AAC Journals.ASM.org

First National Survey of Antibiotic Susceptibility of the *Bacteroides fragilis* Group: Emerging Resistance to Carbapenems in Argentina

Liliana Fernández-Canigia,[a] Mirta Litterio,[a] María C. Legaria,[a] Liliana Castello,[a] Silvia C. Predari,[a] Ana Di Martino,[a] Adelaida Rossetti,[a] Raquel Rollet,[a] Graciela Carloni,[a] Hebe Bianchini,[a] Daniela Cejas,[b] Marcela Radice,[b] Gabriel Gutkind,[b] and the Anaerobe Surveillance Team

Subcomisión de Bacterias Anaerobias, SADEBAC, Asociación Argentina de Microbiología, Ciudad Autónoma de Buenos Aires, Buenos Aires, Argentina,[a] and Facultad de Farmacia y Bioquímica, Universidad de Buenos Aires, Ciudad Autónoma de Buenos Aires, Buenos Aires, Argentina[b]

The antibiotic susceptibility rates of 363 clinical *Bacteroides fragilis* group isolates collected from 17 centers in Argentina during the period from 2006 to 2009 were as follows: piperacillin-tazobactam, 99%; ampicillin-sulbactam, 92%; cefoxitin, 72%; tigecycline, 100%; moxifloxacin, 91%; and clindamycin, 52%. No metronidazole resistance was detected in these isolates during this time period. Resistance to imipenem, doripenem, and ertapenem was observed in 1.1%, 1.6%, and 2.3% of *B. fragilis* group strains, respectively. *B. fragilis* species showed a resistance profile of 1.5% to imipenem, 1.9% to doripenem, and 2.4% to ertapenem. This is the first report of carbapenem resistance in Argentina. The *cfiA* gene was present in 8 out of 23 isolates, all of them belonging to the *B. fragilis* species and displaying reduced susceptibility or resistance to carbapenems (MICs ≥ 4 μg/ml). Three out of eight *cfiA*-positive isolates were fully resistant to carbapenems, while 5 out of 8 isolates showed low-level resistance (MICs, 4 to 8 μg/ml). The inhibition by EDTA was a good predictor of the presence of metallo-β-lactamases in the fully resistant *B. fragilis* strains, but discrepant results were observed for low-level resistant isolates. *B. fragilis* was more susceptible to antimicrobial agents than other *Bacteroides* species. *Bacteroides vulgatus* species was the most resistant to ampicillin-sulbactam and piperacillin-tazobactam, and *B. thetaiotaomicron/ovatus* strains showed the highest level of resistance to carbapenems, with an unknown resistance mechanism. *B. vulgatus* and the uncommon non-*Bacteroides fragilis* species were the most resistant to moxifloxacin, showing an overall resistance rate of 15.1%.

The increasing resistance to antimicrobial agents among anaerobic bacteria has become a global problem in the past 2 decades, particularly within the species that make up the *Bacteroides fragilis* group, the most frequently isolated species in clinical infections, in which resistance to metronidazole, carbapenems, and β-lactam–β-lactamase inhibitor combinations has been reported (14–16, 29).

Antimicrobial susceptibility testing has been recommended only in particular clinical situations and microorganisms (6). Antimicrobial resistance rates vary in different countries and also among the different medical centers within the same country (2, 15, 20). Therefore, both periodic local and national susceptibility studies and the evaluation of new therapeutic agents are necessary to provide data for an appropriate empirical antimicrobial therapy (18).

In Argentina, the Subcomisión de Bacterias Anaerobias de la Asociación Argentina de Microbiología (SADEBAC-AAM) conducts regular monitoring and surveillance of these bacteria. The results of the first national survey of antibiotic susceptibility in the *B. fragilis* group are reported here.

(This study was presented in part at the 50th Interscience Conference on Antimicrobial Agents and Chemotherapy in Boston, MA, September 2010.)

MATERIALS AND METHODS

Bacterial isolates. Between 2006 and 2009, a total of 363 nonduplicate clinical isolates belonging to the *B. fragilis* group were collected from 17 centers in Argentina. The isolates collected from centers in Ciudad Autónoma de Buenos Aires, Argentina, were from the Hospital General de Agudos Dr. E. Tornú (77 isolates), Hospital Nacional de Pediatría Prof. Dr. J. P. Garrahan (70 isolates), Hospital Alemán (69 isolates), Instituto de Investigaciones Médicas Alfredo Lanari-UBA (22 isolates), Hospital de Infecciosas F. J. Muñiz (22 isolates), Sanatorio Mitre (7 isolates), Hospital Dr. P. Piñero (5 isolates), and Sanatorio Mater Dei (2 isolates). In Córdoba, the isolates were from Clínica Reina Fabiola (25 isolates), Hospital Nacional de Clínicas (14 isolates), and Clínica Privada de Río Cuarto (2 isolates). Twenty-one isolates were from the Hospital Dr. J. C. Perrando, Resistencia, in Chaco. Ten isolates were from the Hospital Provincial de Neuquén in Neuquén (10 isolates), and nine isolates were from the HIGA Dr. A Piñero, Junin, in Buenos Aires Province. Two isolates were from the Hospital Eva Perón in San Martín, five isolates were from the Clínica Regional Privada, San Genaro, in Santa Fe, and one isolate was from the Hospital Central Mendoza in Mendoza.

The isolates were recovered from the abdomen (58%), genital tract (13%), blood (12%), skin and soft tissues (8.5%), as well as from other body sites (8%).

The species distribution of the isolates was as follows. A total of 198 isolates were *Bacteroides fragilis*, 69 were *Bacteroides thetaiotaomicron/ovatus*, 30 were *Bacteroides caccae*, and 27 were *Bacteroides vulgatus*. Thirty-nine isolates were less frequently isolated species. Of the 39 isolates, 7 were *Parabacteroides* (*Bacteroides*) *distasonis*, 7 were *Bacteroides*

Received 2 September 2011 **Returned for modification** 2 October 2011
Accepted 23 December 2011
Published ahead of print 9 January 2012
Address correspondence to Liliana Fernández-Canigia, lfcanigia@labdl.com.ar.

doi:10.1128/AAC.05622-11

uniformis, 7 were *Bacteroides stercoris*, and 2 were *Bacteroides merdae*, and 16 isolates of the *Bacteroides fragilis* group could not be fully identified to the species level (*Bacteroides* spp.). The isolates were stored at −70°C in glycerol broth and subcultured onto sheep blood brucella agar for further testing.

Antimicrobial agents. Standard powders of ampicillin, sulbactam, piperacillin, tazobactam, cefoxitin, imipenem, clindamycin, metronidazole, and moxifloxacin antibiotics were kindly supplied by INEI-ANLIS Dr. Carlos G. Malbrán, Argentina. Ertapenem was provided by Merck & Co, West Point, PA. Doripenem was provided by Janssen-Cilag, Argentina, and tigecycline was provided by Wyeth Pharmaceutical, Argentina.

Antimicrobial susceptibility testing. MICs were determined according to the reference agar dilution method of the Clinical and Laboratory Standards Institute (CLSI Document M11-A7) (6) with brucella agar supplemented with 5 μg/ml hemin, 1 μg/ml vitamin K, and 5% laked sheep blood. Agar dilution test plates were inoculated with approximately 10^5 CFU/spot using a Steers multipoint replicator and incubated at 37°C for 48 h in an anaerobic chamber.

The MIC was defined as the lowest concentration of an antimicrobial agent that will markedly reduce growth occurring on the test plate compared with the anaerobic control plate. Reference strains *B. fragilis* ATCC 25285 and *B. thetaiotaomicron* ATCC 29741 were used as controls. Results were recorded only when the MICs corresponding to the control organisms were within the range specified by the CLSI. The breakpoints recommended by the CLSI in CLSI Document M11-A7 (6) were used for most antibiotics. The breakpoints established by the CLSI for carbapenems (imipenem and ertapenem) were used for doripenem, and the breakpoints recommended by the Food and Drug Administration (FDA) were used for tigecycline, as there are no published susceptibility criteria recommendations for this drug (Tygacil package insert; Wyeth Pharmaceuticals). In this study, those isolates displaying carbapenem MICs of 4 μg/ml (susceptible) or 8 μg/ml (intermediate) were considered to have decreased susceptibility.

Phenotypic screening for MBLs. Inhibition of metallo-β-lactamases (MBLs) by EDTA was evaluated in those isolates that displayed decreased susceptibility or resistance to carbapenems. A reduction of at least 3 dilutions of the MIC of imipenem in the presence of 0.4 mM EDTA with respect to the MIC of imipenem alone was considered positive.

Detection of the *cfiA* gene. PCR amplification for detection of the *cfiA* gene was performed in those isolates that displayed resistance or decreased susceptibility to carbapenems, using the following primers (5′-3′) described by Kato et al. (13): GBI-1 (CCCAACTCTCGGACAAAAGTG) and GBI-2 (AGTGAATCGGTGAATCCATG). Total DNA was obtained by the boiling method. PCR amplification was run for 30 cycles, with 1 cycle consisting of 1 min at 95°C and 1 min at 57°C. Twenty-two isolates with carbapenem MICs of ≤2 μg/ml were also included.

RESULTS

Antimicrobial susceptibilities of all isolates expressed as MIC range, MIC_{50}, MIC_{90}, and percentage of susceptibility are summarized in Table 1.

The overall susceptibility rates to β-lactams were as follows: imipenem, 99%; piperacillin-tazobactam, 99%; doripenem, 98%; ertapenem, 96%; ampicillin-sulbactam 92%; and cefoxitin, 72%. The susceptibility profiles for the other antibiotics were as follows: metronidazole, 100%; tigecycline, 100%; moxifloxacin, 91%; and clindamycin, 52%.

B. fragilis was the most susceptible species with respect to the majority of the agents tested and showed the highest rates of susceptibility to ampicillin-sulbactam, cefoxitin, and clindamycin. *B. vulgatus* remained susceptible to carbapenems, despite showing the lowest rates of susceptibility to β-lactam–β-lactamase inhibitor combinations.

Emergence of resistance to carbapenems and piperacillin-tazobactam was observed in this study. For *B. fragilis* isolates, nonsusceptible rates (including resistant and intermediate strains) to imipenem, doripenem, and ertapenem were 1.5%, 2.5% and 4%, respectively, and 0.6%, 2%, and 4.8% for other species of the *B. fragilis* group, with *B. thetaiotaomicron/ovatus* being the most resistant.

Among the resistant isolates, three *B. fragilis* isolates and one *B. thetaiotaomicron/ovatus* isolate recovered from blood, bone, pericardial fluid, and abdominal fluid sources showed carbapenem MICs of ≥16 μg/ml.

Correspondence of the phenotypic screening of the MBLs and genotypic detection of the *cfiA* gene for all the isolates with decreased susceptibility or resistance to carbapenems are shown in Table 2. The *cfiA* gene was present in 8 out of 23 isolates, all corresponding to the *B. fragilis* species. Three out of eight *cfiA*-positive isolates were resistant to carbapenems and showed inhibition by EDTA. The other five isolates showed decreased susceptibility to ertapenem and doripenem, and only three strains scored positive in the phenotypic screening. The *cfiA* gene was not detected in any other species of the group, not even in carbapenem-resistant *B. thetaiotaomicron*/*ovatus*. Moreover, the *cfiA* gene was not detected in the 22 isolates with carbapenem MICs of ≤2 μg/ml.

The overall susceptibility rate to moxifloxacin was 91%. Nevertheless, isolates from the uncommon species [*Parabacteroides* (*Bacteroides*) *distasonis*, *Bacteroides uniformis*, *Bacteroides stercoris*, *Bacteroides merdae*, and *Bacteroides* spp.] and *B. vulgatus* showed the highest moxifloxacin resistance rates, up to 15.4% and 14.8%, respectively, doubling the rates observed in the other species.

No metronidazole- or tigecycline-resistant isolates were observed. However, tigecycline MIC_{90} values among the different species ranged between 0.125 and 1 μg/ml, below the FDA breakpoint. Clindamycin showed the lowest activity among all the antibiotics tested, ranging from 42% for *B. thetaiotaomicron/ovatus* to 74.7% for *B. fragilis* species.

Slight variations were observed in the antimicrobial activity profiles of *B. fragilis* species according to the source of isolate (Table 3). However, for skin and soft tissue isolates, cefoxitin displayed better activity, while clindamycin and moxifloxacin were the least active. Similarly, non-*B. fragilis* species recovered from skin and soft tissues showed higher resistance to clindamycin, moxifloxacin, and also cefoxitin (Table 3).

DISCUSSION

As reported previously elsewhere in the world, variability in the resistance patterns among the species of the *B. fragilis* group and the emergence of resistance to some of the most active β-lactams was observed (2, 8, 12, 16, 17, 27).

In this study, the ampicillin-sulbactam susceptibility rates of *B. fragilis* and non-*B. fragilis* species were 97% and 85%, respectively. No increase in the resistance to ampicillin-sulbactam has been observed compared with previous studies conducted in Argentina, so the combination of these two drugs remains a good therapeutic option for anaerobic bacterial infections (8, 15).

Although carbapenems constitute the most active β-lactams against these microorganisms, the emergence of resistance to carbapenems and also to piperacillin-tazobactam in Argentina is reported here. Imipenem resistance due to metallo-β-lactamases has been reported since 1986 (7); however, it re-

TABLE 1 *In vitro* activity of 10 antimicrobial agents against 363 isolates of the *Bacteroides fragilis* group

Organism (no. of isolates) and antimicrobial agent(s)	MIC (μg/ml)			% of isolates with the indicated susceptibility:		
	Range	50%	90%	Susceptible	Intermediate	Resistant
Bacteroides fragilis (198)						
Ampicillin-sulbactam	0.125–128	1	8	97.0	1.5	1.5
Piperacillin-tazobactam	≤0.03–>512	0.25	2	98.5	0.0	1.5
Cefoxitin	1–256	16	32	82.8	11.1	6.1
Ertapenem (126)	0.06–>64	0.25	4	96.0	1.6	2.4
Imipenem	≤0.015–>64	0.125	0.5	98.5	0.0	1.5
Doripenem (159)	0.03–>64	0.25	1	97.5	0.6	1.9
Clindamycin	≤0.125–>256	1	>256	74.7	2.5	22.7
Metronidazole	≤0.06–4	0.5	1	100.0	0.0	0.0
Moxifloxacin	0.06–64	0.5	2	89.9	2.0	8.1
Tigecycline	≤0.03–4	0.125	1	100	0.0	0.0
Bacteroides thetaiotaomicron/ovatus (69)						
Ampicillin-sulbactam	0.5–32	0.5	1	87.0	8.7	4.3
Piperacillin-tazobactam	≤0.03–64	≤0.03	4	98.6	1.4	0.0
Cefoxitin	4–128	4	32	49.3	27.5	23.2
Ertapenem (49)	0.125–>64	0.12	1	89.8	4.1	4.1
Imipenem	≤0.015–16	≤0.015	0.125	98.6	0.0	1.4
Doripenem (58)	0.125–64	0.125	0.25	96.6	0.0	3.4
Clindamycin	≤0.125–>256	≤0.125	4	42.0	18.8	39.1
Metronidazole	≤0.06–2	≤0.06	0.5	100.0	0.0	0.0
Moxifloxacin	0.125–64	0.125	1	89.9	2.9	7.2
Tigecycline	≤0.03–2	≤0.03	0.125	100.0	0.0	0.0
Bacteroides caccae (30)						
Ampicillin-sulbactam	0.5–16	1	4	90	10	0
Piperacillin-tazobactam	≤0.03–8	0.25	4	100	0	0
Cefoxitin	4–128	16	64	63.3	16.7	20
Ertapenem (19)	0.125–8	0.5	4	94.7	5.3	0
Imipenem	≤0.015–2	0.125	0.5	100	0	0
Doripenem (25)	0.125–8	0.25	1	96	4	0
Clindamycin	≤0.125–>256	1	>256	63.3	10	26.7
Metronidazole	≤0.06–4	0.5	1	100	0	0
Moxifloxacin	0.125–16	0.5	4	86.7	6.7	6.7
Tigecycline	≤0.03–4	0.5	1	100.0	0	0.0
Bacteroides vulgatus (27)						
Ampicillin-sulbactam	0.125–128	4	16	74.1	18.5	7.4
Piperacillin-tazobactam	0.25–512	2	8	96.3	0.0	3.7
Cefoxitin	0.5–128	8	32	81.5	7.4	11.1
Ertapenem (24)	0.125–2	0.5	1	100.0	0.0	0.0
Imipenem	≤0.015–4	0.25	1	100.0	0.0	0.0
Doripenem	0.06–1	0.25	0.5	100.0	0.0	0.0
Clindamycin	≤0.125–>256	0.5	>256	66.7	3.7	29.6
Metronidazole	0.25–4	0.5	1	100.0	0.0	0.0
Moxifloxacin	0.06–128	1	8	81.5	3.7	14.8
Tigecycline	≤0.03–2	0.06	0.5	100.0	0.0	0.0
Uncommon *B. fragilis* group species (39)[a]						
Ampicillin-sulbactam	≤0.125–32	2	16	87.2	10.3	2.6
Piperacillin-tazobactam	≤0.03–64	1	8	97.4	2.6	0.0
Cefoxitin	4–128	16	64	61.5	20.5	17.9
Ertapenem (34)	0.06–16	0.5	2	97.1	0.0	2.9
Imipenem	0.03–4	0.25	0.5	100.0	0.0	0.0
Doripenem (37)	0.125–4	0.25	1	100.0	0.0	0.0
Clindamycin	≤0.125–>256	2	>256	51.3	7.7	41.0
Metronidazole	0.06–4	0.5	2	100.0	0.0	0.0
Moxifloxacin	≤0.03–64	1	16	79.5	5.1	15.4
Tigecycline	≤0.03–4	0.125	1	100.0	0.0	0.0

[a] The 39 uncommon isolates of the *B. fragilis* group species were found to belong to the following species: *Parabacteroides* (*Bacteroides*) *distasonis*, 7 isolates; *Bacteroides uniformis*, 7 isolates; *Bacteroides stercoris*, 7 isolates; *Bacteroides merdae*, 2 isolates; and *Bacteroides* spp., 16 isolates.

TABLE 2 Phenotypic and genotypic characterization of MBLs in carbapenem-resistant isolates and isolates with decreased susceptibility

Organism(s)	Center	MIC (μg/ml)[a]				EDTA inhibition	*cfiA* gene
		ERT	IMI	IMI+EDTA	DOR		
B. fragilis	2	>64	>64	≤0.015	>64	+	+
	1	>64	32	0.125	>64	+	+
	1	32	32	0.06	32	+	+
	1	8	4	≤0.015	4	+	−
	3	8	0.5	0.125	4	−	+
	1	4	2	≤0.015	2	+	−
	1	4	1	0.125	4	+	+
	1	4	1	0.125	4	+	+
	2	4	0.5	0.125	4	−	+
	1	4	0.5	0.125	1	−	−
	2	4	0.5	0.125	1	−	−
	5	4	0.25	0.03	8	+	+
	6	4	0.06	≤0.015	4	−	−
	8	4	0.25	0.03	2	+	−
B. caccae	1	8	2	0.125	8	+	−
	4	4	0.5	0.125	1	−	−
	3	4	0.5	≤0.015	0.5	+	−
B. thetaiotaomicron/ovatus	3	>64	16	8	64	−	−
	4	32	4	0.5	16	+	−
	1	8	2	0.5	4	−	−
	1	8	1	≤0.015	4	+	−
	7	4	0.5	≤0.015	1	+	−
B. stercoris	1	16	4	≤0.015	4	+	−

[a] ERT, ertapenem; IMI, imipenem; DOR, doripenem.

mains infrequent in the United States and Europe, ranging from 0.4% to 1.5%, similar to what was observed in this study (2, 17, 25, 26, 28).

The production of metallo-β-lactamase (5) CfiA was reported to be responsible for resistance to carbapenems in *B. fragilis* as early as 1990 (1, 23). The inhibition by EDTA was a very good predictor for the presence of metallo-β-lactamases in the highly resistant *B. fragilis* strains, but discrepant results were observed for strains with decreased susceptibility. Phenotypic detection could be improved using ertapenem instead of imipenem, especially in

TABLE 3 *In vitro* activity of 10 antimicrobial agents against 363 isolates of *Bacteroides fragilis* grouped by the source of the isolate

Source of isolate and organism (no. of isolates)	% of isolates susceptible to the following antimicrobial agent[a]:									
	AMS	PTZ	FOX	ETP	IMI	DOR	CLI	MXF	TIG	MTZ
Abdomen										
B. fragilis (102)	96	99	83	95	99	98	75	92	100	100
Non-*B. fragilis* species (108)	85	98	65	95	99	99	58	87	100	100
Skin and soft tissues										
B. fragilis (35)	100	100	94	100	100	100	65	79	100	100
Non-*B. fragilis* species (16)	69	94	38	92	100	93	44	81	100	100
Genital tract										
B. fragilis (9)	100	100	67	100	100	100	78	89	100	100
Non-*B. fragilis* species (21)	95	100	76	100	100	100	67	95	100	100
Others[b]										
B. fragilis (15)	87	93	67	92	93	93	80	100	100	100
Non-*B. fragilis* species (15)	87	100	33	92	100	92	13	87	100	100
Blood										
B. fragilis (30)	96	100	90	95	97	96	66	83	100	100
Non-*B. fragilis* species (12)	92	100	67	100	100	100	50	75	100	100

[a] AMS, ampicillin-sulbactam; PTZ, piperacillin-tazobactam; FOX, cefoxitin; ETP, ertapenem; IMI:, imipenem; DOR, doripenem; CLI, clindamycin; MXF, moxifloxacin; TIG, tigecycline; MTZ, metronidazole.

[b] The other sources included bones (11 isolates), pericardial fluid (2 isolates), middle ears (6 isolates), lung (1 isolate), head and neck abscesses (3 isolates), hematomas (3 isolates), and surgical wounds (4 isolates).

low-level CfiA-producing bacteria, as was already described for meropenem by Bogaerts et al. (4).

Only a small percentage of *B. fragilis* strains carrying the *cfiA* gene express the protein at a level high enough to classify the strain as resistant. High-level resistance has been associated with the presence of an insertion element that may provide an efficient promoter immediately upstream of the *cfiA* gene (13, 21, 22). In our samples, the presence of the *cfiA* gene was confirmed in 8 out of 23 isolates of the *B. fragilis* group displaying resistance or decreased susceptibility to carbapenems. The estimated prevalence of *cfiA* in clinically imipenem-susceptible *B. fragilis* isolates (imipenem MICs ≤ 4 μg/ml) was 2.5%, as was described in the literature (31).

The overall moxifloxacin susceptibility rate remained higher than that reported by Snydman et al. and Betriu et al. (65.5% and 86.1%, respectively) (2, 28). In our isolates, moxifloxacin resistance was variable among species and ranged from as low as 6.7% for *B. caccae* to almost double in *B. vulgatus* (14.8%) and the less common species of the group (15.4%). Even at very different levels, the same was previously observed by Snydman et al. (27.4% and 54.7% for *B. fragilis* and *B. vulgatus*, respectively), and Betriu et al. (9% and 25% for *B. fragilis* and *B. uniformis*, respectively). It is noteworthy that an 8-fold increase in MIC_{90} values after 10 years of monitoring was reported. Moxifloxacin activity should be monitored because of the gradual decrease of MIC values to resistance levels (10, 19, 20, 26).

Previous reports have already shown that resistance to both clindamycin and cefoxitin makes their empirical use unacceptable (2, 8, 30). Compared with our last surveys, resistance to clindamycin increased from 16% to 25% within *B. fragilis*, and, even worse, from 44 to 48% in non-*B. fragilis* species (8). It is clear that clindamycin should no longer be used without prior susceptibility testing. For cefoxitin, the range of susceptibility was similar to that of previous studies (global susceptibility about 60%).

In agreement with previous reports, metronidazole and tigecycline are still the most active agents. In our case, MIC_{90} values for tigecycline ranged from 0.125 to 1 μg/ml, well below the 4 to 8 μg/ml values reported by other authors, and unlike Betriu et al. (3) and Grisold et al. (11), no resistance of any kind was found. It is also important to highlight that not a single metronidazole-resistant isolate was found in Argentina, which is different from the situation in European countries and the United States (9, 14, 17, 24, 27).

We want to make doctors and scientists aware of the emergence of *B. fragilis* group isolates resistant to carbapenems and to piperacillin-tazobactam in Argentina. Among these isolates, the presence of the *cfiA* metallo-β-lactamase gene could be responsible for the resistance or decreased susceptibility observed in some isolates of *B. fragilis* sensu stricto. However, additional studies should be performed in order to determine the mechanisms involved in the regulation of the expression of the *cfiA* gene, the clinical relevance of reduced susceptibility, and the challenge to discover clues about the mechanisms responsible for the resistance observed in non-*B. fragilis* isolates.

ACKNOWLEDGMENTS

This study was partially supported by Fundación Alberto J. Roemmers, Buenos Aires, Argentina. M.R. and G.G. are members of Carrera del Investigador Científico, CONICET, Argentina.

We are grateful to Marcelo Galas and members of the Laboratorio de Antimicrobianos, INEI-ANLIS Dr. Carlos G. Malbrán, Argentina, for their encouragement, contributing to the collection of strains, and sending the strains needed to conduct this study.

Members of the Anaerobe Surveillance Team are M. Bottiglieri, M. Rocchi, I. A. Marqués, M. Machain, M. C. Mauro, M. R. Nuñez, and D. Ballester.

REFERENCES

1. **Bandoh K, Muto Y, Watanabe K, Katoh N, Ueno K.** 1991. Biochemical properties and purification of metallo-beta-lactamase from *Bacteroides fragilis*. Antimicrob. Agents Chemother. **35**:371–372.
2. **Betriu C, et al.** 2008. Resistance trends of the *Bacteroides fragilis* group over a 10-year period, 1997 to 2006, in Madrid, Spain. Antimicrob. Agents Chemother. **52**:2686–2690.
3. **Betriu C, Culebras E, Gomez M, Rodriguez-Avial I, Picazo JJ.** 2005. In vitro activity of tigecycline against *Bacteroides* species. J. Antimicrob. Chemother. **56**:349–352.
4. **Bogaerts P, et al.** 2008. Evaluation of a new meropenem-EDTA double-ended Etest strip for the detection of the *cfiA* metallo-beta-lactamase gene in clinical isolates of *Bacteroides fragilis*. Clin. Microbiol. Infect. **14**:973–977.
5. **Bush K, Jacoby GA, Medeiros AA.** 1995. A functional classification scheme for beta-lactamases and its correlation with molecular structure. Antimicrob. Agents Chemother. **39**:1211–1233.
6. **Clinical and Laboratory Standards Institute.** 2007. Methods for antimicrobial susceptibility testing of anaerobic bacteria, 7th ed. Approved standard M11-A7. Clinical and Laboratory Standards Institute, Wayne, PA.
7. **Cuchural GJ, Jr, Malamy MH, Tally FP.** 1986. Beta-lactamase-mediated imipenem resistance in *Bacteroides fragilis*. Antimicrob. Agents Chemother. **30**:645–648.
8. **Fernandez Canigia L, et al.** 2007. Susceptibility trends of *Bacteroides fragilis* group isolates from Buenos Aires, Argentina. Rev. Argent. Microbiol. **39**:156–160.
9. **Gal M, Brazier JS.** 2004. Metronidazole resistance in *Bacteroides* spp. carrying *nim* genes and the selection of slow-growing metronidazole-resistant mutants. J. Antimicrob. Chemother. **54**:109–116.
10. **Golan Y, et al.** 2003. Emergence of fluoroquinolone resistance among *Bacteroides* species. J. Antimicrob. Chemother. **52**:208–213.
11. **Grisold AJ, et al.** 2007. In vitro activity of ten antibiotics, including tigecycline, against *Bacteroides* species in Austria. Eur. J. Clin. Microbiol. Infect. Dis. **26**:525–527.
12. **Hecht DW.** 2006. Anaerobes: antibiotic resistance, clinical significance, and the role of susceptibility testing. Anaerobe **12**:115–121.
13. **Kato N, Yamazoe K, Han CG, Ohtsubo E.** 2003. New insertion sequence elements in the upstream region of *cfiA* in imipenem-resistant *Bacteroides fragilis* strains. Antimicrob. Agents Chemother. **47**:979–985.
14. **Katsandri A, Avlamis A, Pantazatou A, Houhoula DP, Papaparaskevas J.** 2006. Dissemination of *nim*-class genes, encoding nitroimidazole resistance, among different species of Gram-negative anaerobic bacteria isolated in Athens, Greece. J. Antimicrob. Chemother. **58**:705–706.
15. **Litterio M, et al.** 2004. "In vitro" activity of ten antimicrobial agents against anaerobic bacteria. A collaborative study, 1999–2002. Rev. Argent. Microbiol. **36**:130–135. (In Spanish.)
16. **Liu CY, et al.** 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.
17. **Nagy E, Urban E, Nord CE, ESCMID Study Group on Antimicrobial Resistance in Anaerobic Bacteria.** 2011. Antimicrobial susceptibility of *Bacteroides fragilis* group isolates in Europe: 20 years of experience. Clin. Microbiol. Infect. **17**:371–379.
18. **Nguyen MH, et al.** 2000. Antimicrobial resistance and clinical outcome of *Bacteroides* bacteremia: findings of a multicenter prospective observational trial. Clin. Infect. Dis. **30**:870–876.
19. **Oteo-Iglesias J, Alos JI, Gomez-Garces JL.** 2002. Increase in resistance to new fluoroquinolones from 1998 to 2001 in the *Bacteroides fragilis* group. J. Antimicrob. Chemother. **50**:1055–1057.
20. **Papaparaskevas J, et al.** 2008. Moxifloxacin resistance is prevalent among *Bacteroides* and *Prevotella* species in Greece. J. Antimicrob. Chemother. **62**:137–141.
21. **Podglajen I, Breuil J, Bordon F, Gutmann L, Collatz E.** 1992. A silent

carbapenemase gene in strains of *Bacteroides fragilis* can be expressed after a one-step mutation. FEMS Microbiol. Lett. **70**:21–29.
22. **Podglajen I, Breuil J, Collatz E.** 1994. Insertion of a novel DNA sequence, 1S1186, upstream of the silent carbapenemase gene *cfiA*, promotes expression of carbapenem resistance in clinical isolates of *Bacteroides fragilis*. Mol. Microbiol. **12**:105–114.
23. **Rasmussen BA, Gluzman Y, Tally FP.** 1990. Cloning and sequencing of the class B beta-lactamase gene (*ccrA*) from *Bacteroides fragilis* TAL3636. Antimicrob. Agents Chemother. **34**:1590–1592.
24. **Seifert H, Dalhoff A.** 2010. German multicentre survey of the antibiotic susceptibility of *Bacteroides fragilis* group and *Prevotella* species isolated from intra-abdominal infections: results from the PRISMA study. J. Antimicrob. Chemother. **65**:2405–2410.
25. **Snydman DR, Jacobus NV, McDermott LA.** 2008. In vitro activities of doripenem, a new broad-spectrum carbapenem, against recently collected clinical anaerobic isolates, with emphasis on the *Bacteroides fragilis* group. Antimicrob. Agents Chemother. **52**:4492–4496.
26. **Snydman DR, et al.** 2011. Update on resistance of *Bacteroides fragilis* group and related species with special attention to carbapenems 2006–2009. Anaerobe **17**:147–151.
27. **Snydman DR, et al.** 2010. Lessons learned from the anaerobe survey: historical perspective and review of the most recent data (2005–2007). Clin. Infect. Dis. **50**(Suppl. 1):S26–S33.
28. **Snydman DR, et al.** 2007. National survey on the susceptibility of *Bacteroides fragilis* group: report and analysis of trends in the United States from 1997 to 2004. Antimicrob. Agents Chemother. **51**:1649–1655.
29. **Snydman DR, et al.** 2002. National survey on the susceptibility of *Bacteroides fragilis* Group: report and analysis of trends for 1997–2000. Clin. Infect. Dis. **35**:S126–S134.
30. **Wybo I, et al.** 2007. Third Belgian multicentre survey of antibiotic susceptibility of anaerobic bacteria. J. Antimicrob. Chemother. **59**:132–139.
31. **Yamazoe K, et al.** 1999. Distribution of the *cfiA* gene among *Bacteroides fragilis* strains in Japan and relatedness of *cfiA* to imipenem resistance. Antimicrob. Agents Chemother. **43**:2808–2810.