Local and Systemic Effects Following Intraarticular Injection of MM-II Liposomes. Results from Two Nonclinical Toxicity Studies in Rabbits and Dogs

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INTRODUCTION

MM-II is a suspension of large multilamellar liposomes composed of two phospholipids, DMPC and DPPC for intra-articular administration, being developed for its potential to slow down OA disease progression and reduce osteoarthritic pain. Due to their unique characteristics, these liposomes are retained on the cartilage surface, providing long-term lubrication and leading to a reduction in wear of the cartilage.

OBJECTIVE

The purpose of these preclinical studies was to evaluate the toxicity of MM-II when administered via intra-articular (IA) injection into the stifle joint of rabbits and dogs.

METHODS

In two GLP compliant studies, NZW rabbits or Beagle dogs were administered an IA injection of either placebo, saline or low (~20 mg/kg), middle (~40 mg/kg) or high (~60 mg/kg) dose of MM-II. Due to the fixed concentration of MM-II, 3 injections, each of the maximum tolerable volume per injection for each animal species, were injected over a period of a week in order to enable a high exposure of MM-II and provide maximal safety margins for the proposed human dose. Animals were sacrificed either 4 days (interim sacrifice), 14 weeks (terminal sacrifice), or 18 weeks (recovery period) following the last of the 3 injections. Assessment of toxicity was based on mortality, clinical observations, body weight change, food consumption, ophthalmic observations, electrocardiographic (ECG) measurements (dogs only), and clinical and anatomic pathology. As part of this study, blood samples were collected for toxicokinetic (TK) evaluation and synovial fluid samples were collected for clinical pathology evaluations.

RESULTS

Rabbit study

Clinical observation: No MM-II-related clinical observations were noted, and no MM-II-related veterinary treatments were administered. Clinical observations were observed with comparable incidences as in the control groups.

Body Weights: By the end of the dosing phase, body weights relative to both control groups were increased for females at all MM-II dose levels. The increase in body weights was of moderate magnitude and did not result in any other clinical effects and was therefore considered non adverse.

Food Consumption: No MM-II-related effects on food consumption occurred.

Clinical Laboratory Evaluations: No MM-II-related effects on hematology, coagulation, urinalysis, synovial fluid analysis or clinical chemistry test results were identified.

Macroscopic Observations: No MM-II- or procedure-related macroscopic findings were noted.

RESULTS (cont.)

Microscopic Observations: At the Interim Sacrifice timepoint, the primary microscopic findings were hyperplasia of the synovium and/or a mixed inflammatory cell infiltration and occasional inflammatory exudate in the joint space. Synovial hyperplasia was characterized as minimal to slight in severity. Findings were generally more severe in animals that have received MM-II, with similar severities between the different doses. Additionally, in the right stifle joint, fibrosis of the synovium and exudate in the joint space were noted; however, these findings were observed at a similar incidence and severity in animals from all groups and, therefore, were attributed to the injection procedure and not to MM-II. Incidence of severity for terminal sacrifice time point is presented in Figure 1.

Toxicokinetics: After MM-II administration, DMPC exposure, as assessed by mean C_{max} and AUC_{0-72}, was higher than its endogenous baseline level in the placebo and saline control groups. After reaching C_{max}, DMPC concentrations declined largely, through 144 or 552 hours post dosing. DPPC exposure was relatively similar to its endogenous baseline level and to their levels in the placebo and saline control groups.

	MM-II									
Sex	Males					Females				
Group	1	2	3	4	5	1	2	3	4	5
Joint, Stifle, Left										
Number Examined	4	4	4	4	4	4	4	4	4	4
Hyperplasia, synovium										
Minimal	0	0	0	0	0	0	0	1	0	0
Joint, Stifle, Right										
Number Examined	4	4	4	4	4	4	4	4	4	4
Hyperplasia, synovium										
Minimal	1	2	1	0	1	1	0	3	0	1
Inflammation, mixed cell										
Minimal	0	0	0	0	0	1	0	2	0	0

Figure 1. Incidence and Severity of MM-II or Procedure Related Microscopic Findings at Terminal Sacrifice

Dog study

Clinical observation: Limited use of the injected hind limb was observed in several animals post dosing. This observation was transient and considered procedure-related as it was noted for both control and MM-II-administered animals.

Ophthalmic Examinations: No visible lesions were noted during ophthalmic examinations.

Body Weights: No MM-II-related effects occurred on body weights.

Food Consumption: No MM-II-related effects on food consumption occurred.

Electrocardiographic Examinations: Borderline first degree and second-degree atrioventricular block were observed in one saline control and one high dose animals. These were considered incidental and not attributed to MM-II.

Clinical Laboratory Evaluations: No MM-II-related effects on hematology, coagulation, urinalysis, synovial fluid analysis or clinical chemistry test results were identified.

RESULTS (cont.)

Macroscopic Observations: No MM-II or procedure-related macroscopic findings were noted.

Microscopic Observations: All microscopic findings were considered spontaneous and/or incidental because they occurred at a low incidence, were randomly distributed across groups (including concurrent controls), and/or their severity were as expected for Beagle dogs of this age; therefore, they were considered not test article related. Incidence of severity for terminal sacrifice time point is presented in Figure 2.

	MM-II									
Sex	Males					Females				
Dose Group	1	2	3	4	5	1	2	3	4	5
Number Examined	4	4	4	4	4	4	4	3	4	4
Joint, Stifle, Left										
Hyperplasia, synovium										
Minimal	1	1	1	0	3	0	1	1	0	2
Slight	0	0	0	0	0	0	1	0	0	0
Joint, Stifle, Right										
Hyperplasia, synovium										
Minimal	2	1	2	1	3	1	1	0	0	2
Slight	0	0	0	0	0	0	1	0	0	0
Inflammation, mixed cell, synovium										
Minimal	0	1	0	0	1	0	0	0	0	0

Excluded Animal D0704 (Group 3 female sacrificed moribund on Day 10)

Figure 2. Incidence and Severity of MM-II or Procedure Related Microscopic Findings at Terminal Sacrifice

Toxicokinetics: After MM-II administration, DMPC exposure as assessed by mean C_{max} and AUC_{0-72} was higher than its endogenous baseline levels and the placebo and saline control groups. After reaching C_{max}, mean DMPC concentrations declined largely through 312 or 552 hours post dosing. DPPC exposure was relatively similar to its endogenous baseline level and to their levels in the placebo and saline control groups.

CONCLUSIONS

Two non-clinical toxicology studies in rabbits and dogs were conducted for the MM-II liposome suspension. In both studies, MM-II-related findings were considered non-adverse at all dose levels. The NOAEL was determined as 59 mg/kg in the dog study and 66 mg/kg in the rabbit study. Toxicokinetic findings in both studies indicated that DMPC exposure appears to be impacted by MM-II administration, but DPPC is not. The highest doses employed in these studies provide a maximal local safety margin of 12-fold for dogs and 23-fold for MM-II in rabbits of the highest dose being studied in an ongoing phase 2b clinical trial. To conclude, MM-II was found safe with no adverse events and with a NOAEL of the highest dose employed in the study.

CONTACT

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