Appendix 1

Isolation and Culture of Degenerative Tenocytes from Human Rotator Cuff Tendons.

Tendon tissues 3 x 3mm in size were harvested from patients undergoing arthroscopic rotator cuff repair, washed twice in calcium-and magnesium-free phosphate-buffered saline (DPBS) and finely minced into 1 to 2mm fragments (n=3). Cells were isolated by treating with 0.3% collagenase II for 2 hours in Dulbecco's modified Eagle medium (DMEM) containing antibiotic solution (100 U/mL penicillin and 100 mg/mL streptomycin) with gentle agitation. After the same volume of DPBS was added, undigested tissue was removed using a 100-mm nylon sieve, and cells were collected by centrifugation, washed twice, resuspended in DMEM supplemented with 10% fetal bovine serum (FBS) and antibiotic solution (growth medium), and seeded in 100-mm tissue culture dishes at a density of 2 to 5 X 10^4 cells/cm^2 at 37°C in a humidified 5% CO_2 atmosphere. The medium was replaced every 2 to 3 days. When cells reached 60% to 80% confluence, they were detached by incubation for 5 minutes with 0.25% trypsin (Welgene, Daegu, Korea), washed, and then replated at a ratio of 1:3.

Appendix 2

Quantification of growth factors using ELISA.

The levels of EGF (Human EGF Quantikine ELISA Kit, DEG00; R&D Systems, Minneapolis, Minnesota), TGF-β1 (Human TGF-β1 Quantikine ELISA Kit, DB100B; R&D Systems), VEGF (Human VEGF Quantikine ELISA Kit, DVE00; R&D Systems), bFGF (Human FGF basic Quantikine HS ELISA Kit, HSFB00D; R&D Systems), PDGF-AB (Human PDGF-AB Quantikine ELISA Kit, DHD00C; R&D Systems), IGF-1 (Human IGF-1 Quantikine ELISA Kit, DG100; R&D Systems), and CTGF (Human CTGF ELISA Kit, SK00726-01; Aviscera Bioscience, Santa Clara, California) in PPP and PRPs were measured with use of commercially available ELISA kits. All experiments were performed in duplicate according to manufacturer's instructions. The optical densities of the microplate wells were measured with a microplate reader (SpectraMax Plus384; Molecular Devices, Sunnyvale, California). Sample concentrations were obtained by interpolating from the standard curve.

Appendix 3

Real-time reverse transcriptase polymerase chain reaction (RT-PCR).

Total RNA was extracted from tenocytes seeded at a density of 2 x 10^4 cells/cm^2 in the 6-well plate (SPL Lifesciences, Pocheon, Korea) using a HiYield Total RNA mini kit (Real Biotech Corporation, Taiwan) quantified using a NanoDrop ND-100 spectrophotometer (NanoDrop, Wilmington, Delaware). First-strand complementary DNA (cDNA) was synthesized using the Superscript III Reverse Transcription kit (Invitrogen, Carlsbad, California). Briefly, first-strand cDNA was synthesized from cellular RNAs (1 ug) by heating a mixture (1 ug RNA, 1 uL Oligo(dT)20 [50 uM], 1 uL dNTP [10 mM], and up to 10 uL DW) to 65°C for 5 minutes, cooling on ice for 2 minutes, and then adding a mixture containing 2 uL 10X RT buffer, 4 uL MgCl2 (25 mM), 2 uL DTT (0.1 M), 1 uL RNaseOut (40 U/mL), and 1 uL Superscript III Reverse Transcriptase (200 U/mL) (Invitrogen). The reaction mixture was held at 50°C for 50 minutes to promote cDNA synthesis, and the reaction was terminated by heating to 85°C for 5 minutes and then cooling on ice for 2 minutes. Finally, RNase H (1 uL, 2 U/mL) was added and incubated at 37°C for 20 minutes to remove RNA strands from RNA-cDNA hybrids. Synthesized cDNA was used for real-time RT-PCR. To perform real-time PCR utilizing a LightCycler 480 (Roche Applied Science, Mannheim, Germany), Taq-Man Gene Expression Assays (Applied Biosystems, Foster City, California) were used as a probe/primer set specified for IL-1β (assay ID: Hs99999029_m1), IL-6 (assay ID: Hs99999032_m1), COX-2 (assay ID: Hs00153133_m1), mPGES-1 (assay ID: Hs00610420_m1), TNF-α (assay ID: Hs99999043_m1), MMP-1 (assay ID: Hs00899658_m1), MMP-3 (assay ID: Hs00968308_m1), MMP-9 (assay ID: Hs00957555_m1), MMP-13 (assay ID: Hs00233992_m1), TIMP-1 (assay ID: Hs99999139_m1), TIMP-3 (assay ID: Hs00165949_m1), ADAMTS-4 (assay ID: Hs00192708_m1), ADAMTS-5 (assay ID: Hs00199841_m1), IL-4 (assay ID: Hs00174122_m1), IL-10 (assay ID: Hs00961622_m1), VIP (assay ID: Hs00175021_m1), IL-1RN (assay ID: Hs00893626_m1), type I collagen (assay ID: Hs00164004_m1), type III collagen (assay ID: Hs00943809_m1), decorin (assay ID: Hs00754870_s1), tenascin-C (assay ID: Hs00233648_m1), and GAPDH (assay ID: Hs99999905_m1). The PCRs were performed in a final volume of 20 uL containing 10 uL 2X LightCycler480 Probes Master (FastStart Taq DNA polymerase, reaction buffer, dNTP mix [with dUTP instead of dTTP], and 6.4 mM $MgCl_2$) (Roche Applied Science), 1 uL TaqMan Gene Expression Assay (Applied Biosystems), 5 uL cDNA as the template, and 4 uL H_2O using the following program: 95°C for 10 minutes, 60 cycles at 95°C for 10 seconds, and 60°C for 1

minute, followed by 72°C for 4 seconds, and a final cooling at 40°C for 30 seconds. Gene expressions were normalized versus GAPDH as follows: the cycle number at which the transcript of each gene was detectable (threshold cycle, Ct) was normalized against the Ct of GAPDH, which is referred to as $\triangle$Ct. Gene expressions relative to GAPDH are expressed as $2^{-\triangle Ct}$, where $\triangle Ct = C_T$ gene of interest – C_T GAPDH.

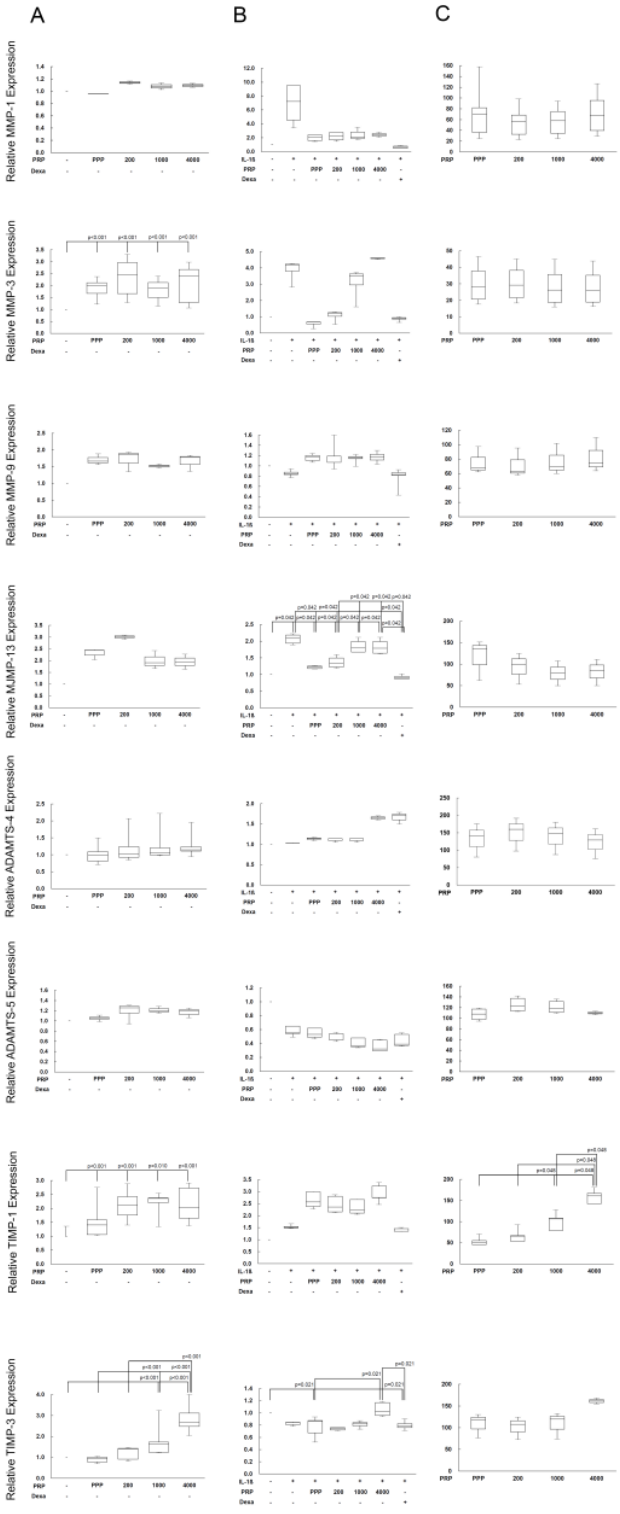
Appendix 4

Western blotting.

Cell protein extracts were prepared from tenocytes seeded at a density of 2 x 10^4 cells/cm^2 in the 6-well plate using PRO-PREP™ protein extraction solution (iNtRON, Sungnam, Korea). Equal amounts of protein extracts for each group were electrophoresed into 10% SDS-PAGE gels. The electrophoresed proteins were blotted onto a PVDF membrane with 0.45μm pore size. The membranes were blocked with TBS-T buffer containing 5% skim milk or 5% BSA for 1 hour at room temperature in 1X Tris-buffered saline with 0.1% tween 20 (TBS-T) followed by incubation with primary antibody overnight at 4°C. Primary antibodies were as follows : MMP-1 antibody (#MAB901, R&D Systems), MMP-3 antibody (#MAB905, R&D Systems), MMP-9 antibody (#2270, Cell signaling technology), MMP13 antibody (ab75606, abcam), ADAMTS-4 antibody (ab84792, abcam), ADAMTS-5 antibody (ab41037, abcam), TIMP-1 antibody (#8946, Cell signaling technology), TIMP-3 antibody (#ab39184, abcam), β-actin antibody (#sc47778, Santacruz Biotechnology). The membranes were washed with TBS-T, and incubated with HRP-conjugated secondary antibody diluted 1:4,000 for 45 minutes. After washing with 1X TBS containing 0.1% Tween 20 (TBS-T), the membranes were scanned using ImageQuant LAS4000 mini (GE Healthcare Life Sciences, Little Chalfont, UK). Densitometric quantitation was analyzed with ImageQuant LAS4000 mini. The protein synthesis levels were normalized to those of beta-actin. Western blotting was also performed with PPP and PRPs to identify proteins in themselves.

Appendix 5.

Effects of PRP on protein synthesis of degradative enzymes & their inhibitors with or without IL-1β treatment.



Appendix 6

Change of pain after PRP injection.

Variable	PRP (n=17)			*P* Value	Steroid (n=17)			*P* Value	*P* Value
Pain at rest									
Preinjection	2.3	±	1.7		2.6	±	2.2		.812
1W	2.4	±	1.8	.776	1.4	±	1.9	.012	.045
1M	1.5	±	1.7	.105	1.2	±	1.5	.012	.518
3M	1.8	±	1.8	.396	1.9	±	2.5	.275	.760
6M	0.8	±	1.5	.004	1.5	±	2.5	.056	.474
Pain on motion									
Preinjection	4.5	±	1.6		4.1	±	1.8		.610
1W	3.2	±	1.8	.006	2.2	±	1.8	.000	.205
1M	2.5	±	1.7	.002	1.8	±	1.8	.001	.634
3M	2.4	±	1.5	.001	2.9	±	2.6	.038	.518
6M	1.5	±	1.4	.000	2.4	±	2.5	.032	.454
Pain at night									
Preinjection	4.5	±	2.4		4.9	±	2.5		.496
1W	4.1	±	2.2	.372	3.2	±	2.7	.003	.150
1M	2.9	±	1.7	.016	2.5	±	1.9	.001	.231
3M	3	±	2.1	.039	2.8	±	2.9	.037	.306
6M	1.8	±	2.2	.004	3.2	±	3.0	.014	.375
Mean pain									
Preinjection	3.8	±	1.4		3.9	±	1.7		.786
1W	3.2	±	1.5	.120	2.2	±	1.7	.000	.014
1M	2.3	±	1.4	.003	1.9	±	1.5	.001	.231
3M	2.4	±	1.6	.002	2.7	±	2.6	.065	.375
6M	1.4	±	1.6	.000	2.5	±	2.5	.017	.433
Worst pain									
Preinjection	7.5	±	1.5		7.2	±	1.7		.586
1W	6.3	±	1.8	.004	5.5	±	2.2	.003	.586
1M	4.5	±	2.2	.001	4.4	±	2.7	.001	.734
3M	4.4	±	2.5	.001	3.9	±	3.5	.002	.786
6M	3.6	±	2.7	.001	4.4	±	3.6	.004	.205

Appendix 7

Change of ROM and strength after PRP injection.

Variable	PRP (n=17)	*P* Value	Steroid (n=17)	*P* Value	*P* Value
Forward flexion, deg					
Preinjection	162.4 ± 19.5		162.6 ± 12.1		.540
1W	168.2 ± 11.7	.083	167.1 ± 10.0	.085	.586
1M	168.8 ± 13.6	.059	164.4 ± 13.2	.473	.496
3M	166.8 ± 15.1	.222	163.0 ± 13.9	.446	.708
6M	167.1 ± 15.1	.461	159.7 ± 14.5	.728	.306
Abduction, deg					
Preinjection	167.1 ± 17.1		165.9 ± 11.5		.474
1W	167.1 ± 21.6	.757	170.6 ± 9.0	.021	.245
1M	161.8 ± 32.3	.505	170.6 ± 11.3	.063	.586
3M	162.9 ± 29.5	.721	168.3 ± 11.1	.264	.760
6M	165.0 ± 23.8	.906	163.8 ± 12.0	.682	.540
External rotation with arm at the side, deg					
Preinjection	52.9 ± 15.7		47.6 ± 10.0		.306
1W	55.3 ± 16.6	.354	49.1 ± 11.5	.334	.812
1M	53.8 ± 18.2	.813	48.5 ± 15.4	.673	1.000
3M	53.2 ± 16.8	.905	51.3 ± 14.7	.412	.708
6M	53.2 ± 16.6	.648	45.9 ± 14.5	.836	.474
Internal rotation, vertebral level					
Preinjection	11.2 ± 2.3		10.6 ± 2.6		.540
1W	11.1 ± 2.6	.914	10.9 ± 2.2	.380	.760
1M	11.2 ± 2.1	.943	11.3 ± 1.8	.135	.160
3M	11.1 ± 2.2	.931	11.4 ± 1.8	.053	.394
6M	11.8 ± 1.7	.147	11.4 ± 1.5	.054	.708
Supraspinatus, lb					
Preinjection	9.6 ± 5.0		10.6 ± 5.9		.812
1W	9.0 ± 4.1	.245	11.6 ± 5.9	.179	.122
1M	12.0 ± 5.2	.076	12.0 ± 5.1	.063	.634
3M	13.8 ± 6.0	.003	10.4 ± 5.3	.460	.022
6M	13.5 ± 5.7	.010	10.7 ± 4.5	.623	.002
Infraspinatus, lb					
Preinjection	9.7 ± 3.8		9.0 ± 4.9		.474
1W	9.6 ± 2.7	.586	10.4 ± 4.8	.088	.259
1M	10.6 ± 3.2	.163	10.7 ± 3.5	.041	.973
3M	12.6 ± 5.3	.005	9.8 ± 4.6	.600	.022
6M	11.0 ± 3.3	.124	9.8 ± 3.9	.205	.518
Subscapularis, lb					
Preinjection	13.3 ± 5.6		12.5 ± 4.4		.838

1W	13.5	±	5.0	.394	13.7	±	4.9	.435	.919
1M	14.7	±	6.1	.093	15.7	±	5.3	.033	.892
3M	16.3	±	5.4	.016	15.0	±	6.1	.484	.073
6M	16.1	±	5.8	.007	14.5	±	5.1	.301	.193

Variable	Affected side(n=17)			Contralateral side(n=17)			*P* Value
Supraspinatus, lb (PRP group)							
Preinjection	9.6	±	5.0	12.9	±	4.8	.045
1W	9.0	±	4.1	12.8	±	4.4	.009
1M	12.0	±	5.2	13.7	±	4.7	.205
3M	13.8	±	6.0	15.4	±	5.2	.394
6M	13.5	±	5.7	14.7	±	4.3	.433
Infraspinatus, lb (PRP group)							
Preinjection	9.7	±	3.8	10.8	±	3.2	.322
1W	9.6	±	2.7	10.8	±	2.3	.218
1M	10.6	±	3.2	11.9	±	3.0	.245
3M	12.6	±	5.3	12.7	±	4.7	.734
6M	11.0	±	3.3	12.6	±	3.6	.394
Subscapularis, lb (PRP group)							
Preinjection	13.3	±	5.6	14.9	±	4.9	.170
1W	13.5	±	5.0	15.4	±	5.1	.182
1M	14.7	±	6.1	16.5	±	5.6	.290
3M	16.3	±	5.4	17.1	±	4.6	.634
6M	16.1	±	5.8	16.3	±	6.4	.683
Supraspinatus, lb (Steroid group)							
Preinjection	10.6	±	5.9	13.2	±	5.5	.131
1W	11.6	±	5.9	13.7	±	5.0	.182
1M	12.0	±	5.1	13.8	±	4.4	.246
3M	10.4	±	5.3	14.0	±	4.4	.102
6M	10.7	±	4.5	14.3	±	4.1	.023
Infraspinatus, lb (Steroid group)							
Preinjection	9.0	±	4.9	11.2	±	4.6	.122
1W	10.4	±	4.8	11.3	±	4.2	.496
1M	10.7	±	3.5	11.5	±	3.3	.539
3M	9.8	±	4.6	11.8	±	4.1	.336
6M	9.8	±	3.9	11.8	±	3.9	.196
Subscapularis, lb (Steroid group)							
Preinjection	12.5	±	4.4	16.4	±	5.2	.041
1W	13.7	±	4.9	17.1	±	4.2	.079
1M	15.7	±	5.3	18.5	±	4.1	.202
3M	15.0	±	6.1	18.3	±	6.3	.223
6M	14.5	±	5.1	16.8	±	4.7	.210

Appendix 8

Change of ASES, Constant, UCLA, DASH, SST, and SPADI Scores after PRP injection.

Variable	PRP (n=17)			*P* Value	Steroid (n=17)			*P* Value	*P* Value
ASES									
Preinjection	65.0	±	10.7		64.3	±	12.1		.634
1W	66.5	±	12.8	.623	76.0	±	14.2	0.000	.002
1M	75.3	±	13.4	.005	82.3	±	13.8	0.000	.049
3M	74.5	±	16.6	.013	75.9	±	23.3	0.044	.586
6M	85.3	±	12.6	.000	77.6	±	22.2	0.017	.193
Constant									
Preinjection	65.7	±	9.3		64.3	±	11.6		.563
1W	67.7	±	8.5	.193	72.7	±	11.8	0.006	.062
1M	72.0	±	11.0	.039	73.7	±	10.3	0.005	.563
3M	75.5	±	13.3	.010	69.4	±	14.7	0.125	.357
6M	79.1	±	9.2	.001	71.4	±	14.1	0.074	.160
UCLA									
Preinjection	20.6	±	3.8		19.9	±	3.2		.474
1W	23.9	±	4.7	.012	25.6	±	4.5	0.001	.150
1M	25.5	±	4.4	.005	27.3	±	4.2	0.001	.306
3M	24.3	±	5.6	.026	22.8	±	10.9	0.170	.563
6M	27.2	±	5.7	.005	25.1	±	8.9	0.014	.634
DASH									
Preinjection	25.1	±	12.3		23.0	±	8.8		.683
1W	26.4	±	13.5	.794	18.9	±	10.9	0.038	.041
1M	22.1	±	12.9	.102	13.4	±	9.3	0.004	.026
3M	20.8	±	14.8	.149	15.4	±	24.0	0.218	.540
6M	13.5	±	10.7	.001	16.9	±	22.6	0.149	.634
SST									
Preinjection	7.4	±	2.7		7.5	±	2.0		.812
1W	7.6	±	2.5	.324	8.5	±	2.2	0.086	.634
1M	8.6	±	2.4	.034	10.4	±	3.2	0.002	.182
3M	8.9	±	2.4	.074	7.8	±	4.2	0.571	.518
6M	10.5	±	3.8	.015	8.7	±	3.9	0.139	.496
SPADI									
Preinjection	36.5	±	14.3		34.4	±	11.3		.838
1W	32.8	±	15.5	.407	20.8	±	12.3	0.000	.009
1M	23.3	±	14.9	.002	16.7	±	11.2	0.000	.231
3M	24.5	±	19.5	.006	20.0	±	23.0	0.026	.357
6M	14.5	±	12.2	.000	20.5	±	22.5	0.014	.290

ASES, American Shoulder and Elbow Surgeons; UCLA, University of California, Los Angeles; DASH, Disability Assessment of Shoulder and Hand; SST, Simple Shoulder Test; SPADI, Shoulder Pain and Disability Index

Appendix 9

Overall function and satisfaction after PRP injection.

Variable	PRP (n=17)	*P* Value	Steroid (n=17)	*P* Value	*P* Value
Overall function					
Preinjection	5.5 ± 1.2		5.8 ± 2.3		.433
1W	5.7 ± 1.4	.382	6.2 ± 1.5	.544	.973
1M	5.9 ± 1.6	.299	6.4 ± 1.5	.255	.708
3M	5.9 ± 2.2	.472	5.2 ± 3.3	.483	.290
6M	7.1 ± 1.8	.004	5.7 ± 3.1	.695	.029
Overall satisfaction					
1W	58.8 ± 26.6	NA	62.4 ± 28.2	NA	.610
1M	69.7 ± 25.2	NA	68.8 ± 20.9	NA	.973
3M	61.2 ± 27.4	NA	59.4 ± 35.1	NA	.892
6M	76.8 ± 20.2	NA	60.6 ± 33.1	NA	.160