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Comparative Pharmacokinetics of BB-K8 and Kanamycin in Dogs and Humans

BERNARD E. CABANA AND JAMES G. TAGGART

Research Division, Bristol Laboratories, Division of Bristol-Myers Co., Syracuse, New York 13201

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Comparative studies in beagle dogs suggested that the pharmacokinetic profiles of BB-K8 and kanamycin are similar. After intravenous (iv) administration to dogs, BB-K8 and kanamycin were rapidly distributed in plasma and tissue fluids; their "apparent" volumes of distribution comprised approximately 23% of the total body volume. Like kanamycin, BB-K8 had a plasma half-life of about 0.8 h which paralleled the urinary excretion rate. Approximately 92% of the dose was excreted as unchanged drug within 6 h of dosing, and clearance appeared to be primarily by glomerular filtration. After intramuscular (im) administration to dogs, BB-K8 and kanamycin were totally and rapidly absorbed; peak concentrations in the plasma occurred 0.5 to 1.0 h after dosing. The kinetic parameters governing the distribution and elimination of BB-K8 and kanamycin after an im dose were similar to those obtained for iv dosing and indicate desirable dose-independent kinetics. A human pharmacokinetic study indicated that the kinetic profiles of BB-K8 and kanamycin are similar in man after im dosing. Like kanamycin, BB-K8 is rapidly absorbed, yielding peak serum concentrations of about 20 μg/ml at 1 h after a 500-mg im dose. The plasma half-life of these two drugs was approximately 2.3 h. Clearance in man was primarily by glomerular filtration, and the urinary excretion of BB-K8 and kanamycin accounted for 83% of the dose.

BB-K8 is a new semisynthetic aminoglycosidic antibiotic derived from kanamycin A (H. Kawaguchi et al., J. Antibiot. [Tokyo], in press) which exhibits broad-spectrum antimicrobial activity, including potent antipseudomonal activity (K. E. Price et al., J. Antibiot. [Tokyo], in press) and activity against some kanamycin-resistant and gentamicin-resistant clinical isolates of *Enterobacteriaceae*. In recent studies with cats (J. C. Reiffenstein et al., in preparation), BB-K8 produced a degree of ototoxicity and renal toxicity similar to that produced by kanamycin when dosed on a weight basis over a 7-day period. In a study based on blood chemistry and histopathology (S. W. Holmes et al., Abstr. 12th Intersci. Conf. Antimicrob. Ag. Chemother., p. 7, 1972), BB-K8 was found to be approximately five times less nephrotoxic than gentamicin, and, in contrast to gentamicin, BB-K8 produced vestibular dysfunction only at the high doses which produced cochlear toxicity.

This report presents a pharmacokinetic comparison of BB-K8 and kanamycin in dogs and humans after parenteral administration.

MATERIALS AND METHODS

In a complete crossover design, BB-K8 and kanamycin were administered intravenously and intramuscularly to four beagle dogs (mean weight, 8.2 ± 0.2 kg) at a dose of 25 mg/kg. Blood samples were taken from an intravenous catheter (Bardic Intracath) implanted into the saphenous vein. Urine was collected cumulatively over a 6- to 8-h period by means of Lucite-coated stainless-steel cannulae surgically implanted within the bladder of the dogs. The urine was collected into flasks placed in an ice bath, and the plasma and urine specimens were kept refrigerated or frozen until bioassayed. Bioassays were made by the cup-plate method (3), with *Bacillus subtilis* as the test organism.

Renal clearance of BB-K8 and creatinine was determined under approximate steady-state conditions in the same beagle dogs. Thirty minutes after oral hydration (30 ml/kg) with tap water, a loading dose of creatinine (800 mg) was given intravenously. Immediately thereafter, continuous infusion of creatinine was initiated at a rate of 5 mg/min, and the infusion was maintained for 3 h. Plasma concentrations of creatinine were determined at 15-min intervals and urine was collected at 30-min intervals. Approximately 1 h after creatinine dosing, the dogs were administered a loading dose of BB-K8 (90 mg),

which was immediately followed by an infusion of BB-K8 at a rate of 1 mg/min for 2 h. Plasma and urine concentrations of BB-K8 and creatinine were determined at 15-min intervals throughout the infusion period. The effect of probenecid on the renal clearance of BB-K8 was determined by the intravenous co-administration of probenecid (25 mg/kg) 60 min after administration of the loading dose of BB-K8. Thereafter, concentrations of BB-K8 and creatinine in plasma and urine were determined at 15-min intervals during the final infusion period. The creatinine concentrations were determined according to the method of Bonsnes and Tausky (1).

The absorption characteristics of BB-K8 and kanamycin were compared in beagle dogs after intramuscular administration of doses of 7.5, 25, and 50 mg/kg. Twelve beagle dogs having a mean weight of 10.1 ± 0.6 kg were employed in a complete crossover design. A similar study was also performed in human volunteers. Twenty-four healthy male volunteers between the ages of 21 and 48 years (median, 24 years) and weighing between 62.3 and 91.0 kg (median, 78.2 kg) were selected for this study. Twelve subjects received one dose of 250 mg of each of the two drugs in a completely randomized crossover design, with at least 48 h between administration of the two drugs. The remaining 12 subjects followed the same procedure with doses of 500 mg of each drug. Blood samples for determinations of drug concentrations in the serum were drawn at 0.25, 0.5, 1, 2, 2.5, 3, 4, 6, 8, 10, and 12 h after dosing. Urine was collected for 24 h, and the amounts of drug excreted in the urine were estimated.

Drug concentrations in plasma were analyzed kinetically by means of a Fortran 4 computer program (IBM Share Program Library, No. 3094) on an IBM 360/30 digital computer. Nonlinear regression analyses of the plasma concentration-time curves and the least-squares estimation of the nonlinear parameters were performed by the method of Marquardt (5), assuming the two-compartment open system model illustrated in Fig. 1 as described by Wagner and Northam (9) and Riegelman et al. (7, 8).

After intravenous administration of BB-K8 or kanamycin, the plasma concentrations of drug were fit to the general biexponential equation

$$C_P = A_1e^{-\alpha t} + B_1e^{-\beta t} \quad \{1\}$$

where C_P is the concentration in plasma (in micrograms per milliliter), t is time (in hours), the rate constants α and β are hybrid rate constants governing drug distribution and elimination, and the sum of $A_1 + B_1$ is equal to drug concentration (C_P^0) at zero time. The rate constants governing distribution (K_{12}, K_{21}) to and from tissue fluids, the rate constant governing drug elimination (K_{el}), and the respective volumes of drug distribution, V_P and V_T (illustrated in the model), were computed by means of kinetic equations previously described (7-10).

The concentrations of drug in plasma after intramuscular administration of either BB-K8 or kanamycin were fit to the following biexponential equation

$$C_P = -A_1e^{-\alpha t} + B_1e^{-\beta t} \quad (2)$$

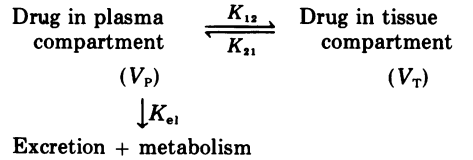

FIG. 1. *Two-compartment open-system model describing the distribution and elimination of drug in dogs.*

where α is the hybrid rate constant governing absorption and distribution of drug and β is the hybrid rate constant describing drug elimination from plasma. The intrinsic absorption rate of BB-K8 and kanamycin in the beagle dog was calculated by the method of Loo and Riegelman (4) as previously described for ampicillin (2). The physiological availability of each drug was determined by comparing the area under the plasma concentration-time curves after intramuscular administration with that obtained after intravenous administration of drug, by use of the following equation

$$\text{percent availability} = \frac{\int_0^\infty C_P dt\ (\text{IM})}{\int_0^\infty C_P dt\ (\text{IV})} \cdot 100 \quad (3)$$

The areas under the plasma concentration-time curves were determined by integration of the appropriate equations describing such curves. The bioavailability of the drugs was also determined by comparing the urinary recovery ratios (F_u) after intramuscular and intravenous doses, by use of the following equation

$$F_u = \frac{\text{percent recovery}_{(IM)}}{\text{percent recovery}_{(IV)}} \quad (4)$$

The time at which peak plasma concentration (T_{max}) was achieved after intramuscular administration of drug was determined from the following equation

$$T_{max} = \frac{\ln A_1\alpha/B_1\beta}{\alpha - \beta} \quad (5)$$

The maximal concentration of drug in plasma (C_{max}) was determined by substitution of the T_{max} value in the appropriate equations describing the plasma concentration-time curves.

The plasma concentrations of BB-K8 or kanamycin obtained in humans after intramuscular administration of drug were fit to the following biexponential equation, assuming a single compartment model

$$C_P = \frac{FD}{V_D} \frac{k_a}{(k_a - k_{el})} (e - k_{el}^{(t - t_0)} - e - k_a^{(t - t_0)}) \quad (6)$$

where F is the fraction of drug absorbed, D is the dose in milligrams per kilogram, V_D is the "apparent" volume of drug distribution (liters per kilogram), k_a and k_{el} are the hybrid rate constants governing drug absorption and elimination, respectively, and t_0 is the absorption lag time. The "apparent" volume of

drug distribution (V_D) was arrived at by use of the following equation, assuming 100% absorption of drug at the muscle site

$$V_D = \frac{Dk_a}{C_0(k_a - k_{el})} \quad (7)$$

where C_0 is the zero time intercept concentration obtained at time t_0.

The renal clearances of (Cl_R) of BB-K8 or kanamycin were determined from the concentrations in plasma and urine from a single intravenous or intramuscular dose, by use of the following equation

$$Cl_R = \frac{f_e \text{ Dose}}{\int_0^\infty C_P dt} = \frac{f_e \text{ Dose}}{\text{Area}} \quad (8)$$

where f_e is the fraction of administered drug excreted unchanged in the urine.

RESULTS

Intravenous and intramuscular studies in dogs. Before the absorption kinetics of any drug can be adequately described, the distribution and elimination kinetics must be defined for that drug after intravenous dosing. The mean concentrations of BB-K8 and kanamycin in plasma of beagle dogs after intravenous administration of 25 mg/kg doses are illustrated in Fig. 2A with their corresponding least-squares nonlinear regression lines. Kinetic analysis of the plasma concentration data indicated a very close similarity between the pharmacokinetic profiles of BB-K8 and kanamycin in beagle dogs (Table 1). BB-K8 and kanamycin were rapidly distributed to the plasma and tissue fluids, their total volume of distribution (V_D) comprising approximately 23 to 25% of the body volume. The plasma half-lives of BB-K8 and kanamycin were similar ($t_{1/2} \simeq 0.7$ to 0.85 h) and paralleled the decline in urinary excretion rate. The urinary excretion half-life of BB-K8 and kanamycin was approximately 45 min in dogs, resulting in approximately 55 to 60% of the dose being excreted within 1 h of dosing. The total extent of urinary excretion of BB-K8 and kanamycin 6 h after an intravenous dose to beagle dogs accounted for approximately 92 and 90% of the dose, respectively. The renal clearance (Cl_R) of BB-K8 and kanamycin in the beagle dogs was 30 to 34 ml/min, or approximately 4 ml per min per kg.

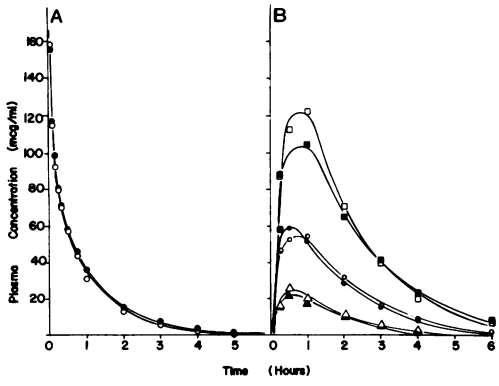

FIG. 2. A, *Mean concentrations of BB-K8* (●) *and kanamycin* (○) *in plasma after intravenous administration of 25 mg/kg doses to beagle dogs.* B, *Mean concentrations of BB-K8 and kanamycin in plasma after intramuscular administration to beagle dogs. BB-K8: 7.5* (▲), *25* (●), *and 50 mg/kg* (■). *Kanamycin: 7.5* (△), *25* (○), *and 50 mg/kg* (□).

The intramuscular administration of BB-K8 and kanamycin to the same beagle dogs at a dose of 25 mg/kg resulted in peak concentrations of 40 to 50 μg/ml at 30 to 60 min after drug administration. The plasma half-life values of BB-K8 and kanamycin were approximately 1.2 and 1.3 h, respectively, and paralleled the decline in urinary excretion rate. Approximately 92% of the administered dose of BB-K8 was excreted in the urine as unchanged drug over an 8-h period. Similar values were obtained with kanamycin. Comparison of the areas under the plasma concentration-time curves for intramuscular dosing to those similarly obtained for intravenous dosing, and comparison of the urinary excretion ratios (equation 5), revealed that the bioavailability of BB-K8 and kanamycin in the beagle dog was about 100%.

Illustrated in Fig. 2B are the mean concentrations of BB-K8 and kanamycin obtained in three separate groups of beagle dogs (four dogs per group) after intramuscular administration of doses of 7.5, 25, and 50 mg/kg. The similarity in the kinetic profile of BB-K8 and kanamycin is rather apparent. Comparison of the kinetic profiles of BB-K8 and kanamycin failed to reveal any significant difference between these two drugs (Table 2). After intramuscular administration, BB-K8 and kanamycin were rapidly absorbed; peak concentrations in the plasma occurred 30 to 45 min after dosing. The absorption half-life of BB-K8 ranged from 0.45 to 0.75 h. Comparison of the peak concentrations (C_{max}) and areas under the plasma concentration-time curves indicated an excellent dose response for both drugs. The plasma half-life of BB-K8 ranged from 1.05 to 1.20 h. Similar values were obtained for kanamycin.

Renal clearance studies in beagle dogs. Renal clearance studies with creatinine and BB-K8 were undertaken in four beagle dogs under steady-state conditions to determine the mode of elimination of BB-K8 in the dog. As described in Materials and Methods, pseudo steady-state conditions were achieved by intravenous bolus followed by slow drug infusion

over a 2- to 3-h period. Comparison of the mean concentrations of creatinine and BB-K8 during the first and last infusion period indicated that steady-state conditions were closely approximated (Table 3). The clearances of creatinine and BB-K8 ranged from 30.0 to 34.3 ml/min and 32.2 to 35.8 ml/min, respectively, in the beagle dog. Thus, it would seem that the renal clearance of creatinine and BB-K8 was approximately 4.0 ml per min per kg in the beagle dog. The co-administration of probenecid (25 mg/kg intravenously) did not alter the renal clearance of BB-K8 in the dog, thereby ruling out any active secretion of BB-K8 by the renal tubules.

Intramuscular studies in humans. Illustrated in Fig. 3 are the results of a crossover study in humans in which 250- and 500-mg doses of BB-K8 and kanamycin were administered intramuscularly. Peak concentrations of approximately 12 and 20 μg/ml were obtained in plasma at 1 h after intramuscular administration of 250- and 500-mg doses of BB-K8 and kanamycin, respectively. Comparison of the pharmacokinetic profiles of both drugs revealed a close similarity (Table 4). In man, BB-K8 and kanamycin appeared to be distributed in approximately 22 to 23% of the body volume ($V_D \simeq 225$ ml/kg). The plasma half-life

TABLE 1. *Parameters for the two-compartment open system model estimated from concentrations of BB-K8 and kanamycin in plasma after intravenous administration of 25 mg/kg doses to beagle dogs*

Agent	Equation	Parameters[a]								
		K_{12}	K_{21}	K_2	Half-life	V_P	V_T	V_D	Cl_R	Area
BB-K8	$68.3e^{-5.75t} + 80.9e^{-0.81t}$	1.74	3.49	1.33	0.85	0.17	0.80	0.25	34	111.8
Kanamycin	$60.0e^{-8.1t} + 92.4e^{-1.0t}$	2.25	5.30	1.54	0.70	0.16	0.70	0.23	30	98.8

[a] K_{12}, K_{21}, and K_2 are expressed as hours^{-1}; the half-life, as hours; V_P, V_T, and V_D, as liters per kilogram; Cl_R, as milliliters per minute; and area, as micrograms·hour per milliliter.

TABLE 2. *Parameters for the two-compartment model estimated from concentrations of BB-K8 or kanamycin in plasma after intramuscular administration to beagle dogs*

Drug	Dose (mg/kg)	T_{max} (h)	C_{max} (μg/ml)	Plasma half-life (h)	k_a (h^{-1})	Absorption half-life (h)	Area (μg·h per ml)
BB-K8	7.5	0.49	22.5	1.05	1.54	0.45	40.6
Kanamycin	7.5	0.49	25.8	1.03	1.73	0.40	41.1
BB-K8	25.0	0.57	59.3	1.14	1.33	0.52	131.5
Kanamycin	25.0	0.68	55.6	0.93	1.38	0.50	140.9
BB-K8	50.0	0.86	102.4	1.20	0.92	0.75	283.9
Kanamycin	50.0	0.76	123.8	1.0	1.26	0.55	311.9

TABLE 3. *Renal clearance of creatinine and BB-K8 and the effect of probenecid on the clearance of BB-K8 in beagle dogs*

Treatment	Creatinine		BB-K8	
	Concn in plasma (μg/ml)	Renal clearance (ml/min)	Concn in plasma (μg/ml)	Renal clearance (ml/min)
Creatinine alone[a]	149.3 ± 10.5	30.0 ± 6.0	—	—
Creatinine + BB-K8[b]	135.8 ± 13.1	31.8 ± 2.5	35.2 ± 2.7	32.2 ± 2.2
Creatinine + BB-K8 + probenecid[c]	133.5 ± 11.7	34.3 ± 8.9	31.5 ± 2.9	35.8 ± 4.0

[a] An intravenous loading dose of creatinine (800 mg) was followed by a continuous infusion (5 mg/min) for 3 h. Concentrations of creatinine during the first hour of the infusion were determined at 15-min intervals in plasma and at 30-min intervals in urine.

[b] Approximately 1 h after creatinine dosing, a loading dose of BB-K8 (90 mg) was administered, and this was immediately followed by an infusion of BB-K8 (1 mg/min) for 2 h. Concentrations of creatinine and BB-K8 in plasma and urine were determined at 15-min intervals during the first hour of the BB-K8 infusion.

[c] At 60 min after the loading dose of BB-K8, 25 mg of probenecid/kg was administered. Concentrations of creatinine and BB-K8 in plasma and urine were determined at 15-min intervals during the second hour of the BB-K8 infusion.

for both drugs ranged from 2.1 to 2.4 h, and the renal clearance was approximately 75 ml/min. The mean renal clearances of creatinine (Cl_{cr}) before and after drug administration in these volunteers were 114 and 117 ml/min, respectively. Comparison of the areas under the plasma concentration-time curves indicated excellent dose proportionality. Like kanamycin the total urinary excretion of BB-K8 (f_e) in 24 h after dosing was approximately 84% of the dose.

DISCUSSION

BB-K8 was shown to exhibit a pharmacokinetic profile similar to that of kanamycin in both dogs and humans. After an intravenous bolus to beagle dogs, BB-K8, like kanamycin, was rapidly distributed in plasma and tissue fluids from which it was eliminated primarily by the kidneys. The apparent volume of drug distribution of BB-K8 ($V_D \simeq 230$ ml/kg) comprised approximately 23% of the body volume, which indicated that the drug was distributed primarily in plasma and extracellular fluids. The plasma half-life of BB-K8 was approximately 0.85 h and paralleled the decline in urinary excretion rate. Approximately 92% of an intravenous dose of BB-K8 was excreted as unchanged drug within 6 h of dosing, indicating that urinary excretion is the major pathway of drug elimination in the dog. The renal clearance of BB-K8 in the dog approximated the clearance of creatinine (Cl_R = 4.0 ml per min per kg), indicating that glomerular filtration is the major excretory pathway for BB-K8. Based on simultaneous BB-K8–creatinine clearance in the absence and presence of probenecid, no evidence of tubular reabsorption or secretion was obtained in the canine studies.

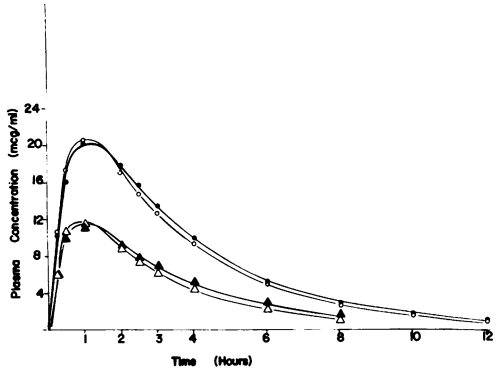

FIG. 3. *Mean concentrations of BB-K8 and kanamycin in serum of human volunteers after intramuscular administration. BB-K8: 250- (▲) and 500-mg (●) doses. Kanamycin: 250- (△) and 500-mg (○) doses.*

The similarity in the kinetic profiles of BB-K8 and kanamycin was not restricted to intravenous dosing but also manifested itself in dogs and humans after intramuscular dosing. In the dog, BB-K8 was rapidly absorbed ($t_{1/2} \simeq$ 0.5 hours), achieving peak plasma concentrations 30 to 45 min after intramuscular dosing. Comparison of the peak plasma concentrations and the areas under the plasma concentration-time curves after intramuscular administration of BB-K8 and kanamycin in doses of 7.5, 25, and 50 mg/kg to beagle dogs indicated excellent dose proportionality. These comparative studies indicated that the kinetic parameters governing the absorption, distribution, and elimination of BB-K8 and kanamycin in the dog were similar (see Table 2). The results further indicate that, within the dose range studied, BB-K8 and kanamycin are eliminated by dose-independent kinetics. The bioavailability of both drugs in the dog was approximately 100%. The total urinary excretion of BB-K8 in dogs after intramuscular dosing accounted for approximately 92% of the dose, indicating that the route of administration of drug did not alter the kinetic profile of BB-K8 or kanamycin.

The kinetic studies performed in humans showed that there is a strong similarity between the kinetic profiles of BB-K8 and kanamycin after intramuscular dosing (Table 4). The kinetic profiles in humans were also similar to those obtained in beagle dogs (Table 2). As in dogs, the apparent volumes of distribution (V_D) of BB-K8 and kanamycin in humans were approximately 22 to 23% of the total body

TABLE 4. *Kinetic parameters estimated in humans after intramuscular administration of BB-K8 and kanamycin*

Drug	Dose (mg)	T_{max} (h)	C_{max} (μg/ml)	Half-life (h)	V_D (ml/kg)	Cl_R (ml/min)	f_e (%)	Area (μg·h per ml)
BB-K8	250	0.90	11.4	2.4	225	70.0	0.83	50.0
Kanamycin	250	0.80	11.9	2.1	217	79.0	0.84	44.2
BB-K8	500	1.15	20.4	2.2	222	74.9	0.83	92.4
Kanamycin	500	1.0	20.6	2.3	236	71.3	0.77	90.0

volume, suggesting that these drugs are primarily distributed in extracellular fluids. These findings are in excellent agreement with those reported for kanamycin in humans by Orme and Cutler (6). The plasma half-life of BB-K8 was approximately 2.3 h and paralleled the urinary excretion rate, indicating that urinary excretion of drug was the rate-determining step in the elimination of BB-K8 in man. The half-life values for kanamycin were similar to those for BB-K8 and are in excellent agreement with those obtained by other investigators (J. T. Doluisio, L. W. Dittert, and J. C. LaPiana, in preparation). Approximately 84% of the administered dose of each aminoglycoside was excreted as unchanged drug in the 24 h after dosing. The renal clearance values (Cl_R) reported in Table 4 are in good agreement with those previously reported for normal adult volunteers (6; Doluisio et al., in preparation) and suggest that BB-K8 is primarily excreted by glomerular filtration. The ratios of BB-K8 clearance to creatinine clearance were 0.65 in these adult volunteers, suggesting that tubular reabsorption does occur in man. These ratio values are in excellent agreement with those reported for kanamycin (6; Doluisio et al., in preparation). Further studies are in progress to confirm these findings.

In conclusion, the results of these studies suggest that the absorption, distribution, and elimination kinetics of BB-K8 and kanamycin are similar in dogs and humans. These studies also indicate that urinary excretion is the main excretory pathway in the elimination of BB-K8. Further, the primary mechanism of renal excretion of the drug is by glomerular filtration. Therefore, careful monitoring of renal and eighth nerve function will be of prime importance in the clinical situation, especially in the high-risk patient population with such predisposing factors as impaired renal function, dehydration, diuretic therapy, and advanced age.

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