Bone & Joint Research

Supplementary Material

10.1302/2046-3758.117.BJR-2021-0358.R2

In vitro drug release and sustainability analysis

To evaluate the rate of the recombinant secretory leucocyte protease inhibitor (rSLPI) release from the fibrin gel comprising 100 µL fibrinogen and 1.0 µg rSLPI, we incubated each gel in 1.0 mL phosphate-buffered saline (PBS) supplemented with 10% fetal bovine serum at 37°C with continuous rotation using a microhybridization incubator for 14 days. The PBS solution was replaced and collected daily. These wash solutions were then subjected to enzyme-linked immunoabsorbent assay (ELISA) for rSLPI (Human SLPI ELISA Kit; J&I Biological, JL13242, China), and these release experiments were carried out three times. The cumulative percentages are presented in Figure a and show that 26.4% (standard deviation (SD) 2.6%) of the total rSLPI in the fibrin gel was released into the PBS-10% fetal bovine serum solution at 24 hours, and that more than 50% of the total rSLPI was released into the wash solution by 72 hours. On the 14th day, 98.1% (SD 4.4%) of the total rSLPI had been released from these gels.

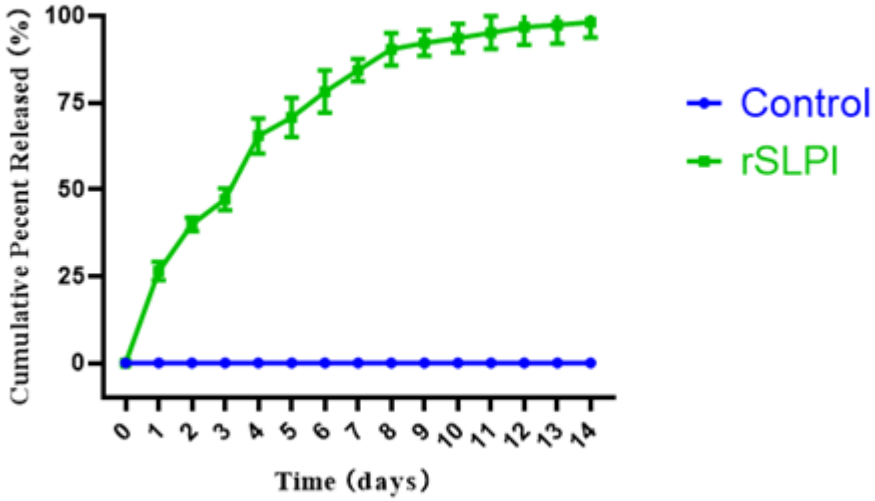

Fig a. Sustained release curve of recombinant secretory leucocyte protease inhibitor (rSLPI).

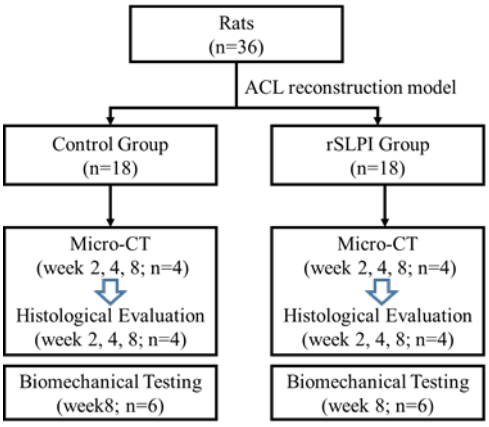

Fig b. The roadmap for this study. ACL, anterior cruciate ligament; rSLPI, recombinant secretory leucocyte proteinase inhibitor.

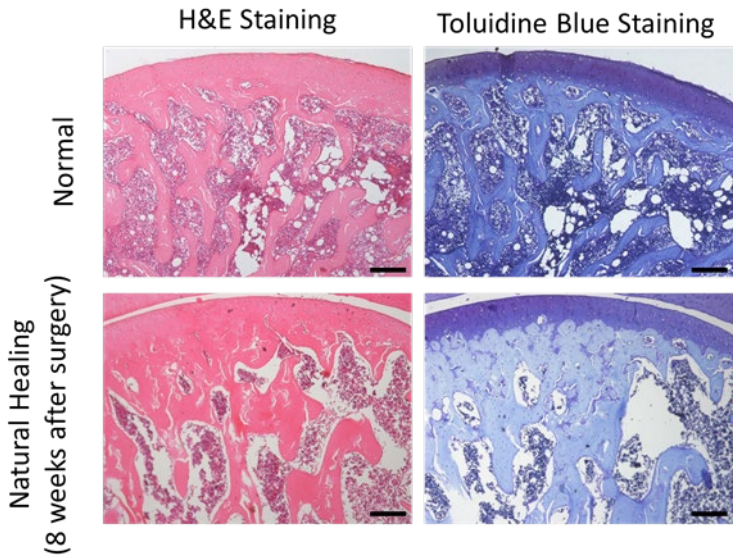

Fig c. Histological staining of specimens from the natural healing group in other studies did not suggest significant osteoarthritis. Bar: 200μm. H&E, haematoxylin and eosin.

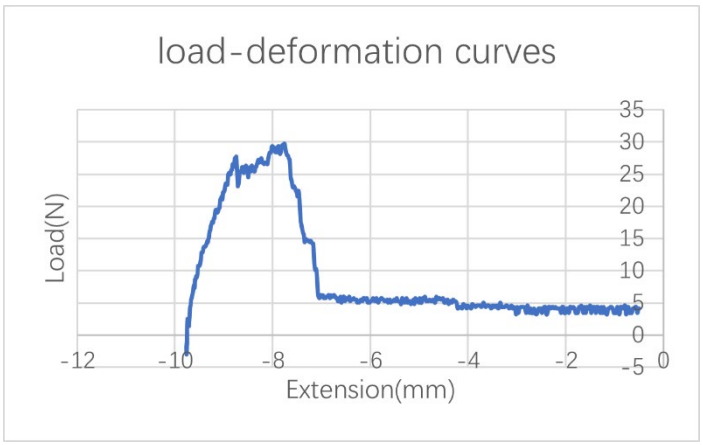

Fig d. The load elongation curve from biomechanical experiment.

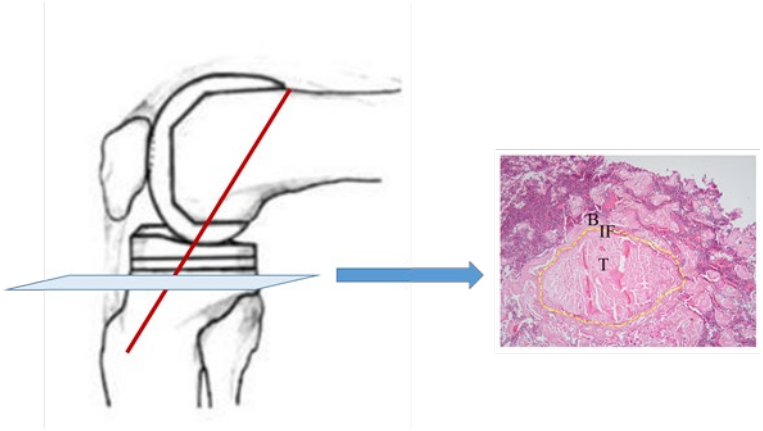

Fig e. Histological section orientation and haematoxylin and eosin-stained section at low magnification (4×). B, bone; IF, interface; T, tendon.

NOTE: Please save this file locally before filling in the table, DO NOT work on the file within your internet browser as changes will not be saved. Adobe Acrobat Reader (available free here) is recommended for completion.

The ARRIVE guidelines 2.0: author checklist

The ARRIVE Essential 10

These items are the basic minimum to include in a manuscript. Without this information, readers and reviewers cannot assess the reliability of the findings.

Item		Recommendation	Section/line number, or reason for not reporting
Study design	1	For each experiment, provide brief details of study design including: a. The groups being compared, including control groups. If no control group has been used, the rationale should be stated. b. The experimental unit (e.g. a single animal, litter, or cage of animals).	Methods: animal study design and surgical procedure Methods: animal study design and surgical procedure
Sample size	2	a. Specify the exact number of experimental units allocated to each group, and the total number in each experiment. Also indicate the total number of animals used. b. Explain how the sample size was decided. Provide details of any *a priori* sample size calculation, if done.	Methods: animal study design and surgical procedure
Inclusion and exclusion criteria	3	a. Describe any criteria used for including and excluding animals (or experimental units) during the experiment, and data points during the analysis. Specify if these criteria were established *a priori.* If no criteria were set, state this explicitly. b. For each experimental group, report any animals, experimental units or data points not included in the analysis and explain why. If there were no exclusions, state so. c. For each analysis, report the exact value of *n* in each experimental group.	Methods: animal study design and surgical procedure Methods: animal study design and surgical procedure Methods: Specimen Collection and Preparation
Randomisation	4	a. State whether randomisation was used to allocate experimental units to control and treatment groups. If done, provide the method used to generate the randomisation sequence. b. Describe the strategy used to minimise potential confounders such as the order of treatments and measurements, or animal/cage location. If confounders were not controlled, state this explicitly.	Methods: animal study design and surgical procedure
Blinding	5	Describe who was aware of the group allocation at the different stages of the experiment (during the allocation, the conduct of the experiment, the outcome assessment, and the data analysis).	Methods: Histological Evaluation
Outcome measures	6	a. Clearly define all outcome measures assessed (e.g. cell death, molecular markers, or behavioural changes). b. For hypothesis-testing studies, specify the primary outcome measure, i.e. the outcome measure that was used to determine the sample size.	Methods: Specimen Collection and Preparation; Biomechanical Testing
Statistical methods	7	a. Provide details of the statistical methods used for each analysis, including software used. b. Describe any methods used to assess whether the data met the assumptions of the statistical approach, and what was done if the assumptions were not met.	Methods: Statistical Analysis Methods: Statistical Analysis
Experimental animals	8	a. Provide species-appropriate details of the animals used, including species, strain and substrain, sex, age or developmental stage, and, if relevant, weight. b. Provide further relevant information on the provenance of animals, health/immune status, genetic modification status, genotype, and any previous procedures.	Methods: animal study design and surgical procedure Methods: animal study design and surgical procedure
Experimental procedures	9	For each experimental group, including controls, describe the procedures in enough detail to allow others to replicate them, including: a. What was done, how it was done and what was used. b. When and how often. c. Where (including detail of any acclimatisation periods). d. Why (provide rationale for procedures).	Methods: animal study design and surgical procedure Methods: animal study design and surgical procedure Methods: animal study design and surgical procedure Methods: animal study design and surgical procedure
Results	10	For each experiment conducted, including independent replications, report: a. Summary/descriptive statistics for each experimental group, with a measure of variability where applicable (e.g. mean and SD, or median and range). b. If applicable, the effect size with a confidence interval.	Results: Biomechanical testing