Category E
10/2018-9/2019 Fiscal Year

Species: Guinea Pig
Studies: MRQ011-PH14 **Number of Animals: 36** **Max Duration: 98 days**

Osteoarthritis (OA)/inflammatory pain

Procedure Summary:

OA pain state that will be treated with the proprietary compound. The use of a negative control group, which will be given only saline, is necessary to determine how well the pain state was induced and how well the test article ameliorates osteoarthritis pain. However, anesthetics (isoflurane) will be used for the surgical procedure for all animals.

Explanation for use of Guinea pigs:

The test article is intended to treat canines and felines and, as such, needs to be tested in animals prior to approval. Guinea pigs with MIA-induced OA are useful studying pain behavior in response to OA. Furthermore, Guinea pigs are relatively easy to handle and have sufficient joint size to evaluate biomarkers. In addition, the Guinea pig has been the most widely used model to evaluate inflammatory biomarkers related with OA. In light of the multifaceted general reasons to use Guinea pig model, it is hoped that this study can establish the model as suitable in particular for assessing the effects of test articles on the pain associated with bearing weight on a weight-bearing joint afflicted with OA. Additionally, the test article has a significantly greater affinity to guinea pig nerve growth factor than to rat nerve growth factor and, as such, testing in rats would not be valid. Finally, due to genetic variations in the targeted protein, the protein homology between Guinea pigs and canine/felines is more closely matched than in rats or mice.

Explanation for why analgesics cannot be used:

Analgesics were not used for two reasons: First, treatment of pain can interfere with the development of the pathology and resultant pain-related behaviors (which would result in those animals not meeting study requirements and being euthanized), and second, these animal models of pain are designed to test novel analgesic agents and as such, require drug-naïve animals.

Category E
10/2018-9/2019 Fiscal Year

Species: Canine
Studies: MRQ008-PH40 **Number of Animals**: 4 **Max Duration**: 14 days/dosing

Inflammatory pain

Procedure Summary:
Beagle dogs underwent unilateral injections of kaolin suspension in order to create a model of inflammatory pain. Lameness and paw edema were assessed ~1 day prior to kaolin injection, and ~3 hours, and 1, 2, 3, 4, 5, 8, 10, 14, 17, 21, 24, and up to 35 days after kaolin injection. Lameness lasted ~14 days following kaolin injection. After minimally a 35 day monitoring period, the dogs were given an additional 14 day washout period, then redosed. Two animals were dosed 5 separate times and two animals dosed twice.

Explanation for use of Dogs:
Dogs are a defined model for the assessment of inflammatory pain and can also serve as a target species for future test article evaluation (that is, dogs may be used as the subjects in this research with the ultimate goal being the development of therapies to treat inflammatory pain in dogs). Further, dogs offer a different dimension to the study of pain and pain therapeutics. While a majority of pain research can be and is done in rodents, rodents notoriously do not show robust affective signs of pain (such as pain-suppressed behaviors), which are key facets of pain in humans. As such, for many later stage investigational pain therapies, assessments in dog models of pain prove to be the most highly translatable preclinical models.

Explanation for why analgesics cannot be used:
The purpose of this study was to determine what pain behaviors are present and over what time course following kaolin injections, as such analgesics could not be administered without compromising the study endpoints. The inflammatory pain state lasts from ~1 to ~2 weeks post-injection. In order to understand the extent and time course of the development of any pain/distress and lameness that develops over the course of this study, animals were assessed frequently by trained staff to determine any changes in lameness and inflammation. This provided a frequent and thorough assessment of the animals' welfare during the course of the study.

The kaolin induced inflammation is expected to resolve over time and so the animals enrolled on this study are expected to return to a baseline state after a period of time. As a result, the animals do not need to be euthanized and can be used again after resolution of the inflammatory pain.

Category E
10/2018-9/2019 Fiscal Year

Species: Rabbits
Studies: APS414-IR99 **Number of Animals**: 6 **Max Duration**: 3 days

Skin Irritation Positive Control

Procedure Summary:
Four abrasions (about 2.5 cm X 2.5 cm) in a # shape in the keratinous layer (avoiding injury to the dermis) in the 2 caudal quadrants were made on each rabbits back. Formaldehyde (positive control) or saline (negative control) was applied directly to the skin via a 2.5 cm X 2.5 cm non-occlusive dressing (e.g. sterile gauze) over intact and abraded skin. The application site patches will be secured by wrapping the animals with a semi-occlusive bandage for 24 ± 0.5 hours. After wrapping was removed, the animal was monitored and irritation severity scored for 48 hours.

Explanation for use of Rabbits:
Irritation has been studied extensively in rabbits, and is an approved model for irritation testing among the regulatory bodies. Rabbits are widely used for testing irritation potential of test materials. Rabbits and humans have similarities in their immune system, particularly their skin reactivity that make them a good screening model for compounds that are intended to come in contact with humans. Rabbits are one of the preferred animal species for irritation testing due to their ability to readily display irritation responses.

Explanation for why analgesics cannot be used:
This study is designed to test the irritation potential of medical devices. The use of anesthetics, analgesics, or tranquilizers to alleviate pain is prohibited due to the potential molecular interaction between the drugs and the compounds associated with the medical device. This interaction may cause a response (inhibitory or synergistic) that could affect the outcome of the study. In order to effectively evaluate the characteristics of the device, the use of medications is contraindicated.

Federal Regulatory agency requiring procedure:
Japanese Ministry of Health, Labour, and Welfare (JMHLW) PFSB/ELD/OMDE Notification No. 0301-20 (2012): Part 5 Test for irritation.