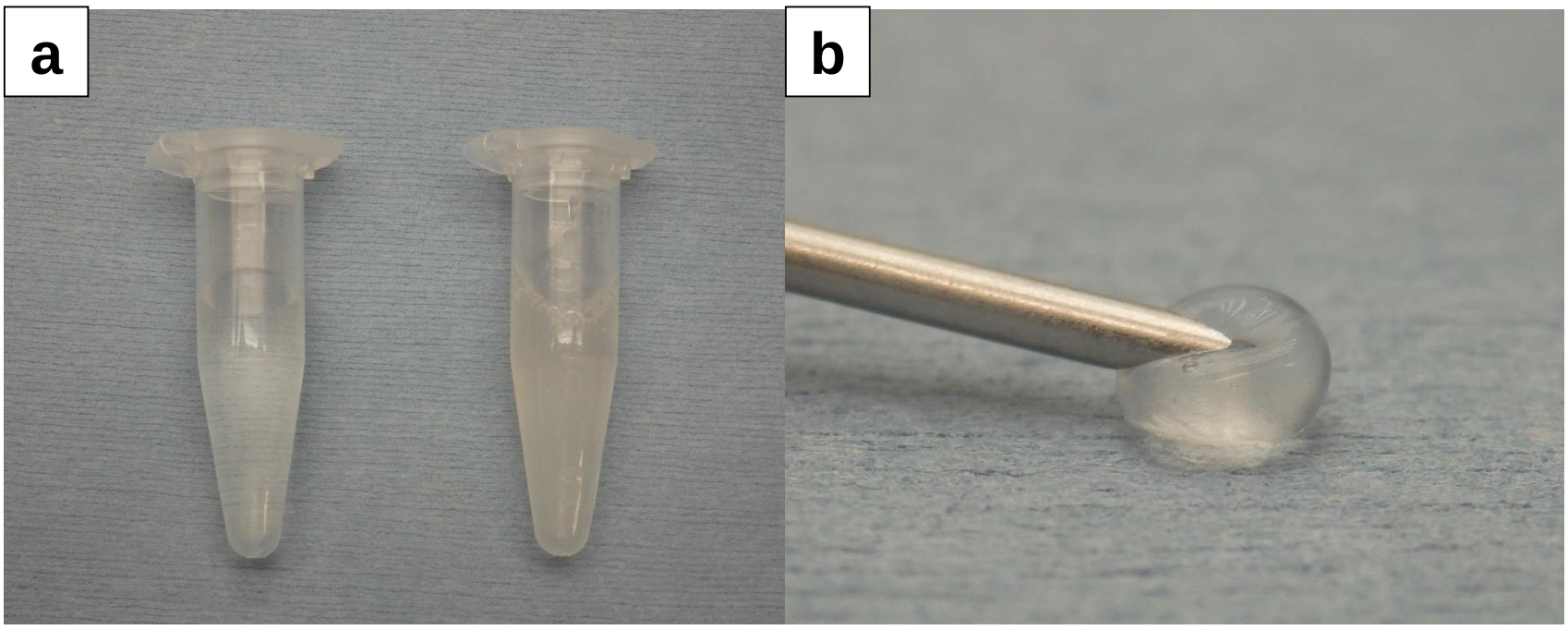
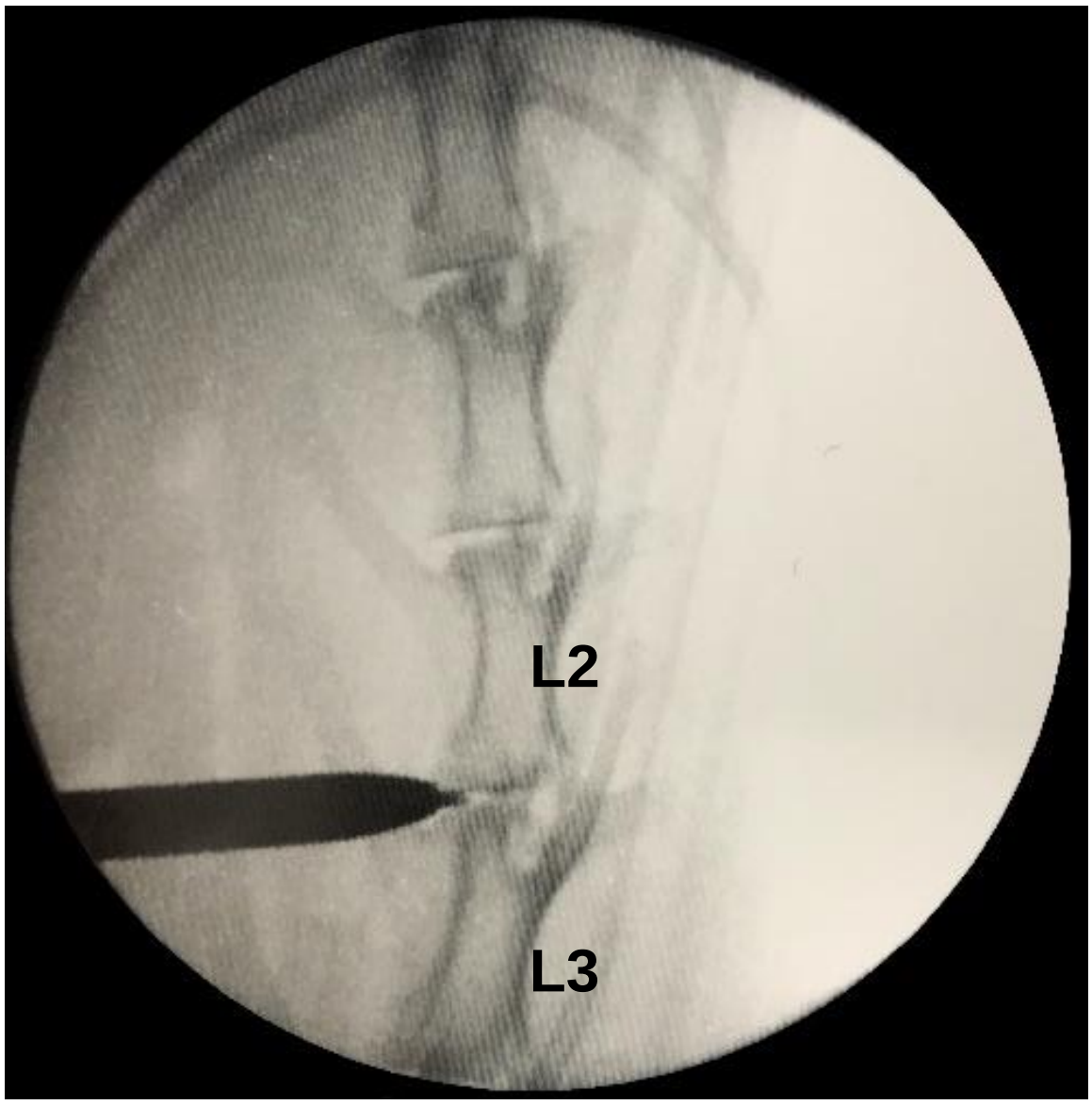
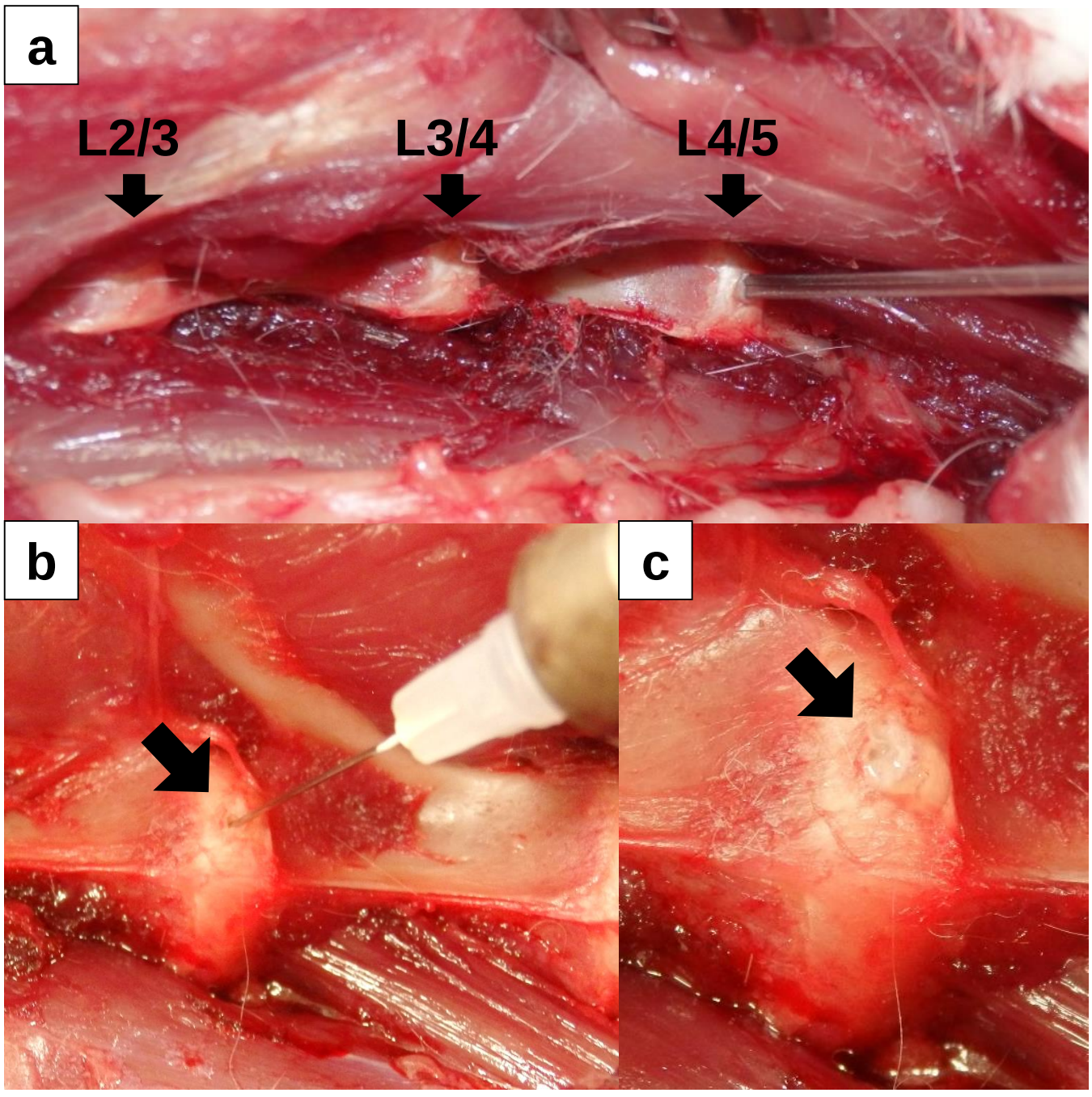


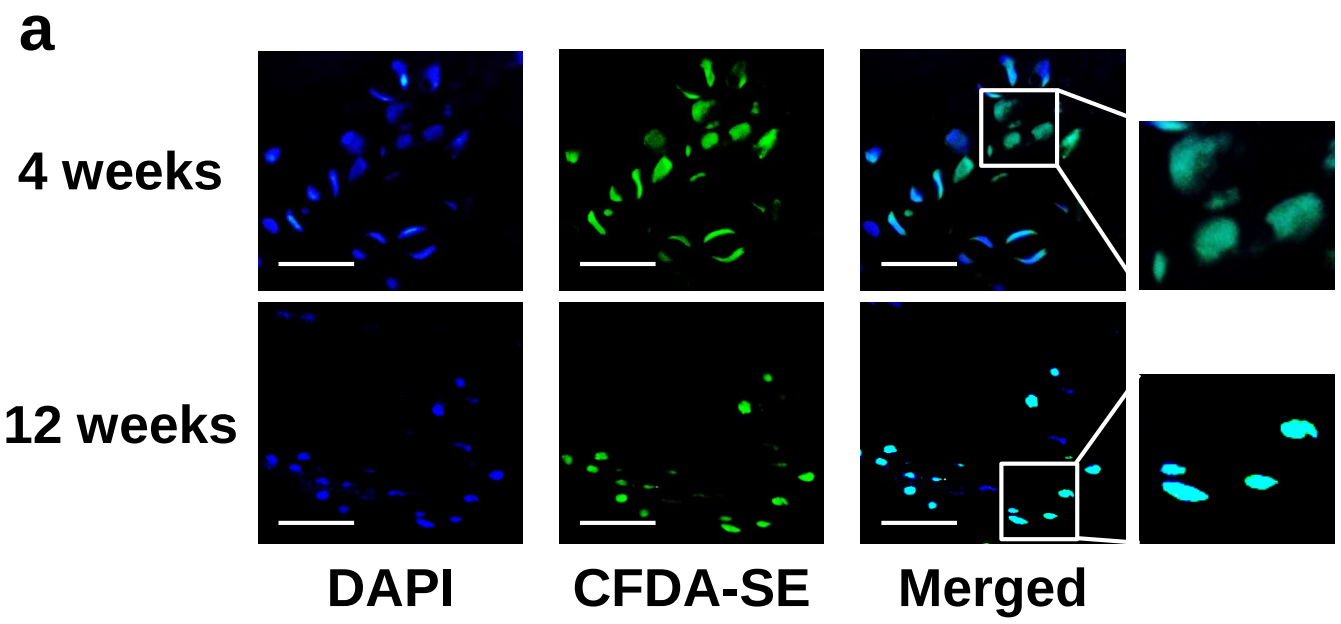
Supplementary Fig. 1. Preparation of ultra-purified alginate (UPAL) solution and gelation. (a): Preparation of 2% (w/v) UPAL solution (left side). Labeled bone-derived mesenchymal stem cells (BMSCs) and unlabeled nucleus pulposus cells (NPCs) were encapsulated in the UPAL solution (right side). (b): UPAL gel beads after pipetting into 102 mM $CaCl_2$ using a 22-gauge needle for gelation.

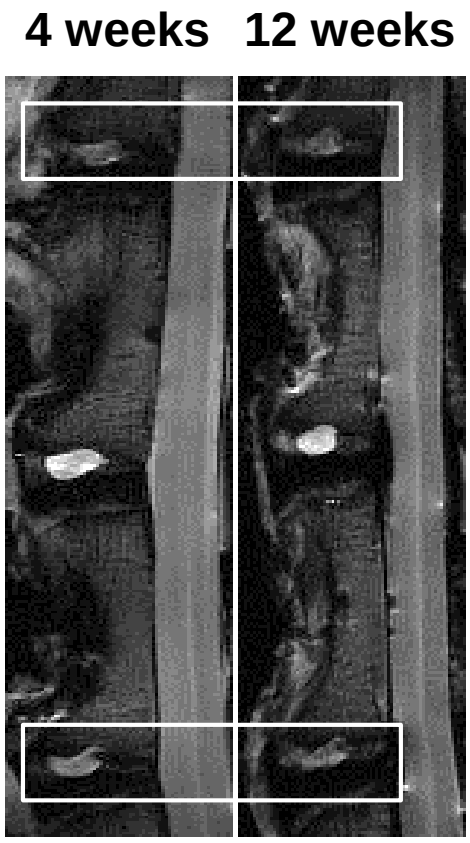


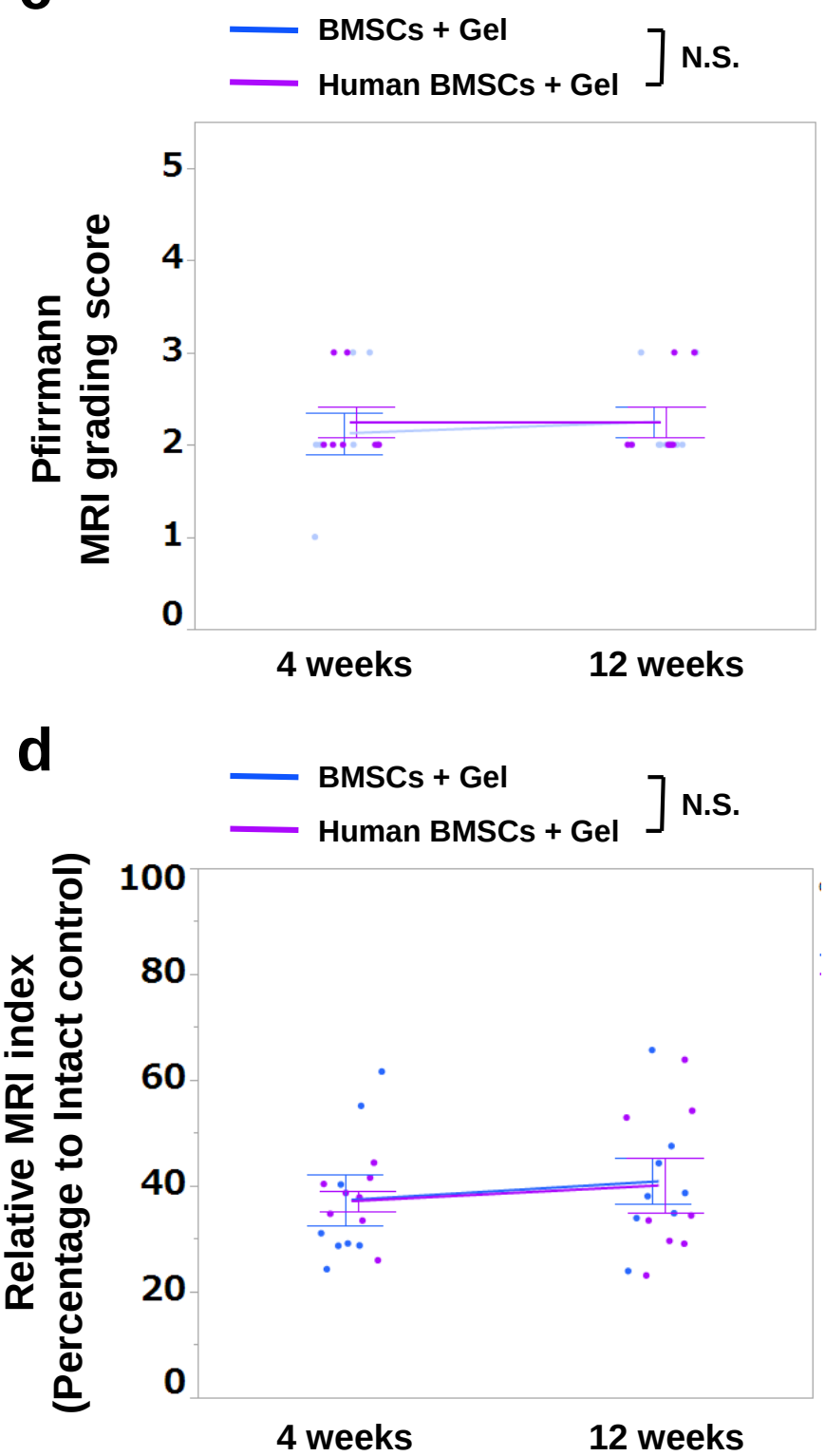
Supplementary Fig. 2. Induction of intervertebral disc (IVD) degeneration. Rabbit IVD (L2/3) was punctured with an 18-guage needle percutaneously under fluorography guidance with a small skin incision and a precut dilator (METRx; Medtronic Sofamor Danek, Memphis, TN).



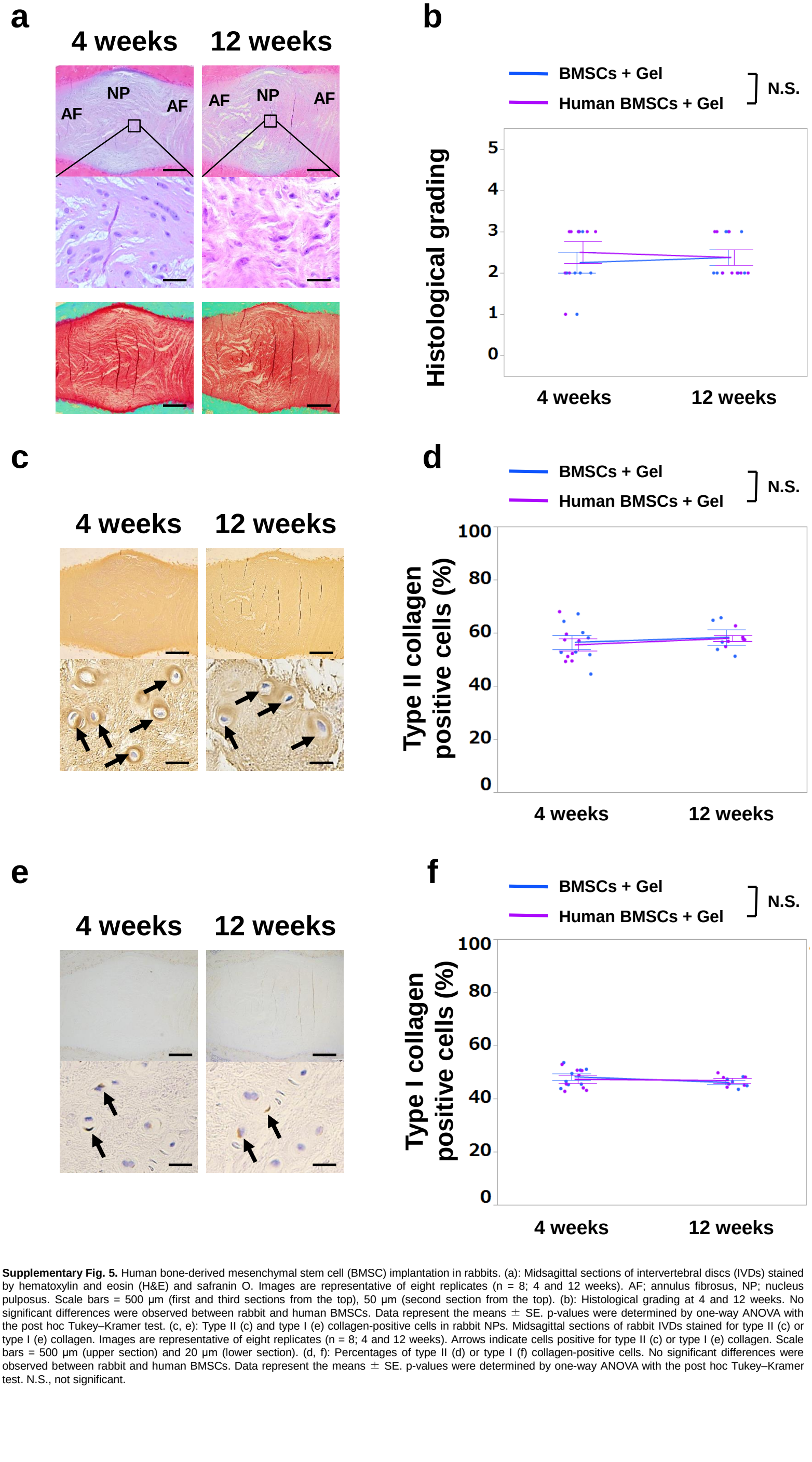
Supplementary Fig. 3. In vivo implantation in rabbit. (a): Rabbit intervertebral discs (IVDs) from L2/3 to L4/5 shown via an antero-lateral retroperitoneal approach. Removal of the degenerated nucleus pulposus (NP) tissue using a micro ear forceps after making a hole using an 18-gauge needle. (b): Ultra-purified alginate (UPAL) solution injection into the IVD defect after discectomy. (c): Gelation after injection of 102 mM CaCl2.



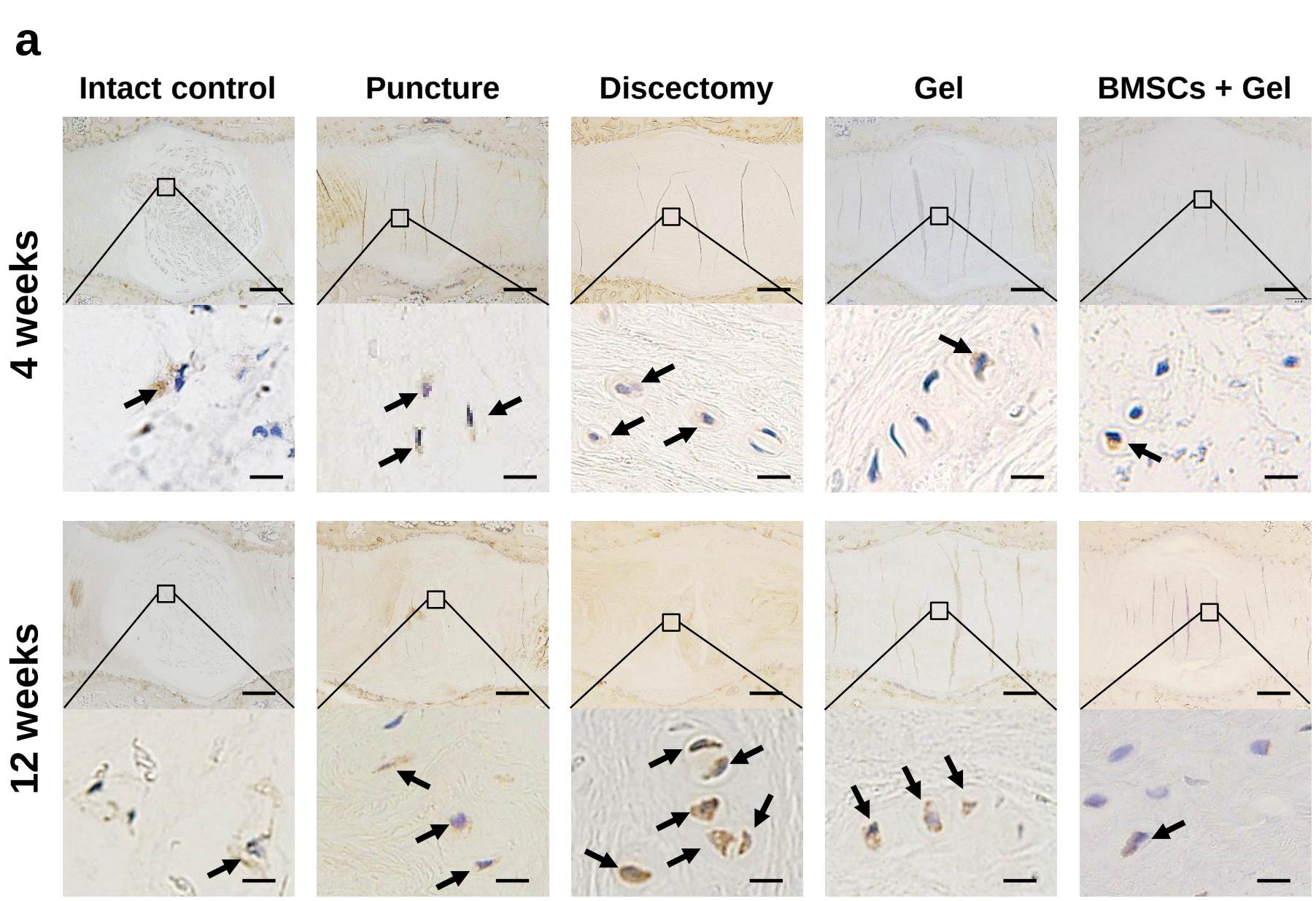


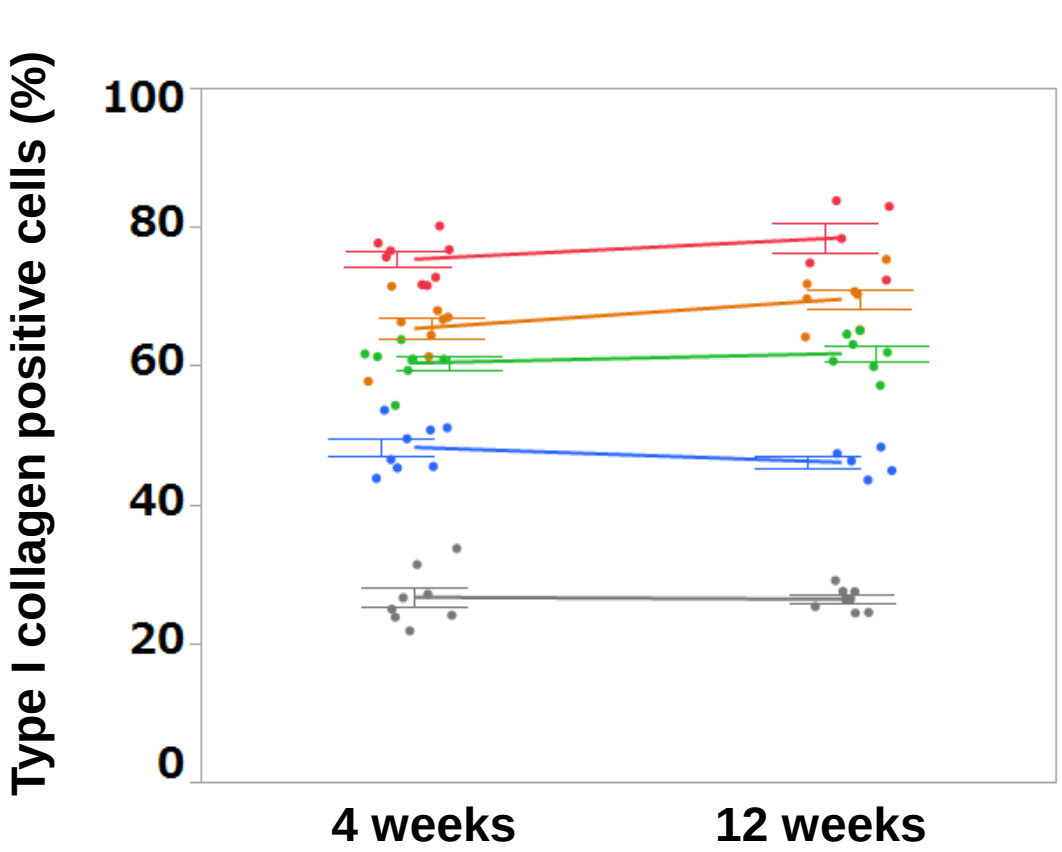


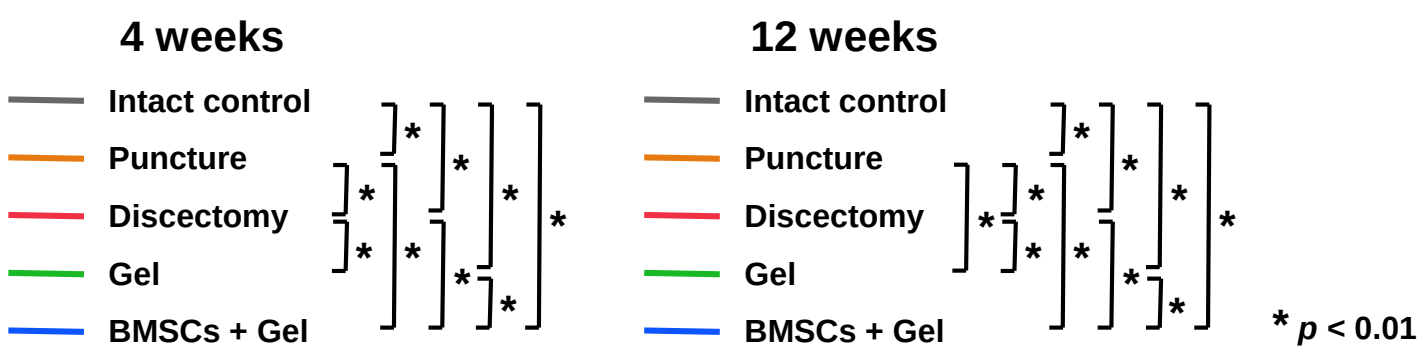
Supplementary Fig. 4. Human bone-derived mesenchymal stem cell (BMSC) labeled with caboxyfluorescein diactetate succinimidyl ester (CFDA-SE) implantation in rabbits. (a): Implanted human BMSCs survived in intervertebral discs (IVDs) at 4 and 12 weeks after surgery. Frozen sections of IVD stained by DAPI. Images are representative of four replicates (n = 4; 4 and 12 weeks). Scale bar = 50 μm. (b): Human-BMSCs combined with ultra-purified alginate (UPAL) gel preserved the water content of degenerated IVDs following discectomy. T2-weighted, midsagittal images of degenerated IVDs at 4 and 12 weeks after surgery. Images are representative of eight replicates. (c): Pfirrmann grading of IVD degeneration. No significant differences were apparent between rabbit and human BMSCs. Data represent the means ± SE (Allogeneic-BMSCs + Gel and Human-BMSCs + Gel n = 8; 4 and 12 weeks). N.S., not significant (d): Magnetic resonance imaging (MRI) index for degenerative alterations of the nucleus pulposus (NP). No significant differences were observed between rabbit and human BMSCs. Data are the means ± SE. p-values were determined by one-way ANOVA with post hoc analysis using the Tukey–Kramer test. N.S., not significant.



Supplementary Fig. 5. Human bone-derived mesenchymal stem cell (BMSC) implantation in rabbits. (a): Midsagittal sections of intervertebral discs (IVDs) stained by hematoxylin and eosin (H&E) and safranin O. Images are representative of eight replicates (n = 8; 4 and 12 weeks). AF; annulus fibrosus, NP; nucleus pulposus. Scale bars = 500 μm (first and third sections from the top), 50 μm (second section from the top). (b): Histological grading at 4 and 12 weeks. No significant differences were observed between rabbit and human BMSCs. Data represent the means ± SE. p-values were determined by one-way ANOVA with the post hoc Tukey–Kramer test. (c, e): Type II (c) and type I (e) collagen-positive cells in rabbit NPs. Midsagittal sections of rabbit IVDs stained for type II (c) or type I (e) collagen. Images are representative of eight replicates (n = 8; 4 and 12 weeks). Arrows indicate cells positive for type II (c) or type I (e) collagen. Scale bars = 500 μm (upper section) and 20 μm (lower section). (d, f): Percentages of type II (d) or type I (f) collagen-positive cells. No significant differences were observed between rabbit and human BMSCs. Data represent the means ± SE. p-values were determined by one-way ANOVA with the post hoc Tukey–Kramer test. N.S., not significant.







Supplementary Fig. 6. Type I collagen-positive cells in rabbit nucleus pulposus (NP). (a) Midsagittal sections of rabbit intervertebral discs (IVDs) stained for type I collagen. Images are representative of eight replicates (Intact control, Puncture, Discectomy, Gel, BMSCs + Gel, n = 8; 4 and 12 weeks). Arrows indicate cells positive for type I collagen. Scale bars = 500 μm (first and third sections from the top) and 20 μm (second and fourth sections from the top). (b): Percentages of type I collagen-positive cells. Data represent the means ± SE. p-values were determined by one-way ANOVA with the post hoc Tukey–Kramer test.

Supplementary Table. 1. Predesigned primer and probe mixes by Applied Biosystems for qRT-PCR

Gene	Gene symbol	Assay ID
HIF-1α	HIF1A	Oc03398626_m1
GLUT-1	SLC2A1	Oc03399482_m1
Brachyury	T	Oc03395780_m1
CDMP-1	GDF5	Oc00433564_m1
TGF-β	TGFB1	Oc04176122_u1
IGF-1	IGF1	Oc04096599_m1
Type II collagen	COL2A1	Oc03396134_m1
Aggrecan *	ACAN	-

* Custom TaqMan ® Gene Expression Assay