Systemic Administration of HMGB1 Peptide Promotes Meniscal Healing via Mobilization of PDGFRα+ Mesenchymal Cells

Kyota Ishibashi[1, 2], Takashi Shimbo[2], Tomomi Kitayama [2], Eiji Sasaki[1], Takahiro Tsushima[1], Yasuyuki Ishibashi[1], Katsuto Tamai[2]
[1]Hirosaki university, Aomori, Japan., [2]Osaka university, Osaka, Japan.
kyota.i@hirosaki-u.ac.jp

Disclosures: Kyota Ishibashi (N), Takashi Shimbo (Stemrim), Tomomi Kitayama (N), Eiji Sasaki (N), Takahiro Tsushima (N), Yasuyuki Ishibashi (N), Katsuto Tamai (Stemrim)

INTRODUCTION: Meniscal injuries often result in poor intrinsic healing capacity due to the limited vascularity of the meniscus. High mobility group box 1 (HMGB1), a nuclear protein released during tissue injury, promotes recruitment of mesenchymal stem cells (MSCs) from bone marrow to sites of damage [1]. We hypothesized that systemic administration of HMGB1-derived peptide (HMGB1-peptide) accelerates meniscal healing by mobilizing Platelet-Derived Growth Factor Receptor α (PDGFRα)+ MSCs.

METHODS: Large medial meniscectomy model in mice was established (**Fig1 a**). HMGB-peptide (5 mg/kg) or saline was administered intravenously for five consecutive days postoperatively (n=20 per group). Meniscal regeneration was assessed at 1, 2, 4, and 6 weeks using gross morphology and histology (Safranin-O staining, Modified Pauli's score). Contribution of bone marrow–derived cells was evaluated using a parabiosis model of PDGFRα-Cre; ROSA-tdTomato and wild-type mice(**Fig2 a**). Involvement of the Stromal Cell-Derived Factor-1 / C-X-C Chemokine Receptor Type 4 (SDF-1/CXCR4) axis was examined by immunostaining and pharmacologic blockade with a CXCR4 antagonist. Single-cell RNA sequencing was performed to assess quality of regenerated tissue.

RESULTS: Mice in HMGB1-peptide group exhibited significantly improved meniscal regeneration compared to controls at 1, 2, 4 and 6 weeks (Modified Pauli scores: HMGB1 group 7.2±1.1 vs control 4.9±1.3 at 6 weeks, $p<0.01$) (**Fig1 b,c**). In the parabiosis model, tdTomato+ cells accounted for ~15% of cells in the regenerated meniscus with HMGB1-peptide, compared to ~1% with saline ($p<0.01$) (**Fig2 b,c**). Immunostaining revealed that SDF-1 was predominantly expressed around vascular endothelial cells adjacent to the injured meniscus. CXCR4 blockade reduced recruitment to ~2%, confirming involvement of SDF-1/CXCR4 signaling ($p<0.01$)(**Fig2 d,e**). Single-cell transcriptomics demonstrated enrichment of collagen and glycosaminoglycan-related pathways in the HMGB1-peptide group, indicating superior matrix composition (**Fig3**).

DISCUSSION: Systemic HMGB1-peptide administration enhances meniscal repair by mobilizing PDGFRα+ bone marrow–derived MSCs through the SDF-1/CXCR4 axis, resulting in improved structural regeneration.

SIGNIFICANCE/CLINICAL RELEVANCE: This novel systemic regenerative therapy offers a promising adjunct to meniscal repair surgery, potentially improving healing outcomes and delaying osteoarthritis progression.

REFERENCES: [1] Tamai K, et al. PDGFRα-positive cells in bone marrow are mobilized by high mobility group box 1 (HMGB1) to regenerate injured epithelia. Proc Natl Acad Sci U S A. 2011 Apr 19;108(16):6609-14. doi: 10.1073/pnas.1016753108.

Figure 1

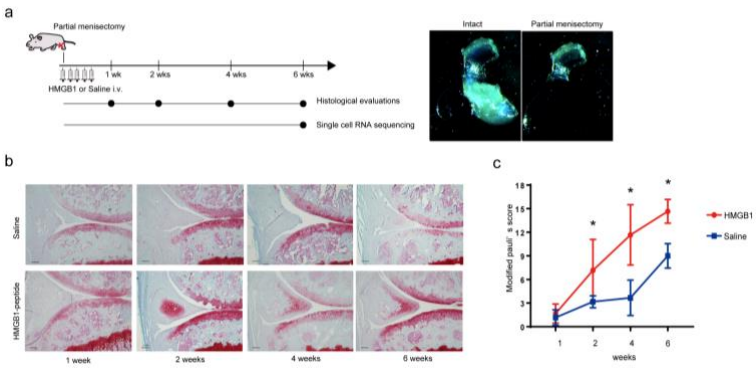


Figure 2

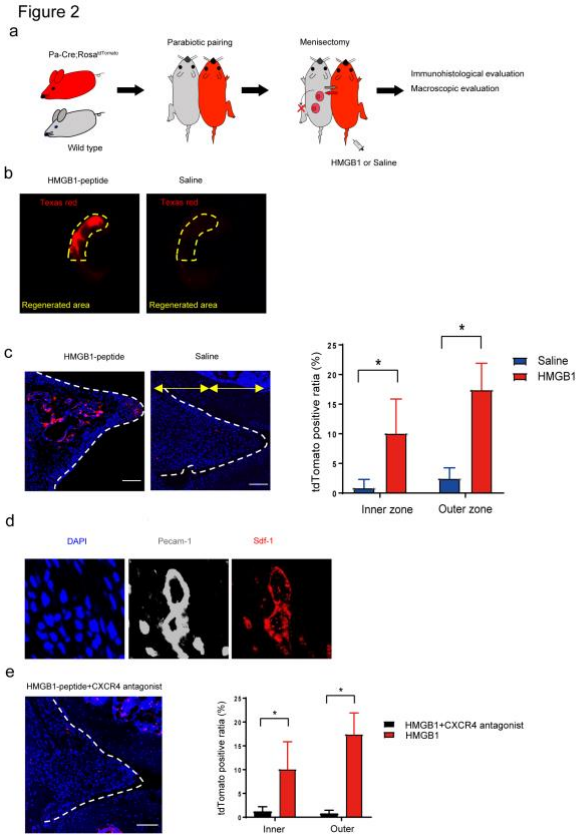
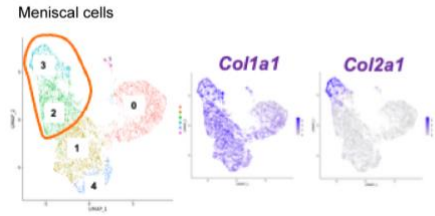


Figure 3



GO analysis enriched in HMGB1-peptide

	Term
1	Extracellular matrix organization
2	Collagen fibril organization
3	Protein complex subunit organization
4	Skeletal system development
5	Response to unfolded protein
6	Collagen metabolic process
7	Cellular response to unfolded protein