AAC Journals.ASM.org

Risk Factors for Resistance to β-Lactam/β-Lactamase Inhibitors and Ertapenem in *Bacteroides* Bacteremia

Janessa M. Smith,[a] Edina Avdic,[a] Pranita D. Tamma,[b] Long Zhang,[c] Karen C. Carroll,[d] Sara E. Cosgrove[e]

Department of Pharmacy, The Johns Hopkins Hospital, Baltimore, Maryland, USA[a]; Department of Pediatrics, Division of Pediatric Infectious Diseases, Johns Hopkins University School of Medicine, Baltimore, Maryland, USA[b]; Johns Hopkins Bloomberg School of Public Health, Department of Epidemiology, Baltimore, Maryland, USA[c]; Department of Pathology, Division of Medical Microbiology, The Johns Hopkins Hospital, Baltimore, Maryland, USA[d]; Department of Medicine, Division of Infectious Diseases, Johns Hopkins University School of Medicine, Baltimore, Maryland, USA[e]

The objective of this study was to determine risk factors for the development of resistance to β-lactams/β-lactamase inhibitors (βL/βLIs) and ertapenem among *Bacteroides* species bacteremia. We conducted a retrospective case-control study of 101 adult patients with *Bacteroides* species bacteremia at a 1,051-bed tertiary care medical center. The duration of exposure to βL/βLIs (odds ratio [OR], 1.25; 95% confidence interval [CI], 1.08 to 2.31) was the only independent risk factor for resistance.

The incidence of *Bacteroides* species bacteremia has increased over the past few decades (1–5). This increase is likely related to enhanced detection resulting from improved anaerobic culture methods and a more complex at-risk patient population (1). Anaerobic bacteremia has been associated with recent surgery, malignancy, and immunosuppression (1, 5–8).

There are 24 species of *Bacteroides* in the *Bacteroides fragilis* group, and they vary considerably in their resistance patterns although rates of resistance to β-lactams have increased among most species (2–4, 9, 10). *B. fragilis* remains the most susceptible while *Bacteroides distasonis* and *Bacteroides thetaiotaomicron* have demonstrated higher rates of resistance to β-lactams (9).

The primary objective of this study was to determine the prevalence of and risk factors for *Bacteroides* species with reduced susceptibility to β-lactams/β-lactamase inhibitors (βL/βLIs) and ertapenem in patients with bacteremia.

We performed a retrospective case-control study at a 1,051-bed tertiary care medical center in Baltimore, Maryland. All adult patients hospitalized between 1 January 2007 and 31 August 2013 with blood cultures growing *Bacteroides* species with β-lactam susceptibility testing available were eligible for inclusion. Gram-negative organisms believed to be anaerobes in positive blood culture bottles were subcultured to CDC anaerobic blood agar, laked kanamycin-vancomycin (LKV) agar, and *Bacteroides* bile esculin (BBE) agar and incubated under anaerobic conditions in an anaerobe chamber. Prior to 2012, organisms that grew on these media were further identified as *Bacteroides* species by the RapID ANA II System (Thermo Fisher Scientific, Waltham, MA, USA). Starting in 2012, organisms were identified via matrix-assisted laser desorption ionization–time of flight mass spectrometry (MALDI-TOF MS) using a Bruker microflex instrument, Biotyper software v3.0, and database v3.1.66 (Bruker Daltonics, Billerica, MA).

Antibiotic susceptibility testing was performed via Etest strips (bioMérieux). For the purpose of this study, all isolates with MICs interpreted as intermediate or resistant to amoxicillin-clavulanate, piperacillin-tazobactam, and/or ertapenem according to Clinical and Laboratory Standards Institute (CLSI) recommendations, were classified as resistant (11). For a majority of isolates, the MIC was not available, so isolates were evaluated based on their susceptibility to the respective antibiotic. Ertapenem is the only carbapenem routinely tested against anaerobes at our institution.

Patients were identified using the TheraDoc clinical surveillance software system. Cases were defined as patients with blood isolates of *Bacteroides* species resistant to βL/βLIs and/or ertapenem. Patients who had positive blood cultures for *Bacteroides* species but did not have susceptibility testing performed were excluded. Three controls were matched to each case patient by year of positive culture. A random number generator was used to select controls.

Baseline characteristics of cases and controls were compared using chi-square testing and Fisher's exact test for categorical variables, as appropriate, and the Wilcoxon rank sum test for continuous variables. Variables with a P value of ≤ 0.20 were entered into a multivariable logistic regression model and automatically selected using a backwards stepwise approach. Data were analyzed using Stata statistical software v12.0 (Stata Corp LP, TX). This study was approved by the Johns Hopkins University School of Medicine Institutional Review Board with a waiver of informed consent.

There were 159 patients with *Bacteroides* bacteremia identified during the study period. The identified isolates and their corresponding resistance rates are listed in Table 1. Of these, 26 (16.0%) patients had resistant *Bacteroides*. Amoxicillin-clavulanate was the most common agent to which isolates were resistant (11.5%), followed by ertapenem (7.0%) and piperacillin-tazobactam (6.8%).

Received 8 January 2015 **Returned for modification** 2 February 2015
Accepted 14 May 2015

Accepted manuscript posted online 1 June 2015

Citation Smith JM, Avdic E, Tamma PD, Zhang L, Carroll KC, Cosgrove SE. 2015. Risk factors for resistance to β-lactam/β-lactamase inhibitors and ertapenem in *Bacteroides* bacteremia. Antimicrob Agents Chemother 59:5049–5051. doi:10.1128/AAC.00046-15.

Address correspondence to Janessa M. Smith, jsmit276@jhmi.edu.

E.A., P.D.T., L.Z., K.C.C., and S.E.C. contributed equally to this article.

doi:10.1128/AAC.00046-15

TABLE 1 Resistance by species

Bacteroides species[a]	No. of isolates	No. (%) resistant to amoxicillin-clavulanate	No. (%) resistant to piperacillin-tazobactam	No. (%) resistant to ertapenem
B. caccae	12	3 (25.0)	0	0
B. distasonis	6	1 (16.0)	0	1 (16.0)
B. eggerthii	4	0	0	0
B. fragilis	71	8 (11.3)	2 (2.8)	5 (7.0)
B. loescheii	1	0	0	0
B. melaninogenicus	1	0	0	0
B. stercoris	1	0	0	0
B. thetaiotaomicron	43	3 (7.0)	6 (14.0)	2 (4.7)
B. uniformis	15	3 (20.0)	0	0
B. ureolyticus	2	0	0	0
B. vulgatus	3	0	0	0

[a] A total of 159 *Bacteroides* bacteremia isolates were identified during the study period.

There were five isolates (19%) that were resistant to ertapenem but not βL/βLIs.

There were 101 patients included in the case-control analysis (26 case patients and 75 control patients). Only one suitable control patient was found for one of the case patients. The characteristics of these patients are summarized in Table 2. Of note, 23 (88.5%) cases and 48 (64%) controls were considered health care-associated infections (defined as positive blood culture >48 h after hospitalization or surgical procedure or hospitalization within 30 days of a positive culture). Factors associated with resistance on univariable analysis included time at risk (odds ratio [OR], 1.07; 95% confidence interval [CI], 1.02 to 1.12), health care-associated infection (OR, 4.3; 95% CI, 1.18 to 15.7), and duration of exposure to βL/βLIs (OR, 1.5; 95% CI, 0.93 to 2.45). On multivariable analysis, the only independent predictor of resistance was duration of therapy with βL/βLIs prior to infection (OR, 1.25; 95% CI, 1.08 to 2.31) and after being adjusted for time at risk.

A higher proportion of case patients died in the hospital (n = 10 [38.5%]) than control patients (n = 4 [5.3%]; $P < 0.001$). There was a higher percentage of concurrent Gram-negative bacteremia in case patients (n = 4 [15.3%]) than in control patients

TABLE 2 Risk factors for resistance to *Bacteroides* species

Characteristic[b]	Cases (n = 26)[a]	Controls (n = 75)[a]	OR (95% CI)	P value
Age (yr)	60 (50–67)	61 (47–71)	1.00 (0.98–1.03)	0.79
Pitt score	2 (0–4)	1 (0–2)	1.13 (0.95–1.34)	0.16
Health care-associated infections	23 (88.5)	46 (60.5)	4.21 (1.18–15.7)	0.03
ICU admissions at time of positive culture	13 (50)	22 (29.33)	2.41 (0.96–6.02)	0.06
Source of bacteremia				
Abdominal surgery	17 (65.38)	35 (47.30)	2.05 (0.81–5.19)	
Perforation	3 (11.54)	5 (6.67)	1.83 (0.40–8.24)	
Biliary procedure	0 (0)	2 (2.67)	1.17 (0–15.29)	
SSTI	1 (3.85)	6 (8.00)	0.46 (0.05–4.01)	
Concurrent Gram-negative bacteremia	4 (15.38)	4 (5.33)	2.87 (0.89–8.42)	0.12
Source controls[c]	2 (7.69)	6 (8.00)	0.95 (0.88–1.08)	1.0
Comorbidity				
Diabetes	5 (19.23)	12 (16.22)	1.23 (0.39–3.90)	
GI disease[d]	2 (7.69)	7 (9.33)	0.81 (0.16–4.17)	
Malignancy	9 (34.62)	35 (47.30)	0.57 (0.23–1.46)	
Solid organ transplant	2 (7.69)	1 (1.33)	6.17 (0.54–71.05)	
Liver disease	4 (15.38)	4 (5.56)	3.09 (0.71–13.40)	
Time at risk[e] (days)	5.83 (3.04–16.92)	3.07 (0.80–6.61)	1.07 (1.02–1.12)	0.005
Duration of prior exposure to β-lactam (days)[f]				
βL/βLI exposure	13 (8–15)	5 (2–9)	1.5 (0.93–2.45)	0.008
Ertapenem exposure	14 (2–19)	10 (7–27)	0.95 (0.81–1.12)	0.044

[a] Values shown are median (IQR) or number (%).
[b] βL/βLI, β-lactam/β-lactamase inhibitor; GI, gastrointestinal; ICU, intensive care unit; SSTI, skin and soft tissue infection.
[c] Defined as no evidence of ongoing contamination and/or an undrained collection of infection.
[d] Includes ulcerative colitis, Crohn's disease, diverticulosis.
[e] The time (days) from admission to positive culture.
[f] Within 90 days before positive culture.

($n = 4$ [5.3%]), but this difference was not statistically significant ($P = 0.12$).

Since we started routine antibiotic susceptibility testing in 2010, we have not documented metronidazole resistance among *Bacteroides* species isolates and therefore limited our evaluation to β-lactams. Resistance rates in this study were similar to what has previously been reported for amoxicillin-clavulanate but were higher for ertapenem and piperacillin-tazobactam (4, 9). Karlowsky et al. reported that 2.3%, 0.5%, and 12.7% of *B. fragilis* species were intermediate or resistant to ertapenem, piperacillin-tazobactam, and amoxicillin-clavulanate, respectively (4). In a report by Snydman et al., the proportions of resistant *B. fragilis* and *B. thetaiotaomicron* isolates from 2008 to 2009 to βL/βLIs and ertapenem were <5% (9). Resistance rates for *Bacteroides* spp. may vary among other regions, and higher resistance rates have been reported outside North America (12).

The duration of exposure to βL/βLIs was an important risk factor for resistance in our study. Each additional day of βL/βLI therapy was associated with a 25% increased risk of developing a resistant *Bacteroides* isolate. Similarly, Nguyen et al. found a significant association between *in vitro* resistance to β-lactams and previous exposure to β-lactam antianaerobic agents (e.g., ticarcillin-clavulanate, piperacillin, cefotetan, ampicillin-sulbactam) within the previous 14 days (6). We did not find exposure to metronidazole to be protective against β-lactam resistance, as more case patients were exposed to metronidazole than were control patients (19.2% versus 6.7%, respectively).

We found a higher mortality rate in patients with resistant *Bacteroides* species. This result was likely confounded by more severe underlying disease with a higher percentage of patients with concurrent Gram-negative bacteremia, admissions to the ICU, and longer hospital stays prior to the first positive culture in case patients. In our study, 90% of patients who died received initial appropriate therapy. Only one patient received combination antianaerobic therapy with a βL/βLI and metronidazole. Of the 10 patients with resistant *Bacteroides* spp. that died, 4 (40%) were treated with βL/βLIs and 4 (40%) were treated with metronidazole. The remaining 2 patients received ertapenem. Because of these small numbers, we are unable to draw any meaningful conclusions about the role of empirical combination antianaerobic therapy.

Resistance patterns vary among regions and institutions; therefore, the single center design limits the generalizability of our findings. We were limited by a small sample size, so we grouped together the βL/βLI agents for analysis. Grouping these agents together makes it difficult to determine if different risk factors for resistance to amoxicillin-clavulanate and piperacillin-tazobactam exist.

Duration of exposure to βL/βLIs is a significant risk factor for the development of resistance to these classes of agents. It is important to use these agents judiciously in an effort to decrease the development of resistance to anaerobic organisms. Routine antimicrobial susceptibility testing and the development of an antibiogram for *Bacteroides* species can ensure that appropriate empirical therapy is selected.

REFERENCES

1. **Lassmann B, Gustafson DR, Wood CM, Rosenblatt JE.** 2007. Reemergence of anaerobic bacteremia. Clin Infect Dis **44:**895–900. http://dx.doi.org/10.1086/512197.
2. **Snydman DR, McDermott L, Cuchural GJ, Hecht DW, Iannini PB, Harrell LJ, Jenkins SG, O'Keefe JP, Pierson CL, Rihs JD, Yu VL, Finegold SM, Gorbach SL.** 1996. Analysis of trends in antimicrobial resistance patterns among clinical isolates of *Bacteroides fragilis* group species from 1990 to 1994. Clin Infect Dis **23**(Suppl)**:**S54–S65. http://dx.doi.org/10.1093/clinids/23.Supplement_1.S54.
3. **Snydman DR, Jacobus NV, McDermott LA, Ruthazer R, Goldstein EJC, Finegold SM, Harrell LJ, Hecht DW, Jenkins SG, Pierson C, Venezia R, Rihs J, Gorbach SL.** 2002. National survey on the susceptibility of *Bacteroides fragilis* group: report and analysis of trends for 1997-2000. Clin Infect Dis **35**(Suppl)**:**S126–S134. http://dx.doi.org/10.1086/341934.
4. **Karlowsky JA, Walkty AJ, Adam HJ, Baxter MR, Hoban DJ, Zhanel GG.** 2012. Prevalence of antimicrobial resistance among clinical isolates of *Bacteroides fragilis* group in Canada in 2010-2011: CANWARD Surveillance Study. Antimicrob Agents Chemother **56:**1247–1252. http://dx.doi.org/10.1128/AAC.05823-11.
5. **Salonen JH, Eerola E, Meurman O.** 1998. Clinical significance and outcome of anaerobic bacteremia. Clin Infect Dis **26:**1413–1417. http://dx.doi.org/10.1086/516355.
6. **Nguyen MH, Yu VL, Morris AJ, McDermott L, Wagener MW, Harrell L, Snydman DR.** 2000. Antimicrobial resistance and clinical outcome of *Bacteroides* bacteremia: findings of a multicenter prospective observational trial. Clin Infect Dis **30:**870–876. http://dx.doi.org/10.1086/313805.
7. **Wilson JR, Limaye AP.** 2004. Risk factors for mortality in patients with anaerobic bacteremia. Eur J Clin Microbiol Infect Dis **23:**310–316. http://dx.doi.org/10.1007/s10096-004-1111-y.
8. **Chen YM, Lee HC, Chang CM, Chuang YC, Ko WC.** 2001. *Clostridium* bacteremia: emphasis on the poor prognosis in cirrhotic patients. J Microbiol Immunol Infect **34:**113–118.
9. **Snydman DR, Jacobus NV, McDermott LA, Golan Y, Goldstein EJ, Harrell L, Jenkins S, Newton D, Pierson C, Rosenblatt J, Venezia R, Gorbach SL, Queenan AM, Hecht DW.** 2011. Update on resistance of *Bacteroides fragilis* group and related species with special attention to ertapenem 2006-2009. Anaerobe **17:**147–151. http://dx.doi.org/10.1016/j.anaerobe.2011.05.014.
10. **Hecht DW.** 2004. Prevalence of antibiotic resistance in anaerobic bacteremia: worrisome developments. Clin Infect Dis **39:**92–97. http://dx.doi.org/10.1086/421558.
11. **Clinical and Laboratory Standards Institute.** 2012. Performance standards for antimicrobial susceptibility testing; 22nd informational supplement. CLSI M100-S22. Clinical and Laboratory Standards Institute, Wayne, PA.
12. **Lie C-Y, Huang Y-T, Liao C-H, Yen L-C, Lin H-Y, Hsueh P-R.** 2008. Increasing trends in antimicrobial resistance among clinically important anaerobes and *Bacteroides fragilis* isolates causing nosocomial infections: emerging resistance to carbapenems. Antimicrob Agents Chemother **52:**3161–3168. http://dx.doi.org/10.1128/AAC.00355-08.