REVIEW

Robenacoxib in the treatment of pain in cats and dogs: safety, efficacy, and place in therapy

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Abstract: Robenacoxib is a novel nonsteroidal anti-inflammatory drug (NSAID) of coxib class developed for the control of inflammation and pain in dogs and cats. It shows high selectivity for the cyclooxygenase-2 (COX-2) enzyme in rats, cats, and dogs. Robenacoxib is available in both injectable and tablet formulations. This review initially focuses on the preclinical pharmacology of robenacoxib in rats that includes its high affinity for COX-2 enzyme and weaker and rapidly reversible binding for COX-1 enzyme in in vitro and ex vivo models of inflammation and its pharmacokinetics in the blood and inflammatory exudate, selective tissue distribution, and safety. These basic pharmacological profiles highlight the suitability of robenacoxib for use in target species, such as cats and dogs. Since the level of expression and activity of COX enzymes is species specific, COX-2-selective inhibition and the resultant effects of coxibs must be studied in target species. The pharmacological and toxicological profiles of robenacoxib in cats and dogs have been discussed prior to reviewing its clinical efficacy and safety. Large, multicenter field trials conducted in cats and dogs demonstrated the noninferior efficacy and safety of robenacoxib compared with noncoxib NSAIDs used in dogs and cats. These trials investigated the efficacy of robenacoxib against various acute and chronic painful conditions. Robenacoxib produced superior efficacy to placebo and COX-2 preferential inhibitors in postsurgical cats. The tissue-selective anti-inflammatory activity of robenacoxib has been demonstrated in dogs with osteoarthritis. Robenacoxib has also been shown to be safe in healthy dogs and cats receiving antihypertensive drugs and loop diuretics that could cause renal injury. The developmental objective of coxibs, comparable efficacy but superior safety to less selective/nonselective NSAIDs, is well established with robenacoxib in preclinical studies. More studies need to be conducted to fully explore the benefits of robenacoxib in clinical subjects.

Keywords: robenacoxib, preclinical pharmacology, clinical studies, analgesia, tolerability

Introduction

Historically nonsteroidal anti-inflammatory drugs (NSAIDs) have been demonstrated to inhibit the synthesis of prostaglandins for the control of inflammation, pain, and hyperthermia in mammals. They inhibit the cyclooxygenase (COX) enzyme, which is responsible for the production of prostaglandins causing inflammation and pain.[1,2] The COX enzyme exists in the following two isoforms: COX-1 and COX-2.[3] A simplified description of the functional activity of these isozymes is that COX-1 is primarily involved in the production of physiological prostaglandins stimulating and maintaining the normal body functions such as regulation of gastric acid and mucus production, platelet production, and renal homeostasis and that COX-2 expression is induced and is responsible for the production of prostaglandins that are important for signaling

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inflammation, pain, and pyrexia.[4] Nonspecific inhibition of COX enzymes by traditional NSAIDs such as ibuprofen, naproxen, and aspirin leads to various adverse effects (AEs) in therapeutic doses: classic AEs include gastric ulceration and bleeding and renal damage. Recent research has been directed at the selectivity of NSAIDs inhibiting COX-2 over COX-1 (coxibs) in order to minimize the AEs at doses that produce the beneficial (therapeutic) effects. However, there is substantial evidence that some of the specific functions of the two COX enzymes may crossover, and therefore, COX-2 activity is also important for some physiological functions.[5] This again depends on the fact that the level of expression and activity of COX enzymes is species specific, and therefore, COX-selective inhibition and the resultant effects of specific NSAIDs must be separately studied in different species of animals.[6,7]

Robenacoxib is a novel NSAID developed for the control of inflammation, pain, and hyperthermia in dogs and cats. It is described as "coxib" type of NSAID as it demonstrates highly selective and targeted inhibition of the COX-2 enzyme in animals.[8,9] Preclinical studies in rats show its high affinity for COX-2 enzyme and a weaker and rapidly reversible binding for COX-1 enzyme in in vitro systems and ex vivo models of inflammation. The basic pharmacological profiles of robenacoxib in rats highlight its potential for use in the target species, such as dogs and cats. This article initially reviews the preclinical pharmacology of robenacoxib in rats. Then, its basic pharmacology and toxicology in cats and dogs will be discussed prior to reviewing its safety and efficacy in clinical studies.

Preclinical studies in rats

King et al[10] conducted an extensive study on preclinical pharmacology of robenacoxib in rats. Robenacoxib binds with high affinity and dissociates slowly from COX-2 compared with a weaker binding and a rapid reversible inhibition of COX-1 in rats. The potency and highly targeted selectivity for the COX-2 inhibition of robenacoxib have been demonstrated in both in vitro assays, using purified COX enzymes, and clinically relevant ex vivo systems such as the whole blood and inflammatory exudate assays in rats. The COX-1:COX-2 IC_{50} ratio was 27:1 in enzyme preparations and >967:1 in cellular assays.

Robenacoxib produced the classic, beneficial NSAID effects in vivo, by reducing the carrageenan-induced rat paw edema, lipopolysaccharide (LPS)-induced pyrexia, and nociception induced by Randall–Selitto assay. It showed comparable efficacy with an archetypal NSAID, diclofenac, and other COX-2-selective inhibitors, such as etoricoxib

and lumiracoxib in these in vivo pharmacodynamic models of inflammation, pain, and pyrexia. The researchers did not find a significant difference between the effective dose $(ED)_{50}$ values of robenacoxib in this study and other coxibs from the published data, for the inhibition of LPS-induced pyrexia and carrageenan-induced paw edema. In Randall–Selitto assay that measures altered pain sensitivity/hyperalgesia of inflamed rat paws, the maximal effective dose of robenacoxib was significantly higher (30 mg/kg, after oral [PO], 2 hours after administration) than that of diclofenac (3 mg/kg, PO, 2 hours after administration). Other coxibs such as etoricoxib also were required in high doses to reduce hyperalgesia in this model.[11]

Randall–Selitto assay measures mechanical withdrawal thresholds in inflamed paws of rats. The paw withdrawal reflexes to noxious stimuli mainly indicate spinal nociceptive processing eliciting immediate motor responses limiting the duration of stimulus.[46] Pain is different from this nociception in that it elicits more complex behaviors to noxious stimuli due to the involvement of the supraspinal and higher brain centers in processing the stimuli.[47]

Mechanisms that produce hyperalgesia of the inflamed rat paw are diverse: extensive research on pharmacological activity of diclofenac suggests that it possesses multiple mechanisms of action, such as inhibition of substance P and *N*-methyl-D-aspartate receptor-mediated hyperalgesia and alteration of interleukin-6 production, beyond the primary mechanism of COX inhibition.[12] In this study, the efficacy of diclofenac at a much lower dose than robenacoxib against paw hyperalgesia could be due to its multimodal action. In addition, the researchers of the study suggest that the higher dosage requirement of robenacoxib (similar to other coxibs) could be due to more COX-2 independent mechanisms involved in this model of nociception.

After oral administration in rats, robenacoxib was rapidly absorbed and maximum plasma concentrations (C_{max}) were obtained in 1 hour (T_{max}), suggesting that the drug can produce rapid onset of effect. Robenacoxib is highly bound to plasma proteins (99.9±0%) in rats. One important criterion for newer coxib class of NSAIDs is to maintain the comparable efficacy of traditional nonselective NSAIDs with higher safety profiles.[13] Robenacoxib (2 mg/kg, PO) preferentially distributes into inflammatory exudate in zymosan-induced tissue cage model of inflammation, compared with blood. The area under the drug concentration versus time curve (AUC) for the exudate is significantly higher than for blood. The elimination half-life $(t_{1/2}$ el$)$ and mean residence time (MRT) of robenacoxib in the exudate are longer than that

of blood. The longer residence time coupled with persistent higher concentrations at sites of inflammation compared with short $t_{1/2}$ el and lower AUC in blood indicate that robenacoxib might have insignificant effect on constitutive prostaglandins in well-perfused organs such as the kidney, heart, and liver and in vasculature.[10]

Comparative effects of robenacoxib and diclofenac on renal function of water-overloaded rat model have been studied over 6 hours of their oral administration. The dose of both drugs, 30 mg/kg, was greater than that required to completely inhibit PGE_2 in in vivo models of inflammation. Compared with the control (vehicle), robenacoxib had no significant effect on urine volume, urine PGE_2, and urine creatinine concentration, whereas diclofenac significantly reduced the urine volume and PGE_2 concentration. Although it was a slight rise, robenacoxib significantly increased the serum creatinine concentrations compared to the control. This slight increase in serum creatinine levels in the absence of inflammation and at doses much higher than that required to produce therapeutic effects may not be clinically important.

The gastrointestinal safety of robenacoxib has been studied at a dose (100 mg/kg, PO) 250 times greater than ED_{50} required for minimizing carrageenan-induced paw edema in rats. The incidence and diameter of gross gastric lesions were significantly less than that observed with diclofenac (100 mg/kg, PO). There was no significant difference between robenacoxib and control (vehicle) groups. Intestinal mucosal integrity has been evaluated by quantifying the ^{51}Cr-ethylenediaminetetraacetic acid (EDTA) excreted in urine after robenacoxib (30 mg/kg, PO) administration in rats for 4 days; ^{51}Cr-EDTA is a standard marker used to assess the altered intestinal permeability due to reduced prostaglandin synthesis by NSAIDs.[14] Increased urinary excretion of orally administered ^{51}Cr-EDTA after test NSAID administration indicates an increase in intestinal permeability due to prostaglandin inhibition. Rats administered with robenacoxib had significantly lower ^{51}Cr-EDTA concentrations in the urine compared to those administered with diclofenac. These findings suggest that robenacoxib possesses significantly superior and wider margin of gastrointestinal safety than diclofenac due to its high affinity for the COX-2 enzyme and sparing of COX-1 enzyme.

The effect of robenacoxib on platelet-derived thromboxane B_2 (T_xB_2), synthesis of which is primarily dependent on the functional COX-1, has been compared with diclofenac and control (vehicle) at 30 and 100 mg/kg oral doses in whole blood assays. Serum concentrations of T_xB_2 were significantly higher in the robenacoxib group than in the diclofenac group, at both doses. At 30 mg/kg dose, there was no

significant difference in serum T_xB_2 concentrations between robenacoxib (180±4.4 ng/mL) and control (310±32.6 ng/mL) groups. However, at 100 mg/kg dose, serum T_xB_2 concentrations of the robenacoxib group (60.2±7.6 ng/mL) were significantly lower than that of the control group (310±32.6 ng/mL; $P<0.01$). This could be due to possible but a trivial role of COX-2 in TxB_2 synthesis[15] and the potential for limited inhibition of COX-1 at higher (toxic) doses by robenacoxib.

Preclinical studies in cats

Cats, dogs, and horses are more susceptible to AEs of NSAIDs than humans and rats.[10] It is essential to evaluate the efficacy and safety of coxibs prior to their clinical use in these animals; also, simple extrapolation of safety benefits of coxibs from human and rodent pharmacological studies may not be optimally translated to other mammals.

In cats, the inhibitory actions of robenacoxib on COX enzymes have been studied in in vitro feline whole blood assays.[8] These assays have been considered as the most relevant in vitro models that reflect the actions of NSAIDs in vivo.[7,16] The mean IC_{50} values of robenacoxib for COX-1 and COX-2 were 28.9 and 0.058 μm, respectively. The classical index of COX selectivity, expressed as the ratio of the IC_{50} value for COX-1 and COX-2, was 502.3. These COX enzyme inhibition profiles suggest that robenacoxib is a highly selective COX-2 inhibitor NSAID in cats. In another study, Schmid et al[17] compared the potency of robenacoxib to selectively inhibit the COX-2 enzyme with that of two other NSAIDs licensed for use in cats, ketoprofen, and meloxicam and with the reference NSAID, diclofenac. Feline whole blood assays were used to characterize the COX enzyme inhibition potencies of these NSAIDs. Based on IC_{50} values, it has been reported that robenacoxib was highly selective for COX-2, both diclofenac and meloxicam were only slightly preferential for COX-2, and ketoprofen was selective for COX-1. For 95% of COX-2 inhibition levels, robenacoxib produced only 12.4% inhibition of COX-1 whereas meloxicam caused 72.7% inhibition of COX-1. The relative percentage inhibition of COX-2 and COX-1 for ketoprofen was 50 and 97.7, respectively. These relative inhibition potencies for COX-1 and COX-2 further demonstrate the efficacy of robenacoxib at selectively targeting the COX-2 while sparing COX-1.

The in vitro whole blood assay findings have been confirmed by ex vivo inhibition of feline plasma PGE_2 and serum T_xB_2 as surrogates of COX-2 and -1 inhibition, respectively. Both orally (1–2 mg/kg) and subcutaneously ([SC] 2 mg/kg) administered robenacoxib significantly reduced the plasma PGE_2 concentrations and produced a nonsignificant effect on

serum T_xB_2 concentrations compared to the negative (vehicle) control.[17] The time course of COX-2 inhibition (PGE_2) in the blood plasma was significantly shorter than that of meloxicam. This is further confirmed by in vivo pharmacokinetics of robenacoxib showing rapid plasma clearance and shorter residence time and $t_{1/2}$ el in the blood following both routes (SC and PO) of administration in healthy cats in the same study. These ex vivo and in vivo study findings suggest that robenacoxib is COX-2 selective but with a short duration in the central compartment (blood).

Giraudel et al[9] investigated the efficacy of robenacoxib against kaolin-induced paw inflammation in 10 cats. The researchers used a pharmacokinetic/pharmacodynamic (PK/PD) approach to demonstrate the magnitude and time course of effect of robenacoxib following administration of a single dose (2 mg/kg, SC). Since the main aim of this preclinical study was to find an effective dosage regime of robenacoxib in cats, clinically relevant endpoints of the inflammation model such as pain scores, locomotion, and lameness scores have been evaluated; changes in skin temperature and body temperature were also assessed as surrogates of inflammation and hyperalgesia. The onset of responses was very rapid, and the peak responses for all endpoints occurred between 2.6 and 3.5 hours after injecting the drug. The duration of drug responses ranged from 4.6 to 8.1 hours. Maximum robenacoxib plasma concentration was achieved in 0.88±0.13 hours (mean ± SD). The apparent total blood clearance was high (10.10±1.85 mL/kg/min) with a short mean $t_{1/2}$ el of 1.87 hours. The PK/PD simulations suggested twice daily dosing for maintaining efficacy following SC administration of a single dose. In an extensive field (clinical) trial in cats, robenacoxib (1–2.4 mg/kg, PO) produced analgesic and anti-inflammatory effects equivalent to ketoprofen following once daily dosing.[18] In retrospect, it appears that robenacoxib produces long-lasting analgesia despite its high body clearance and a short half-life in the blood.

Pelligand et al[19,20] conducted PK/PD modeling of robenacoxib in feline tissue cage model of inflammation to determine its disposition in the inflammatory exudate, in addition to the blood, and to determine its COX enzyme selectivity profiles using serum T_xB_2 and exudate PGE_2 as markers of COX-1 and -2 activities, respectively. After intravenous (IV), SC (2 mg/kg), and oral (6 mg/kg) administrations, the MRT in the blood was short (0.4, 1.9, and 3.3 hours, respectively) with a rapid clearance compared to the longer MRT (~24 hours regardless of the route of administration) in the tissue cage exudate. The exudate PK parameters showed that robenacoxib attains maximum and highest concentrations

following SC administration than the other two routes. The time to reach maximum exudate concentrations was longer after SC (7.1±1.8 hours) and oral (9.6±6.5 hours) administrations than after IV injection (4.4±2.6 hours), despite a nonsignificant difference in the MRT values (23.3, 23.5, and 25.9 hours) between the three routes of administration.

Robenacoxib is highly bound to plasma proteins (>98%), in both cat and dog plasma.[21] Similar to many other traditional nonselective NSAIDs, robenacoxib possesses a carboxylic acid moiety and differs from many other coxibs in lacking a sulfur-containing group in its chemical structure.[10] The acidic moiety serves as a major binding group (ionic binding) with plasma proteins. Since exudate is the protein-rich filtrate of plasma that leaks into sites of inflammation, NSAIDs with high degree of plasma protein binding can be easily carried to and accumulate in high concentrations at sites of inflammation.[7] The comparative PK of robenacoxib in the blood and tissue cage inflammatory exudate suggests that it can readily enter into and persists longer at sites of inflammation in elevated concentrations, despite a short $t_{1/2}$ el in the blood. This explains the long-lasting inhibition of exudate PGE_2 with a negligible and transient effect on serum T_xB_2 following a single dose (2 mg/kg) of robenacoxib in cats.[19] Using the same inflammation model in cats, Pelligand et al[20] reported that ketoprofen, another acidic NSAID with COX-1 selectivity, significantly suppresses serum T_xB_2 levels for 24 hours, although it produces a persistent reduction in exudate PGE2 concentrations similar to robenacoxib.

Clinical studies in cats

The clinical efficacy and tolerability of oral robenacoxib, as a tablet formulation, in cats have been studied in two large multicenter field trials in Europe (n=155) and Japan (n=68).[18,22] Ketoprofen (1 mg/kg), a selective COX-1 inhibitor approved for short-term use in cats, has been used as an active control in these randomized, blinded, and similar clinical trials with noninferiority design. The dosages of robenacoxib, 1–2.4 mg/kg, once or twice daily, have been chosen based on the preclinical studies that evaluated the efficacy of robenacoxib against kaolin-induced soft tissue inflammation in cats.[9] The dose has been predicted to produce 80% of inhibition of COX-2 in a whole blood assay. As anecdotal PK data in cats suggest reduced bioavailability of oral robenacoxib if given with entire daily ration study, cats were fed only a third of the daily ration when receiving robenacoxib tablets. Ketoprofen was given with entire ration; both treatments were given for 5 or 6 days to cats with signs of acute pain and inflammation due to either muscular or skeletal disorders. Treatment

efficacies have been evaluated by veterinary investigators on days 0, 2, and 4 and by the cat owners (unblinded) on each of the treatment days. A numerical rating scale (NRS) that scores signs of pain at palpation of the inflamed area, intensity of inflammation based on classical signs, and level of mobility on a 0–3 descriptor construct has been used to compare the primary treatment efficacies by clinical investigators. Cat owners assessed the response variables such as changes in the level of activity, behavior, interaction with owner, and other persons/animals and scored using the 0–3 score descriptors on the NRS. In one of the two studies,[18] there were no significant differences between the treatment groups for the efficacy variables assessed by both the clinical investigators and the cat owners. Also, there was no significant difference in the efficacy between once daily and twice daily robenacoxib groups. The study by Sano et al[22] did not find significant difference between the treatment groups for the clinical investigators' assessment of efficacy, but robenacoxib has been shown to be superior to ketoprofen for the owners' assessment of cats' activity and social interaction.

Emesis and diarrhea were the common AEs reported in all of the three treatment groups, and there was no significant difference between the treatments for the reported AE. Pretreatment illness has been deemed to be the cause of most of these AEs. No significant differences were found between baseline (day 0) and post-treatment (day 4 or 5) values for hematology variables such as red blood cell (RBC), white blood cell, and platelet counts, hematocrit, and hemoglobin concentration. Small nonsignificant increases in plasma urea and creatinine, potassium, and sodium concentrations have been detected in all three treatment groups. Overall, these studies indicate that once daily oral robenacoxib (1–2.4 mg/kg) produces noninferior efficacy and tolerability compared with the active control ketoprofen (1 mg/kg). The developmental objective of COX-2-specific inhibitors such as superior safety than less/nonselective NSAIDs has not been fully explored in these two short-term studies due to many confounding factors including inconsistencies among the sample population and evaluation methods.

Robenacoxib (1.03–2.4 mg/kg, PO) produced superior efficacy to placebo for the control of postoperative pain and inflammation in cats (n=167) undergoing forelimb onychectomy with ovariohysterectomy or castration.[23] Robenacoxib was administered 30 minutes prior to surgery and once daily for 2 days after surgery in this study. No significant changes in hepatic, hematological, and renal biomarkers indicating classic NSAID toxicity have been detected with this dosage regime related to surgery. The postoperative analgesic

efficacy of preoperative robenacoxib (2 mg/kg, SC) and buprenorphine (0.02 mg/kg, SC) alone or in combination has been evaluated in cats for 24 hours after ovariohysterectomy.[24] Cats were premedicated with medetomidine and ketamine and anesthetized with propofol and isoflurane. Robenacoxib alone produced better efficacy than buprenorphine, and its combination with buprenorphine did not provide additional analgesia. Buprenorphine has been shown to be less effective when administered by SC route than by intramuscular or IV injections in cats.[25,26] Although robenacoxib could produce noticeable analgesia in its own right, as demonstrated in other studies, comparison of its analgesic efficacy with SC buprenorphine obscures the result of this study.

Preoperative robenacoxib (2 mg/kg, SC) showed superior efficacy to meloxicam (0.3 mg/kg, SC) in reducing pain scores for 22 hours of cats after soft tissue and orthopedic surgeries in a multicenter, randomized clinical trial.[27] Pain and inflammation scores at the injection site were significantly higher in the meloxicam group than in the robenacoxib group. No adverse clinical signs have been reported in both groups. Follow-up treatment with oral robenacoxib tablets (1–2.4 mg/kg) for 9 days, in addition to a single preoperative dose, has not been found to be beneficial in further improving the efficacy scores compared to placebo group.[28] However, no adverse clinical signs have been detected after 9 days of its oral use in cats.

The clinical safety of oral robenacoxib has been evaluated in cats with osteoarthritis (OA) in a multicenter, randomized, and blinded clinical trial.[29] Robenacoxib (n=95) at a dosage of 1.0–2.4 mg/kg, PO, once daily for 28 days has been compared with placebo (n=99) for changes in body weight, clinical chemistry, hematology, and urinalyses at the completion of the study. A subset of the study cats (n=40) had chronic kidney disease (CKD) concurrent with OA. There were no significant differences in the outcome variables between the two groups of OA cats. Robenacoxib did not cause clinically detected evidence of damage to the gastrointestinal tract, liver, or kidney (reflected through the variables described above) of study cats, including the subgroup of cats with pre-existing CKD, compared to baseline values and the placebo-treated cats. The most frequent AE was vomiting, which occurred with similar frequency across treatment groups. Similar findings have been reported from a previous preclinical study on the safety of robenacoxib, in a tablet formulation, in healthy young cats.[30] Robenacoxib has been administered once daily for two study periods, such as 28 and 42 days. The dosages tested in both studies were higher than the current recommended dosage, 1–2.4 mg/kg, PO, q24. In addition to clinical chemistry, hematology, and

urinalyses in life, the researchers investigated the gross and histopathological changes of different vital organs and structures after euthanizing the cats at the end of the study periods. No significant toxicity of robenacoxib has been detected in any of the study outcome measures.

The renal safety of robenacoxib has been assessed in healthy cats administered angiotensin-converting enzyme inhibitors (ACEIs) and loop diuretics (LDs).[31] The combination of NSAIDs with ACEI and LD (triple whammy) could cause acute kidney injury in humans with systemic hypertension, congestive heart failure, and/or CKD.[32] Once daily administration of robenacoxib (1–2.4 mg/kg, PO) and benazepril (ACEI – 0.5–1.0 mg/kg, PO), either singly or in combination, for 7 days did not reduce the glomerular filtration rate (GFR) estimated from the plasma clearance of iohexol. In cats treated with an LD, furosemide (0.5 mg/kg, SC, twice daily) GFR was increased by benazepril but decreased by robenacoxib compared to the control cats. This treatment effect on GFR was influenced by the sex of the cat. Robenacoxib and its combination with benazepril significantly inhibited the increase in plasma aldosterone caused by furosemide. These studies have been conducted in healthy cats: it will be interesting to investigate the renal effects of robenacoxib in combination with ACEI in cats with CKD and OA.

Preclinical studies in dogs

In dogs, the inhibitory actions of robenacoxib on COX enzymes have been studied in in vitro whole blood assays.[33] The COX inhibitory potency of robenacoxib has been compared with that of other reference NSAIDs in dogs. Based on IC_{50} values, the relative potency order of COX-2 inhibition was robenacoxib > deracoxib > nimesulide > S$^+$ carprofen > meloxicam > etodolac > R-carprofen > ketoprofen. The relative potency for COX-1 inhibition was ketoprofen > meloxicam > nimesulide > etodolac > deracoxib > robenacoxib > S$^+$ carprofen > R-carprofen. The in vivo pharmacokinetics of oral robenacoxib at different doses (0.5, 1.0, 2.0, 4.0, and 8.0 mg/kg, tablet form) indicate that it is rapidly absorbed and eliminated with a short mean terminal blood half-life (0.6–0.91 hours). The results of the in vitro blood assays and in vivo PK and ex vivo PD studies in beagle dogs corroborate with the findings of preclinical studies in cats that robenacoxib, in clinically recommended dosages (1–2 mg/kg), possesses high potency for COX-2 but low potency for COX-1 inhibition and a short duration of action to inhibit PGE_2 in the central compartment.

Jung et al[21] developed an analytical method for the determination of robenacoxib in dog plasma in order to determine its blood concentration-time profiles and PK profiles after IV, SC and oral administration. Robenacoxib showed good bioavailability after oral (in fasted dogs) and SC administration and rapidly attained peak plasma concentrations in 0.25–0.5 hours after dosing. Food reduced the bioavailability of oral robenacoxib tablets. Robenacoxib is highly bound to plasma proteins (>98%) similar to that of cats and rats.

In a model of acute stifle synovitis induced by intra-articular injection of sodium urate crystals, dose–response and blood concentration–response of robenacoxib have been compared with placebo (negative control) and meloxicam (0.2 mg/kg; positive control) in dogs.[34,35] Both oral and SC robenacoxib (0.25, 0.5, 1.0, 2.0, and 4.0 mg/kg) produced dose-dependent improvement in force plate measures of gait and subjective measures of clinical orthopedic examination. The onset of effect and time to maximum effect of robenacoxib at higher dose (4 mg/kg) were faster than that of meloxicam (0.2 mg/kg, SC). There were no significant differences between 0.2 mg/kg meloxicam and 1 or 2 mg/kg robenacoxib dosage groups for any outcome measures. At all of the test doses, robenacoxib significantly inhibited COX-2 (measured in terms of ex vivo PGE_2 levels) without markedly affecting COX-1 (measured as ex vivo serum T_XB_2). Although there was no significant difference in analgesia between both drugs, meloxicam (0.2 mg/kg, SC) only moderately inhibited ex vivo PGE2 in plasma indicating lower selectivity for COX-2 than robenacoxib.[34] The researchers conclude that robenacoxib at dosages 1–2 mg/kg produces analgesia equivalent to 0.2 mg/kg meloxicam. In healthy beagle dogs, robenacoxib administered at daily dosages as high as 40 mg/kg for 1 month or 10 mg/kg daily for 6 months caused no evidence of toxicity indicating that it has a high safety index in dogs.[36]

Clinical studies in dogs

Efficacy and tolerability of robenacoxib have been compared with meloxicam, a reference NSAID in dogs, for the control of pain and inflammation in dogs undergoing soft tissue and orthopedic surgeries in two large separate multicenter, randomized, and blinded field trials.[37,38] Dogs received a single, preoperative dose of robenacoxib (2 mg/kg) or meloxicam (0.2 mg/kg) SC followed by daily oral doses (robenacoxib 1–2 mg/kg or meloxicam 0.1 mg/kg) for 12 and 15 days after soft tissue and orthopedic surgeries, respectively. In both studies, no significant differences in pain scores, obtained primarily by clinical investigators using a Glasgow pain scale, have been observed between the two treatments. None of the dogs in both groups required rescue analgesic therapy. The secondary efficacy variables such as changes in demeanor

and mobility, assessed by the dog owners supported the findings of primary efficacy variables. Both treatments were well tolerated without causing changes in buccal mucosal bleeding time. These studies indicate that robenacoxib could produce analgesia comparable to meloxicam and that both NSAIDs are well tolerated after oral dosing for 15 days in dogs undergoing soft tissue and orthopedic surgeries.

Friton et al[39] assessed the efficacy and safety of robenacoxib tablets in a prospective, multicenter, placebo controlled trial in dogs (n=239) undergoing soft tissue surgery. Robenacoxib, administered at a target dose of 2 mg/kg prior to surgery and once daily for two postoperative days, produced better analgesia than a placebo: analgesia was measured in terms of the need for rescue analgesia after surgery. No significant changes in the mean values for hematology and serum chemistry have been found in both groups. The most frequently reported AEs in both groups were emesis and diarrhea for a short period, similar to those reported in cats. Though it was not considered clinically relevant, a statistically significant rise in BUN/creatinine ratio has been detected in the robenacoxib group.

Robenacoxib has been compared with carprofen, a preferential COX-2 inhibitor and a commonly used NSAID in dogs, to establish the efficacy against chronic OA in dogs.[40] Both robenacoxib (1–2 mg/kg, Onsior® tablet; Novartis Santé Animale SA, Huningue, France) and carprofen (2–4 mg/kg, Rimadyl® tablet; Pfizer Inc, New York, NY, USA) were administered once daily for 12 weeks in dogs with OA of one or more joints that had been diagnosed for at least 3 weeks. NRSs were used to assess the efficacy of both treatments at days 0, 7, 14, 28, 56, and 84. Robencoxib showed efficacy noninferior to carprofen in improving the pain scores and functional disabilities of dogs. Small yet significant decreases in RBC count and changes in clinical chemistry from the baseline values have been found in both groups at the exit (84 days) of the study. Similar study has been conducted in Japan[41] to establish the efficacy and safety of robenacoxib in local dog breeds living in different conditions and geographical locations than Europe. Robenacoxib (1–2 mg/kg, tablet) and carprofen (3.5–5 mg/kg, tablet) were administered to dogs with OA once daily for 28 days. Robenacoxib produced noninferior efficacy and tolerability to carprofen for the clinical improvement of OA.

The tissue-selective anti-inflammatory activity of robenacoxib has been explored in dogs (n=34) with OA secondary to failure of the cranial cruciate ligament.[42] Oral robenacoxib (1 mg/kg, once daily [SID]) administered for 28 days decreased the lameness scores and improved the radiographic scores of arthritic joints. There was no significant reduction

in levels of serum C-reactive protein (CRP), which is an acute phase protein and highly sensitive indicator of inflammation, in response to treatment: but the levels of CRP in the synovial fluid sampled from the affected joint, decreased significantly after 28 days. This study supports the findings from preclinical studies in which robenacoxib produced persistent inhibition of PGE_2 in the inflammatory exudate despite rapid clearance from plasma.

Similar to the study in cats,[31] the renal safety of robenacoxib has been assessed in healthy dogs administered with ACEI and LD.[43] Robenacoxib, administered separately or in combination with benazepril (ACEI) with or without furosemide (LD), produced no evidence of acute renal injury.

The safety of interchangeable use of robenacoxib tablets (Onsior®) and solution for injection (20 mg/mL of robenacoxib) has been assessed in healthy dogs[44] for three 20-day cycles. The dosages ranged from 2 to 12 mg/kg, SID. The safety variables assessed were clinical observations indicating general health of the dog, body weight, food consumption, neurological examinations, injection site scoring, buccal mucosal bleeding time, and clinical pathology, gross, and microscopic examinations of GIT, liver, and kidney. No significant effect of the treatment was found on physical and neurological examinations. Injection site edema with erythema and skin thickening with granulation have been noticed 1–3 days after injection. Hematology, clinical chemistry, and urinalysis variables did not differ significantly between treatment and control groups.

Gross examination of the GIT revealed red discoloration of multiple segments in a few dogs. An ulcer in jejunum in one dog and cecal inflammation and hemorrhage in two dogs have been found. No evidence of renal and hepatic toxicities and coagulation abnormalities were noticed. This study recommends that interchangeable use of robenacoxib formulations is safe, despite minor and transient abnormalities observed. The safety of IV robenacoxib (2 and 4 mg/kg, bolus) has been compared with its administration by SC route at the recommended dose (2 mg/kg) for analgesia in healthy dogs.[45] No significant abnormalities in cardiovascular variables, buccal mucosal bleeding time, and hematology have been reported.

Conclusion

Several preclinical studies in rats, cats, and dogs demonstrated the high specificity of robenacoxib for COX-2 enzyme and its selective distribution and persistence at sites of inflammation. Rapid clearance of robenacoxib from the blood and consequently its transient effects on COX enzymes in the blood have been attributed for its wide margin of

safety, in addition to its COX-2 selectivity. Robenacoxib attains peak concentrations and persists longer in inflammatory exudate due to the acidic moiety in its structure and consequent high affinity for plasma proteins that carry the drug to sites of inflammation. This explains the longer efficacy after a single dose of robenacoxib, despite its rapid clearance from the blood. Large, multicentre clinical trials in cats and dogs demonstrated the noninferior efficacy and tolerability of robenacoxib (2 mg/kg, SC and PO) compared with noncoxib NSAIDs, against a variety of acute and chronic painful conditions. Preoperative robenacoxib (2 mg/kg, SC) showed superior efficacy to meloxicam, a preferential COX-2 inhibitor, after soft tissue and orthopedic surgeries in cats. Long-term administration of robenacoxib tablets (1–2 mg/kg) in cats with OA and concurrent CKD has been shown to be safe. Simultaneous administration of robenacoxib with potential nephro-toxic drugs did not cause acute renal injury in healthy dogs and cats. In summary, the efficacy and safety of robenacoxib have been well established in preclinical and clinical studies in cats and dogs. More studies are needed to fully explore the benefits of robenacoxib in cats and dogs.

Disclosure

The authors report no conflicts of interest in this work.

References

1. Vane JR. Inhibition of prostaglandin synthesis as a mechanism of action for aspirin-like drugs. *Nat New Biol*. 1971;231(25):232–235.
2. Smith JB, Willis AL. Aspirin selectively inhibits prostaglandin production in human platelets. *Nat New Biol*. 1971;231(25):235–237.
3. Xie WL, Chipman JG, Robertson DL, Erikson RL, Simmons DL. Expression of a mitogen-responsive gene encoding prostaglandin synthase is regulated by mRNA splicing. *Proc Natl Acad Sci U S A*. 1991;88(7):2692–2696.
4. Flower RJ. The development of COX2 inhibitors. *Nat Rev Drug Discov*. 2003;2(3):179–191.
5. Curry SL, Cogar SM, Cook JL. Nonsteroidal antiinflammatory drugs: a review. *J Am Anim Hosp Assoc*. 2005;41(5):298–309.
6. Brideau C, Van Staden C, Chan CC. *In vitro* effects of cyclooxygenase inhibitors in whole blood of horses, dogs, and cats. *Am J Vet Res*. 2001;62(11):1755–1760.
7. Giraudel JM, Diquelou A, Lees P, Toutain PL. Development and validation of a new model of inflammation in the cat and selection of surrogate endpoints for testing anti-inflammatory drugs. *J Vet Pharmacol Ther*. 2005;28(3):275–285.
8. Giraudel JM, Toutain PL, King JN, Lees P. Differential inhibition of cyclooxygenase isoenzymes in the cat by the NSAID robenacoxib. *J Vet Pharmacol Ther*. 2009;32(1):31–40.
9. Giraudel JM, King JN, Jeunesse EC, Lees P, Toutain PL. Use of a pharmacokinetic/pharmacodynamic approach in the cat to determine a dosage regimen for the COX-2 selective drug robenacoxib. *J Vet Pharmacol Ther*. 2009;32:18–30.
10. King JN, Dawson J, Esser RE, et al. Preclinical pharmacology of robenacoxib: a novel selective inhibitor of cyclooxygenase-2. *J Vet Pharmacol Ther*. 2009;32(1):1–17.
11. Riendeau D, Percival MD, Brideau C, et al. Etoricoxib (MK-0663): preclinical profile and comparison with other agents that selectively inhibit cyclooxygenase-2. *J Vet Pharmacol Ther*. 2001;296(2):558–566.
12. Gan TJ. Diclofenac: an update on its mechanism of action and safety profile. *Curr Med Res Opin*. 2010;26(7):1715–1731.
13. Brune K, Furst DE. Combining enzyme specificity and tissue selectivity of cyclooxygenase inhibitors: towards better tolerability? *Rheumatology*. 2007;46(6):911–919.
14. Bjarnason I, Smethurst P, Fenn CG, Lee CE, Menzies IS, Levi AJ. Misoprostol reduces indomethacin-induced changes in human small intestinal permeability. *Dig Dis Sci*. 1989;34(3):407–411.
15. Karim S, Habib A, Levy-Toledano S, Maclouf J. Cyclooxygenase-1 and -2 of endothelial cells utilize exogenous or endogenous arachidonic acid for transcellular production of thromboxane. *J Biol Chem*. 1996;271(20):12042–12048.
16. Patrignani P, Panara MR, Greco A, et al. Biochemical and pharmacological characterization of the cyclooxygenase activity of human blood prostaglandin endoperoxide synthases. *J Vet Pharmacol Ther*. 1994;271(3):1705–1712.
17. Schmid VB, Seewald W, Lees P, King JN. *In vitro* and *ex vivo* inhibition of COX isoforms by robenacoxib in the cat: a comparative study. *J Vet Pharmacol Ther*. 2010;33(5):444–452.
18. Giraudel JM, Gruet P, Alexander DG, Seewald W, King JN. Evaluation of orally administered robenacoxib versus ketoprofen for treatment of acute pain and inflammation associated with musculoskeletal disorders in cats. *Am J Vet Res*. 2010;71(7):710–719.
19. Pelligand L, King JN, Toutain PL, Elliott J, Lees P. Pharmacokinetic/pharmacodynamic modelling of robenacoxib in a feline tissue cage model of inflammation. *J Vet Pharmacol Ther*. 2012;35(1):19–32.
20. Pelligand L, King JN, Hormazabal V, Toutain PL, Elliott J, Lees P. Differential pharmacokinetics and pharmacokinetic/pharmacodynamic modelling of robenacoxib and ketoprofen in a feline model of inflammation. *J Vet Pharmacol Ther*. 2014;37(4):354–366.
21. Jung M, Lees P, Seewald W, King JN. Analytical determination and pharmacokinetics of robenacoxib in the dog. *J Vet Pharmacol Ther*. 2009;32(1):41–48.
22. Sano T, King JN, Seewald W, Sakakibara N, Okumura M. Comparison of oral robenacoxib and ketoprofen for the treatment of acute pain and inflammation associated with musculoskeletal disorders in cats: a randomised clinical trial. *Vet J*. 2012;193(2):397–403.
23. King S, Roberts ES, Roycroft LM, King JN. Evaluation of oral robenacoxib for the treatment of postoperative pain and inflammation in cats: results of a randomized clinical trial. *ISRN Vet Sci*. 2012;2012:794148.
24. Staffieri F, Centonze P, Gigante G, De Pietro L, Crovace A. Comparison of the analgesic effects of robenacoxib, buprenorphine and their combination in cats after ovariohysterectomy. *Vet J*. 2013;197(2):363–367.
25. Steagall PVM, Monteiro-Steagall BP, Taylor PM. A review of the studies using buprenorphine in cats. *J Vet Intern Med*. 2014;28(3):762–770.
26. Kongara K, Jhonson L, Kells NJ, Johnson CB, Dukkipati R, Mitchinson SL. Alteration of electroencephalographic responses to castration in cats by administration of opioids. *J Vet*. 2014;1(1):38–42.
27. Kamata M, King JN, Seewald W, Sakakibara N, Yamashita K, Nishimura R. Comparison of injectable robenacoxib versus meloxicam for peri-operative use in cats: results of a randomised clinical trial. *Vet J*. 2012;193(1):114–118.
28. Speranza C, Schmid V, Giraudel JM, Seewald W, King JN. Robenacoxib versus meloxicam for the control of peri-operative pain and inflammation associated with orthopaedic surgery in cats: a randomised clinical trial. *BMC Vet Res*. 2015;11(1):79.
29. King JN, King S, Budsberg SC, et al. Clinical safety of robenacoxib in feline osteoarthritis: results of a randomized, blinded, placebo-controlled clinical trial. *J Feline Med Surg*. 2016;18(8):632–642.
30. King JN, Hotz R, Reagan EL, Roth DR, Seewald W, Lees P. Safety of oral robenacoxib in the cat. *J Vet Pharmacol Ther*. 2012;35(3):290–300.
31. King JN, Panteri A, Graille M, Seewald W, Friton G, Desevaux C. Effect of benazepril, robenacoxib and their combination on glomerular filtration rate in cats. *BMC Vet Res*. 2016;12(1):124.

32. Thomas MC. Diuretics, ACE inhibitors and NSAIDs – the triple whammy. *Med J Aust*. 2000;172(4):184–185.

33. King JN, Rudaz C, Borer L, Jung M, Seewald W, Lees P. *In vitro* and *ex vivo* inhibition of canine cyclooxygenase isoforms by robenacoxib: a comparative study. *Res Vet Sci*. 2010;88(3):497–506.

34. Schmid VB, Spreng DE, Seewald W, Jung M, Lees P, King JN. Analgesic and anti-inflammatory actions of robenacoxib in acute joint inflammation in dog. *J Vet Pharmacol Ther*. 2010;33(2):118–131.

35. Borer LR, Seewald W, Peel JE, King JN. Evaluation of the dose-response relationship of oral robenacoxib in urate crystal-induced acute stifle synovitis in dogs. *J Vet Pharmacol Ther*. 2017;40(2):148–157.

36. King JN, Arnaud JP, Goldenthal EI, et al. Robenacoxib in the dog: target species safety in relation to extent and duration of inhibition of COX-1 and COX-2. *J Vet Pharmacol Ther*. 2011;34(3):298–311.

37. Gruet P, Seewald W, King JN. Evaluation of subcutaneous and oral administration of robenacoxib and meloxicam for the treatment of acute pain and inflammation associated with orthopedic surgery in dogs. *Am J Vet Res*. 2011;72(2):184–193.

38. Gruet P, Seewald W, King JN. Robenacoxib versus meloxicam for the management of pain and inflammation associated with soft tissue surgery in dogs: a randomized, non-inferiority clinical trial. *BMC Vet Res*. 2013;9:92.

39. Friton G, Thompson CM, Karadzovska D, King S, King JN. Efficacy and safety of oral robenacoxib (tablet) for the treatment of pain associated with soft tissue surgery in client-owned dogs. *BMC Vet Res*. 2017;13(1):197.

40. Reymond N, Speranza C, Gruet P, Seewald W, King JN. Robenacoxib vs. carprofen for the treatment of canine osteoarthritis; a randomized, noninferiority clinical trial. *J Vet Pharmacol Ther*. 2012;35(2):175–183.

41. Edamura K, King JN, Seewald W, Sakakibara N, Okumura M. Comparison of oral robenacoxib and carprofen for the treatment of osteoarthritis in dogs: a randomized clinical trial. *J Vet Med Sci*. 2012;74(9):1121–1131.

42. Bennett D, Eckersall PD, Waterston M, et al. The effect of robenacoxib on the concentration of C-reactive protein in synovial fluid from dogs with osteoarthritis. *BMC Vet Res*. 2013;9(1):42.

43. Panteri A, Kukk A, Desevaux C, Seewald W, King JN. Effect of benazepril and robenacoxib and their combination on glomerular filtration rate in dogs. *J Vet Pharmacol Ther*. 2017;40(1):44–56.

44. Toutain CE, Heit MC, King SB, Helbig R. Safety evaluation of the interchangeable use of robenacoxib (Onsior™) tablets and solution for injection in dogs. *BMC Vet Res*. 2017;13:359.

45. Desevaux C, Marotte-Weyn AA, Champeroux P, King JN. Evaluation of cardiovascular effects of intravenous robenacoxib in dogs. *J Vet Pharmacol Ther*. 2017;40(6):e62–e64.

46. Julius D, Basbaum AI. Molecular mechanisms of nociception. *Nature*. 2001;413(6852):203–210.

47. Basbaum A, Jessell T. The perception of pain. In: Kandel ER, Schwartz JH, Jessell TM, editors. *Principles of Neural Science*. 4th ed. New York: McGraw-Hill, Health Professions Division; 2000: 472–491.