

Gradient mechanical microenvironment promotes interface regeneration by regulating Ca²⁺ channel

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INTRODUCTION: The natural tendon-bone interface achieves stress dispersion through a gradient arrangement of collagen fibers and gradual mineralization². However, after surgical repair, tissue homogenization leads to stress concentration, causing re-rupture or impaired regeneration³, which remains a major clinical challenge. This study aims to develop an implantable magnetically responsive gradient piezoelectric hydrogel system that mimics the structure and mechanical properties of the natural interface. By synergistically regulating multiple physical fields, it seeks to achieve stress dispersion and structural regeneration at the tendon-bone interface.

METHODS: We developed a magnetically responsive composite hydrogel system by integrating electrospinning and dynamic magnetic field regulation. The system comprises magnetically controlled short fibers (CMSFs) and methacrylated gelatin (GelMA), engineered as follows: 1. Gradient Structure Programming: CMSFs were directionally aligned using an external static magnetic field, generating a biomimetic gradient arrangement within the hydrogel; 2. Multi-Field Coupling Design: Piezoelectric BaTiO₃ nanoparticles (NPs) were incorporated to enable dynamic magnetic field-induced mechanical vibrations, achieving synergistic "magnetic-force-electric" multiphysics coupling. The system was implanted in a rat Achilles tendon injury model (n = 96, ethics committee-approved) (Figure 1). Regenerative outcomes were assessed at 2 and 6 weeks via Micro-CT, histology, tensile testing, and gait analysis. RNA sequencing and gene/protein expression analysis were performed to elucidate the healing-promoting mechanisms (Figure 2). Data are expressed as mean ± SD from ≥3 independent replicates. Prism 9 (v9.0.0) was used for Student's t-test and ANOVA, with p ≤ 0.05 considered significant.

RESULTS SECTION: The experimental group demonstrated significant advantages in interface regeneration: 1. Structural regeneration: The newly formed interface developed a continuous four-layer structure (tendon → fibrocartilage → calcified cartilage → bone), with collagen fiber alignment restored to over 70% of the natural state; Micro-CT 3D reconstruction showed a 12.7% increase in bone density compared to the control group (n = 5 in each group). 2. Mechanical properties: At 6 weeks, the ultimate tensile strength recovered to 79.8% of the normal value, significantly higher than the blank control group, representing a 58.5% increase over the blank control (n = 10 in each group, P = 0.0001). 3. Functional recovery: Gait analysis at 6 weeks post-surgery showed no significant changes in contact force among all groups (P > 0.05) (Figure 3), indicating that foot weight-bearing capacity was partially restored by 6 weeks (n = 5 in each group). 4. Molecular mechanism: Gradient mechanical vibration promoted stable proliferation of interface stem cells via the CalcR pathway; piezoelectric signals activated Ca²⁺ influx, facilitating differentiation into tendon, fibrocartilage, and bone lineages (n = 3 in each group).

DISCUSSION: This study achieved three major breakthroughs through an innovative magnetically responsive gradient piezoelectric hydrogel system: (1) establishing in situ mechanical regulation for implantable devices, overcoming the technical challenge of biomimetic gradient structures; (2) validating the clinical feasibility of structural interface regeneration in animal models; and (3) elucidating the molecular mechanism by which the "magnetic-force-electric" multiphysics fields regulate interface regeneration via the CalcR and Ca²⁺ signaling pathways. These findings not only provide a new paradigm for tendon-to-bone repair but also offer core principles that can be extended to the regeneration of other gradient tissues (such as periodontal ligament-to-bone and muscle-to-tendon), holding significant theoretical and practical value. Despite the significant achievements of this study, several issues require further exploration: 1. The precision control of the magnetic field; 2. Long-term biocompatibility: It is essential to extend the observation period and further validate in large animal models whether the long-term retention of Fe₃O₄ NPs affects tissue metabolism.

SIGNIFICANCE/CLINICAL RELEVANCE: (1-2 sentences): This study pioneers a magneto-electro-mechanical coupling strategy to dynamically regulate tendon-bone interface regeneration microenvironments, with potential applications in other interface tissue injuries.

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IMAGES AND TABLES:

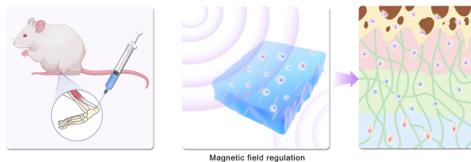


Figure 1. Achilles tendon-bone injury model.

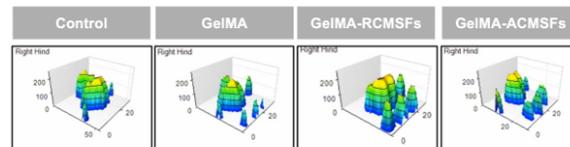


Figure 3. Gait pressure distribution diagrams of each group.

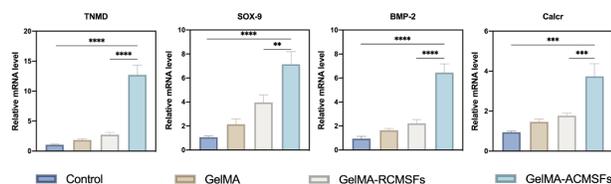


Figure 2. Relative expression of genes in the regulatory mechanism of Gel-ACMSFs.