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In Vitro Activity and β-Lactamase Stability of a New Penem, CGP 31608

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The in vitro activity of CGP 31608, a new penem, against aerobic and anaerobic organisms was evaluated and compared with those of other beta-lactams. CGP 31608 inhibited *Escherichia coli*, *Klebsiella pneumoniae*, *K. oxytoca*, *Proteus mirabilis*, *Citrobacter diversus*, and *Salmonella*, *Shigella*, *Aeromonas*, and *Yersinia* spp. with MICs for 50% of the strains (MIC_{50}s) of 2 to 4 μg/ml and MIC_{90}s of 4 μg/ml, compared with cefotaxime, ceftazidime, aztreonam, and imipenem MICs of <0.25 μg/ml. MIC_{90}s were 8 μg/ml for *Enterobacter* species and *C. freundii*, for which other agents had MICs of 32 μg/ml, except imipenem, which had equal activity. The MIC_{90} for *Proteus vulgaris*, *Morganella morganii*, *Providencia stuartii*, and *Providencia rettgeri* was 8 μg/ml, compared with <2 μg/ml shown by the other agents. *Acinetobacter* species resistant to other agents except imipenem were inhibited by 4 μg/ml, as were *Pseudomonas aeruginosa*, including piperacillin-, ceftazidime-, and gentamicin-resistant isolates. The MIC for *P. cepacia*, *P. fluorescens*, and *P. acidovorans* was ≤8 μg/ml, but that for *P. maltophilia* was ≥128 μg/ml. Hemolytic streptococci A, B, C, G, and F were inhibited by <1 μg/ml, but the MIC for *Streptococcus faecalis* was ≥32 μg/ml. MICs for *Staphylococcus aureus* methicillin-susceptible and -resistant strains were ≤1 μg/ml, as were those for methicillin-susceptible and -resistant *S. epidermidis*. *Bacteroides fragilis* and *Clostridium* species and *Fusobacterium* spp. were inhibited by ≤4 μg/ml. CGP 31608 was not hydrolyzed by plasmid beta-lactamases TEM-1, TEM-2, SHV-1, PSE-1, OXA-2, PSE-4, or by *S. aureus*. Chromosomal beta-lactamases of type Ia in *Enterobacter cloacae* P99 and *Morganella morganii*, Ic in *P. vulgaris*, K-1 in *K. oxytoca*, and Id in *P. aeruginosa* also did not hydrolyze CGP 31608. It inhibited TEM-1, but the 50% inhibitory concentration was 14.2 μg/ml compared with 0.15 μg/ml for the P99 enzyme. CGP 31608 induced beta-lactamases in *P. aeruginosa*, *E. cloacae*, *C. freundii* and *Providencia rettgeri*, but there was no increase in MICs for the isolates and it did not select strains derepressed for beta-lactamase production. Synergy of CGP 31608 and gentamicin was found against 90% *P. aeruginosa*, 60% *Enterobacter cloacae*, and 50% *Serratia marcescens* strains. No synergy was found with rifampin. A postantibiotic effect was found against *E. coli*.

Although penem antibiotics have been known since the pioneering chemical syntheses of Woodward (3, 16), few agents of this class have achieved clinical success. Several penems have undergone preclinical investigation. These include SCH 29482, SCH 34343, FCE 22101, and FCE 22891 (1, 7, 10, 11, 15). Penems and carbapenems, such as imipenem, have been shown to inhibit a broad range of microorganisms, depending upon the substituent side chains (2, 4, 5). In general, the agents have been extremely stable against attack by beta-lactamases but are susceptible to hydrolysis by dehydropeptidases which are present in kidney, lung, and intestinal tissue of different animal species (5). We investigated the activity of a new penem, CGP 31608, (5*R*,6*S*)-2-(1′-aminoalkyl)-6-(hydroxyalkyl)penem-3-carboxylic compound (6) (Fig. 1), in comparison with those of other beta-lactam agents currently available.

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MATERIALS AND METHODS

CGP 31608 was provided by CIBA-GEIGY, Basel, Switzerland. All other agents were gifts from their manufacturers. The organisms used were isolates from patients at hospitals in the Columbia University system, and included isolates were retained because of known beta-lactamase activity or resistance to beta-lactam antibiotics or both.

Antimicrobial susceptibility tests. Antimicrobial activity was measured by an agar dilution method with Mueller-Hinton agar unless otherwise specified. A final inoculum of 10^5 CFU was applied with a replicating device. Broth dilution tests were performed with 5×10^5 CFU in tubes containing 1 ml. Incubation of agar and broth test tubes was at 35°C for 18 h. The susceptibility of *Neisseria* and *Haemophilus* spp. was determined with chocolate Mueller-Hinton agar in the presence of 5% CO_2. The susceptibility of streptococci was determined with Mueller-Hinton agar supplemented with 5% sheep blood, and the susceptibility of anaerobic species was determined with brucella agar supplemented with sheep blood, hemin, and vitamin K. Incubation of anaerobic cultures was for 48 h in GasPak jars (BBL Microbiological Systems, Cockeysville, Md.). The susceptibility of methicillin-resistant staphylococci was determined on Mueller-Hinton agar or broth supplemented with 5% NaCl; isolates for which the oxacillin MIC was >8 μg/ml were considered resistant. The MIC was defined as the lowest concentration of antibiotic that inhibited the development of visible growth on agar or in broth. The MBC was determined by subculture of 0.01 ml from clear tubes onto

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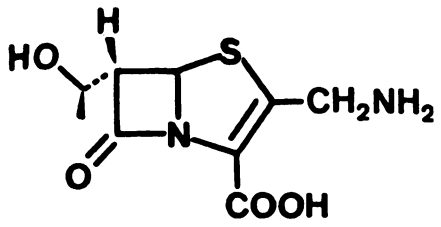

FIG. 1. Structure of CGP 31608 [(5*R*,6*S*)-2-(1'-aminoalkyl)-6-(hydroxyalkyl)penem-3-carboxylic acid].

antibiotic-free agar plates which indicated a 99.9% reduction in CFU. All tests were run with control strains from the American Type Culture Collection, Rockville, Md.

Beta-lactamase assays and inhibition studies. The presence of beta-lactamases in the clinical isolates was determined by the nitrocefin assay (9). Enzymes used to analyze the stability of CGP 31608 and other agents were prepared as described previously (9). The beta-lactamase stability was determined by spectrophotometric assay as described previously (9) and by use of a microbiological assay with *Pseudomonas aeruginosa* as the assay organism (9). Inhibition assays were performed with nitrocefin as substrate and by varying the concentrations of different inhibitors used to calculate 50% inhibitory (I_{50}) concentrations.

Induction of beta-lactamases. One strain each of *P. aeruginosa*, *Enterobacter cloacae*, *Citrobacter freundii*, and *Providencia rettgeri* were grown overnight in broth and diluted 100-fold to achieve exponential growth. During incubation at 37°C, inducers were added at concentrations equal to one-fourth the MIC, one-half the MIC, and the MIC, and incubation was continued for 2 h. Bacteria were harvested by centrifugation, washed in 0.05 M potassium phosphate buffer (pH 7), and subsequently disrupted by sonication. Cellular debris were removed by centrifugation, and the samples were dialyzed at 4°C for 24 h against phosphate buffer. Beta-lactamase activity was determined by using nitrocefin as substrate, with activity based on micromoles of substrate hydrolyzed per milligram of protein.

Selection of resistant mutants. To determine the frequency of spontaneous point resistance to CGP 31608, 5×10^9 organisms of different species were exposed to the drug in agar at four and eight times the MIC. Plates were incubated at 37°C and read at 24 and 48 h. A total of seven *Escherichia coli*, four *Klebsiella pneumoniae*, six *K. oxytoca*, three *Proteus vulgaris*, seven *C. freundii*, four *E. cloacae*, seven *Morganella morganii*, six *P. aeruginosa*, six *Serratia marcescens*, and five *Staphylococcus aureus* strains were tested to select mutants for which the MICs were increased.

Postantibiotic suppression effect. Exponentially growing bacteria at 10^6 CFU were exposed to a concentration of antibiotic at twice the MBC for 2 h. Samples were removed, diluted 10,000-fold in fresh antibiotic-free medium, and placed in a 37°C shaking water bath. Samples were removed every 2 h and plated on agar to determine the CFU remaining and the rate of regrowth. Control organisms not exposed to antibiotic were processed in an identical fashion.

Synergy studies. Synergy was determined by using a checkerboard agar method with concentrations of drugs varying twofold. Synergy was defined as a fractional inhibitory concentration of ≤0.5, and antagonism was defined as fractional inhibitory concentration of ≥4.

RESULTS

The comparative in vitro activity of CGP 31608 and other agents against gram-negative aerobic species is shown in Table 1. Overall, CGP 31608 had a very narrow range of inhibitory values for most species. The MIC of CGP 31608 for 90% of the strains (MIC_{90}) of *E. coli*, *K. pneumoniae*, *K. oxytoca*, *C. diversus*, *Hafnia alvei*, *P. vulgaris*, *Salmonella* spp. (including *S. typhi*), *Shigella* spp., *Aeromonas hydrophila*, and *Yersinia enterocolitica* was 4 μg/ml. Imipenem, SCH 34343, cefotaxime, ceftazidime, and aztreoman inhibited most of these species at ≤1 μg/ml, excluding *P. vulgaris* for cefotaxime. The CGP 31608 MIC_{90}s against species such as *C. freundii*, *E. cloacae*, and *E. aerogenes* were 8 μg/ml, which was comparable or superior to the MICs of cefotaxime, ceftazidime, and aztreonam, and superior to those of piperacillin. Imipenem had MICs against *E. cloacae* similar to those of CGP 31608 but was more active than CGP 31608 against *C. freundii* and *E. aerogenes*. Imipenem, ceftazidime, and aztreonam were more active than CGP 31608 against *Serratia marcescens*. CGP 31608 had MIC_{90}s for *Morganella morganii* and *Providencia* species of 8 μg/ml compared with imipenem, which had MICs of 4 and 2 μg/ml. Piperacillin-resistant *Proteus*, *Morganella* and *Providencia* spp. were inhibited by ≤8 μg of CGP 31608 per ml.

Haemophilus influenzae, *Neisseria gonorrhoeae*, *N. meningitidis* (five isolates not shown), and *Branhamella catarrhalis* were inhibited by ≤2 μg of CGP 31608 per ml, which was generally 2-fold higher than for imipenem and 8- to 16-fold higher than for cefotaxime, ceftazidime, and aztreonam. The majority of these isolates were beta-lactamase producing, and several chromosomally penicillin-resistant *N. gonorrhoeae* strains were included. CGP 31608 had excellent activity against *P. aeruginosa* and other *Pseudomonas* species, with the exception of *P. maltophilia*, inhibiting the majority at 4 μg/ml. Other *Pseudomonas* species were inhibited, including piperacillin- and gentamicin-resistant isolates and isolates for which the ceftazidime and aztreonam MICs were 32 μg/ml. Furthermore, CGP 31608 inhibited *Acinetobacter calcoaceticus* subspp. *lwoffi* and *anitratus* at ≤4 μg/ml, which was superior to the performance of all agents except imipenem.

Of note, there was a narrow range of susceptibility to CGP 31608 for pseudomonads, as there was for members of the family *Enterobacteriaceae*.

Table 2 illustrates the activity of CGP 31608 against gram-positive and anaerobic species in comparison with imipenem, SCH 34343, and cefotaxime. CGP 31608 inhibited methicillin-susceptible *S. aureus* strains at concentrations of ≤0.03 to 0.5 μg/ml. It had activity comparable to that of imipenem and SCH 34343 and was 4- to 8-fold more active than cefotaxime. At 2 μg/ml, CGP 31608 inhibited methicillin-resistant *S. aureus* strains which were resistant to imipenem and SCH 34343. Some methicillin-resistant *S. epidermidis* strains that were resistant to the other agents were inhibited by CGP 31608. Hemolytic streptococcal groups A, B, C, F, and G and *Streptococcus bovis* were inhibited by 0.5 to 1 μg/ml, but *S. faecalis* and *S. faecium* (eight isolates not shown) were resistant (CGP 31608 MICs, 16 to 32 μg/ml). CGP 31608 was minimally less active than imipenem and SCH 34343 against some *S. pneumoniae* strains but inhibited, at 2 μg/ml, penicillin G-resistant (MIC, >1 μg/ml) isolates. It had excellent inhibitory activity against viridans group streptococci such as *S. mutans*, *S. sanguis*, and *S. mitis* and was comparable in activity to imipenem. *Listeria monocytogenes* strains were inhibited,

TABLE 1. Comparative activity of CGP 31608 and other agents against gram-negative bacteria

Organism (no. of isolates)	Agent	MIC (μg/ml)		
		Range	50%	90%
E. coli[a] (30)	CGP 31608	2–4	4	4
	Imipenem	≤0.12–0.5	0.25	0.5
	SCH 34343	0.25–0.5	0.25	0.5
	Cefotaxime	≤0.12–1	≤0.12	≤0.12
	Ceftazidime	0.12–1	0.5	0.5
	Aztreonam	≤0.12–0.25	≤0.12	0.15
	Piperacillin	8–>128	128	>128
K. pneumoniae[a] (25)	CGP 31608	2–8	4	4
	Imipenem	0.12–0.5	0.12	0.5
	SCH 34343	0.25–2	0.25	0.5
	Cefotaxime	≤0.12–0.5	≤0.12	0.5
	Ceftazidime	≤0.12–0.5	0.12	0.5
	Aztreonam	≤0.12–0.5	≤0.12	0.12
	Piperacillin	2–>128	128	>128
K. oxytoca[a] (25)	CGP 31608	4–8	4	4
	Imipenem	0.12–4	0.12	0.5
	SCH 34343	0.25–2	0.25	0.5
	Cefotaxime	≤0.12–4	≤0.12	0.5
	Ceftazidime	0.12–2	≤0.12	0.5
	Aztreonam	0.12–16	0.25	1
	Piperacillin	2–>128	128	>128
C. diversus (20)	CGP 31608	2–4	2	4
	Imipenem	0.12–2	0.12	0.25
	SCH 34343	0.12–1	0.25	0.5
	Cefotaxime	≤0.12	≤0.12	≤0.12
	Ceftazidime	<0.12–0.5	≤0.12	0.25
	Aztreonam	≤0.12	≤0.12	≤0.12
	Piperacillin	2–128	4	8
C. freundii[a] (25)	CGP 31608	2–8	4	8
	Imipenem	0.12–2	0.5	2
	SCH 34343	0.25–2	0.5	1
	Cefotaxime	0.25–>64	0.25	32
	Ceftazidime	0.12–32	0.5	8
	Aztreonam	0.25–32	0.5	8
	Piperacillin	1–>128	8	>128
E. cloacae[a] (30)	CGP 31608	4–8	4	8
	Imipenem	0.12–16	2	8
	SCH 34343	0.25–8	2	4
	Cefotaxime	0.12–>32	8	>32
	Ceftaxidime	0.12–>32	8	32
	Aztreonam	0.12–>32	2	8
	Piperacillin	2–>128	32	>128
E. aerogenes[a] (20)	CGP 31608	2–8	4	8
	Imipenem	0.25–8	0.5	2
	SCH 34343	0.25–8	0.1	4
	Cefotaxime	≤0.12–32	0.12	8
	Ceftazidime	≤0.12–32	0.5	8
	Aztreonam	≤0.12–16	0.12	4
	Piperacillin	2–>128	4	32
E. agglomerans[a] (10)	CGP 31608	4–8	4	8
	Imipenem	≤0.12–2	0.12	1
	SCH 34343	0.5–4	0.5	2
	Cefotaxime	≤0.12–4	0.25	2
	Ceftazidime	≤0.12–1	0.25	0.5
	Aztreonam	≤0.12–1	0.25	0.5
	Piperacillin	1–8	2	8
H. alveii[a] (10)	CGP 31608	4	4	4
	Imipenem	0.5	0.5	0.5
	SCH 34343	0.5–1	0.5	1

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TABLE 1—*Continued*

Organism (no. of isolates)	Agent	MIC (μg/ml)		
		Range	50%	90%
	Cefotaxime	0.5–1	0.5	1
	Ceftazidime	0.5–1	0.5	1
	Aztreonam	0.12–25	0.12	0.25
	Piperacillin	4–128	16	128
S. marcescens[a] (30)	CGP 31608	8–16	8	8
	Imipenem	0.12–4	0.12	1
	SCH 34343	0.5–16	4	8
	Cefotaxime	0.12–16	1	8
	Ceftazidime	0.12–8	012	2
	Aztreonam	0.12–8	0.25	2
	Piperacillin	2–>128	32	>128
P. mirabilis (30)	CGP 31608	2–16	4	8
	Imipenem	0.12–4	0.5	1
	SCH 34343	0.25–2	0.5	1
	Cefotaxime	≤0.12	≤0.12	≤0.12
	Ceftazidime	≤0.12	≤0.12	≤0.12
	Aztreonam	≤0.12	≤0.12	≤0.12
	Piperacillin	1–>128	1	64
M. morganii[a] (25)	CGP 31608	4–16	4	8
	Imipenem	0.12–4	1	4
	SCH 34343	0.12–4	1	2
	Cefotaxime	≤0.12–16	≤0.12	4
	Ceftazidime	≤0.12–8	<0.12	1
	Azteonam	≤0.12–2	0.12	0.5
	Piperacillin	1–128	1	64
P. vulgaris[a] (20)	CGP 31608	4–8	4	4
	Imipenem	0.12	0.5	2
	SCH 34343	0.5–4	0.5	1
	Cefotazime	≤0.12–8	0.12	8
	Ceftazidime	≤0.12–2	≤0.12	0.25
	Aztreonam	≤0.12	≤0.12	≤0.12
	Piperacillin	1–>128	8	64
P. penneri[a] (10)	CGP 31608	4–8	4	8
	Imipenem	0.5–2	0.5	1
	SCH 34343	0.5–2	0.5	1
	Cefotaxime	1–32	1	8
	Ceftazidime	≤0.12–1	≤0.12	1
	Aztreonam	≤0.12	≤0.12	≤0.12
	Piperacillin	1–128	4	32
P. stuartii[a] (20)	CGP 31608	2–8	4	8
	Imipenem	0.5–4	1	2
	SCH 34343	0.5–2	1	2
	Cefotaxime	≤0.12–2	0.12	0.5
	Ceftazidime	≤0.12–1	0.25	0.5
	Aztreonam	≤0.12–0.5	≤0.12	0.5
	Piperacillin	2–>128	4	>128
P. rettgeri[a] (20)	CGP 31608	2–8	4	8
	Imipenem	0.25–2	0.5	2
	SCH 34343	0.25–2	0.5	1
	Cefotaxime	≤0.12–2	≤0.12	≤0.12
	Ceftazidime	≤0.12–1	≤0.12	≤0.12
	Aztreonam	≤0.12–0.5	≤0.12	≤0.12
	Piperacillin	0.5–>128	2	>128
Salmonella spp.[a] (25)	CGP 31608	4–8	4	4
	Imipenem	≤0.12–0.25	0.12	0.25
	SCH 34343	0.25–1	0.25	0.5
	Cefotaxime	≤0.12–0.25	≤0.12	0.25
	Ceftazidime	≤0.12–0.5	0.5	
	Aztreonam	≤0.12–0.25	≤0.12	0.25
	Piperacillin	8–>128	32	>128

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TABLE 1—*Continued*

Organism (no. of isolates)	Agent	MIC (μg/ml)		
		Range	50%	90%
Shigella spp.[a] (25)	CGP 31608	4–8	4	4
	Imipenem	≤0.12–0.5	0.12	0.5
	SCH 34343	0.12–1	0.25	0.25
	Cefotaxime	≤0.12–0.25	≤0.12	0.5
	Ceftazidime	≤0.12–0.25	0.12	0.5
	Aztreonam	≤0.12–0.25	≤0.12	0.25
	Piperacillin	8–>128	32	>128
A. hydrophila[a] (10)	CGP 31608	1–4	1	4
	Imipenem	≤0.12–0.25	≤0.12	0.25
	SCH 34343	0.12–0.5	0.12	0.5
	Cefotaxime	≤0.12–2	≤0.12	0.5
	Ceftazidime	≤0.12–2	≤0.12	0.25
	Aztreonam	≤0.12–0.5	≤0.12	0.5
	Piperacillin	2–32	8	16
Y. enterocolitica[a] (10)	CGP 31608	1–4	2	4
	Imipenem	≤0.12–0.25	0.12	0.25
	SCH 34343	0.12–0.5	0.25	0.5
	Cefotaxime	≤0.12	≤0.12	≤0.12
	Ceftazidime	≤0.12–0.25	≤0.12	0.25
	Aztreonam	≤0.12–0.5	0.25	0.5
	Piperacillin	0.5–128	4	128
H. influenzae[a] (15)	CGP 31608	0.5–1	0.5	1
	Imipenem	0.25–1	0.5	1
	SCH 34343	0.25–1	0.5	1
	Cefotaxime	≤0.12	≤0.12	≤0.12
	Ceftazidime	≤0.12	≤0.12	≤0.12
	Aztreonam	≤0.12	≤0.12	≤0.12
	Piperacillin	0.5–128	4	32
N. gonorrhoeae[a] (15)	CGP 31608	0.5–2	0.5	1
	Imipenem	0.12–0.5	0.12	0.5
	SCH 34343	0.25–1	0.25	0.5
	Cefotaxime	≤0.12	≤0.12	≤0.12
	Ceftazidime	≤0.12	≤0.12	≤0.12
	Aztreonam	≤0.12	≤0.12	≤0.12
	Piperacillin	0.5–128	4	32
B. catarrhalis[a] (10)	CGP 31608	0.25–1	0.25	1
	Imipenem	≤0.12–32	≤0.12	0.5
	Cefotaxime	≤0.12–0.5	≤0.12	0.5
	Ceftazidime	≤0.12–0.5	≤0.12	0.5
	Aztreonam	≤0.12–0.25	≤0.12	≤0.12
	Piperacillin	0.12–32	0.5	8
P. aeruginosa[a] (57)	CGP 31608	2–16	2	4
	Imipenem	0.5–8	2	4
	SCH 34343	>64	>64	>64
	Ceftazidime	0.5–32	4	16
	Aztreonam	1–64	8	32
	Piperacillin	2–>128	64	>128
	Gentamicin	0.5–>32	4	>32
	Amikacin	1–>64	8	32
P. cepacia[a] (20)	CGP 31608	0.12–4	1	2
	Imipenem	1–>128	4	8
	Ceftazidime	0.12–8	1	4
	Aztreonam	1–64	32	64
	Piperacillin	1–16	4	8
	Gentamicin	1–>32	8	32
P. maltophilia[a] (20)	CGP 31608	32–>128	64	>128
	Imipenem	>128	>128	>128
	Ceftazidime	1–>128	16	64
	Aztreonam	>64	>64	>64

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TABLE 1—*Continued*

Organism (no. of isolates)	Agent	MIC (μg/ml)		
		Range	50%	90%
	Piperacillin	16–>128	128	>128
	Gentamicin	1–>16	16	>16
P. acidovorans[a] (10)	CGP 31608	0.25–1	0.25	0.5
	Imipenem	0.25–1	0.5	0.5
	Ceftazidime	0.5–8	1	4
	Piperacillin	1–>128	16	128
P. fluorescens[a] (10)	CGP 31608	0.5–16	2	8
	Imipenem	0.12–8	1	4
	Ceftazidime	1–8	2	8
	Piperacillin	4–128	4	32
Pseudomonas spp., other[a,b] (10)	CGP 31608	2–8	2	4
	Imipenem	0.5–2	1	2
	Ceftazidime	1–8	4	8
	Piperacillin	4-128	16	128
Acinetobacter spp.[a] (30)	CGP 31608	1–4	2	4
	Imipenem	0.12–4	1	2
	SCH 34343	0.5–8	4	8
	Cefotaxime	4–>32	8	>32
	Ceftazidime	1–>32	8	>32
	Aztreonam	8–>32	16	>32
	Piperacillin	4–>128	16	32

[a] Resistant to ampicillin.
[b] Includes *P. putida*, *P. stutzeri*, and *P. diminuta*.

but the JK group *Corynebacterium* organisms were resistant, as they were to the other agents.

CGP 31608 inhibited anaerobic organisms at <4 μg/ml. It was more active than cefoxitin, cefotaxime, or piperacillin against *Bacteroides* species. Its MICs for *Clostridium difficile* were 4 and 8 μg/ml, compared with 1 and 2 μg/ml for *C. perfringens*. Against the peptococci, peptostreptococci, fusobacteria, and eubacteria, imipenem MICs generally were lower.

Effect of assay conditions. The in vitro activity of CGP 31608 against 30 members of the family *Enterobacteriaceae* and 30 *P. aeruginosa*, 30 *S. aureus*, *S. epidermidis*, and *S. faecalis* strains did not differ more than twofold when determined in Mueller-Hinton, brain heart infusion, and Columbia agar. Its MICs for *S. aureus* and members of the family *Enterobacteriaceae* determined under anaerobic conditions were within a factor 2 of the MICs determined aerobically. Broth dilution MICs for *S. aureus*, *E. coli*, *K. pneumoniae*, *S. marcescens*, *M. morganii*, and *P. aeruginosa* (five isolates of each species) were within a factor 2 of the agar MIC results.

The CGP 31608 MBCs were within a factor 4 of the MICs for *E. coli*, *K. pneumoniae*, *E. cloacae*, *S. marcescens*, *P. vulgaris*, and *P. mirabilis* (five isolates of each tested) (Table 3). There was a fourfold increase in MBC for methicillin-susceptible *S. aureus* strains and a fourfold increase in MBC for methicillin-resistant isolates. For *P. aeruginosa*, the MBCs were eightfold higher than the MICs. Increasing the inoculum size from 10^5 to 10^7 CFU did not increase the agar MICs for *S. aureus*, *E. coli*, *K. pneumoniae*, *E. cloacae*, *P. mirabilis*, *S. marcescens*, and *P. vulgaris* (five isolates each), and for only four of 30 isolates of *P. aeruginosa* was there a fourfold increase in MICs at 10^7 CFU compared with 10^5 CFU.

The effect of serum upon the activity of CGP 31608 was determined for two isolates each of *S. aureus*, *E. coli*, *K. pneumoniae*, *E. cloacae*, and *P. aeruginosa*. In 50% normal human serum, the MICs and MBCs were identical or within a factor 2 lower or higher than the MICs and MBCs determined in Mueller-Hinton broth. For the same strains, the MICs determined in sterile filtered urine (pH 5.5) were within a factor 2 of the MICs determined in broth (Table 4).

Permeability studies. The role of permeability in the activity of CGP 31608 was analyzed by using beta-lactam-permeable strains of *P. aeruginosa* and *E. coli* obtained from Zimmermann (17) and Richmond et al., respectively (12). For *P. aeruginosa*, there was a fourfold difference in CGP 31608 MICs between the parent and the mutant (Table 5). MICs for the permeable *E. coli* strains were the same as that for the parent strain.

Activity against strains possessing characterized beta-lactamases. At 4 μg/ml, CGP 31608 inhibited strains of *P. aeruginosa* which contained PSE-1, PSE-2, PSE-3, and PSE-4 beta-lactamases. In contrast, the cefotaxime MICs exceeded 64 μg/ml for these isolates, and the MIC of cefoperazone for the PSE-4-containing strain was 128 μg/ml. CGP 31608 had an MIC of 4 μg/ml for *E. cloacae* P99, whereas the MICs of cefotaxime, ceftazidime, and aztreonam exceeded 32 μg/ml. The *K. oxytoca* containing the K-1 beta-lactamase for which the aztreonam MIC was 32 μg/ml was inhibited by 4 μg of CGP 31608 per ml, and an *E. coli* that had a high copy number of the plasmid coding for

TABLE 2. Comparative activity of CGP 31608 and other agents against gram-positive and anaerobic species

Organism (no. of isolates)	Agent	MIC (μg/ml)		
		Range	50%	90%
S. aureus, methicillin susceptible (32)	CGP 31608	≤0.03–0.5	0.12	0.5
	Imipenem	≤0.03–0.25	0.3	0.25
	SCH 34343	0.03–0.5	0.06	0.5
	Cefotaxime	0.5–4	1	4
S. aureus, methicillin resistant (22)	CGP 31608	0.12–2	0.12	2
	Imipenem	0.5–16	1	16
	SCH 34343	0.25–16	1	16
	Cefotaxime	16–>32	>32	>32
S. epidermidis, methicillin susceptible (26)	CGP 31608	0.06–0.5	0.12	0.25
	Imipenem	≤0.03–0.25	0.06	0.12
	SCH 34343	≤0.03–0.25	0.12	0.25
	Cefotaxime	0.25–8	2	8
S. epidermidis, methicillin resistant (23)	CGP 31608	0.12–1	0.12	0.25
	Imipenem	0.12–8	2	8
	SCH 34343	0.12–8	2	8
	Cefotaxime	>32	>32	>32
Streptococcus pyogenes (25)	CGP 31608	0.12–1	0.25	0.5
	Imipenem	≤0.03	≤0.03	≤0.03
	SCH 34343	≤0.03	≤0.03	≤0.03
	Cefotaxime	≤0.03	≤0.03	≤0.03
S. agalactiae (20)	CGP 31608	0.12–0.5	0.12	0.5
	Imipenem	≤0.03	≤0.03	≤0.03
	SCH 34343	≤0.03	≤0.03	≤0.03
	Cefotaxime	≤0.03–1	0.06	0.25
Streptococcus group C, F, G (30)	CGP 31608	0.03–1	0.25	1
	Imipenem	0.03–0.5	0.12	0.5
	SCH 34343	0.03–0.5	0.12	0.5
	Cefotaxime	0.03–1	0.12	0.5
S. bovis (20)	CGP 31608	0.12–0.5	0.5	0.5
	Imipenem	≤0.12	≤0.12	≤0.12
	SCH 34343	≤0.12	≤0.12	≤0.12
	Cefotaxime	≤0.12	≤0.12	≤0.12
S. faecalis (20)	CGP 31808	16–32	16	32
	Imipenem	0.5–8	0.5	1
	SCH 34343	2–8	4	8
	Cefotaxime	>32	>32	>32
S. pneumoniae (17)[a]	CGP 31608	≤0.12–2	0.12	2
	Imipenem	≤0.03–2	≤0.03	2
	SCH 34343	≤0.03–2	≤0.03	2
	Cefotaxime	≤0.03–1	≤0.03	1
Viridans group streptococci (18)	CGP 31608	0.25–1	0.5	0.5
	Imipenem	≤0.03–1	≤0.03	1
	SCH 34343	0.03–4	1	4
	Cefotaxime	≤0.03–1	0.06	0.25
L. monocytogenes (20)	CGP 31608	<0.03–4	0.5	1
	Imipenem	≤0.03–4	0.06	1
	SCH 34343	≤0.03–4	0.03	1
	Cefotaxime	8–>32	8	>32
Corynebacterium JK group (10)	CGP 31608	>32	>32	>32
	Imipenem	>32	>32	>32
	SCH 34343	>32	>32	>32
	Cefotaxime	>32	>32	>32
Clostridium spp.[b] (20)	CGP 31608	1–8	2	4
	Imipenem	≤0.03–4	0.03	1
	SCH 34343	≤0.03–2	0.03	2
	Cefotaxime	0.25–64	2	4

Continued on following page

TABLE 2—*continued*

Organism (no. of isolates)	Agent	MIC (μg/ml)		
		Range	50%	90%
B. fragilis (50)	CGP 31608	1–8	1	4
	Imipenem	0.03–1	0.03	0.25
	SCH 34343	0.03–1	0.06	0.25
	Cefotaxime	8–>64	>64	>64
	Cefoxitin	2–>32	8	64
	Piperacillin	8–>128	64	>128
Bacteroides spp., other[c] (25)	CGP 31608	0.5–8	1	2
	Imipenem	0.03–2	0.03	0.5
	SCH 34343	0.03–1	0.03	0.5
	Cefotaxime	4–>64	32	>64
	Cefoxitin	4–64	16	64
	Piperacillin	8–>128	32	>128
Eubacterium spp. (10)	CGP 31608	0.5–1	0.5	1
	Imipenem	<0.03–0.25	0.12	0.12
Peptococci (10)	CGP 31608	0.5–1	0.5	1
	Imipenem	≤0.03–0.25	≤0.03	0.12
Peptostreptococci (10)	CGP 31608	0.12–1	0.12	0.5
	Imipenem	≤0.03–0.25	0.06	0.12
Propionibacterium spp. (10)	CGP 31608	0.06–0.5	0.25	0.5
	Imipenem	≤0.03	≤0.03	≤0.03
Fusobacterium spp. (10)	CGP 31608	0.5–1	0.5	1
	Imipenem	0.03–0.5	0.03	0.5

[a] Includes penicillin-resistant (MIC, > 1 μg/ml) isolates.
[b] Includes *C. perfringens*, *C. tetani*, *C. novyi*, *C. difficile*, and *C. ramosum*.
[c] Includes *B. thetaiotaomicron*, *B. melaninogenicus*, *B. bivius*, *B. disiens*, *B. ovatus*, *B. vulgatus*, and *B. distasonis*.

TEM-2 showed a cefoperazone MIC of >128 μg/ml but was inhibited by 4 μg of CGP 31608 per ml.

Beta-lactamase stability and inhibitory activity. CGP 31608 was not hydrolyzed by the majority of plasmid and chromosomal beta-lactamases (Table 6). The V_{max} for cephaloridine with these enzymes ranged from 15 to 7,560 μmol/min per μg of protein.

CGP 31608 inhibited the hydrolysis of nitrocefin by a variety of beta-lactamases (Table 7). It showed poor inhibition of the K-1, PSE-1, and PSE-4 beta-lactamases. CGP 31608 was an effective inhibitor of the chromosomal type Ia P99 beta-lactamase compared with cefotaxime and imipenem but a much less effective inhibitor of TEM-1, the plasmid enzyme, than clavulanic acid (Table 8).

Beta-lactamase induction. The effect of exposure to CGP 31608 of single isolates of *P. aeruginosa*, *E. cloacae*, *C. freundii*, and *Providencia rettgeri* was determined. The beta-lactamase activity was determined by the nitrocefin assay (Table 9). There was an increase in beta-lactamase

TABLE 3. Comparison of CGP 31608 MICs and MBCs and the effect of inoculum size[a]

Organism	MIC (μg/ml) at[b]:		MBC (μg/ml) at:	
	10^5 CFU	10^7 CFU	10^5 CFU	10^7 CFU
S. aureus	0.25	0.5	0.25	1
S. aureus (methicillin resistant)	0.25	0.5	1	4
E. coli	4	4	4	4
K. pneumoniae	4	4	4	4
S. marcescens	8	8	8	8
P. vulgaris	4	4	4	4
P. mirabilis	4	4	4	4
P. aeruginosa	2	2	4	32

[a] Determined in Mueller-Hinton broth.
[b] Median MIC for five of six isolates of each species. Median of 30 *P. aeruginosa* isolates for MICs and 10^5 and 10^7 CFU.

TABLE 4. Activity of CGP 31608 in serum and urine compared with broth[a]

Organism[b]	Broth		Serum		Urine	
	MIC	MBC	MIC	MBC	MIC	MBC
S. aureus (methicillin resistant)	0.12	0.12	0.06	0.12	0.12	0.12
S. aureus	0.06	0.06	0.06	0.06	0.06	0.06
E. coli	2	2	1	2	2	2
E. coli	4	4	4	4	4	4
K. pneumoniae	4	4	4	4	2	4
K. pneumoniae	4	4	2	4	4	4
E. cloacae	4	8	4	4	4	8
E. cloacae	8	8	4	8	8	8
P. aeruginosa	4	4	2	2	4	4
P. aeruginosa	2	2	2	4	2	4

[a] Broth was Mueller-Hinton, serum was 50% pooled heat-inactivated normal serum, and urine was pooled and filtered with a 0.45-μm-pore-size Millipore filter. MICs and MBCs one expressed in micrograms per millilter.
[b] All organisms were beta-lactamase producers. Two isolates of each strain were used.

TABLE 5. Activity of CGP 31608 against permeability mutants of *E. coli* and *P. aeruginosa*

Organism	MIC (μg/ml)					
	CGP 31608	Imipenem	SCH 34343	Ticarcillin	Cefotaxime	Aztreonam
E. coli						
Parent	8	0.5	0.25	8	0.125	0.25
Mutants						
DC 1	4	0.5	0.25	4	0.015	0.03
DC 2	8	0.25	0.25	0.5	0.015	0.03
DC 3	8	0.5	0.25	4	0.05	0.125
DC 13	8	0.5	0.25	4	0.03	0.03
P. aeruginosa						
Parent	1	1	NT[a]	4	16	1
Mutant	0.125	0.125	NT	0.25	4	0.06

[a] NT, Not tested since inactive against *P. aeruginosa*.

TABLE 6. Stability of CGP 31608 to attack by beta-lactamases

Enzyme source	Trivial name	Richmond-Sykes classification[a]	Relative hydrolysis rate[b]			
			CGP 316-8	Imipenem	Cefotaxime	Aztreonam
E. coli	TEM-1	III	<0.1	<0.1	<0.1	<0.1
E. coli	TEM-2	III	<0.1	<0.1	<0.1	<0.1
K. pneumoniae	SHV-1	III	<0.1	<0.1	<0.1	<0.1
K. pneumoniae	HMS-1	III	<0.1	<0.1	<0.1	<0.1
P. aeruginosa	OXA-2	V	<0.1	<0.1	3	<0.1
P. aeruginosa	PSE-1	V	<0.1	<0.1	<0.1	<0.1
P. aeruginosa	PSE-4	V	<0.1	<0.1	1	<0.1
P. aeruginosa	PSE-2	V	<0.1	<0.1	5	1
E. cloacae	P99	Ia	<0.1	<0.1	1	<0.1
M. morganii		Ia	<0.1	<0.1	3	<0.1
P. vulgaris		Ic	<0.1	<0.1	42	<0.1
P. penneri		Ic	<0.1	<0.1	21	<0.1
P. aeruginosa	Sabath-Abraham	Id	<0.1	<0.1	<0.1	<0.1
K. oxytoca	K-1	IV	<0.1	<0.1	3	12
P. mirabilis		II	<0.1	<0.1	<0.1	<0.1
E. coli	CEP-1	III	<0.1	<0.1	<0.1	<0.1
E. coli	Carb 3	V	<0.1	<0.1	<0.1	<0.1
S. aureus			<0.1	<0.1	<0.1	<0.1
B. catarrhalis			<0.1	<0.1	<0.1	<0.1

[a] Reference 13.
[b] Rate relative to cephaloridine at 100.

TABLE 7. Inhibition of the hydrolysis of beta-lactamases by CGP 31608[a]

Beta-lactamase name	Source organism	Richmond-Sykes classification	% Inhibition of nitrocefin hydrolysis
TEM-1	*E. coli*	IIIa	76
SHV-1	*Klebsiella* spp.	IIIa	95
P99	*E. cloacae*	Ia	99
–[b]	*M. morganii*	Ia	80
–	*P. vulgaris*	Ic	99
Sabath-Abraham	*P. aeruginosa*	Id	86
K-1	*Klebsiella* spp.	IV	17
PSE-1	*P. aeruginosa*	V	0
PSE-4	*P. aeruginosa*	V	0
OXA-2	*P. aeruginosa*	V	93
–	*P. penneri*	Ic	97

[a] CGP 31608 was preincubated with the enzymes for 10 min before the nitrocefin was added. Both CGP 31608 and nitrocefin were present at 100 μM.
[b] –, No trivial name.

activity for all isolates, but the increase was lowest with *E. cloacae*.

Selection of resistant mutants. The CGP 31608 MICs were 2 to 8 μg/ml for seven *E. coli*, four *K. pneumoniae*, six *K. oxytoca*, three *P. vulgaris*, seven *C. freundii*, four *E. cloacae*, seven *M. morganii*, six *P. aeruginosa*, six *S. marcescens*, and five *S. aureus* strains. No organisms were isolated with four- or eightfold greater MICS. Exposure of parent and mutant isolates to cefotaxime, ceftazidime, and aztreonam caused selection of strains derepressed for beta-lactamase

TABLE 8. I_{50} of CGP 31608 and other beta-lactams against the P99 and TEM-1 beta-lactamases

Beta-lactamase	I_{50}[a] (μg/ml)				
	CGP 31608	Imipenem	SCH 3343	Cefotaxime	Clavulanic acid
P99	0.15	0.7	0.4	0.35	5.7
TEM-1	14.2	6.4	6.7	5.7	0.02

[a] I_{50}, 50% inhibitory concentration.

TABLE 9. Effect of CGP 31608 on beta-lactamase production

Organism	Relative increase in beta-lactamase activity at following concn (μg/ml)[a]:		
	1	4	8
P. aeruginosa	15.8	11.4	15.2
E. cloacae	1.5	1.5	3.2
C. freundii	18.4	12.2	32.2
P. rettgeri	91.3	82.3	123.7

[a] Ratio of beta-lactamase activity in induced cells to activity in uninduced cells. Beta-lactamase activity was determined by the nitrocefin assay and calculated as international units per milligram of protein per milliliter.

production. These organisms were tested against CGP 31608 and imipenem (Table 10). The MICs of both CGP 31608 and imipenem remained identical in comparison with the increase in the MIC of the selecting agent.

Killing data and postantibiotic suppression. The killing activity of CGP 31608 was studied with a beta-lactamase-producing *E. coli* strain resistant to ampicillin, cefazolin, cefoperazone, and trimethoprim and with a *P. aeruginosa* strain resistant to carbenicillin and gentamicin. The CGP 31608 MIC was 4 μg/ml and the MBC was 8 μg/ml; the imipenem MIC was 0.25 μg/ml and the MBC was 0.5 μg/ml for the *E. coli* strain. Exposure for 1 h to CGP 31608 produced a decrease in CFU comparable to the decrease with imipenem (Table 11). CGP 31608 produced a kill effect at a concentration only twice the MBC, which was comparable to that with imipenem 30-fold above the MBC. A 2-h exposure at 15 μg/ml caused a major decrease in CFU for CGP 31608 and imipenem. With the *P. aeruginosa* strain for which the CGP 31608 MBC was 16 μg/ml and the imipenem MBC was 4 μg/ml, there were equal decreases in CFU for CGP 31608 compared with imipenem.

The postantibiotic effect (PAE) of CGP 31608 and imipenem were also studied for the above organisms. A PAE was found for *E. coli* (Table 12). We did not find a PAE for the *P. aeruginosa* strain when the exposure was for 1 h, and a short PAE compared with the PAE for imipenem.

Synergy studies. The effect of combination of CGP 31608 and azlocillin, gentamicin, rifampin, and ciprofloxacin was studied with *P. aeruginosa*, *E. cloacae*, and *S. marcescens* (Table 13). Antagonism was seen only for azlocillin with one *S. marcescens* and one *P. aeruginosa* strain. Gentamicin was most likely to act synergistically with CGP 31608, ciprofloxacin was less likely, and rifampin and azlocillin were primarily indifferent, as tested against *P. aeruginosa* and *S. marcescens*.

DISCUSSION

The first penem was synthesized in 1975 (3, 16), but the compound was destroyed by beta-lactamases, probably because of the type of acyl side chain present on C-6. Penems were conceived as compounds that would combine properties of penams and cephems, in which the reactivity produced by the double bond between C-3 and C-4 of the cephem would be present in a five-membered ring. The knowledge gained from the analysis of the naturally occurring carbapenem thienamycin showed that introduction of a hydroxyethyl substituent at C-6 would yield a chemically stable, beta-lactamase-resistant agent (5, 8).

To date, only a few penems have been extensively investigated despite the plethora of compounds found in the patent literature. The well-known penems are SCH 29482, SCH 34343, FCE 22101, and FCE 22891 (1, 4, 7, 10, 11, 15). These agents have been demonstrated to inhibit a wide range of aerobic and anaerobic organisms (1, 7, 10, 11, 15) but lack the activity against *P. aeruginosa* which the carbapenem imipenem possesses.

TABLE 11. Cumulative log kill of CGP 31608 and imipenem against *E. coli* and *P. aeruginosa*

Organism	Antibiotic	Concn (μg/ml)	Decrease in log CFU/ml at:	
			1 h	2 h
E. coli 8340[a]	CGP 31608	15	1.8	2.7
		30	1.8	ND[b]
	Imipenem	15	2.2	4
		30	3.9	ND
P. aeruginosa 9209[c]	CGP 31608	15	1.4	1.2
		30	1.4	ND
	Imipenem	15	1.5	1.6
		30	1.6	ND

[a] CGP 31608 MBC, 8 μg/ml; imipenem MBC, 0.5 μg/ml.
[b] ND, Not determined.
[c] CGP 31608 MBC, 16 μg/ml; imipenem MBC, 4 μg/ml.

TABLE 12. PAE of CGP 31608 and imipenem against *E. coli* and *P. aeruginosa*

Organism	Antibiotic	Concn (μg/ml)	PAE (h) after exposure for:	
			1 h	2 h
E. coli 8384[a]	CGP 31608	15	1	2.5
		30	1.9	ND[b]
	Imipenem	15	3.2	3.6
P. aeruginosa 9209[c]	CGP 31608	15	0.8	0.5
		30	0.5	ND
	Imipenem	15	0.5	3.2
		30	1.4	ND

[a] CGP 31608: MIC, 4 μg/ml; MBC, 8 μg/ml. Imipenem: MIC, 0.25 μg/ml; MBC, 0.5 μg/ml.
[b] ND, Not determined.
[c] CGP 31608: MBC, 16 μg/ml; imipenem: MBC, 4 μg/ml.

TABLE 10. Activity of CGP 31608 against bacteria constitutively producing high-level beta-lactamase

Organism	MIC (μg/ml)[a]									
	CGP 31608		Imipenem		Cefotaxime		Ceftazidime		Aztreonam	
	P	M	P	M	P	M	P	M	P	M
P. aeruginosa	4	4	4	4	32	>128	2	32	4	32
E. cloacae	4	4	4	4	2	64	2	64	2	64
C. freundii	4	4	2	2	1	32	1	32	1	32
M. morganii	8	8	4	4	2	16	1	16	0.5	8
K. oxytoca	4	4	0.5	0.5	1	16	0.5	16	0.5	16

[a] P, Parent; M, mutant isolate.

TABLE 13. Effect of combination of CGP 31608 with other antimicrobial agents

Organism (no. of isolates)	No. of isolates showing effect when CGP 31608 combined with:											
	Azlocillin			Gentamicin			Rifampin			Ciprofloxacin		
	S[a]	I[a]	A[a]	S	I	A	S	I	A	S	I	A
P. aeruginosa (11)	0	10	1	9	2	0	0	11	0	6	5	0
E. cloacae (10)	5	5	0	6	4	0	0	10	0	2	8	0
S. marcescens (11)	0	10	1	5	6	0	1	10	0	0	11	0

[a] S, Synergy; I, indifference; A, antagonism. Method was agar checkerboard.

This study illustrates that CGP 31608 has an extremely broad spectrum of antibacterial activity, inhibiting most members of the family *Enterobacteriaceae* at ≤4 μg/ml, including species resistant to ampicillin, cefazolin, cefoperazone, and piperacillin. It also has excellent activity (MIC_{90}, 4 μg/ml) against *P. aeruginosa*, including selected isolates resistant to gentamicin, amikacin, piperacillin, ceftazidime, and aztreonam.

In general, the MIC_{90}s of cefotaxime, ceftazidime, and aztreonam for the highly susceptible members of the family *Enterobacteriaceae* are lower (0.25 μg/ml) than those of CGP 31608 (2 to 4 μg/ml), and imipenem usually is fourfold more active than CGP 31608. Conversely, CGP 31608 and imipenem both inhibit isolates of *E. cloacae*, *C. freundii*, *S. marcescens*, and *P. aeruginosa* at 4 to 8 μg/ml, although the above agents have MICs in the range of 16 to 64 μg/ml.

The gram-positive activity of CGP 31608 is impressive and comparable to the published activity of cephalosporins and isoxazolyl penicillins against staphylococci and streptococci (4), and CGP 31608 is more active than cefotaxime against staphylococci and minimally less active against streptococci. Of particular note is its activity against methicillin-resistant *S. aureus* and *S. epidermidis* strains. It inhibits isolates resistant to imipenem. This is a new aspect of these agents. The mechanism is unknown and could be due to failure to induce a new penicillin-binding protein 2′ or to affinity for the new enzymes. Studies are in progress to determine this. Unlike imipenem, CGP 31608 does not inhibit enterococci, *S. faecalis*, or *S. faecium*, and the explanation for this is unknown.

Similarly to the other penems and to carbapenems, CGP 31608 has excellent in vitro activity against anaerobic cocci and bacilli and inhibits isolates resistant to cefoxitin, cefotaxime, and piperacillin. As with all beta-lactams, MICs against certain anaerobes, *C. difficile* and *Bacteroides theataiotaomicron*, are higher, i.e., 4 to 8 μg/ml, than against *C. perfringens* and *B. fragilis*.

CGP 31608 has excellent beta-lactamase stability with both chromosomal and plasmid enzymes. Although it induces beta-lactamase activity as do other penems, cephamycins, carbapenems and clavams, the induced strains remain susceptible to the compound. Furthermore, CGP 31608 does not select isolates which constitutively produce high levels of beta-lactamase as do certain cephalosporins (14). CGP 31608 inhibits beta-lactamase-producing isolates selected by exposure to other beta-lactams.

CGP 31608 showed a PAE for *E. coli* at concentrations that would be achieved in humans (O. Zak, personal communication), but the effect was minor with *P. aeruginosa*. Higher concentrations may demonstrate the effect similar to that noted with imipenem tested against *P. aeruginosa*. It is also possible that a study of a larger number of isolates will show the effect.

Overall, these studies demonstrate the excellent in vitro properties of this novel penem. Further pharmacological and clinical studies will establish the clinical utility of this agent in comparison with other beta-lactams. Clearly, this penem has very impressive in vitro activity against staphylococci, including methicillin-resistant bacteria, and against *P. aeruginosa*, and this would make it of interest in an era of increased problems with these species.

LITERATURE CITED

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