COMPARATIVE *IN VITRO* ACTIVITY OF LY 127935 (6059-S), SEVEN CEPHALOSPORINS, THREE AMINOGLYCOSIDES, CARBENICILLIN, AND TICARCILLIN

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LY 127935 (6059-S), a new semi-synthetic beta-lactam antibiotic was tested simultaneously with 6 cephalosporins, 3 aminoglycosides, carbenicillin and ticarcillin against 398 clinical isolates of Gram-negative bacilli and Gram-positive cocci. Many of the organisms were selected for study because of known resistance to one or more of the clinically available antibiotics tested. *Escherichia coli, Klebsiella, Serratia* and *Providencia* were susceptible to LY 127935. Some resistant strains of *Enterobacter, Proteus, Pseudomonas aeruginosa* and *Acinetobacter* were also resistant to LY 127935, but many of the strains resistant to other antibiotics were susceptible to LY 127935. The activity of LY 127935 against *Staphylococcus aureus, Streptococcus pneumoniae, Streptococcus pyogenes, Streptococcus viridans* and *Streptococcus bovis* was similar to that of cephalexin and cephradine. LY 127935 was not active against methicillin-resistant *S. aureus* nor enterococcus.

LY 127935 (6059-S) is a new semi-synthetic beta-lactam antibiotic. It differs from the cephalosporin antibiotics in the replacement of sulfur atom in the six-membered dihydrothiazine ring of the cephalosporin nucleus by an oxygen atom.[2] This compound was synthesized in the Shionogi Research Laboratories of Osaka, Japan. Preliminary studies indicated that this compound has wide antimicrobial spectrum including organisms that are resistant to the cephalosporins, *e. g.* indole-positive *Proteus* species, *Enterobacter* species, *Serratia marcescens, Pseudomonas aeruginosa* and *Bacteroides fragilis* (T. YOSHIDA, M. NARISADA, S. MATSUURA, W. NAGATA & S. KUWAHARA. Program Abstr. 18th Intersci. Conf. Antimicrob. Agents & Chemother. Atlanta, Georgia, Abstr. no. 151, 1978). Studies in animals showed that the pharmacology of this compound was similar to that of cefazolin but its protein binding was about half of cefazolin (S. MATSUURA, T. YOSHIDA, K. SUGENO, Y. HARADA, M. HARADA & S. KUWAHARA. Program Abstr. 18th Intersci. Conf. Antimicrob. Agents & Chemother. Atlanta, Georgia, Abstr. no. 152, 1978).

In order to put this compound in perspective, especially in its relative usefulness among the many clinically available antibiotics, we compared the *in vitro* activity of LY 127935 (6059-S) with seven cephalosporins, three aminoglycosides, carbenicillin and ticarcillin against many species of Gram-positive cocci and Gram-negative bacilli, including many organisms that were known to be resistant to one or more of the following: cephalosporin, gentamicin, tobramycin, amikacin, carbenicillin and ticarcillin.[4~7]

Materials and Methods

Organisms

Three hundred ninety eight clinical isolates were studied. Many of these isolates were selected for

testing because of their known resistance to one or more antibiotics used in this study. The organisms studied were *Escherichia coli* (40 strains), *Klebsiella* species (40 strains), *Enterobacter* species (40 strains), *Serratia* species (40 strains), *Proteus* species (41 strains), *Providencia* species (17 strains), *Acinetobacter* species (11 strains), *Pseudomonas aeruginosa* (40 strains), *Staphylococcus aureus* (60 strains including 10 methicillin-resistant strains), *Streptococcus pneumoniae* (35 strains), *Streptococcus pyogenes* (14 strains), *Streptococcus viridans* and *Streptococcus bovis* (10 strains), and enterococcus (10 strains).

Antimicrobial Susceptibility Testing

The minimal inhibitory concentration (MIC) of LY 127935 (6059-S), cephalothin, cephapirin, cefazolin, cefamandole, cephalexin, cephradine, carbenicillin, ticarcillin, gentamicin, tobramycin, and amikacin for a given strain of organism was determined concurrently using the same inoculum by the WHO-ICS agar dilution method.[1] Two-fold dilutions of antibiotics from 64 μg/ml to 0.125 μg/ml were incorporated into MUELLER-HINTON agar (supplemented with 5% human erythrocytes for testing *S. pneumoniae*). An agar plate with no antibiotic served as a control. The inoculum was 0.002 ml of a 10^{-2} dilution of an 18-hour culture in MUELLER-HINTON broth, delivered by a Steers replicator to the surface of agar plates.[3] The MIC was the minimal antibiotic concentration that inhibited growth completely or allowed only growth of one colony after incubation at 37°C for 18 hours.

Table 1. Comparative *in vitro* activity of 12 antibiotics against Gram-negative bacilli.

Organism (Number of strains)	Antibiotics	Minimal inhibitory concentrations (μg/ml)			
		Range	MIC_{50}*	MIC_{75}*	MIC_{90}*
E. coli (40)	LY 127935	≤0.125~1	≤0.125	0.25	0.5
	Cephalothin	8~>64	16	32	64
	Cephapirin	4~>64	32	64	>64
	Cefazolin	1~>64	2	8	16
	Cefamandole	0.25~>64	1	16	64
	Cephalexin	4~>64	8	16	32
	Cephradine	4~>64	16	16	32
	Carbenicillin	2~>64	8	>64	>64
	Ticarcillin	2~>64	8	>64	>64
	Gentamicin	1~>64	2	8	16
	Tobramycin	0.5~64	2	16	32
	Amikacin	1~64	4	16	32
Klebsiella (40)	LY 127935	≤0.125~4	0.25	0.25	0.5
	Cephalothin	2~>64	4	32	>64
	Cephapirin	2~>64	8	32	>64
	Cefazolin	1~>64	2	8	>64
	Cefamandole	0.5~>64	1	16	>64
	Cephalexin	4~>64	8	8	>64
	Cephradine	4~>64	16	16	>64
	Carbenicillin	32~>64	>64	>64	>64
	Ticarcillin	32~>64	>64	>64	>64
	Gentamicin	0.5~>64	1	2	32
	Tobramycin	0.5~>64	1	2	32
	Amikacin	1~8	2	2	4
Enterobacter (40)	LY 127935	≤0.125~>64	0.25	4	64
	Cephalothin	4~>64	>64	>64	>64
	Cephapirin	4~>64	>64	>64	>64
	Cefazolin	2~>64	>64	>64	>64
	Cefamandole	1~>64	4	>64	>64
	Cephalexin	8~>64	>64	>64	>64
	Cephradine	4~>64	>64	>64	>64
	Carbenicillin	8~>64	32	>64	>64
	Ticarcillin	2~>64	16	>64	>64
	Gentamicin	0 5~64	2	16	64
	Tobramycin	0 5~>64	2	32	32
	Amikacin	1~16	4	4	8

(to be cont'd)

Table 1. (cont'd)

Organism (Number of strains)	Antibiotics	Minimal inhibitory concentrations (μg/ml)			
		Range	MIC_{50}*	MIC_{75}*	MIC_{90}*
Serratia (40)	LY 127935	≤0.125~8	0.5	1	4
	Cephalothin	64~>64	>64	>64	>64
	Cephapirin	>64	>64	>64	>64
	Cefazolin	32~>64	>64	>64	>64
	Cefamandole	1~>64	64	>64	>64
	Cephalexin	64~>64	>64	>64	>64
	Cephradine	64~>64	>64	>64	>64
	Carbenicillin	1~>64	4	>64	>64
	Ticarcillin	2~>64	16	>64	>64
	Gentamicin	0.25~>64	2	16	64
	Tobramycin	0.5~>64	4	32	64
	Amikacin	1~>64	4	8	32
Proteus (41)	LY 127935	≤0.125~>64	≤0.125	0.25	32
	Cephalothin	2~>64	16	>64	>64
	Cephapirin	2~>64	8	>64	>64
	Cefazolin	2~>64	8	>64	>64
	Cefamandole	≤0.125~>64	2	>64	>64
	Cephalexin	8~>64	16	>64	>64
	Cephradine	8~>64	32	>64	>64
	Carbenicillin	≤0.125~>64	1	16	>64
	Ticarcillin	≤0.125~>64	1	32	>64
	Gentamicin	0.5~>64	4	16	64
	Tobramycin	0.5~>64	4	8	64
	Amikacin	2~>64	16	16	64
Providencia (17)	LY 127935	≤0.125~4	≤0.125	0.25	4
	Cephalothin	64~>64	>64	>64	>64
	Cephapirin	>64	>64	>64	>64
	Cefazolin	>64	>64	>64	>64
	Cefamandole	0.5~>64	4	4	>64
	Cephalexin	64~>64	>64	>64	>64
	Cephradine	64~>64	>64	>64	>64
	Carbenicillin	1~>64	2	8	>64
	Ticarcillin	0.5~>64	1	8	>64
	Gentamicin	2~>64	8	16	32
	Tobramycin	4~>64	16	32	64
	Amikacin	1~>64	4	8	32
P. aeruginosa (40)	LY 127935	4~64	16	16	32
	Cephalothin	>64	>64	>64	>64
	Cephapirin	>64	>64	>64	>64
	Cefazolin	>64	>64	>64	>64
	Cefamandole	>64	>64	>64	>64
	Cephalexin	>64	>64	>64	>64
	Cephradine	>64	>64	>64	>64
	Carbenicillin	64~>64	>64	>64	>64
	Ticarcillin	8~>64	32	>64	>64
	Gentamicin	2~>64	8	>64	>64
	Tobramycin	1~>64	2	8	>64
	Amikacin	4~64	8	16	16
Acinetobacter (11)	LY 127935	2~>64	32	32	64
	Cephalothin	>64	>64	>64	>64
	Cephapirin	>64	>64	>64	>64
	Cefazolin	>64	>64	>64	>64
	Cefamandole	64~>64	>64	>64	>64
	Cephalexin	>64	>64	>64	>64
	Cephradine	>64	>64	>64	>64
	Carbenicillin	16~>64	16	16	64
	Ticarcillin	8~>64	16	16	32
	Gentamicin	0.25~16	1	8	16
	Tobramycin	0.25~16	2	4	4
	Amikacin	1~8	1	8	8

* MIC_{50}, MIC_{75}, MIC_{90}: MIC needed to inhibit 50%, 75% and 90% of strains respectively.

Results

The comparative *in vitro* activity of 12 antibiotics against Gram-negative bacilli is listed in Table 1. All strains of *E. coli, Klebsiella, Serratia* and *Providencia* tested including many strains resistant to the cephalosporins, aminoglycosides, carbenicillin and ticarcillin were susceptible to LY 127935. Some resistant strains of *Enterobacter, Proteus, P. aeruginosa* and *Acinetobacter* were also resistant to LY 127935, but many of the strains resistant to other antibiotics were susceptible to LY 127935.

Table 2. Comparative *in vitro* activity of 12 antibiotics against Gram-positive cocci.

Organism (Number of strains)	Antibiotics	Minimal inhibitory concentration (μg/ml)			
		Range	MIC_{50}*	MIC_{75}*	MIC_{90}*
S. aureus (50)	LY 127935	4~32	8	8	16
	Cephalothin	≤0.125~1	0.25	0.5	0.5
	Cephapirin	≤0.125~2	0.25	0.5	0.5
	Cefazolin	≤0.125~4	0.5	1	2
	Cefamandole	≤0.125~2	1	1	1
	Cephalexin	4~32	8	8	16
	Cephradine	2~32	8	8	16
	Carbenicillin	0.25~64	16	32	64
	Ticarcillin	0.5~32	8	16	32
	Gentamicin	≤0.125~8	1	2	4
	Tobramycin	≤0.125~>64	1	2	4
	Amikacin	2~64	8	16	32
S. aureus (methicillin-resistant) (10)	LY 127935	>64	>64	>64	>64
	Cephalothin	32~>64	>64	>64	>64
	Cephapirin	16~64	32	32	32
	Cefazolin	64~>64	>64	>64	>64
	Cefamandole	8~16	16	16	16
	Cephalexin	>64	>64	>64	>64
	Cephradine	>64	>64	>64	>64
	Carbenicillin	64~>64	>64	>64	>64
	Ticarcillin	>64	>64	>64	>64
	Gentamicin	>64	>64	>64	>64
	Tobramycin	64~>64	>64	>64	>64
	Amikacin	8~>64	64	>64	>64
S. pneumoniae (35)	LY 127935	≤0.125~8	1	1	1
	Cephalothin	≤0.125~0.25	≤0.125	≤0.125	≤0.125
	Cephapirin	≤0.125	≤0.125	≤0.125	≤0.125
	Cefazolin	≤0.125~0.25	≤0.125	≤0.125	≤0.125
	Cefamandole	≤0.125~0.25	≤0.125	≤0.125	≤0.125
	Cephalexin	0.5~2	1	1	2
	Cephradine	≤0.125~2	1	1	2
	Carbenicillin	≤0.125~2	≤0.125	0.25	0.5
	Ticarcillin	≤0.125~2	0.5	0.5	0.5
	Gentamicin	16~64	32	32	64
	Tobramycin	8~>64	64	>64	>64
	Amikacin	32~>64	>64	>64	>64
S. pyogenes (14)	LY 127935	0.5~4	1	1	2
	Cephalothin	≤0.125	≤0.125	≤0.125	≤0.125
	Cephapirin	≤0.125	≤0.125	≤0.125	≤0.125
	Cefazolin	≤0.125	≤0.125	≤0.125	≤0.125
	Cefamandole	≤0.125	≤0.125	≤0.125	≤0.125
	Cephalexin	≤0.125~2	0.5	0.5	0.5
	Cephradine	≤0.125~1	≤0.125	≤0.125	0.25
	Carbenicillin	≤0.125	≤0.125	≤0.125	≤0.125
	Ticarcillin	≤0.125~0.5	≤0.125	≤0.125	0.25
	Gentamicin	16~64	32	32	32
	Tobramycin	16~>64	64	64	64
	Amikacin	64~>64	>64	>64	>64

(to be cont'd)

Table 2. (cont'd)

Organism (Number of strains)	Antibiotics	Minimal inhibitory concentration (μg/ml)			
		Range	MIC_{50}*	MIC_{75}*	MIC_{90}*
S. viridans & *S. bovis* (10)	LY 127935	0.5~4	4	4	4
	Cephalothin	≤0.125~0.25	≤0.125	0.25	0.25
	Cephapirin	≤0.125~0.25	≤0.125	≤0.125	≤0.125
	Cefazolin	≤0.125	≤0.125	≤0.125	≤0.125
	Cefamandole	≤0.125	≤0.125	≤0.125	≤0.125
	Cephalexin	0.5~4	2	4	4
	Cephradine	≤0.125~2	1	2	2
	Carbenicillin	≤0.125~0.5	≤0.125	0.5	0.5
	Ticarcillin	0.25~0.5	0.5	0.5	0.5
	Gentamicin	0.5~64	8	16	32
	Tobramycin	2~>64	16	64	>64
	Amikacin	8~>64	64	>64	>64
Enterococcus (10)	LY 127935	>64	>64	>64	>64
	Cephalothin	16~>64	32	32	32
	Cephapirin	16~64	16	32	32
	Cefazolin	16~>64	32	32	64
	Cefamandole	32~>64	32	64	64
	Cephalexin	>64	>64	>64	>64
	Cephradine	64~>64	>64	>64	>64
	Carbenicillin	32~64	64	64	64
	Ticarcillin	16~32	32	32	32
	Gentamicin	32~>64	>64	>64	>64
	Tobramycin	64~>64	>64	>64	>64
	Amikacin	>64	>64	>64	>64

* MIC_{50}, MIC_{75}, MIC_{90}: MIC needed to inhibit 50%, 75% and 90% of strains respectively.

The comparative *in vitro* activity of 12 antibiotics against Gram-positive cocci is listed in Table 2. The *in vitro* activity of LY 127935 against *S. aureus* was similar to that of cephalexin and cephradine; it was not active against methicillin-resistant *S. aureus*. All strains of *S. pneumoniae, S. pyogenes, S. viridans* and *S. bovis* tested were susceptible to LY 127935. Again, the activity of LY 127935 against these organisms was similar to that of cephalexin and cephradine. All strains of enterococcus were resistant to LY 127935.

Discussion

This new beta-lactam compound, LY 127935 (6059-S) has a very broad antimicrobial spectrum against Gram-negative bacilli and Gram-positive cocci. It not only was active against organisms that were resistant to the cephalosporins including *P. aeruginosa,* but also organisms that were resistant to one or more of the aminoglycosides including amikacin. The vast majority of multi-resistant organisms in this study were susceptible to LY 127935. Only methicillin-resistant *S. aureus* and enterococcus were totally resistant to LY 127935.

If the susceptibility of *B. fragilis* to LY 127935 in the preliminary study can be confirmed by further testing, this compound certainly is one of the newer antibiotics with very broad antimicrobial spectrum. If this new beta-lactam antibiotic proves to be relatively free of toxic adverse reactions similar to the cephalosporins, it would certainly be worth clinical trials.

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