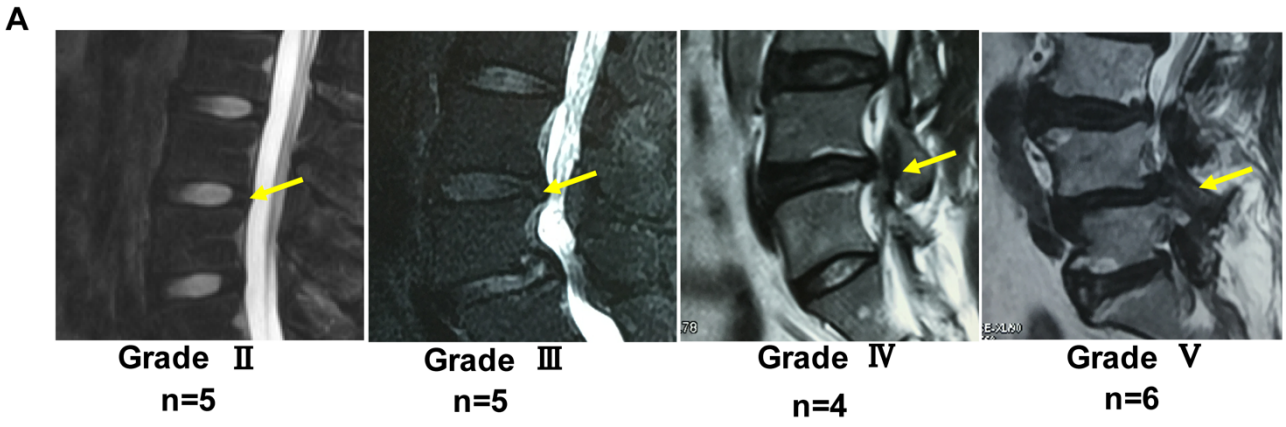
Supplementary tables and figures

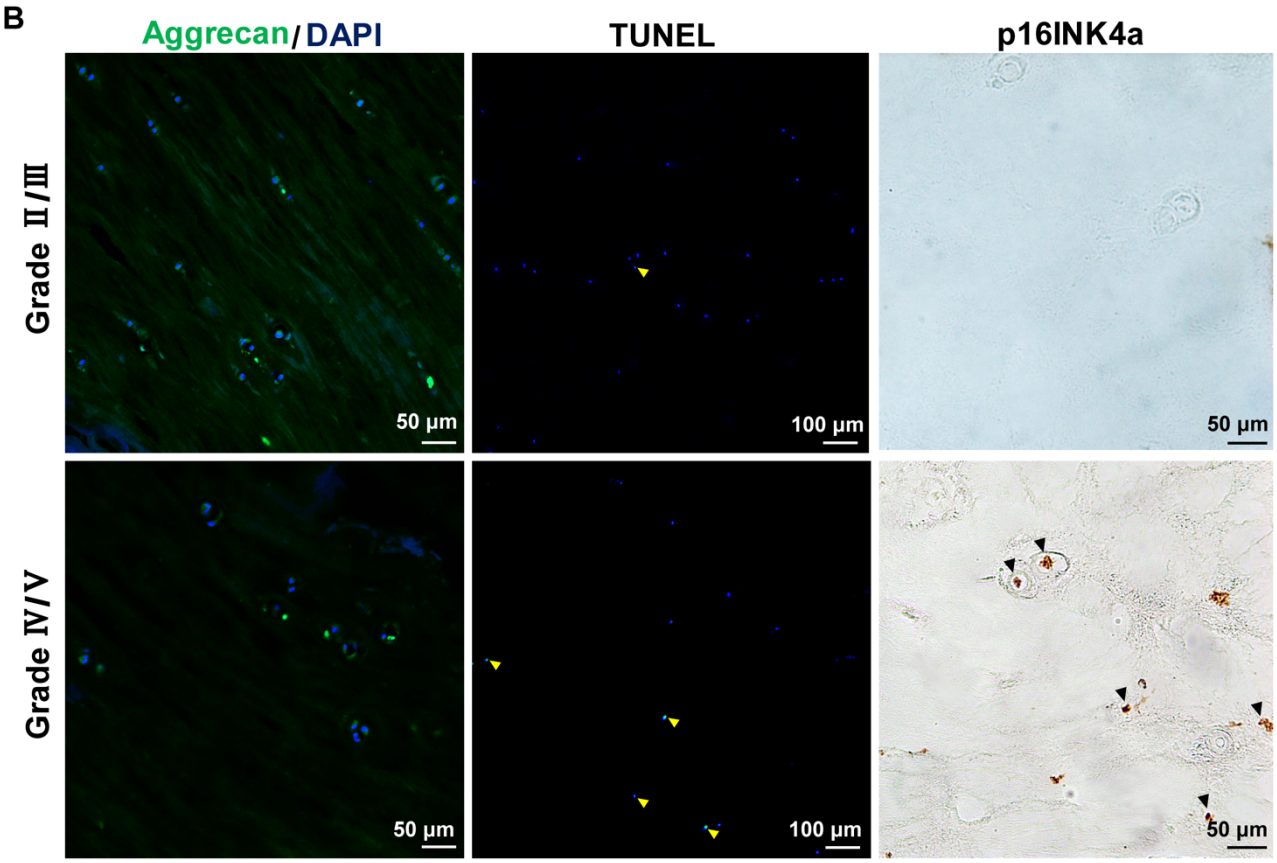
Table S1. Demographic data of patients.

Patients NO.	Age	Gender	Level	Pfirrmann grading
Grade II/III group				
1	6	M	L3/4	II
2	16	F	L3/4	II
3	13	F	L2/3	II
4	12	F	L3/4	II
5	25	M	L5/S1	II
6	45	F	L4/5	III
7	61	F	L4/5	III
8	53	M	L4/5	III
9	28	M	L4/5	III
10	32	F	L4/5	III
Grade IV/V group				
11	52	M	L4/5	IV
12	36	F	L4/5	IV
13	31	M	L4/5	IV
14	26	M	L4/5	IV
15	71	F	L4/5	V
16	33	M	L5/S1	V
17	34	M	L5/S1	V
18	54	F	L5/S1	V
19	42	F	L4/5	V
20	58	M	L4/5	V

Table S2. Sequences of primers used for qRT-PCR.

Gene (Homo)	Sequence (5′–3′)	
Progerin	Sense	GCAACAAGTCCAATGAGGACCA
	Antisense	CATGATGCTGCAGTTCTGGGGGCT CTGGAC
LMNA	Sense	CTACACCAGCCAACCCAGAT
	Antisense	ACTGAGTCAAGGGTCTTGCG
GAPDH	Sense	AATGGGCAGCCGTTAGGAAA
	Antisense	GCGCCCAATACGACCAAATC
Gene (Rattus)	Sequence (5′–3′)	
Acan	Sense	GCGATGCCACCTTGGAAATC
	Antisense	AGTCCAGTGTGTAGCGTGTG
Col2a1	Sense	AGTCCAGTGTGTAGCGTGTG
	Antisense	ACCCCTCTCTCCCTTGTCAC
Mmp13	Sense	GTGACTCTTGCGGGAATCCT
	Antisense	CAGGCACTCCACATCTTGGT
Opa1	Sense	GCCCTTCCCAGTTCAGAAGA
	Antisense	GGTGTACCCGCAGTGAAGAA
Dbp	Sense	GAAAAGGAGCGCAAGGCAAC
	Antisense	CGTATTCCACGTCCCCGAAA
Mfn1	Sense	CAAAGAAGGCCATCACTGCG
	Antisense	TCCGATCAAGTTCCGGGTTC
Mfn2	Sense	GAATCGGCACAGAGGAGACC
	Antisense	AAGTGCTTGAGAGGGGAAGC
Fis1	Sense	ACCCAAGCGTGCTTTCTGTA
	Antisense	TCATCCCTTACCACGCAACC
Tfam	Sense	TTCCAGGGGGCTAAGGATGA
	Antisense	CACACTGCGACGGATGAGAT
GAPDH	Sense	AGTGCCAGCCTCGTCTCATA
	Antisense	GATGGTGATGGGTTTCCCGT
Gene (Mus)	Sequence (5′–3′)	
Progerin	Sense	CTATTGCATGCTTCTCCTCAG
	Antisense	TGAGCGCAGGTTGTACTCAG


A
Grade Ⅱ
n=5
Grade Ⅲ
n=5
Grade Ⅳ
n=4
Grade Ⅴ
n=6


B
Aggrecan/DAPI
TUNEL
p16INK4a
Grade Ⅱ/Ⅲ
Grade Ⅳ/Ⅴ
50 μm
100 μm
50 μm
50 μm
100 μm
50 μm

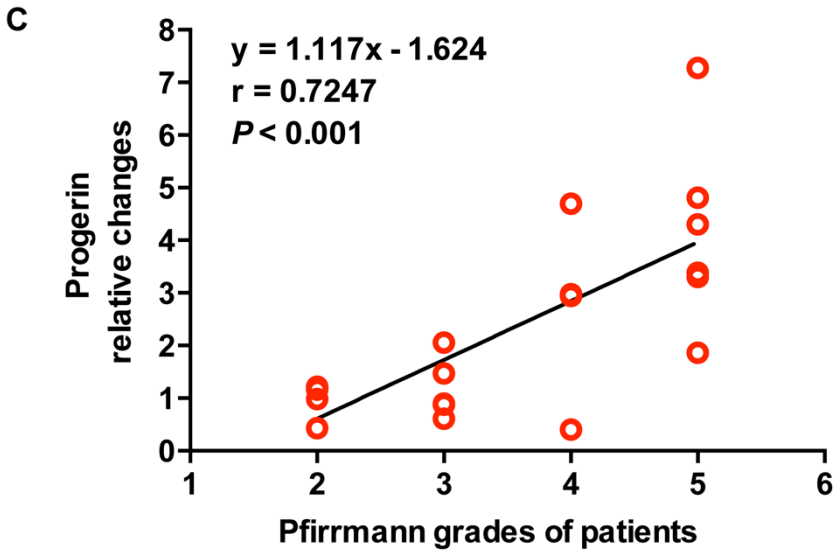

C
y = 1.117x - 1.624
r = 0.7247
P < 0.001
Progerin relative changes
Pfirrmann grades of patients
0
1
2
3
4
5
6
7
8
1
2
3
4
5
6

Figure S1. Human IDD NP tissues display an increased number of apoptotic NP cells and matrix degradation. (A) NP tissues were collected from patients and classified according to the modified Pfirrmann grading system: grades II (n = 5), grades III (n = 5), grades IV (n = 4) and V (n = 6). Arrows indicate MRI images of NP biopsies. **(B)** Representative images showing aggrecan (green) expression, TUNEL-positive cells (green), and p16INK4a (brown) in NP tissue sections from the Grade II/III and Grade IV/V groups. Nuclei were stained with DAPI. TUNEL, terminal deoxynucleotidyl transferase-mediated dUTP nick-end labelling. **(C)** Correlation between progerin expression and Pfirrmann grade (n = 20).

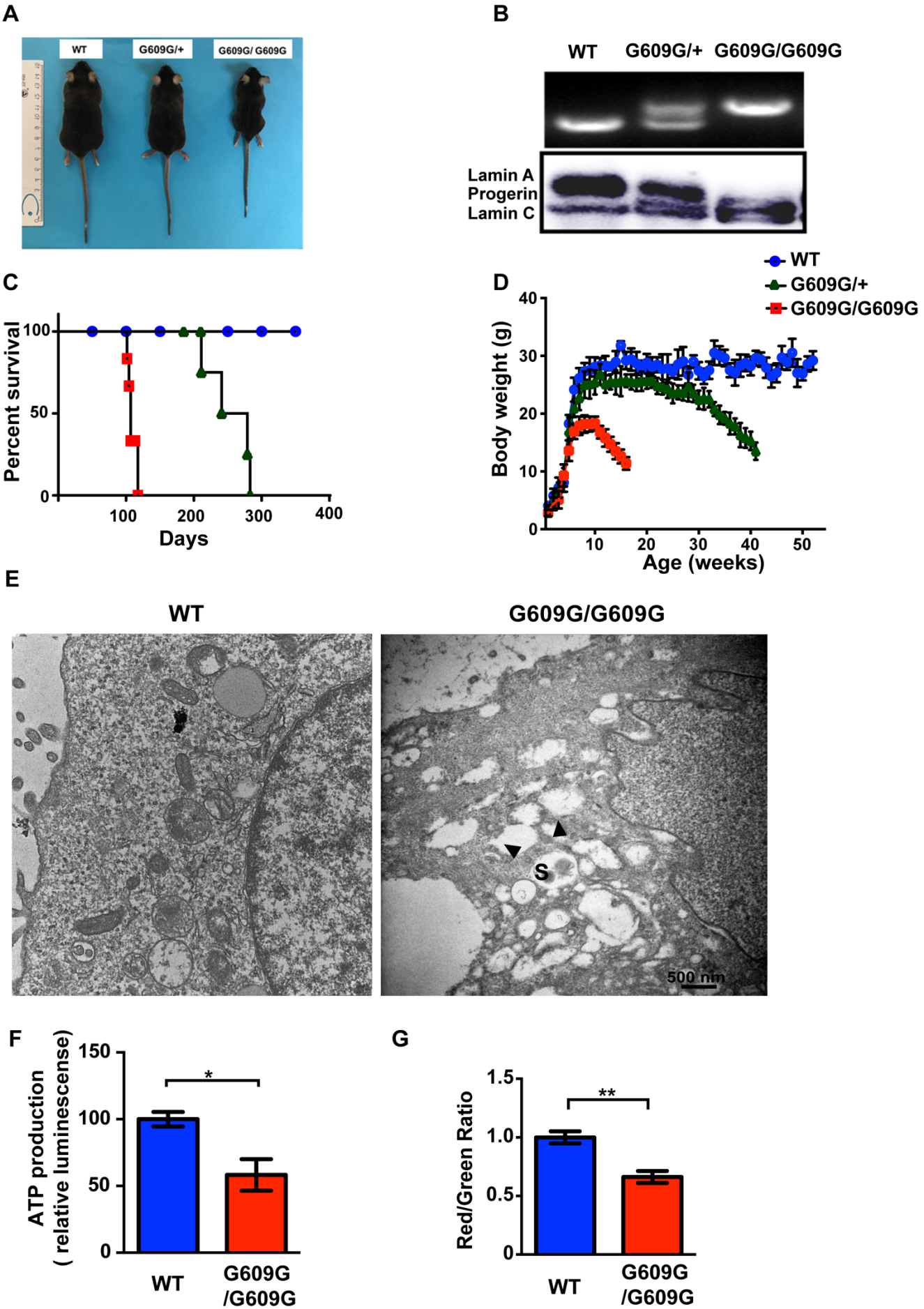

A
WT
G609G/+
G609G/ G609G
B
WT
G609G/+
G609G/G609G
Lamin A
Progerin
Lamin C
C
Percent survival
100
50
0
100
200
300
400
Days
D
WT
G609G/+
G609G/G609G
Body weight (g)
40
30
20
10
0
10
20
30
40
50
Age (weeks)
E
WT
G609G/G609G
S
500 nm
F
ATP production
(relative luminescense)
150
100
50
0
*
WT
G609G
/G609G
G
Red/Green Ratio
1.5
1.0
0.5
0
**
WT
G609G
/G609G

Figure S2. LMNA G609G/G609G mice display decreased lifespan extension and impaired mitochondrial function in disc. (A) Representative photographs of 4-month-old WT, heterozygous (G609G/+), and homozygous (G609G/G609G) mice. **(B)** qRT-PCR and Western blotting analyses of the indicated genotypes. **(C)** Kaplan–Meier survival plots of homozygous (n = 6), heterozygous (n = 6), and WT (n = 6) mice. **(D)** Cumulative plots of body weight *versus* age; n = 3. **(E)** Representative TEM images of mitochondria in NP cells from the WT and G609G/G609G mice group; S, swollen mitochondria. **(F)** ATP production in the WT and G609G/G609G groups; n = 3; $*P < 0.05$. **(G)** JC-1 staining. The red: green fluorescence ratio reflects changes in the mitochondrial membrane potential in the WT and G609G/G609G groups; n = 3; $**P < 0.01$. Data represent mean ± SEM.

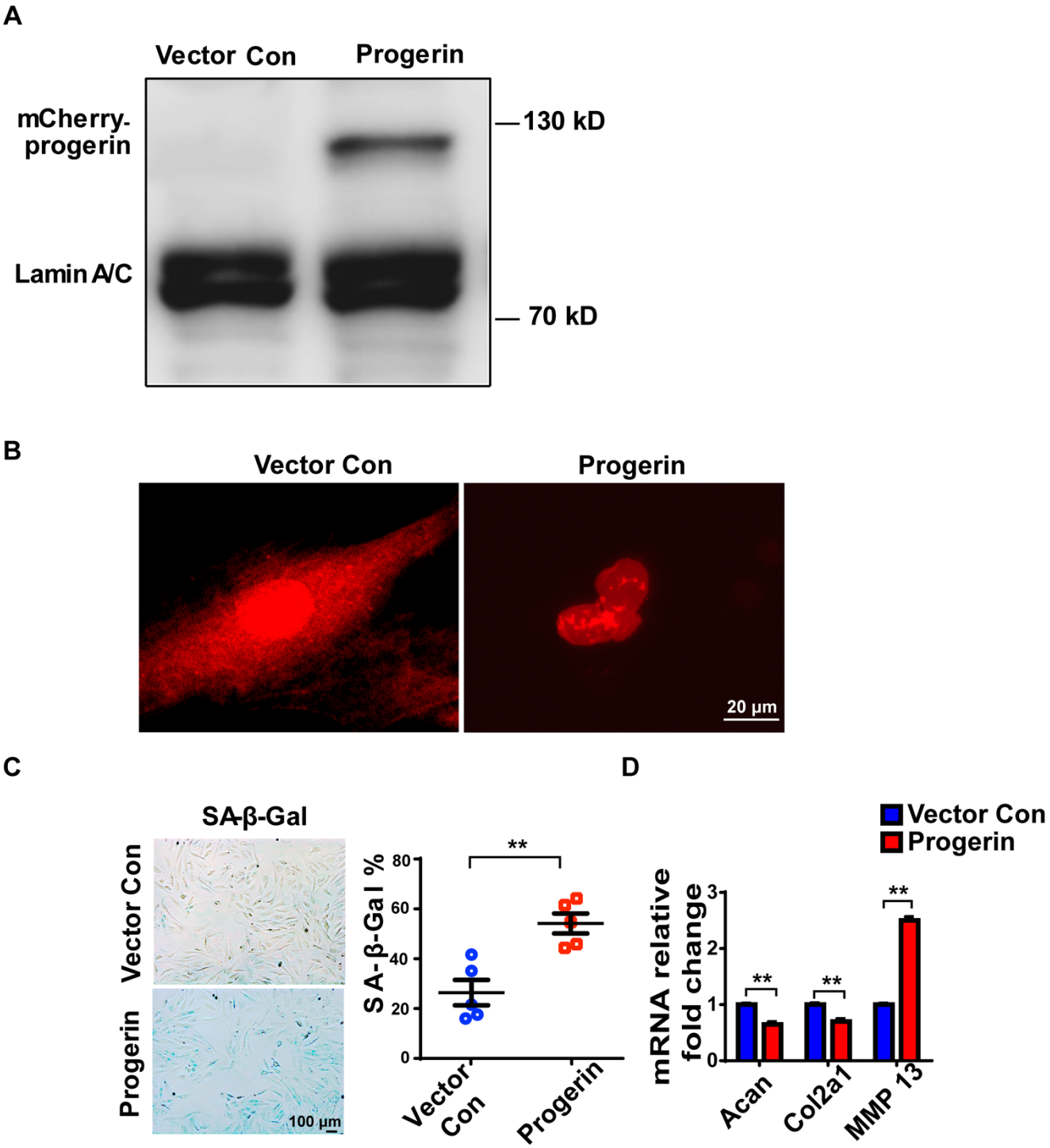


Figure S3. Progerin overexpression causes nucleus deformation and induces senescence and apoptosis of NP cells. (A) Rat NP cells were transduced with lentivirus expressing the vector (vector con) or mCherry-progerin (progerin). Western blotting analysis using an anti-Lamin A/C antibody of rat NP cells from the vector con or progerin group. **(B)** Representative fluorescence images of rat NP cells from the vector con or progerin group. **(C)** Frequency of senescent (SA-β-gal-positive; blue) cells from the vector con and progerin groups; n = 5; $^{**}P < 0.01$. **(D)** qRT-PCR

analysis of the mRNA levels of Acan, Col2a1, and MMP-13 in NP cells from the vector con and progerin groups; n = 3; $^{**}P < 0.01$. Data represent mean ± SEM.

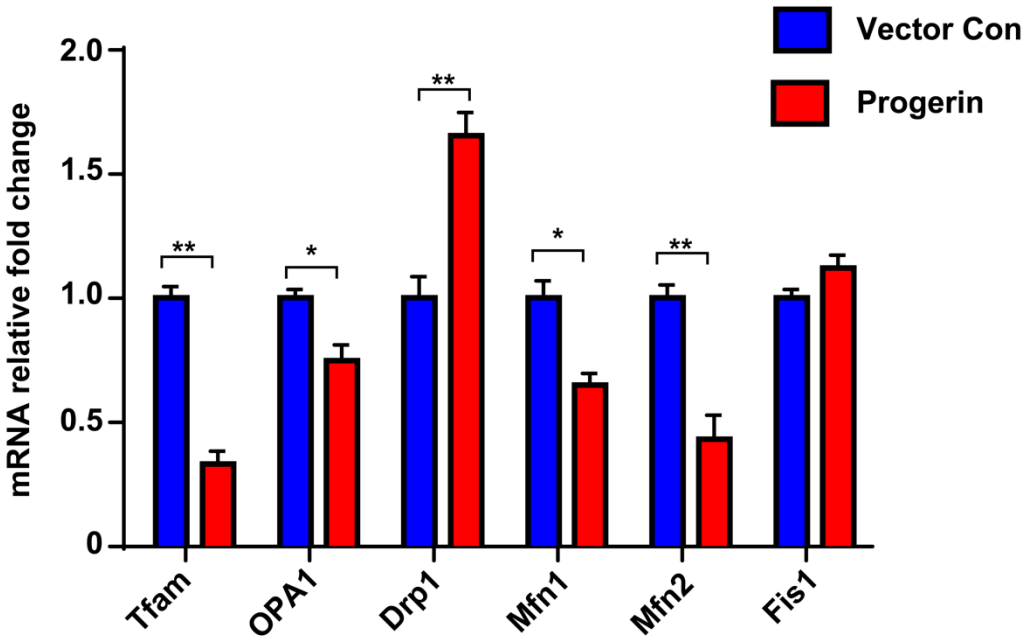


Figure S4. Effect of progerin on the expression of genes related to mitochondrial biogenesis. qRT-PCR analysis of the expression of *Tfam*, *OPA1*, *Drp1*, *Mfn1*, *Mfn2*, and *Fis1* in rat NP cells from the vector con and progerin groups; n = 3; **P* < 0.05, ***P* < 0.01. Data represent mean ± SEM.

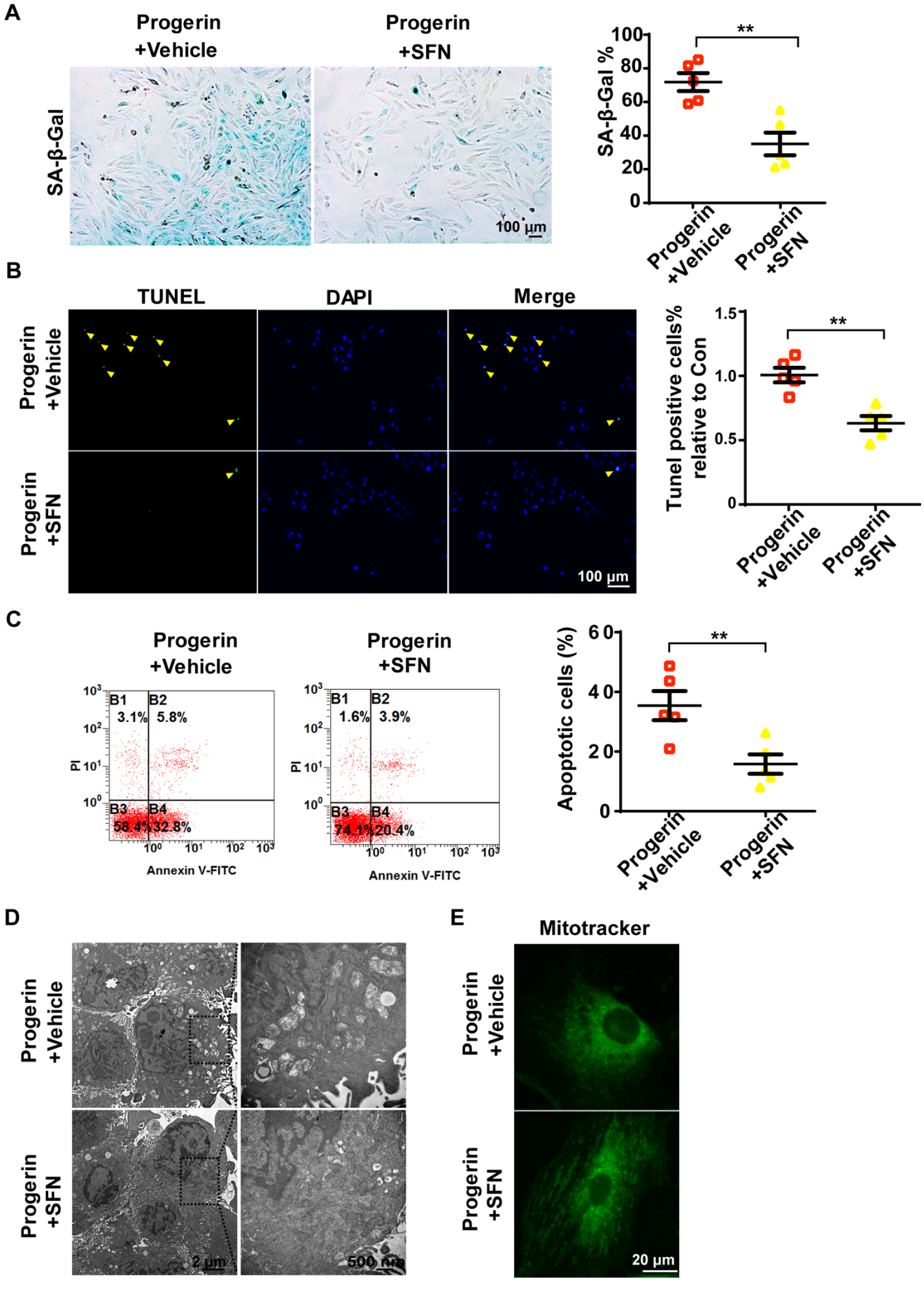

A
Progerin +Vehicle
Progerin +SFN
SA-β-Gal
100 μm
SA-β-Gal %
100
80
60
40
20
0
**
Progerin +Vehicle
Progerin +SFN
B
TUNEL
DAPI
Merge
Progerin +Vehicle
Progerin +SFN
100 μm
Tunel positive cells% relative to Con
1.5
1.0
0.5
0
**
Progerin +Vehicle
Progerin +SFN
C
Progerin +Vehicle
Progerin +SFN
PI
Annexin V-FITC
B1 3.1%
B2 5.8%
B3 58.4%
B4 32.8%
B1 1.6%
B2 3.9%
B3 74.1%
B4 20.4%
Apoptotic cells (%)
60
40
20
0
**
Progerin +Vehicle
Progerin +SFN
D
Progerin +Vehicle
Progerin +SFN
2 μm
500 nm
E
Mitotracker
Progerin +Vehicle
Progerin +SFN
20 μm

Figure S5. SFN attenuates progerin-induced cell death and mitochondrial deformation. (A) Frequency of senescent (SA-β-gal-positive; blue) cells in vehicle-treated progerin-expressing rat NP cells (progerin+vehicle) and SFN-treated progerin-expressing rat NP cells (progerin+SFN); n = 5; $^{**}P < 0.01$. **(B)** Numbers of TUNEL-positive cells in the progerin+vehicle and progerin+SFN groups; n = 5; $^{**}P < 0.01$. Nuclei were stained with DAPI (blue). (**C**) Representative flow cytometry dot plots of apoptosis after Annexin V-FITC/PI dual staining. The relative number of apoptotic cells was smaller in the progerin+SFN compared to the progerin+vehicle group; n = 5; $^{*}P < 0.05$. **(D)** Representative TEM images of the mitochondria of NP cells from the vehicle-treated and SFN-treated progerin groups. **(E)** Representative fluorescence images of mitochondria in NP cells from the progerin+vehicle and progerin+SFN groups. TUNEL, terminal deoxynucleotidyl transferase-mediated dUTP nick-end labelling. Data represent mean ± SEM.