Pharmacological Evaluation of Amikacin in Neonates

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The in vitro bacterial susceptibilities and pharmacokinetic properties of amikacin (BB-K8) were studied in newborn infants. Gram-negative bacteria and *Staphylococcus aureus* isolated from neonates were uniformly susceptible to 10 μg or less of amikacin per ml, and five *Escherichia coli* strains resistant to kanamycin were inhibited and killed by 5 μg or less of amikacin per ml. Mean peak serum concentrations of 17 to 20 μg/ml were observed 30 min after 7.5-mg/kg amikacin doses, and accumulation of drug in serum was not detected after repeated doses for 5 to 7 days. Intravenous infusion of amikacin over a 20-min period resulted in extremely low peak serum levels in four of eight infants studied. Serum half-life values were correlated inversely with postnatal age and renal clearances of amikacin. The volumes of drug distribution indicate that amikacin remains primarily in the extracellular fluid space of neonates. The lack of efficacy and safety data preclude the use of amikacin in neonates at this time.

Past experience has shown that microbial resistance to antibiotics emerges commonly in closed population areas such as nurseries and neonatal intensive care units where these drugs are used routinely. As a result, it is imperative to constantly monitor susceptibilities of commonly encountered pathogens in these high risk units and to evaluate new antimicrobial agents that offer promise for treatment of neonatal bacterial infections. Amikacin is a new aminoglycoside with broad antimicrobial activity similar to that of gentamicin (7). The drug is not a suitable substrate for the common plasmid-associated enzymes responsible for microbial resistance to the other aminoglycosides. Studies in adults have demonstrated that the pharmacokinetic properties of amikacin are similar to those of kanamycin (3).

Because of its potential usefulness in neonatal bacterial infections caused by organisms resistant to kanamycin and gentamicin, the antimicrobial activity against gram-negative bacteria isolated from neonates and the clinical pharmacology of amikacin in full-term and premature infants were studied.

MATERIALS AND METHODS

The susceptibility of 304 organisms cultivated from blood, cerebrospinal fluid, and urine of sick neonates and from stool cultures of healthy babies was determined by agar-plate dilution using a multiple inoculation apparatus (5). The inoculum was 10^5 bacteria/ml. The lowest concentration of antibiotic inhibiting visible growth on Mueller-Hinton agar after 18 h of incubation at 37 C was taken as the minimal inhibitory concentration. Amikacin concentrations tested were 20, 15, 10, 5, and 2.5 μg/ml.

The comparative minimal inhibitory and bactericidal concentrations of amikacin for 10 strains of *Escherichia coli* were studied by the tube dilution technique using an inoculum of 10^5 bacteria/ml. After 18 h of incubation in Mueller-Hinton broth, the lowest concentration of antibiotic inhibiting visible growth was taken as the minimal inhibitory concentration. Dilutions showing no visible growth were subcultured onto eosin methylene blue agar. The highest dilution without growth on agar was taken as the minimal bactericidal concentration.

Amikacin. Concentrations of amikacin in serum and urine were assayed by a modification of the micromethod of Simon and Yin (6) using *Bacillus subtilis* as the test organism. Standard amikacin curves were prepared in an identical fashion to the test samples. Specimens obtained from infants receiving either penicillin or ampicillin in addition to amikacin were treated with penicillinase (Neutropin, Riker Laboratories, Inc.) to inactivate penicillin activity in the assay system.

Serum half-life determinations. The equation for the regression line of the log of serum amikacin concentrations was calculated by the method of the least mean squares. The half-life was determined by dividing the $\log_{10} 2$ by the slope of the line.

Amikacin distribution and plasma clearance. The volume of distribution (V_D) of amikacin, expressed in milligrams per kilogram, was calculated using the formula $V_D = $ (dosage [mg/kg]/area × K) × 1,000, where K is the elimination rate constant and area represents the area under the serum concentration-time curve, which is formulated by successive trapezoidal approximation (4). The plasma clearance (Clp) of amikacin in milliliters per minute per 1.73 M² was determined using the formula Clp = (total dos-

age/area) × (1,000 μg/60 min) × (1.73 M²/a), where a is surface area in M².

Study patients. Patients were selected from the nurseries of Parkland Memorial Hospital, Dallas, Tex. Written informed parental consent was obtained for all infants. Babies treated with penicillin and either gentamicin or kanamycin for suspected bacterial infections had one intramuscular dose of the aminoglycoside substituted with amikacin. An additional 16 infants were treated with an initial 10-mg/kg amikacin dose followed by 7.5-mg/kg doses given every 12 h for 5 days. Serum samples for bioassay were obtained by heel or finger stick technique on days 1 and 5 of therapy. Eight infants who were receiving amikacin intramuscularly were given one dose intravenously over a 20-min period, and multiple serum samples after both routes of administration were obtained on successive days.

All patients were evaluated clinically for evidence of adverse reactions, and complete blood count, urinalysis, blood urea nitrogen, serum creatinine, and serum glutamic oxalotransaminase determinations were performed at initiation and completion of therapy.

RESULTS

Bacterial susceptibilities. The amikacin susceptibilities of gram-negative bacteria are presented in Table 1. The percentages of strains inhibited by 10 μg or less of amikacin per ml were similar to those inhibited by 5 μg of gentamicin and 10 μg of kanamycin per ml, with the exception of *Pseudomonas aeruginosa* strains against kanamycin.

Kanamycin and amikacin susceptibilities of 10 *E. coli* strains cultivated from cerebrospinal fluid of neonates with meningitis were performed by tube dilution. Kanamycin minimal inhibitory and minimal bactericidal concentration values of 20 μg/ml or greater were observed

for five strains. These kanamycin-resistant *E. coli* strains were inhibited and killed by 5 μg or less of amikacin per ml (Table 2).

Clinical pharmacological studies. Serum concentration-time curves were determined in 32 infants given a single 7.5-mg/kg dose of amikacin (Fig. 1). The curves for infants with birth weights under 2,000 g were similar to those for infants weighing 2,000 g or more at birth. Mean peak serum levels of 17 to 20 μg/ml were detected 0.5 h after the dose in all infants regardless of birth weight. Individual peak values ranged from 10 to 26 μg/ml. Mean values of 3.3 to 5.3 μg/ml were observed 12 h after the dose.

To assure that peak serum amikacin concentrations did not fall below the therapeutic range (15 to 25 μg/ml) or accumulate to potentially toxic levels, infants were treated with an initial 10-mg/kg dose followed by 7.5-mg/kg doses administered every 12 h. This dosage schedule was selected because preliminary studies had shown that peak values after 10-mg/kg amikacin doses were in the range of 20 μg/ml. The 12-h dose interval represents approximately two half-lives, during which about 75% of amikacin disappears from serum. Thus, subsequent amikacin doses should be 75% of the initial dose or 7.5 mg/kg, if peak serum concentrations are to be maintained in the range of 20 μg/ml. This was tested by measuring peak and trough (8 and/or 12 h) serum levels in infants receiving this dosage schedule for 5 days (Fig. 2). Peak serum concentrations on day 5 of therapy were in the desired range, and accumulation of drug in serum was not observed.

Urinary excretion. The average excretion of amikacin in 12 h, expressed as percentage of the administered dose, was 40% for the first 3 days

TABLE 1. *In vitro susceptibility studies of gentamicin, kanamycin, and amikacin*

Strains (no. tested)	Cumulative % of strains inhibited at given antibiotic concn (μg/ml)														
	Gentamicin					Kanamycin					Amikacin				
	0.6	1.25	2.5	$\geqslant$5	10	2.5	5	10	15	$\geqslant$20	2.5	5	10	15	>20
E. coli (62)	22	80	96	100	100	35	79	84	95	95	49	84	96	100	
Proteus (32)	1	53	94	100		38	72	94	100		22	66	97	100	
Enterobacter (32)	72	88	94	100		44	75	88	94	100	72	88	97	97	100
Klebsiella (32)	69	88	94	97	100	50	75	88	94	100	72	88	94	100	
Pseudomonas (32)	3	3	31	84	88	3	3	3	6	6	3	47	81	91	94
Salmonella (32)	50	81	100			38	97	100			53	91	97	100	
Shigella (32)	6	88	91	100		16	81	91	100		6	72	100		
Staphylococcus Aureus (17)	94	100				88	88	100			69	82	100		

of life, 29% from 4 to 6 days, and 50% for infants 7 days of age and older. Urinary amikacin concentrations were greatest during the first 4 h after the dose and in the oldest babies. Urinary levels of 50 to 650 μg/ml were observed.

Serum half-life. Serum amikacin half-life

TABLE 2. *Susceptibility of E. coli strains to kanamycin and amikacin*

E. coli strains tested	Kanamycin		Amikacin	
	MIC[a] (μg/ml)	MBC[b] (μg/ml)	MIC (μg/ml)	MBC (μg/ml)
KLC-AT	>80	>80	0.6	1.25
GM-MC	>80	>80	2.5	5.0
BBG-SL	80	>80	5.0	5.0
OVL-ME	80	>80	1.25	1.25
207-DA	20	20	5.0	5.0
N18-SL	10	10	1.25	2.5
B43-DA	5	10	2.5	2.5
B75-DA	2.5	2.5	2.5	5.0
DM-CI	2.5	5	1.25	2.5
BC-MO	2.5	2.5	1.25	2.5

[a] MIC, Minimal inhibitory concentration.
[b] MBC, Minimal bactericidal concentration.

values were inversely correlated with gestational and chronological ages (Fig. 3) and ranged from 7 to 8 h in low-birth-weight infants 1 to 3 days of age to 4 or 5 h in full-term infants 7 days of age or older.

Volume of distribution and plasma clearance. The volumes of amikacin distribution during the first 3 days of life were largest in low-birth-weight infants (Table 3). With increasing postnatal age the volumes diminished in the low-birth-weight infants and increased in the normal-birth-weight babies. The plasma clearance of amikacin did not change significantly during the neonatal period.

Intravenous versus intramuscular administration. Serum levels were measured after both an intramuscular and an intravenous amikacin dose (7.5 mg/kg) in eight infants. Serum concentration-time curves were comparable after the two routes of administration in four infants. In the other four infants the curves after the intravenous dose were flat, and peak serum concentrations of only 4 to 6 μg/ml were detected. By contrast, the peak serum values after intramuscular administration in these babies were 14 to 20 μg/ml.
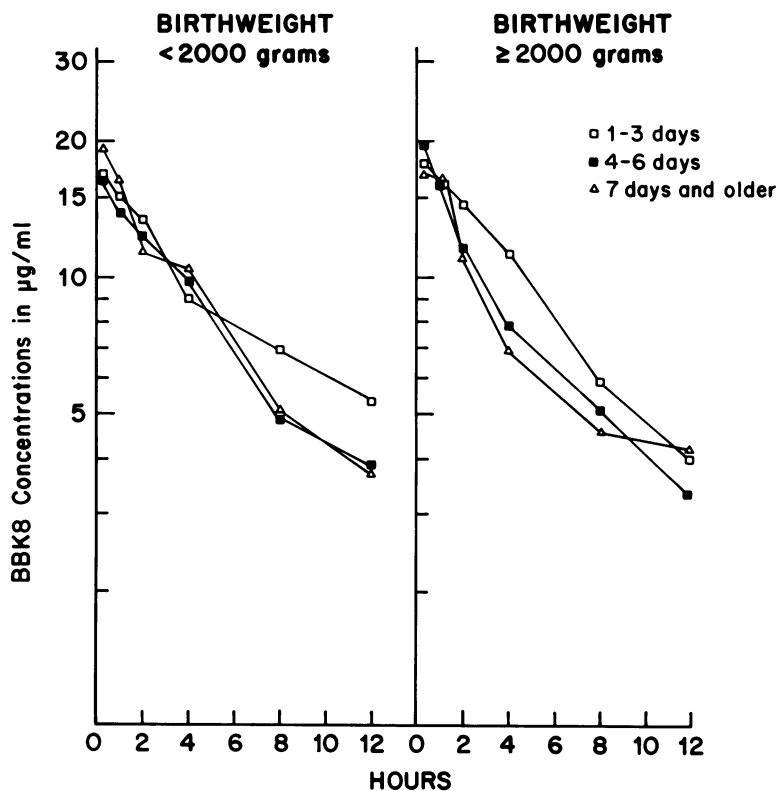

FIG. 1. *Serum concentration-time curves in neonates who received a single intramuscular 7.5-mg/kg dose of amikacin (BBK8).*
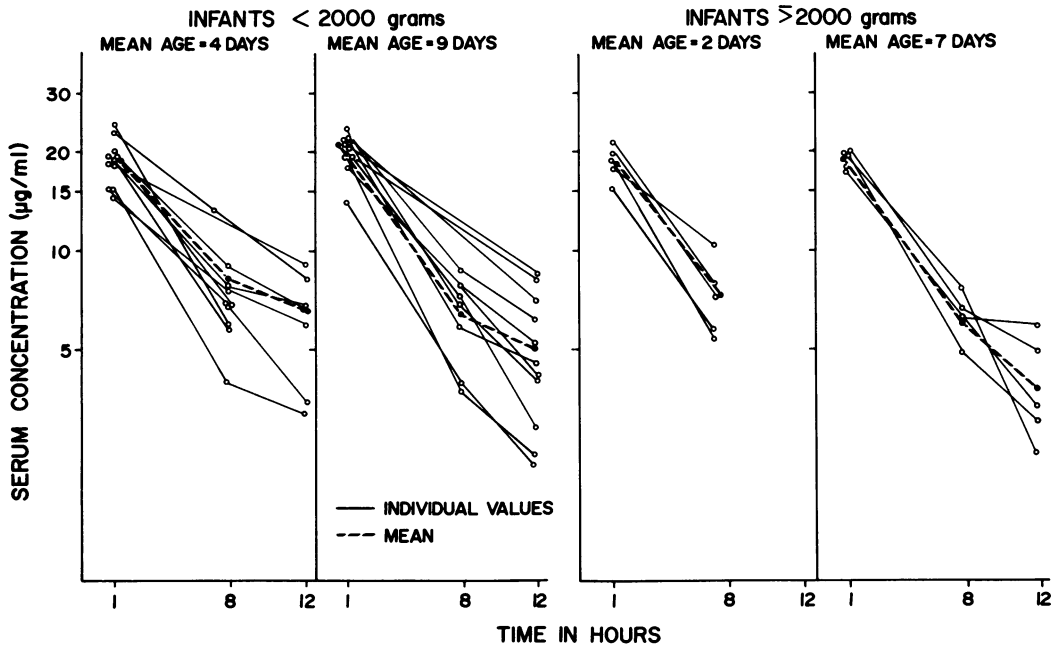
FIG. 2. *Serum concentration-time curves in neonates on days 1 and 5 of therapy. Dosage schedule was 10 mg of amikacin per kg initially, followed by 7.5-mg/kg doses administered intramuscularly every 12 h.*
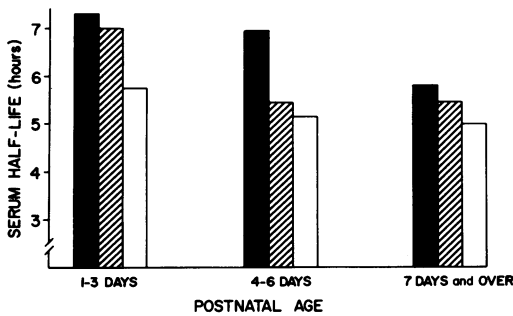

FIG. 3. *Serum amikacin half-life values in relationship to gestational and chronological ages. Gestation periods: closed bar, 30–33 weeks; striped bar, 34–37 weeks; open bar, 38 weeks and over.*

TABLE 3. *Pharmacokinetic properties of amikacin in newborn infants[a]*

Infant groups		Vol of distri-bution (ml/kg)	Plasma clear-ance (ml/mm/1.73 M²)	Serum half-life (h)
Birth-weight (g)	Age (days)			
<2,000				
	1 to 4	563	21.3	7.1
	4 to 7	568	22.8	6.1
	>7	502	24.6	5.5
2,000	1 to 4	509	27.0	6.5
	4 to 7	567	29.8	5.1
	>7	594	36.4	4.9

[a] Five or six infants were studied in each age group for the <2,000- and ≥2,000-g weight categories.

There was no evidence of acute hematologic or renal toxicity after multiple amikacin doses for up to 7 days, and the drug was well tolerated by all study infants.

DISCUSSION

The in vitro amikacin susceptibilities of commonly encountered gram-negative pathogens cultivated from neonates were similar to those for gentamicin. Kanamycin-resistant *E. coli* strains were inhibited and killed by low concentrations of amikacin.

Mean peak serum amikacin levels after an initial 10-mg/kg dose and subsequent 7.5-mg/kg doses given every 12 h were approximately 20

μg/ml, and accumulation of drug in serum after repeated administration was not observed. Peak serum values were independent of birth weight and chronological age, which is different from our previous studies with kanamycin where peak concentrations were correlated with birth weight and postnatal age (5a). Erratic amikacin levels after 20-min intravenous infusions were observed in four of eight infants studied. Because these four babies had the smallest birth weights it is possible that the volume of drug distribution in these infants after

intravenous administration is greater than that after an intramuscular dose. Studies are underway to investigate this possibility.

These pharmacokinetic data suggest that amikacin is distributed within the extracellular fluid space of neonates. The amikacin distribution volumes are similar to the bromide spaces in babies reported by Cassady (2) and to the ticarcillin distribution volumes measured by Nelson and co-workers (Nelson, Shelton, and Kusmiesz, J. Pediatr., in press). The larger volumes in low-birth-weight infants during the first week of life, with subsequent smaller values in the later neonatal period, contrasted with enlarging amikacin distribution volumes in full-term babies throughout the first month of life, most likely reflect the normal physiological changes in the extracellular fluid space during the neonatal life (1, 2). The calculated volumes of distribution of kanamycin were larger than those of amikacin in newborn infants 1 to 4 days of age, whereas the values beyond 4 days were comparable for the two drugs (5a).

On the basis of these studies, amikacin may prove useful for therapy of neonatal infections caused by gram-negative bacteria resistant to kanamycin. A dosage schedule of an initial 10-mg/kg dose followed by 7.5-mg/kg doses administered intramuscularly every 12 h results in peak serum values that are within the thera-peutic and safe range. Studies of safety and efficacy are currently underway in four nurseries in North, Central, and South America. Until these studies are completed, amikacin should only be used for specific clinical conditions where kanamycin and gentamicin are contraindicated and where the infant can be observed closely for evidence of drug toxicity.

LITERATURE CITED

1. Cassady, G. 1966. Plasma volume studies in low birth-weight infants. Pediatrics 38:1020–1027.
2. Cassady, G. 1970. Bromide space studies in infants of low birhtweight. Pediatr. Res. 4:14–24.
3. Clarke, J. T., R. D. Libke,C. Regamey, and W. M. Kirby. 1974. Comparative pharmacokinetics of amikacin and kanamycin. Clin. Pharmacol. Ther. 15:610–616.
4. Dost, F. H. 1968. Grundlagen der Pharmacokinetic, p. 155–190. Georg Thieme Verlag, Stuttgart.
5. Haltalin, K. C., A. H. Markley, and E. Woodman. 1973. Agar plate dilution method for routine antibiotic susceptibility testing in a hospital laboratory. Am. J. Clin. Pathol. 60:384–394.
5a. Howard, J. B., and G. H. McCracken. 1975. Reappraisal of kanamycin usage in neonates. J. Pediatr. 86:949–956.
6. Simon, H. J., and E. J. Yin. 1970. Microbioassay of antimicrobial agents. App. Microbiol. 19:573–579.
7. Yu, P. K. W., and J. A. Washington. 1974. Comparative in vitro activity of three aminoglycosidic antibiotics: BB-K8, kanamycin, and gentamicin. Antimicrob. Agents Chemother. 4:133–139.