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Survey of Anaerobic Susceptibility Patterns in Canada

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The in vitro activity of penicillin, cefoxitin, moxalactam, ticarcillin, clindamycin, chloramphenicol, and metronidazole against 590 anaerobic isolates collected from five Canadian hospitals during 1984 was determined by an agar dilution technique. Cefoxitin, clindamycin, chloramphenicol, and metronidazole were very active against most of the isolates. No major regional differences in the susceptibility patterns were observed.

Routine susceptibility testing of anaerobes has been of limited usefulness in the immediate treatment of patients with anaerobic infections, because of the delay in obtaining results (5). Because most general bacteriology laboratories do not routinely perform susceptibility testing of anaerobic bacteria, it is necessary for reference laboratories to do periodic surveys to detect major changes in susceptibility profiles and to provide susceptibility patterns useful for a rational basis for empirical therapy. In recent years, the in vitro antimicrobial susceptibility pattern of anaerobic bacteria seems to have undergone gradual change (1–3, 7–10, 13, 17, 18, 21, 25). The purpose of this study was to determine susceptibility patterns of anaerobic bacteria in Canada.

The anaerobic strains were obtained from nonduplicate clinically significant isolates collected from March 1984 to October 1984 by five Canadian medical centers: Victoria General Hospital, Halifax, Nova Scotia; Hôpital Saint-Luc, Montreal, Quebec; Toronto Western Hospital, Toronto, Ontario; Vancouver General Hospital, Vancouver, British Columbia; and Saint-Boniface General Hospital, Winnipeg, Manitoba. The isolates were sent to the Hôpital Saint-Luc laboratory, where the identity of the strains was confirmed by established methods (14, 24) and antimicrobial susceptibility testing was performed.

The MICs were determined by the proposed standard reference agar dilution procedure for antimicrobial susceptibility testing of anaerobic bacteria using Wilkins-Chalgren agar (20). The following laboratory-standard antibiotic powders were tested: penicillin G (Ayerst Laboratories, Montreal, Quebec, Canada), cefoxitin (Merck Frosst Canada Inc., Pointe-Claire, Quebec, Canada), moxalactam (Eli Lilly & Co., Indianapolis, Inc.), ticarcillin (Beecham Laboratories, Pointe-Claire, Quebec, Canada), chloramphenicol (Parke Davis Canada Inc., Brockville, Ontario, Canada), clindamycin (The Upjohn Co., Kalamazoo, Mich.), and metronidazole (Rhône Poulenc Pharma Inc., Montreal, Quebec, Canada). All data were stored, retrieved, and analyzed by using database management and statistical programs developed for the TRS-80 model 4 (Tandy Corp., Fort Worth, Tex.) computer. MIC breakpoints, above which the organisms were considered to be resistant, were established for each of the antimicrobial agents (11, 22, 26). The lower and higher breakpoints, respectively, were as follows: penicillin G, 16 and 32 U/ml; cefoxitin, 16 and 32 μg/ml; moxalactam, 16 and 32 μg/ml; ticarcillin, 64 and 128 μg/ml; chloramphenicol, 8 and 16 μg/ml; clindamycin, 4 and 8 μg/ml; and metronidazole, 8 and 16 μg/ml.

Susceptibility results were available on 590 of the 722 isolates collected. The 590 strains were isolated from blood (16.1%), normally sterile body fluids and tissues (30.8%), the female genital tract (6.8%), and abdominal and wound infections (46.3%). A total of 132 strains received could not be tested: 48 isolates did not grow upon subculturing, 67 were heavily contaminated, and 17 failed to grow on Wilkins-Chalgren agar.

The results of the combined data from the five centers for all the species tested are shown in Table 1. With breakpoints of 16 and 32 U/ml, 37 and 15% of the isolates of the *Bacteroides fragilis* group were resistant to penicillin. However, 90% of the 260 strains were β-lactamase producers, so that these arbitrary breakpoints, although widely used in the literature, may not be clinically relevant. Ticarcillin, moxalactam, and cefoxitin possessed good activity against the *B. fragilis* group of organisms; these results are in agreement with published surveys (7, 9, 26). Tally et al. (26) found rates of resistance to cefoxitin of 16 and 3% at the lower and higher breakpoints, respectively, whereas we observed rates of 21 and 2%. As these authors (26) have pointed out in the past, the MICs of cefoxitin for most isolates cluster around 16 μg/ml, so that a single twofold-dilution change in the MIC can result in a large fraction of the isolates becoming resistant. This may explain the wide variation in rates of resistance to cefoxitin observed in different studies. The 0.3% rate of resistance to clindamycin was lower than the rates previously observed in several North American (4, 7, 8, 10, 13, 17) and European (1, 9, 21) surveys. In 1984, 2.5 times more clindamycin was used per capita in the United States than in Canada (Intercontinental Medical Statistics, December 1985 report). This different antimicrobial prescribing pattern may partly explain the low resistance rate observed in our survey. As expected, chloramphenicol and metronidazole were uniformly active. There was variability in the resistance rates among the various species of the *B. fragilis* group (Table 2), with *B. fragilis* and *Bacteroides vulgatus* being more susceptible to the β-lactam agents than

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TABLE 1. Comparative in vitro activity of seven antimicrobial agents against anaerobic bacteria

Organism (no. of isolates)	Antimicrobial agent	MIC (μg/ml)[a]			% Resistant[b]
		Range	50%	90%	
Bacteroides fragilis group (260)	Penicillin	0.5–>128	16	>128	37 (15)
	Cefoxitin	0.5–>128	8	32	21 (2)
	Moxalactam	0.25–>128	1	64	15 (12)
	Ticarcillin	0.25–>128	32	>128	13 (12)
	Clindamycin	≤0.06–>128	0.25	2	0.7 (0.3)
	Chloramphenicol	0.5–8	4	4	0 (0)
	Metronidazole	0.2–4	1	2	0 (0)
Bacteroides spp.[c] (35)	Penicillin	≤0.06–32	1	16	11 (0)
	Cefoxitin	≤0.06–128	1	32	14 (6)
	Moxalactam	≤0.06–>128	4	64	17 (14)
	Ticarcillin	≤0.06–64	2	64	0 (0)
	Clindamycin	≤0.06–2	0.12	1	0 (0)
	Chloramphenicol	≤0.06–16	2	4	0 (0)
	Metronidazole	≤0.06–4	0.5	4	0 (0)
Clostridium perfringens (78)	Penicillin	≤0.06–1	<0.06	0.5	0 (0)
	Cefoxitin	0.12–16	1	1	0 (0)
	Moxalactam	≤0.06–2	0.12	0.5	0 (0)
	Ticarcillin	≤0.06–2	0.5	1	0 (0)
	Clindamycin	≤0.06–64	0.25	2	1 (1)
	Chloramphenicol	≤0.06–128	2	4	1 (1)
	Metronidazole	≤0.06–2	0.5	1	0 (0)
Clostridium spp.[d] (109)	Penicillin	≤0.06–>128	0.25	4	7 (7)
	Cefoxitin	≤0.06–>128	2	64	19 (17)
	Moxalactam	≤0.06–>128	4	64	23 (17)
	Ticarcillin	≤0.06–>128	4	64	8 (7)
	Clindamycin	≤0.06–>128	0.5	8	13 (8)
	Chloramphenicol	≤0.06–64	2	8	1 (1)
	Metronidazole	≤0.06–>128	0.5	1	1 (1)
Fusobacterium spp. (7)	Penicillin	≤0.06–>128	1	>128	38 (38)
	Cefoxitin	0.25–16	4	64	0 (0)
	Moxalactam	0.5–32	4	32	13 (13)
	Ticarcillin	≤0.06–>128	4	>128	38 (13)
	Clindamycin	≤0.06–32	0.5	32	13 (13)
	Chloramphenicol	≤0.06–4	2	4	0 (0)
	Metronidazole	≤0.06–1	0.5	1	0 (0)
Peptococcus spp. (50)	Penicillin	≤0.06–4	≤0.06	0.25	0 (0)
	Cefoxitin	≤0.06–16	0.25	4	0 (0)
	Moxalactam	≤0.06–64	1	32	12 (4)
	Ticarcillin	≤0.06–32	0.5	4	0 (0)
	Clindamycin	≤0.06–>128	≤0.06	4	10 (8)
	Chloramphenicol	≤0.06–8	2	4	0 (0)
	Metronidazole	≤0.06–>128	1	>128	16 (16)
Peptostreptococcus spp. (13)	Penicillin	≤0.06–2	≤0.06	2	0 (0)
	Cefoxitin	≤0.06–2	0.12	2	0 (0)
	Moxalactam	≤0.06–8	0.5	8	0 (0)
	Ticarcillin	≤0.06–8	1	4	0 (0)
	Clindamycin	≤0.06–2	0.25	1	0 (0)
	Chloramphenicol	2–8	2	4	0 (0)
	Metronidazole	≤0.06–>128	0.5	128	15 (15)
Proprionibacterium acnes (8)	Penicillin	≤0.06–0.25	≤0.06	0.25	0 (0)
	Cefoxitin	0.12–0.25	0.12	0.12	0 (0)
	Moxalactam	0.5–1	0.5	1	0 (0)
	Ticarcillin	0.25–0.5	0.5	0.5	0 (0)
	Clindamycin	≤0.06–0.25	0.12	0.25	0 (0)
	Chloramphenicol	0.25–1	1	1	0 (0)
	Metronidazole	>128	>128	>128	100 (100)

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TABLE 1—*Continued*

Organism (no. of isolates)	Antimicrobial agent	MIC (μg/ml)[a]			% Resistant[b]
		Range	50%	90%	
Veillonella spp. (7)	Penicillin	0.12–32	4	32	14 (0)
	Cefoxitin	1–16	2	16	0 (0)
	Moxalactam	4–64	8	64	14 (14)
	Ticarcillin	2–>128	32	>128	14 (14)
	Clindamycin	≤0.06–2	≤0.06	2	0 (0)
	Chloramphenicol	0.5–4	2	4	0 (0)
	Metronidazole	1–4	1	1	0 (0)

[a] 50 and 90%, MIC for 50 and 90% of isolates tested, respectively.
[b] Percent resistant at the lower (higher) breakpoints. See the text.
[c] *B. melaninogenicus* (10 strains), *B. ruminicola* (6 strains), *B. bivius* (4 strains), *B. ureolyticus* (3 strains), *B. capillosus* (1 strain), and *Bacteroides* spp. (11 strains).
[d] *C. ramosum* (15 strains), *C. difficile* (12 strains), *C. bifermentans* (11 strains), *C. sordellii* (9 strains), *C. tertium* (8 strains), *C. butyricum* (7 strains), *C. clostridiiforme* (6 strains), *C. innocuum* (5 strains), *C. paraputrificum* (3 strains), and *Clostridium* spp. (33 strains).

the indole-positive species, *Bacteroides thetaiotaomicron* and *Bacteroides ovatus*, and *Bacteroides distasonis*. Our findings corroborate observations made by others (12, 15, 22). The breakdown of antimicrobial resistance of the *B. fragilis* group by center revealed that the proportions of the various species of the group collected by each cooperating hospital were similar; furthermore, no major regional differences in the susceptibility patterns were observed (data not shown).

Recent surveys have emphasized increased resistance to penicillin among non-*fragilis*-group *Bacteroides* spp. (10, 16–18) and related this finding to β-lactamase production (19, 23). Our findings confirmed these reports, as 11% of our isolates were resistant to 16 U of penicillin per ml and 52% were β-lactamase producers, but also uncovered increased resistance to cefoxitin (14%) and moxalactam (17%). Resistance to these latter agents has been reported in only a few isolates, and the reason for our relatively high resistance rates is unclear.

The seven antibiotics were predictably active against *Clostridium perfringens*, but the other *Clostridium* species exhibited variable degrees of resistance to the β-lactams and clindamycin. Increased resistance of these clostridia was previously documented (18, 24, 27).

Although the number of strains tested was very small, the observation of penicillin, ticarcillin, clindamycin, and cefoxitin resistance among the *Fusobacterium* spp. isolates suggests that these organisms, considered widely susceptible to most anti-anaerobic antibacterial agents, may have a changing susceptibility pattern. Of interest, two of the seven strains were β-lactamase producers.

Among the anaerobic gram-positive cocci, 15.8% of the strains required >8 μg of metronidazole per ml and 6.3% required >8 μg of clindamycin per ml for inhibition. Resistance to metronidazole is well documented (18, 25), but resistance to clindamycin has been rare among these organisms. The poor activity of metronidazole against *Proprionibacterium acnes* is in agreement with previously published data (6). Of interest, *Veillonella* spp. isolates demonstrated a susceptibility pattern similar to that of the gram-negative anaerobic bacilli.

In summary, the antimicrobial agents tested were very active against the clinically significant anaerobic isolates examined. There were no major regional differences in the susceptibility patterns observed with each group of organisms.

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TABLE 2. Resistance rates of *B. fragilis* group species

Species (no. of isolates)	% Resistance[a] to antimicrobial agent[b]						
	PEN	CFX	MOX	TIC	CLIN	CHL	MET
Bacteroides fragilis (153)	29.4 (17.6)	6.5 (0)	3.3 (2.6)	16.3 (14.4)	0.6 (0.6)	0 (0)	0 (0)
Bacteroides thetaiotaomicron (35)	64.7 (11.8)	82.4 (8.8)	35.3 (29.4)	5.9 (5.9)	0 (0)	0 (0)	0 (0)
Bacteroides ovatus (27)	65.4 (7.7)	34.6 (7.7)	34.6 (26.9)	7.7 (7.7)	0 (0)	0 (0)	0 (0)
Bacteroides vulgatus (22)	9.1 (9.1)	0 (0)	0 (0)	13.6 (4.5)	0 (0)	0 (0)	0 (0)
Bacteroides distasonis (18)	44.4 (22.2)	33.3 (0)	61.1 (50.0)	16.7 (16.7)	0 (0)	0 (0)	0 (0)
Bacteroides uniformis (5)	0 (0)	0 (0)	20 (0)	0 (0)	0 (0)	0 (0)	0 (0)

[a] Percent resistant at the lower (higher) breakpoints.
[b] PEN, Penicillin; CFX, cefoxitin; MOX, moxalactam; TIC, ticarcillin; CLIN, clindamycin; CHL, chloramphenicol; MET, metronidazole.

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