Nonsteroidal anti-inflammatory drugs are used in humans and other animals because of their analgesic, anti-inflammatory, and antipyretic properties. The primary site for the toxic effects of NSAIDs is the gastrointestinal tract; it has been reported that adverse effects occur in 20% to 50% of people taking NSAID medication.1–5 Complications such as gastroduodenal erosion, ulceration, perforation, and stricture and delayed ulcer healing in people, horses, dogs, and rats have been reported.6–13 Complication rates in dogs receiving NSAIDs can be high. In 1 study,7 treatment with the NSAID derocoxib resulted in gastrointestinal tract perforation in 29 dogs, of which 20 died or were euthanized. In another study,10 17 of 24 dogs that were given ketoprofen, meloxicam, or carprofen at therapeutic dosages for 1 month developed lesions in the gastrointestinal tract. Endoscopically, lesions were observed to be most severe in the region of the gastric antrum.19

The NSAID carprofen is a propionic acid derivative.14 Carprofen is COX-2 selective but is only 1.75 times as selective for COX-2 as it is for COX-1; at recommended doses, carprofen inhibits phospholipase A2.

**Objective**—To evaluate the effects of carprofen and meloxicam on conductance and permeability to mannitol and on the histologic appearance of sections of canine gastric mucosa.

**Sample**—Gastric mucosa from 6 mature mixed-breed dogs.

**Procedures**—Sections of gastric mucosa were mounted in Ussing chambers, and carprofen (40 or 400 µg/mL [CAR40 and CAR400, respectively]), meloxicam (8 or 80 µg/mL [MEL8 and MEL80, respectively]), or no drug (controls) was added to the bathing solution. For all sections, conductance was calculated every 15 minutes for 240 minutes and flux of mannitol was calculated for 3 consecutive 1-hour periods; histologic examination was performed after the experiment. The area under the conductance-time curve for each chamber was calculated. Values of conductance X time, flux of mannitol, and the frequency distribution of histologic findings were analyzed for treatment effects.

**Results**—For CAR400- and MEL80-treated sections, conductance X time was significantly higher than that for control and MEL8-treated sections. The effect of CAR40 treatment was not different from that of any other treatment. Over the three 1-hour periods, mannitol flux increased significantly in MEL80-, CAR40-, and CAR400-treated sections but not in MEL8-treated or control sections. Major histologic changes including epithelial cell sloughing were limited to the CAR400-treated sections.

**Conclusions and Clinical Relevance**—In the gastric mucosa of dogs, carprofen and meloxicam increased in vitro conductance and permeability to mannitol. At a concentration of 400 µg/mL, carprofen caused sloughing of epithelial cells. Carprofen and meloxicam appear to compromise gastric mucosal integrity and barrier function in dogs. (Am J Vet Res 2011;72:570–577)
Carprofen is highly protein bound in the blood and undergoes hepatic metabolism before being secreted in the bile into the duodenum, which allows enterohepatic recirculation. In dogs, 70% to 80% of a PO or IV administered dose of carprofen is eliminated in the faeces, with the remainder eliminated in the urine.17, 18

The enolic acid NSAID meloxicam is COX-2 selective and is 12 times as selective for COX-2 as it is for COX-1 in dogs.15 The specificity of meloxicam for the COX-2 isoenzyme is decreased at high doses.18 Meloxicam is well absorbed after oral administration, is metabolized in the liver, and undergoes extensive enterohepatic recirculation. Both unchanged meloxicam and its metabolites are primarily eliminated in the faeces.18

The mechanism by which NSAIDs cause gastrointestinal tract injury is not completely understood. All NSAIDs reduce prostaglandin synthesis via inhibition of the COXs (COX-1 and COX-2), thereby reducing the gastric mucosal protective effects of prostaglandins.11,19 Oxygen free radical–mediated lipid peroxidation is also considered important, as is the role of nitric oxide.20,21 Direct chemical damage is induced by altering the permeability of gastric mucosal cells to hydrogen ions, resulting in ion trapping, cellular injury, and ultimately cell death.22 Uncoupling of oxidative phosphorylation in the mitochondria and depletion of ATP has also been demonstrated.23 The nonselective NSAID aspirin significantly decreases prostanoid concentrations in the gastric and duodenal mucosa, compared with the effects of a COX-2–selective NSAID such as carprofen.9 The NSAIDs can also induce platelet aggregation and neutrophil adherence in the gastrointestinal tract.24 Interestingly, COX inhibition can increase leukotriene synthesis, and leukotrienes may damage gastric mucosa by promoting tissue ischemia and inflammation.25,26

To our knowledge, the effect of carprofen and meloxicam on the integrity and barrier function of the gastric mucosa of dogs has not been studied. The purpose of the in vitro study of this report was to measure the conductance and permeability to mannitol and to describe the histologic appearance of canine gastric mucosa in the presence of carprofen and meloxicam.

Materials and Methods

Harvesting of gastric mucosa sections—Six healthy research dogs with no prior involvement in research and no abnormalities detected via physical examination were the source of gastric mucosa sections. The dogs were used in accordance with the Louisiana State University Institutional Animal Care and Use Committee policy, and approval was obtained for the study protocol. For each dog, anaesthesia was induced with 3% thiopental (5 mL/kg IV, to effect) and maintained with 1% isoflurane in oxygen delivered via an endotracheal tube. Immediately after anesthetic induction, the body of the stomach was isolated, harvested, and washed in Krebs solution. Euthanasia of each dog had been scheduled for reasons unrelated to the study, and euthanasia was then immediately performed with an IV overdose injection of sodium pentothal. The tissue was then moved to a stripping pan filled with 400 mL of iced, carbogenated (95% O2 and 5% CO2) Krebs solution, and the mucosa was separated from the muscular layer via sharp dissection. The mucosa was cut into 11 sections (each approx 3 cm2). Ten sections were each randomly assigned to 1 of 10 Ussing chambers (aperture, 3.14 cm2). All sections from a given dog were processed at the same time. The remaining section was immediately placed in neutral-buffered 10% formalin for later histologic examination.

Ussing chamber experiments—Each section of mucosa was clamped as a flat sheet between the 2 halves of an acrylic chamber. Each hemichamber was filled with 15 mL of Krebs solution. The Krebs solution was continuously carbogenated (with 95% O2 and 5% CO2) and circulated in water-jacketed reservoirs. The temperature of the solution was maintained at 37°C, and the pH of the solution maintained at 7.4. The tissue was allowed to equilibrate for 30 minutes prior to treatment with carprofen or meloxicam or prior to use as a control section. Treatments were randomly assigned to the sections in the 10 chambers (2 chambers/treatment). Sections in the pairs of chambers were each treated with CAR40, CAR400, MEL8, or MEL80 or received no drug treatment (control sections); however, not all sections maintained their integrity to the end of the experiment. In all 4 treatment groups, high concentrations of carprofen and meloxicam were used to maximize the confidence of obtaining a significant result. The maximum plasma concentration following oral administration of 25 mg of carprofen in Beagles is 18.7 μg/mL, and 25 mg represents half the recommended clinical dose of carprofen. The plasma concentration of carprofen is directly proportional to the dose. A treatment concentration of 400 μg of carprofen/mL is approximately 10 times as great as the plasma concentration achieved with a full, single, daily dose in vivo, and is realistic in terms of potential clinical overdose. Meloxicam plasma concentrations were similarly derived from studies in dogs.27, 29

Conductance—The potential difference was measured by use of agar bridges connected to Ag-AgCl voltage electrodes. If the potential difference was between −1.0 and 1.0 mV, tissues were current clamped at 100 μA for 5 seconds and the potential difference was recorded. The potential difference was short circuited through the voltage electrodes with a voltage clamp that corrected for fluid resistance. The short circuit current was measured by use of a separate pair of agar bridges connected to Ag-AgCl current electrodes. The potential difference and short circuit current were recorded every 15 minutes for 240 minutes. Conductance was calculated from the potential difference and short circuit current by use of Ohm’s law as follows:

Potential difference (mV) = Resistance (Ω) X short circuit current (μA)
Conductance (millisiemen) = 1/Resistance.

Mannitol flux—Tritiated (3H) mannitol (10 μCi/mL) was added to the mucosal bathing solution 15 minutes after mounting of each section. A postequilibration sample (0.1 mL) was collected from the mucosal solution 30 minutes after addition of 3H-mannitol (45 min-

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cells without erosion or ulceration of the underlying epithelium was defined as detachment of surface epithelial sloughing without subsequent development of erosions to compensate for areas of sloughing. For purpuric mucosal epithelial cells into the gastrointestinal lumen and not used in Ussing chamber procedures was evaluated to verify preservation of the mucosa from the chamber and immediately fixed in neutral-buffered 10% formalin for later histologic examination. Fixed segments of gastric mucosa were trimmed, embedded in paraffin, and sectioned at a thickness of 5 μm. Sections were mounted on slides and stained with H&E stain. Sections were evaluated via light microscopy by 1 evaluator (TWM), a board-certified veterinary pathologist who was unaware of the treatment assignment of each section. Mucosa obtained from the stomach of each dog prior to the start of the experiment and not used in Ussing chamber procedures was evaluated to rule out the presence of preexisting disease. Sections removed from the Ussing chambers at the end of the experiment were evaluated to verify preservation of the tissue. All sections were examined for inflammation, sloughing of surface epithelium, edema, and necrosis of cells within the gastric glands.

Because the treatments were done on isolated tissues, an influx of inflammatory cells did not occur during the experiments. The level of inflammation was determined by subjective scoring of H&E-stained slides examined via light microscopy by the same board-certified veterinary pathologist (TWM) who examined the experimental sections. If < 20% of lamina propria contained inflammatory cells (as determined via visual inspection), the level of inflammation was considered normal background inflammation. If 20% to 60% of the lamina propria contained inflammatory cells (as determined via visual inspection), the level of inflammation was considered moderate. If ≥ 61% of the lamina propria contained inflammatory cells (as determined via visual inspection), the level of inflammation was considered severe.

Within the gastrointestinal tract, the mucosal epithelial lining cells have a limited life span and are released or sloughed into the gastrointestinal lumen at the end of their life span. When these cells are released, the adjacent epithelial cells slide along the basement membrane to maintain continuity of the mucosal barrier and prevent any gaps or erosions from forming. Under conditions of increased stress or increased damage to the mucosal epithelial cells, the rate of turnover of mucosal epithelial cells into the gastrointestinal lumen increases. To a certain extent, the adjacent mucosal epithelial cells are able to slide along the basement membrane to compensate for areas of sloughing. For purposes of this study, the amount of this type of epithelial sloughing without subsequent development of erosions was evaluated histologically. Sloughing of mucosal epithelium was defined as detachment of surface epithelial cells without erosion or ulceration of the underlying mucosal epithelium. Sloughing of the surface epithelium was classified as follows: minimal, ≤ 10% of the mucosal surface had adjacent sloughed epithelial cells; mild, 11% to 20% of the mucosal surface had adjacent sloughed epithelial cells; moderate, 21% to 50% of the mucosal surface had adjacent sloughed epithelial cells; and severe, ≥ 51% of the mucosal surface had adjacent sloughed epithelial cells.

Necrosis of cells within the gastric glands was classified as follows: absent, no glands contained necrotic cells; mild, < 5% of gastric glands contained necrotic cells; moderate, 5% to 10% of gastric glands contained necrotic cells; and severe, ≥ 11% of gastric glands contained necrotic cells.

Statistical analysis—For all analyses, a value of P ≤ 0.05 was considered significant.

Data from the 0- and 15-minute time points (ie, during the 30-minute equilibration period) were not used for analysis. Conductance at 30 to 240 minutes was plotted against time for each chamber, and the AUC was calculated by use of the trapezoid method. Sections from each dog that received the same treatment were considered replicates. The AUC was the response variable used for the statistical analysis. The mean ± SEM AUC (conductance X time) for each treatment was calculated. The conductance X time data were normally distributed as verified by failure to reject the null hypothesis of normality at a value of P ≤ 0.05 (Shapiro-Wilk statistic). The conductance X time was compared for a fixed effect of treatment by use of a mixed-effect linear model that included the random variance of dog across treatments. Where differences were significant at a value of P ≤ 0.05, post hoc comparisons were made with the Scheffé adjustment to maintain α at 0.05.

The mucosal-to-serosal flux of mannitol was calculated for three 1-hour periods: 60 to 120 minutes, 120 to 180 minutes, and 180 to 240 minutes. Sections from each dog that received the same treatment were considered replicates. The mean ± SEM mannitol flux for each period was calculated for each treatment and was used as the response variable for statistical analysis. The data were normally distributed as verified by failure to reject the null hypothesis of normality at a value of P ≤ 0.05 (Shapiro-Wilk statistic). The mannitol flux was compared for fixed effects of treatment and period by use of a mixed-effect linear model that included the random variance of dog across treatments and periods. Where differences were significant at a value of P ≤ 0.05, post hoc comparisons were made with the Scheffé adjustment to maintain α at 0.05.

The frequency distributions of histologic findings from untreated and treated mucosal sections were compared by use of a Fisher exact test. The null hypothesis of similar distributions was rejected against a 2-sided hypothesis at a value of P ≤ 0.05. Computer software programs were used for the analysis.

Results

Conductance—In general, the conductance in the canine gastric mucosal sections increased over time; this effect was most pronounced in CAR400- and MEL80-treated sections (Figure 1). Significant (P <

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Results

Conductance—In general, the conductance in the canine gastric mucosal sections increased over time; this effect was most pronounced in CAR400- and MEL80-treated sections (Figure 1). Significant (P <
0.05) differences in mean conductance X time among treatments were detected (Table 1). The CAR400- and MEL80-treated sections had significantly (P < 0.05) higher values of conductance X time, compared with values for untreated sections and MEL8-treated sections. The effect of CAR40 treatment on conductance did not differ from the effects of other drug or control treatments. It should be noted that duplicate treatments (CAR40, CAR400, MEL8, MEL80, or no drug treatment [control sections]) were randomly assigned to the sections in the 10 chambers (2 chambers/treatment). However, not all sections maintained their integrity to the end of the experiment, and the number of sections in each group consequently varied.

Mannitol flux—Mannitol flux did not change significantly among the 3 consecutive 1-hour periods.
Table 3—Distribution of histologic findings for sections of gastric mucosa (obtained from 6 dogs) that received no treatment (control) or CAR40, CAR400, MEL8, or MEL80 treatment for 240 minutes in Ussing chamber experiments.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Sloughing of epithelium</th>
<th>Necrosis of cells within gastric glands</th>
<th>Inflammation</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Minimal</td>
<td>Mild</td>
<td>Moderate</td>
</tr>
<tr>
<td>Control (12)</td>
<td>42</td>
<td>50</td>
<td>8</td>
</tr>
<tr>
<td>CAR40 (10)</td>
<td>50</td>
<td>25</td>
<td>25</td>
</tr>
<tr>
<td>CAR400* (10)</td>
<td>9</td>
<td>9</td>
<td>27</td>
</tr>
<tr>
<td>MEL8 (8)</td>
<td>40</td>
<td>40</td>
<td>20</td>
</tr>
<tr>
<td>MEL80 (11)</td>
<td>30</td>
<td>50</td>
<td>10</td>
</tr>
</tbody>
</table>

Data are reported as the percentage of sections in each category. *Indicates that the distribution of the histologic findings are significantly (P ≤ 0.05) different from untreated sections. See Table 1 for remainder of key.

Discussion

In the study of this report, in vitro treatment with carprofen and meloxicam altered the electrical conductance and permeability to mannitol of sections of canine gastric mucosa, indicating that the treatments compromised the mucosal integrity and resulted in loss of barrier function. In addition, carprofen caused sloughing of epithelial cells, which suggested a loss of viability of those cells.

Mechanisms that could be responsible for the increase in conductance and permeability to mannitol of the gastric mucosa after treatment with carprofen include changes in transcellular and paracellular ion transport (which alter the transepithelial voltage) or alterations to intercellular tight junctions (which determine the resistance and integrity of epithelia).

Sloughing of the epithelial cells from the mucosal surface was also likely, in part, responsible for the increase in electrical conductance and permeability by allowing indiscriminant passage of ions and molecules of mannitol through areas devoid of epithelial cells. Additional mechanisms are likely to be in effect because increases in electrical conductance and permeability to mannitol were also detected after treatment with meloxicam, despite absence of epithelial sloughing. According to Ohm’s law, electrical conductance increases proportionally with short circuit current flowing through the conductor (ie, the gastric mucosa in our study), as long as the transepithelial potential difference across the conductor is unchanged. Transport of chloride, sodium, and potassium ions account for most of the in vitro short circuit current across the epithelium of the intestinal mucosa, and carprofen may have altered the transport of 1 or more of these ions, resulting in an increase in electrical conductance.

Cell-to-cell integrity and barrier function are maintained by junctional complexes composed of tight junctions, adherens junctions, and desmosomes including the protein zonula occludens. Carprofen and meloxicam may have increased electrical conductance and permeability to mannitol through interference with the intercellular tight junctions of the gastric mucosa. Aspirin increases the transport of fluorescein-labeled dextran by locally disrupting and decreasing the expression of zonula occludens proteins in rat gastric mucosal cell lines, and a similar mechanism may be responsible for the increased mannitol flux in the carprofen- and meloxicam-treated sections in our study. Interestingly, in the low-dose carprofen-treated (CAR40) group, de-

for untreated sections (P = 0.070) or MEL8-treated sections (P = 0.098). However, there were significant increases in flux among the three 1-hour periods when sections received the MEL80 (P = 0.003), CAR40 (P = 0.001), and CAR400 (P = 0.033) treatments (Figure 2; Table 2).

Histologic findings—No evidence of preexisting disease that might have excluded a dog from the study was noted during histologic examination of any of the sections that were fixed immediately after harvest. The inflammation in the sections that were fixed immediately after harvest was minimal and was consistent with the typical background level of inflammation expected in normal tissue. Among the sections that received drug or control treatments in the Ussing chambers, there was minimal surface epithelial sloughing in all but 1 section, which had mild sloughing (Figure 3). Necrosis of cells within the gastric glands was absent in all but 1 section, which had mild changes. Inflammation was minimal in all but 1 section, which had moderate inflammation. Spirochetes were detected in small numbers in 1 section and in large numbers in another.

Compared with findings in untreated control sections, the distribution of categories for the extent of surface epithelial sloughing was significantly (P = 0.002) different in CAR400-treated sections (Figure 1; Table 3). In sections treated with CAR400, sloughing was mostly severe and moderate; in control sections, sloughing was mostly minimal and mild. The distributions of sloughing severity categories for the other treated sections did not differ from findings in the control sections. However, it is interesting to note that in sections that had severe sloughing, underlying mucosal cells provided an intact epithelial coverage and there was no erosion or exposure of the lamina propria.

The distributions of categories for the extent of necrosis and inflammation in any of the treated sections did not differ significantly from findings in the untreated control sections (Table 3). Overall, inflammation was mild, and sections had amounts of superficial or deep lymphocytic-plasmacytic infiltrates expected in gastric mucosal tissue of healthy dogs; there was an occasional neutrophilic focus in 3 sections. A small number of spirochetes was detected in 20 sections (from 4 dogs), whereas a large number of spirochetes was detected in 5 sections (from 3 dogs). Mild edema of the lamina propria was seen in 1 CAR400-treated section.
fects in mucosal integrity were detected by fluxes, but were not apparent electrophysiologically. Flux is a more sensitive indicator of alterations in permeability, and this is likely to be a reflection of the different physiological mechanisms involved in the NSAID-induced changes in mucosal integrity.11,13-15

Oral administration of indomethacin uncouples oxidative phosphorylation and depletes ATP in the mitochondria of epithelial cells of the intestinal mucosa of rats.23 This results in increased cytosolic calcium concentration, which adversely affects the functional integrity of intracellular tight junctions.23 Uncoupling of oxidative phosphorylation and calcium release from storage vesicles in cultures of human colonic cells caused by increased intracellular concentration of calcium that subsequently compromised functional integrity of the tight junctions has also been reported.33 A simultaneous increase in electrical conductance and permeability to mannitol was also detected in colonic cell monolayers.35 If carprofen and meloxicam had a similar effect and caused uncoupling of oxidative phosphorylation in the mitochondria of gastric mucosal cells of dogs, compromised functional integrity of intracellular tight junctions may explain, in part, the increase in electrical conductance and permeability observed in our study, although this warrants further investigation.

Other investigators have shown that NSAIDs cause direct chemical damage by altering the permeability of gastric mucosal cells to hydrogen ions, which results in ion trapping, cellular injury, and ultimately cell death.32 Acetylsalicylate induces topical damage as a result of accumulation of the ionized NSAID in gastric epithelial cells, which causes ion trapping.36 It may be that ion trapping is responsible, in part, for the increased conductance and permeability detected in canine gastric mucosa sections following carprofen and meloxicam treatments in our in vitro study.

Increased platelet aggregation, neutrophil adherence in the gastrointestinal tract, leukotriene-mediated gastric mucosal ischemia and inflammation, reduced gastric mucosal protective effects of prostaglandins, and alterations to nitric oxide synthesis are important in the pathogenesis of NSAID-induced gastropathy.9,20,24-26 The nonselective NSAIDs, such as aspirin, significantly decrease prostanoid concentrations in the gastric and duodenal mucosa, compared with the effects of a selective COX-2 NSAID, such as carprofen.9 Although some of the mechanisms of NSAID-induced gastropathy, such as effects on prostaglandin synthesis, nitric oxide synthesis, and neutrophil and leukocyte activities, can only be investigated in vivo, mechanisms that cause a direct effect via ion trapping or uncoupling of oxidative phosphorylation are most relevant in the in vitro experiments performed in our study.

Carprofen-treated sections had significantly more sloughing of cells from the surface epithelium of the gastric mucosal sections than did untreated sections. The mechanism of this damage was not investigated in the present study. In Ussing chamber experiments, a gradual increase in the permeability of rat intestinal mucosa was detected over a 180-minute period.37 The gradual increase in permeability to mannitol may be a result of sloughing of cells from the intestinal mucosa.37 However, this occurred in the face of an extensive repair process (reduction in villi index and nucleoapical distance),37 which was not observed in the canine gastric mucosal sections used in our study. Although some cell loss may be attributed to the time spent in the chamber, the changes that were observed in our study—in the absence of any repair—were likely to be induced by the treatment and not simply an effect of time.

Similar histologic observations of mucosal sloughing in carprofen-treated canine colonic mucosa have been previously reported.36 In that study,36 mild epithelial sloughing was observed in 5 of 63 (7.9%) sections, moderate sloughing was observed in 17 of 63 (27.0%) sections, and severe sloughing was observed in 41 of 63 (65.0%) sections. No sloughing of cells was detected within the mucosal glands in 39 of 63 (61.9%) treated sections; sloughing to some extent was detected in 38.1% of sections.36 Edema was not evident in 16 of 63 (25.4%) sections but was rated mild in 37 (58.7%) and moderate in 10 (15.9%) sections.36 It was suggested that sloughing of cells and erosions likely contributed to an increase in electrical conductance and permeability recorded in carprofen-treated sections, compared with findings in untreated control samples.36

In the present study, histologic examination of all sections of gastric mucosa at the end of the experiment was performed to verify viability and preservation of structural integrity in the control sections and to compare the histologic findings from the carprofen- and meloxicam-treated sections. Sloughing of cells from the surface epithelium, sloughing of cells within the mucosal glands, and edema in the colonic mucosa of rats after 60 minutes of immersion in an Ussing chamber was detected in another study.37 In that study, severe but not mild histologic changes were associated with increased permeability. Edema and mild sloughing of surface epithelial cells may represent epithelial loss within the limits of repair of the epithelium and do not indicate complete loss of integrity or barrier function.36 In the study reported here, the epithelium was intact (without erosions) in most of the control sections (11/12 sections had mild or minimal sloughing) and edema was not detected in the control sections. The distributions of categories for the extent of necrosis and inflammation were not significantly different between the untreated control sections and any of the treated sections. Hence, these mild histologic changes, along with relatively stable conductance and permeability through the duration of the experiment, suggest that barrier function and integrity of the control gastric mucosa sections were adequately preserved.

Similarly, minimal histologic changes were observed in the sections that received the lower-dose meloxicam treatment (MEL8), and conductance and mannitol permeability did not change in these sections throughout the experiment. It would appear that MEL8 treatment did not cause a detrimental effect on the gastric mucosa. In contrast, of the 10 sections that received the high-dose carprofen treatment (CAR400), 55% and 27% had severe and moderate sloughing, respectively. This may also be a function of the aforementioned mechanisms that are likely to alter electrical conductance and mannitol flux, thereby compromising...
the mucosal integrity; however, conclusions regarding clinical relevance cannot be drawn.

Meloxicam is in the enolic-acid group of NSAIDs, whereas carprofen is a propionic acid derivative. The 2 drugs have basic differences in their chemical composition. These compositional differences may account for the differences in direct mucosal effects and resultant histologic appearance of the gastric mucosa sections treated with these drugs, particularly in the higher-dose treatment groups. Carprofen is highly protein bound in the circulation and undergoes hepatic metabolism before being secreted in the bile and delivered to the duodenum, which allows enterohepatic recirculation. In dogs, biliary secretion predominates; following IV administration, 70% of a dose of carprofen is excreted in feces and 8% to 13% of that dose is excreted in urine. This may, in part, explain why a direct effect of carprofen on the mucosa, resulting in mucosal damage, is associated with the lower portions of the small intestine clinically. Bile also moves into the stomach. In 1 study in dogs treated with NSAIDs, the pyloric antrum was the most severely affected region of the stomach, a finding that supports a direct effect of the secretion of carprofen in bile. Enterohepatic recirculation results in repeated exposure of the intestinal mucosa to an administered NSAID, and the combination of an NSAID with bile appears to be much more damaging to the mucosa than the effect of an NSAID alone. Therefore, clinical use of an NSAID that undergoes enterohepatic recirculation may increase the likelihood of intestinal ulceration.

In the study of this report, 1 section of mucosa was placed in formalin immediately after it was harvested from the stomach of each dog and was examined for the microscopic presence of preexisting gastric disease. Although it was assumed that the histologic findings in a given section were representative of the entire stomach from which it was harvested, that may not have been the case. Certain of these sections contained parasites, but the extent of inflammation was rarely more than mild. For the purposes of this experiment, and given the notable effects of the higher dose of carprofen, we do not believe preexisting disease was a factor in the study results.

The concentrations of carprofen and meloxicam in the solutions applied to the gastric mucosa tissue sections were 40 and 400 μg/mL and 8 and 80 μg/mL, respectively. Because the effect of carprofen on canine gastric mucosa has not been investigated to our knowledge, we began our experiments with high concentrations of carprofen and meloxicam to maximize the confidence of obtaining a significant result. Although a plasma concentration in vivo cannot be directly extrapolated to a solution concentration in a tissue bath in vitro, we used the information currently available on plasma concentrations achieved after administration of carprofen and meloxicam in dogs. The maximum plasma concentration after an orally administered dose of 25 mg of carprofen in Beagles is 18.7 μg/mL. Based on the expected body weight and size for the breed, that orally administered dose corresponds to approximately 2.2 mg/kg, half the total recommended daily dose of carprofen in dogs. The plasma concentration of carprofen is directly proportional to the dose. A concentration of 400 mg of carprofen/mL is approximately 10 times as great as the plasma concentration achieved following administration of a full, single, daily dose in vivo and is realistic in terms of potential clinical overdosage. Meloxicam plasma concentrations were similarly derived from studies in dogs. Whether the effect of a specific plasma concentration of carprofen or meloxicam on the gastric mucosa of dogs in vivo is the same as the effect of the corresponding concentration on gastric mucosa sections in vitro is unknown. Also, whether repeated administration of carprofen or meloxicam has a cumulative damaging effect on the colonic mucosa of dogs in vivo is also unknown.

In the in vitro study of this report, carprofen and meloxicam increased in vitro electrical conductance and permeability to mannitol in sections of canine gastric mucosa. In addition, carprofen caused sloughing of epithelial cells, which suggests a loss of viability of these cells. On the basis of these findings, it appears that carprofen and meloxicam can compromise the integrity and barrier function of the gastric mucosa in dogs. Further studies are needed to investigate the effects of these drugs in clinical patients.

References


b. Metacam, Boehringer Ingelheim, Ingelheim/Rhein, Germany.
c. BX41 microscope, Olympus America Inc, Center Valley, Pa.
d. PROC UNIVARIATE, PROC MIXED, and PROC FREQ, version 9.1, SAS Institute Inc, Cary, NC.