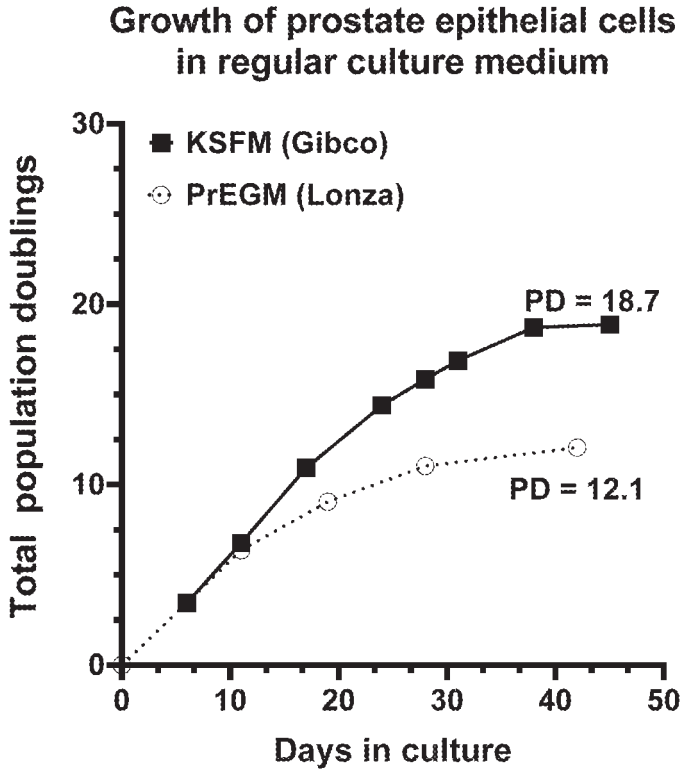
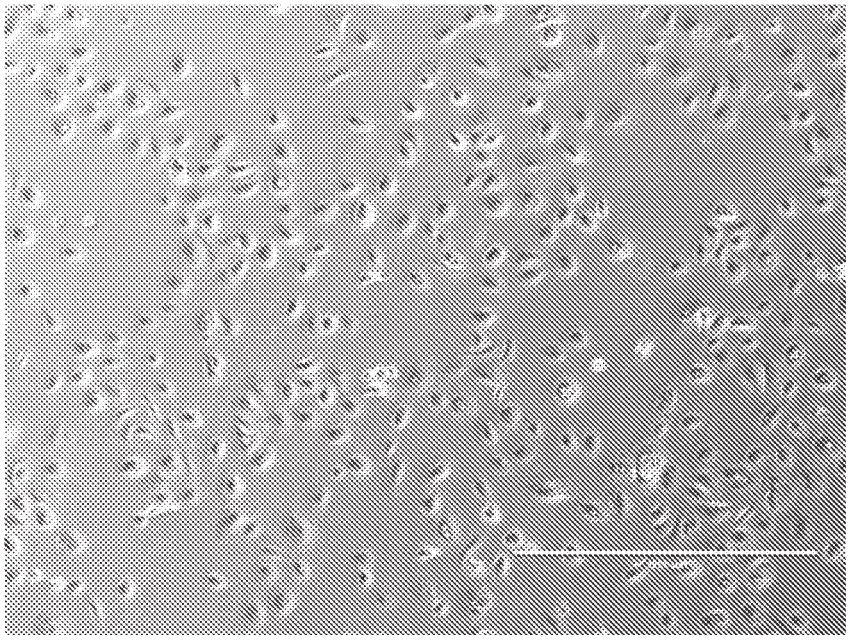
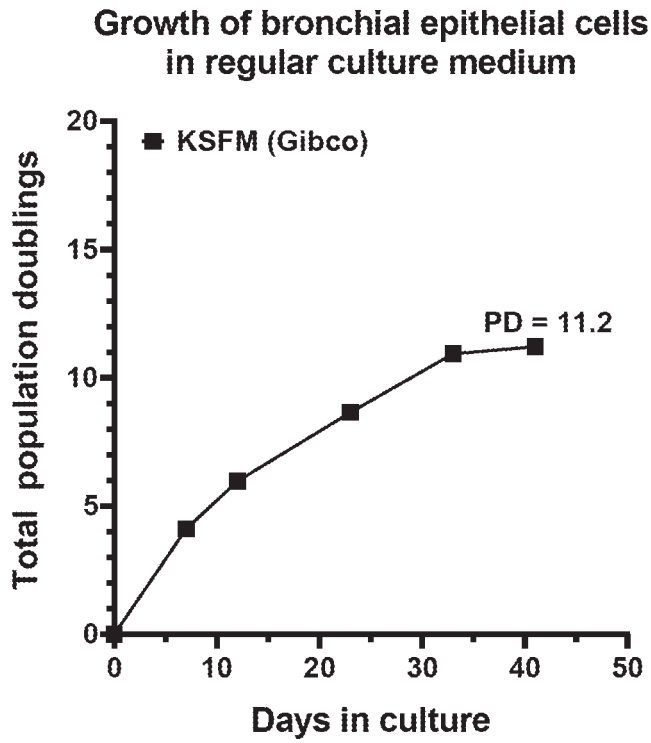
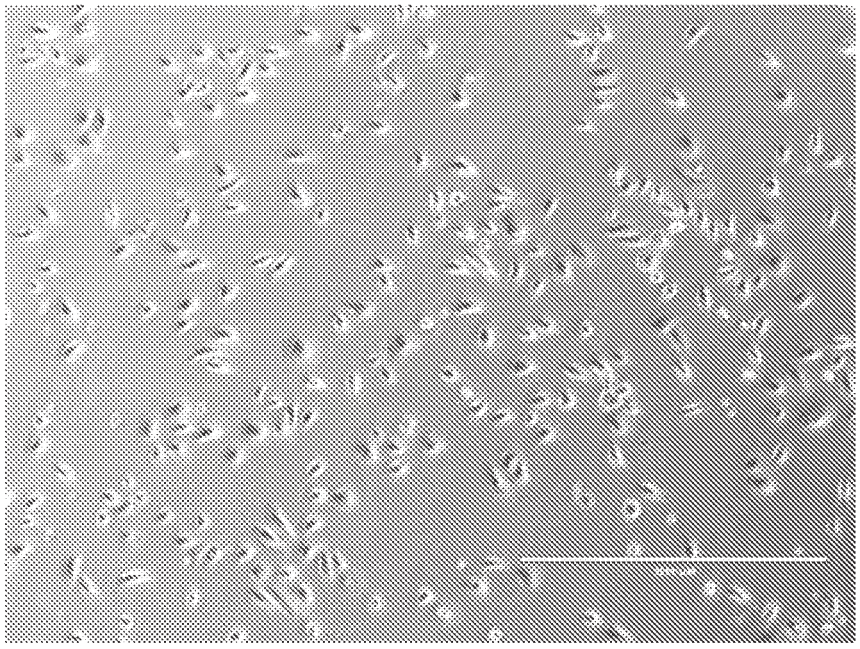




Prostate epithelial cells in KSFM at PD 18

Fig. 1





Bronchial epithelial cells in KSFM at PD 11

Fig. 2

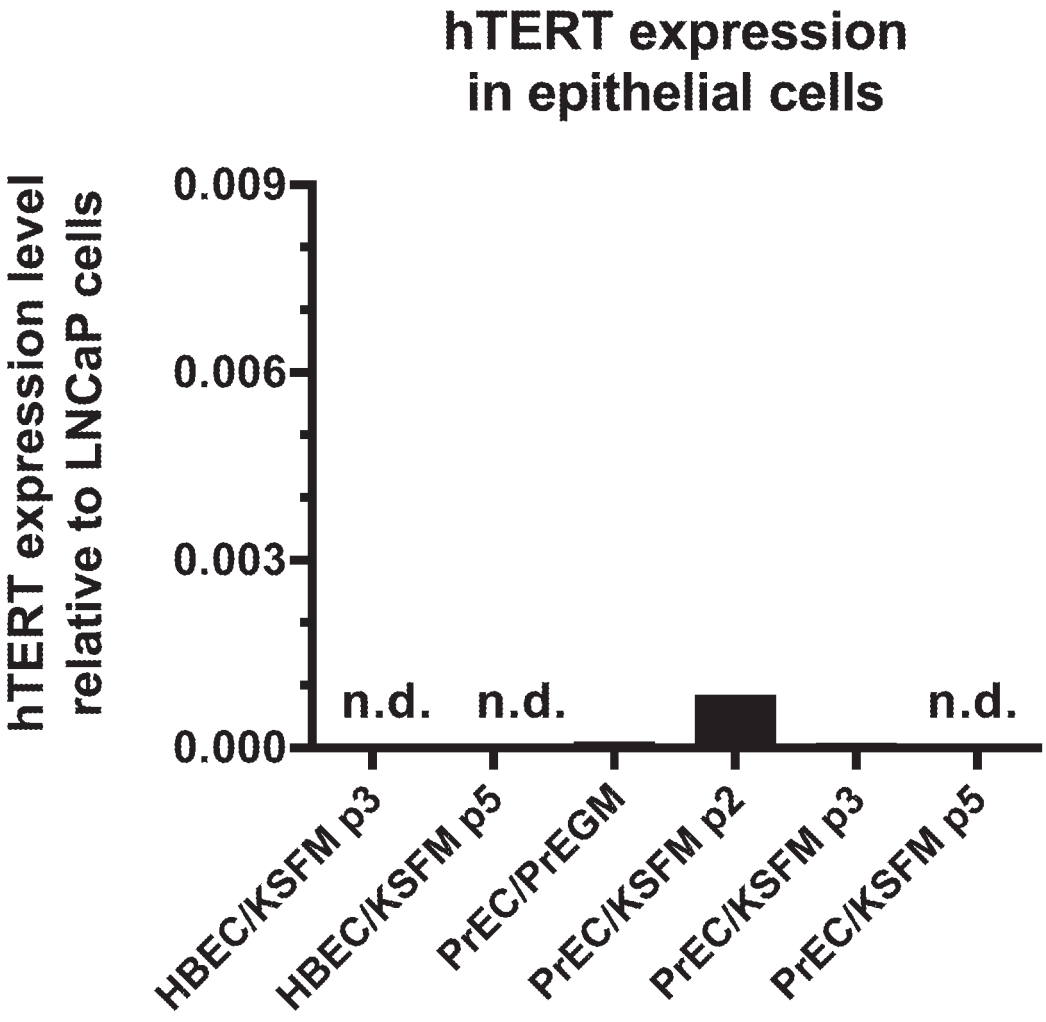

hTERT expression
in epithelial cells
hTERT expression level
relative to LNCaP cells
0.009
0.006
0.003
0.000
n.d.
n.d.
n.d.
HBEC/KSFM p3
HBEC/KSFM p5
PrEC/PrEGM
PrEC/KSFM p2
PrEC/KSFM p3
PrEC/KSFM p5

Fig. 3A

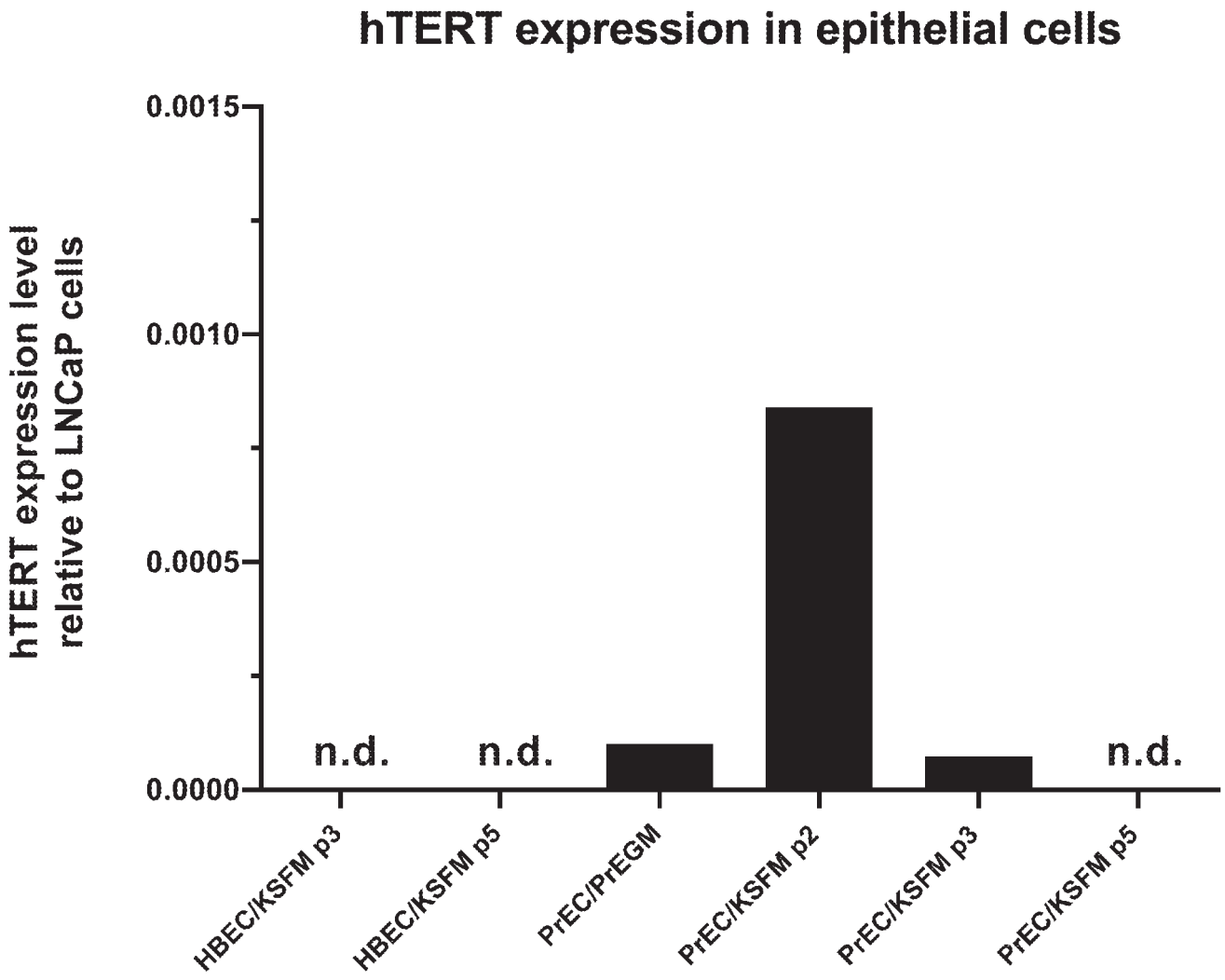

hTERT expression in epithelial cells
hTERT expression level relative to LNCaP cells
0.0015
0.0010
0.0005
0.0000
n.d.
n.d.
n.d.
HBEC/KSFM p3
HBEC/KSFM p5
PrEC/PrEGM
PrEC/KSFM p2
PrEC/KSFM p3
PrEC/KSFM p5

Fig. 3B

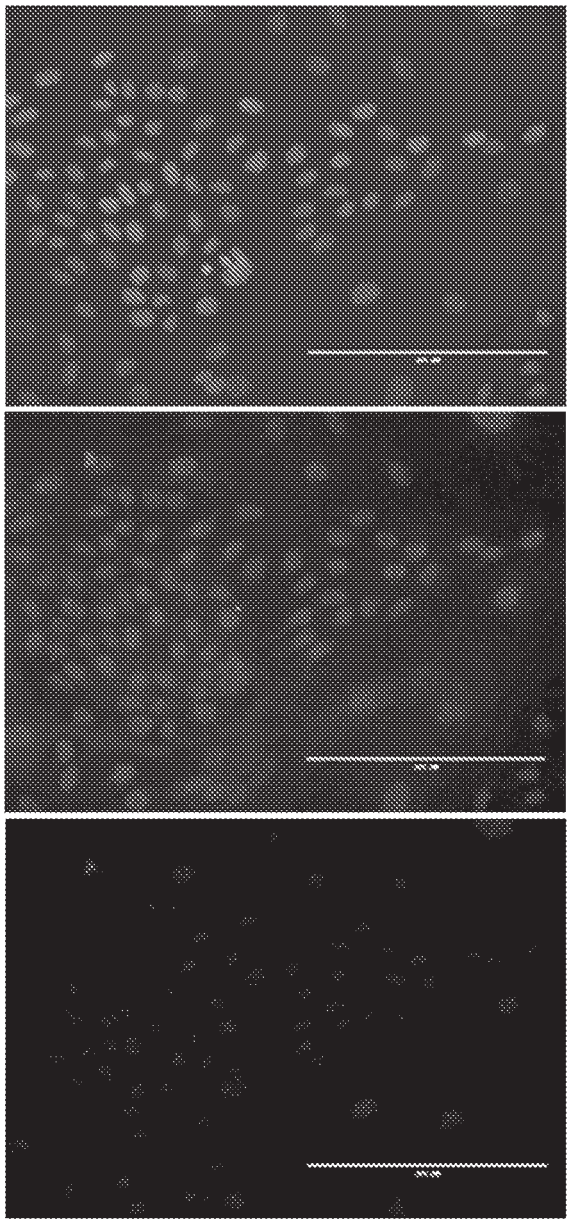


Fig. 4

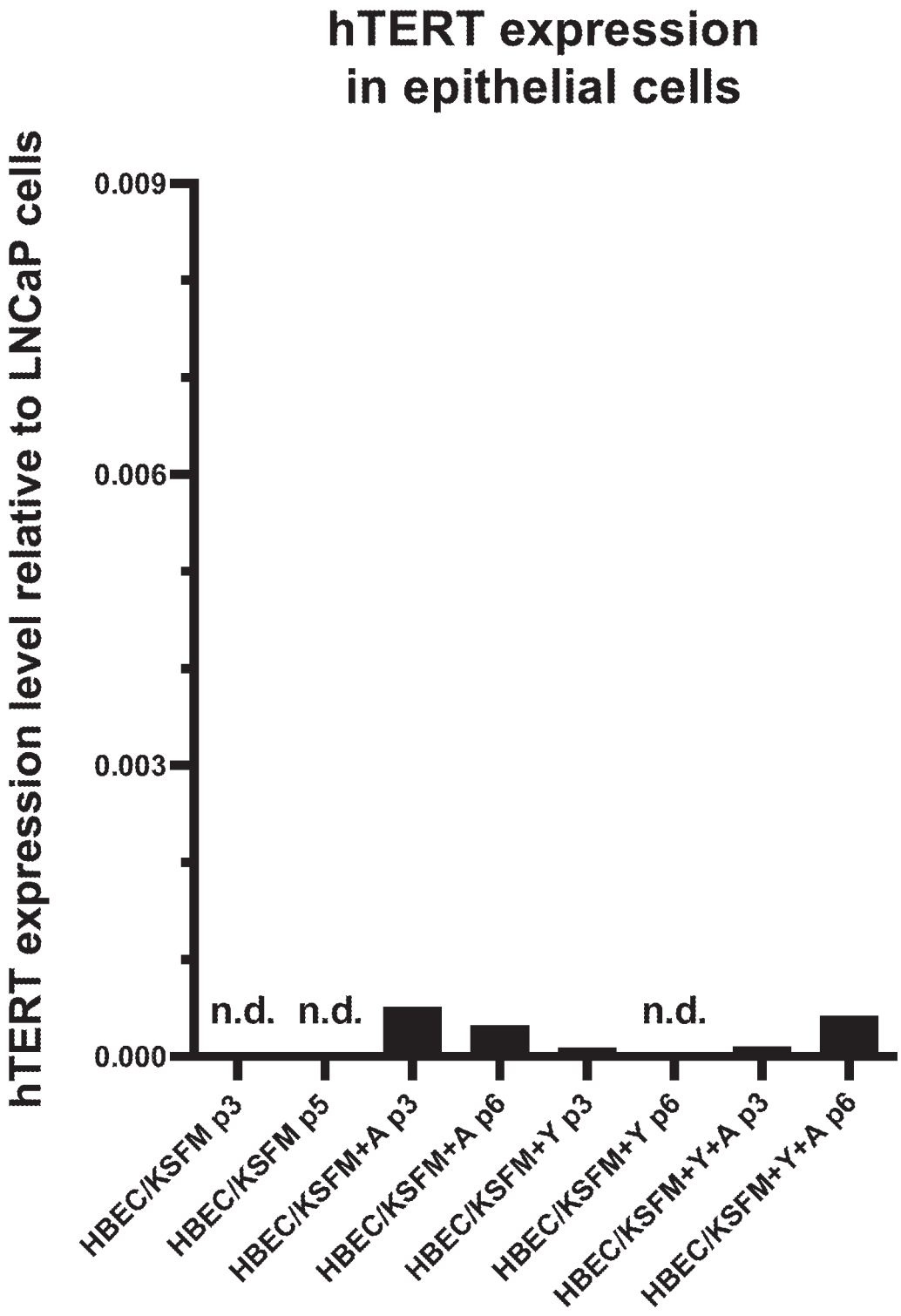

hTERT expression
in epithelial cells
hTERT expression level relative to LNCaP cells
0.009
0.006
0.003
0.000
n.d.
n.d.
n.d.
HBEC/KSFM p3
HBEC/KSFM p5
HBEC/KSFM+A p3
HBEC/KSFM+A p6
HBEC/KSFM+Y p3
HBEC/KSFM+Y p6
HBEC/KSFM+Y+A p3
HBEC/KSFM+Y+A p6

Fig. 5A

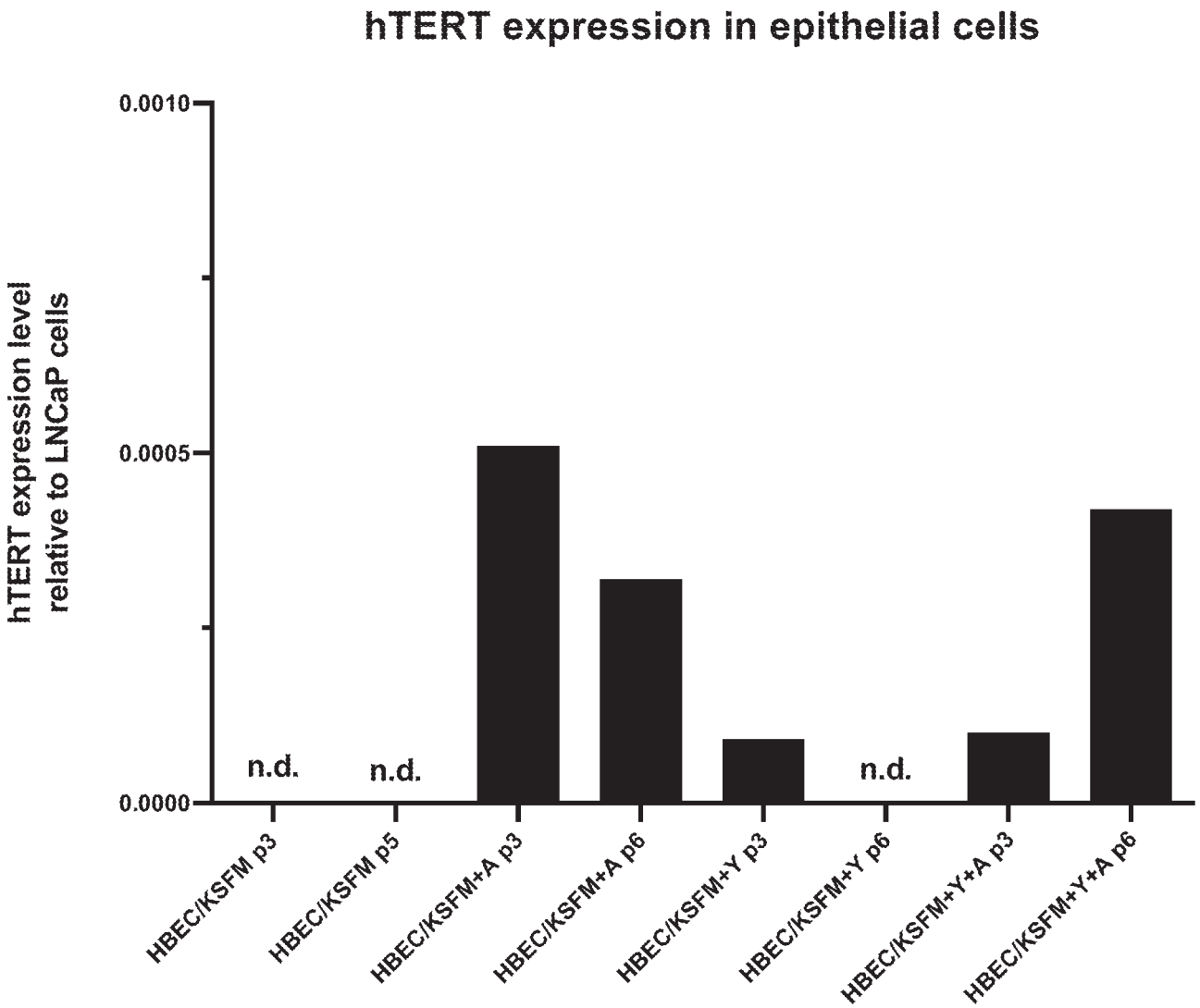

hTERT expression in epithelial cells
hTERT expression level relative to LNCaP cells
0.0010
0.0005
0.0000
n.d.
n.d.
n.d.
HBEC/KSFM p3
HBEC/KSFM p5
HBEC/KSFM+A p3
HBEC/KSFM+A p6
HBEC/KSFM+Y p3
HBEC/KSFM+Y p6
HBEC/KSFM+Y+A p3
HBEC/KSFM+Y+A p6

Fig. 5B

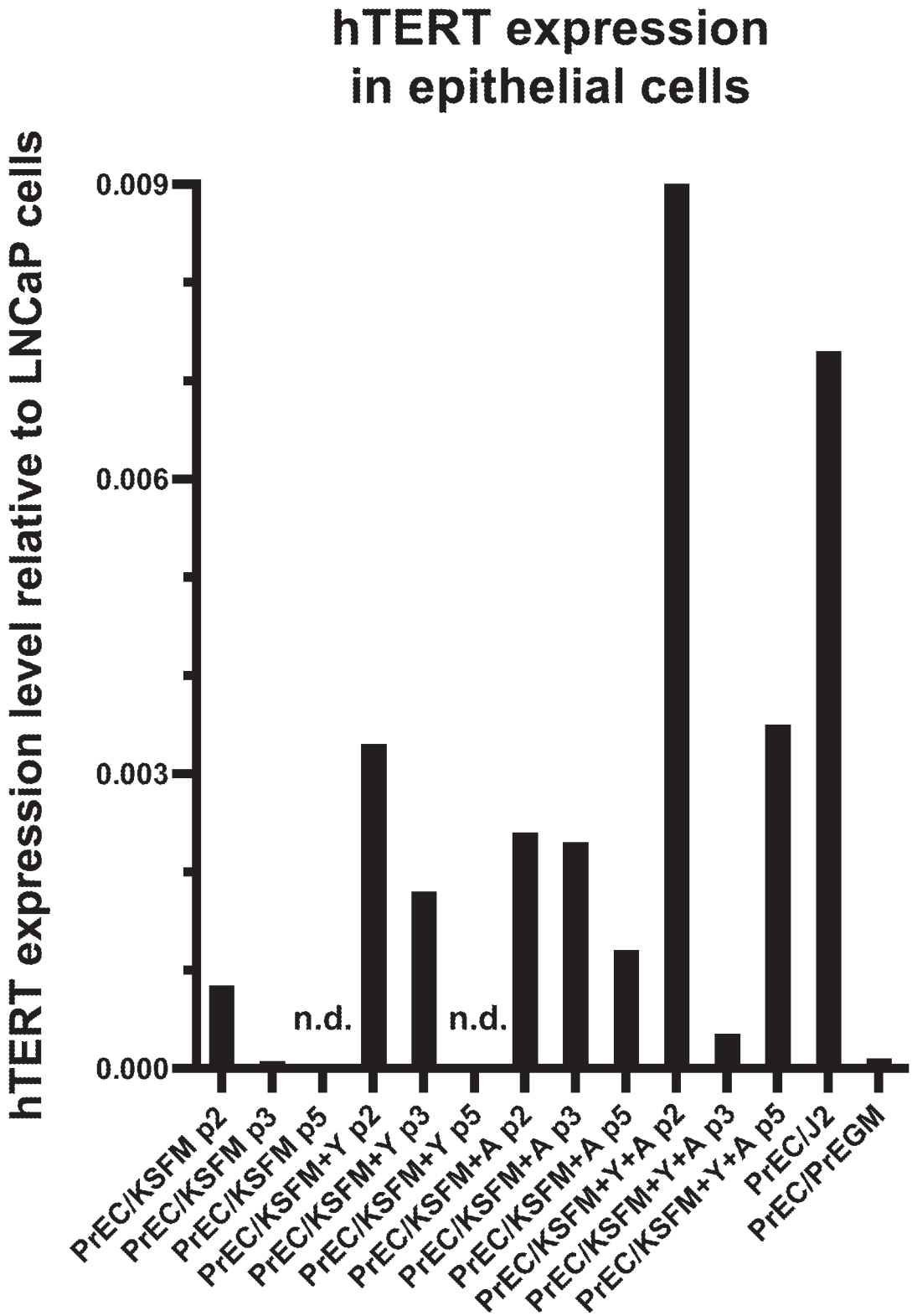

hTERT expression
in epithelial cells
hTERT expression level relative to LNCaP cells
0.009
0.006
0.003
0.000
n.d.
n.d.
PrEC/KSFM p2
PrEC/KSFM p3
PrEC/KSFM p5
PrEC/KSFM+Y p2
PrEC/KSFM+Y p3
PrEC/KSFM+Y p5
PrEC/KSFM+A p2
PrEC/KSFM+A p3
PrEC/KSFM+A p5
PrEC/KSFM+Y+A p2
PrEC/KSFM+Y+A p3
PrEC/KSFM+Y+A p5
PrEC/J2
PrEC/PrEGM

Fig. 6A

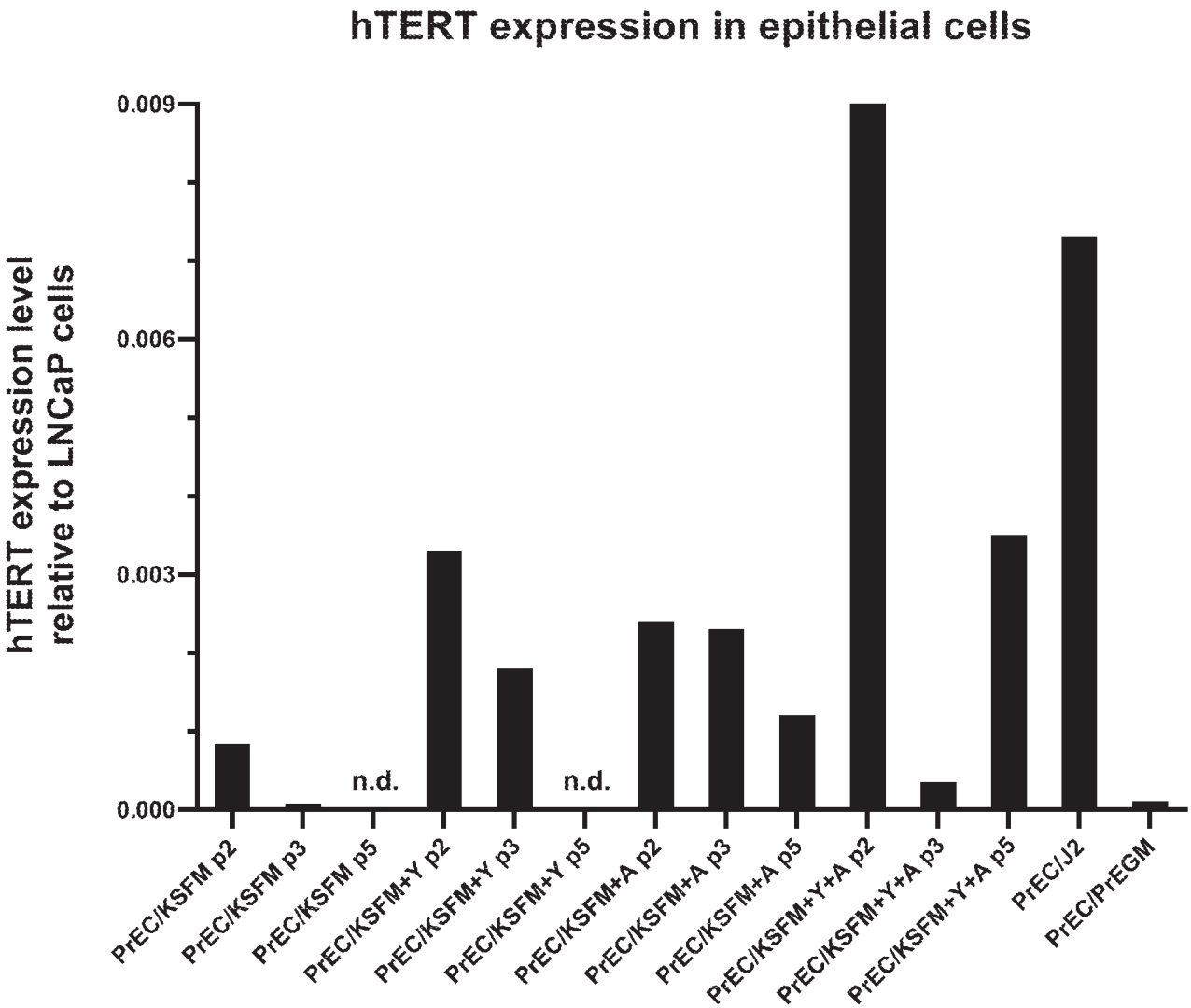
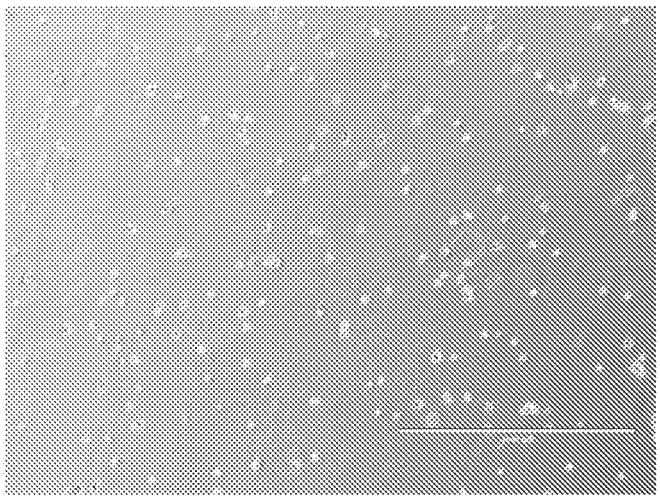
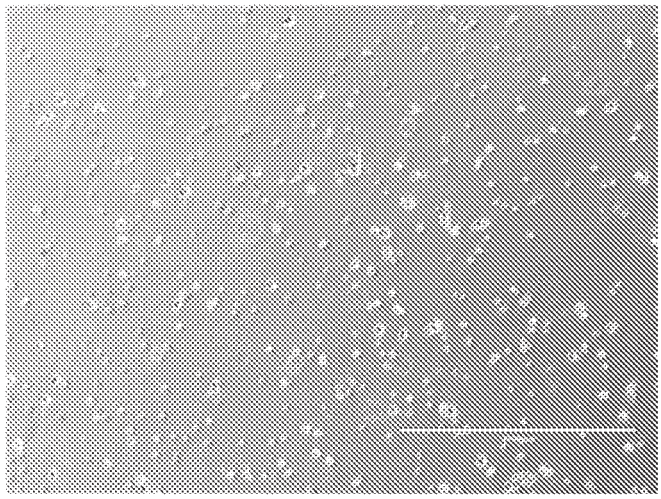

hTERT expression in epithelial cells
hTERT expression level relative to LNCaP cells
0.009
0.006
0.003
0.000
n.d.
n.d.
PrEC/KSFM p2
PrEC/KSFM p3
PrEC/KSFM p5
PrEC/KSFM+Y p2
PrEC/KSFM+Y p3
PrEC/KSFM+Y p5
PrEC/KSFM+A p2
PrEC/KSFM+A p3
PrEC/KSFM+A p5
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PrEC/KSFM+Y+A p5
PrEC/J2
PrEC/PrEGM

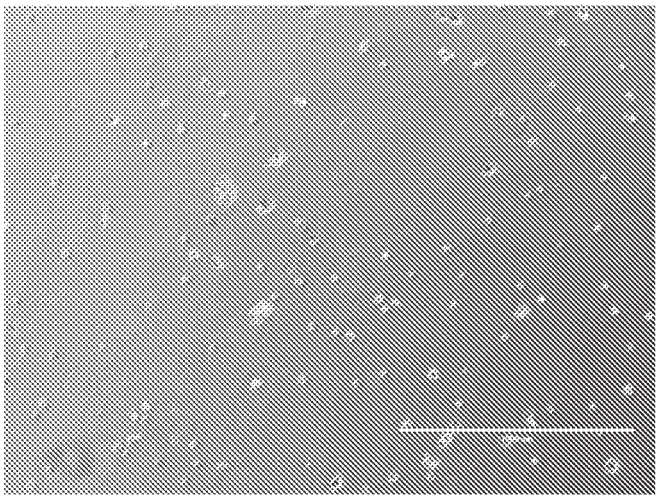
Fig. 6B



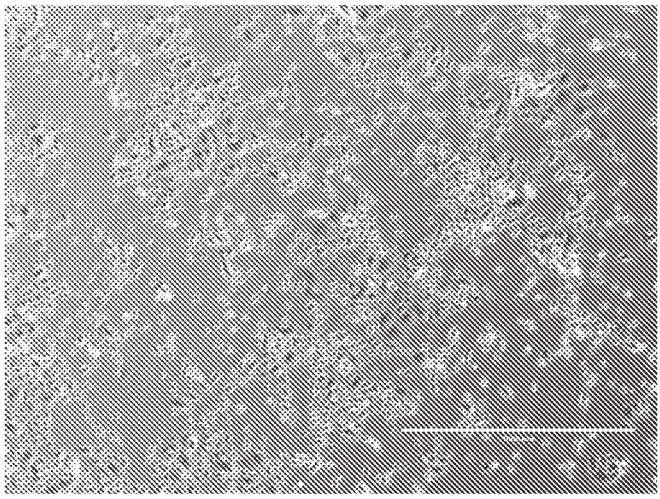
PrEC in KSFM+A, passage 6, day 1



day 2

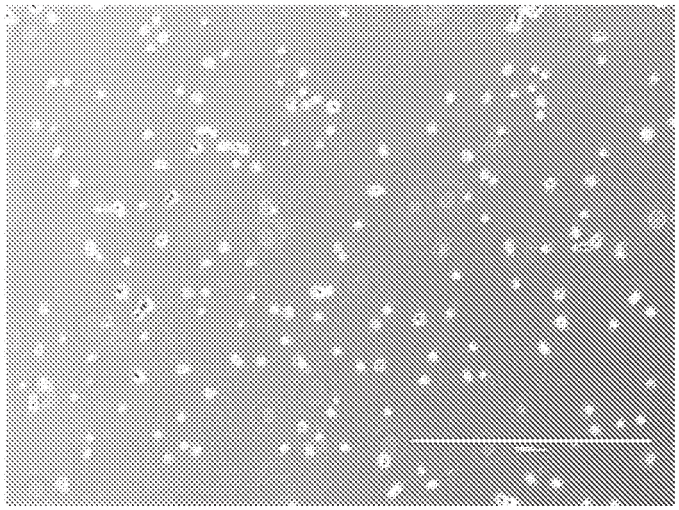


day 3

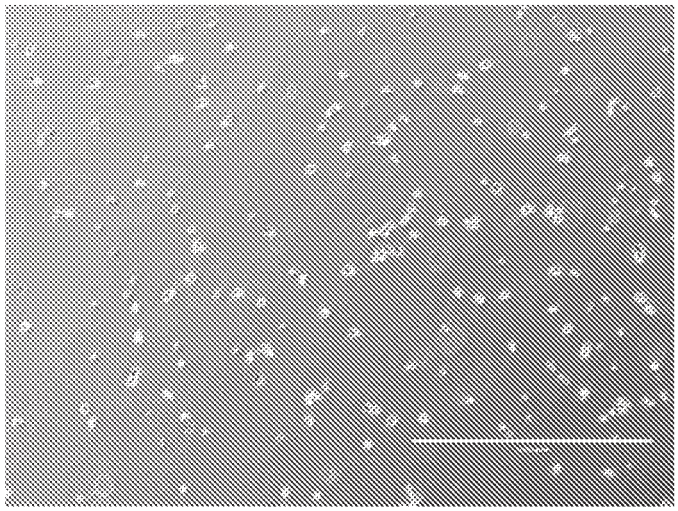


day 8

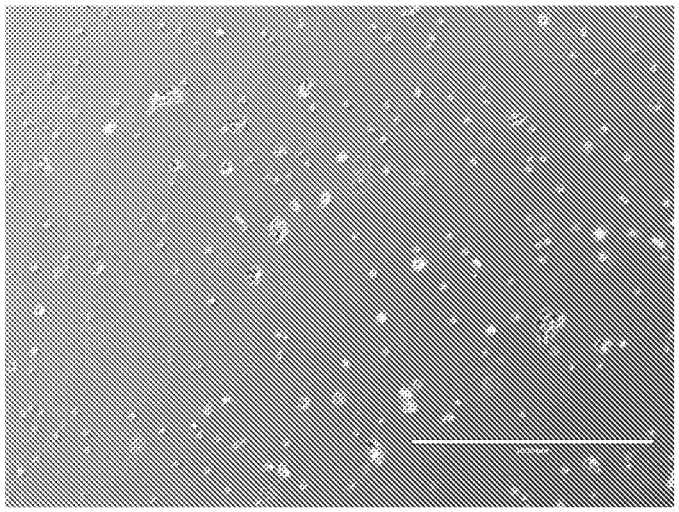
Fig. 7



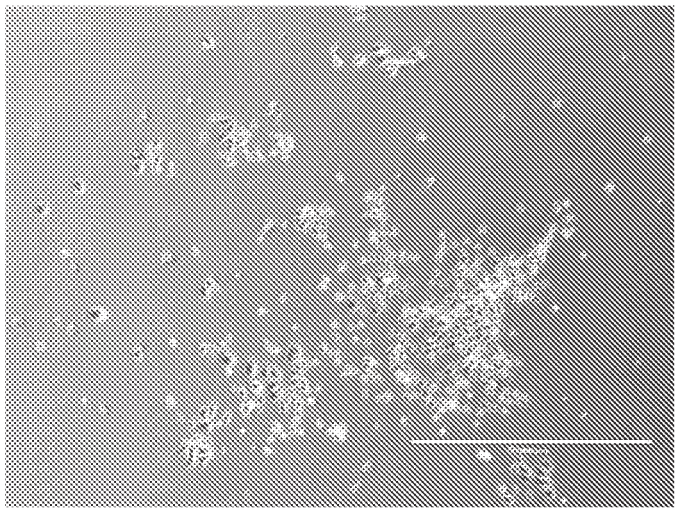
HBEC in KSFM+A, passage 7, day 1



day 2

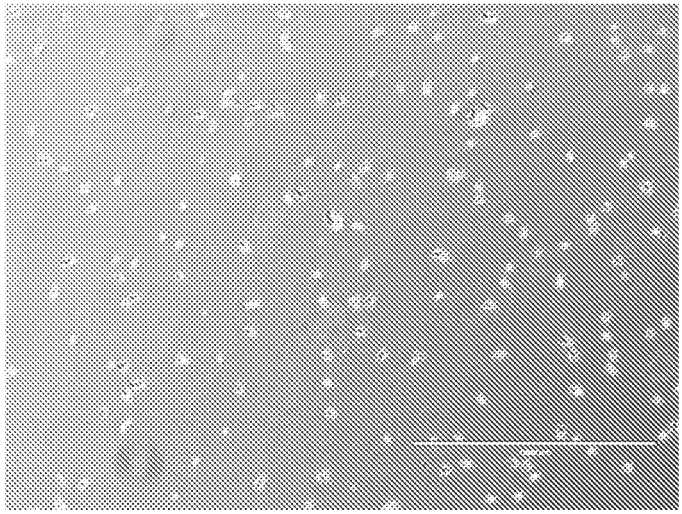


day3

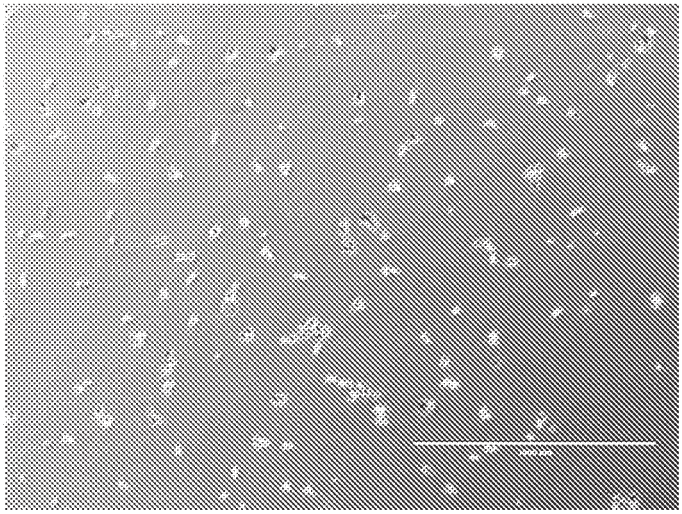


day 7

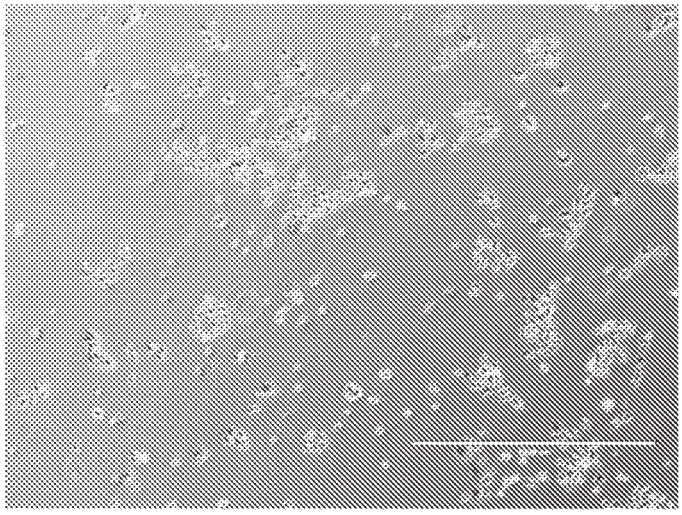
Fig. 8



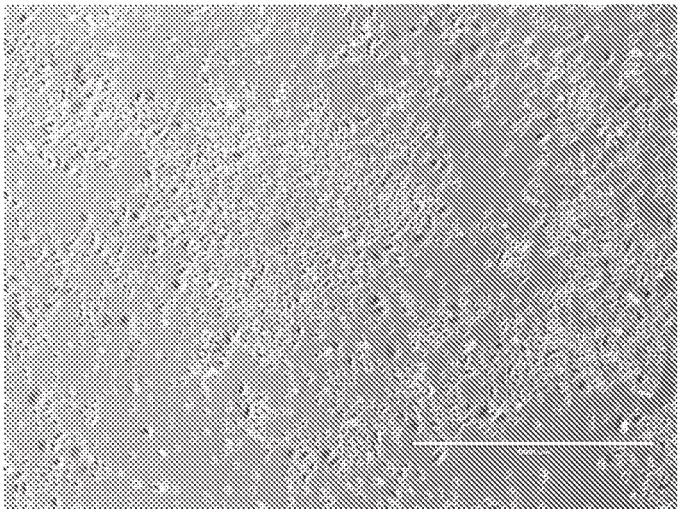
PrEC in KSFM+Y+A, passage 7, day 1



day 2

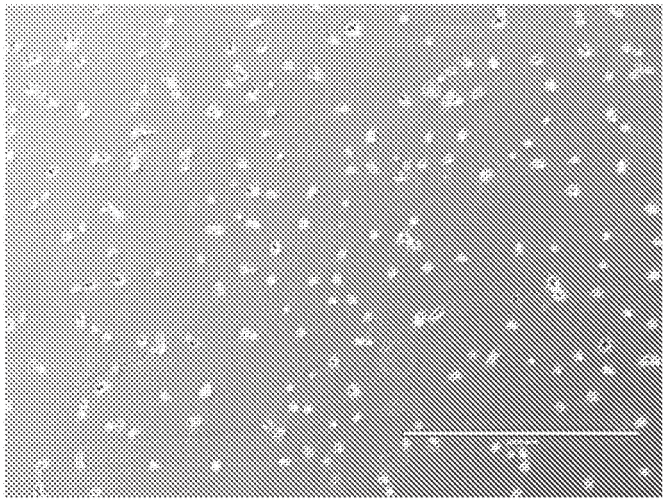


day 4

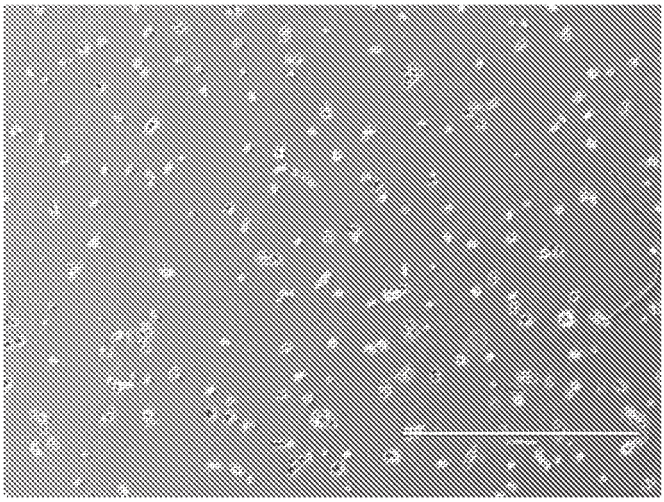


day 7

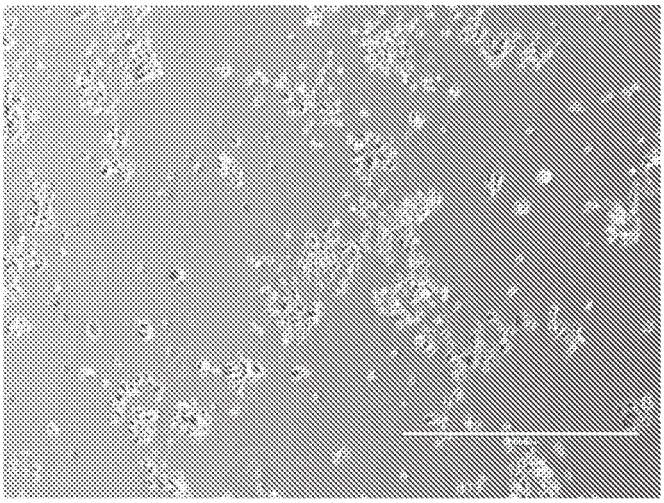
Fig. 9



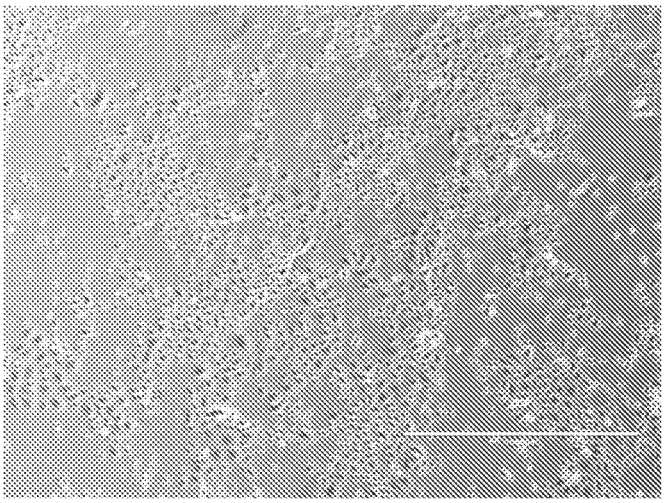
HBEC in KSFM+Y+A, passage 7, day 1



day 2

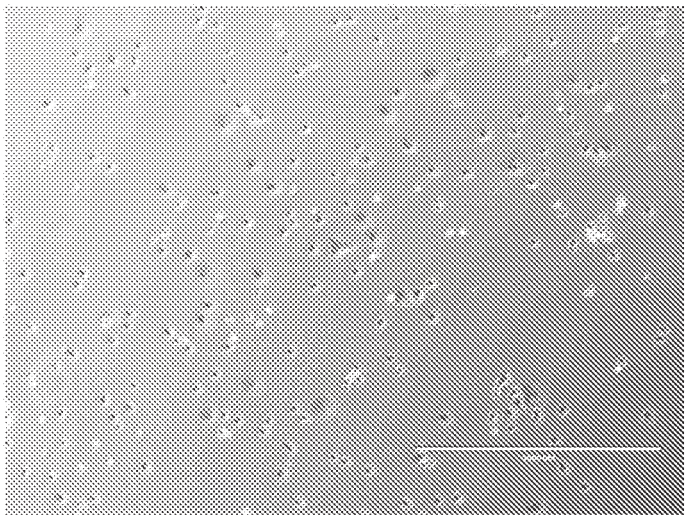


day 4

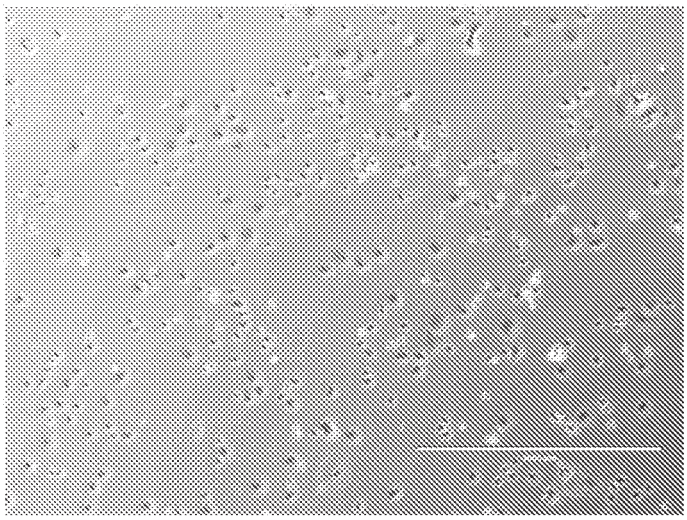


day 7

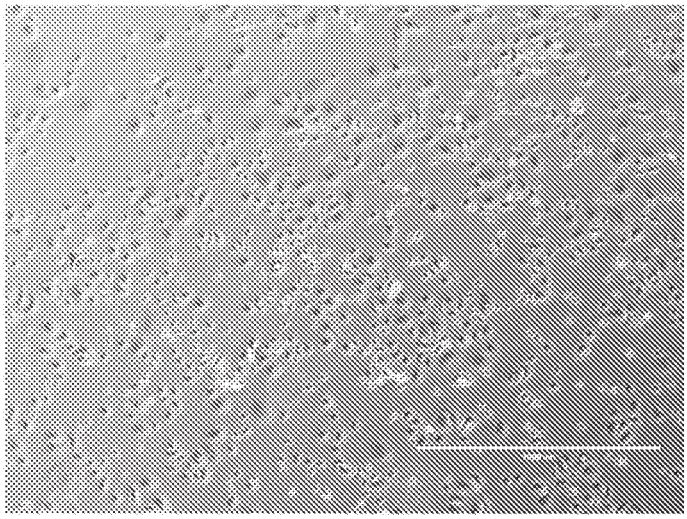
Fig. 10



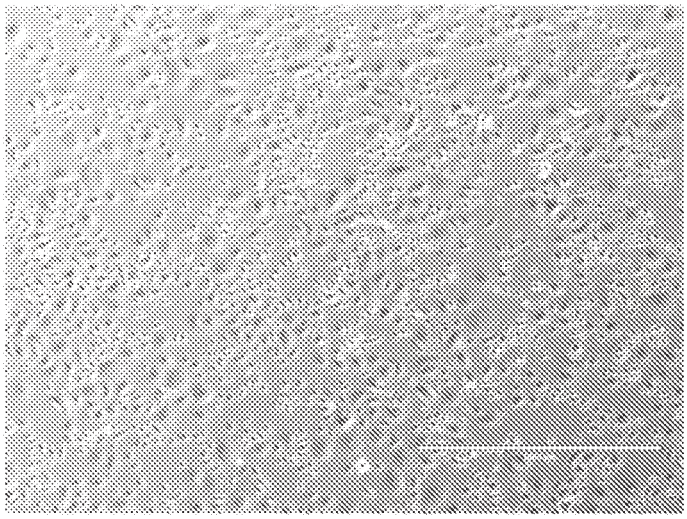
PrEC in KSFM+Y+A+Collagen
passage 7, day 1



day 2

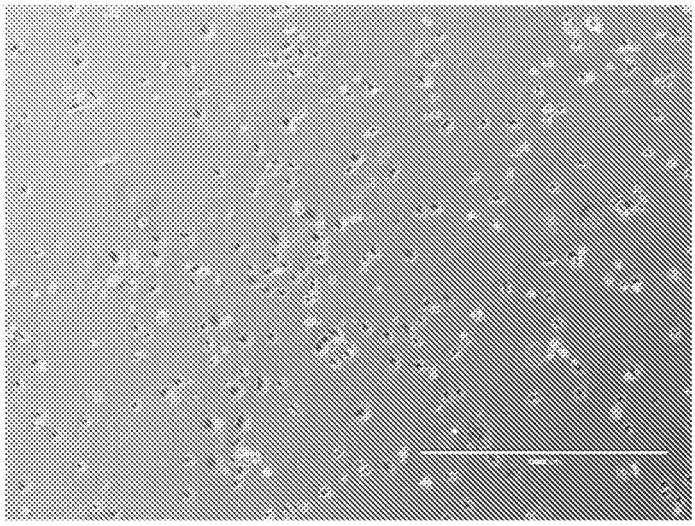


day 4

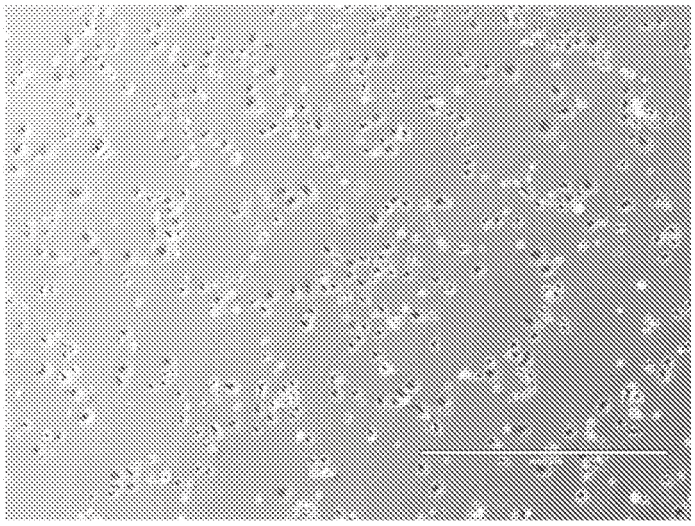


day 5

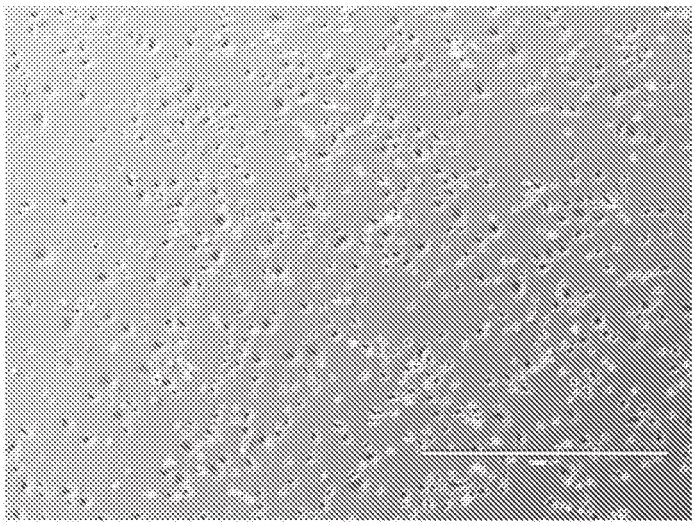
Fig. 11



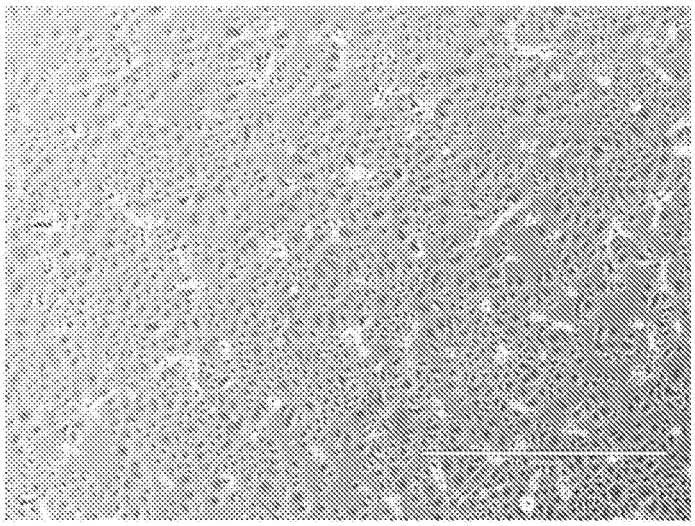
HBEC in KSFM+Y+A+Collagen
passage 7, day1



day 2

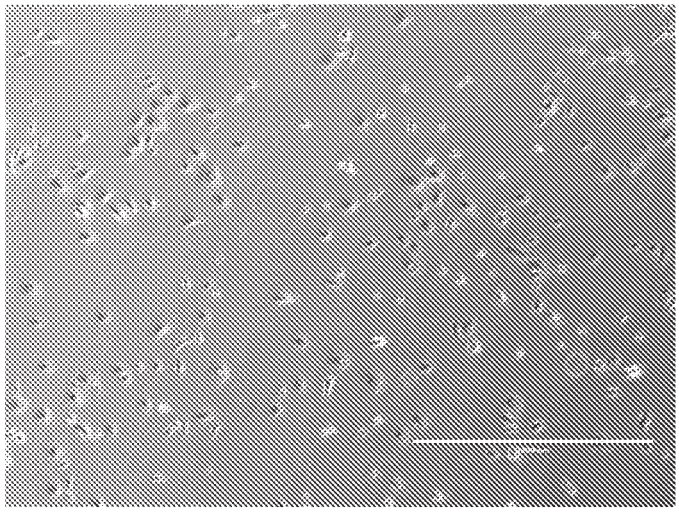


day 3

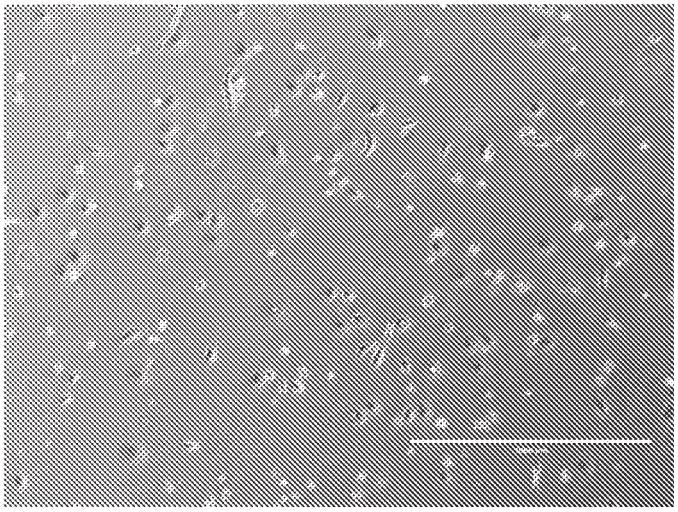


day 6

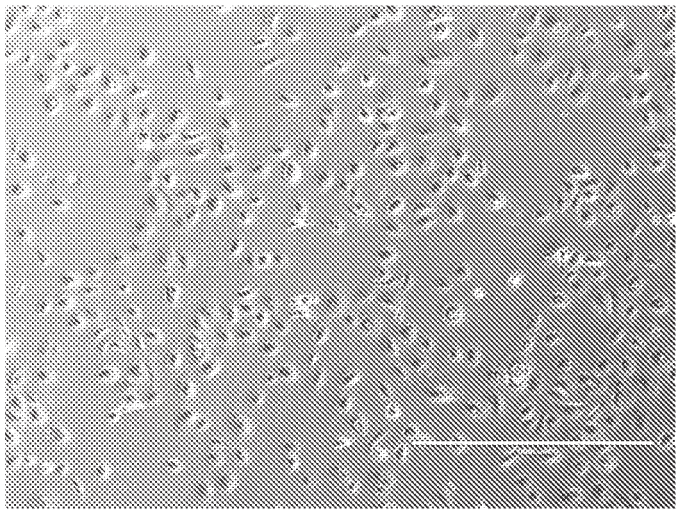
Fig. 12



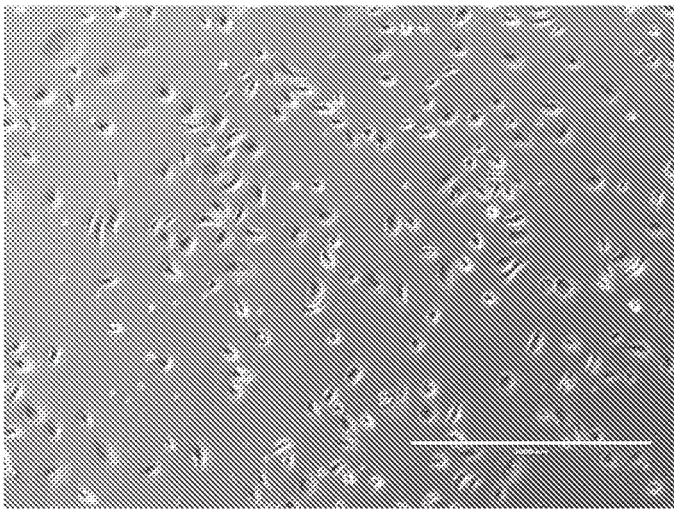
PrEC in KSFM+Y, passage 7, day 1



HBEC in KSFM+Y, passage 6, day 1



PrEC in KSFM+Y, passage 7, day 9



HBEC in KSFM+Y, passage 6, day 8

Fig. 13

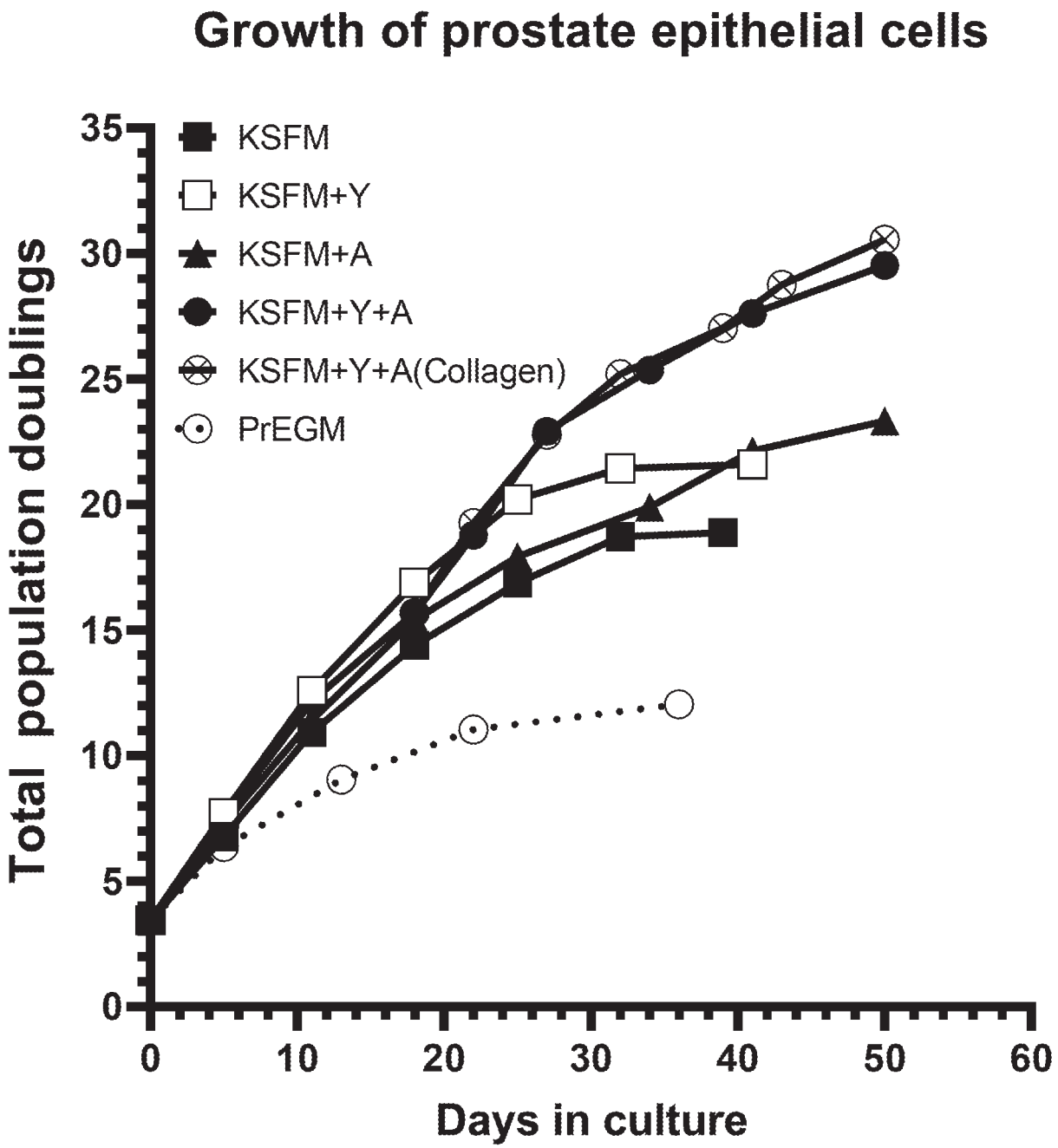

Growth of prostate epithelial cells
KSFM
KSFM+Y
KSFM+A
KSFM+Y+A
KSFM+Y+A(Collagen)
PrEGM
Total population doublings
0
5
10
15
20
25
30
35
0
10
20
30
40
50
60
Days in culture

Fig. 14

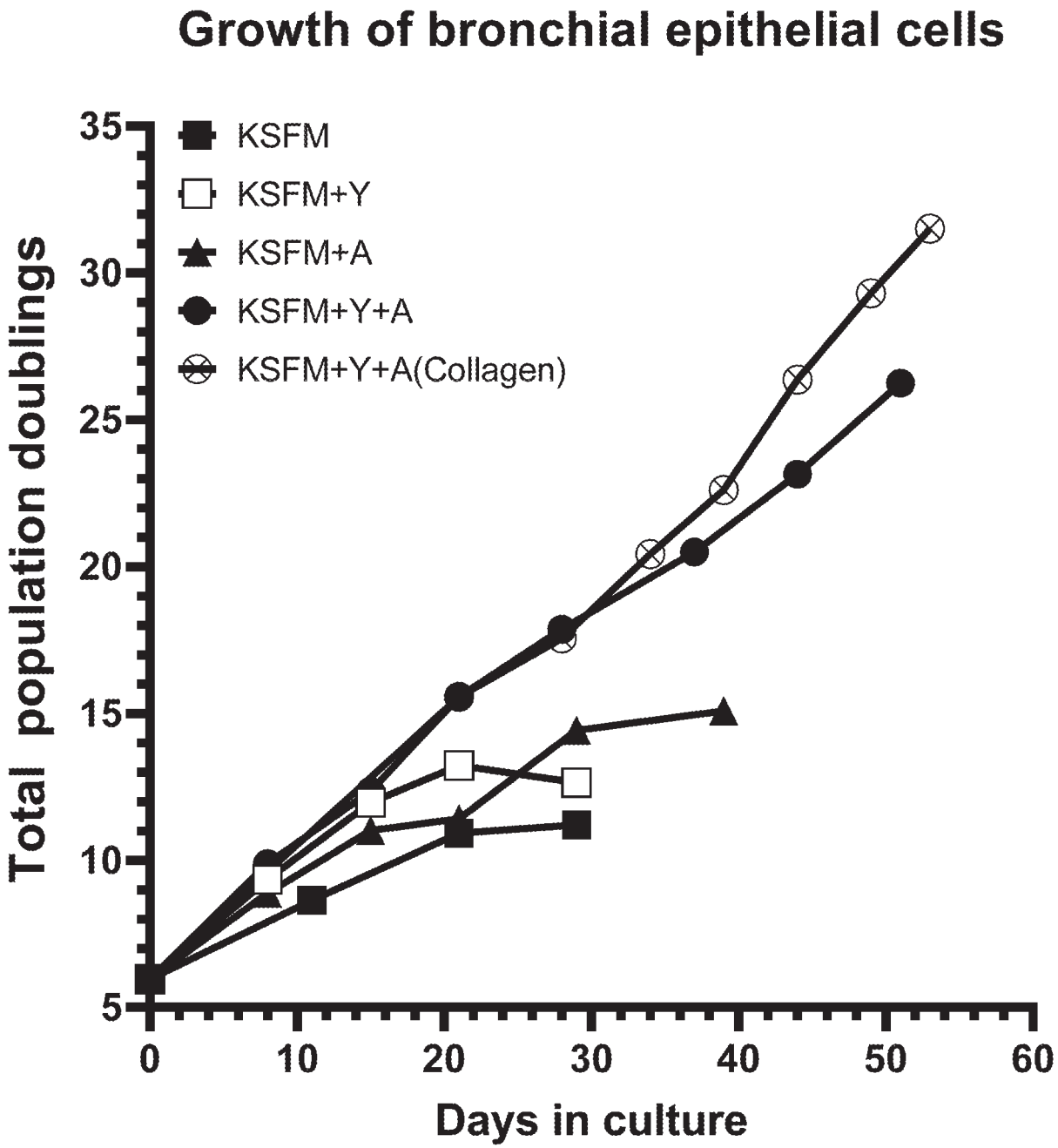

Growth of bronchial epithelial cells
KSFM
KSFM+Y
KSFM+A
KSFM+Y+A
KSFM+Y+A(Collagen)
Total population doublings
35
30
25
20
15
10
5
0
10
20
30
40
50
60
Days in culture

Fig. 15

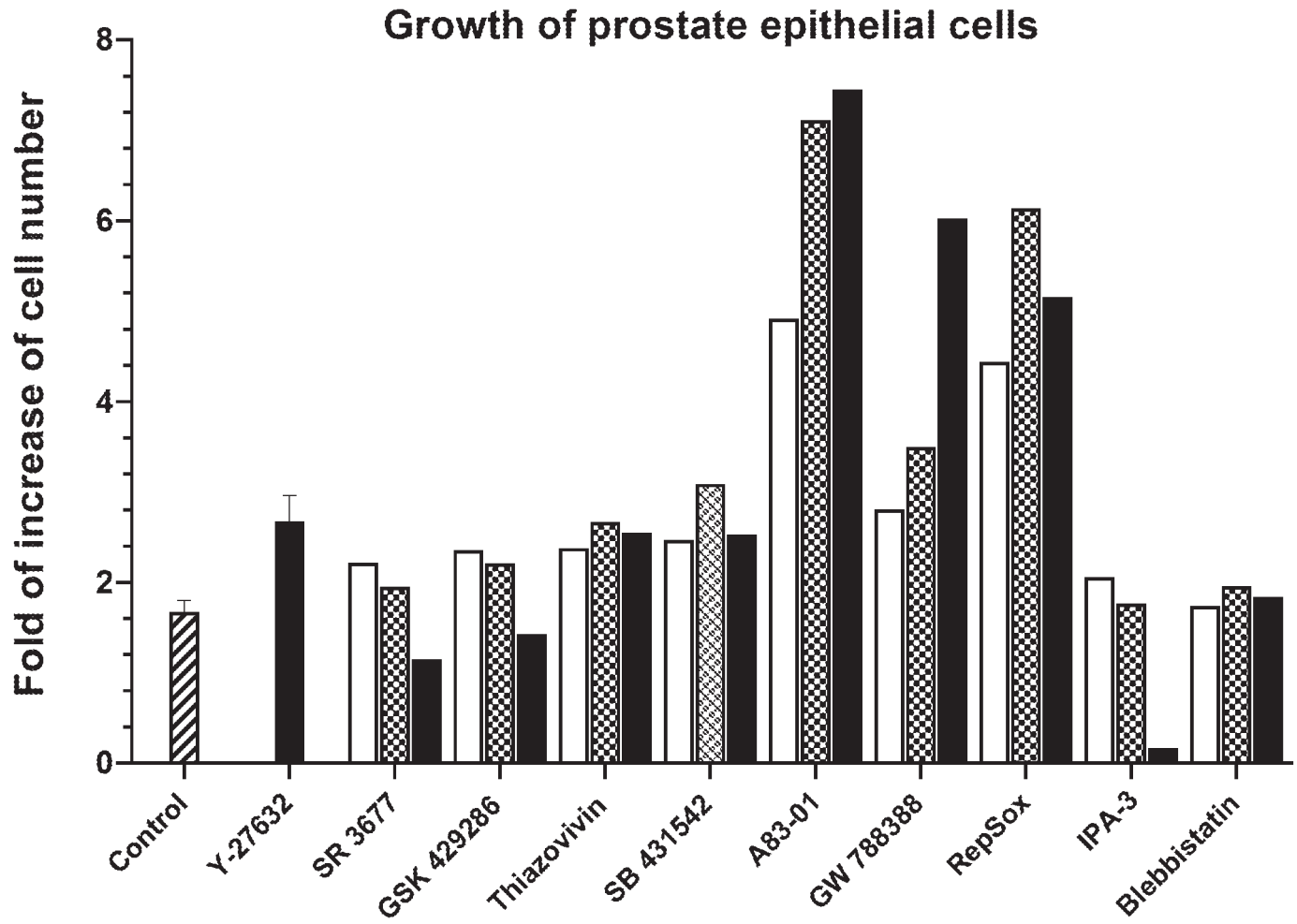

Growth of prostate epithelial cells
Fold of increase of cell number
8
6
4
2
0
Control
Y-27632
SR 3677
GSK 429286
Thiazovivin
SB 431542
A83-01
GW 788388
RepSox
IPA-3
Blebbistatin

Fig. 16

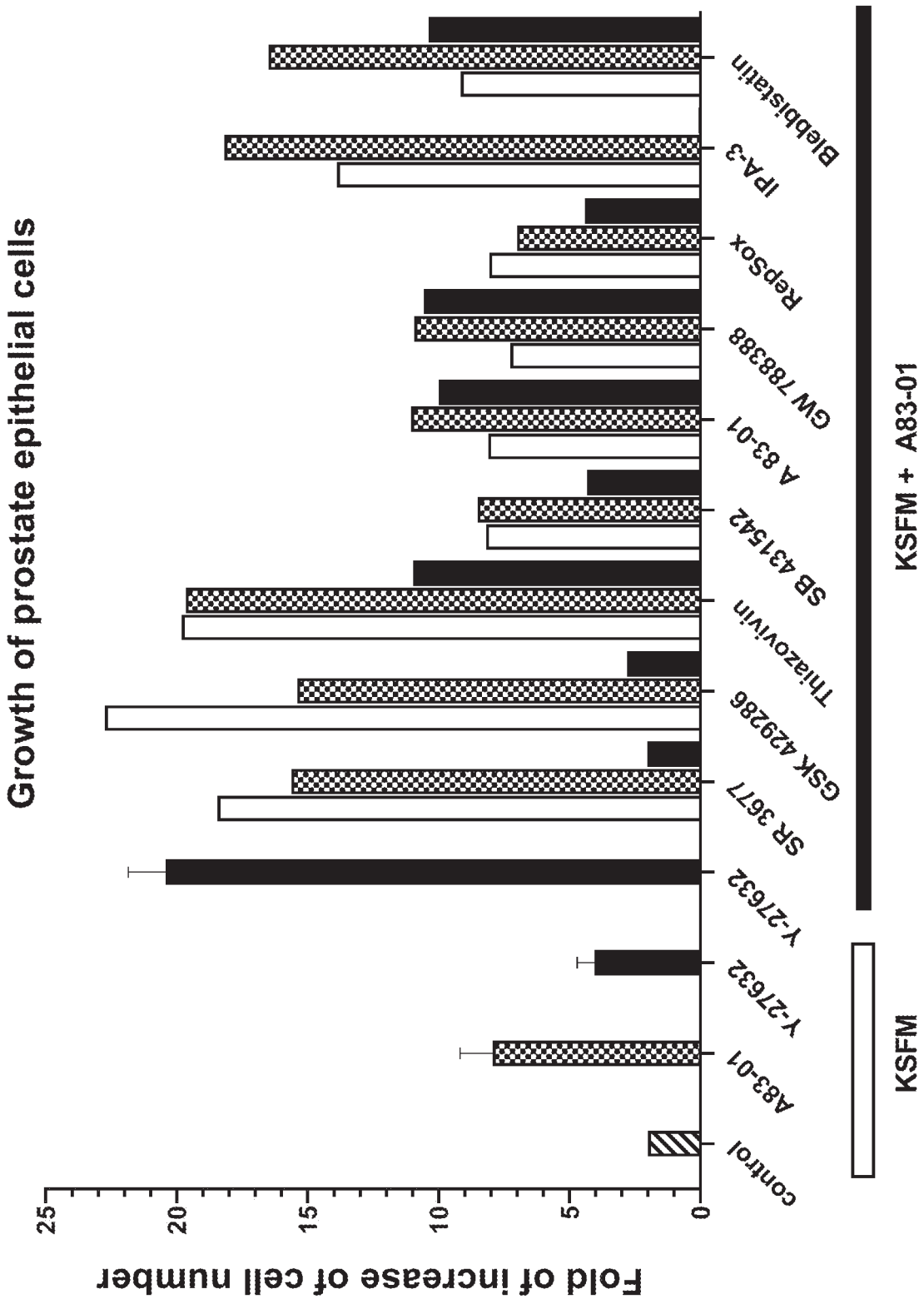

Growth of prostate epithelial cells
Fold of increase of cell number
0
5
10
15
20
25
control
A83-01
Y-27632
Y-27632
SR 3677
GSK 429286
Thiazovivin
SB 431542
A 83-01
GW 788388
Repsox
IPA-3
Blebbistatin
KSFM
KSFM + A83-01

Fig. 17

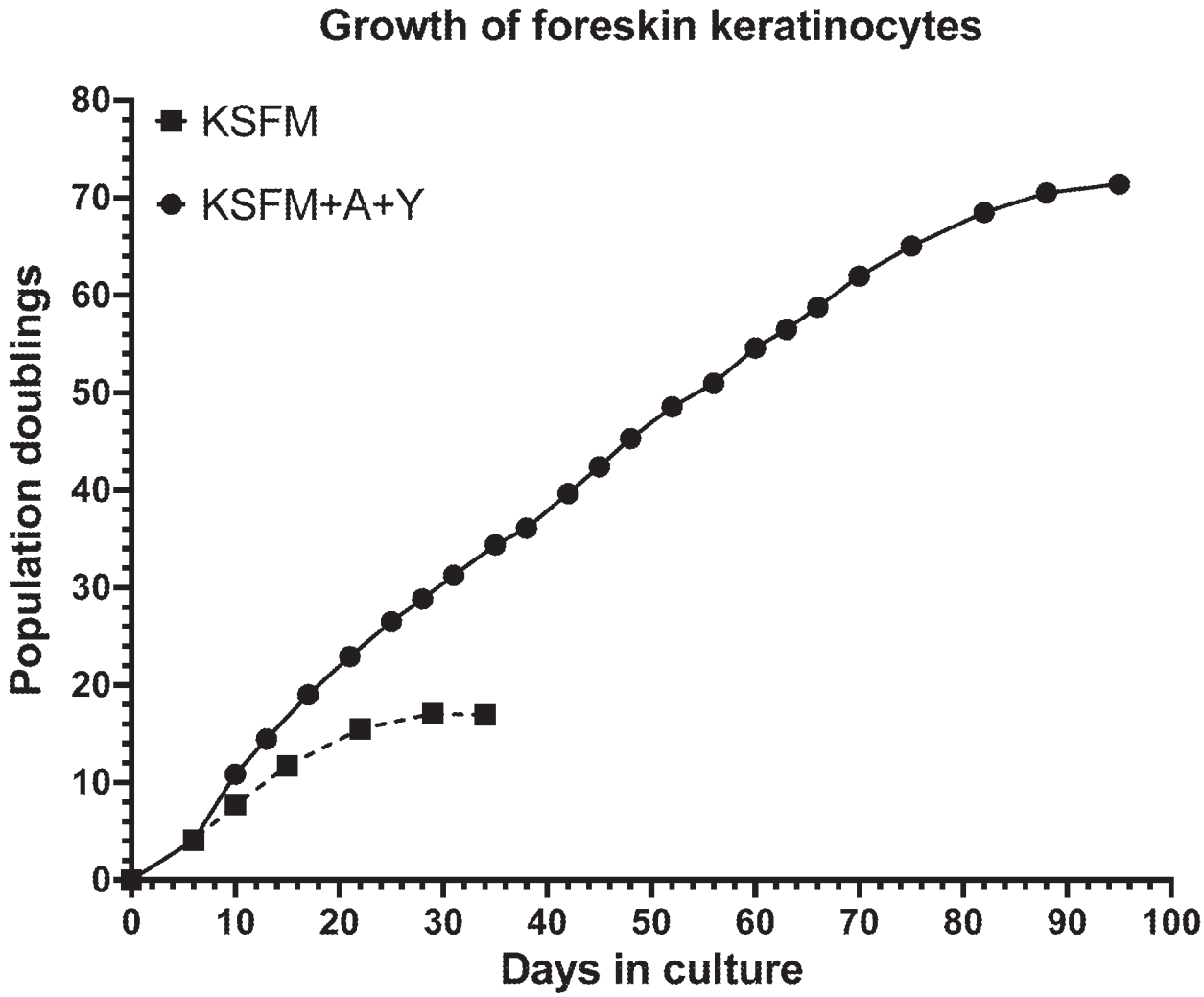

Growth of foreskin keratinocytes
KSFM
KSFM+A+Y
Population doublings
Days in culture
0
10
20
30
40
50
60
70
80
90
100

Fig. 18

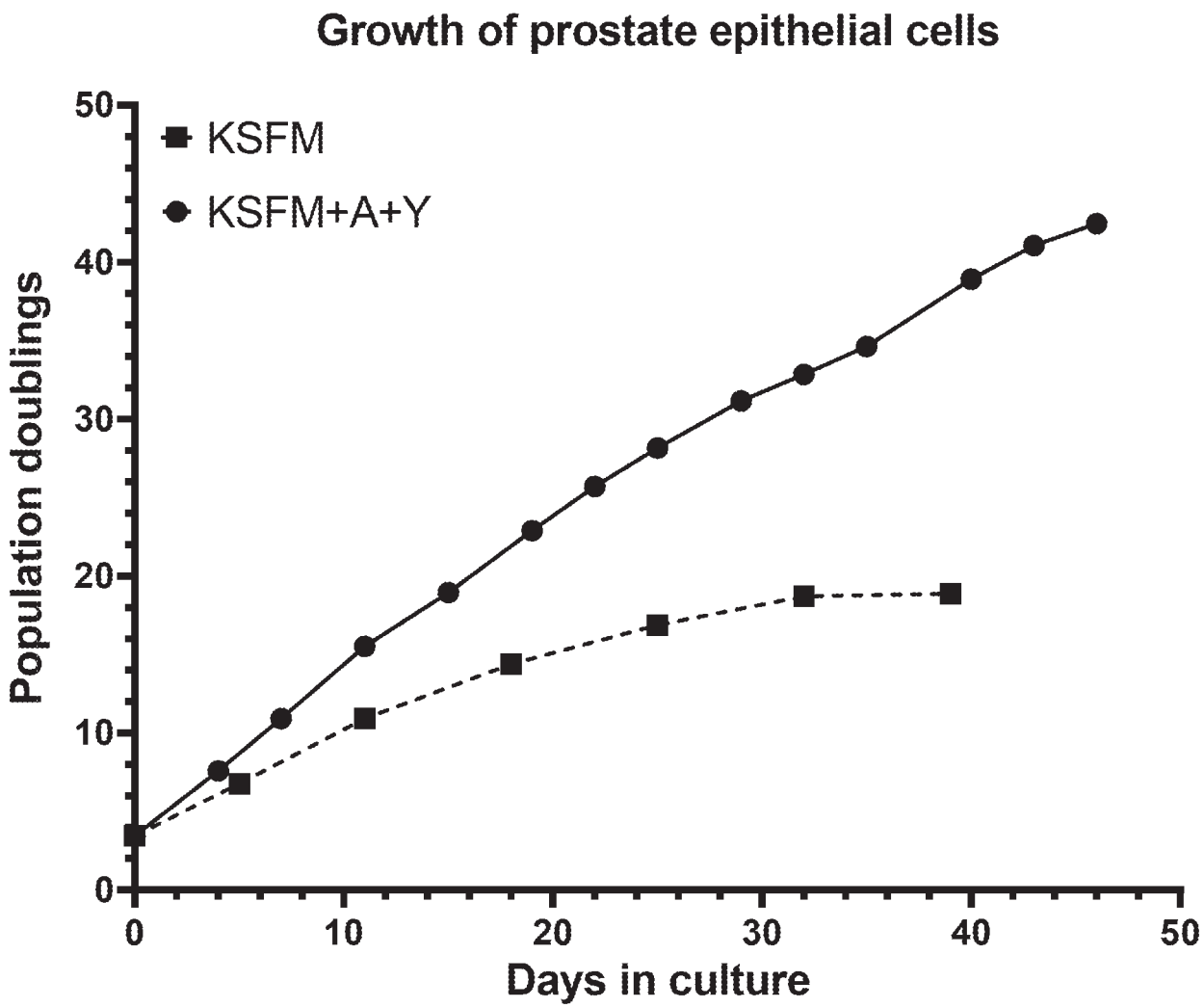

Growth of prostate epithelial cells
KSFM
KSFM+A+Y
Population doublings
Days in culture
0
10
20
30
40
50

Fig. 19

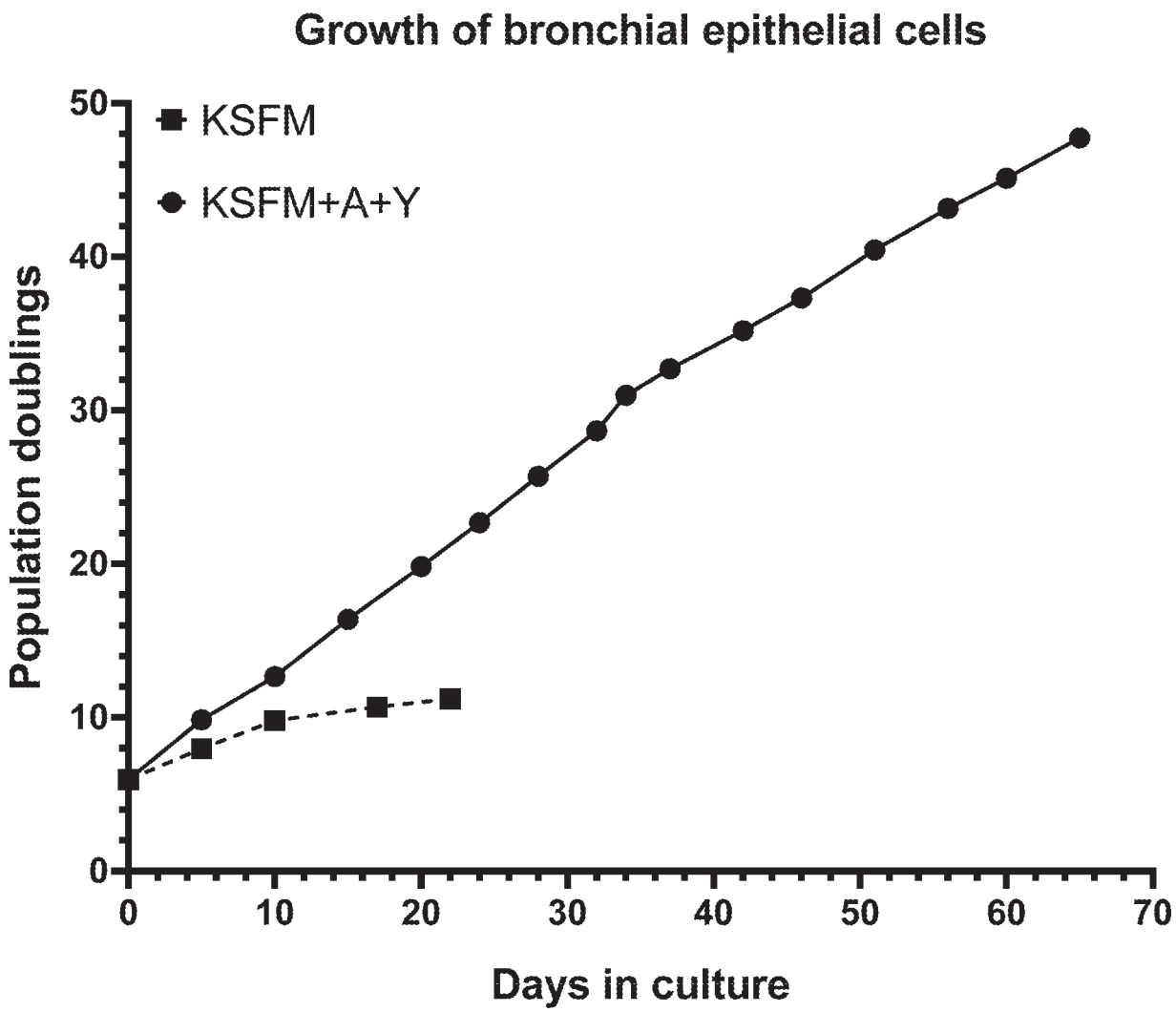

Growth of bronchial epithelial cells
KSFM
KSFM+A+Y
Population doublings
0
10
20
30
40
50
0
10
20
30
40
50
60
70
Days in culture

Fig. 20

Karotypes of HFK, HBEC and PrEC cells			
Cell Line	Donor Age	Early Passage in KSFM+A+Y	Late Passage in KSFM+A+Y
HFK	0 year	46,XY, P3 (13.5 PD)	46,XY, P19 (62.0 PD)
HBEC	49 years	46,XX, P4 (11.1 PD)	46,XX, P16 (45.1 PD)
PrEC	26 years	46,XY, P3 (13.5 PD)	46,XY, P13 (41.1 PD)

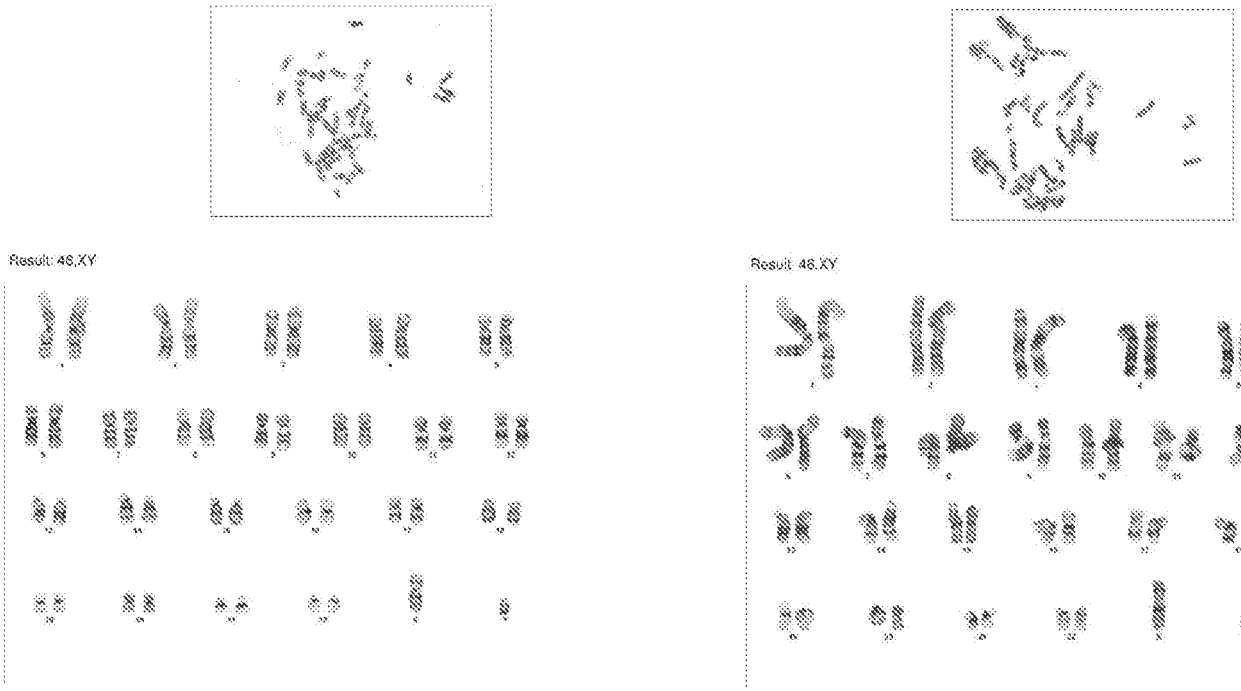


Fig. 21

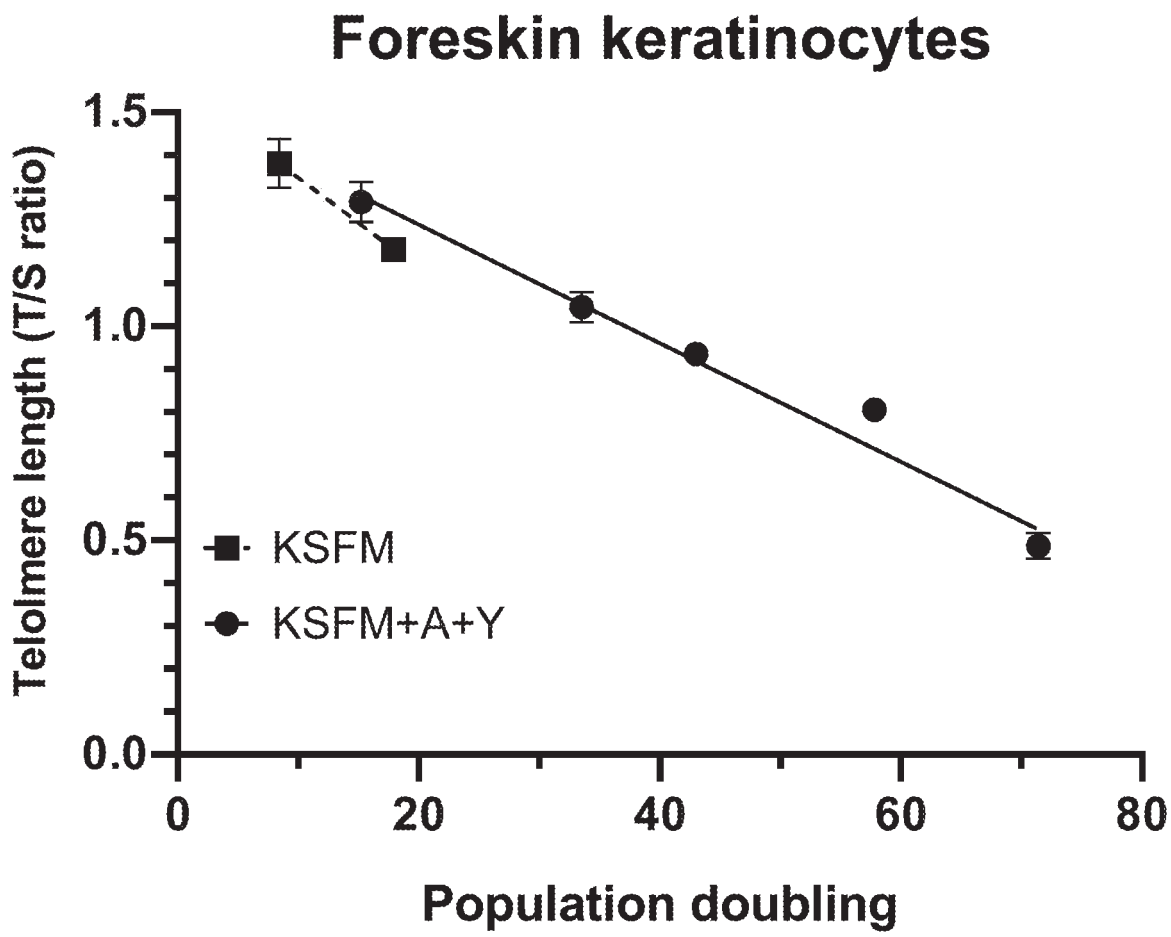

Foreskin keratinocytes
Telolmere length (T/S ratio)
1.5
1.0
0.5
0.0
KSFM
KSFM+A+Y
0
20
40
60
80
Population doubling

Fig. 22

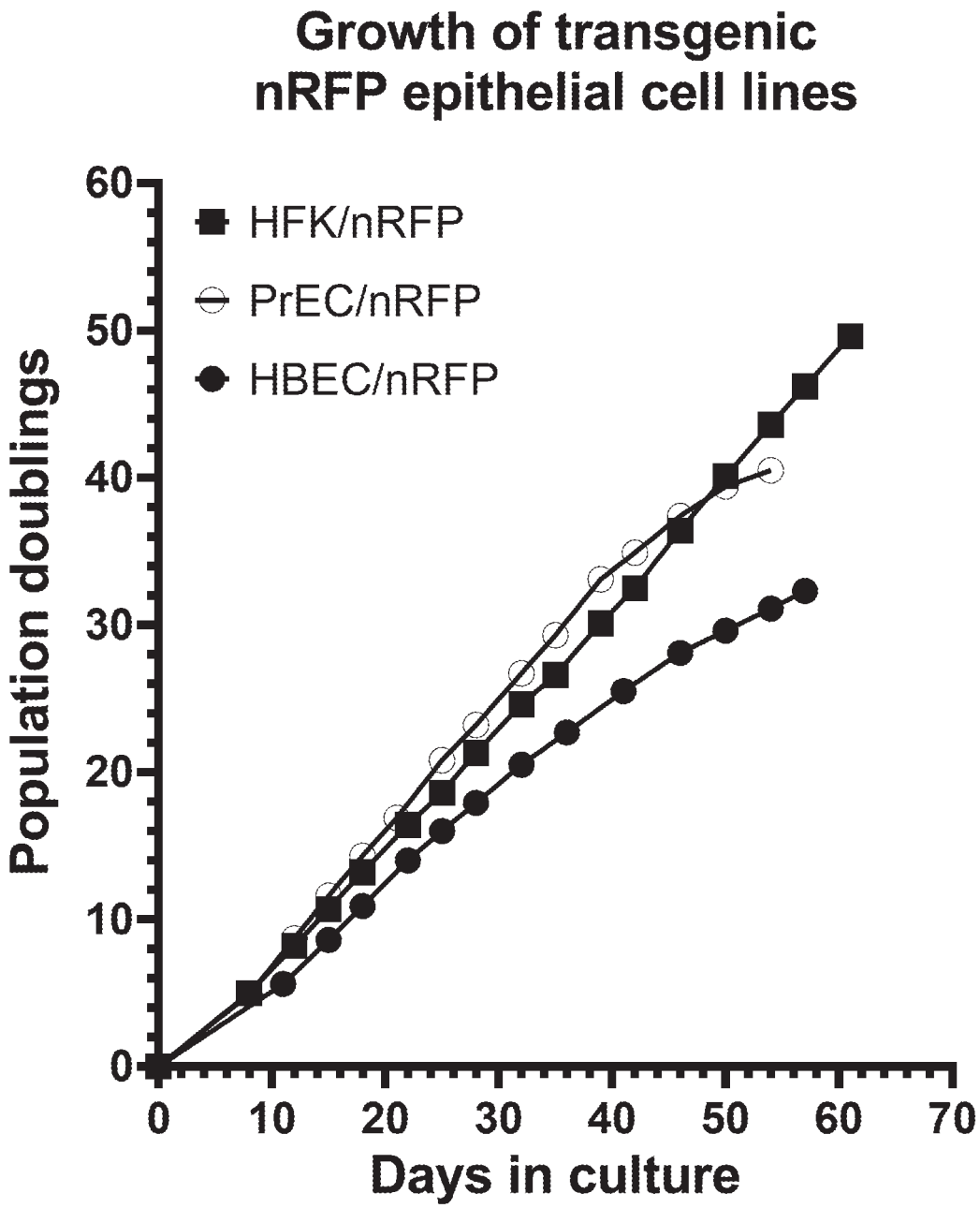

Growth of transgenic
nRFP epithelial cell lines
HFK/nRFP
PrEC/nRFP
HBEC/nRFP
Population doublings
0
10
20
30
40
50
60
0
10
20
30
40
50
60
70
Days in culture

Fig. 23

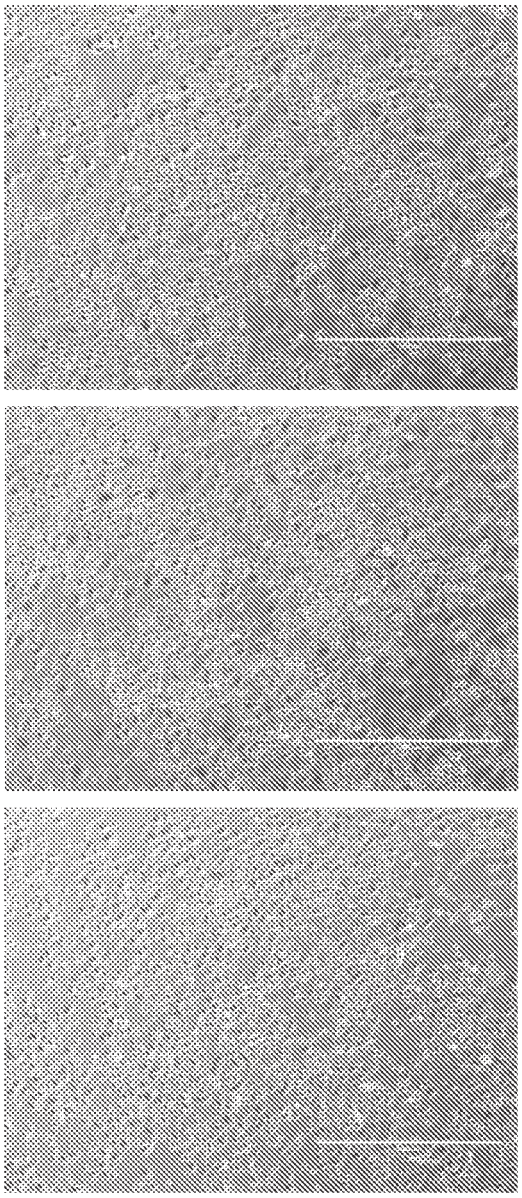


Fig. 24

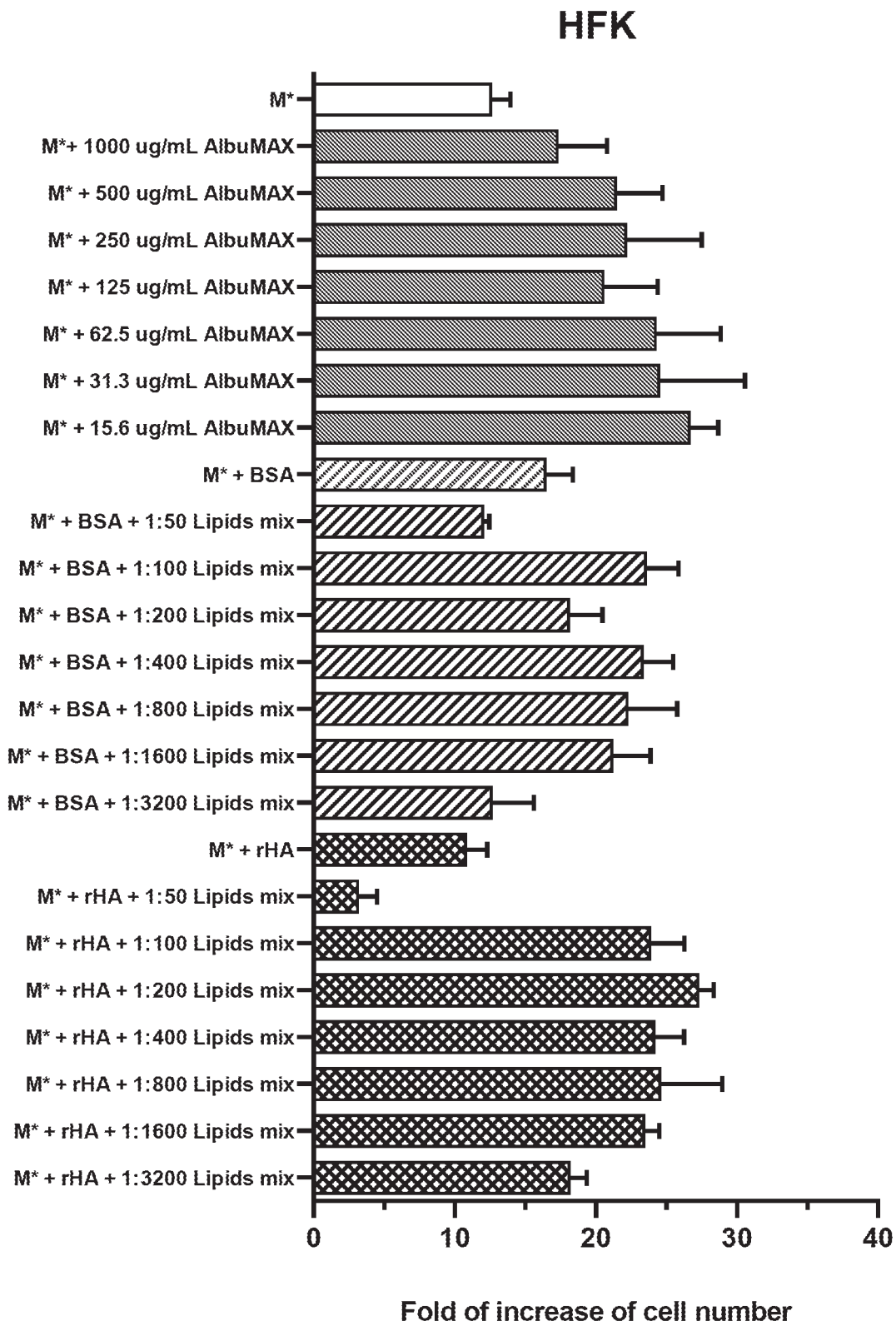

HFK
M*
M*+ 1000 ug/mL AlbuMAX
M* + 500 ug/mL AlbuMAX
M* + 250 ug/mL AlbuMAX
M* + 125 ug/mL AlbuMAX
M* + 62.5 ug/mL AlbuMAX
M* + 31.3 ug/mL AlbuMAX
M* + 15.6 ug/mL AlbuMAX
M* + BSA
M* + BSA + 1:50 Lipids mix
M* + BSA + 1:100 Lipids mix
M* + BSA + 1:200 Lipids mix
M* + BSA + 1:400 Lipids mix
M* + BSA + 1:800 Lipids mix
M* + BSA + 1:1600 Lipids mix
M* + BSA + 1:3200 Lipids mix
M* + rHA
M* + rHA + 1:50 Lipids mix
M* + rHA + 1:100 Lipids mix
M* + rHA + 1:200 Lipids mix
M* + rHA + 1:400 Lipids mix
M* + rHA + 1:800 Lipids mix
M* + rHA + 1:1600 Lipids mix
M* + rHA + 1:3200 Lipids mix
0
10
20
30
40
Fold of increase of cell number

Fig. 25

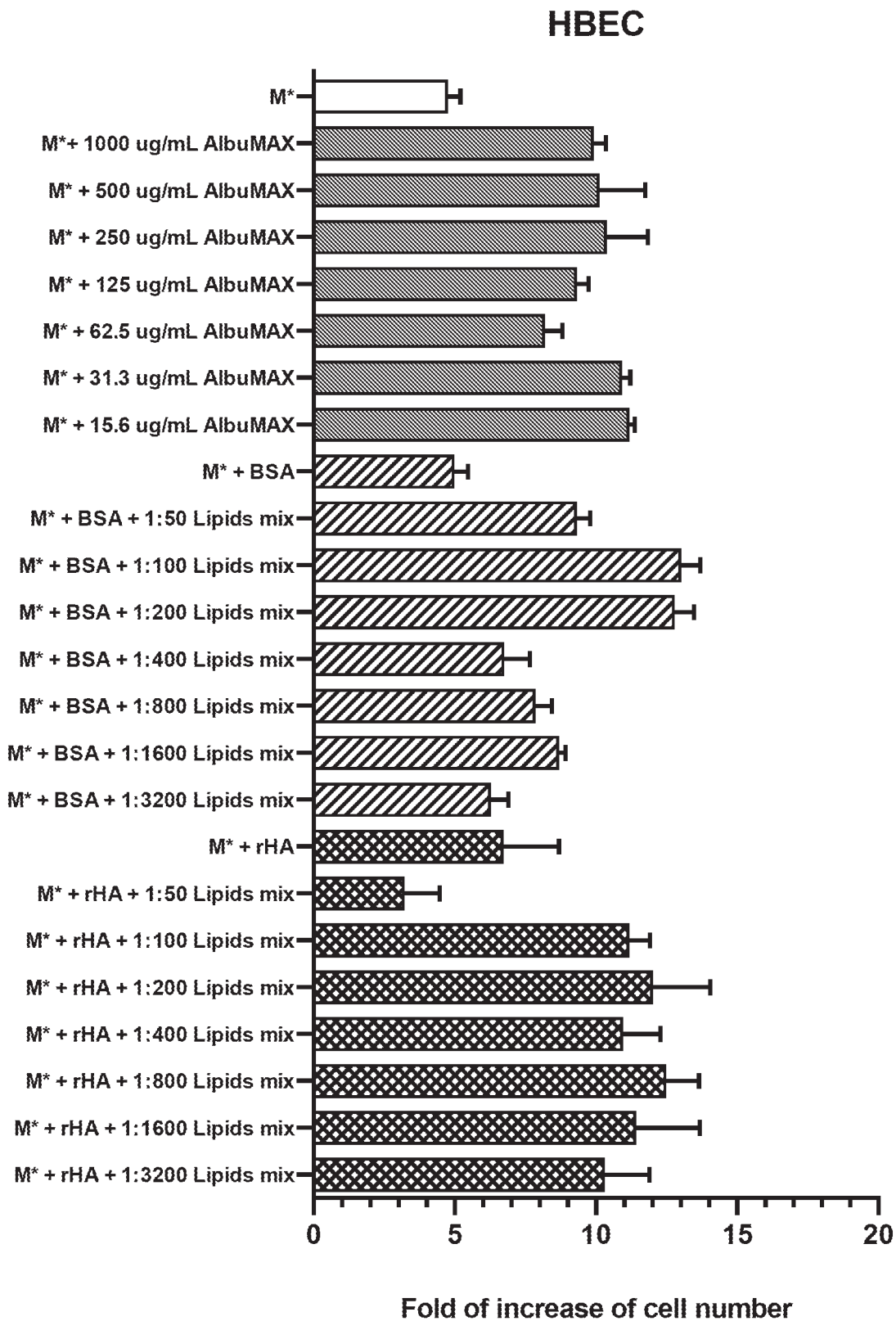

HBEC
M*
M*+ 1000 ug/mL AlbuMAX
M* + 500 ug/mL AlbuMAX
M* + 250 ug/mL AlbuMAX
M* + 125 ug/mL AlbuMAX
M* + 62.5 ug/mL AlbuMAX
M* + 31.3 ug/mL AlbuMAX
M* + 15.6 ug/mL AlbuMAX
M* + BSA
M* + BSA + 1:50 Lipids mix
M* + BSA + 1:100 Lipids mix
M* + BSA + 1:200 Lipids mix
M* + BSA + 1:400 Lipids mix
M* + BSA + 1:800 Lipids mix
M* + BSA + 1:1600 Lipids mix
M* + BSA + 1:3200 Lipids mix
M* + rHA
M* + rHA + 1:50 Lipids mix
M* + rHA + 1:100 Lipids mix
M* + rHA + 1:200 Lipids mix
M* + rHA + 1:400 Lipids mix
M* + rHA + 1:800 Lipids mix
M* + rHA + 1:1600 Lipids mix
M* + rHA + 1:3200 Lipids mix
0
5
10
15
20
Fold of increase of cell number

Fig. 26

Gene Name	KSFM p2	KSFM p6	KSFM+A+Y, p2	KSFM+A+Y, p13	KSFM+A+Y, p23
Genes that are involved in stress response and senescence					
AKT1	1	26.0	-2.6	-3.1	-3.2
ATM	1	6.1	-3.2	-1.6	-3.2
CDKN2A	1	16.3	-2.5	2.5	1.8
GADD45A	1	5.0	-6.2	2.2	-1.8
GLB1	1	4.8	-7.8	-1.1	1.1
PLAU	1	3.8	-19.4	-7.1	-28.8
SERPINE1	1	3.3	-16.1	-2.3	-13.7
SOD2	1	5.8	-1.0	1.2	-1.2
Adhesion molecules and intermediate filament protein					
FN1	1	4.7	-3821.7	-2149.8	-982.3
THBS1	1	2.8	-99.0	-3.3	-16.3
VIM	1	4.1	-153.3	-310.8	-8192.0
Genes that are upregulated in KSFM+A+Y					
CDKN2B	1	-226.0	1.2	1.9	4.5
CITED2	1	3.2	-1.4	8.5	8.5
CREG1	1	1.0	1.5	1.9	3.1
ID1	1	-1.0	4.4	3.3	11.2
MAP2K6	1	-2.3	-1.6	33.4	22.6
IGFBP3	1	12.7	9.3	455.1	2019.8
IGFBP5	1	2.0	1.8	57.3	2.6

Fig. 27

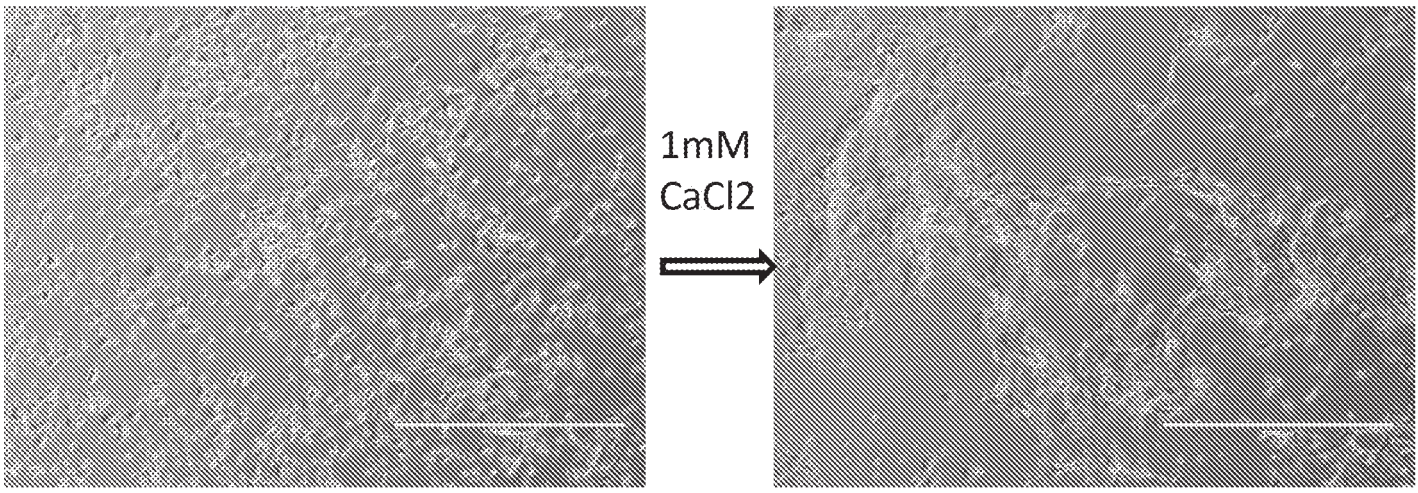


Fig. 28

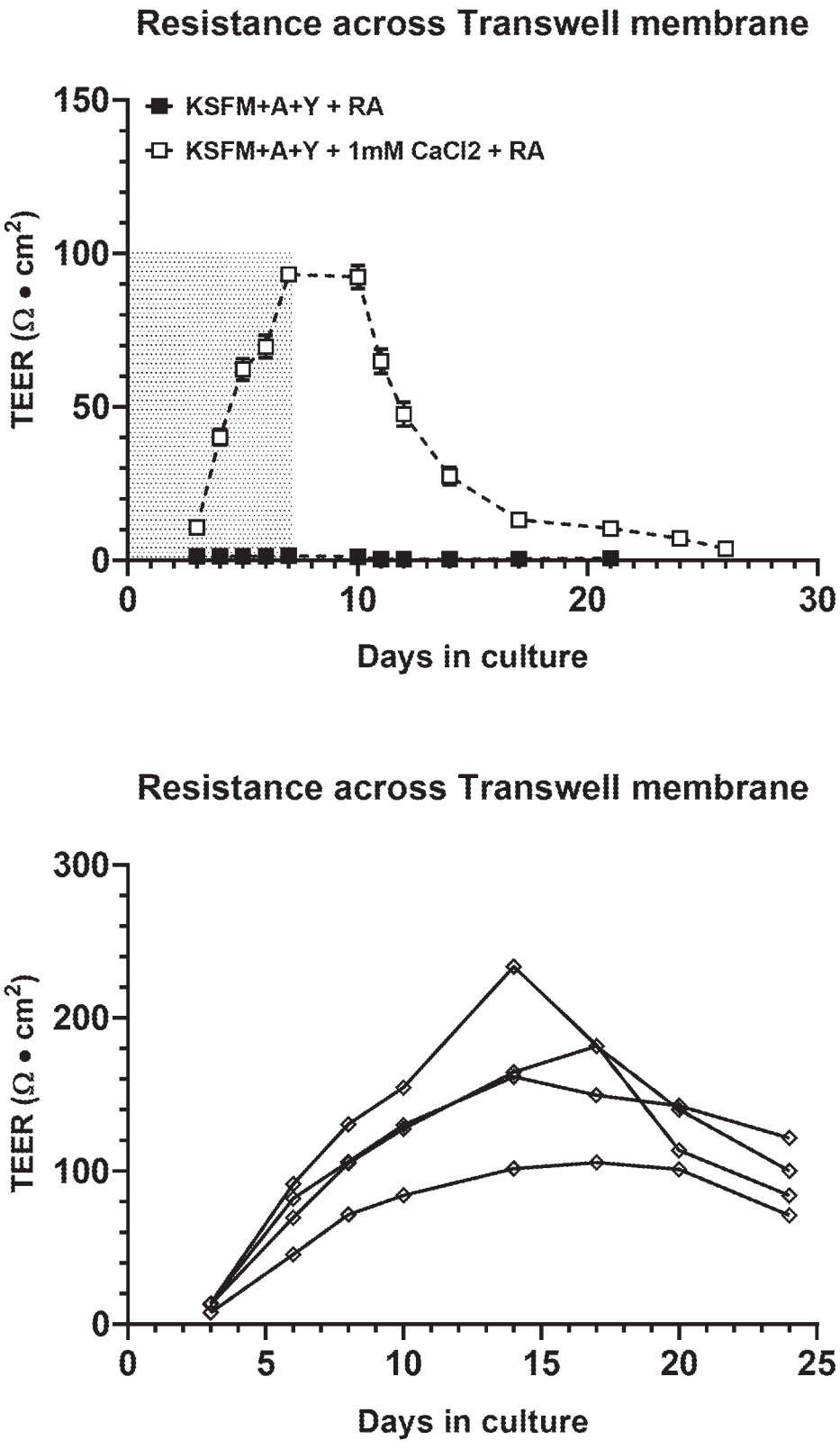

Resistance across Transwell membrane
KSFM+A+Y + RA
KSFM+A+Y + 1mM CaCl2 + RA
TEER (Ω • cm²)
150
100
50
0
0
10
20
30
Days in culture
Resistance across Transwell membrane
TEER (Ω • cm²)
300
200
100
0
0
5
10
15
20
25
Days in culture

Fig. 29

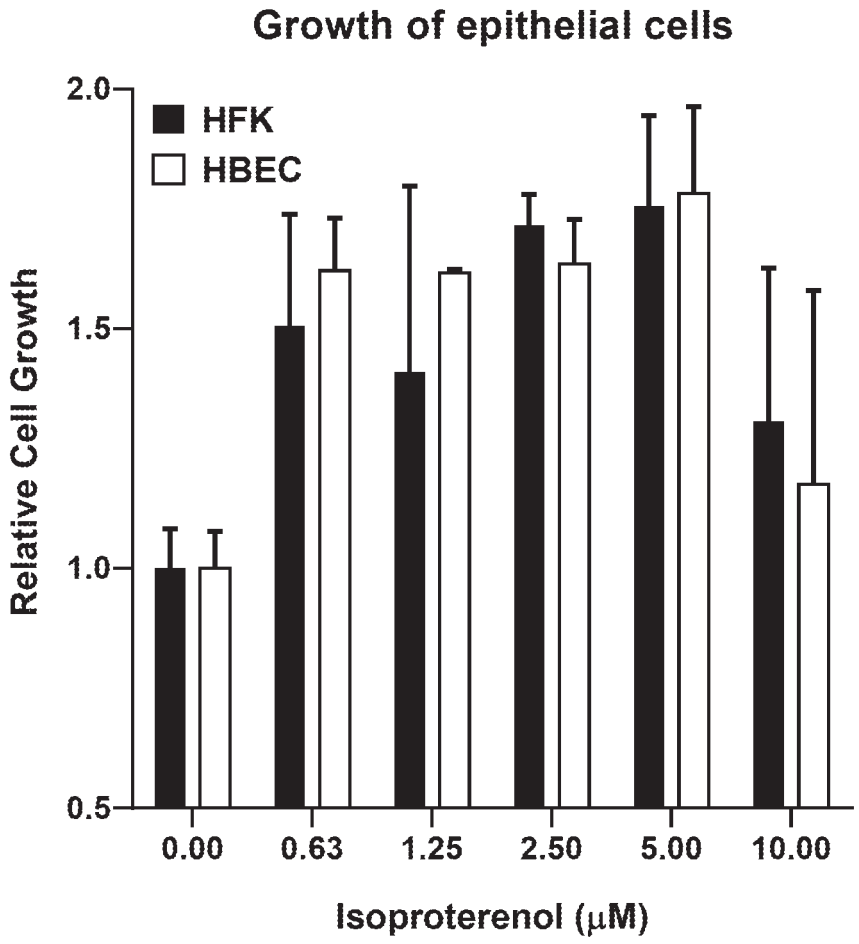

Growth of epithelial cells
HFK
HBEC
Relative Cell Growth
2.0
1.5
1.0
0.5
0.00
0.63
1.25
2.50
5.00
10.00
Isoproterenol (μM)

Fig. 30

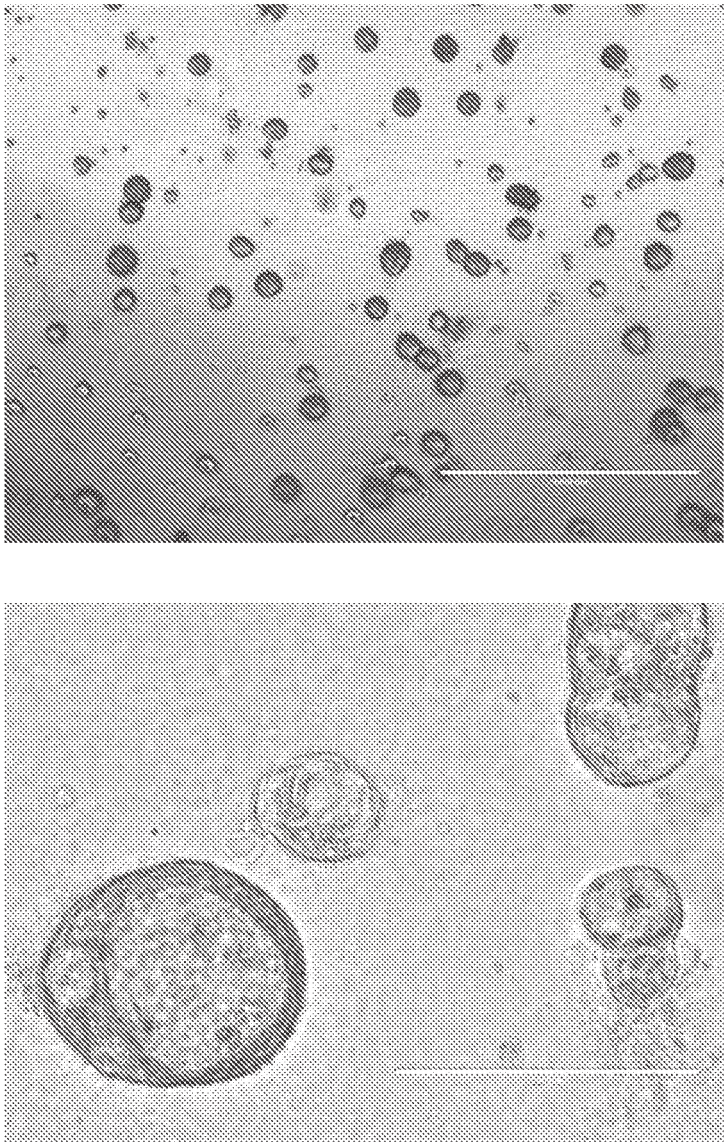


Fig. 31

Fig. 32A

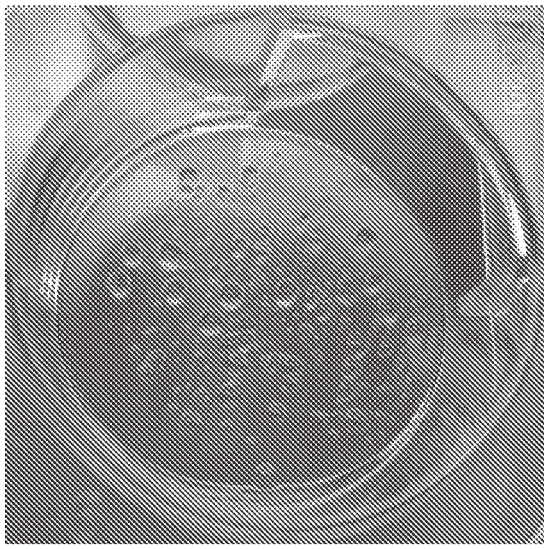


Fig. 32B

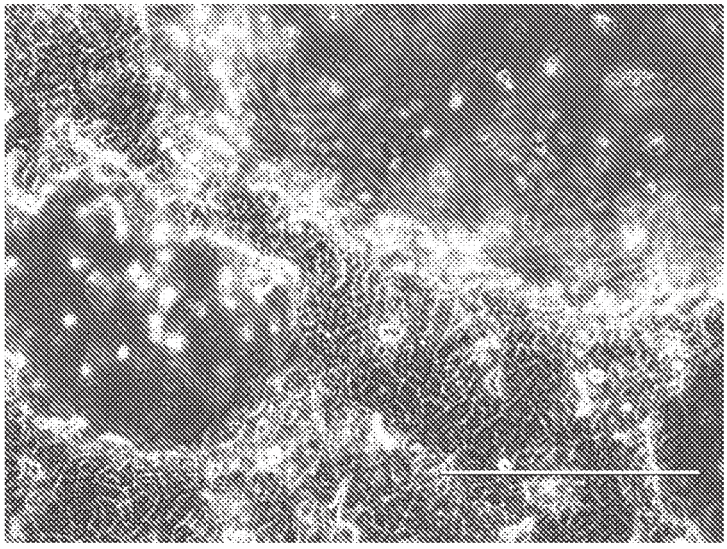


Fig. 32C

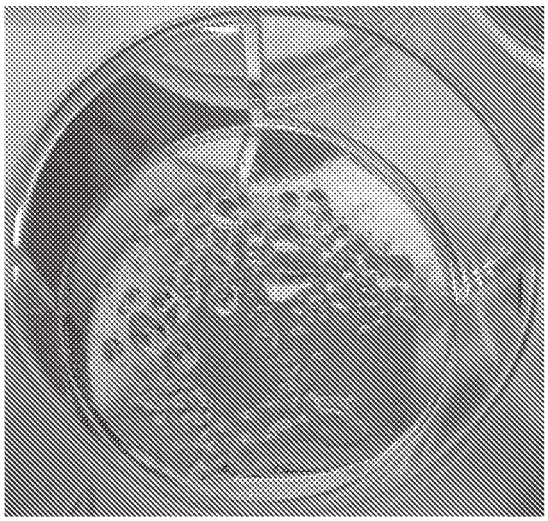
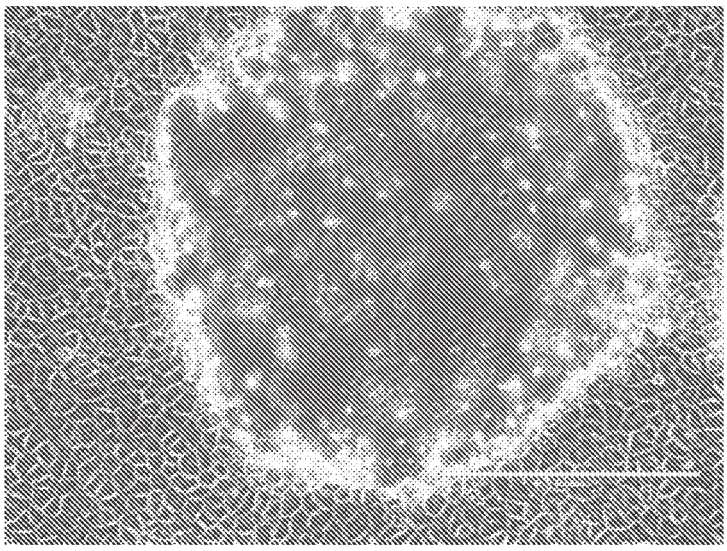
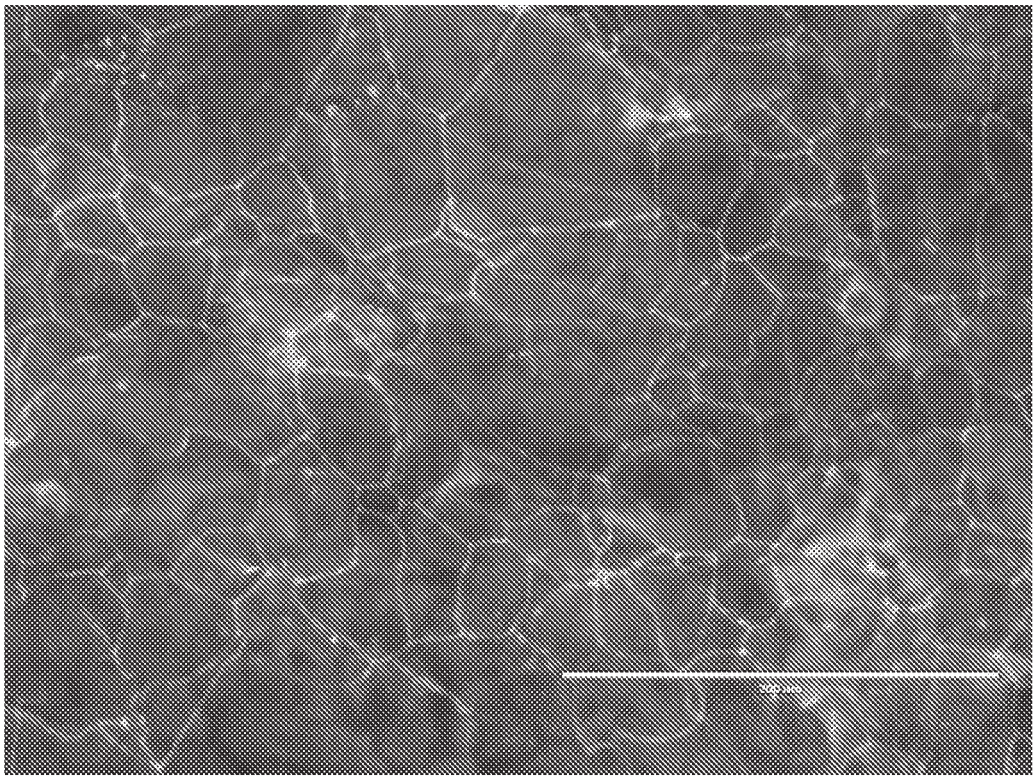


Fig. 32D





ZO-1 immunostaining

Fig. 33

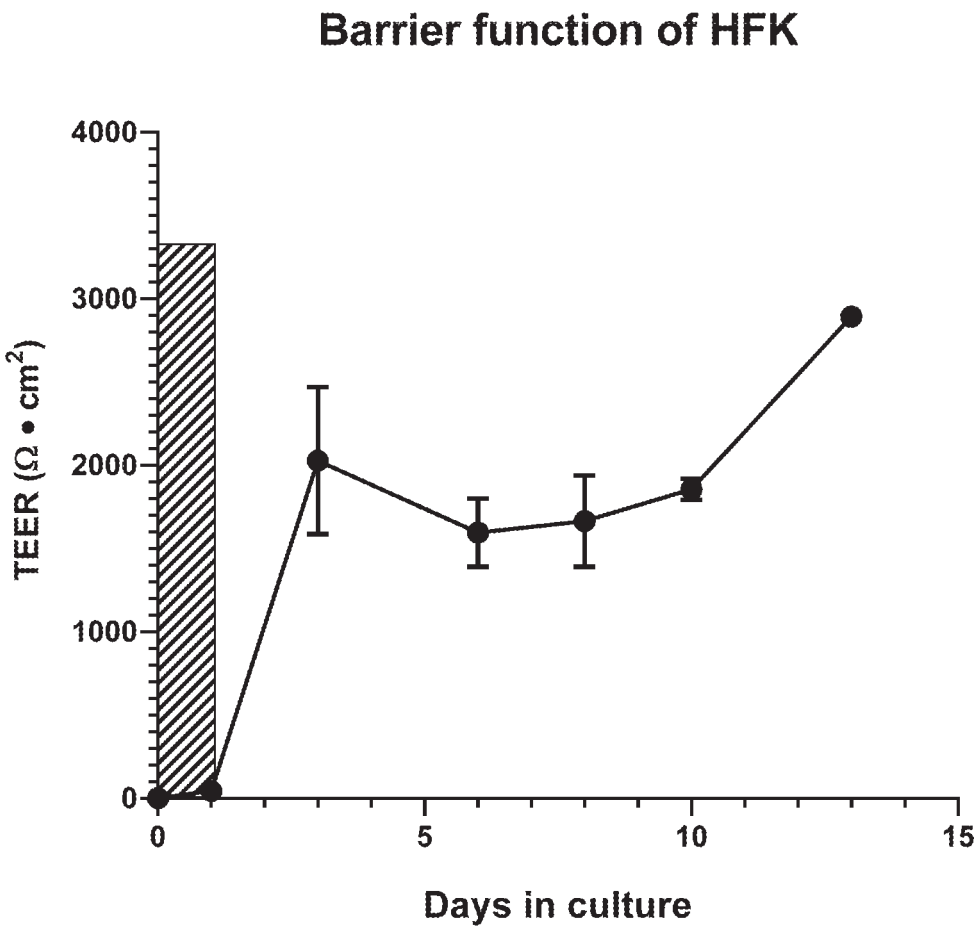

Barrier function of HFK
4000
3000
2000
1000
0
TEER (Ω • cm²)
0
5
10
15
Days in culture

Fig. 34

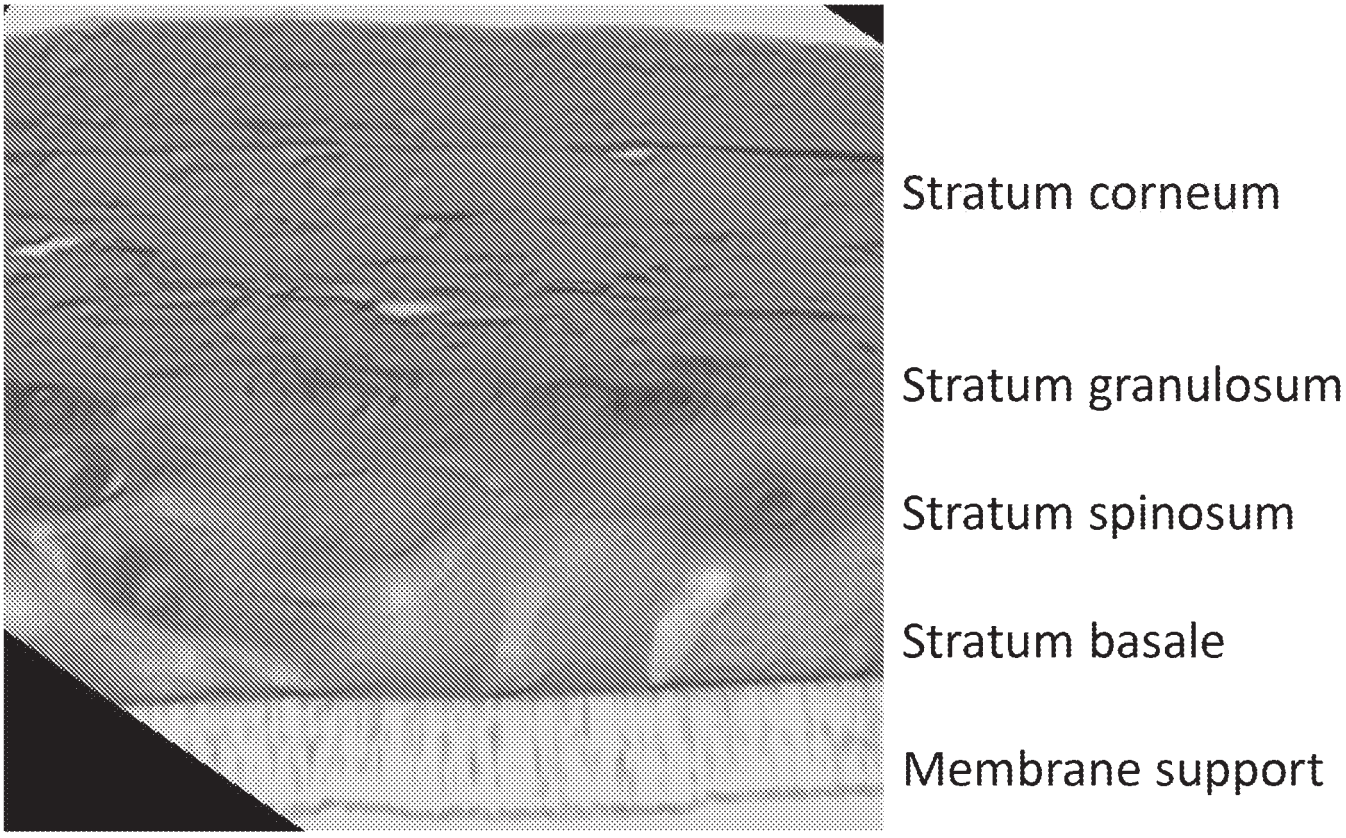

Stratum corneum
Stratum granulosum
Stratum spinosum
Stratum basale
Membrane support

Fig. 35

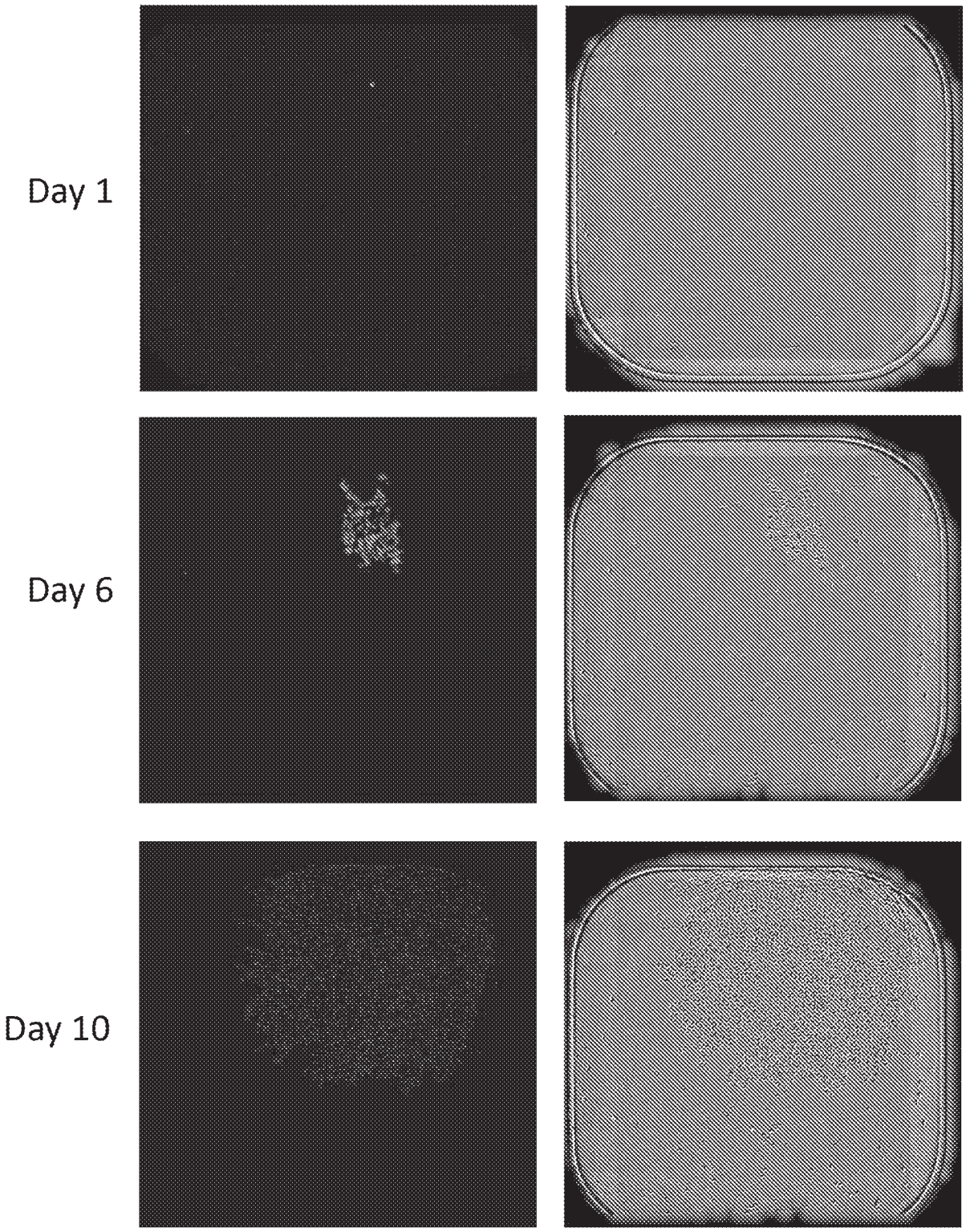


Fig. 36

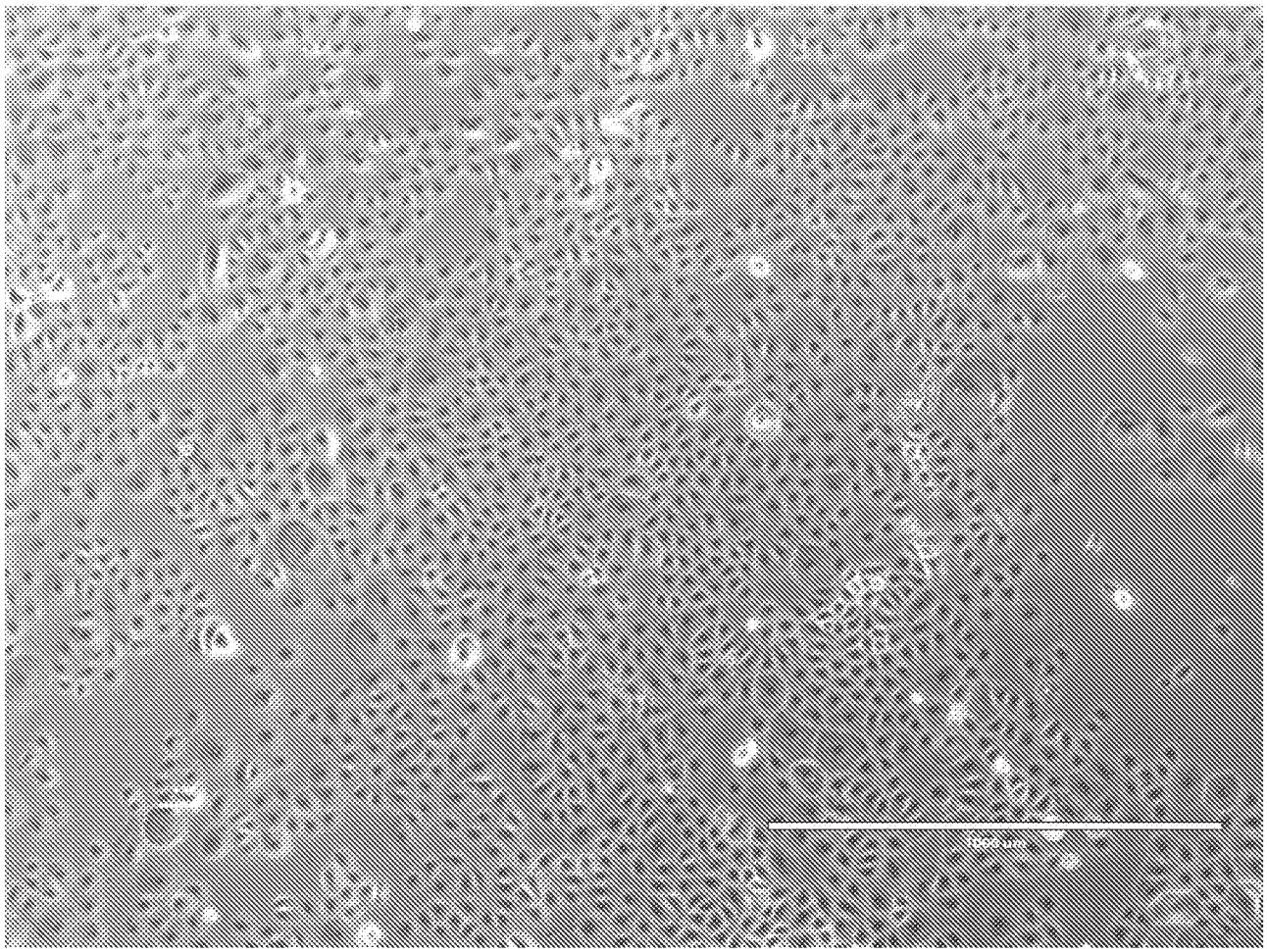


Fig. 37

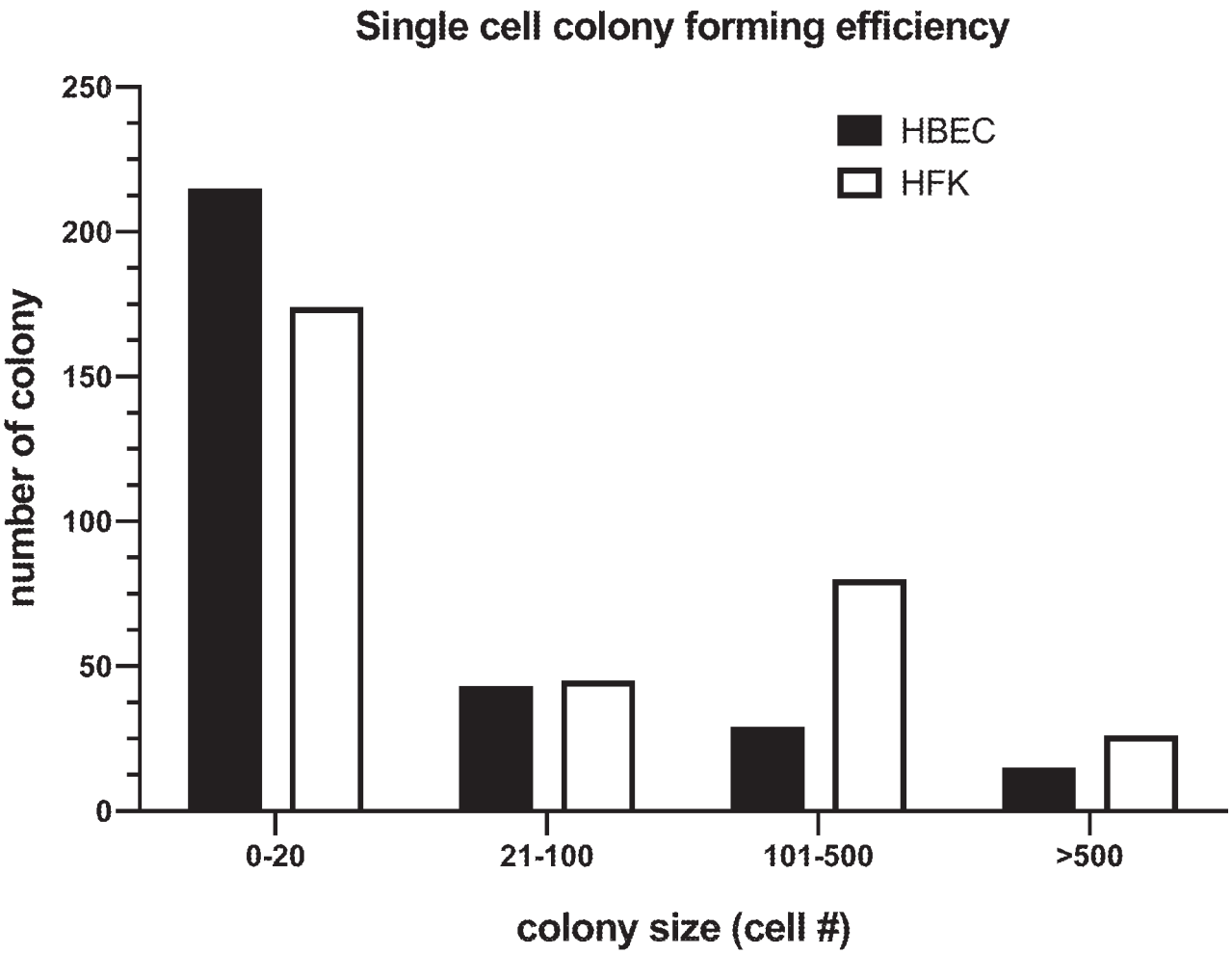

Single cell colony forming efficiency
HBEC
HFK
number of colony
250
200
150
100
50
0
0-20
21-100
101-500
>500
colony size (cell #)

Fig. 38