

PHARMACOKINETICS AND BIOAVAILABILITY OF KETOPROFEN AFTER SINGLE DOSE INTRAVENOUS AND INTRAMUSCULAR ADMINISTRATION IN CATTLE

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Pharmacokinetics of Ketoprofen was determined following single dose intravenous (IV) and intramuscular (IM) administration at the dose rate of 3 mg/kg body weight in cow calves. Drug concentration in plasma was determined using High Performance Liquid Chromatography. Following single dose intravenous administration, the drug was rapidly distributed ($t_{1/2\alpha}$: 0.10 ± 0.01 h) and eliminated ($t_{1/2\beta}$: 1.55 ± 0.05 h; Cl_B : 4.82 ± 0.16 ml/min/kg) from the body. Following intramuscular administration, the drug was rapidly absorbed (C_{max} : 6.15 ± 0.24 μ g/ml; T_{max} : 5 h) and eliminated ($t_{1/2\beta}$: 3.40 ± 0.05 h) from the body. The mean residence time (MRT) and bioavailability following intramuscular administration were 4.22 ± 0.07 h and 77.31 ± 2.23 %, respectively. The pharmacokinetic profile indicated that ketoprofen can be used to treat various inflammatory conditions cow calves.

Key words:

Ketoprofen, Pharmacokinetics, Cattle

Ketoprofen is an aryl propionic acid derivative, non-selective cyclooxygenase (COX) inhibitor non-steroidal anti-inflammatory drug (NSAID) with two enantiomers *viz.* S(+) ketoprofen and R(-) ketoprofen (Sweetman, 2002). It produces reversible COX inhibition by competing with the substrate, arachidonic acid, for the active site of the enzyme (Vane *et al.*, 1998). Commercially available products contain a racemic (50:50) mixture of the two enantiomers. In veterinary medicine, ketoprofen is used to lower body temperature in animals with fever, to relieve

respiratory signs in calf and piglet pneumonia and to relieve pain in conditions as diverse as equine colic and joint diseases of the horse and dog, as well as for the control of traumatic and postoperative pain in all species (Lees *et al.*, 2004). The present investigation was undertaken to establish intramuscular bioavailability, and pharmacokinetic parameters of ketoprofen following intravenous (i/v) and intramuscular (i/m) administration at dose rate of 3 mg/kg body weight in cow calves.

MATERIALS AND METHODS

The study was conducted in six clinically healthy crossbred (Holstein-Friesian x Kankrej) male calves, aged between 6 to 12 months with the average body weight of 91 kg. The calves were kept at Instructional Farm, College of Veterinary Science and Animal Husbandry, Anand Agricultural University, Gujarat State, India. Calves were housed in experimental calf pen two weeks prior to experiment for acclimatization; the animals were fed concentrates, green fodder and roughage and had free access to water. All essential and standard managemental measures were followed to keep the calves free from stress.

The study was planned in a crossover design with ketoprofen administered both i/v and i/m to each animal with a 15 days washout period between administrations. Ketoprofen injection (Neoprofen[®], racemic mixture; 100 mg/ml; Vetnax Ranbaxy Fine Chemicals Limited, New Delhi) was administered i/v at the dose rate of 3 mg/kg through left jugular vein, while i/m injection was given in the lateral deep neck muscles. Blood samples

were collected into heparinized tubes from intravenous catheter (Venflon, 22 x 0.9 x 25 mm) fixed into the right jugular vein at 0, 2, 5, 10, 15, 30, 45 min and 1, 2, 4, 8, 12, 18, 24 h after i/v administration. Following i/m administration the blood samples were collected at 0, 5, 10, 15, 30, 45 min and 1, 2, 4, 8, 12, 18, 24, and 36 h. Plasma was separated after centrifugation (15 min, 2000g, 25 °C) of blood samples and stored at -40 °C and analyzed within 24 h.

Plasma Ketoprofen concentration was analyzed by reverse-phase high performance liquid chromatography (HPLC) as per the validated method described by Wanwimolruk *et al.* (1991), with minor modifications. Chromatographic separation was performed by using reverse phase C₁₈ column at room temperature and effluent was monitored through UV detector at 258 nm with flow rate of mobile phase kept at 1.5 ml/min. The retention time of ketoprofen was 10.4 min. Calibration curve were prepared daily by adding known amount of ketoprofen to blank unfortified plasma for the expected range of concentrations from 0.05 to 50 µg/ml, and accepted only if it had a R² value > 0.99. The lower level of quantification (LLOQ) was 0.05 µg/ml. The

lowest mean recovery of ketoprofen from plasma was 81.45% found at 0.05 µg/ml. Precision and accuracy were determined with known concentrations of 0.05, 1.0, and 50 µg/ml in plasma (5 replicates each/day). The intraday and inter-day coefficients of variation were satisfactory and were under acceptable limits with relative standard deviations (RSD) less than 10%.

Pharmacokinetic parameters were determined for each calf by non-compartmental analysis with commercial software (PK solution version 2.0, USA). Following oral administration of the drug, maximum concentration (C_{max}) and time to reach the maximum concentration (T_{max}) were determined from the concentration-time curve. Intramuscular bioavailability of the drug was calculated using the following formula (Wagner, 1967).

$$F (\%) = [\text{AUC (IM)} \times t_{1/2\beta} (\text{IV})] / [\text{AUC (IV)} \times t_{1/2\beta} (\text{IM})] \times 100$$

RESULTS AND DISCUSSION

The mean plasma concentration-time curves for ketoprofen after single dose i/v and i/m administration (3 mg/kg) in calves are presented in Figure I. Following i/v administration, plasma drug concentration of

Table I. Pharmacokinetic parameters of ketoprofen after single i/v and i/m administration (3.0 mg/kg body weight) in calves (n=6)

Pharmacokinetic parameters	Unit	Intravenous (Mean±S.E.)	Intramuscular (Mean±S.E.)
C _{p0}	µg/ml	34.13±2.57	-----
C _{max}	µg/ml	-----	6.15±0.24
T _{max}	h	-----	0.50±0.00
t _{1/2α}	h	0.10±0.01	-----
t _{1/2β}	h	1.55±0.05	3.40±0.05
AUC	µg.h/ml	10.42±0.32	17.72±0.39
AUMC	µg.h ² /ml	12.37±0.38	74.93±2.02
V _{d(area)}	L/kg	0.64±0.03	----
V _{d(ss)}	L/kg	0.35±0.02	0.72±0.02
Cl _B	ml/kg/min	4.82±0.16	2.83±0.06
MRT	h	1.20±0.06	4.22±0.07
F	%	-----	77.31±2.23

Abbreviations: C_{p0}: theoretical plasma concentration of drug at zero-time; C_{max}: Maximum drug concentration; T_{max}: observed time for C_{max}; t_{1/2α}: distribution half life; t_{1/2β}: elimination half life; AUC: area under plasma drug concentration-time curve; AUMC: area under first moment of curve; V_{d(area)}: apparent volume of distribution; V_{d(ss)}: volume of distribution at steady state; Cl_B: total body clearance; MRT: mean residence time; F: bioavailability.

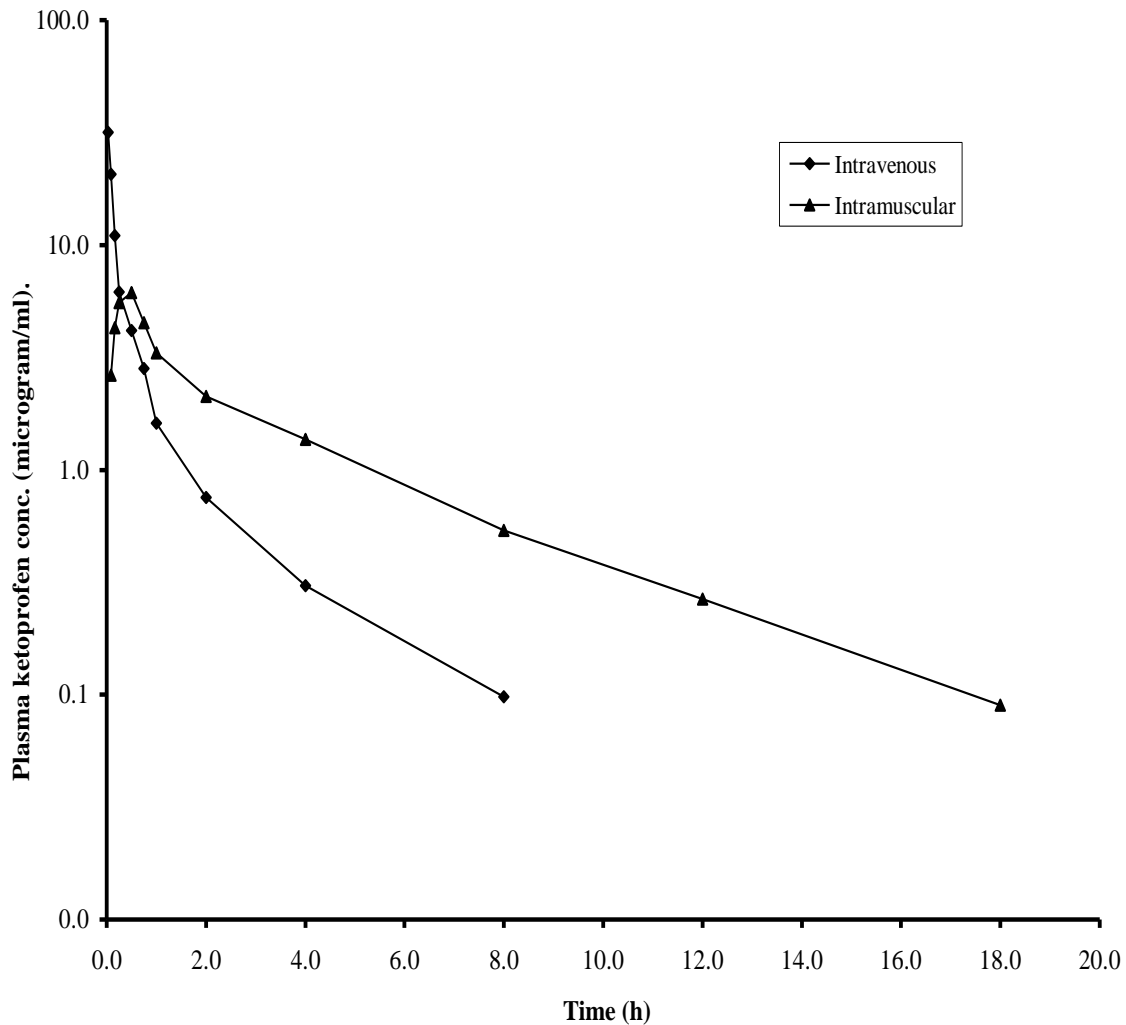


Figure I. Plasma concentration versus time profile curve of ketoprofen @ 3.0 mg/kg of body weight following single dose i/v and i/m administrations (n=6)

31.63±1.71 µg/ml was achieved at 2 min and declined gradually to 0.098±0.004 µg/ml at 8 h. Following i/m administration, mean peak plasma concentration (C_{max}) of ketoprofen (6.15±0.24 µg/ml) was achieved at T_{max} of 0.50 h (30 min).

Pharmacokinetic parameters derived following i/v and i/m administration of the drug are presented in the Table I. Rapid distribution and a slower elimination were observed after i/v administration of ketoprofen with high distribution rate constant (7.50±0.86 /h) and low elimination rate constant (0.45±0.01 /h). Higher value of distribution rate constant and lower distribution half life (0.10±0.01 h) indicate that ketoprofen is rapidly distributed from central to peripheral compartment. The distribution half-life ($t_{1/2\alpha}$), elimination half-

life ($t_{1/2\beta}$), apparent volume of distribution ($V_{d(\text{area})}$), area under plasma drug concentration-time curve (AUC), and total body clearance (Cl_B) were 0.10±0.01 h, 1.55±0.05 h, 0.64±0.03 L/kg, 10.42±0.32 µg.h/ml and 4.82±0.16 ml/kg/min, respectively. The elimination half-life (1.55 h) of ketoprofen following i/v route observed in the present study is in agreement with that reported in mare (1.63 h; Sams *et al.*, 1995) but more rapid elimination of drug has been reported following i/v administration in sheep (0.86 h; Landoni *et al.*, 1999) and horse (1.02 h; Owens *et al.*, 1995). Such variations in elimination half-life might be attributable to interspecies and inter-individual variation. Values reported for volume of distribution at steady state (0.35±0.02 L/kg) following intravenous administration in present study is in

accordance with that for goat (Arifah *et al.*, 2003) and cows (Igarza *et al.*, 2004). The moderate volume of distribution following i/v administration of ketoprofen in cow calves in the present study is expected as ketoprofen is also highly bound to plasma proteins, similar to most NSAIDs. The total body clearance (Cl_B) following i/v administration observed in the present study is in agreement with the reported values for both enantiomers of ketoprofen in sheep (Landoni *et al.*, 1999) and goat (Arifah *et al.*, 2003).

Following i/m administration, the area under curve (AUC), elimination half-life, total body clearance and systemic bioavailability (F) were 17.72 ± 0.39 $\mu\text{g}\cdot\text{h}/\text{ml}$, 3.40 ± 0.05 h, 2.83 ± 0.06 ml/kg/min and 77.31 ± 2.23 %, respectively. The longer elimination half-life of ketoprofen (3.40 ± 0.05 h) following i/m administration observed in the present study indicates that the drug being continuously absorbed during the elimination phase also. Bioavailability of ketoprofen (77.31 %) obtained in the present study is similar to 71-96 % reported in human (Jamali and Brocks, 1990). However, in camel (Alkatheeri *et al.*, 1999) and Japanese quail birds (Graham *et al.*, 2005) it was reported to be 121.1 and 56 %, respectively showing high inter-species variation. Longer elimination half-life, extensive volume of distribution at steady state and slower body clearance of ketoprofen following i/m administration as compared to i/v administration found in present study, makes it more suitable for intramuscular use in calves.

CONCLUSIONS

Pharmacokinetics and intramuscular bioavailability of non-steroidal anti-inflammatory drug ketoprofen (3 mg/kg) was investigated in six crossbred cow calves after its single dose intravenous and intramuscular administration in a cross-over design study. Drug concentration in plasma was determined by high performance liquid chromatography and the data obtained were subjected to non-compartmental analysis. Following i/v and i/m administration of ketoprofen, values of elimination half-life (1.55 ± 0.05 and 3.40 ± 0.05 h), volume of distribution of drug at steady state

(0.35 ± 0.02 and 0.72 ± 0.02 L/kg) and total body clearance (289.28 ± 9.31 and 169.75 ± 3.51 ml/h/kg) were determined, respectively. The systemic bioavailability of ketoprofen following intramuscular administration in the calves was 77.31 per cent. Good bioavailability and plasma concentration profile with pharmacokinetic parameters supports intramuscular use of ketoprofen in calves as convenient approach.

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