Grapiprant is a selective antagonist of the EP4 receptor, whose physiological ligand is prostaglandin E2 (PGE2). The EP4 receptor is one of four G-protein coupled receptors (EP1, EP2, EP3 and EP4) that mediate the action of prostaglandin E2 (PGE2). The EP4 receptor mediates PGE2-elicited sensitization of sensory neurons (Southall and Vasko, 2001) and studies have demonstrated that EP4 is a major receptor in mediating pain associated with both rheumatoid and osteoarthritis (Clark et al., 2008 and Chen et al., 2010) and in inflammation (Lin et al., 2006 and Nakao et al., 2007). Grapiprant is under development for use in humans and dogs for the control of pain and inflammation associated with osteoarthritis. The study described here was undertaken to evaluate the potential toxicity and systemic exposure of grapiprant in Beagle dogs and also to assess the reversibility of any observed changes.

Grapiprant was administered orally by gavage, once daily, for 9 consecutive months to Beagle dogs at doses of 0 (0.5% methylcellulose), 1, 6, and 50 mg/kg/day in a dose volume of 5 mL/kg. Four animals/sex were used in each dose group and 2 additional animals/sex were used in the 50 mg/kg dose group for recovery purposes in the study. Clinical signs and food consumption were assessed daily. Body weight was recorded weekly. Ophthalmologic examinations, electrocardiograms, and clinical pathology analysis and urinalysis were conducted at several time-points during the dosing and recovery phases. Serum drug concentrations of grapiprant were measured on Day 1 (50 mg/kg only) and Week 38. At the end of the dosing or recovery period, dogs were euthanized and necropsied. After gross examination, selected organs were weighed, and a comprehensive set of tissues was collected and processed for microscopic examination.

Grapiprant at doses given daily up to 50 mg/kg (more than 10X the anticipated dose for dogs with osteoarthritis) for 9 months resulted in minimal toxicity, with no mortality, or effects on body weight, food consumption, ophthalmic exams, electrocardiograms, hematology, coagulation, organ weights, or gross pathology. Treatment was associated with mild gastrointestinal signs such as soft formed stools, stool with mucus, and occasional blood seen in the stool. Emesis was seen sporadically and watery stool was seen sporadically with prolonged use. Treatment was also associated with mild and reversible decreases in total protein and albumin over time, with incidence increasing as dose increased. Calcium decreases were also seen but were considered secondary to diet and low albumin levels. Serum drug concentrations were dose proportional from 1 to 6 mg/kg and more than dose proportional from 6 to 50 mg/kg. The only histopathology of note was a mild mucosal regeneration of the ileum in one dog at 50 mg/kg.

Treatment with grapiprant was well-tolerated when given at doses up to 50 mg/kg for 9 months.

**ABSTRACT OT-16**

Grapiprant is a selective antagonist of the EP4 receptor, whose physiological ligand is prostaglandin E2 (PGE2). The EP4 receptor is one of four G-protein coupled receptors (EP1, EP2, EP3 and EP4) that mediate the action of prostaglandin E2 (PGE2). The EP4 receptor mediates PGE2-elicited sensitization of sensory neurons (Southall and Vasko, 2001) and studies have demonstrated that EP4 is a major receptor in mediating pain associated with both rheumatoid and osteoarthritis (Clark et al., 2008 and Chen et al., 2010) and in inflammation (Lin et al., 2006 and Nakao et al., 2007). Grapiprant is under development for use in humans and dogs for the control of pain and inflammation associated with osteoarthritis. The study described here was undertaken to evaluate the potential toxicity and systemic exposure of grapiprant in Beagle dogs and also to assess the reversibility of any observed changes.

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Treatment with grapiprant was well-tolerated when given at doses up to 50 mg/kg for 9 months.

**INTRODUCTION**

Osteoarthritis affects up to 20% of all dogs over one year old (Johnson, 1997). It is a progressive disease with degradation of articular cartilage and inflammatory arthropathies. It can affect dogs of all ages, sizes, and breeds, although it is most common in large breed and overweight dogs (Roush et al., 2010). Treatment of dogs with arthritis is focused on reduction of pain and inflammation. Non-steroidal anti-inflammatory drugs that inhibit the cyclooxygenase enzymes are currently the most frequently recommended treatment. Grapiprant, a treatment with a different mechanism of action that also decreases pain and inflammation, is being developed for the treatment of osteoarthritis in dogs. It belongs to a new class of drugs, the piprants. This study describes a safety study for grapiprant, a small molecule, orally available prostaglandin EP4 receptor antagonist.

Prostaglandin E2 (PGE2), a product of the arachidonic acid pathway, is an important mediator that contributes to inflammatory pain. The EP4 receptor is one of four G-protein coupled receptors (EP1, EP2, EP3, and EP4) that mediate the action of PGE2.

The EP4 receptor mediates PGE2-elicited sensitization of sensory neurons (Southall and Vasko, 2001) and studies have demonstrated that EP4 is a major receptor in mediating pain associated with both rheumatoid and osteoarthritis (Clark et al., 2008 and Chen et al., 2010). Grapiprant is a selective antagonist of the EP4 receptor which is under development for the control of pain and inflammation associated with osteoarthritis in dogs. This study was undertaken to evaluate the safety and systemic exposure of grapiprant when administered orally, once daily, for 9 consecutive months, to Beagle dogs. The study also was designed to assess the reversibility of any toxic changes.

Grapiprant Uniquely Targets the Prostaglandin EP4 Receptor
**MATERIALS & METHODS**

The study was conducted under Good Laboratory Practices (GLP).

Grapiprant was administered orally by gavage as a methylcellulose suspension, once daily, for 9 consecutive months to laboratory Beagle dogs at doses of 0 (0.5% methylcellulose), 1, 6, and 50 mg/kg/day in a dose volume of 5 mL/kg.

- Each group (n=8) had 4 animals/sex with an additional 2 animals/sex in the 50 mg/kg/day dose group.
- Clinical signs and food consumption were assessed daily.
- Body weight was recorded weekly.
- Ophthalmologic examination was performed on Weeks 20, 38, and 4.
- Urinalyses were performed on Week 37 of the dosing phase & Week 4 or 5 of the recovery phase.
- Organ weights, coagulation, and serum chemistry parameters were monitored on Weeks 13, 26, 39, & Week 4 or 5 of the recovery phase.
- Urinalyses were performed on Week 37 of the dosing phase & Week 3 or 4 of the recovery phase.
- Electrocardiograms were measured at 0.5, 1, 2, 4, and 24 hours post-dose on Day 1 and one day during Week 38.

**RESULTS**

**Grapiprant at doses given daily up to 50 mg/kg for 9 months resulted in minimal toxicity.**

- No drug related effects on:  
  - Body weight  
  - Ophthalmic exams  
  - Hematology  
  - Organ weights  
  - Clinical signs restricted to mild gastrointestinal signs associated with treatment  
  - Soft stool  
  - Occasional blood in stool  
  - Sporadic watery stool with prolonged use  
  - Clinical pathology: minor changes seen (no effect on liver enzymes, BUN/creatinine, platelet function, etc.)

- Mild and reversible decreases in total protein and albumin were seen over time with increased incidence as dose increased
- Calcium decreases secondary to low protein
- Colorless homogenous 2.5-10 micrometer spheres (too numerous to count) seen in urine sediment of some females at 50 mg/kg dose
- Histopathology: minor finding (no effects seen on liver, kidney, stomach)

Serum drug concentrations were dose proportional from 1 to 6 mg/kg and more than dose proportional from 6 to 50 mg/kg. There were no gender differences in the systemic exposure to grapiprant, and no evidence of drug accumulation.

**SUMMARY**

Treatment with grapiprant was well-tolerated when given at doses up to 50 mg/kg for 9 months to Beagle dogs. Treatment was associated with mild gastrointestinal signs and with mild and reversible decreases in serum total protein and albumin over time, with incidence increasing as dose increased.

Even with the high dose and long duration of this study, there were no treatment effects on liver or kidney function, or gross or histopathological findings of the liver, kidney, or stomach, or on coagulation parameters. The only histopathology of note was a mild mucosal regeneration of the ileum seen in one dog at 50 mg/kg.

The relative lack of toxic effects with grapiprant compared to those seen with non-steroidal anti-inflammatory drugs working via the inhibition of the cyclooxygenase enzymes is not surprising. Grapiprant, which shows selective antagonism of the EP4 receptor, does not interfere with the production of prostanooids, and therefore will not affect the other prostanooid receptor pathways, as occurs with cyclooxygenase inhibitor drugs.

The grapiprant doses tested in this study are more than 10X the anticipated 2 mg/kg dose for dogs with osteoarthritis. A pharmacokinetic study to determine the relative bioavailability of the final formulation (flavored tablets) compared to the methylcellulose suspension administered in this study is underway.

**References**