Toxicity Study of Recombinant Human Platelet-Derived Growth Factor-BB Hydrated Bovine Type I Collagen in Decorated Femur Model in Rabbits---a Safety Study Model for Rotator Cuff Repair

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Statement of Purpose: A previous ovine model demonstrated that a bovine type I collagen matrix hydrated with recombinant human platelet-derived growth factor-BB (rhPDGF-BB) significantly enhanced rotator cuff repair in a dose response study[1]. The purpose of this study was to assess the systemic and local toxicity of the same bovine type I collagen matrix hydrated with rhPDGF-BB. However in order to assess the safety of this device, taking into account anatomical concerns of smaller animals, a decorticated femur model in rabbits was designed to determine the safety of the device for rotator cuff repair.

Methods: 48 rabbits, 24 males and 24 females, weighing 3-4 kg, were randomized into 4 groups of 6 males and 6 females for 1 and 6 week time points. After exposing the femur through a lateral incision along the axis of the femur, a 1.6 cm "footprint" area was marked on the femur using the third trochanter as an anatomical reference. The periosteum within the marked area was then removed and a superficial decortication was performed with irrigation. Two holes (about 1.2~1.6 mm dia.) were drilled at the center of the decorticated area (Figure 1a). The surgery was performed bilaterally.

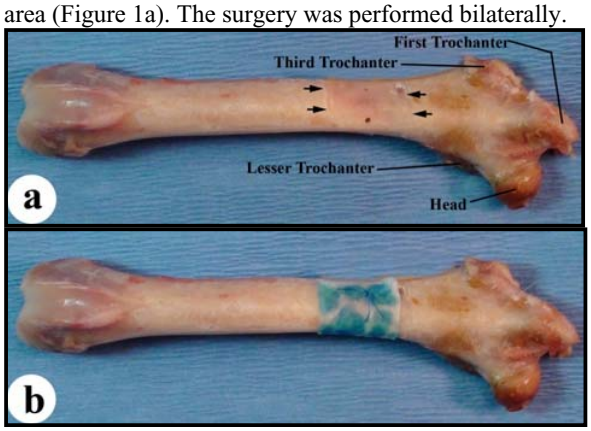

Figure 1. Scheme of placement and stabilization of test materials

Group 1 served as a sham control where a 5-0 Prolene suture was threaded through the two holes and tied on the top of femur. Group 2 was implanted with the type I collagen matrix hydrated with sodium acetate buffer (Reference Control) wrapped around the decorticated "footprint" and stabilized with the Prolene suture through the two holes on the femur (Figure 1b). Groups 3, and 4 were implanted with the type I collagen matrix hydrated with approximately 100 and 1000 µg rhPDGF-BB, respectively. The incision was closed in layers. The rhPDGF-BB was provided by BioMimetic Therapeutics, Inc., Franklin, TN and the bovine type I collagen matrix was provided by Integra, Plainsboro, NJ. Three males and three females from each group were euthanized after 1-week and 6-week implantation. Toxicological endpoints included: health observations, body weights, hematology, coagulation, clinical chemistry, organ weights, and gross and microscopic pathology. The implantation sites and selected systemic tissues were microscopically evaluated based on two main categories: Inflammation and Tissue Response. The scale was set up as below:

0 = Absent; 1 = Minimal/Slight; 2 = Mild; 3 = Moderate; 4 = Marked/Severe

Results: There were some statistical differences observed in body weight and parameters of clinical pathology between the test and control groups, however these were attributed to individual variability. There were no test article related toxicologically significant findings for body weights, hematology, coagulation, clinical chemistry, and organ weights. No specific adverse histopathological systemic findings were identified across dose and time groups. The implant sites were scored microscopically based upon two categories as shown in Table 1.

Table 1. Histopathological scoring of implant sites

Time Points	Categories of Scoring	Sham	Reference Control	Test Article (Dose 1)	Test Article (Dose 2)
1 W	Inflammation	3.5 ± 1.0	3.7 ± 1.0	3.8 ± 0.8	4.5 ± 0.5
	Tissue Response	3.1 ± 1.2	3.1 ± 1.3	3.8 ± 1.2	4.3 ± 1.3
6 W	Inflammation	7.2 ± 1.9	9.9 ± 3.6	9.4 ± 2.4	10.0 ± 2.1
	Tissue Response	7.9 ± 2.0	4.9 ± 1.6	9.4 ± 2.2	9.0 ± 2.0

Kruskal-Wallis One Way ANOVA on Ranks was used to compare across the groups at each time point and each category. No statistically significant different was found on both Test Articles after comparing with Reference control at both time points on inflammation. No statistically significant difference was found between Test Articles and controls at 1 week on tissue response, but tissue response scores from both Test Articles were statistically higher than Reference control at 6 week due to the trend of myofiber, fibrocartilage, and bone regeneration, which are the expected mechanism of action for rhPDBF-BB. The type I collagen matrix was partially degraded at 1 week and not detected at 6 weeks by histology.

Conclusions: The attachment site of the supraspinatus tendon and decorticated humeral head is the site of healing for rotator cuff repair. Based on FDA guidelines for local safety, a decorticated femur model in rabbits was utilized to approximate the delivery of the type I collagen bovine matrix hydrated with rhPDGF-BB. Under the conditions of this study, there were no specific adverse systemic, local histopathology or toxicity observations attributed to the type I collagen matrix hydrated with rhPDGF-BB following 1 and 6 weeks post-bilateral implantation. .

References: 1. Hee CK., 56th Annual Meeting of the Orthopaedic Research Society, Poster No. 1172