Population pharmacokinetic analysis of blood concentrations of robenacoxib in dogs with osteoarthritis


1. Introduction

Robenacoxib is a novel and highly cyclooxygenase (COX)-2 selective member of the COXIB group of non-steroidal anti-inflammatory drugs (NSAIDs). Robenacoxib has been developed for use in cats and dogs, and is currently approved (Onsior® injection and tablets) in many countries for the treatment of pain and inflammation associated with chronic osteoarthritis (OA) and orthopedic and soft tissue surgery.

The physicochemical characteristics of this drug and the resulting pharmacological properties (e.g., COX-2 selective, tissue selective and high safety index) in the dog have been described recently (Jung et al., 2009; King et al., 2009, 2010, 2011; Silber et al., 2010). Robenacoxib’s good efficacy profile in young healthy Beagle dogs in a preclinical inflammation model (Schmid et al., 2010) was confirmed in field trials (Gruet et al., 2011; Reymond et al., 2012). However, because certain other NSAIDs are sensitive to pharmacogenetic and metabolic variability within and between dog breeds (Paulson et al., 1999; Fleischer et al., 2008; Martinez et al., 2009; Cox et al., 2011), it was judged important to evaluate the pharmacokinetic variability of robenacoxib in the target population of dogs with osteoarthritis (OA).

The objective of the present study was therefore to investigate the pharmacokinetic variability of robenacoxib using a population analysis and to identify whether covariates such as sex, age and breed, or kidney and liver variables, played a significant role in the variability of exposure.

2. Materials and methods

All three studies were conducted in France after authorization by the French authorities (AFSSA) and in compliance with Good Clinical Practice (VICH Guideline 9, 2000). Prior written owner’s consent was obtained for all dogs.
2.1. Animal phase of study COXFRA101

This study was a multi-center (13 veterinary clinics in different geographical locations in France), parallel-group design, randomized and blinded field dose establishment study comparing three different dosages of robenacoxib (0.5–1 mg/kg, 1–2 mg/kg, and 2–4 mg/kg). The test item consisted of tablets containing 10 mg or 40 mg robenacoxib (Onsior® tablets, Novartis Animal Health, Basel, Switzerland). The tablets were administered once daily orally at the same time as feeding (either mixed with the food or administered into the mouth at the time of feeding). Mean (range) dosages of robenacoxib were 0.72 (0.51–1.00) mg/kg in Group 1, 1.42 (1.00–1.91) mg/kg in Group 2 and 3.03 (2.14–3.95) mg/kg in Group 3. The positive control (meloxicam, Metacam® ad us. Vet., Oral suspension, 32 mL) was dosed according to manufacturer's recommendations: 0.2 mg/kg on the first day and then 0.1 mg/kg on subsequent days. Dogs were dosed once daily for 28 days.

The inclusion criteria were: dogs older than 6 weeks, with a bodyweight between 5 and 80 kg and with clinical signs of OA of any joint as evidenced by clinical signs of at least 3 weeks duration (e.g. lameness, morning stiffness, disuse atrophy, decreased range of motion in a joint, and crepitus), lameness and pain at palpation/mobilization diagnosed on Day 0 and confirmation of OA by X-ray. A thorough investigation of the animals’ clinical condition supported by blood chemistry and hematological analyses was performed. The exclusion criteria were: lameness associated with neoplasia, lameness associated with a primary neurological disorder or a known immunological disorder, surgery of any joint in the previous 60 days, animals intended for breeding or known to be pregnant or lactating, dogs with severe concomitant disorders (e.g. kidney, liver or gastrointestinal tract) that may have interfered with the evaluation of response to treatment, dogs which received local or systemic NSAIDs within 14 days prior to their inclusion in the study, dogs which received corticosteroids within 30 days (systemic) or 90 days (intra-articular) prior to their inclusion in the study and dogs which received polysulfated glycosaminoglycans, glucosamine or chondroitin sulfate within 30 days prior to their inclusion in the study. Dogs were allocated to treatment groups by random. Case allocation was stratified according to investigator. No separate randomization was made for sex, age or bodyweight of the dog. Blood samples (2 mL) for robenacoxib assay were collected from a vein into tubes containing EDTA on Days 2, 7, and 14 (one sample a day). The precise time of blood collection, as well as the time of the last treatment administration, was recorded. Samples were frozen and stored at −20 °C until shipment. A total of 67 dogs (17 dogs in each of the robenacoxib groups and 16 dogs in the meloxicam group) were included in this study. Samples were taken from all dogs but only animals treated with robenacoxib (51 dogs) were included in the population analysis. The overall mean ± SD age of the dogs included in this study was 9.4 ± 3.6 years with a range of 0.9–17.9 years. The mean ± SD bodyweight of the dogs was 30.2 ± 10.1 kg with a range of 6.3–57.0 kg.

2.2. Animal phase of study COXFRA102

This study was very similar to study COXFRA101 and only differences between both studies are highlighted in this section. It was a multi-center (14 veterinary clinics in different geographical locations in France), parallel-group design, randomized and blinded field dose establishment study comparing three different dosages of robenacoxib (0.25–0.5, 0.5–1 mg/kg and 1–2 mg/kg, all dosed twice daily) and the same dosage (0.2 mg/kg on Day 1 and subsequently 0.1 mg/kg) of the positive control, meloxicam. Dogs were dosed for 28 days. Blood samples (2 mL) for robenacoxib assay were collected from a vein into tubes containing EDTA but only on two days (7 and 14, one sample each). A total of 81 dogs (20 dogs in each group but 21 dogs in group 0.5–1 mg/kg) were included in this study. The test item administered to these animals consisted of tablets containing 5 mg or 20 mg robenacoxib (Onsior® tablets, Novartis Animal Health, Basel, Switzerland). The tablets were administered as a twice daily intake by the oral route at the same time as feeding (either mixed with the food or administered into the mouth at the time of feeding). Mean (range) dosages were 0.35 (0.25–0.47) mg/kg in Group 5, 0.68 (0.50–0.89) mg/kg in Group 6 and 1.49 (1.00–1.88) mg/kg in Group 7. The overall mean ± SD age of the dogs included in this study was 9.8 ± 3.2 years with a range of 0.8–16.2 years. The mean ± SD bodyweight of the dogs was 29.3 ± 13.1 kg with a range of 5.2–78.5 kg.

2.3. Animal phase of study COXFRA106

Details on the experimental phase of this study were published previously (Silber et al., 2010). This study was a multi-center field study (20 veterinary clinics in different geographical locations in France) with the objective to evaluate the pharmacokinetics of robenacoxib in blood and stifle joint synovial fluid in dogs suffering from OA after multiple (for 7 days) or single oral administration. In addition to the synovial fluid sample, blood samples (2 mL) were taken at 5 of the following time points: 0.25, 0.5, 1, 2, 3, 5, 7, 10, 16 or 24 h after treatment administration. The administration of robenacoxib preceding the start of the sampling campaign (last administration for the first group of dogs and single administration for the second group) was performed by the investigator by gavage and under fasted condition in the veterinary clinic. The exact times of tablet administration and sampling were recorded for all animals. Totals of 47 animals were included in the first group of dogs (7 days of treatment) and 56 in the second (single administration), but only 41 and 54, respectively, were included in the population analysis due to missing pharmacokinetic data or treatment incompliance. The test item consisted of flavored tablets containing 20 mg or 40 mg robenacoxib (Onsior® tablets, Novartis Animal Health, Basel, Switzerland). Mean (range) dosages were 1.47 (1.03–1.99) mg/kg in the first group of dogs and 1.42 (1.03–2.00) mg/kg in the second group. The mean ± SD age of the dogs was 7.7 ± 3.3 years with a range of 0.7–14.3 years. The mean ± SD bodyweight of the dogs was 33.9 ± 12.5 kg with a range of 10.0–72.7 kg.

2.4. Analytical Phase

Blood samples were collected into tubes containing EDTA and stored at approximately −20 °C prior to analysis. Determination of robenacoxib blood concentrations involved an initial analysis by high pressure liquid chromatography – ultra violet, covering the range of 500–20,000 ng/mL and, if required, a subsequent analysis by liquid chromatography – mass spectrometry, covering the range of 2–100 ng/mL (Jung et al., 2009). The analytical method was validated using quality control spiked matrix specimens run with each sequence of unknown samples, and independent of calibration standards. Samples containing robenacoxib in blood were shown to be stable at approximately −20 °C for 5 months. The Lower Limit of Quantification (LLOQ) of the analytical method in blood was 2 ng/mL.

2.5. Data analysis and model evaluation

Non-linear mixed effects modeling of the data was performed using NONMEM version VI 2.0 (Icon Development Solutions, Ellicott City, Maryland, USA). The first order conditional estimation method (FOCE) was used for all analyses in combination with the Laplacian method for categorical data when needed. Model selection was based on statistical significance between competing models using the objective function value (OFV) obtained from...
different between two nested models is approximately \( \chi^2 \)-distributed and a difference in OFV of 3.84 corresponds to \( p < 0.05 \) for one degree of freedom (and 6.63 corresponding to \( p < 0.01 \)). Graphical assessment was performed using the R-based software Xpose version 4.1 (Jonsson and Karlsson, 1999) in R version 2.8.1 (The R Foundation for Statistical Computing, Vienna, Austria).

2.6. Pharmacokinetic model development

Based on previous results (Silber et al., 2010) two- and three-compartment models were evaluated for the description of the distribution and elimination of robenacoxib. Different models were evaluated for the description of the absorption phase including 1st-order absorption, a combination of 1st- and 0-order absorption as well as a mixture model. Inter-individual variabilities (IIV) were included as far as possible under the restriction that an IIV on apparent clearance was present in order to later on derive the exposure variability based on the post-hoc apparent clearance estimates. IIVs were expressed using exponential models to obtain log-normal distributions. Inter-occasion variability could not be tested due to the study layout – only one data point per occasion in studies COXFRA101/102 and different dogs per arm in study COXFRA106. An additive model was used to describe the residual variability on log-transformed data. This model corresponds approximately to a proportional model on normal data.

2.7. Inclusion of data below the limit of quantification

Different approaches for inclusion of data below the LLOQ (BLQ) were evaluated including setting the values BLQ to half of the Lower Limit of Quantification (LLOQ/2), also known as the M2 method (Beal, 2001). When the M2 method was used, the first observation in each dog which was BLQ during the elimination phase was set to LLOQ/2 and the subsequent observations were discarded. If a series of observations BLQ were observed during the absorption phase, the last of these values was kept at LLOQ/2. The M3 method, which estimates the likelihood that the BLQ observations are below the LLOQ, was also evaluated as this method has been shown to give estimates the likelihood that the BLQ observations are below the LLOQ/2 and the subsequent observations were discarded. If a series of observations BLQ were observed during the absorption phase, the last of these values was kept at LLOQ/2. The M3 method, which estimates the likelihood that the BLQ observations are below the LLOQ, was also evaluated as this method has been shown to give less biased parameter estimates compared to the M2 method (Beal, 2001). For this method, all observations BLQ were kept in the data set.

2.8. Covariate search for apparent volume and clearance

After structural model development, parameter differences between studies were investigated before the covariate search on apparent clearance and central volume was performed. Age, bodyweight, sex, and breed were initially considered and thereafter kidney and liver variables.

The statistical results presented in the paper were derived using the inclusion of covariates in the population model using forward inclusion (\( p < 0.05 \)) and backward exclusion (\( p > 0.01 \)) to correct for multiple comparisons (c.f. Wahby et al., 2002).

2.9. Statistical results and graphical display of the exposure-parameter relations

Most figures in this paper were created based on the empirical Bayes estimates (EBEs) of apparent clearance from NONMEM. Based on the assumption of dose- and time-linearity of robenacoxib, the exposure (or area under the curve AUC) was derived by

\[ \text{AUC} = \text{Dose/Clearance} \]

for a dosage in the range of 1–2 mg/kg (except if stated otherwise).

The statistical results presented in the paper do not rely on the EBEs thereby avoiding problems due to possible shrinkage.

3. Results

3.1. Study demographics

The pharmacokinetic data of three studies were pooled (see details in Table 1). There were no missing values for the demographic variables age, bodyweight, sex, and breed (a summary is given in Table 2) with only minor differences between studies. As can be seen in Fig. 1 the data were sparse in the COXFRA101 and 102 studies with only one sample per occasion per individual at two or three occasions. The COXFRA106 study comprised five samples during 24 h post dosing on one occasion.

3.2. Pharmacokinetics

Similar to Silber et al. (2010), a two-compartment model was found to describe the disposition of robenacoxib in blood well (see Fig. 1 and Supplemental Fig. 2). In order to capture the absorption kinetics and their variability, different models were tested. A transit compartment model (see Supplemental Fig. 1 for details) showed the largest drop in OFV, but lead to a numerically very instable model, which made it impossible to extend it and thus to do a covariate search. Splitting the dog population into two groups, one with faster and one with slower absorption, lead to a numerically identifiable and robust model, using a sequential 0- and 1st-order absorption (see parameters in Table 3 and Fig. 1 and Equations in Supplemental Table 1). Due to the sparsity of the data in two of the three studies, only two inter-individual variabilities could be added; one for absorption duration and one for apparent clearance. The latter was preferred over apparent volume as the goal of this investigation was to estimate variability in exposure (AUC) based on the EBEs of apparent clearance (see Section 2). The M3 method to handle data BLQ did not converge successfully. Bergstrand and Karlsson (2009) showed similar levels of bias when removing all BLQ values or when setting some of them to LLOQ/2 (M2). Therefore the final result was obtained ignoring all BLQ values.

3.3. Assessment of dose adjustment by age, bodyweight and sex

In order to investigate the common assumption that apparent clearance scales with bodyweight, the initial model did not include dose adjustment by bodyweight (BW). A model based assessment was performed with stepwise inclusion and exclusion of age, bodyweight and sex to explain variability in apparent clearance and central volume (see Section 2 for details) and assess the necessity for dose adjustment according to these characteristics. Only two additions were found to explain part of the variability and to significantly improve the model (\( p < 0.01 \)): the influence of bodyweight on both apparent clearance and volume.

\[ \text{Cl}/\text{F} = \text{C}_{10}/\text{F} \ast (\text{BW}/35)^{0.88} \]

\[ \text{Vc}/\text{F} = \text{V}_{0}/\text{F} \ast (\text{BW}/35)^{1.35} \]

Note that the exponent for the effect of bodyweight on apparent clearance was 0.88 with the 95% confidence interval including 1 and the exponent for the effect on apparent central volume was 1.35, being estimated significantly different from 1 (\( p < 0.01 \)).

The covariate search initially included sex on apparent clearance as well (\( p < 0.05 \)), but the estimated 15% lower exposure for females was not statistically significant when assessing the back-
ward step \( p > 0.01 \) and was also considered not to be clinically significant.

Final model structure and equations are provided in Fig. 2 and Supplemental Table 1, respectively.

Fig. 3 underlines the finding that no correlation between age and exposure was apparent and no significant difference in exposure was observed between sexes.

Using the final model (with only bodyweight as covariance) we also tested if there was a significant study difference for apparent clearance but no statistically significant effect \( p > 0.05 \) was found.

The final parameter values are given in Table 3. Note that the relative standard errors (RSE) provide a measure of the accuracy of the 14 parameter estimates (which are consistently below or up to 40%), whereas the inter-individual variabilities provide a measure of the differences between individuals with an estimate of 23% for apparent clearance.

3.4. Effect of bodyweight on exposure (robenacoxib AUC)

The current recommended dosage of 1–2 mg/kg led to four different dose groups considering the study population (5.2–78 kg): 10, 20, 40, and 80 mg per dog. Fig. 4 shows the exposure (AUC) derived from the EBs of apparent clearance versus the actual dose and the normalized dosage per bodyweight. The exposure range

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**Table 1**

Study design and pharmacokinetic sampling times.

<table>
<thead>
<tr>
<th>Study</th>
<th>Population</th>
<th>Treatment</th>
<th>Dosages (mg/kg)</th>
<th>Sampling times (days)</th>
</tr>
</thead>
<tbody>
<tr>
<td>COXFRA101</td>
<td>33 m/18f = 51</td>
<td>28 days, once daily</td>
<td>0.5–1; 1–2; or 2–4</td>
<td>2, 7, and 14 (1 sample each)</td>
</tr>
<tr>
<td>COXFRA102</td>
<td>32 m/28f = 60</td>
<td>28 days, twice daily</td>
<td>0.25–0.5; 0.5–1; or 1–2</td>
<td>7 and 14 (1 sample each)</td>
</tr>
<tr>
<td>COXFRA106A</td>
<td>18 m/25f = 43</td>
<td>7 days, once daily</td>
<td>1–2</td>
<td>7 (5 samples in 24 h)</td>
</tr>
<tr>
<td>COXFRA106B</td>
<td>22 m/32f = 54</td>
<td>1 single dose</td>
<td>1–2</td>
<td>0 (5 samples in 24 h)</td>
</tr>
</tbody>
</table>

**Table 2**

Age, bodyweight, sex, and breed.

<table>
<thead>
<tr>
<th>性</th>
<th>年龄 (年)</th>
<th>身体重量 (kg)</th>
<th>最小值</th>
<th>平均值</th>
<th>最大值</th>
</tr>
</thead>
<tbody>
<tr>
<td>性别</td>
<td>年龄 (年)</td>
<td>身体重量 (kg)</td>
<td>最小值</td>
<td>平均值</td>
<td>最大值</td>
</tr>
<tr>
<td>育种</td>
<td>最小值</td>
<td>平均值</td>
<td>最大值</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Fig. 1.** Overview of experimental data and model fit. The columns provide experimental data, individual and population prediction of the model, respectively. The data are split into four groups (rows), i.e., studies COXFRA101, COXFRA102, and COXFRA106 single dose and multiple doses. The model reflects the variable absorption patterns and the general variability of the data.

was similar for the different dose groups. The plot versus the normalized (mg/kg) dosage shows that the 2-fold difference in dosage between dogs obtaining 1 mg/kg and others with a dosage of 2 mg/kg dominates the variability observed in exposure compared to the remaining unexplained variability in apparent clearance (estimated to be 23% – see Table 3). The necessity to adjust the dose (mg) by bodyweight (mg/kg) is very apparent when plotting exposure versus bodyweight for a fixed dose of 40 mg for all dogs (see Fig. 5 right panel) resulting in an approximately 11-fold difference in exposure. On the other hand administration of exactly 1.4 mg/kg (median dosage in the given studies) shows less differences in exposure for the various dogs, but not substantially less than with the dosage adjustment to 1–2 mg/kg (Fig. 5).

3.5. Assessment of relation between kidney/liver variables and exposure (robenacoxib AUC)

Analogous to the investigation of age, bodyweight and sex on explaining the variability in apparent clearance (and thus AUC), six different variables used clinically to assess the health of the kidney or liver were assessed to explain the remaining variability: plasma concentrations of total protein, urea and creatinine, and activities of aspartate-aminotransferase, alanine-aminotransferase, and gamma-glutamyl transferase. None significantly improved the OFV of the model ($p < 0.01$) and the graphical display of the relation between exposure and the respective markers did not reveal any correlation (see Fig. 6).

<table>
<thead>
<tr>
<th>Table 3</th>
<th>Pharmacokinetic parameters.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Unit</td>
<td>Value</td>
</tr>
<tr>
<td>Absorption duration ($D_{0(1)}$)**</td>
<td>h</td>
</tr>
<tr>
<td>Absorption rate ($ka_{1(1)}$)**</td>
<td>1/h</td>
</tr>
<tr>
<td>Absorption duration ($D_{0(2)}$)**</td>
<td>h</td>
</tr>
<tr>
<td>Absorption rate ($ka_{2(2)}$)**</td>
<td>1/h</td>
</tr>
<tr>
<td>Population proportion ($A^1$)**</td>
<td>45%</td>
</tr>
<tr>
<td>Apparent central volume ($V_{c0/F}$)**</td>
<td>L</td>
</tr>
<tr>
<td>Apparent intercomp. clearance ($Q/F$)**</td>
<td>L/h</td>
</tr>
<tr>
<td>Apparent peripheral volume ($V_{p/F}$)**</td>
<td>L</td>
</tr>
<tr>
<td>Rel. bioavailability with food</td>
<td>73.8%** Fixed</td>
</tr>
<tr>
<td>Proportional residual error ($e^2$)**</td>
<td>96%</td>
</tr>
<tr>
<td>IVV absorption duration ($v_{D0}$)**</td>
<td>98%</td>
</tr>
<tr>
<td>IVV apparent clearance ($v_{Cl}$)**</td>
<td>23%</td>
</tr>
<tr>
<td>Exponent BW on $Cl/F$</td>
<td>0.88</td>
</tr>
<tr>
<td>Exponent BW on $Vc/F$</td>
<td>1.35</td>
</tr>
</tbody>
</table>

** RSE... relative standard error (i.e., accuracy of parameter estimates).
** IIV... inter-individual variability (% CV).
** Mixture model to cover high variability in absorption.
** Typical values for a dog of 35 kg bodyweight.
** Fixed parameter value based on Jung et al. (2009).

### Fig. 2. Sketch of the final model structure used to represent the dynamics of robenacoxib concentrations measured in plasma (as part of the central compartment). Model equations can be found in the Supplementary Table 1.

### Fig. 3. Exposure versus age and sex. Including age and sex could not substantially explain the variability in apparent clearance (and thus exposure) and did not significantly improve the model ($p > 0.01$). Studies are denoted by O COXFRA101, △ COXFRA102, and +COXFRA106.

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3.6. Assessment of dose adjustment for different breeds or breed groups

The large number of different breeds (62) and therefore the low number of animals per breed \((\frac{1}{3} / \text{breed})\) excluded a thorough analysis for all breeds separately. Based on published literature (Cox et al., 2011) we investigated if German Shepherds or Labradors would show significantly different apparent clearance/exposure than the remaining breeds. Both breeds showed non-significant differences to the main population and point estimates of the differences of 15% or less. Fig. 7 (right panel) shows the distribution of exposure (AUC, based on EBEs of apparent clearance) for breeds with at least two animals compared to the 5th and 95th percentile of the population with no breeds being apparent outliers.

Grouping the breeds according to the Fédération Cynologique Internationale (FCI) yielded 9 groups with approximately 19 animals per group. Model based analysis of the differences between these breed groups did not yield any significant differences \((p > 0.01)\). This is consistent with the left panel of Fig. 7 showing the generally wide span of exposure within breed groups and also that all the inter-quartile range intervals lie within the 5th and 95th percentile of the overall population.

4. Discussion

Although there is limited information in the literature on pharmacogenetic differences between dog breeds, it has been demonstrated that the NSAIDs celecoxib and mavacoxib show high within and/or between-breed variability in the dog (Cox et al., 2011; Fleischer et al., 2008; Paulson et al., 1999). In healthy Beagle dogs, a marked polymorphism in the metabolism of celecoxib was detected, with at least two populations of dogs which were termed “poor metabolizers” and “extensive metabolizers” (Paulson et al., 1999). The polymorphism was attributed to differences in the rate of liver metabolism by CYPs. In dogs with OA, mavacoxib had relatively high between-subject variability, and associations between apparent clearance \((\text{Cl/F})\) and age, breed and notably bodyweight were observed (Cox et al., 2011). Furthermore for mavacoxib in dogs with OA, 5% of dogs had very long terminal half-lives and higher apparent clearance was observed in two breeds, German Shepherds and Labrador Retrievers. Age of dog was also an important covariate in the population pharmacokinetic variability of mavacoxib in dogs with OA (Cox et al., 2011). Because of the potential impact any variability could have on efficacy and safety, and because most pre-clinical data for robenacoxib were obtained in young Beagle dogs (Jung et al., 2009; Schmid et al., 2010; King et al., 2011), it was judged important to document the pharmacokinetic variability of robenacoxib in the target population (frequently large-breed elderly dogs) and to document whether covariates such as sex, age and breed play a significant role in this variability.

Using normal statistical analysis it would not have been possible to obtain an estimate of the individual exposures based on the available data in order to assess the influence of the various covar-
iates mentioned before. The population pharmacokinetic model selected enabled the effective analysis of the pooled data from all three studies. Although the variable absorption was difficult to model, it helped to distinguish the variability in absorption time from variability in apparent clearance which would have been impossible with only trough values.

The main limitation of the modeling was the high residual error estimated by the model indicating that there could be a problem in assessing and correctly allocating the IIV to different parameters. However, other model structures and software tools were used as well to fit the same data (not shown) and all results provided similar conclusions.

Fig. 6. Exposure versus total protein (PROT), urea, creatinine (CREA), aspartate-aminotransferase (AST), alanine-aminotransferase (ALT) and gamma glutamyl-transferase (GGT). The panels show the relation between exposure (AUC) and the respective kidney or liver variable. None of them was strongly correlated or significantly improved the model ($p > 0.01$) (study symbols as Fig. 2).

Fig. 7. Variability in exposure for grouped breeds and for individual breeds ($n > 2$). The dashed line denotes the median AUC (1.28 h mg/mL) and the gray area the range between 5th and 95th percentile. No significant differences ($p > 0.01$) could be found between breed groups. Breeds are sorted according to mean bodyweight of the breed. Selected breeds were also tested (see text) – showing no significant changes ($p > 0.01$).
The thorough investigation of the influence of bodyweight (range 5.2–78 kg) on exposure clearly showed that a dose adjustment by bodyweight and the classical assumption that apparent clearance is proportional to bodyweight seems to hold true for robenacoxib. The number of very light animals was probably not sufficient to definitely rule out a possible under-estimation of the true apparent clearance for small dogs (Martinez et al., 2009). However the fact that animals receiving 20, 40 or 80 mg robenacoxib had very similar exposure ranges confirms that, for this compound, apparent clearance is roughly proportional to bodyweight for a wide range of bodyweights and that a dosage of 1–2 mg/kg provides similar exposure levels across the whole target population.

To our knowledge, biologically relevant gender differences in pharmacokinetics are very rare and a correlation between gender and exposure was therefore not expected. The covariate analysis confirmed that gender was not a significant covariate to explain the variability of apparent clearance. No sex difference in pharmacokinetic variables was previously reported in healthy young Beagle dogs (Jung et al., 2009).

No correlation between age and exposure was apparent and no significant differences between breeds could be demonstrated. Although this result is not a definitive proof that breed differences in the pharmacokinetics of robenacoxib do not exist, the analysis clearly showed that for breeds well represented in this study and for which a breed effect could be demonstrated with the NSAID mavacoxib i.e., German Shepherds and Labradors (Cox et al., 2011), no significant pharmacokinetic differences to the main population could be identified. This finding suggests that despite the fact that robenacoxib is heavily metabolized in the liver there does not seem to be a high level of between-breed polymorphism in its metabolism.

No evidence for effects of kidney or liver variables on robenacoxib exposure was observed. However, the measured values were mostly within the normal range, the exception being moderately elevated ALT values at baseline in some groups, indicating that only relatively few dogs had severe kidney or liver problems – reflecting the inclusion criteria set for the three studies. Therefore our available data do not preclude the possibility that severe kidney or liver dysfunction could have an impact on robenacoxib exposure in dogs. Mild to moderate liver or renal impairment had no significant effect on the pharmacokinetics of lumiracoxib, an analog of robenacoxib, in humans (Rordorf et al., 2005).

Since no evidence for age, breed, sex, kidney or liver effects on the exposure of robenacoxib was evidenced, it is concluded that the most probable reason for the previously observed higher exposure of robenacoxib in a population of mixed-breed OA dogs compared to healthy young Beagle dogs is cytochrome P450 inhibition due to chronic inflammation (Silber et al., 2010).

Overall, the population pharmacokinetic analysis performed showed that the 1–2 mg/kg dosage provides consistent robenacoxib exposure in a wide range of canine patients. No adjustment of dose for special dog populations seems necessary.

Acknowledgement

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.rvsc.2013.04.021.

References


Conflict of interest

The authors of the manuscript are Novartis employees.