Pharmacokinetics of meloxicam in adult goats: a comparative study of subcutaneous, oral and intravenous administration

U Karademir, H Erdogan, M Boyacioglu, C Kum, S Sekkin & M Bilgen

To cite this article: U Karademir, H Erdogan, M Boyacioglu, C Kum, S Sekkin & M Bilgen (2016) Pharmacokinetics of meloxicam in adult goats: a comparative study of subcutaneous, oral and intravenous administration, New Zealand Veterinary Journal, 64:3, 165-168, DOI: 10.1080/00480169.2015.1124811

To link to this article: http://dx.doi.org/10.1080/00480169.2015.1124811
Pharmacokinetics of meloxicam in adult goats: a comparative study of subcutaneous, oral and intravenous administration

U Karademir*§, H Erdogan†, M Boyacioglu*, C Kum*, S Sekkin* and M Bilgen‡

Abstract

AIMS: To determine the plasma disposition of meloxicam in goats following S/C, oral or I/V administration at a single dose of 0.5 mg/kg bodyweight.

METHODS: Five healthy Saanen goats, aged 12–14 months and weighing 35–40 kg, were used for a three phase cross-over design with a 10-day washout period, with meloxicam administered I/V, then orally and S/C. Heparinised blood samples (5 mL) were collected from all animals prior to drug administration (0 hours) and subsequently up to 96 hours. Concentrations of meloxicam in plasma were measured using high performance liquid chromatography. Concentration-time curves were fitted and pharmacokinetic parameters were estimated for each administration group.

RESULTS: Subcutaneous administration of meloxicam exhibited unique plasma distribution characteristics that differed from oral and I/V administration. Mean peak plasma concentrations were greater (1.91 (SD 0.39) vs 0.71 (SD 0.17) µg/mL) and the time to reach them shorter (3.20 (SD 1.64) vs 14.33 (SD 2.19) hours) following S/C compared with oral administration (p<0.05). The terminal half-life was longer (15.16 (SD 4.74) hours) following S/C compared with oral administration vs 10.69 (SD 1.49) hours) and the MRT was shorter (15.67 (SD 3.27) hours vs 24.33 (SD 3.12) hours) following S/C than oral administration (p<0.05), but bioavailability was similar (98.24 (SD 9.62) vs 96.49 (SD 10.71)%).

CONCLUSION AND CLINICAL RELEVANCE: Subcutaneous administration of meloxicam resulted in long-term presence of drug at high concentration in goat plasma. This unique plasma disposition characteristic may offer an advantage in some clinical cases towards potentially improving the treatment efficacy in goats.

KEY WORDS: Goats, meloxicam, NSAID, pharmacokinetics

Introduction

Nonsteroidal anti-inflammatory drugs (NSAID) are used in domestic animal species for treating a number of inflammatory conditions, including musculo-skeletal disorders, soft tissue injuries, mastitis and pneumonia (Mathews 2002; Friton et al. 2005). The common mechanism of action for this class of drugs is attributed to the inhibition of cyclooxygenase which blocks the biosynthesis of prostaglandins, prostacyclins and thromboxanes (Botting 2006). The action is preferential but not specific to cyclooxygenase - 2 isoenzyme whose expression is induced by inflammatory stimuli under various pathophysiological conditions (Mitchell et al. 1993; Curry et al. 2005). The drug meloxicam belongs to the enolic acid class of NSAID, and has analgesic, anti-inflammatory, and antipyretic properties.

Plasma dispositions of meloxicam have been reported in the literature when administered to ruminants by oral and I/V routes (Shukla et al. 2007; Coetzee et al. 2009; Ingvast-Larsson et al. 2010; Stock et al. 2013). However, to the best of our knowledge, there are no published data for the S/C administration of meloxicam to ruminants, as this would have important implications for its rational therapeutic use in these species. Thus, in this study, the plasma distribution characteristics of meloxicam following S/C administration were determined in goats. The temporal profile of the concentration in plasma was compared against those obtained following oral and I/V administration, with the drug dose kept identical for all three routes.

Materials and methods

The study was approved by the Animal Ethics Committee of Adnan Menderes University (Aydin, Turkey). It was carried out using a three-phase cross over design with a 10-day washout period. Five healthy Saanen goats, aged 12–14 months and weighing 35–40 kg were acquired from the Agricultural Faculty of the University of Adnan Menderes (Isikli Koyu, Aydin, Turkey) and identified by specific name tags attached to their ears. The animals were housed in single pens and fed twice daily at 08:00 and 16:00

| AUC | Area under the concentration curve from time zero to infinity |
| C_max | Maximum plasma concentration |
| MRT | Mean residence time |
| NSAID | Nonsteroidal anti-inflammatory drug(s) |
| t_1/2 | Terminal half-life |
with a mixture of hay and concentrate feed. Water was supplied ad libitum and mineral licks were provided with free access. Meloxicam was administered to each animal via the S/C, oral or I/V route at three different time points separated by 10 days of washout period. The amount of drug for each administration route was prepared from commercial formulations for delivery at a single dose of 0.5 mg/kg bodyweight. This dose was consistent with previous studies using goats and sheep (Shukla et al. 2007; Ingvast-Larsson et al. 2010; Stock et al. 2013); and those used in calves (Coetzee et al. 2009).

During the study, the animals were evaluated visually for their behaviour, feeding habits, and ruminations assessed by auscultation.

**Treatment groups and blood sampling**

One day before starting the treatment with meloxicam, a catheter was surgically placed in the jugular vein of each animal for blood withdrawal. The catheter remained in place and was flushed with 2% heparin solution throughout the study. Drug-free blood samples (5 mL) were collected from all animals into heparin-containing tubes and then centrifuged at 3000g for 20 minutes. The resulting plasma was transferred to plastic tubes, stored at −20°C and later used for analytical method development, validation purposes or reference measurements.

The first phase of the study involved I/V administration. Meloxicam (Bavet Meloxicam, solution for injection, 5 mg/mL, Bavet, Istanbul, Turkey) was delivered through the catheter and then flushed with 10 mL of saline solution (0.9% NaCl). Blood samples were collected into heparin-containing tubes prior to drug administration (0 hours) and at 0.25, 0.5, 1, 2, 3, 4, 5, 6, 7, 8, 10, 12, 16, 24, 32, 48, 56, 72 and 96 hours. A 10 day washout period from the time of the drug delivery was allowed, as this time duration was reported to be adequate for the clearance of the drug from the plasma in ruminants (Coetzee et al. 2009; Ingvast-Larsson et al. 2010; Stock et al. 2013).

In the second phase, the goats received oral meloxicam suspension (Metacam, oral suspension, 1.5 mg/mL, Boehringer Ingelheim Vetmedica, Malmö, Sweden) placed on the back of the tongue and then flushed with 10 mL of saline solution (0.9% NaCl). Blood samples were collected into heparin-containing tubes prior to drug administration (0 hours) and at 0.25, 0.5, 1, 2, 3, 4, 5, 6, 7, 8, 10, 12, 16, 24, 32, 40, 48, 56, 72 and 96 hours. A 10 day washout period from the time of the drug delivery was allowed, as this time duration was reported to be adequate for the clearance of the drug from the plasma in ruminants (Coetzee et al. 2009; Ingvast-Larsson et al. 2010; Stock et al. 2013).

Immediately after collection, each blood sample was centrifuged at 3000g for 20 minutes and the resulting plasma was transferred to plastic tubes and stored at −20°C for analysis.

**Analytical procedures**

The analytical procedures were performed using a liquid-liquid phase extraction followed by high performance liquid chromatography, as modified from Baert and De Backer (2002). Firstly, the protocol was tested and validated using drug-free plasma samples mixed with a known amount of meloxicam. Subsequently plasma samples collected from the goats after administration of meloxicam were analysed for meloxicam content via its parent compound (C14H13N3O4S2).

Stock solution (100 µg/mL) of pure standard of meloxicam (Batch No: MX1259FT06; Dr. Reddy’s Laboratories Ltd., Banjara Hills, Hyderabad, Telangana, India) was first prepared using acetonitrile as solvent. The resulting solution was then further diluted to produce 0.05, 0.5, 1, 2.5, 5 and 10 µg/mL solutions for calibration of standard curves and to add to drug-free plasma samples to determine recovery.

For meloxicam analysis, drug-free plasma samples (0.5 mL) were spiked with meloxicam standards to produce final concentrations of 0.05, 0.1, 0.5, 1, 2.5, 5 and 10 µg/mL. Hydrochloric acid (150 µL, 37%, 12 M) was added, and after vortexing for 15 seconds, the solution was mixed with ethyl acetate (4 mL) and shaken on a slow rotary mixer for another 15 minutes. After centrifugation at 2000g for 15 minutes, the organic phase (3 mL) was transferred to a thin-walled 10 mL-conical glass tube and evaporated to dryness at 45°C in a sample concentrator (Maxi-dry plus, Heto Holten, Allerod, Denmark). The dry residue was dissolved in 250 µL of mobile phase and 35 µL of this solution was injected into the chromatographic system. The mobile phase consisted of acetonitrile-deionised water (65:35, v/v) and was delivered (Agilent 1100 Series QuatPump, Waldron, Germany) at a flow rate of 1 mL/minute. An inertsil ODS-3 analytical column (5 µm, 4.6 mm × 150 mm, GL Sciences, Tokyo) was used in the analyses. The eluate was continuously monitored using a photodiode array detector (Agilent 1100 Series, PDA, Waldron, Germany) at a wavelength of 355 nm.

The analytical method used for measuring the concentration of meloxicam in goat plasma was validated prior to starting the study using recommendations developed by the Veterinary International Conference on Harmonization (VICH; Anonymous 1999a, 1999b). The analytes were identified by comparison with the retention times of the pure reference standards. Recovery of the molecule under study was measured by comparison of the peak areas from spiked plasma samples with the areas resulting from the direct injection of analytical standard prepared in acetonitrile. The inter- and intra-assay precisions of the extraction and chromatography procedures were evaluated by processing six replicates of the aliquots of the drug-free goat plasma samples containing known amounts of the drugs on six different days. Calibration graphs were prepared (linear range 0.05–10 µg/mL meloxicam). The detection limit of the molecule was established using high performance liquid chromatography analysis of blank plasma spiked with the analytical standard, measuring the baseline noise at the retention time of the peak. The mean baseline noise at the peak retention time plus 3 SD was defined as the detection limit. The mean baseline noise plus 5 SD was defined as the limit of quantification (Shrivastava and Gupta 2011).

**Pharmacokinetic and statistical analysis of data**

A curve was fitted to the plasma concentration versus time data using the WinNonlin software program (Win Nonlin version 5.2, Pharsight Corporation, Mountain View, CA, USA). Pharmacokinetic parameters for each drug treatment were estimated using non-compartmental model analysis (Ingvast-Larsson et al. 2010). The maximum plasma concentration (Cmax) and time to reach maximum concentration (tmax) were obtained from the plotted concentration-time curves. The area under the curve (AUC) and mean residence time (MRT) from time zero to infinity were calculated by trapezoidal rule (Gibaldi and Perrier 2007). Terminal half-life (t1/2) was calculated as:

\[ t_{1/2} = \frac{\ln(2)}{\lambda} \]

where λ represents the first-order rate constant associated with the terminal portion of the curve.
Summary data for the pharmacokinetic parameters were tabulated and analysed using nonparametric (Kruskal Wallis and Mann-Whitney U) tests, due to the small group size, using SPSS ver. 17.0 for Windows (SPSS Inc., Chicago USA).

Results

The analytical method used to extract and quantify the plasma concentration of meloxicam by chromatographic analysis was validated before processing the experimental samples. This analysis yielded a standard curve that was linear between 0.05 and 10 µg/mL meloxicam with a correlation coefficient of 0.999. The limits of detection and quantification were 0.01 and 0.05 µg/mL, respectively. For six replicates of aliquots, with known concentrations the CV was <15%, and the accuracy ranged from 76 to 84%. The mean extraction recovery was 80.87% (inter assay CV=4.09%). The parent molecule of meloxicam (C14H13N3O4S2) was detected after all the routes of drug administrations.

The animals tolerated the experimental procedures as evidenced by the exhibition of normal behaviour, feeding habits and ruminations assessed by auscultation. The plasma concentrations versus time curves following S/C, I/V and oral administration are presented in Figure 1 and the pharmacokinetic parameters are summarised in Table 1.

The terminal half-life was longer following S/C or I/V than oral administration, but the MRT was shorter following S/C or I/V than oral administration (p<0.05). Peak plasma concentrations were greater and the time to reach them was shorter following S/C compared with oral administration (p<0.05). The bioavailability of meloxicam did not differ following S/C or oral administration, and AUC and area under the moment concentration curve were similar following oral, S/C and I/V administration.

Discussion

This study aimed to determine the plasma disposition of meloxicam in healthy goats following S/C administration and to compare the pharmacokinetic parameters with those following oral and I/V administration at the same dose of 0.5 mg/kg body-weight. The normal behaviour exhibited by the animals following the experimental procedures showed that the S/C administration appeared as safe as oral and I/V use of meloxicam.

Key pharmacokinetic parameters were calculated from the data for meloxicam administered via different routes. Bioavailability was similar following S/C and oral administrations, but the value after oral treatment was higher than that reported previously (79%) by Ingvast-Larsson et al. (2010). Those authors worked with Swedish landrace breed of goats and the drug administration time relative to the feeding regimen was not given. The feeding protocol influences the absorption of NSAID as drugs bind to the feed and may be the reason for the difference in bioavailability.

Nevertheless the results of the current study following oral and I/V administration agree with other previously published studies, confirming that our experimental procedures and measurement protocols were properly implemented. Following I/V administration of the same dose of meloxicam, Ingvast-Larsson et al. (2010) reported similar values for $t_{1/2}$ (10.09 hours), MRT (13.90 hours) and AUC (29.73 µg.hour/mL).

Table 1. Pharmacokinetic parameters measured following subcutaneous, oral and intravenous administration of 0.5 mg/kg meloxicam to five healthy goats.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Subcutaneous</th>
<th>Oral</th>
<th>Intravenous</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean ± SD</td>
<td>Min, Max</td>
<td>Mean ± SD</td>
<td>Min, Max</td>
</tr>
<tr>
<td>Terminal half-life (hours)</td>
<td>15.16±4.74$^a$</td>
<td>11.53, 23.35</td>
<td>10.69±1.49$^y$</td>
</tr>
<tr>
<td>Mean residence time (hours)</td>
<td>15.67±2.37$^a$</td>
<td>13.93, 19.78</td>
<td>24.33±3.12$^y$</td>
</tr>
<tr>
<td>T$_{max}$ (hours)</td>
<td>3.20±1.64$^a$</td>
<td>1.00, 5.00</td>
<td>14.33±2.19$^y$</td>
</tr>
<tr>
<td>Maximum plasma concentration (µg/mL)</td>
<td>1.91±0.39$^a$</td>
<td>1.4, 2.47</td>
<td>0.71±0.17$^y$</td>
</tr>
<tr>
<td>AUC (µg.hour/mL)</td>
<td>24.65±5.41$^a$</td>
<td>16.28, 33.18</td>
<td>24.21±7.54</td>
</tr>
<tr>
<td>AUMC (µg.hour$^2$/mL)</td>
<td>546.42±155.20</td>
<td>375.06, 794.40</td>
<td>642.03±244.69</td>
</tr>
<tr>
<td>Volume of distribution (mL/kg)</td>
<td>N/A</td>
<td>N/A</td>
<td>N/A</td>
</tr>
<tr>
<td>Clearance (mL/hour/kg)</td>
<td>N/A</td>
<td>N/A</td>
<td>N/A</td>
</tr>
<tr>
<td>Bioavailability (%)</td>
<td>98.24±9.62</td>
<td>85.44, 114.97</td>
<td>96.49±10.71</td>
</tr>
</tbody>
</table>

$x,y,z$ Values in same row differ (p<0.05) based on Kruskal Wallis and Mann-Whitney U tests. 
AUC=area under the concentration curve from time zero to infinity; AUMC=area under the moment concentration curve from time zero to infinity; N/A=not applicable; 
$T_{max}$=time to reach maximum plasma concentration.
The AUC in goats is lower than reported in sheep (55.23 μg.hour/mL; Stock et al. 2013) and cattle (49.40 μg.hour/mL; Coetzee et al. 2009), implying that meloxicam is eliminated at a faster rate in goats than other ruminant species. Meloxicam is primarily metabolised into inactive polar metabolites (including 5′-hydroxymethyl and 5′-carboxy) in the liver (Mosher et al. 2011), and goats are known to possess higher activities of drug-metabolising biotransformation enzyme cytochrome P450 2C9 (Szotáková et al. 2004). This may explain the higher clearance measured in this study.

Results following oral administration in this study were also similar to those reported by Ingvast-Larsson et al. (2010) for t1/2 (11.80 hours), MRT (25.40 hours), AUC (23.24 μg.hour/mL) and Cmax (0.73 μg/mL). The time to reach maximum plasma concentration reported here (14.33 hours) was shorter than in sheep (19 hours) but longer than calves (11.6 hours), but Cmax was lower than in both sheep (1.72 μg/mL) and calves (3.10 μg/mL) (Coetzee et al. 2009; Stock et al. 2013). Recirculation within the intestinal lumen can be another reason for the slower elimination of a drug from the system. In sheep, meloxicam is absorbed mostly distal to the four-compartment gastric tract, primarily in the intestine, but is also stored within the rumen (Stock et al. 2013). In light of this, meloxicam absorption appears to be relatively rapid in goats, also possibly starting at more proximal section of the gastrointestinal tract.

The results for AUC and area under the moment concentration curve in this study were similar following all three treatments, although there were differences in t1/2 and MRT. As shown in Figure 1, S/C administration of meloxicam resulted in a shorter time to reach a higher peak plasma concentration than oral administration, suggesting relatively fast absorption and shorter clearance compared with the oral administration in goats.

In conclusion, the plasma disposition of meloxicam exhibited unique characteristics following S/C, oral and I/V routes of administration in goats. The behaviours of the critical parameters describing the concentration kinematics should be taken into account for increasing the treatment efficacy of meloxicam in goats. Plasma concentrations reached a peak faster after S/C than oral administration, and lasted for a similar time. If this is reflected at the site of action, S/C meloxicam may be a preferable route to oral administration, particularly in acute cases.

Acknowledgements

The authors would like to thank Prof. Dr. Ibrahim Gencsoylu for his expert advice with the experimental procedures carried out during the study.

References


Coetzee JF, KuKanich B, Mosher R, Allen PS. Pharmacokinetics of intravenous and oral meloxicam in ruminant calves. Veterinary Therapeutics 10(4), E1–8, 2009


*Non-peer-reviewed

Submitted 3 March 2015
Accepted for publication 18 November 2015
First published online 26 November 2015