In Vitro Studies with UK-18,892, a New Aminoglycoside Antibiotic

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The antibacterial activity of UK-18,892, a new semisynthetic aminoglycoside, was examined against aminoglycoside-susceptible and aminoglycoside-resistant clinical isolates of gram-negative bacilli and *Staphylococcus aureus*. UK-18,892 had a similar degree of activity to those of amikacin and kanamycin A against aminoglycoside-susceptible bacteria but was less potent than gentamicin against all isolates except *Providencia* spp. UK-18,892 was highly active against aminoglycoside-resistant bacteria, inhibiting 93% of the 268 isolates examined at 12.5 µg/ml. Amikacin was similarly active, whereas gentamicin inhibited only 14% of these isolates at 12.5 µg/ml.

The aminoglycosides are among the most useful drugs currently available for the treatment of infections due to gram-negative bacilli. Their utility is being threatened, however, by the appearance of resistant bacteria, the majority of which possess enzymes capable of inactivating aminoglycosides by acetylation of amino groups and by adenylation or phosphorylation of hydroxyl groups. UK-18,892 (Fig. 1) is a new semisynthetic aminoglycoside derived from kanamycin A, which is active against *Staphylococcus aureus* and a wide range of gram-negative bacilli, including strains known to possess aminoglycoside-inactivating enzymes (7). The purpose of this paper is to report in greater detail the in vitro antibacterial activity of UK-18,892 against clinical isolates of a variety of aminoglycoside-susceptible and aminoglycoside-resistant bacteria and to compare its activity with those of kanamycin A, gentamicin, and amikacin.

MATERIALS AND METHODS

Bacterial isolates. Organisms used in this study were all clinical isolates obtained from hospitals in the United Kingdom and continental Europe, the majority being acquired during 1975 and 1976. Isolates were classified as aminoglycoside resistant if the minimum inhibitory concentrations (MICs) of gentamicin and/or kanamycin A were greater than 6.25 or 12.5 µg/ml, respectively. *Pseudomonas aeruginosa* isolates were classified as resistant on the basis of resistance to gentamicin alone.

Determination of MICs. MICs were determined by using a standard agar plate technique. Organisms were grown in brain heart infusion broth (Difco Laboratories) at 37°C for 18 h. Cultures were diluted in brain heart infusion broth and inoculated onto Diagnostic Sensitivity Test Agar (Oxoid Ltd.) containing serial doubling dilutions of antibiotic, using a multi-

point inoculator (Denley Instruments) giving an inoculum of approximately 10^4 colony-forming units. After incubation at 37°C for 18 h, the MIC was recorded as the lowest concentration of antibiotic which completely inhibited visible growth of the organisms.

MBCs. Organisms were grown as described for MIC determinations. Cultures were diluted in brain heart infusion broth, and approximately 2×10^4 colony-forming units was inoculated into 0.5-ml portions of serial doubling dilutions of the antibiotic in brain heart infusion broth. The tubes were incubated at 37°C for 18 h, and the MIC was recorded. Samples (10 µl) were taken from each tube in which no growth was visible and spread onto Diagnostic Sensitivity Test Agar plates. These were then incubated for 18 h at 37°C. The MBC was the lowest concentration of antibiotic which totally prevented growth on subculture.

Aminoglycosides. Amikacin was a laboratory reference sample generously provided by Bristol Laboratories. Kanamycin A (Kannasyn-Winthrop Laboratories) and gentamicin (gentamicin sulfate Roussel C-UCLAF) were obtained commercially. UK-18,892 was research material prepared as the sulfate. Concentrations of antibiotics are expressed in terms of aminoglycoside base.

RESULTS

Activity against aminoglycoside-susceptible bacteria. The in vitro activity of UK-18,892 against 187 isolates of gram-negative bacilli and 31 isolates of *S. aureus* is shown in Fig. 2. The MICs of UK-18,892 were 6.25 µg/ml or less against all isolates of *S. aureus*, *Escherichia coli*, *Klebsiella* spp., *Proteus mirabilis*, indole-positive *Proteus* spp., *Enterobacter* spp., *Providencia* spp., *Serratia* spp., and *P. aeruginosa*. Amikacin exhibited a similar degree of activity, also inhibiting all of these isolates at 6.25 µg/ml.

The antibacterial potency of kanamycin A was very similar to that of UK-18,892 against *P.*

FIG. 1. UK-18,892, kanamycin A, and amikacin.

mirabilis, indole-positive *Proteus* spp., *Klebsiella* spp., and *S. aureus*, but against most isolates of *Providencia* spp., *Enterobacter* spp., and *E. coli*, the MICs of kanamycin A were 25 to 50% higher than those of UK-18,892. However, all of these isolates were inhibited by 6.25 μg of kanamycin A per ml, whereas 12.5 $\mu\text{g}/\text{ml}$ was necessary to inhibit all of the *Serratia* spp. isolates and more than 90% of the *P. aeruginosa* isolates were resistant to 12.5 μg of kanamycin A per ml. The MICs of gentamicin were 1.56 $\mu\text{g}/\text{ml}$ or less against all isolates of *Proteus* spp., *Enterobacter* spp., *E. coli*, and *S. aureus*; 6.25 $\mu\text{g}/\text{ml}$ or less against *Klebsiella* spp., *Serratia* spp., and *P. aeruginosa*; and 12.5 $\mu\text{g}/\text{ml}$ or less against *Providencia* spp.

Activity against aminoglycoside-resistant bacteria. Table 1 shows the activity of UK-18,892 in comparison with those of gentamicin, kanamycin A, and amikacin against aminoglycoside-resistant isolates of gram-negative bacilli and *S. aureus*. UK-18,892 inhibited over 90% of all isolates of *Klebsiella* spp. and *Enterobacter* spp. at 3.1 $\mu\text{g}/\text{ml}$ and over 95% of *E. coli*, *P. aeruginosa*, and *Serratia* spp. at 6.25 $\mu\text{g}/\text{ml}$. Fifty percent or more of all other clinical isolates examined were inhibited by 12.5 μg of UK-18,892 per ml. Amikacin exhibited similar activity against these isolates, whereas gentamicin (except against *S. aureus*) and kanamycin A were considerably less active.

MBC of UK-18,892. The MBCs of UK-18,892, gentamicin, and amikacin were determined against isolates of *E. coli*, *S. aureus*, *P. aeruginosa*, *P. mirabilis*, and *Klebsiella* spp. In general, the MBCs of all three aminoglycosides tested were about twofold higher than the corresponding MICs. This is in agreement with data

FIG. 2. Activity of UK-18,892 against aminoglycoside-susceptible bacteria. Symbols (and numbers of isolates): ●, *E. coli* (25); ○, *P. aeruginosa* (32); ▲, *S. aureus* (31); △, *P. mirabilis* (35); ■, *Enterobacter* spp. (15); ▼, indole-positive *Proteus* spp. (22); ▽, *Providencia* spp. (12); □, *Serratia* spp. (17); X, *Klebsiella* spp. (29).

published by other workers for gentamicin and amikacin (5).

DISCUSSION

UK-18,892 (Fig. 1) is a synthetic derivative of kanamycin A which exhibits potent activity in vitro against *S. aureus* and a broad spectrum of gram-negative bacilli. Against aminoglycoside-susceptible bacteria in general, UK-18,892 was either equiactive with (*Proteus* spp., *Klebsiella* spp., and *S. aureus*) or up to 1.5 times more potent than (*Providencia* spp., *Enterobacter* spp., and *E. coli*) the parent aminoglycoside, kanamycin A. Against *P. aeruginosa*, however, UK-18,892 was considerably more potent than kanamycin A.

The low activity of kanamycin A against *P. aeruginosa* is expected and may be attributable to the widespread occurrence of 3'-phosphorylating activity in this genus (3, 10). Kanamycin A is inactivated by this enzyme, whereas UK-18,892 is not (7). Amikacin showed very similar activity to UK-18,892 against all aminoglyco-

TABLE 1. Activities of UK-18,892, amikacin, gentamicin, and kanamycin A against aminoglycoside-resistant clinical isolates

Isolates	No. of strains	Compound	Cumulative % inhibited at increasing concn ($\mu\text{g/ml}$)							
			0.2	0.39	0.78	1.56	3.125	6.25	12.5	25
<i>E. coli</i>	16	Gentamicin			13	44	44	44	50	63
		Kanamycin A					6	6	6	6
		Amikacin			6	38	89	94	94	94
		UK-18,892			6	19	75	94	94	94
<i>Klebsiella</i> spp.	56	Gentamicin	2	4	9	9	11	14	20	43
		Kanamycin A					7	7	7	13
		Amikacin			7	88	96	98	100	
		UK-18,892			7	63	96	98	100	
<i>Enterobacter</i> spp.	44	Gentamicin					2	2	7	44
		Kanamycin A								5
		Amikacin			37	88	98	98	100	
		UK-18,892			5	84	98	98	100	
<i>Providencia</i> spp.	21	Gentamicin							19	33
		Kanamycin A					5	10	19	24
		Amikacin				52	71	76	86	90
		UK-18,892				14	71	76	86	95
<i>Serratia</i> spp.	56	Gentamicin								13
		Kanamycin A				2	14	14	14	14
		Amikacin			2	39	89	100		
		UK-18,892				13	71	100		
<i>Acinetobacter</i> spp.	10	Gentamicin			10	10	10	10	10	30
		Kanamycin A							20	30
		Amikacin				30	40	50	50	70
		UK-18,892				10	50	50	60	60
<i>S. aureus</i>	8	Gentamicin	50	63	75	75	75	75	75	100
		Kanamycin A								0
		Amikacin			25	25	63	75	88	100
		UK-18,892			25	25	38	38	50	63
<i>P. aeruginosa</i>	33	Gentamicin								6
		Kanamycin A								0
		Amikacin			9	30	79	94	94	94
		UK-18,892			27	58	85	91	94	94
<i>P. mirabilis</i>	10	Gentamicin				20	30	30	40	80
		Kanamycin A								0
		Amikacin					20	70	80	80
		UK-18,892					10	60	80	80
<i>Proteus</i> spp. (indole positive)	14	Gentamicin						7	7	14
		Kanamycin A					7	7	7	7
		Amikacin			7	14	93	93	100	
		UK-18,892				14	86	86	93	100

side-susceptible organisms examined in this study. The MICs and comparative potencies of gentamicin, amikacin, and kanamycin A reported here are in agreement with data published by other workers (2, 6, 9).

To predict clinical efficacy, the antibacterial activity of these antibiotics must be considered together with the safe and achievable human

serum concentrations of the individual aminoglycosides. Although higher concentrations have been reported (1, 4), we regard 6.25 $\mu\text{g/ml}$ for gentamicin and 12.5 $\mu\text{g/ml}$ for UK-18,892, amikacin, and kanamycin A as representative of clinically acceptable serum concentrations for these aminoglycosides, i.e., as both effective antibacterially and safe. Although not yet evalu-

ated against infections in humans, pharmacological and toxicological studies of UK-18,892 in animals indicate that clinically acceptable serum concentrations of this aminoglycoside will be similar to those of amikacin (unpublished data). On this basis, UK-18,892, amikacin, kanamycin A, and gentamicin should be effective against all of these bacteria, with the exception of kanamycin A against *P. aeruginosa* and gentamicin against *Providencia* spp. Clinical studies have in fact shown that gentamicin and amikacin possess similar clinical efficacies against susceptible bacteria (8).

UK-18,892 was also active in vitro against the majority of the aminoglycoside-resistant bacteria examined. At a clinically acceptable concentration (12.5 µg/ml), 93% of the 268 isolates were susceptible to UK-18,892. Amikacin showed similar activity (94% sensitive), whereas kanamycin A was practically inactive and gentamicin probably had a clinically useful level of activity against only about 10% of these isolates (Table 1).

In view of its potent activity in vitro and in vivo (7; unpublished results) against both aminoglycoside-susceptible and aminoglycoside-resistant bacteria, it is concluded that UK-18,892 is a potentially valuable injectable agent for the treatment of serious bacterial infections.

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