28.6%)	6 (42.9%)	8 (57.1%)	0 (0%)	0 (0%)	2 (14.3%)
Clostridium bifermentans (10)	1 (10%)	7 (70%)	0 (0%)	9 (90%)	3 (30%)	7 (70%)	3 (30%)	1 (10%)	1 (10%)	2 (20%)
Clostridium histolyticum (7)	0 (0%)	7 (100%)	0 (0%)	6 (85.7%)	1 (14.3%)	4 (57.1%)	3 (42.9%)	0 (0%)	0 (0%)	0 (0%)
Clostridium difficile (6)	1 (16.7%)	6 (100%)	1 (16.7%)	5 (83.3%)	3 (50%)	3 (50%)	3 (50%)	0 (0%)	0 (0%)	1 (16.7%)
Clostridium septicum (5)	1 (20%)	4 (80%)	0 (0%)	3 (60%)	1 (20%)	1 (20%)	4 (80%)	0 (0%)	0 (0%)	0 (0%)
Clostridium paraputrificum (3)	1	3	0	1	2	2	1	0	0	1
Clostridium baratii (2)	0	2	0	1	1	2	0	0	0	0
Clostridium butyricum (2)	1	2	1	0	0	0	2	0	0	0
Clostridium sporogenes (2)	2	2	1	1	0	2	0	0	0	0
Clostridium hastiforme (1)	0	1	0	1	0	1	0	1	1	1
Clostridium sordellii (1)	0	1	0	1	0	0	1	0	0	0
Clostridium subterminale (1)	0	1	0	1	0	1	0	0	0	0
Clostridium symbiosum (1)	0	1	0	1	0	0	1	0	0	1
Clostridium tertium (1)	0	0	0	1	1	1	0	0	0	0

Percentages were described in *Clostridium* species with more than 5 cases.
**Clostridium* species: *Clostridium* species that were not definitely identified to the species level.
Abbreviations: DM, diabetes mellitus; GI, gastrointestinal; HD, hemodialysis.

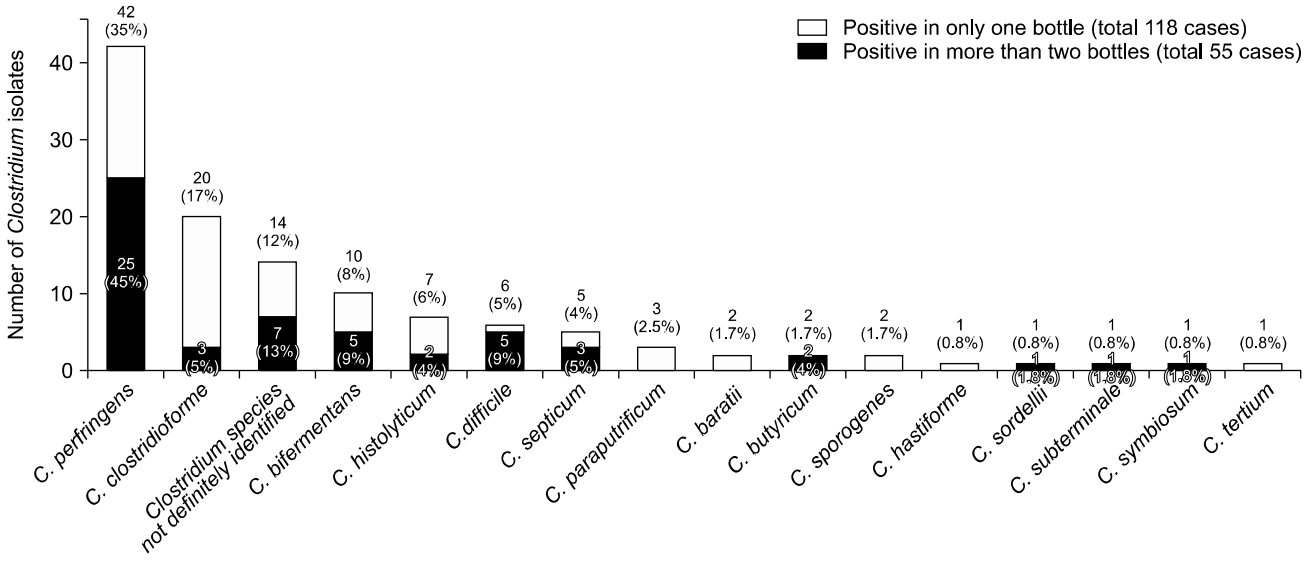

Fig. 1. The frequency of each *Clostridium* species in 118 clostridial isolates.

The proportion of the patients with neoplasms in CB (87/114, 76.3%) was significantly higher than those in non-CB (5,237/10,242, 51.1%, $P<0.01$) (Table 2).

Thirty-two patients had polymicrobial infection, with *Escherichia coli* as the most common co-isolated species. Two patients were concurrently infected with two different *Clostridium* species. Of these 32 patients with polymicrobial bacteremia, 24 were male (75.0%), 22 had underlying neoplasms (68.8%), 24 suffered from gastrointestinal symptoms (75.0%), underlying diabetes mellitus was found in five cases (15.6%), and one subject was on hemodialysis (3.1%). All these epidemiologic characteristics did not show significant differences from cases with monomicrobial bacteremia (*P*=0.35, 0.24, 0.84, 0.89, 0.52, respectively). The mean age of patients with polymicrobial infection was 57.5 years (SD=13.8), and did not show significant difference from patients with monomicrobial bacteremia, which had a mean age of 58.5 years (SD=15.8) (Student's T-test, *P*=0.76). The mortality rates of the patients with polymicrobial bacteremia at days 2, 8, and 30 were 6.3%, 9.4%, and 25.0%, respectively, and also did not show statistically significant differences from 82 subjects with monomicrobial bacteremia (8.5%, 15.9%, and 26.8%) (*P*=0.68, 0.37, and 0.84) (Table 3).

The mortality rates of CB at days 2, 8, and 30 were 7.9% (9/114), 14.0% (16/114), and 26.3% (30/114), respectively. In 87 patients with neoplasms, the mortality rates at days 2 and 8 were 9.2% and 16.1%, which seemed to be higher than the rate of 3.7% and 7.4% in 27 patients without neoplasms. However, probably due to low patient numbers, this did not show to be statistically significant (*P*=0.36 and 0.26, respectively). In 17 patients with diabetes mellitus, the mortality rates at days 2 and 8 were 11.8% and 17.6%, and the rates were 7.2% and 13.4% in 97 patients without DM (*P*=0.52 and 0.64, respectively). The mortality rates at days 2 and 8 were 8.3% and 14.3% in 84 patients with gastrointestinal symptoms, which showed little difference with the 30 patients without gastrointestinal symptoms (6.7% and 13.3%) (*P*=0.77 and 0.90, respectively).

'Appropriate' antibiotics were given to 97 patients (85.1%), but 'inappropriate' antibiotics were administered to 14 patients (12.3%), and no antibiotics were given to three patients (2.6%). Of the patients who received 'appropriate' antibiotics, the most commonly used antimicrobial agents were carbapenem with or without cilastatin, piperacillin-tazobactam, metronidazole, and clindamycin.

However, the mortality rates were not significantly different between patients receiving 'appropriate' antibiotic therapy and those receiving 'inappropriate' antibiotics for CB: 8.2% and 14.4% of 2-day and 8-day mortality rates in patients receiving

Table 2. The proportion of patients with neoplasms in CB and non-CB cases during the 11-years study period

Patient No.	CB	non-CB	Total
With neoplasms	87 (76.3%)	5,237 (51.1%)	5,324 (51.4%)
Without neoplasms	27 (23.7%)	5,005 (48.9%)	5,032 (45.6%)
Total	114	10,242	10,356

$P<0.01$, Chi-square test.
Abbreviation: CB, clostridial bacteremia.

Table 3. Comparison of epidemiologic and clinical characteristics of polymicrobial and monomicrobial infection in patients with CB

Patient group (n)	Mean Age (SD)	Male	Neoplasms	GI Symptoms	DM	HD	Mortality (2 day)	Mortality (8 day)	Mortality (30 day)
Polymicrobial bacteremia (32)	57.5 (13.8)	24 (75.0%)	22 (68.8%)	24 (75.0%)	5 (15.6%)	1 (3.1%)	2 (6.3%)	3 (9.4%)	8 (25.0%)
Monomicrobial bacteremia (82)	58.5 (15.9)	54 (65.9%)	65 (79.3%)	60 (73.2%)	12 (14.6%)	5 (6.1%)	7 (8.5%)	13 (15.9%)	22 (26.8%)
Total (114)	58.2 (15.2)	78 (68.4%)	87 (76.3%)	84 (73.7%)	17 (14.9%)	6 (5.3%)	9 (7.9%)	16 (14.0%)	30 (26.3%)

Abbreviations: CB, clostridial bacteremia; SD, standard deviation; GI, gastrointestinal; DM, diabetes mellitus; HD, hemodialysis.

'appropriate' therapy and 7.1% and 14.3% of 2-day and 8-day mortality rates in those receiving 'inappropriate' antibiotics (P= 0.89 and 0.99, respectively). In 87 patients with neoplasms, the mortality rates of 76 patients receiving 'appropriate' antibiotics were 9.2% at day 2 and 15.8% at day 8, while the mortality rates of 11 patients on 'inappropriate' antibiotics were 9.1% at day 2 and 18.2% at day 8 (P=0.99 and 0.84, respectively). Of the 27 patients without neoplasms, the mortality rate was 4.8% (one subject) at day 2 and 9.5% (two subjects, accumulated) at day 8 in 21 patients that received 'appropriate' antibiotics, while in the 'inappropriate' group, no subject expired at day 2 nor day 8 (P=0.70 and 0.58, respectively).

DISCUSSION

Clostridium species have been found occasionally in polymicrobial bacteremia. In this study, 28.1% (32/114) of CB patients had polymicrobial bacteremia. Other studies have reported variable rates of polymicrobial bacteremia in CB patients, ranging from 8% to 39% [6,7,12,15]. *Clostridium* species that were the most associated with polymicrobial bacteremia in ratio included *C. difficile* (50%, 3/6) as the major *Clostridium* species (recovered in more than five cases) followed by *C. perfringens* (38.1%, 16/42). Among the minor clostridial species (less than five cases), *C. tertium*, *C. paraputrificum*, and *C. baratii* seemed to be closely associated with polymicrobial bacteremia (Table 1).

In our study, the most common medical conditions in patients with CB were neoplasms (87/114, 76.3%), and the proportion of the patients with neoplasm in CB was significantly higher than those in non-CB (Table 2). The most common type of neoplasm was gastrointestinal malignancy (60/87, 69.0%), and the second most common type was hematologic malignancy (14/87, 16.1%). In previous reports, the most commonly related malignancies were gastrointestinal, genitourinary and hematologic malignancies [4,7,10,12,16]. Our results are consistent with previous reports, although the ethnic group is limited to the Korean population in our study. Other known underlying conditions included diabetes mellitus and hemodialysis [4,7,10,12,16]. As for DM, 15.5% (17/110, age over 30 years) of patients had diabetes as an underlying condition in our study and was comparable with 15.3%, the prevalence of diabetes mellitus in South Korea (age and sex corrected) based on the Korean National Health and Nutrition Survey in 2008-2010 (http://knhanes.cdc.go.kr/). Six patients (5.3%) were on hemodialysis, which seems to be higher than the estimated rate (0.64%, 309,000/48,000,000) in the general Korean population based on one report by the Korea Institute for Health and Social Affairs (http://repository.kihasa.re.kr:8080/handle/201002/1106); however, the rate of patients on hemodialysis in the general population is not well established in South Korea.

In our study, healthcare-associated CB accounts for 47% (55/118) of CB cases, which is higher than 31% that were reported in a previous report [9]. Healthcare-associated bacteremia was most frequently found with *C. bifermentans* (70%, 7/10). More than 50% was related with healthcare-associated bacteremia in *C. histolyticum* (57.1%, 4/7) and *C. difficile* (50%, 3/6) in major clostridial species. In minor species, *C. baratii*, *C. sporogenes*, *C. hastiforme*, *C. subterminale*, *C. tertium*, and *C. paraputrificum* seemed to be related to healthcare-associated infection (Table 1).

The mortality rate of CB at days 2, 8, and 30 was 7.9%, 14.0%, and 26.3%, respectively, which is lower than 32% at day 2 and 59% at day 28 that were reported in a previous report [12]. Although it is controversial whether early antibiotic therapy for CB could decrease the mortality rate or not [5,12,15,17], it seems that the difference of appropriate antibiotic coverage rate in early phase of bacteremia between the previous study (64%) [12] and our study (85.1%) might be related with the difference of the mortality rate. But, direct comparison is not justifiable due to difference in clinical settings. In our study, patients receiving 'appropriate' antibiotic therapy for CB did not show

lower 2-day and 8-day mortality rates compared to patients on 'inappropriate' antibiotics. Two patients who had no neoplasm and received 'appropriate' antibiotics expired within 8 days from CB. These patients suffered from extensive intestinal necrosis due to acute mesenteric infarction and severe heart failure due to acute myocardial infarction; the cause of death might not have been due to the antimicrobial therapy.

The method we used to identify *Clostridium* species has its limitations. Of the 40 isolates that were identified with the Vitek and the ANI card, 6 cases failed to identify to the species level. This is apparent in previous study [18], which evaluated this device with 44 known strains of *Clostridium*; nine failed to be identified to the species level, and five failed to be identified as genus *Clostridium*. And even in the 30 cases that were identified to the species level, two were identified as incorrect species. Among the 78 isolates identified with the Vitek 2 and the ANC card, eight cases could not be identified to the species level in our study. This is apparently improved over its predecessor. Another study suggests that Vitek 2 can be exact in identifying *Clostridium* accompanied with additional tests [19]. However, this device also has its limitation as a sole method; this study failed to identify known *Clostridium* strains to species level in 7 out of 27 cases with Vitek 2 alone. Additional testing (e.g., catalase, oxidase, and pigment, etc.) was required to identify to the species level.

This is the largest study of CB in Asia. We described the frequency of CB in blood culture at a large tertiary hospital in Korea. Neoplasms especially from the gastrointestinal tract or from hematologic origin and hemodialysis were considered to be risk factors of blood stream clostridial infection. Early appropriate antibiotic coverage of CB was not definitely associated with lower mortality in our study. The limitations of our study were that it was conducted in a single institution, and was done by retrospective methods. Further studies are warranted for evaluating the risks increasing invasive clostridial infection and factors associated with mortality.

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=국문초록=

11년간 단일 의료기관에서 발생한 *Clostridium* 균혈증 사례들에 대한 분석

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배경: *Clostridium*은 혐기성 균에 의한 균혈증 중에서 두번째로 많은 균이다. *Clostridium* 균혈증 환자는 신속한 항균치료를 받지 못할 경우 높은 사망률을 보인다고 알려져 있다. 본 연구의 저자들은 11년간 한 3차 병원에서 검출된 *Clostridium* 균혈증 사례들에 대해 그 임상적, 미생물학적 특성을 확인하고 환자가 사망에 이른 경우와 연관된 위험요인을 분석하였다.

방법: 2002년부터 2012년까지 11년간 혈액 배양 검사에서 *Clostridium* 균이 배양된 모든 증례의 의무기록과 검사 결과를 후향적으로 분석하였다. 환자의 나이, 성별, 기저질환, 항생제 사용 여부, 그리고 예후를 확인하였다. 투약된 항생제는 *Clostridium* 균에 대한 감수성에 따라 적절한 항생제와 불충분한 항생제로 분류하였다.

결과: 혈액 배양에서 *Clostridium* 균이 배양된 적이 있는 환자는 11년간 114명이었으며, 총 118건의 *Clostridium* 균주가 동정되었다. 114명 환자의 기저상태 중에서 종양이 76.3%로 가장 많은 비중을 차지하였으며, 위장관 증상은 73.7%, 당뇨는 14.9%, 혈액 투석이 5.3%였다. 118건의 균주 중 *C. perfringens*가 35.6%로 가장 많았다. 28.1%의 환자에서는 한 차례의 혈액 배양에서 두 종류 이상의 균주가 함께 동정되었다. 적절한 항생제가 사용된 환자는 85.1%였다. 균 검출 2일, 8일, 30일째의 환자 사망률은 각각 7.9%, 14.0%, 26.3%였다.

결론: 종양을 가진 환자나 혈액투석을 받고 있는 환자에서 *Clostridium* 균혈증의 발생 위험이 높다는 점이 확인되었다. 본 연구에서는 *Clostridium* 균혈증 환자에서 적절한 항생제의 조기 투여와 낮은 조기 사망률은 유의한 연관성을 보이지 않았다. [Ann Clin Microbiol 2015;18:126-132]

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