Pharmacokinetics of Intravenous and Oral Flunixin in Alpacas

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Abstract

Six adult alpacas were administered 2.2mg/kg flunixin (a non-steroidal anti-inflammatory drug) intravenously (IV), then orally. These animals were randomly assigned to two treatment groups, using an open, single-dose, four week, randomized cross-over design. Blood samples were taken over a 72 hour period of time, processed and frozen at -80 degrees C until analysed by a high performance liquid chromatography. Pharmacokinetic parameters for flunixin IV and oral administration were determined by a non-compartmental analysis with the assistance of the program Phoenix WinNonLin v6.

The maximum plasma concentration obtained was 30.2+/- 2.5 ug/ml IV with a mean half-life of 9.65 hours with a range of 8-11.3hours. The clearance(Cl) was 78.4+/- 19.8 ml/hr/kg and the volume of distribution (Vdss) was 0.59+/- 0.115 L/kg and the mean residence time (MRT) was7.65+/-0.79 hours and the area under the curve (AUC) was 29.4+/-6.6 ug*hr/ml. The oral half-life was much longer 16.2 +/- 5.4 hours, Cl/Fas (380+/- 162) L/hr/kg and the Varea/F was 9.13+/- 6.44 L/kg. The bioavailability was erratic and ranged from 12-27%. As is seen with other drugs administered orally to alpacas, oral absorption is not a reliable route of administration due to destruction of the drug through enzymatic pathways and there is evidence of hepatic or intestinal recycling leading to secondary peaks and longer half-lives.

Introduction

Non-steroidal anti-inflammatory drugs are commonly used in veterinary medicine as an analgesic, anti-inflammatory and antipyretic therapy. These drugs are currently being administered to small animals, horses, cattle and even llamas with a species specific drug study. Alpacas have not been studies as a species for treatment. This study was completed to develop a dosage regimes specific to alpacas that has not been extrapolated from other species such as horses and cattle. This could cause a problem as water metabolism in alpacas is different than that of true rumen species. (Rubsamen & Engelhardt, 1975). This may affect all pharmacokinetic parameters resulting in slow metabolism and excretion. The major difference in species is metabolism and excretion, and this category of drug is heavily metabolized and slowly excreted in other species, therefore dosage parameters must be established for alpacas.(Kopcha & Ahl, 1989). The half-life of flunixin is different in cattle, horses and other species which affects the therapy and dosage administration time of the drug. These drugs have a narrow therapeutic index that could cause over-dosing and serious side effects such as renal and hepatic toxicity and gastrointestinal ulcers. The purpose of this study was to determine the pharmacokinetics of flunixin meglumine in alpacas and establish a dosage guideline for veterinarians.

Materials and Methods

Six adult alpacas ranging in weight (51-94kg) were administered a single dose of 2.2mg/kg flunixin (a non-steroidal anti-inflammatory drug) intravenously (IV), then orally. These animals were randomly assigned to two treatment groups, using an open, single-dose, four week, randomized cross-over design. Blood samples were taken over a 72 hour period of time from a separate catheter, processed and frozen at -80 degrees C until analysed by a high performance liquid chromatography.

Materials and Methods cont.

HPLC Assay

Plasma samples were analyzed by high performance liquid chromatography using a published method (Hardee & Lai, 1982) with modifications (Navarre et. al., 2001). The HPLC system consisted of a Waters pump and auto-injector and a variable wavelength Jasco UV-VIS detector interfaced with an integrator. The mobile phase consists of acetonitrile, methanol and 1 % acetic acid solution (40:30:30) and the elution was done on a C-18 column at a wavelength of 254 nm, and a flow rate of 1.2 ml/minute. The amount of flunixin in each sample was quantified based on a peak area ratio method with respect to the internal standard, naproxen.

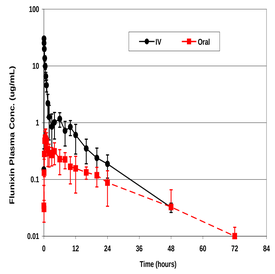
The standard curve was prepared for flunixin plasma concentrations of 0.002 to 100 ug /mL. The inter- and intra-day variation was < 8%. The extraction efficiency for flunixin and the internal standard were 97% and 99%, respectively.

Extraction procedure:

To 0.5 ml of plasma 1.5 ml of acetonitrile containing (1 □g/ml) naproxen was added in order to precipitate the proteins. The samples were then vortexed for 30 seconds and then centrifuged at 2500 rpm for 15 minutes. The supernatant was transferred to a fresh tube. To the remaining pellet, 1 ml of acetonitrile was added followed by vortexing for 15 seconds and centrifuging at 2500 rpm for 5 minutes. Both the supernatants were pooled together and evaporated to dryness, at 38°C, under a gentle stream of nitrogen. The residue remaining after evaporation was reconstituted in 0.75 ml of mobile phase and a 50 □l aliquot of the same was injected into the HPLC system.

Pharmacokinetic Analysis:

Pharmacokinetics parameters for flunixin following single IV and oral administration were determined by non-compartmental analysis with the assistance of the program Phoenix WinNonLin v6. The terminal decline in the LN plasma concentrations was used to identify the elimination rate constant (λz) based on 1/y weighting. The linear/log trapezoidal rule method was applied to estimate the AUC and its moment, AUMC. Total body clearance (Cl), apparent total body clearance after oral dosing (Cl/F), volumes of distribution (Varea, Vss), and mean residence time (MRT) were obtained from individual values of AUC, AUMC, and t1/2. An estimate of the extent of oral absorption was made based on the ratio of AUCs for oral to IV. No compartmental pharmacokinetic analysis was performed since some IV and oral plasma concentration profiles showed secondary peaks suggesting drug redistribution or recycling.



Pharmacokinetics of IV and oral flunxin

Table 1

Route	Subject	Dose	Cmax	AUCt	AUC	t1/2	Cl	Varea	Vss	MRT
		(mg/kg)	(ug/mL)	(ug*hr/mL)	(ug*hr/mL)	(hr)	(mL/hr/kg)	(L/kg)	(L/kg)	(hr)
IV	A	2.2	29.4	25.6	26.0	9.89	84.6	1.207	0.605	7.15
IV	B	2.2	35.0	38.3	38.6	7.52	57.0	0.619	0.427	7.49
IV	C	2.2	28.7	32.4	32.5	8.77	67.8	0.858	0.615	9.08
IV	D	2.2	27.9	19.0	19.4	11.57	113.4	1.893	0.780	6.88
IV	E	2.2	30.2	32.1	32.5	8.64	67.6	0.843	0.539	7.96
IV	F	2.2	29.8	26.8	27.4	11.52	80.2	1.332	0.591	7.37
Mean			30.2	29.0	29.4	9.65	78.4	1.125	0.593	7.65
SD			2.5	6.7	6.6	1.65	19.8	0.458	0.115	0.79
%CV			8.3	23.1	22.5	17.1	25.2	40.7	19.4	10.3
GeoMean			30.1	28.3	28.7	9.53	76.5	1.053	0.584	7.62

Cmax = maximum observed plasma concentration
AUCt = area under the plasma concentration versus time curve till the last measurable time
AUC = area under the plasma concentration versus time curve till infinity
t1/2 = half-life
Cl = total body clearance
Varea = volume of distribution based on AUC
Vss = apparent volume of distribution at steady-state
MRT = mean residence time

Table 2

Route	Subject	Dose	Cmax	Tmax	AUCt	AUC	t1/2	Cl/F	Varea/F	MRT	MRTabs	F
		(mg/kg)	(ug/mL)	(hr)	(ug*hr/mL)	(ug*hr/mL)	(hr)	(L/hr/kg)	(L/kg)	(hr)	(hr)	
Oral	A	2.2	0.341	3.00	3.03	3.33	23.2	661	22.09	23.15	15.99	0.128
Oral	B	2.2	0.609	0.75	7.72	8.22	20.4	268	7.88	28.77	21.28	0.213
Oral	C	2.2	0.347	0.50	6.57	7.77	18.0	283	7.37	27.42	18.34	0.239
Oral	D	2.2	0.601	0.33	4.40	4.54	9.9	485	6.90	13.34	6.46	0.234
Oral	E	2.2	0.917	0.50	8.55	8.81	15.2	250	5.48	17.29	9.32	0.271
Oral	F	2.2	0.637	0.75	6.50	6.55	10.4	336	5.05	13.41	6.04	0.239
Mean			0.575	0.97	6.13	6.54	16.2	380	9.13	20.56	12.91	0.221
SD			0.214	1.01	2.07	2.18	5.4	162	6.44	6.86	6.49	0.049
%CV			37.2	103.5	33.7	33.4	33.2	42.5	70.6	33.3	50.3	22.2
GeoMean			0.542	0.72	5.79	6.18	15.4	356	7.91	19.59	11.47	0.215

Cmax = maximum observed plasma concentration
Tmax = time of maximum plasma concentration
AUCt = area under the plasma concentration versus time curve till the last measurable time
AUC = area under the plasma concentration versus time curve till infinity
t1/2 = half-life
Cl/F = apparent total body clearance following oral administration
Varea /F= apparent volume of distribution based on AUC after oral administration
MRT = mean residence time
MRT = mean residence absorption time
F = extent of oral absorption

Results

Following a single 2.2mg/kg intravenous dose of flunixin, the maximum plasma concentration obtained was 30.2+/- 2.5 ug/ml IV with a mean half-life of 9.65 hours and a range of 8-11.3 hours. The clearance(Cl) was 78.4+/- 19.8 ml/hr/kg and the volume of distribution (Vdss) was 0.59+/- 0.115 L/kg and the mean residence time (MRT) was7.65+/-0.79 hours and the area under the curve (AUC) was 29.4+/-6.6 ug*hr/ml. The same oral dose of flunixin (2.2mg/kg) was administered and the half-life was much longer 16.2 +/- 5.4 hours, Cl/F as (380+/- 162) L/hr/kg and the Varea/F were 9.13+/- 6.44 L/kg. Larger intersubject variations were observed in the oral route pharmacokinetic parameters with coefficient of variations as high as 37.2% with Cmax and 103.5% with Tmax. (See Table I and II and Graph I)

Discussion

The intravenous half-life of flunixin in alpacas (8-11.3 hours) was much larger than llamas, (1.47 +/- 0.61 hr), sheep (3.4-3.8hr) and cattle (3.1 and 8.1 hrs) and horses (1.5 and 4.2 hrs). There was greater assay sensitivity to the terminal phase which may account for a difference in half-lives, but there is a distinct difference between llamas and alpacas which may be due to a difference in pharmacokinetic modelling. Clearance is the same order of magnitude as llamas.

The bioavailability was erratic and ranged from 12-27% with a mean of 22.1% and a large standard deviation. As is seen with other drugs administered orally to alpacas, oral absorption is not a reliable route of administration due to destruction of the drug through enzymatic pathways and there is evidence of hepatic or intestinal recycling leading to secondary peaks and longer half-lives.

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