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Comparative Pharmacokinetics of Theophylline in Camels (*Camelus dromedarius*) and Goats (*Caprus hircus*)

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With 1 figure and 2 tables

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Summary

A comparative randomized crossover study was conducted to determine the pharmacokinetics of theophylline in male and female camels (*Camelus dromedarius*) and goats (*Caprus hircus*). Theophylline is an established 'probe drug' to evaluate the drug metabolizing enzyme activity of animals. It was administered by the intravenous (i.v.) route and then intramuscularly (i.m.) at a dose of 2 mg/kg. The concentration of the drug in plasma was measured using a high-performance liquid chromatography (HPLC) technique on samples collected at frequent intervals after administration. Following i.v. injection, the overall elimination rate constant (λ_z) in goats was 0.006 ± 0.00076/min and in camels was 0.0046 ± 0.0008/min ($P < 0.01$). The elimination half-life ($t_{1/2\lambda z}$) in goats (112.7 min) was lower than in camels (154.7 min) ($P < 0.01$). The apparent volume of distribution (V_z) and the total body clearance (Cl) in goats were 1440.1 ± 166.6 ml/kg and 8.9 ± 1.4 ml/min/kg, respectively. The corresponding values in camels were 1720.3 ± 345.3 ml/kg and 6.1 ± 1.0 ml/min/kg, respectively. After i.m. administration, theophylline reached a peak plasma concentration (C_{max}) of 1.8 ± 0.1 and 1.7 ± 0.2 μg/ml at a post-injection time (T_{max}) of 67.5 ± 8.6 and 122.3 ± 6.7 min in goats and camels, respectively. The mean bioavailability (F) in both goats and camels was 0.9 ± 0.2. The above data suggest that camels eliminate theophylline at a slower rate than goats.

Introduction

The camel (*Camelus dromedarius*) is an important animal in pastoral societies, where it is used in draught and transportation, and together with goats (*Caprus hircus*) constitute a significant source of milk, hide, meat and wool. Recent years have witnessed a surge in pharmacological research in these species, mostly in the field of pharmacokinetics [for recent reviews see Ali et al. (1996a) and Al-Qarawi and Ali (2000)].

It has been established that there are important differences between camels and other ruminant species in the activities and distribution of drug metabolizing enzyme activities (Ali and Elsheikh, 1992a, b), and in the metabolic pathways of certain drugs (Ali et al., 1996b). In general, and for many drugs, it has been shown that camels biotransform

and excrete drugs at a rate significantly lower than other domestic animals (Oukessou et al., 1990; Ali et al., 1998).

Several drugs, such as antipyrine (Vessel, 1979), metronidazole (Loft, 1990), and theophylline (Alberola et al., 1993) are employed as 'probe agents' for the study of drug disposition in man and animals. We have recently studied the comparative kinetic behaviour of the 'probe' drug metronidazole in camels, sheep and goats (Ali et al., 1999) and found the elimination half-life ($t_{1/2\lambda z}$) of the drug to be longer and its systemic clearance (Cl) slower in camels than in sheep or goats.

In the present work we compared the kinetic profile of another 'probe' drug, theophylline, in camels and goats. As far as we are aware, little or no kinetic study of theophylline in these species has ever been reported. Differences consistent with the previously obtained data on the *in vitro* activity of drug metabolizing enzymes in the two ruminant species (Ali and Elsheikh, 1992a,b) and with the metronidazole results (Ali et al., 1999) were found.

Materials and Methods

Animals

Six (three males and three females) healthy desert camels (4–5 years old, 300–350 kg) and six Omani goats (2–3 years old, 17–24 kg) were housed in separate shaded barns at the United Arab Emirates University farm in Al-Ain, United Arab Emirates, and provided with dried hay and water *ad libitum*. Concentrates and salt blocks were also provided.

Treatments

A randomized crossover study was conducted. The six animals in each species were divided, at random, into two groups. The first group was injected intravenously (i.v.) into the jugular vein with theophylline (2 mg/kg). The drug was administered intramuscularly (i.m.) into thigh muscle in the second group at the same dose. Blood (about 5 ml) was collected in heparinized tubes prior to the injections, and thereafter at 5, 10, 20, 30, 40, 60, 120, 180, 240, 300, 360, 480 and 600 min after the injection. The collected blood was centrifuged at 900 ***g*** for 15 min at 5°C and the separated plasma was stored at –70°C. After 3 weeks, the same treatment was repeated according to the crossover design, and blood and plasma were obtained as before.

Drug measurements

Theophylline concentrations in plasma were measured using a high-performance liquid chromatography (HPLC) method described previously (Zweers-Zeilmaker et al., 1996). Briefly, the extraction procedure involved the addition of ethyl acetate and ammonia solution, the evaporation of the ethyl acetate layer to dryness, and reconstitution in methanol. An aliquot (100 μl) of the latter was injected into an HPLC column (C18 Genesis Column, 4 μm particles, length 25 cm, internal diameter 4.6 mm) fitted on an HPLC system with an ultraviolet detector (Shimadzu, Japan) set at 273 nm. The mobile phase was water:methanol (70:30 v/v), and was used at a flow rate of 1 ml/min. The inter-assay coefficient of variation was less than 10 % ($n = 6$).

Kinetic analysis

Theophylline plasma concentration–time data obtained for each animal were analysed using a program for non-linear regression analysis (MedUSA). For the i.v. data, the appropriate pharmacokinetic model was determined by visual examination of individual concentration–time curves and by application of Akaike's Information Criterion (Yamaoka et al., 1978). The plasma concentration–time relationship was best estimated as a two-compartment open model. The i.m.

injection data were analysed by adopting a one-compartment open model. Semilogarithmic plots of mean plasma concentrations versus time were constructed and the terminal linear (elimination) components of the decay curves were determined (Gibaldi and Perrier, 1982). Least squares regression analysis was carried out on this terminal decay component and it was extrapolated to time 0 (Steel and Torrie, 1980). Using the method of residuals, the rapid decay components (distribution) of the curve were determined (Baggot, 1977; Gibaldi and Perrier, 1982). All graphs for theophylline yielded bi-exponential curves described by the equation: $C = C_1e^{-\lambda 1t} + C_ze^{-\lambda zt}$, where C is the drug plasma concentration at any time t, C_1 and C_z are the zero time intercepts of the plasma concentrations determined by extrapolation of the distribution and elimination phases, respectively, of the disposition curve, and λ_1 and λ_z are the distribution and elimination rate constants, respectively. From the constants so derived, pharmacokinetic parameters were calculated according to the equations previously described (Baggot, 1977; Gibaldi and Perrier, 1982).

Statistical analysis

The harmonic means of the half-lives and the means ± standard deviation of the other parameters were calculated. The Wilcoxon rank sum test (Gad and Weil, 1986) was used to test for significant differences in the half-lives. Differences between means were evaluated using Student's *t*-test. *P*-values less than 0.05 were considered significant.

Results

Akaike's Information Criterion test indicated that a two-compartment model best represented the plasma concentration versus time data after i.v. injection of theophylline in camels and goats. Following i.m. administration, the data were best represented by a one-compartment model.

The pharmacokinetic parameters calculated after i.v. administration of theophylline in camels and goats are presented in Table 1. Figure 1 shows the comparative profile of mean plasma theophylline concentrations following i.v. and i.m. administration in camels and goats. A higher body clearance and subsequently a shorter elimination half-life and larger area under the curve were seen for goats compared with camels ($P < 0.01$). There

Table 1. Pharmacokinetic parameters (mean ± standard deviation) of theophylline in camels and goats after a single intravenous injection at the dose rate of 2 mg/kg bodyweight

Pharmacokinetic parameter	Camels ($n = 6$)	Goats ($n = 6$)	Level of significance
C_1 (μg/ml)	4.08 ± 1.82	2.82 ± 0.53	NS
C_z (μg/ml)	3.28 ± 1.32	3.71 ± 0.46	NS
$t_{1/2\lambda 1}$ (min) (harmonic mean)	5.79	4.22	NS
$t_{1/2\lambda z}$ (min) (harmonic mean)	154.72	112.73	$P < 0.01$
λ_1 (/min)	0.15 ± 0.029	0.16 ± 0.019	NS
λ_z (/min)	0.0046 ± 0.0008	0.006 ± 0.00076	$P < 0.01$
AUC (μg/min/ml)	857.23 ± 219.46	575.25 ± 64.32	$P < 0.01$
V_c (ml/kg)	772.17 ± 148.92	841.14 ± 95.46	NS
V_z (ml/kg)	1720.31 ± 345.28	1440.06 ± 166.54	NS
Cl (ml/min/kg)	6.14 ± 0.98	8.94 ± 1.35	$P < 0.01$

NS, not significant at the 95 % confidence limits; C_1 and C_z, zero time intercepts of the plasma concentrations determined by extrapolation of the distribution and elimination phases of the disposition curve, respectively; λ_1 and λ_z, distribution and elimination rate constants, respectively; $t_{1/2\lambda 1}$, distribution half-life; $t_{1/2\lambda z}$, elimination half-life; AUC, area under the curve; Cl, total body clearance; V_c, volume of the central compartment; V_z, volume of distribution calculated by the area method.

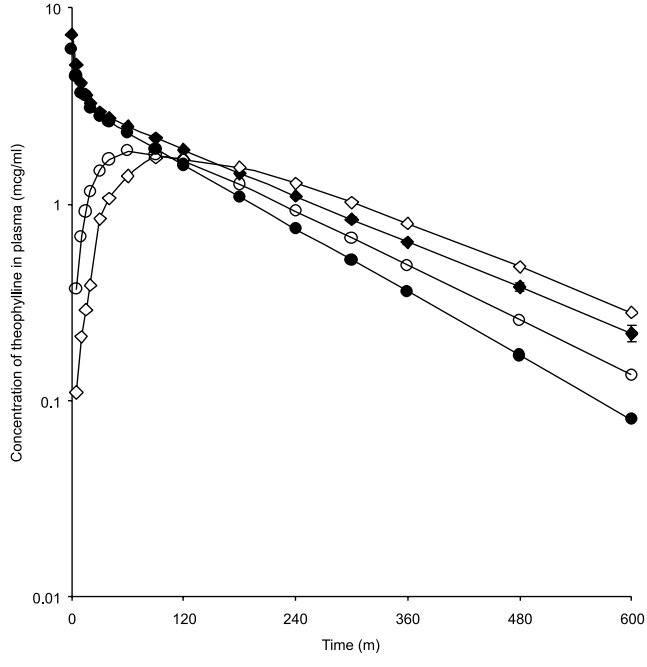

Fig. 1. Semilogarithmic plot of mean plasma concentration versus time data for theophylline after intravenous administration at a dose of 2 mg/kg in camels (◆) and goats (●), or intramuscular injection at a similar dose (◇ and ○, respectively). The standard error of the mean, not drawn, represented 10% or less of the mean values.

were no significant differences between the two species for the volume of the central compartment and the volume of distribution calculated by the area method.

Table 2 reports the pharmacokinetic parameters obtained following i.m. administration of theophylline in camels and goats. Similar theophylline peak plasma levels were reached in camels and goats. However, the time needed to reach the maximum plasma concentration (T_{max}) was shorter in goats compared with camels ($P < 0.001$). The values

Table 2. Pharmacokinetic parameters (mean ± standard deviation) of theophylline in camels and goats after a single intramuscular injection at the dose rate of 2 mg/kg bodyweight

Pharmacokinetic parameter	Camels ($n = 6$)	Goats ($n = 6$)	Level of significance
C_{max} (μg/ml)	1.72 ± 0.18	1.84 ± 0.14	NS
T_{max} (min)	122.32 ± 6.71	67.46 ± 8.59	$P < 0.001$
$t_{1/2ab}$ (min) (harmonic mean)	50.10	18.92	$P < 0.001$
$t_{1/2\lambda z}$ (min) (harmonic mean)	164.99	133.31	$P < 0.05$
MRT (min)	311.88 ± 28.12	225.05 ± 41.26	$P < 0.01$
AUC (μg/min/ml)	649.64 ± 92.29	494.38 ± 79.81	$P < 0.02$
F	0.89 ± 0.16	0.89 ± 0.19	NS

NS, not significant at the 95 % confidence limits; C_{max}, maximal plasma concentration; T_{max}, time of C_{max}; $t_{1/2\lambda z}$, elimination half-life; MRT, mean residence time; F, bioavailability of the drug obtained as $AUC_{i.m.}/AUC_{i.v.}$ where $AUC_{i.m.}$ and $AUC_{i.v.}$ are the areas under the curve of the drug after intramuscular or intravenous injection of the drug, respectively.

of the elimination half-life and the mean residence time were significantly smaller in goats ($P < 0.05$ and $P < 0.01$, respectively). Whereas the area under the curve value was larger in camels, bioavailability (F) showed no significant difference between the two species.

Discussion

Theophylline plasma concentration versus time data following i.v. and i.m. administration in camels and goats were best described by a two-compartment model. Data from the present study have clearly shown that camels eliminate theophylline at a rate significantly slower than that of goats. This drug is commonly used to evaluate the activity of drug metabolizing enzymes especially cytochrome P-450 (Loft, 1990; Alberola et al., 1993). Previous *in vitro* studies have indicated lower activity of hepatic mixed function oxidases in camels when compared with goats (Ali and Elsheikh, 1992a,b; Raza et al., 1998).

The present results corroborate these *in vivo* results, and are also in agreement with other kinetic data obtained from camels and goats given paracetamol (Ali et al., 1996b), sulphadimidine (Elsheikh et al., 1991), and metronidazole (Ali et al., 1999). In the paracetamol study, the kinetic differences between camels and goats were not limited to the slower clearance and longer half-life of the drug in camels, but also extended to differences in the metabolic pathway of the drug in the two species. In camels the predominant metabolite of paracetamol was the sulphate, whereas it was the glucuronide conjugate in the goats (Ali et al., 1996b). It has also been reported that the hepatic capacity to clear caffeine from the systemic circulation is similar between sheep and cattle, but that the preferred routes of metabolism differ; the predominant metabolite being theophylline in sheep, and parathanxine in cattle (Danielson and Golsteyn, 1996). In another study, camels, unlike horses, were found not to biotransform phenylbutazone to oxyphenbutazone to any appreciable extent (Kadir et al., 1997). We have also previously found that camels metabolize and excrete the diuretic drug furosemide at a rate significantly slower than that in horses (Dyke et al., 1996; Ali et al., 1998). In the present work, unfortunately, it was not feasible to measure the concentration and kinetics of theophylline metabolites in the two species. Important information might have been obtained about the metabolic pathways of this drug in the two species. Further studies in these aspects are warranted.

The kinetic parameters of theophylline in camels in the present work differ significantly from those in horses. The elimination half-life in camels (2.6 h) is shorter than those values calculated for horses [average of 15–17 h (Errecalde et al., 1984) and 14.8 h (Ingvast-Larsson et al., 1985)]. The value of the volume of distribution obtained for camels in this study (772.2 ml/kg) is smaller than those reported for horses by Errecalde et al. (1984) (850–900 ml/kg) and by Ingvast-Larsson et al. (1985) (1020 ml/kg).

In conclusion, the present data suggest that camels eliminate theophylline at a slower rate than goats, which provides further evidence that the metabolism of xenobiotics via mixed function oxidases is significantly slower in camels than in goats.

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