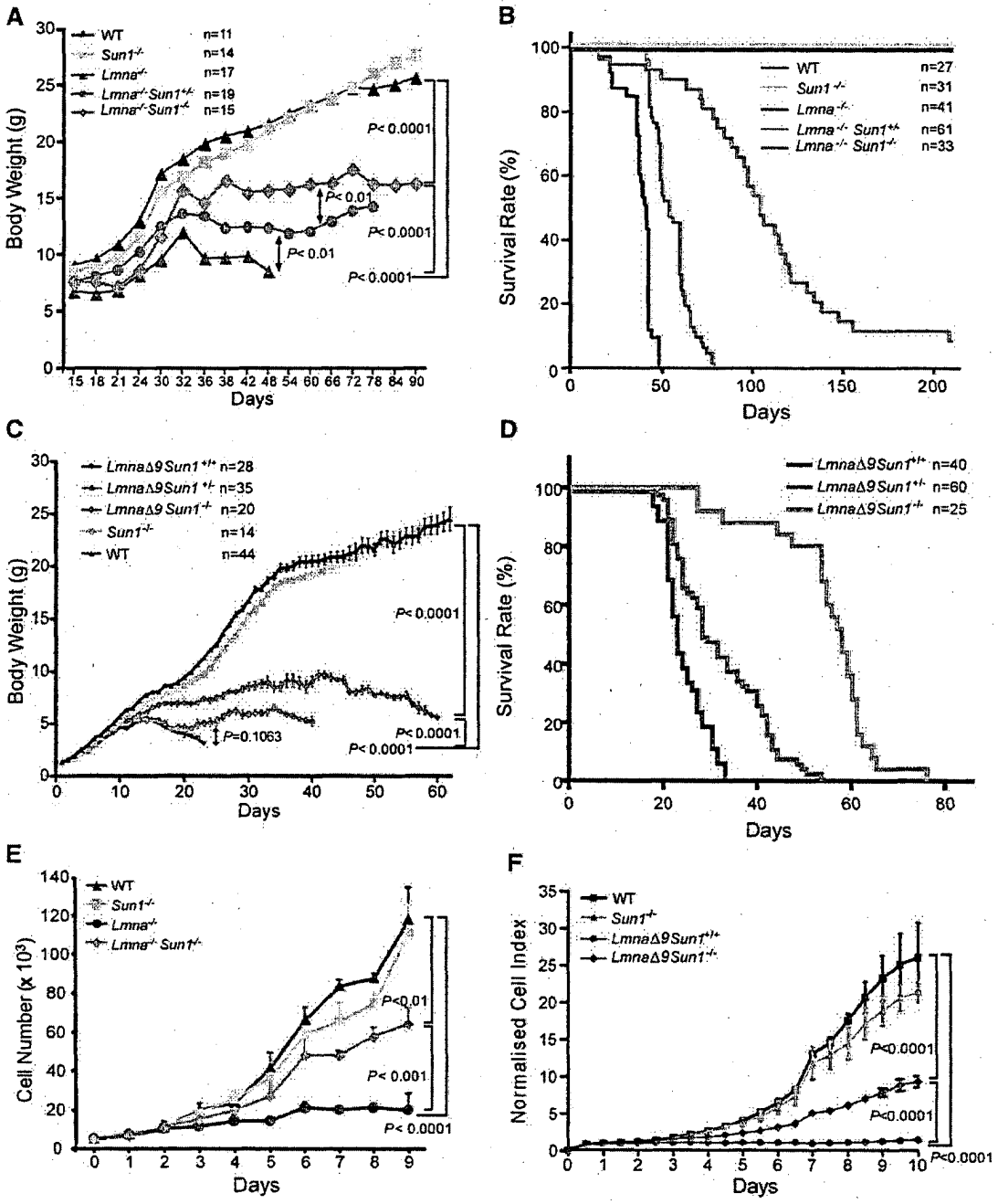

A
Body Weight (g)
Days
WT n=11
Sun1-/- n=14
Lmna-/- n=17
Lmna-/-Sun1+/- n=19
Lmna-/-Sun1-/- n=15
P< 0.0001
P< 0.01
P< 0.0001
P< 0.01
P< 0.0001
B
Survival Rate (%)
Days
WT n=27
Sun1-/- n=31
Lmna-/- n=41
Lmna-/- Sun1+/+ n=61
Lmna-/- Sun1-/- n=33
C
Body Weight (g)
Days
LmnaΔ9Sun1+/+ n=28
LmnaΔ9Sun1+/- n=35
LmnaΔ9Sun1-/- n=20
Sun1-/- n=14
WT n=44
P< 0.0001
P=0.1063
P< 0.0001
P< 0.0001
D
Survival Rate (%)
Days
LmnaΔ9Sun1+/+ n=40
LmnaΔ9Sun1+/- n=60
LmnaΔ9Sun1-/- n=25
E
Cell Number (x 10³)
Days
WT
Sun1-/-
Lmna-/-
Lmna-/- Sun1-/-
P<0.01
P< 0.001
P< 0.0001
F
Normalised Cell Index
Days
WT
Sun1-/-
LmnaΔ9Sun1+/+
LmnaΔ9Sun1-/-
P<0.0001
P<0.0001
P<0.0001

FIGURE 1

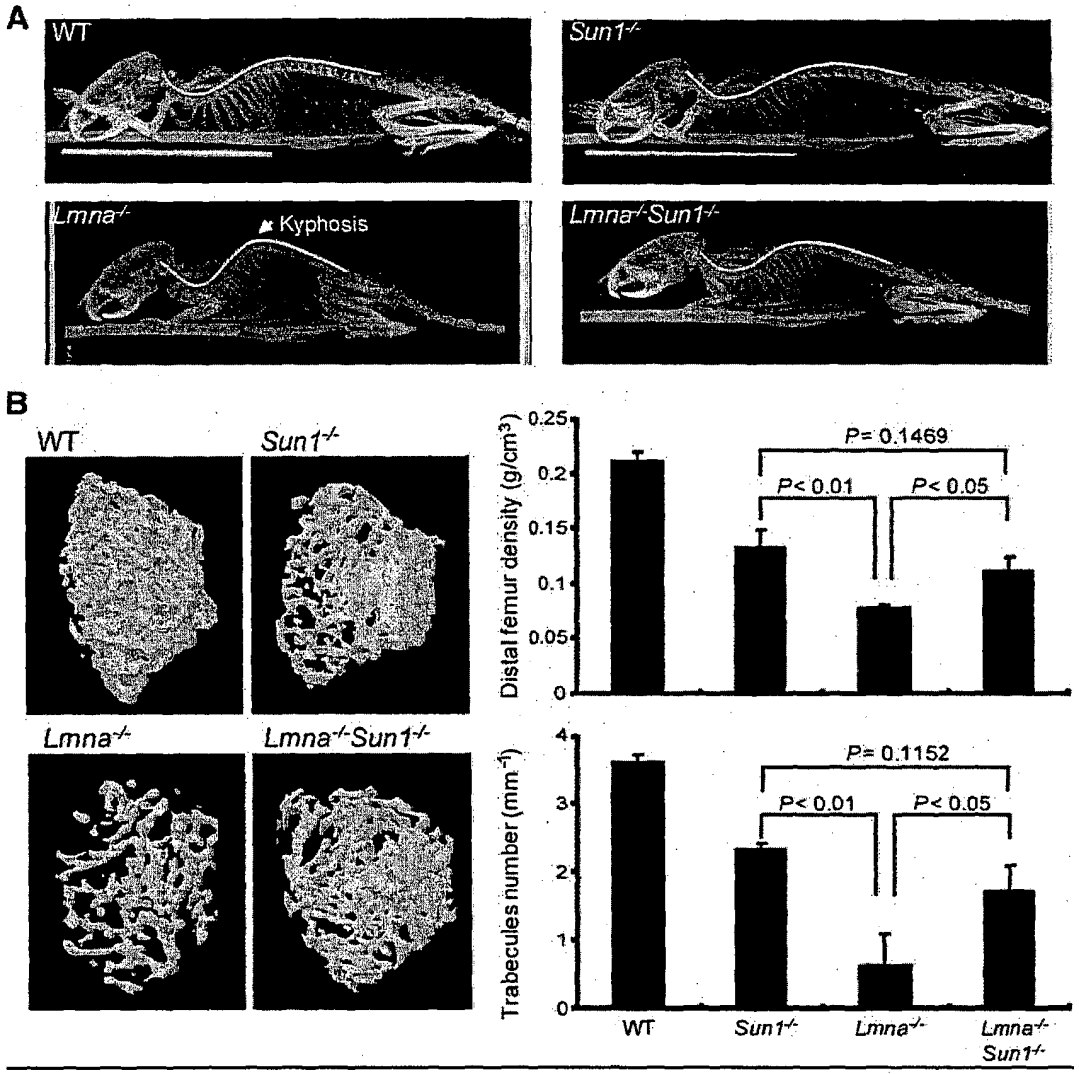

A
WT
Sun1-/-
Lmna-/-
Kyphosis
Lmna-/-Sun1-/-
B
WT
Sun1-/-
Lmna-/-
Lmna-/-Sun1-/-
Distal femur density (g/cm3)
0.25
0.2
0.15
0.1
0.05
0
P= 0.1469
P< 0.01
P< 0.05
Trabecules number (mm-1)
4
3
2
1
0
P= 0.1152
P< 0.01
P< 0.05
WT
Sun1-/-
Lmna-/-
Lmna-/-
Sun1-/-

FIGURE 2

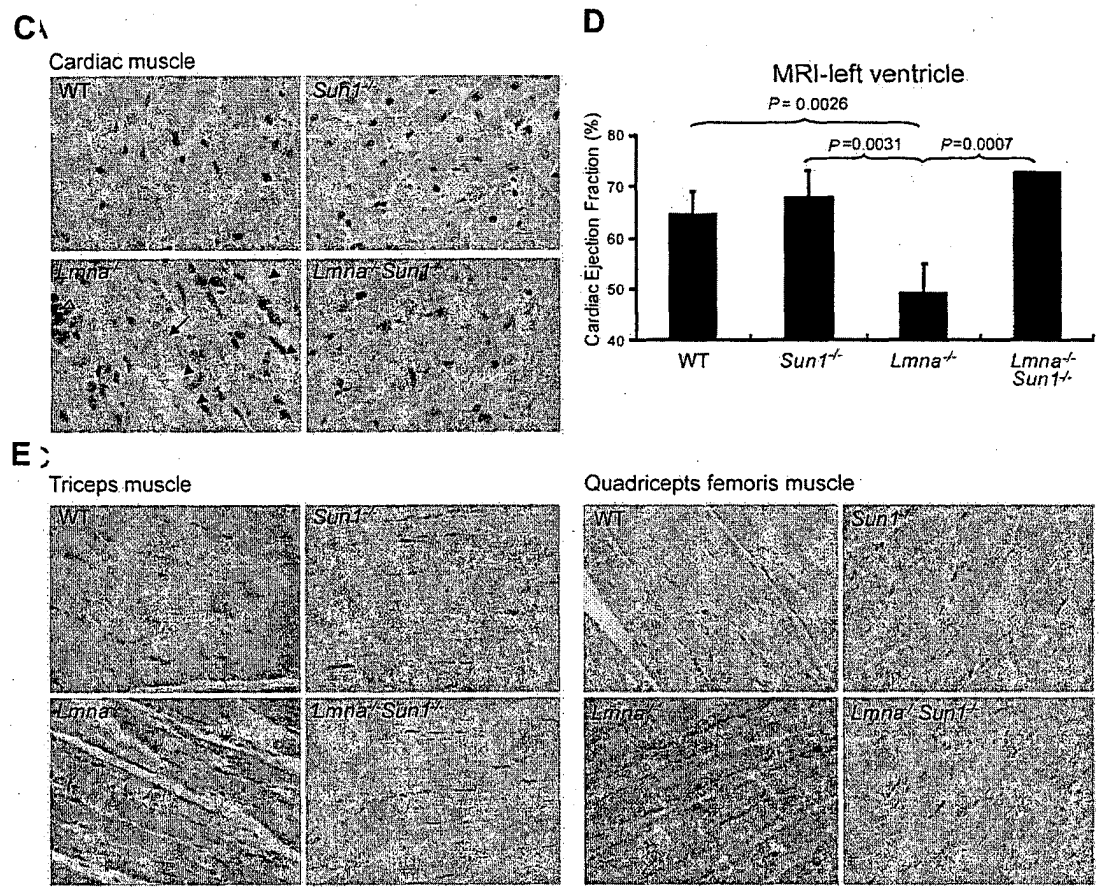

C
Cardiac muscle
WT
Sun1-/-
Lmna-/-
Lmna-/- Sun1-/-
D
MRI-left ventricle
P= 0.0026
P=0.0031
P=0.0007
Cardiac Ejection Fraction (%)
80
70
60
50
40
WT
Sun1-/-
Lmna-/-
Lmna-/- Sun1-/-
E
Triceps muscle
Quadricepts femoris muscle
WT
Sun1-/-
Lmna-/-
Lmna-/- Sun1-/-

FIGURE 2

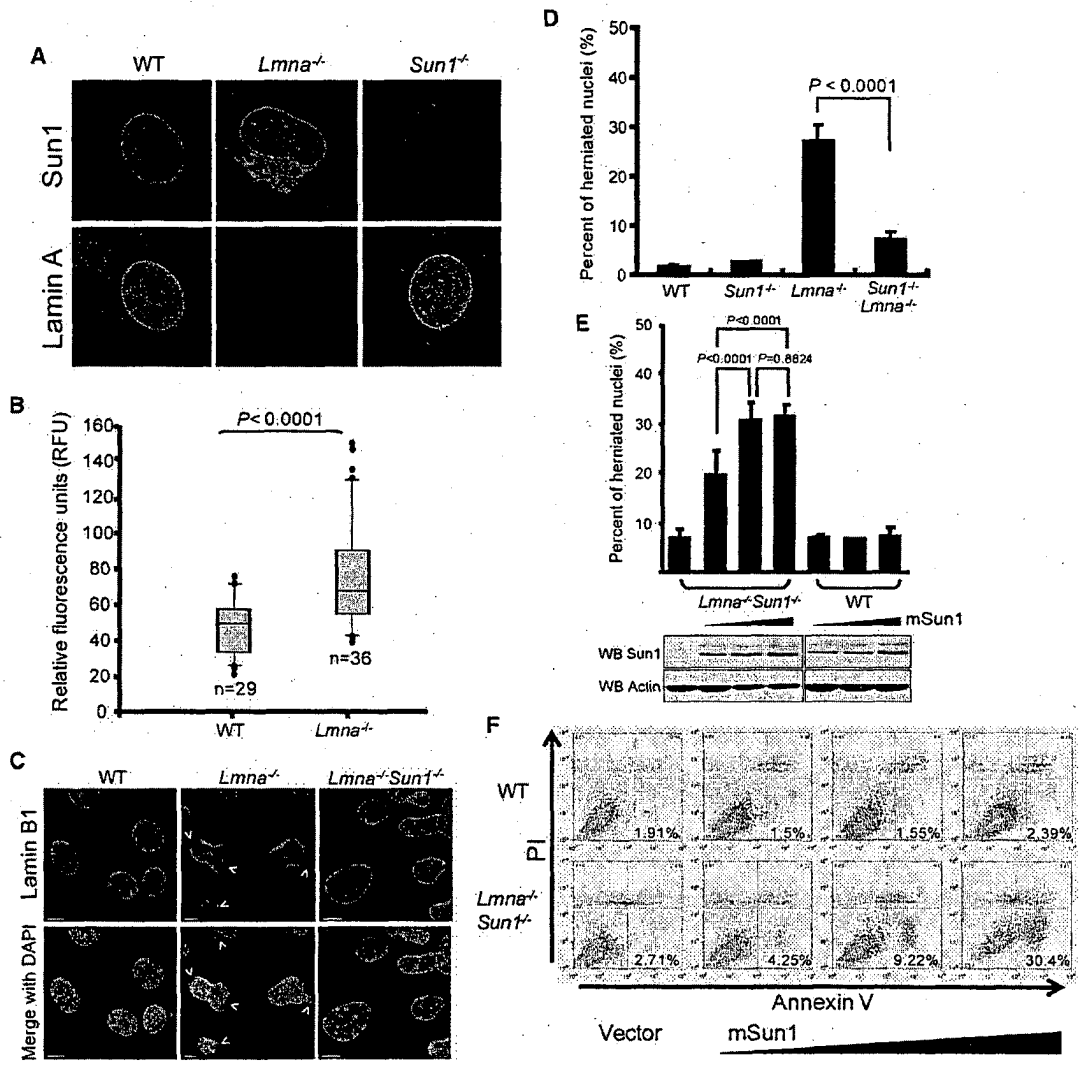

A
WT
Lmna-/-
Sun1-/-
Sun1
Lamin A
B
Relative fluorescence units (RFU)
P< 0.0001
n=29
n=36
WT
Lmna-/-
C
WT
Lmna-/-
Lmna-/-Sun1-/-
Lamin B1
Merge with DAPI
D
Percent of herniated nuclei (%)
P < 0.0001
WT
Sun1-/-
Lmna-/-
Sun1-/- Lmna-/-
E
Percent of herniated nuclei (%)
P<0.0001
P<0.0001
P=0.8624
Lmna-/-Sun1-/-
WT
mSun1
WB Sun1
WB Actin
F
WT
Lmna-/- Sun1-/-
PI
1.91%
1.5%
1.55%
2.39%
2.71%
4.25%
9.22%
30.4%
Annexin V
Vector
mSun1

FIGURE 3

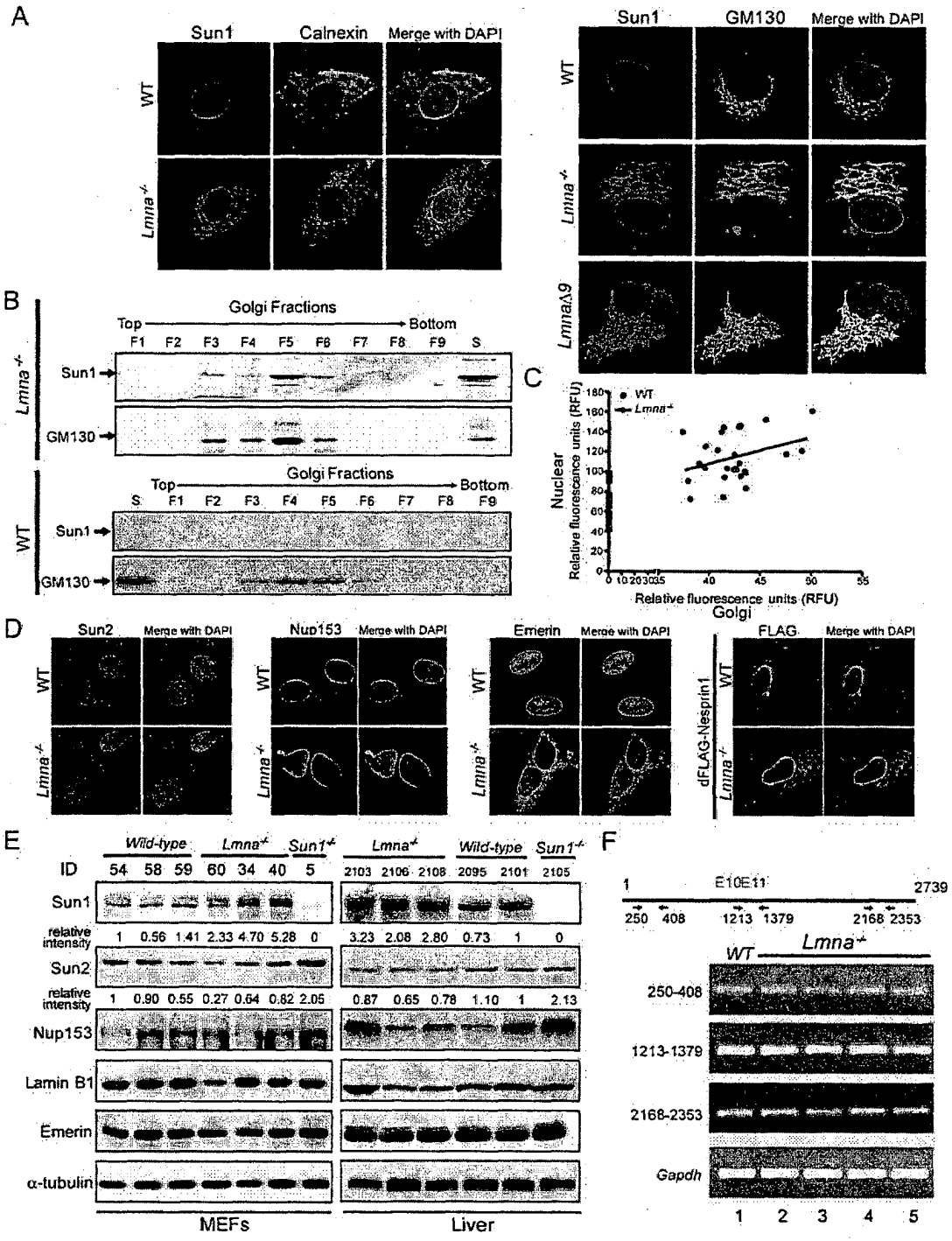

A
Sun1
Calnexin
Merge with DAPI
WT
Lmna-/-
Sun1
GM130
Merge with DAPI
LmnaΔ9
B
Golgi Fractions
Top
Bottom
F1 F2 F3 F4 F5 F6 F7 F8 F9 S
Sun1
GM130
C
WT
Lmna-/-
Nuclear Relative fluorescence units (RFU)
Relative fluorescence units (RFU)
Golgi
D
Sun2
Nup153
Emerin
FLAG
Merge with DAPI
dFLAG-Nesprin1
E
Wild-type
Lmna-/-
Sun1-/-
ID
54 58 59 60 34 40 5
2103 2106 2108 2095 2101 2105
Sun1
relative intensity
1 0.56 1.41 2.33 4.70 5.28 0
3.23 2.08 2.80 0.73 1 0
Sun2
relative intensity
1 0.90 0.55 0.27 0.64 0.82 2.05
0.87 0.65 0.78 1.10 1 2.13
Nup153
Lamin B1
Emerin
α-tubulin
MEFs
Liver
F
1
E10E11
2739
250 408 1213 1379 2168 2353
WT
Lmna-/-
250-408
1213-1379
2168-2353
Gapdh
1 2 3 4 5

FIGURE 4

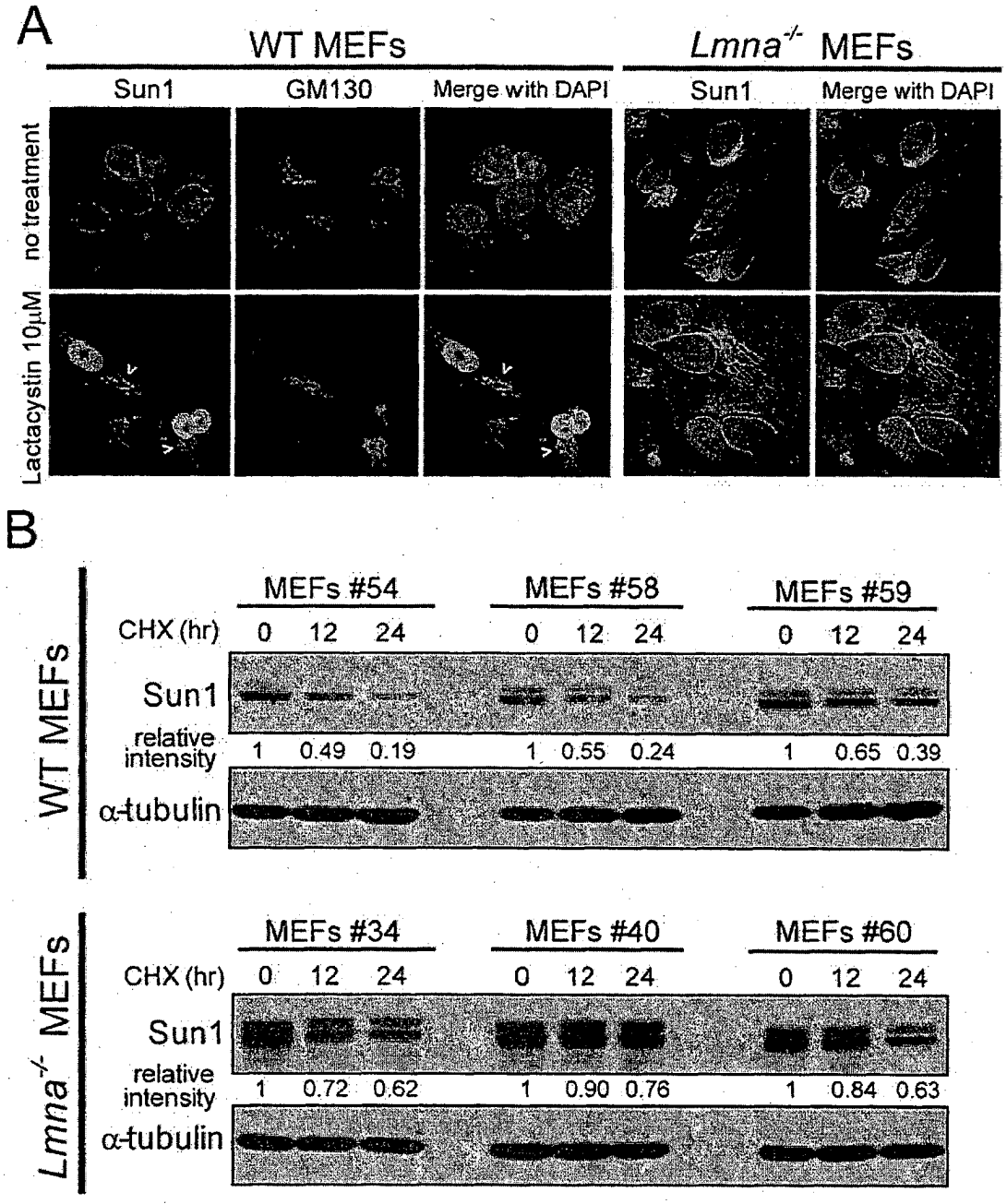

A
WT MEFs
Lmna-/- MEFs
Sun1
GM130
Merge with DAPI
Sun1
Merge with DAPI
no treatment
Lactacystin 10μM
B
WT MEFs
MEFs #54
MEFs #58
MEFs #59
CHX (hr)
0 12 24 0 12 24 0 12 24
Sun1
relative intensity
1 0.49 0.19 1 0.55 0.24 1 0.65 0.39
α-tubulin
Lmna-/- MEFs
MEFs #34
MEFs #40
MEFs #60
CHX (hr)
0 12 24 0 12 24 0 12 24
Sun1
relative intensity
1 0.72 0.62 1 0.90 0.76 1 0.84 0.63
α-tubulin

FIGURE 5

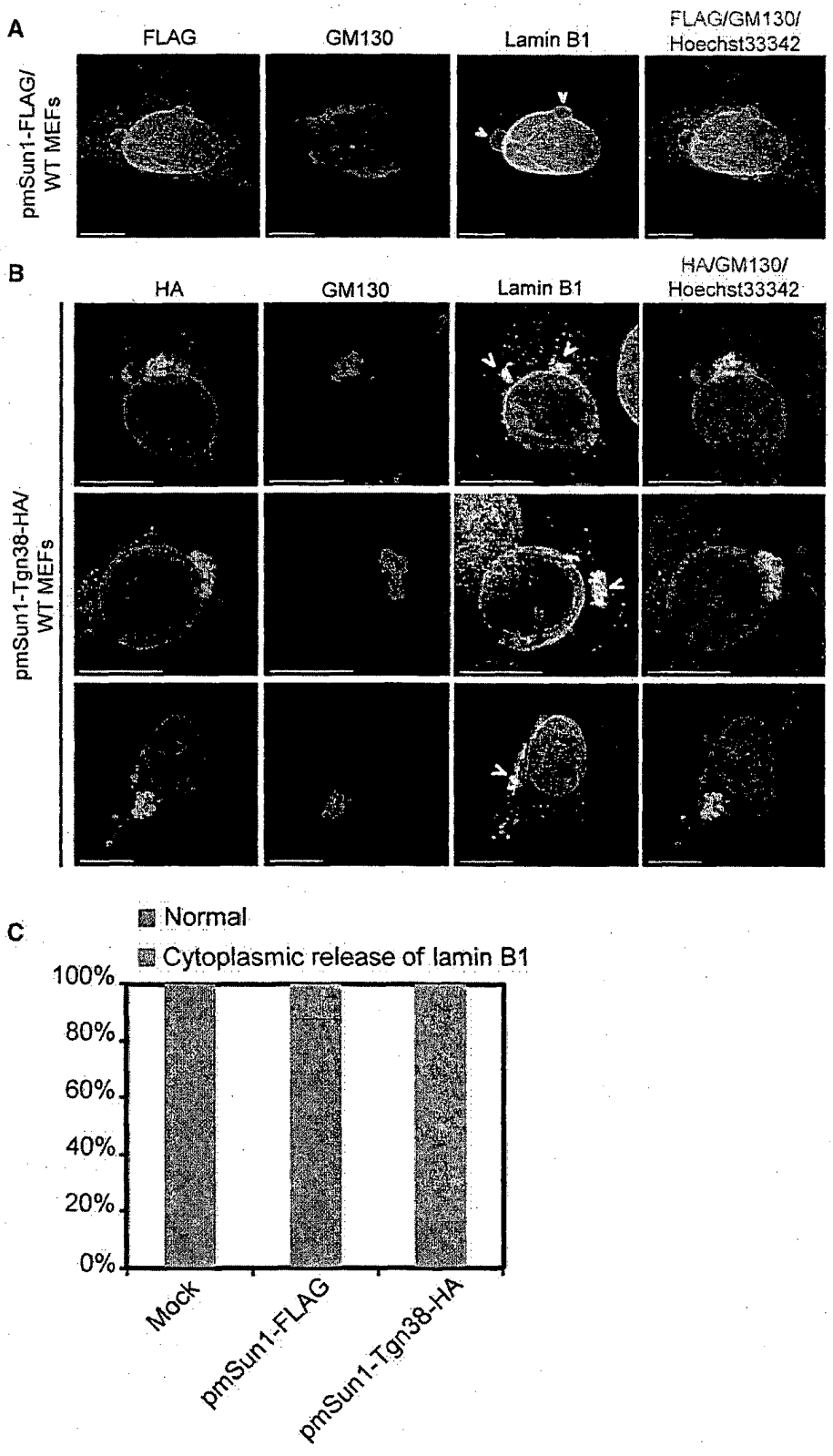

A
FLAG
GM130
Lamin B1
FLAG/GM130/
Hoechst33342
pmSun1-FLAG/
WT MEFs
B
HA
GM130
Lamin B1
HA/GM130/
Hoechst33342
pmSun1-Tgn38-HA/
WT MEFs
C
Normal
Cytoplasmic release of lamin B1
100%
80%
60%
40%
20%
0%
Mock
pmSun1-FLAG
pmSun1-Tgn38-HA

FIGURE 6

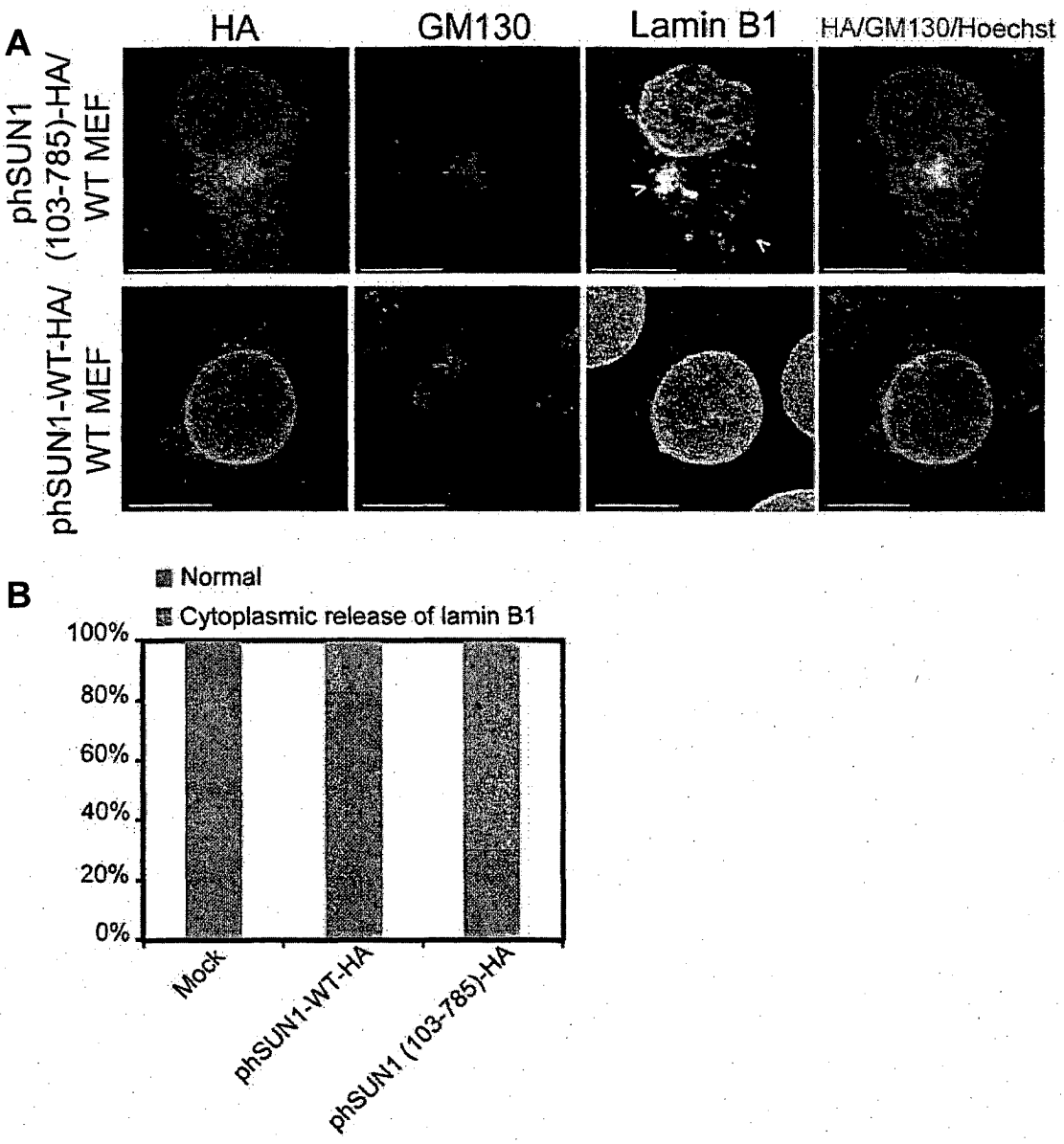

A
HA
GM130
Lamin B1
HA/GM130/Hoechst
phSUN1 (103-785)-HA/ WT MEF
phSUN1-WT-HA/ WT MEF
B
Normal
Cytoplasmic release of lamin B1
100%
80%
60%
40%
20%
0%
Mock
phSUN1-WT-HA
phSUN1 (103-785)-HA

FIGURE 7

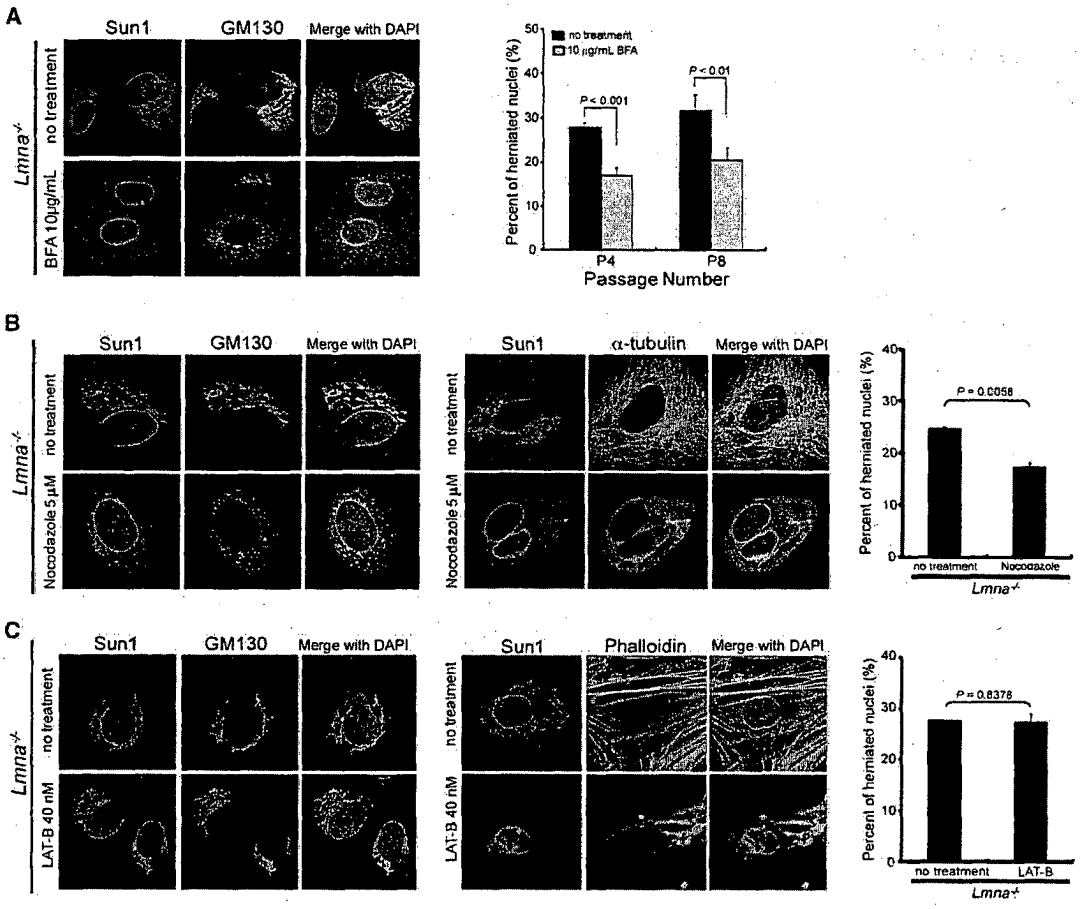
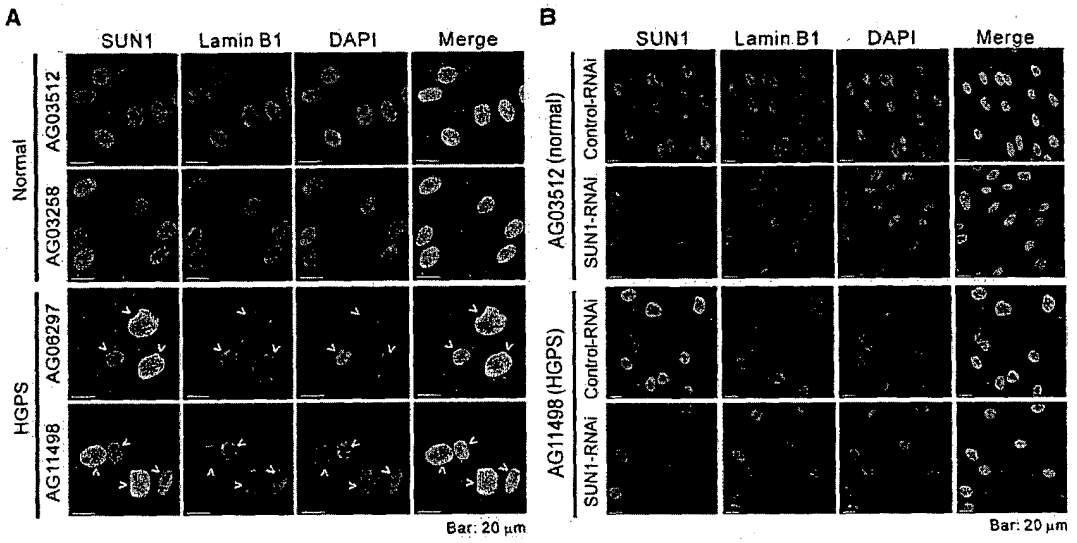
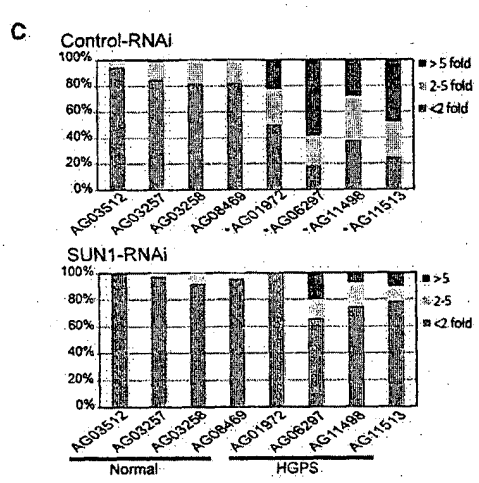
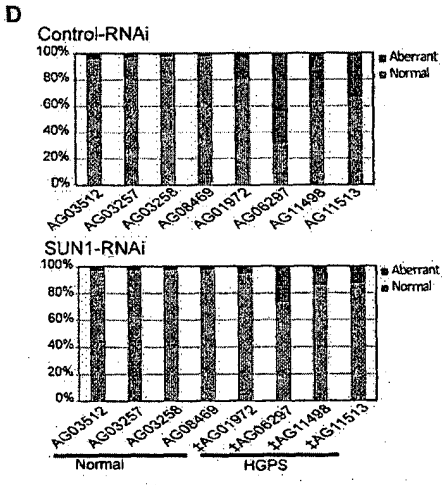

A
Sun1
GM130
Merge with DAPI
Lmna-/-
no treatment
BFA 10μg/mL
no treatment
10 μg/mL BFA
P < 0.001
P < 0.01
Percent of herniated nuclei (%)
P4
P8
Passage Number
B
Sun1
GM130
Merge with DAPI
Sun1
α-tubulin
Merge with DAPI
Nocodazole 5 μM
P = 0.0058
no treatment
Nocodazole
Lmna-/-
C
Sun1
GM130
Merge with DAPI
Sun1
Phalloidin
Merge with DAPI
LAT-B 40 nM
P = 0.8378
no treatment
LAT-B
Lmna-/-

FIGURE 8


A
SUN1
Lamin B1
DAPI
Merge
Normal
AG03512
AG03258
HGPS
AG06297
AG11498
Bar: 20 μm
B
SUN1
Lamin B1
DAPI
Merge
AG03512 (normal)
Control-RNAi
SUN1-RNAi
AG11498 (HGPS)
Control-RNAi
SUN1-RNAi
Bar: 20 μm


C
Control-RNAi
100%
80%
60%
40%
20%
0%
>5 fold
2-5 fold
<2 fold
AG03512
AG03257
AG03258
AG08469
*AG01972
*AG06297
*AG11498
*AG11513
SUN1-RNAi
>5
2-5
<2 fold
Normal
HGPS


D
Control-RNAi
Aberrant
Normal
SUN1-RNAi
‡AG01972
‡AG06297
‡AG11498
‡AG11513
Normal
HGPS

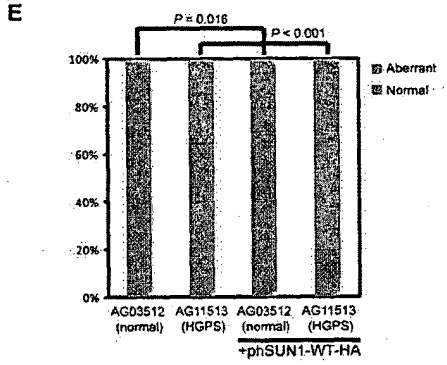

E
P = 0.016
P < 0.001
Aberrant
Normal
AG03512 (normal)
AG11513 (HGPS)
AG03512 (normal)
AG11513 (HGPS)
+phSUN1-WT-HA

FIGURE 9

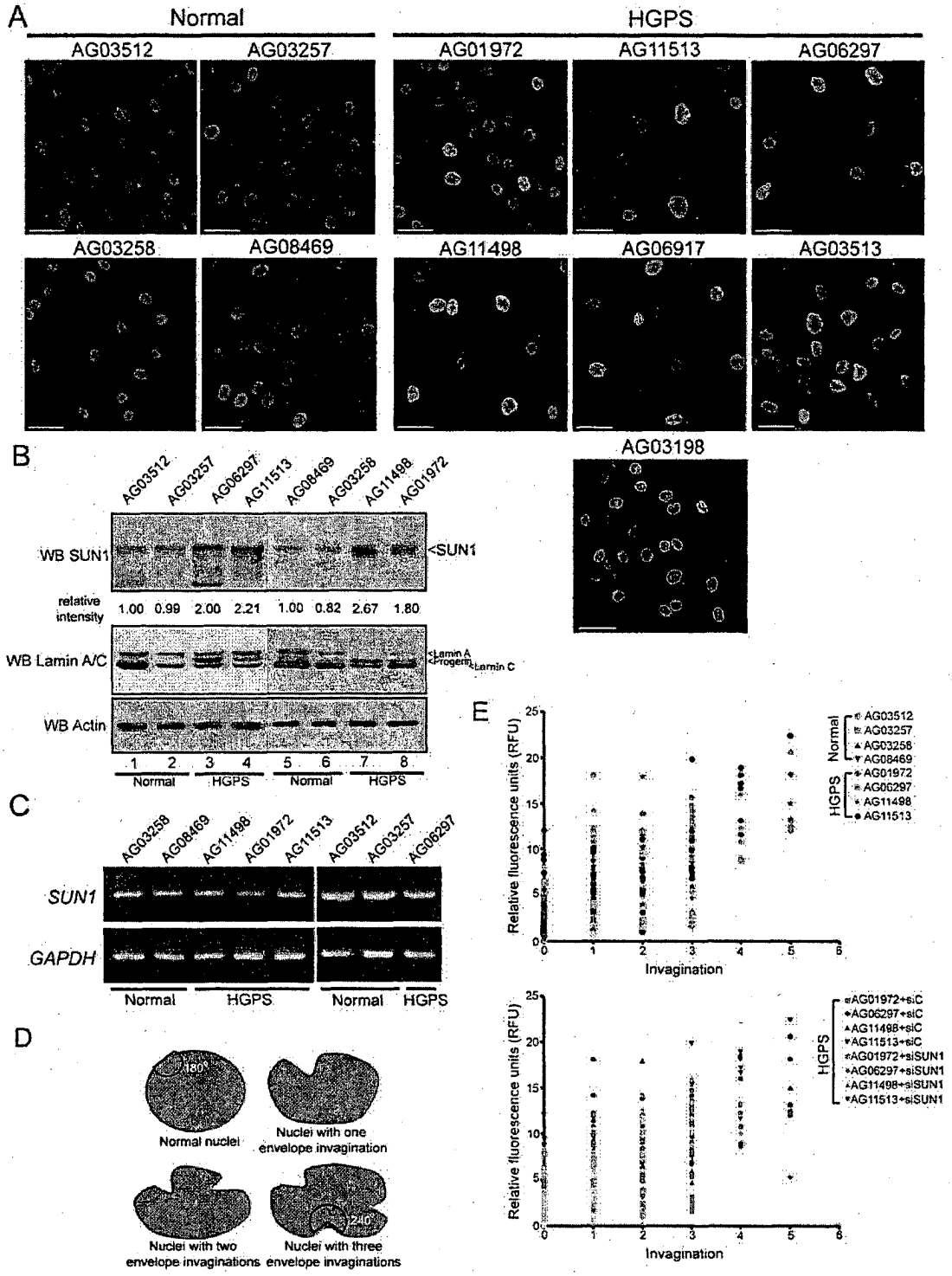

A
Normal
HGPS
AG03512
AG03257
AG01972
AG11513
AG06297
AG03258
AG08469
AG11498
AG06917
AG03513
B
AG03198
WB SUN1
<SUN1
relative intensity
1.00 0.99 2.00 2.21 1.00 0.82 2.67 1.80
WB Lamin A/C
<Lamin A
<Progerin
<Lamin C
WB Actin
1 2 3 4 5 6 7 8
Normal HGPS Normal HGPS
C
SUN1
GAPDH
Normal HGPS Normal HGPS
D
180°
240°
Normal nuclei
Nuclei with one envelope invagination
Nuclei with two envelope invaginations
Nuclei with three envelope invaginations
E
Relative fluorescence units (RFU)
Invagination
Normal
HGPS
AG03512
AG03257
AG03258
AG08469
AG01972
AG06297
AG11498
AG11513
AG01972+siC
AG06297+siC
AG11498+siC
AG11513+siC
AG01972+siSUN1
AG06297+siSUN1
AG11498+siSUN1
AG11513+siSUN1

FIGURE 10

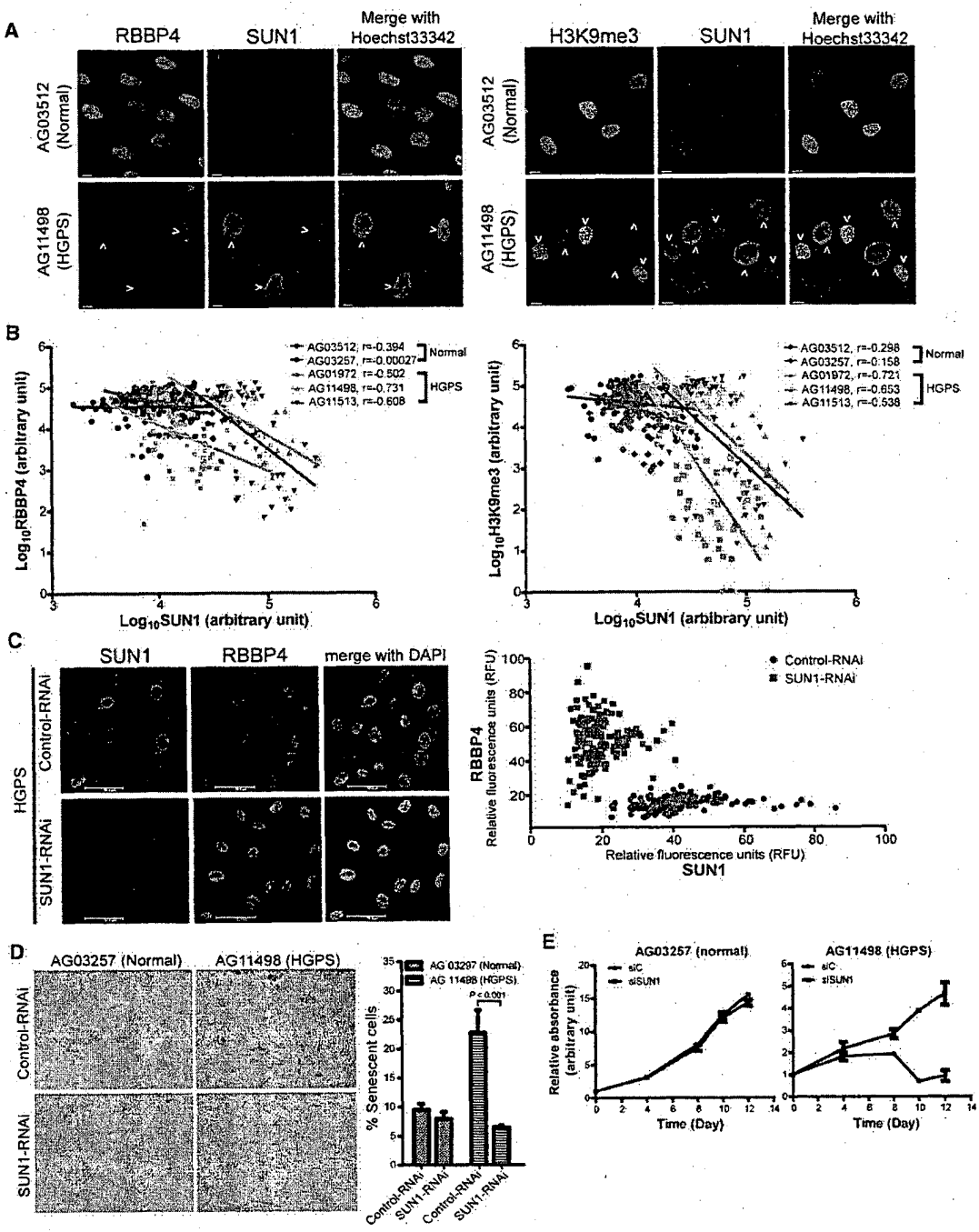

A
RBBP4
SUN1
Merge with Hoechst33342
H3K9me3
AG03512 (Normal)
AG11498 (HGPS)
B
AG03512, r=-0.394
AG03257, r=-0.00027
AG01972, r=-0.502
AG11498, r=-0.731
AG11513, r=-0.608
Normal
HGPS
Log10RBBP4 (arbitrary unit)
Log10SUN1 (arbitrary unit)
AG03512, r=-0.298
AG03257, r=-0.158
AG01972, r=-0.721
AG11498, r=-0.653
AG11513, r=-0.538
Log10H3K9me3 (arbitrary unit)
C
SUN1
RBBP4
merge with DAPI
HGPS
Control-RNAi
SUN1-RNAi
Relative fluorescence units (RFU)
RBBP4
SUN1
D
AG03257 (Normal)
AG11498 (HGPS)
% Senescent cells
P < 0.001
E
AG03257 (normal)
AG11498 (HGPS)
siC
siSUN1
Relative absorbance (arbitrary unit)
Time (Day)

FIGURE 11

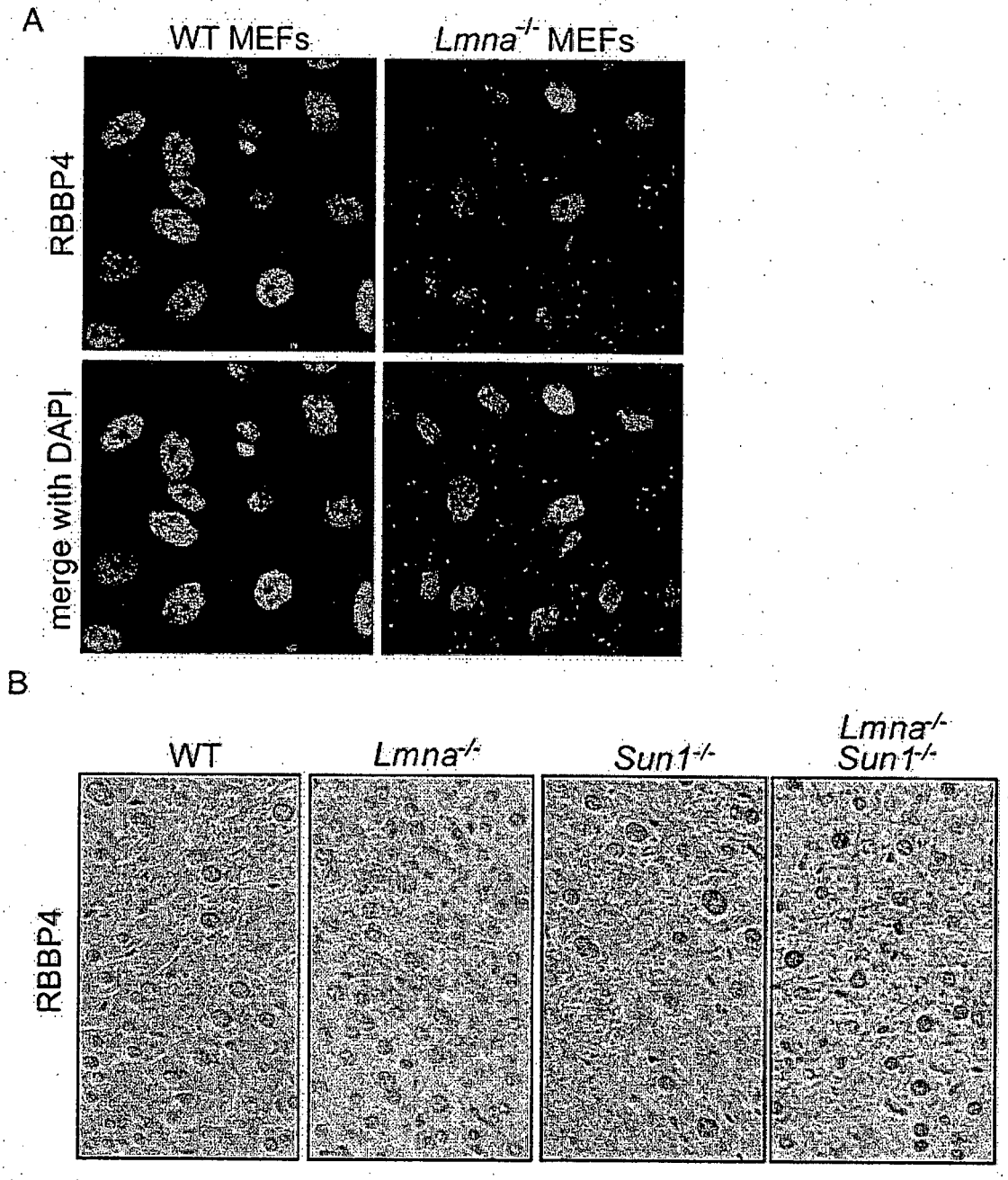

A
WT MEFs
Lmna-/- MEFs
RBBP4
merge with DAPI
B
WT
Lmna-/-
Sun1-/-
Lmna-/- Sun1-/-
RBBP4

FIGURE 12

A. Human Sun1-specific siRNAs that have been shown experimentally to deplete human cells of Sun1 protein.

Target Sequence (mRNA)	Designation	Sun1 depletion at 48h post transfection
GGUAACUGCUGGGCAUUUA	J-025277-05 UNC84A (SUN1)	++
GGUACCAGUUUGUUACUUU	J-025277-06 UNC84A (SUN1)	+++
GCGCUCAGUUCCAGCUAUU	J-025277-07 UNC84A (SUN1)	++
GAAAAGACCCGACGACACA	J-025277-08 UNC84A (SUN1)	++++
All four siRNA oligonucleotides combined		++++

B

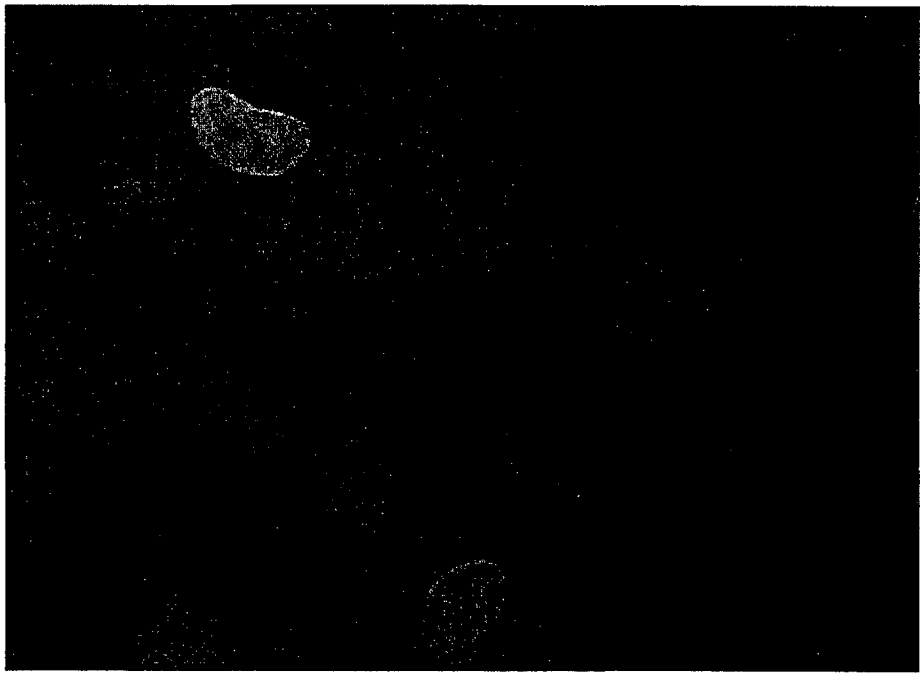


FIGURE 13