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PHARMACOKINETICS OF TOBRAMYCIN FOLLOWING A SINGLE INTRAMUSCULAR ADMINISTRATION IN KAGHANI GOATS (*CAPRA HIRCUS*)

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Summary

Prawez, S., R. Raina, A. K. Srivastava, N. K. Pankaj, P. K. Verma & D. J. Dimitrova, 2007. Phamacokinetics of tobramycin following a single intramuscular administration in Kaghani goats (*Capra hircus*). *Bulg. J. Vet. Med.*, **10**, No 2, 95–101.

The pharmacokinetics and safety of tobramycin sulphate solution was studied in Kaghani goats (*Capra hircus*) following a single intramuscular administration at a dose rate of 3 mg/kg body weight. The blood plasma level of tobramycin was determined by using *Bacillus subtilis* ATCC 6633 as test organism and the lower limit of detection was 0.1 µg/mL. Plasma concentration-time curve was analysed via non-compartmental model based on statistical moment theory (SMT). Serum blood urea nitrogen (BUN) and serum creatinine evaluation was carried out before and on 3rd & 7th days after the tobramycin administration. Following single i.m. administration the drug was rapidly absorbed with peak plasma concentration (C_{max}) of 21.76±0.79 µg/mL at time (T_{max}) 0.43±0.04 h. The values of plasma biological half-life ($t_{½}\lambda$), area under plasma concentration-time curve ($AUC_{0\rightarrow\infty}$) and mean residence time (MRT) were 3.13±0.24 h, 70.02±5.16 µg.h/mL and 4.18±0.24 h, respectively. Laboratory tests detected a slight raise of values of BUN and serum creatinine on days 3rd and 7th but within normal range. It was concluded that the single i.m. administration of tobramycin at 3 mg/kg body weight may provide adequate 12-hour plasma concentration levels to treat most susceptible Gram-positive and Gram-negative infections in goats.

Key words: goats, pharmacokinetics, tobramycin

INTRODUCTION

Tobramycin is a naturally occurring deoxykanamycin with antimicrobial and pharmacokinetic properties similar to those of gentamicin. It is, however, more active against *Pseudomonas aeruginosa,* and against ⅔rd of gentamicin resistant strains (Pennington & Stone, 1974; Szwed *et al.*, 1974; Brogden *et al.*, 1976; Simon *et al.*, 1976; Prescott & Baggot, 1994). Like other aminoglycosides, tobramycin is ototoxic and nephrotoxic. It appears to be less nephrotoxic than gentamicin probably due to the lesser extent of its accumulation in the renal cortical tissue (Luft *et al.*, 1978; Bille & Glauser, 1981). To avoid such toxicity and to ensure adequate

therapeutic drug concentrations (Jernigan *et al.*, 1988) once daily administration of tobramycin is currently applied in human clinical practice (Murry *et al.*, 1999) and for treating tissue infection in horses (Tudor *et al.*, 1999). The aminoglycosides have concentration-dependent bactericidal activity and the peak concentration (C_{max}) to minimum inhibitory concentrations (MIC) ratio (C_{max}/MIC) is the PK/PD parameter best correlated with clinical efficacy (Smith *et al.*, 2001; Scaglione, 2002). The efficacy of aminoglycosides drugs also correlates with the achievement of C_{max}/MIC >10 or C_{max}/MIC=8–10 (Blaser *et al.,* 1987; Xiong *et al.*, 1997; Toutain, *et al.*, 2002). The pharmacokinetics of tobramycin have been reported in humans (Regamey *et al.*, 1973; Dyas *et al.*, 1983), dogs (Dimitrova *et al.*, 2004), cats (Jernigan *et al.*, 1988), rabbits (Beam & Allen, 1977) and camels (Hadi *et al.*, 1994) but not in goats particularly in concern with i.m. administration (Prescott & Baggot, 1994).

The aim of present study was to determine the pharmacokinetic parameters and renal safety following a single intramuscular administration of tobramycin in Kaghani goats (*Capra hircus*).

MATERIALS AND METHODS

Animals

The study was performed on clinically healthy Kaghani goats, 4–6 years old and weighing from 30 to 50 kg. All dewormed animals were housed in hygienic departmental shed twenty days prior to commencement of experiment for acclimatization and allowed free access to pasture, *ad libitum* water and received once daily concentrate feed ration. No treatments were performed within two weeks before the study's initiation. A day prior to commencement of experiment, the goats underwent through physical and clinical examinations.

Experimental design

After a overnight fasting, the goats were administered a single intramuscular injection of tobramycin sulphate (Tobraneg^{TM}, Eli Lilly & Company, USA) at a dose rate of 3 mg/kg body weight in the gluteal muscles. Blood samples of 6–7 mL were obtained directly from *v. jugularis* using disposal needles (G 18, L 45 mm). The blood samples were collected in heparinized test tubes just prior to and at hours 0.04, 0.08, 0.17, 0.33, 0.50, 0.75, 1.0, 1.5, 2, 3, 4, 6, 9 and 12 after drugs administration. Blood samples were centrifuged for 15 minutes at 3000 rpm to get plasma and stored at -20^0 C until analysis, usually within 2–3 days.

To study the renal safety of tobramycin, biochemical parameters (blood urea nitrogen, BUN and serum creatinine) were determined in blood samples obtained from *v. jugularis* just prior to drug administration and on days 3rd, and 7th after drug administration. The blood samples were allowed to clot, serum was separated by centrifugation and used for biochemical parameters analysis.

Assay procedure

Plasma concentrations of tobramycin were microbiologically determined by assay technique using *Bacillus subtillis* ATCC 6633 as a test organism (Arret *et al.*, 1971) by taking overnight growth of test organisms after adjusting the CFU/mL spectrophotometerically (Simon & Jongyin, 1970). The standard solutions of drug were prepared in plasma collected from untreated goats or in phosphate buffered saline (PBS). Comparing both ap-

proaches, no statistically significant differences were found. Hence, standards prepared in PBS as well as in plasma were used for the assay technique whose validity has been previously documented (Sahs & Joynt, 1976; Beam & Allen, 1977). The lower limit of detection by this method was 0.1 μg.mL^{-1} in plasma with a correlation coefficient (r^2) of 0.988±0.004. The standard curve of tobramycin in plasma was linear between 0.1 and 4.0 μg/mL. Each sample was diluted to the extent that its zone of inhibition came in linear range. Semi-logarithmic plots of the zone of inhibition versus standard tobramycin concentrations in serum was subjected to linear regression analysis and concentrations of the drug were determined using the intercept and slope of the regression line.

The variation coefficient of the technique for the intrassay and interassay precission was less than 10% in the range of standard concentrations.

Biochemical analysis

For *in vitro* BUN (DAM method), and serum creatinine (Alkaline picrate method) estimation, Qualigens^T Diagnostic kits were used (GlaxoSmithkline Pharmaceuticals Ltd., Mumbai, India).

Pharmacokinetic analysis

Pharmacokinetic analysis of the data was performed using non-compartmental model based on statistical moment theory (Gibaldi & Perrier, 1982). For peak plasma concentration (C_{max}) and time of peak concentration (T_{max}) observed values were taken. The zero time intercept (C_z) and the apparent terminal rate constant λ_z, were determined by linear regression of the last four to five points on the terminal phase of logarithmic serum concentrations vs. time curve. The terminal elimination half-life ($t_{½}\lambda$), was calculated as In $2/\lambda_z$ and its harmonic mean has also been estimated (Lam *et al.*, 1985). The area under the plasma concentration-time curve (AUC) for the time (t) at which the final measurable concentration was obtained (AUC_{0-t}) was calculated by the linear trapezoidal rule. The AUC from the final time point to infinity ($AUC_{0-\infty}$) was estimated as the ratio of the final observed concentration/λ_z. The total area under the concentration-time curve ($AUC_{0-\infty}$) was calculated by addition of AUC_{0-t} and $AUC_{t-\infty}$. The area under the first moment curve (AUMC) was the area under the curve of the product of time and the plasma drug concentration vs. time from time zero to infinity.

RESULTS

The physical and clinical examination of goats showed that they were in good health. The mean plasma tobramycin concentrations following single i.m. administration of 3 mg/kg are presented in Fig.1 whereas the values of pharmacokinetic variables and biochemical tests parameters (BUN and serum creatinine) are shown in Tables 1 and 2. Tobramycin was rapidly absorbed following i.m. administration, and detected in plasma within 2.5 minutes and reached its peak plasma concentration (C_{max}) 21.76±0.79 μg.mL^{-1} after time (t_{max}) 0.43±0.04 h and thereafter gradually decreased to lower concentration by the 12th h. In this experiment tobramycin concentrations were above the limit of detection (0.10μg.mL^{-1}) up to the time of last sample (12th hour). The elimination half-life ($t_{½}\lambda$), the area under plasma drug concentration-time curve ($AUC_{0-\infty}$) and the mean residence time (MRT) were estimated to be 3.13±0.24 h, 70.02±5.16 μg.h/mL and 4.18 ± 0.24 h, respectively.

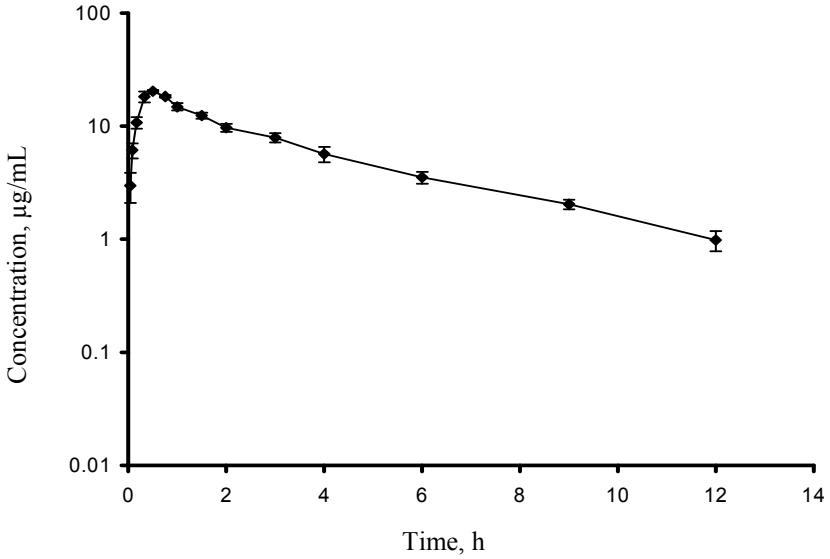

Fig. 1. The mean ± SEM plasma concentration vs. time profile of tobramycin in five Kaghani goats following a single intramuscular administration of tobramycin sulphate at 3 mg/kg body weight.

Table 1. Pharmacokinetic parameters (mean±SEM) of tobramycin in Kaghani goats (n=5) after a single i.m. administration of tobramycin sulphate at a dose of 3 mg/kg body weight – noncompartmental analysis

Parameters	Units	Values
$t_{1/2}\lambda$	h	3.13±0.24 (3.06)[#]
MRT	h	4.18±0.24
$AUC_{0\rightarrow\infty}$	µg.h/mL	70.02±5.16
$AUMC_{0\rightarrow\infty}$	µg.h^2/mL	296.00±38.16
C_{max}	µg/mL	21.76±0.79
T_{max}	h	0.43±0.04

– harmonic mean; $t_{1/2}\lambda$– elimination half-life; MRT – mean residence time; $AUC_{0\rightarrow\infty}$ – area under the concentration *vs*. time curve from hour 0 to infinity; $AUMC_{0\rightarrow\infty}$ area under the first moment curve from hour 0 to infinity ; C_{max} – peak plasma concentration after oral administration ; T_{max} – time to reach peak plasma concentration.

DISCUSSION

Tobramycin is eliminated by renal excretion (glomerular filtration) unchanged in urine. The biological half-life ($t_{1/2}\lambda$= 3.13±0.24 h) was found similar to that in llamas 3.68 h (Christensen *et al.*, 1996) and camels 3.15 – 3.35 h (Hadi *et al.*, 1994) and longer than the half-life reported in dogs 1.17–1.34 h (Szwed *et al.*, 1974), cats 1.17–1.84 h (Jernigan *et al.*, 1988) and humans 1.64-2.15 h (Regamey

Table 2. Blood serum chemistry on days 0, 3 and 7 following a single i.m. administration of tobramycin sulphate at a dose rate of 3 mg/kg to five Kaghani goats

Goats (n=5)	Blood urea nitrogen (mg/dL)*			Serum creatinine (mg/dL)**		
	Day 0	Day 3	Day 7	Day 0	Day 3	Day 7
1	12.00	18.00	19.00	0.88	0.93	1.05
2	14.00	15.00	16.00	1.16	1.40	1.16
3	13.50	13.75	15.00	1.18	1.08	1.17
4	16.00	17.00	15.00	1.13	1.10	1.50
5	13.00	15.00	17.50	1.37	1.00	0.92

* normal value – 10 to 20 mg/dL; ** normal value – < 1.60 mg/dL (colorimetric method).

et al., 1973; Dyas *et al.*, 1983). The observed species variation may be due to lower filtration rates than in humans, dogs and cats (Beam & Allen, 1977).

The plasma mean residence time (MRT) values are similar to values in camels 4.23 h (Hadi *et al.*, 1994) whereas higher than the values in rabbits 1.58 h (Lashev & Dimitrova, 2002) and dogs 1.06 h (Dimitrova *et al.*, 2004) after i.v. administration.

Following single i.m. administration of tobramycin on days 3rd and 7th there were slight increases in BUN and serum creatinine, however, the values ranged within the normal limits in goats (Table 2).

Aminoglycosides' optimal bactericidal activity is achieved when C_{max}/MIC = 8 – 10 (Blaser *et al.,* 1987; Xiong *et al.*, 1997; Toutain, *et al.*, 2002). High C_{max}/MIC ratios are also related to reduced emergence of adaptive aminoglycoside-resistant pathogens (Xiong *et al.*, 1997). The reported MICs for gram negative bacteria isolated from cattle were 0.25–2.0 $\mu g.mL^{-1}$ (Ziv *et al.*, 1981). Thus tobramycin levels found in this study produced high C_{max}/MIC ratios.

In conclusion, the pharmacokinetic parameters of tobramycin sulphate after its single i.m. administration at 3 mg/kg body weight may provide adequate 12 hour plasma concentration levels to treat most susceptible Gram-positive and Gram-negative infections in goats.

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