

Supplementary Figure and Legend

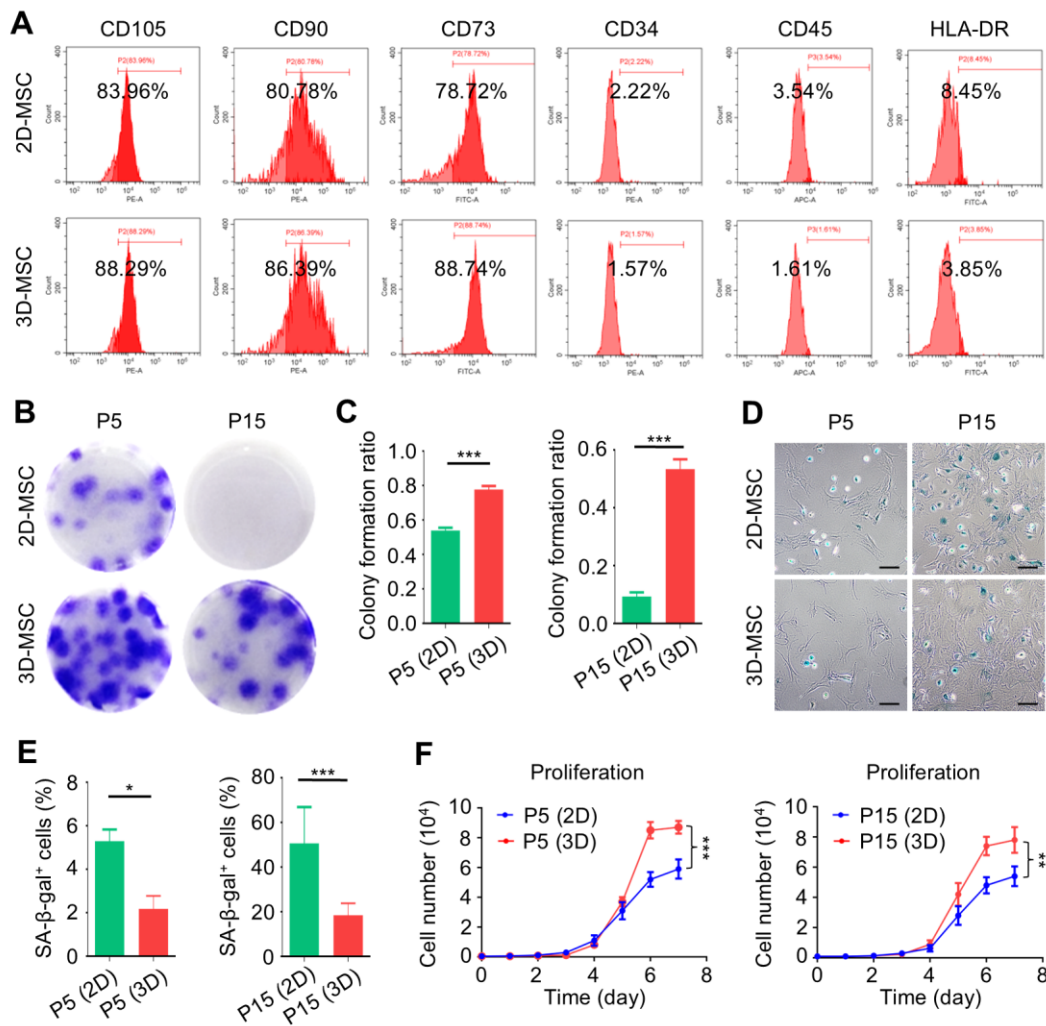


Figure S1. Three-dimensional culture rescues replicative senescence of MSCs despite extensive passages. (A) Flow cytometric analysis of surface markers of MSCs. (B) Colony formation of MSCs analyzed by crystal violet staining. (C) Quantification of colony forming units of MSCs over total seeded cells. (D) Cell senescence of MSCs analyzed by SA-β-gal staining. Scale bars = 100 μm. (E) Quantification of senescent percentages of MSCs. (F) Proliferation rate of MSCs analyzed by CCK8. *N* = 3 per group. Mean ± SD. *, *P* < 0.05; **, *P* < 0.01; ***, *P* < 0.001. Two-tailed unpaired Student's *t* test.

Supplementary Table

Table S1. Key resources table.

REAGENT or RESOURCE	SOURCE	IDENTIFIER
Antibodies		
FITC Mouse Anti-Human CD73	BD Bioscience	Cat# 561254, RRID: AB_10894209
PE Mouse anti-Human CD105	BD Bioscience	Cat# 560839, RRID: AB_2033932
PE Mouse Anti-Human CD90	BD Bioscience	Cat# 561970, RRID: AB_395970
PE Mouse Anti-Human HLA-DR	BD Bioscience	Cat# 555561, RRID: AB_395943
PE Mouse Anti-Human CD34	BD Bioscience	Cat# 550761, RRID: AB_393871
APC Mouse Anti-Human CD45	BD Bioscience	Cat# 555485, RRID: AB_398600
PE anti-human CD3 Antibody	BioLegend	Cat# 300308, RRID: AB_314044
Recombinant anti-Human CD3 mAb	Novoprotein	Cat# GMP-A018, RRID: N/A
Recombinant anti-Human CD28 mAb	Novoprotein	Cat# GMP-A063, RRID: N/A
PE anti-human CD4 Antibody	BioLegend	Cat# 357404, RRID: AB_312696
IFN gamma Monoclonal Antibody (4S.B3), PerCP-Cyanine5.5	Thermo Fisher Scientific	Cat# 45-7319-41, RRID: AB_10718246
IL-4 Monoclonal Antibody (8D4-8), APC	Thermo Fisher Scientific	Cat# 17-7049-41, RRID: AB_10853508
FITC anti-human IL-17A Antibody	BioLegend	Cat# 512303, RRID: AB_961391
Rat anti-CD4 Monoclonal Antibody	Absin	Cat# abs174032
Rabbit anti-IFN- γ Polyclonal Antibody	Absin	Cat# abs119966
Rabbit anti-IL-17 Polyclonal Antibody	Absin	Cat# abs121447
Anti-YAP1 Antibody	Abcam	Cat# ab56701, RRID: AB_2219140
Alexa Fluor 488 Donkey Anti-Rabbit IgG(H+L)	Yeasen Biotechnology	Cat# 34206ES60, RRID: N/A
Alexa Fluor 488 Donkey Anti-Rat IgG(H+L)	Yeasen Biotechnology	Cat# 34406ES60, RRID: N/A

Alexa Fluor 594 Goat Anti-Rabbit IgG(H+L)	Yeasen Biotechnology	Cat# 33112ES60, RRID: N/A
Alexa Fluor 594 Goat Anti-Mouse IgG(H+L)	Yeasen Biotechnology	Cat# 33212ES60, RRID: N/A
Goat Anti-Mouse IgG Antibody, HRP-conjugated	Signalway	Cat# L3032, RRID: AB_895481
Anti GAPDH Mouse Monoclonal Antibody	CWBio	Cat# CW0100, RRID: AB_2801390
Anti β -Tubulin Antibody	Abcam	Cat# ab314069, RRID: N/A
Chemicals, Peptides, and Recombinant Proteins		
Hoechst 33342	Yeasen Biotechnology	Cat# 40731ES10
Trypsin	Gibco	Cat# 15050057
Lymphoprep	MP biomedicals	Cat# 0916922-CF
Prostaglandin E2	MedChemExpress	Cat# HY-101952
Verteporfin	MedChemExpress	Cat# HY-B0146
Red Blood Cell Lysis Buffer	Solarbio	Cat# R1010
Dextran sulfate sodium salt	MP Biomedicals	Cat# 02160110-CF
Paraformaldehyde	Sigma-Aldrich	Cat# 158127
Streptozotocin	MP Biomedicals	Cat# 02100557-CF
Crystal violet	Solarbio	Cat# C8470
Triton X-100	Solarbio	Cat# T8200
Methanol	Kemao	N/A
Ethanol	Fuyu	N/A
Goat serum	Boster	Cat# AR0009
Matrigel	Corning	Cat# 356231
Dimethyl sulfoxide	Solarbio	Cat# D8370
TRIzol	Invitrogen	Cat# 15596026
Tris (Hydroxymethyl) Aminomethane	Solarbio	Cat# T8060
Sodium dodecylsulfate	Solarbio	Cat# S8010
Glycine	Solarbio	Cat# G8200
Bovine serum albumin	Gemini Bio	Cat# 700-100P
20x TBS buffer	Coolaber	Cat# SL1327
SWE Fast High Resolution Electrophoresis Buffer (Dry Powder)	Servicebio	Cat# G2081
PVDF, 0.2 μ m, 26.5 cm x 3.75 m	Millipore	Cat# ISEQ00010
Critical Commercial Assays		
Cell counting kit-8	Yeasen Biotech	Cat# 40203ES60
Human Regulatory T Cell Staining Kit	Thermo Fisher Scientific	Cat# 88-8999-40
Human TGF- β 1 ELISA kit	Neobioscience	Cat# EHC107b
Human IL-6 ELISA kit	Neobioscience	Cat# EHC007
Mouse TNF- α ELISA kit	Neobioscience	Cat# EMC102a
Mouse C-peptide ELISA Kit	Fankewei	Cat# F2580-A

BCA Protein Quantitative Kit	Beyotime	Cat# P0010
Cell Total RNA isolation Kit	Foergene	Cat# RE03113
PrimeScript™ RT reagent Kit	Takara	Cat# RR036A
TB Green® Premix Ex Taq II kit	Takara	Cat# RR820L
Hematoxylin and Eosin Staining Kit	Beyotime	Cat# C0105M
Senescence β -Galactosidase Staining Kit	Beyotime	Cat# C0602
RIPA buffer (high)	Beyotime	Cat# P0013B
Enhanced Chemiluminescence Kit	Amersham Biosciences	N/A
PAGE Gel Fast Preparation Kit	Epizyme Biotech	Cat# PG112
Medium		
α -MEM	Gibco	Cat# 12000063
Cell recovery solution	Corning	Cat# 354253
FBS	Gibco	Cat# 16140071
RPMI 1640	Gibco	Cat# 11875093
Penicillin-Streptomycin	Gibco	Cat# 115140122
PBS	Sigma-Aldrich	Cat# P5493
Experimental Models: Cell Lines		
N/A		
Experimental Models: Organisms/Strains		
Mouse: C57BL/6J	Laboratory Animal Center of the Fourth Military Medical University	N/A
Oligonucleotides		
qRT-PCR primers	Sangon Biotech	N/A
Recombinant DNA		
N/A		
Software and Algorithms		
Gen5	Bio-Tek	RRID: SCR_017317
GraphPad Prism	GraphPad	RRID: SCR_002798
ImageJ 1.47	National Institute of Health	RRID: SCR_003070
CytExpert Software2.4	Beckman Coulter	RRID: SCR_017217
Bio-Rad CFX Maestro	Bio-Rad	N/A
Adobe Photoshop	Adobe Systems Software Ireland Ltd	N/A
Microsoft Office	Microsoft	N/A
OLYMPUS FLUOVIEW FV31S-SW	OLYMPUS	N/A
OlyVIA/XV Imaging	Olympus Soft Imaging Solutions GmbH	N/A
BGISEQ-500	https://www.bgi.com/global/company/careers/bgi-launches-its-desktop-sequencer-bgiseq-500/	RRID:SCR_017979

DESeq2	https://bioconductor.org/packages/release/bioc/html/DESeq2.html	RRID: SCR_015687
R (4.3.0)	https://www.r-project.org/	RRID:SCR_001905