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Comparative In Vitro Activity of Ceftriaxone Against Anaerobic Bacteria

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The in vitro activity of ceftriaxone was compared with those of other recently introduced beta-lactam antimicrobial agents (cefoperazone, cefotaxime, and moxalactam) and with those of cefoxitin, clindamycin, and metronidazole against 227 strains of anaerobic bacteria. The data obtained in this investigation suggest that ceftriaxone, like a majority of the new beta-lactam antimicrobial agents, may be of limited value in the treatment of serious infections involving anaerobic bacteria.

Anaerobic bacteria are recognized as important pathogens in a significant percentage of human infections. The *Bacteroides fragilis* group, the most common anaerobic organisms found in clinical specimens, as well as other anaerobic bacteria, are often resistant to commonly used antimicrobial agents (6, 7). The last few years have seen a proliferation of beta-lactam antibiotics resulting from the modification of the side chains of the penicillin, cephalosporin, and cephamycin nuclei. Several newer ("third-generation") parenteral cephalosporins with expanded antibacterial spectra have been developed. Moxalactam, a novel 1-oxacephem, is a new beta-lactam antibiotic with reported potent and broad-spectrum activity. In a previous report, we found the overall activity of these new beta-lactams to be relatively poor against anaerobic bacteria (16).

Ceftriaxone (Ro 13-9904), a 2-aminothiazoylmethoxyimino semisynthetic cephalosporin, has recently been shown to have good in vitro and in vivo activities against many genera of bacteria (2, 4, 10, 18, 19). What distinguishes ceftriaxone from the other newer cephalosporins is its unusually long plasma half-life, which is 4 to 10 times longer than that of other cephalosporins (1, 2, 17, 20). In the present investigation, the in vitro activity of ceftriaxone against 227 strains of anaerobic bacteria was compared with those of cefoperazone, cefotaxime, moxalactam, metronidazole, clindamycin, and cefoxitin.

All bacterial strains were randomly selected, recent isolates from human clinical material at Wadsworth Veterans Administration Medical Center, Los Angeles, Calif. These bacteria were identified with established procedures (11, 21). Standard laboratory antimicrobial powders were diluted as recommended by the manufacturers and prepared fresh for each experiment. Minimal inhibitory concentrations (MICs) were determined by a blood agar plate dilution method previously described (21) and are summarized in Tables 1 and 2. The MIC was interpreted as the lowest concentration of each antimicrobial agent permitting no growth, one discrete colony, or a barely visible haze. Broth dilution tests were performed with methods described previously (21) to determine the bactericidal activity of each antimicrobial agent against members of the *B. fragilis* group. A summary of broth dilution susceptibility studies comparing the MICs and minimal bactericidal concentrations (MBCs) of these agents is shown in Table 3. The MBC was interpreted as the lowest concentration of antimicrobial agent that resulted in >99.9% killing of the organisms in the original inoculum after 48 h of incubation at 37°C.

This study is an extension of a prior investigation in which the in vitro activities of the new beta-lactam antibiotics against anaerobic bacteria were studied (16). Many of the observations made in our previous investigation were confirmed in this study with different strains. Cefoxitin remained the most active of the cephalosporins, inhibiting 91% of the anaerobic bacteria at its achievable serum level (Table 2).

Metronidazole and moxalactam were the only antimicrobial agents with MICs and MBCs within a fourfold dilution for all of the strains. In only one instance (clindamycin versus *Bacteroides distasonis*) was the MBC greater than eightfold higher than the MIC.

All of the antimicrobial agents were active against all of the strains of *Clostridium perfringens* and *Veillonella* sp. at their achievable serum levels. This is in contrast to a previous report which described the poor activity of cef-

TABLE 1. Susceptibility of anaerobic bacteria to ceftriaxone

Microorganism (no. of strains)	Susceptibility to ceftriaxone (μg/ml)		
	Range	MIC_{50}[a]	MIC_{90}[b]
Actinomyces sp. (7)[c]	≤0.06–2	0.5	2
Bacteroides fragilis (20)	4–>256	32	256
Bacteroides melaninogenicus subsp. *melaninogenicus* (10)	0.12–16	1	16
Bacteroides thetaiotaomicron (17)	2–>256	128	>256
Other *Bacteroides* spp. (65)[d]	≤0.06–>256	4	128
Clostridium difficile (10)	4–64	16	32
Other *Clostridium* spp. (34)[e]	0.12–>256	4	64
Eubacterium sp. (4)[f]	≤0.06–256	0.5	256
Fusobacterium nucleatum (10)	≤0.06–2	0.5	2
Other *Fusobacterium* spp. (12)[g]	≤0.06–>256	≤0.06	>256
Lactobacillus sp. (3)[h]	≤0.06–0.5	≤0.06	0.5
Peptococcus sp. (12)[i]	0.12–64	4	8
Peptostreptococcus sp. (8)[j]	0.5–16	0.5	16
Propionibacterium sp. (5)[k]	0.25–128	1	128
Streptococcus intermedius (7)	0.25–2	1	2
Veillonella parvula (3)	2	2	2

[a] Minimum concentration of ceftriaxone required to inhibit 50% of the strains.
[b] Minimum concentration of ceftriaxone required to inhibit 90% of the strains.
[c] *Actinomyces ondontolyticus* (three strains), *Actinomyces* sp. (four strains).
[d] *Bacteroides asaccharolyticus* (three strains), *B. bivius* (three strains), *B. capillosus* (five strains), *B. disiens* (four strains), *B. distasonis* (four strains), *B. melaninogenicus* subsp. *intermedius* (six strains), *B. oralis* (four strains), *B. ovatus* (five strains), *B. pneumosintes* (two strains), *B. putredinis* (two strains), *B. ruminicola* subsp. *brevis* (six strains), *B. ruminicola* subsp. *ruminicola* (three strains), *B. uniformis* (one strain), *B. ureolyticus* (six strains), *B. vulgatus* (six strains), *Bacteroides* sp. (five strains).
[e] *Clostridium butyricum* (one strain), *C. cadaveris* (one strain), *C. clostridiiforme* (four strains), *C. innocuum* (three strains), *C. paraputrificum* (three strains), *C. perfringens* (four strains), *C. ramosum* (five strains), *C. septicum* (three strains), *C. sordellii* (one strain), *C. sporogenes* (three strains), *C. subterminale* (three strains), *C. tertium* (three strains).
[f] *Eubacterium alactolyticum* (one strain), *E. lentum* (three strains).
[g] *Fusobacterium mortiferum* (three strains), *F. naviforme* (one strain), *F. necrophorum* (four strains), *F. prausnitzii* (one strain), *F. varium* (three strains).
[h] *Lactobacillus catenaforme* (three strains).
[i] *Peptococcus asaccharolyticus* (four strains), *P. magnus* (five strains), *P. prevotii* (three strains).
[j] *Peptostreptococcus anaerobius* (four strains), *P. micros* (two strains), *P. productus* (two strains).
[k] *Propionibacterium acnes* (three strains), *P. granulosum* (two strains).

triaxone against various clostridial species, including *C. perfringens* (15).

Clindamycin, cefoxitin, and metronidazole were the most active agents against the *B. fragilis* group, inhibiting 98 to 100% of the strains at concentrations which can be achieved in serum. Of the remaining agents tested against the *B. fragilis* group, moxalactam was the most active, with a 90% MIC of 128 μg/ml, whereas cefotaxime, cefoperazone, and ceftriaxone required >256 μg/ml for the same degree of inhibition. Some investigators have found ceftriaxone, moxalactam, and cefotaxime to be more active against the *B. fragilis* group than they were observed to be in this study (3, 5, 8, 12, 14).

Cefoperazone and moxalactam were distinctly less active against *Bacteroides distasonis, Bacteroides thetaiotaomicron*, and *Bacteroides ovatus* than against the other two members of the *B. fragilis* group. Cefotaxime and ceftriaxone were much less active against *B. fragilis* than the other antimicrobial agents; both agents have been shown to be susceptible to *Bacteroides* beta-lactamases (15, 23).

Metronidazole was the only antimicrobial agent active against all strains of *Fusobacterium* at its achievable serum level. Strains of *Fusobacterium mortiferum* and *Fusobacterium varium* were resistant to the other antimicrobial agents.

Although not an obligate anaerobe, *Streptococcus intermedius* was included in this investigation because it often fails to grow aerobically upon initial isolation from clinical material (21). Of the seven strains of *S. intermedius*, four were resistant to metronidazole, three were resistant to cefoxitin, and one strain each was resistant to moxalactam and clindamycin.

Metronidazole and ceftriaxone were the most active compounds against strains of *Clostridium difficile*, requiring concentrations of 2 and 64 μg/ml, respectively, to inhibit 100% of the strains.

TABLE 2. Percentage of strains susceptible to achievable serum concentrations

Anaerobic bacterium (no. of strains)	% Susceptible to indicated achievable serum level (μg/ml) of[a]						
	Cefoxitin (32)	Cefoperazone (32)	Cefotaxime (16)	Ceftriaxone (16)	Clindamycin (8)	Metronidazole (16)	Moxalactam (32)
Bacteroides fragilis group (53)	92	47	23	28	98	100	64
Other *Bacteroides* spp. (59)	98	92	95	85	98	100	86
Clostridium difficile (10)[b]	50	60	0	60	60	100	40
Clostridium perfringens (4)	100	100	100	100	100	100	100
Other *Clostridium* spp. (30)	83	80	63	70	73	93	83
Fusobacterium sp. (22)	86	82	82	82	91	100	82
Gram-negative anaerobic cocci (3)	100	100	100	100	100	100	100
Gram-positive anaerobic cocci (27)	96	85	100	96	85	85	93
Streptococcus intermedius	100	57	100	100	86	43	86
Gram-positive, nonsporeforming bacilli (19)	95	84	84	84	100	53	84
Total % susceptible	91	77	68	70	91	93	79

[a] Serum levels were selected on the basis of the peak blood concentration of an antimicrobial agent that can be achieved 30 to 60 min after administration of a maximal safe dosage. These levels were based on information supplied by the manufacturers (cefotaxime and ceftriaxone) and criteria used by Finegold (6) and Kirby et al. (13).

[b] There is no implication intended that serum levels are important in the case of this organism.

Several strains of *C. difficile* were resistant to all of the remaining beta-lactam antibiotics and clindamycin.

The poor activity of metronidazole against gram-positive, nonsporeforming bacilli agrees with published reports (9, 22). In addition, two of the four strains of *Clostridium clostridiiforme* were resistant to metronidazole (MIC, >256 μg/ml).

The data obtained in this investigation suggest that ceftriaxone, like a majority of the new beta-lactam antimicrobial agents, may be of limited value in the treatment of serious infections involving anaerobic bacteria.

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TABLE 3. Comparative inhibitory and bactericidal activities against 25 strains of the *B. fragilis* group

Anaerobic bacterium (no. of strains)	Range of minimal concn of following antimicrobial agent:													
	Cefoxitin		Cefoperazone		Cefotaxime		Ceftriaxone		Clindamycin		Metronidazole		Moxalactam	
	MIC	MBC	MIC	MBC	MIC	MBC	MIC	MBC	MIC	MBC	MIC	MBC	MIC	MBC
B. fragilis (10)	2–32	4–32	4–>256	8–>256	2–128	4–>256	4–256	8–>256	≤0.062–2	0.125–8	0.25–1	0.5–2	0.5–64	1–128
B. thetaiotaomicron (8)	2–32	4–128	2–>256	8–>256	2–>256	4–>256	2–>256	8–>256	0.25–4	0.5–4	0.25–0.5	0.25–1	1–128	1–128
B. distasonis (3)	4–16	16	1–8	8–16	0.5–1	1–8	1–8	2–8	0.25–16	4–32	0.25–0.5	0.25–0.5	8–64	16–256
B. ovatus (3)	16–64	64–128	128–256	32–128	32–128	128	128–256	128–256	4–8	4–8	0.25	0.5–1	8–32	8–32

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