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In Vitro Activities of 15 Oral β-Lactams against *Klebsiella pneumoniae* Harboring New Extended-Spectrum β-Lactamases

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The activities of 15 oral β-lactams against *Klebsiella pneumoniae* harboring new extended-spectrum β-lactamases were studied. All compounds were affected by these enzymes, especially by the SHV derivatives. Except for ceftibuten, the compounds with the greatest intrinsic activity were more affected by the presence of these enzymes than were older compounds with moderate intrinsic activity.

Bacteria harboring new extended-spectrum β-lactamases derived by mutation (11, 13) from TEM-1, TEM-2, or SHV-1 have been described in Europe, the United States, South America, North Africa, Japan, China, and Malaysia (1–8, 10, 11). The activities of these enzymes against β-lactam antibiotics has been widely studied, although there are only a few reports concerning the new oral β-lactams. The aim of this study was to assess the in vitro activities of 15 oral β-lactams in comparison with reference β-lactams against bacteria producing some of these new enzymes.

Thirty-nine clinical isolates of *Klebsiella pneumoniae* containing TEM-3 (23 strains), TEM-7 (4 strains), SHV-3 (4 strains), SHV-4 (5 strains), and SHV-5 (3 strains) were compared with the reference strains *K. pneumoniae* 2222 (4), *Escherichia coli* C_1a Nal^r (9), and *E. coli* C_1a Nal^r containing the reference β-lactamases TEM-1 (R111), TEM-2 (RP4), TEM-3 (pCF204), TEM-4 (pUD16), TEM-7 (pIF100), SHV-1 (R1010), SHV-2 (pMG229), SHV-3 (pUD18), SHV-4 (pUD21), and SHV-5 (pAFF2) (11).

Standard antimicrobial reference powders were obtained

TABLE 1. MICs of oral cephalosporins against *E. coli* C_1a Nal^r transconjugants containing reference β-lactamases

Drug	MIC (μg/ml) against *E. coli*:									
	C_1a Nal^r	C_1a Nal^r transconjugants containing:								
		TEM-1	TEM-2	TEM-3	TEM-4	TEM-7	SHV-1	SHV-2	SHV-3	SHV-4	SHV-5
Cephalexin	4	8	8	16	16	8	8	32	256	32	128
Cephradine	8	16	16	32	16	16	16	32	128	32	128
Cefadroxil	8	8	8	16	16	8	8	32	64	32	128
Cefuroxime	4	4	4	64	64	4	16	16	32	8	32
Cefaclor	1	4	8	32	32	2	64	32	128	32	128
Cefatrizine	1	8	32	64	64	8	64	64	128	64	258
BMY 28100	2	4	64	64	64	4	64	>256	256	64	256
Cefcanel	2	16	32	64	32	8	16	128	64	32	128
Cefotiam	0.25	0.25	0.5	2	2	0.5	1	8	16	2	8
Cefixime	0.25	0.25	0.25	4	8	0.25	0.25	1	1	8	32
Cefetamet	0.25	0.25	0.25	4	8	0.25	1	1	2	1	2
Tigemonam	0.25	0.5	1	32	64	8	32	2	32	64	256
Cefpodoxime	0.25	0.5	0.5	64	32	1	8	8	32	8	32
Loracarbef	0.5	2	2	8	16	1	8	32	64	8	64
Ceftibuten	0.12	0.12	0.12	0.25	0.25	<0.06	0.5	0.5	0.25	1	8
Cefotaxime	0.6	0.06	0.06	8	4	0.12	0.06	4	4	1	4
Ceftazidime	0.12	0.25	0.5	16	8	8	1	4	8	16	64
Aztreonam	0.06	0.06	0.2	4	4	0.25	0.12	2	2	32	128
Moxalactam	0.06	0.12	0.12	0.5	0.25	0.12	0.25	0.12	0.5	0.12	0.5
Meropenem	0.0078	0.0078	0.0078	0.015	0.0078	0.0078	0.015	0.0078	0.0078	0.0078	0.015
Imipenem	0.06	0.03	0.06	0.25	0.03	0.06	0.06	0.06	0.03	0.03	0.06

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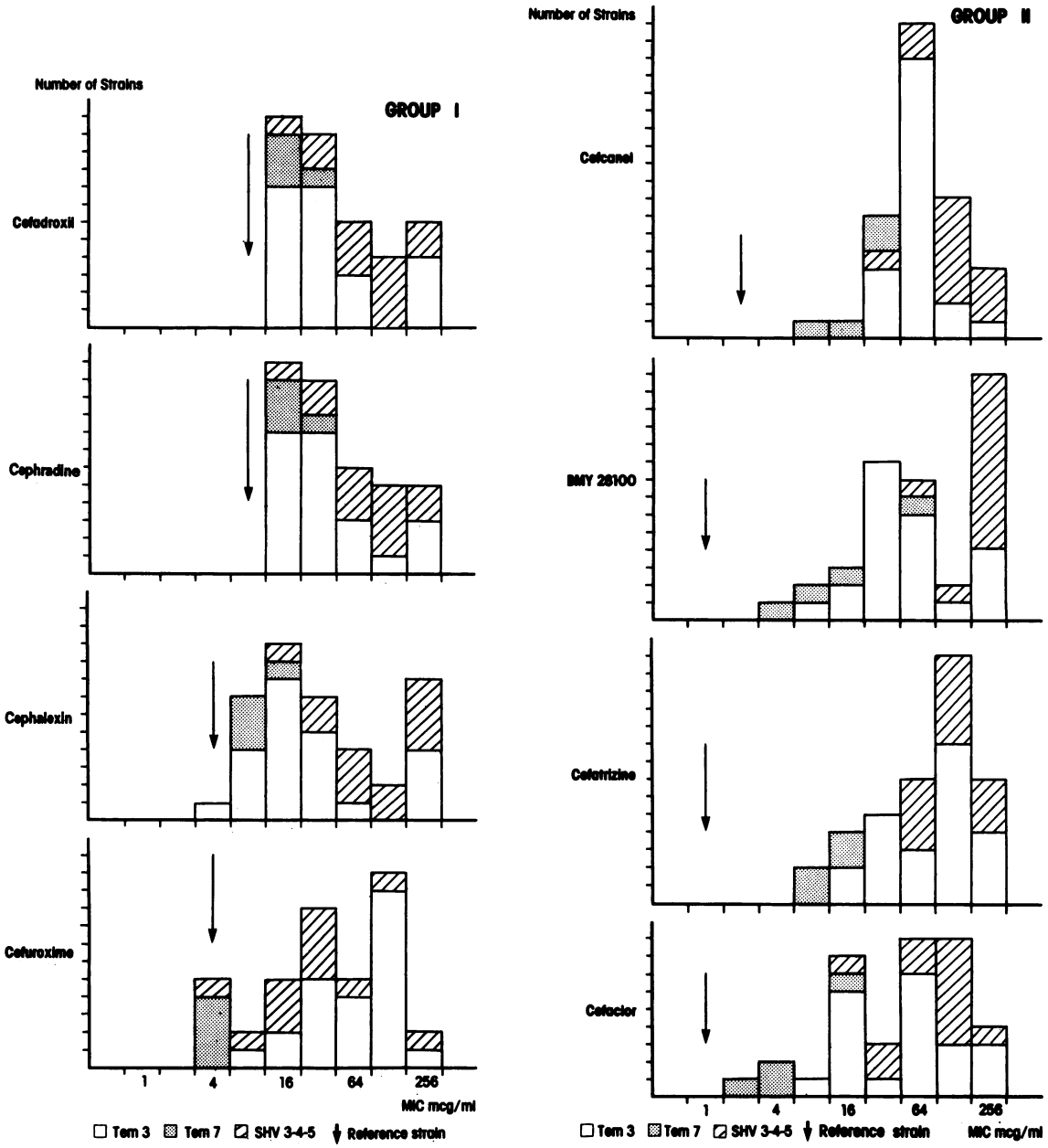

FIG. 1. Susceptibility to the oral cephalosporins cefotaxime, moxalactam, and aztreonam of clinical isolates of *K. pneumoniae* producing extended-spectrum β-lactamases. Four groups (I to IV) were arbitrarily assigned according to the intrinsic activities of the oral cephalosporins against *K. pneumoniae* 2222, with group I possessing the least intrinsic potency and group IV possessing the most.

as follows: cephradine, tigemonam, and aztreonam (Squibb, Paris-La Défense, France); cefuroxime, cephalexin, and ceftazidime (Glaxo, Paris, France); cefadroxil (Bristol-Myers, Paris, France); cefaclor, loracarbef, and moxalactam (Eli Lilly, Saint-Cloud, France); cefatrizine (Allard, Paris, France); cefcanel (Astra, Nanterre, France); BMY 28100 (Bristol-Myers); cefpodoxime (Diamant, Paris-La Défense, France); cefotiam (Takeda, Puteaux, France); cefixime (Pharmuka, Paris, France); cefetamet (Produits Roche, Neuilly-sur-Seine, France); ceftibuten (Schering-Unilabo, Levallois-Perret, France); cefotaxime (Roussel, Paris, France); meropenem (I.C.I. Pharma, Cergy-Pontoise, France); and imipenem (Merck-Sharp & Dohme-Chibret, Paris, France).

MICs were determined on Mueller-Hinton agar (Diagnostics Pasteur, Paris, France) with an inoculum of 10^4 CFU per spot (Steers replicator) after 18 h of incubation at 37°C in air. The antibiotics were separated into four groups according to their intrinsic activities against the reference strain *K. pneumoniae* 2222.

This study was conducted with *K. pneumoniae* because most of the extended-spectrum β-lactamases are currently isolated from nosocomial strains of *K. pneumoniae* (2–4, 7, 8, 10–12). Since these wild-type strains may possess additional mechanisms of resistance (permeability barrier), the MICs of all β-lactams were also tested in an isogenic system with *E. coli* C_1a Nal^r (9) containing different extended-spectrum β-lactamases (Table 1).

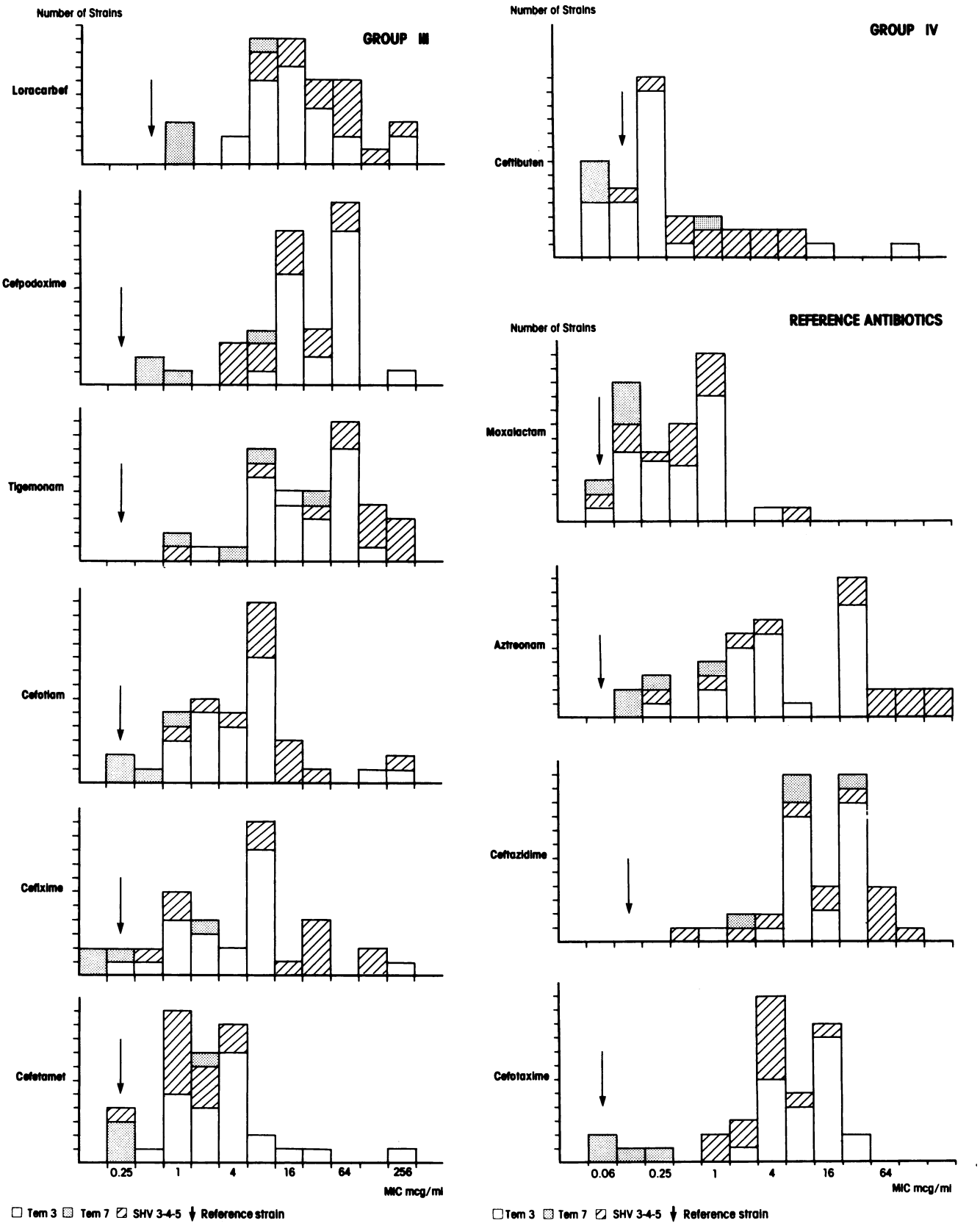

FIG. 1—*Continued*

Figure 1 shows the MICs of the various β-lactams against *K. pneumoniae* 2222 and the clinical isolates of *K. pneumoniae* containing the different extended-spectrum β-lactamases. The oral monobactam, tigemonam, and oral cephalosporins with a 7α-oxyimino or vinyl side chain (cefuroxime, cefixime, cefetamet, cefpodoxime, and ceftibuten) were more active than were other oral cephalosporins. For oral β-lactams except cefuroxime, cefpodoxime, and cefetamet, MICs were generally higher for strains with the SHV derivatives SHV-3, SHV-4, and SHV-5 than for strains with the TEM derivatives TEM-3 and TEM-7. Despite the fourfold-greater intrinsic activities of drugs in group II than of drugs in group I, activities against resistant strains were similar. β-Lactams in group III with similar intrinsic MICs could be divided into two subgroups: (i) loracarbef, cefpodoxime, and tigemonam, which were 32- to 256-fold less active against the resistant strains; and (ii) cefotiam, cefixime, and cefetamet, which were only 4- to 32-fold less active. Ceftibuten, the only member of group IV, was clearly more active than were other β-lactams against *K. pneumoniae* producing TEM derivatives, with virtually no change in the MICs; there was a 4- to 64-fold increase in the MICs against *K. pneumoniae* producing SHV derivatives. This increased activity may have been due to the presence at the C-7 position of a carboxyethylidene radical instead of the methoxyimino substituent that is present in other broad-spectrum cephalosporins.

In comparison with the isogenic *E. coli* strain containing the various extended-spectrum β-lactamases (Table 1), some of the *K. pneumoniae* isolates displayed an unusually high level of resistance (especially for antibiotics in group III; Fig. 1). This was difficult to explain solely by the presence of the enzyme. As inferred from their resistance to cefoxitin (≥16 μg/ml), 13 of the 39 *K. pneumoniae* isolates tested (5 of 23 TEM-3 producers and 7 of 12 SHV derivative producers) were likely to possess an additional mechanism of resistance, i.e., a permeability barrier (4).

Oral β-lactams with an increase in activity of up to 64-fold and an extended spectrum in comparison with that of older cephalosporins have become available. However, increased activity against susceptible strains does not always correlate with activity against strains producing extended-spectrum β-lactamases. Despite relatively low MICs, often within the susceptibility range, the activities of these antibiotics still remain to be demonstrated in vivo.

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