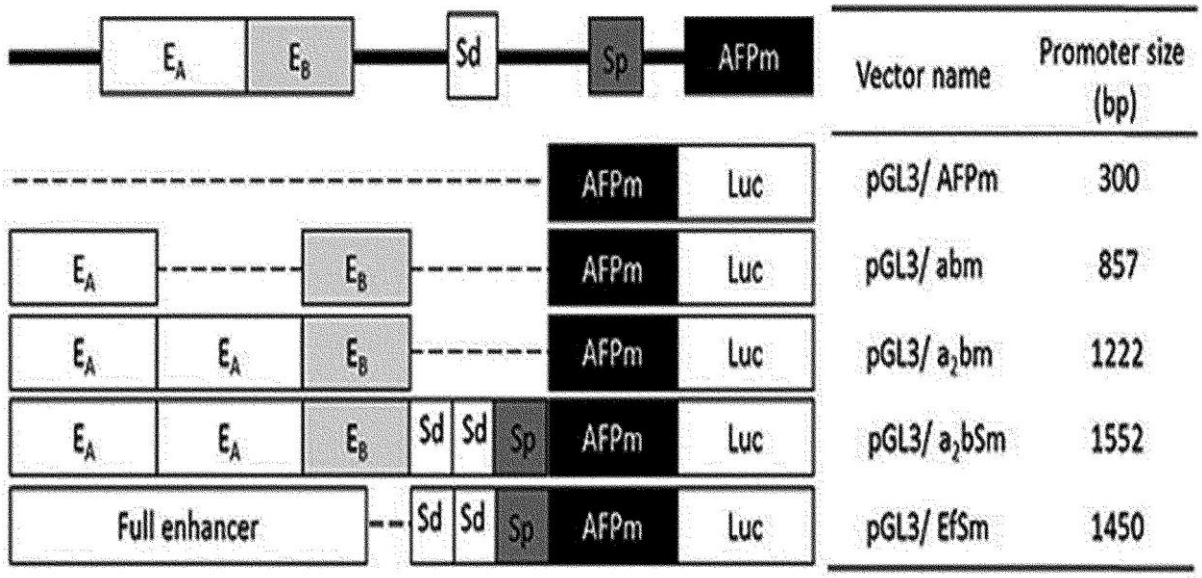
DRAWINGS

FIG. 1




| Vector name | Promoter size (bp) |
|---|---|
| pGL3/ AFPm | 300 |
| pGL3/ abm | 857 |
| pGL3/ $a_2$bm | 1222 |
| pGL3/ $a_2$bSm | 1552 |
| pGL3/ EfSm | 1450 |

FIG. 2

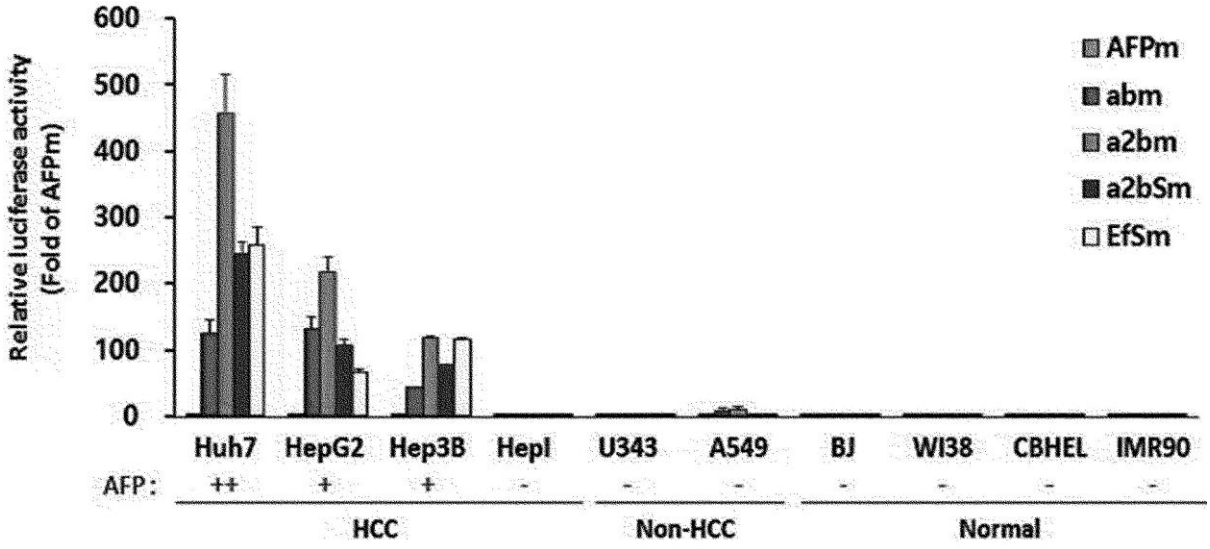


FIG. 3

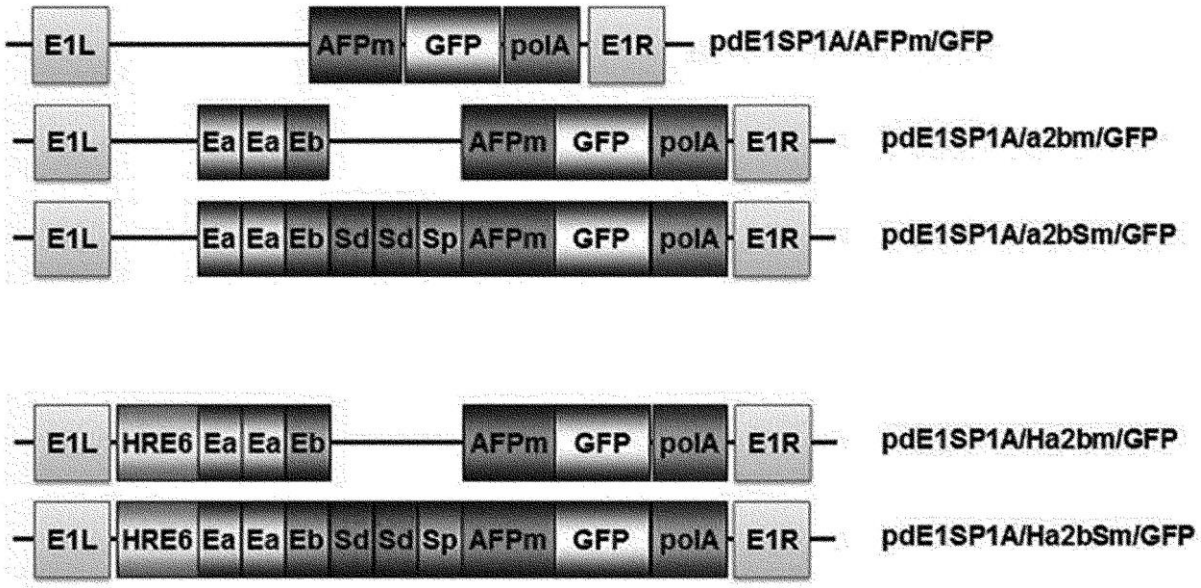

E1L
AFPm
GFP
polA
E1R
pdE1SP1A/AFPm/GFP
E1L
Ea
Ea
Eb
AFPm
GFP
polA
E1R
pdE1SP1A/a2bm/GFP
E1L
Ea
Ea
Eb
Sd
Sd
Sp
AFPm
GFP
polA
E1R
pdE1SP1A/a2bSm/GFP
E1L
HRE6
Ea
Ea
Eb
AFPm
GFP
polA
E1R
pdE1SP1A/Ha2bm/GFP
E1L
HRE6
Ea
Ea
Eb
Sd
Sd
Sp
AFPm
GFP
polA
E1R
pdE1SP1A/Ha2bSm/GFP

FIG. 4

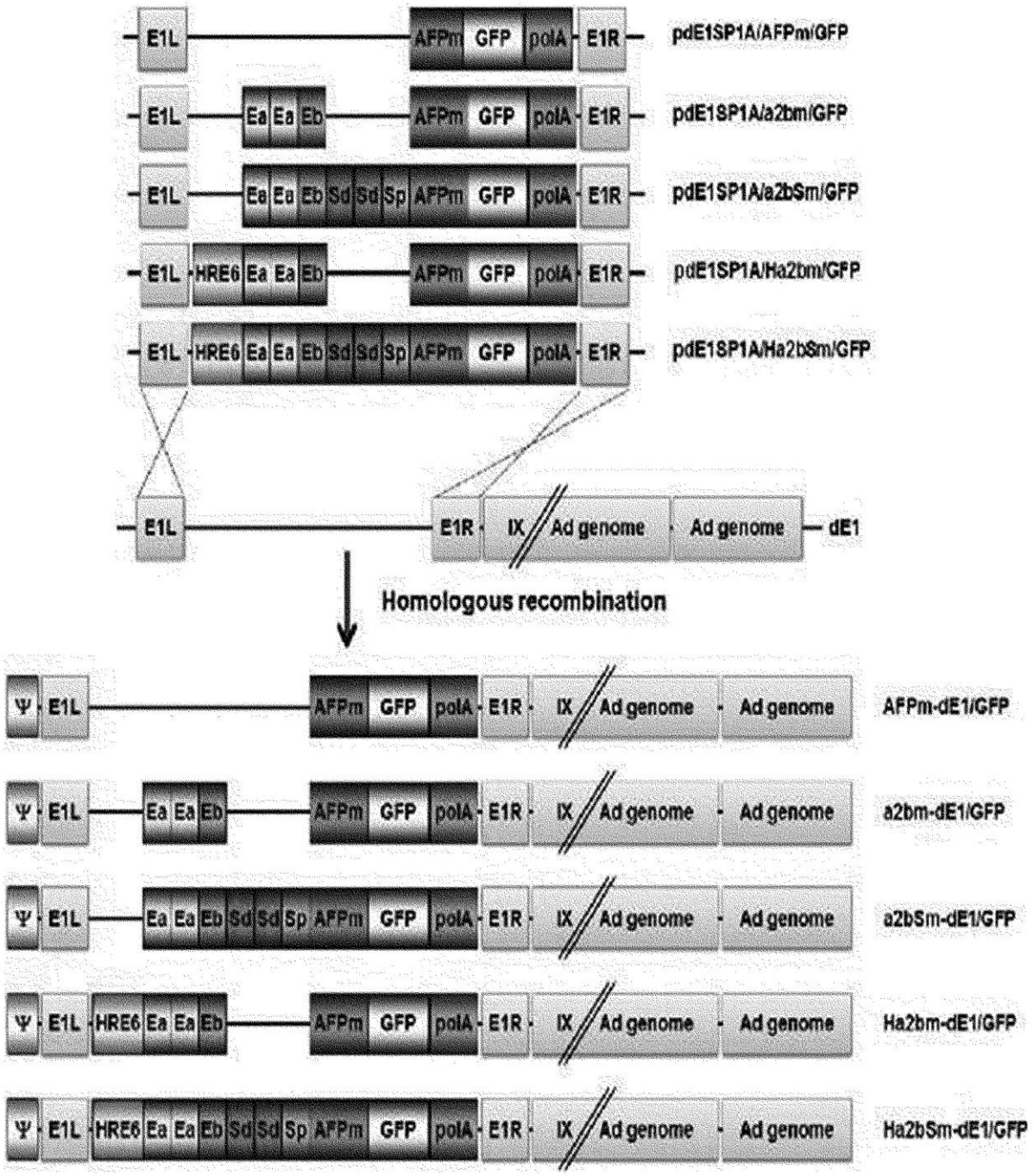

E1L
AFPm
GFP
polA
E1R
pdE1SP1A/AFPm/GFP
Ea
Eb
pdE1SP1A/a2bm/GFP
Sd
Sp
pdE1SP1A/a2bSm/GFP
HRE6
pdE1SP1A/Ha2bm/GFP
pdE1SP1A/Ha2bSm/GFP
IX
Ad genome
dE1
Homologous recombination
Ψ
AFPm-dE1/GFP
a2bm-dE1/GFP
a2bSm-dE1/GFP
Ha2bm-dE1/GFP
Ha2bSm-dE1/GFP

FIG. 5

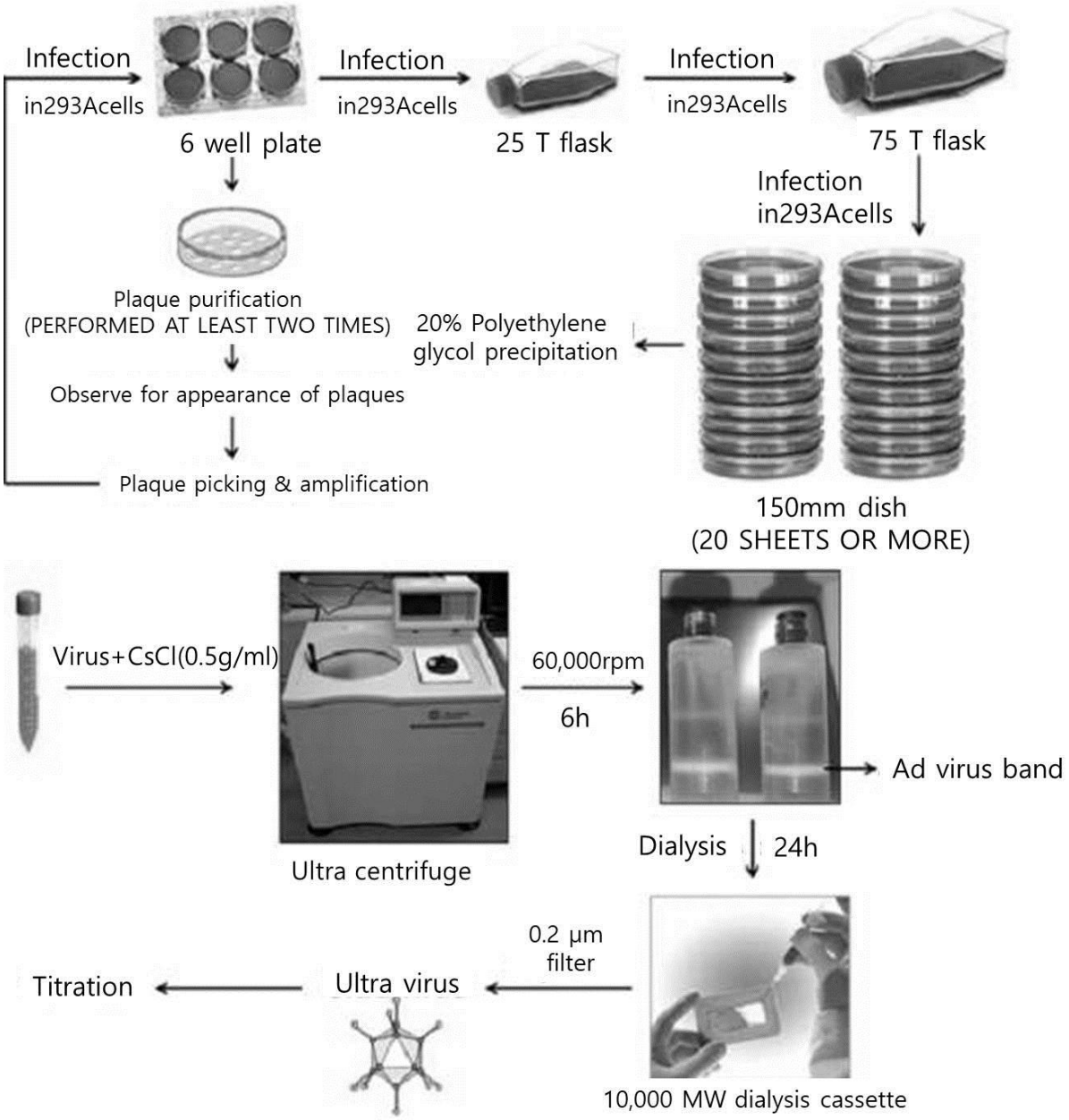

Infection
in293Acells
6 well plate
Infection
in293Acells
25 T flask
Infection
in293Acells
75 T flask
Infection
in293Acells
150mm dish
(20 SHEETS OR MORE)
20% Polyethylene
glycol precipitation
Plaque purification
(PERFORMED AT LEAST TWO TIMES)
Observe for appearance of plaques
Plaque picking & amplification
Virus+CsCl(0.5g/ml)
60,000rpm
6h
Ultra centrifuge
Ad virus band
Dialysis
24h
10,000 MW dialysis cassette
0.2 μm
filter
Ultra virus
Titration

FIG. 6A

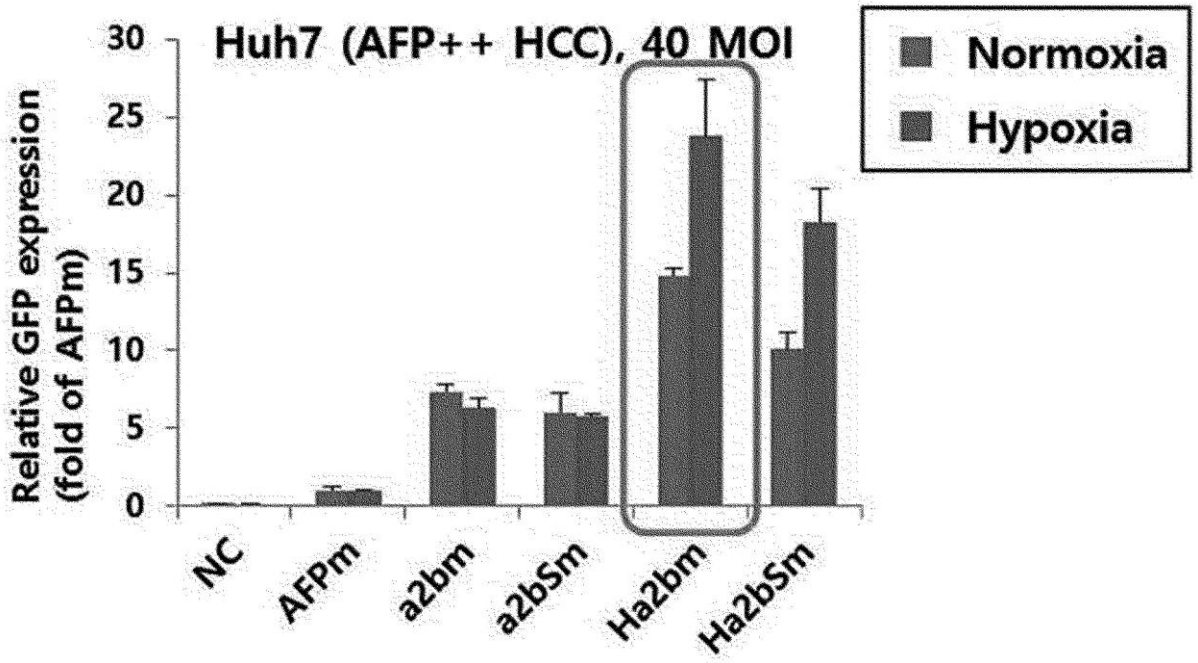

Huh7 (AFP++ HCC), 40 MOI
Relative GFP expression (fold of AFPm)
30
25
20
15
10
5
0
NC
AFPm
a2bm
a2bSm
Ha2bm
Ha2bSm
Normoxia
Hypoxia

FIG. 6B

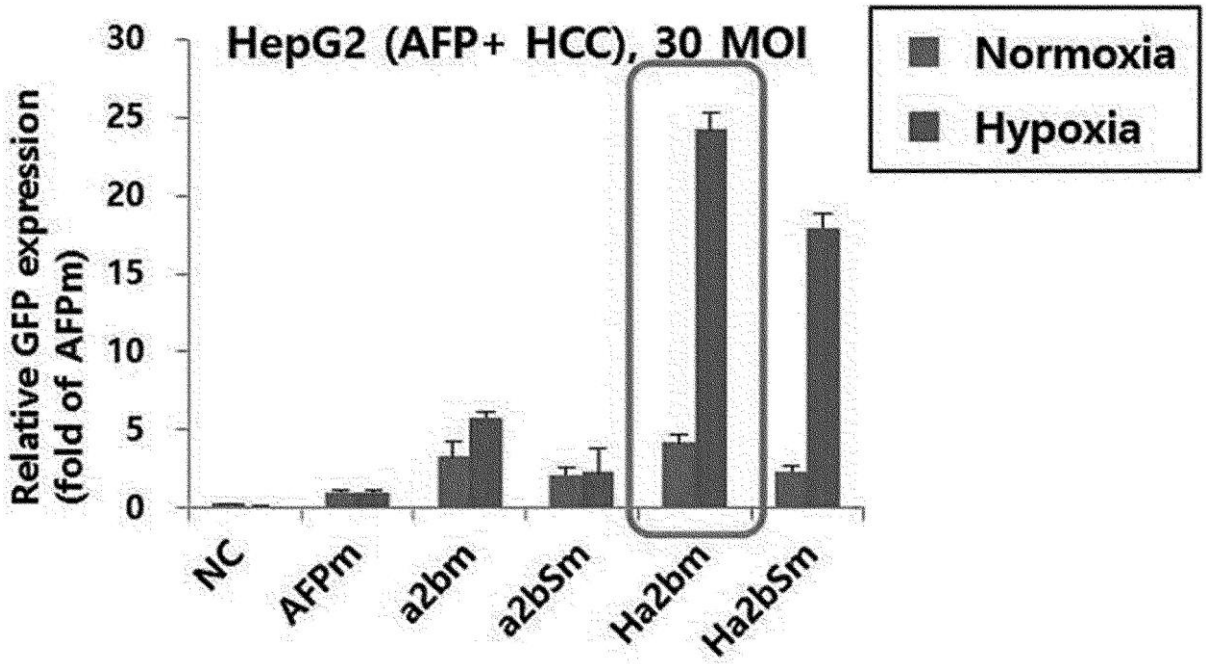

HepG2 (AFP+ HCC), 30 MOI
Relative GFP expression (fold of AFPm)
30
25
20
15
10
5
0
NC
AFPm
a2bm
a2bSm
Ha2bm
Ha2bSm
Normoxia
Hypoxia

FIG. 6C

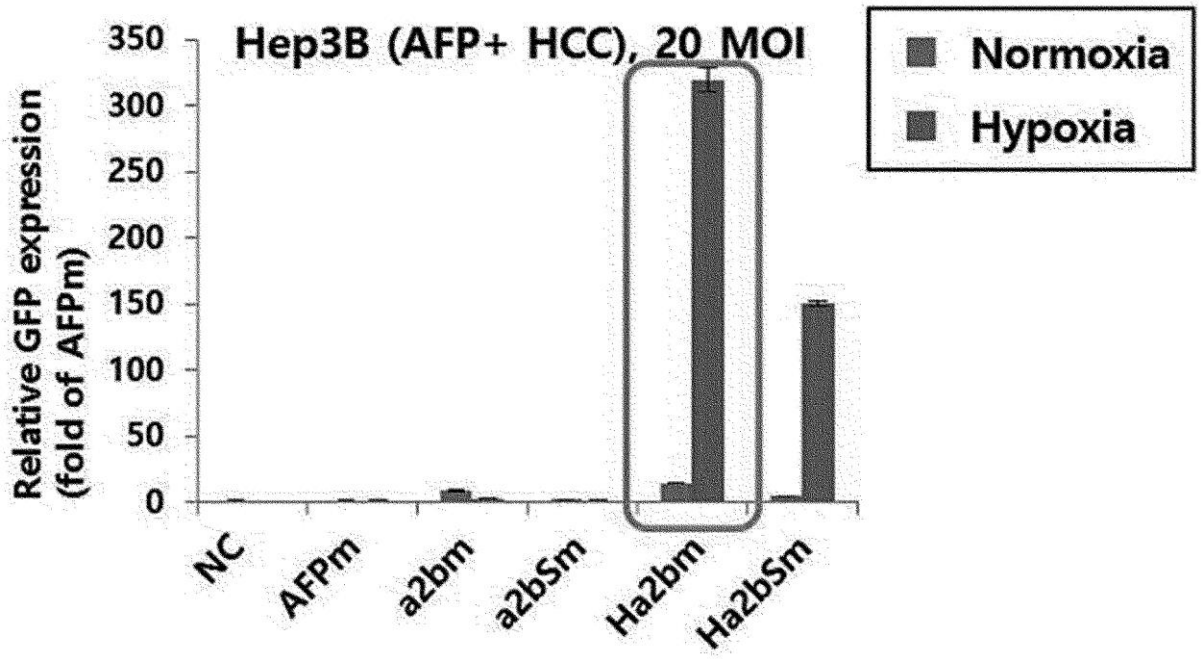

Hep3B (AFP+ HCC), 20 MOI
Relative GFP expression (fold of AFPm)
350
300
250
200
150
100
50
0
NC
AFPm
a2bm
a2bSm
Ha2bm
Ha2bSm
Normoxia
Hypoxia

FIG. 6D

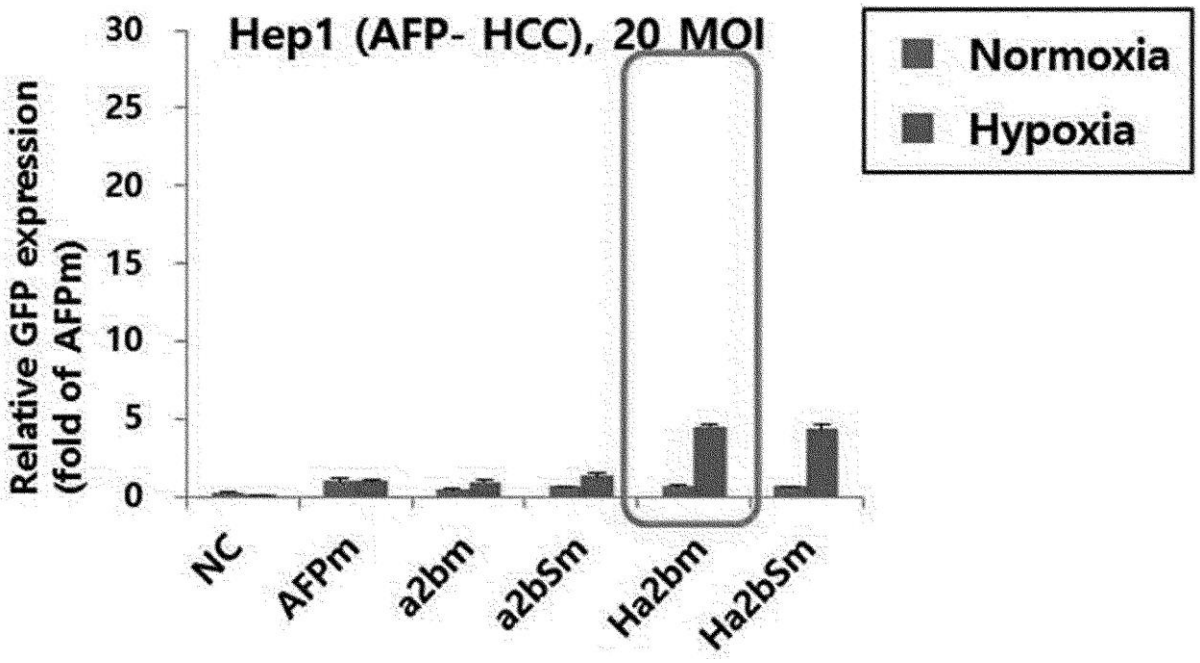

Hep1 (AFP- HCC), 20 MOI
Relative GFP expression (fold of AFPm)
30
25
20
15
10
5
0
NC
AFPm
a2bm
a2bSm
Ha2bm
Ha2bSm
Normoxia
Hypoxia

FIG. 6E

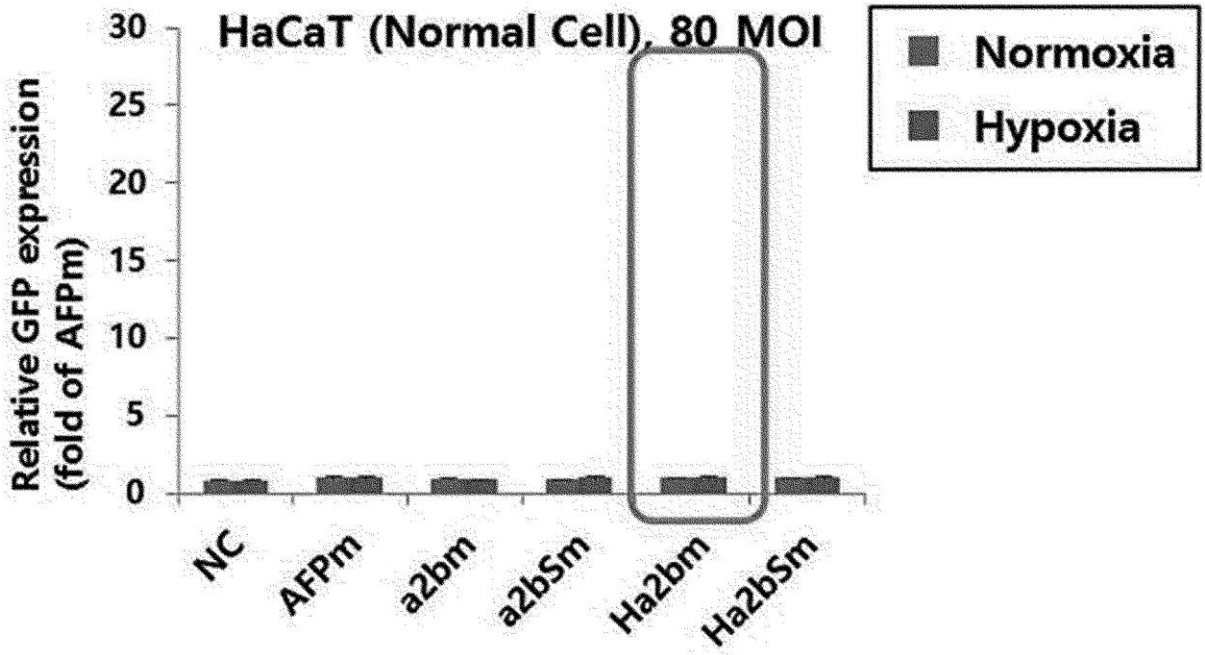

HaCaT (Normal Cell), 80 MOI
Relative GFP expression (fold of AFPm)
30
25
20
15
10
5
0
NC
AFPm
a2bm
a2bSm
Ha2bm
Ha2bSm
Normoxia
Hypoxia

FIG. 6F

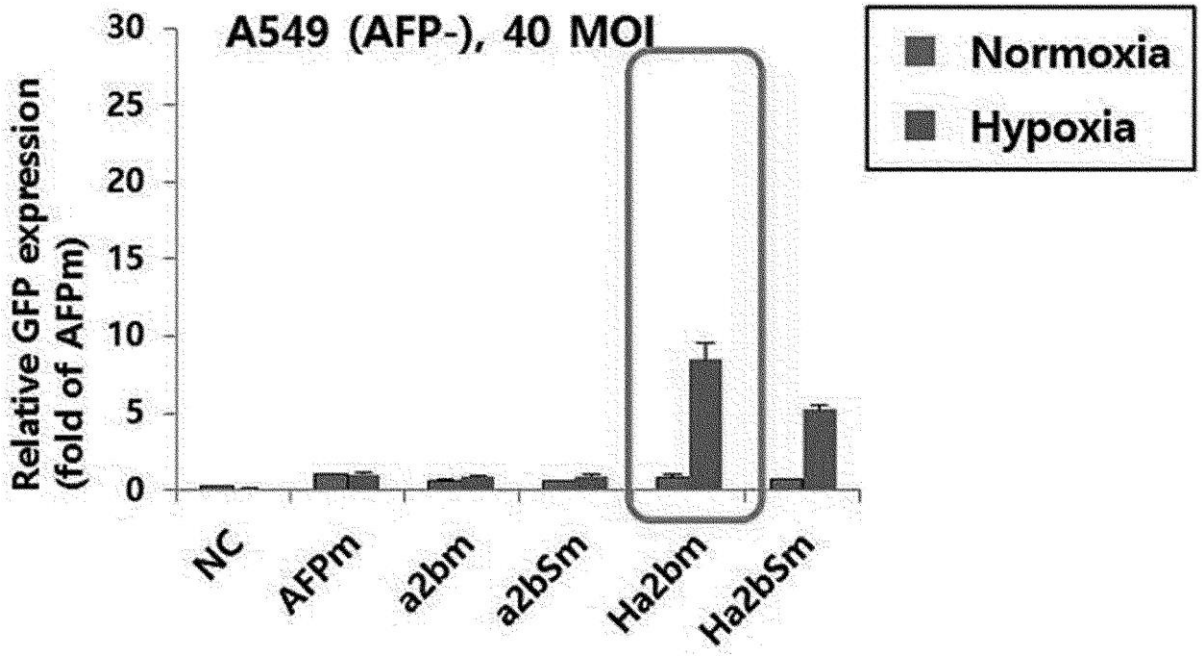

A549 (AFP-), 40 MOI
Relative GFP expression (fold of AFPm)
30
25
20
15
10
5
0
NC
AFPm
a2bm
a2bSm
Ha2bm
Ha2bSm
Normoxia
Hypoxia

FIG. 7

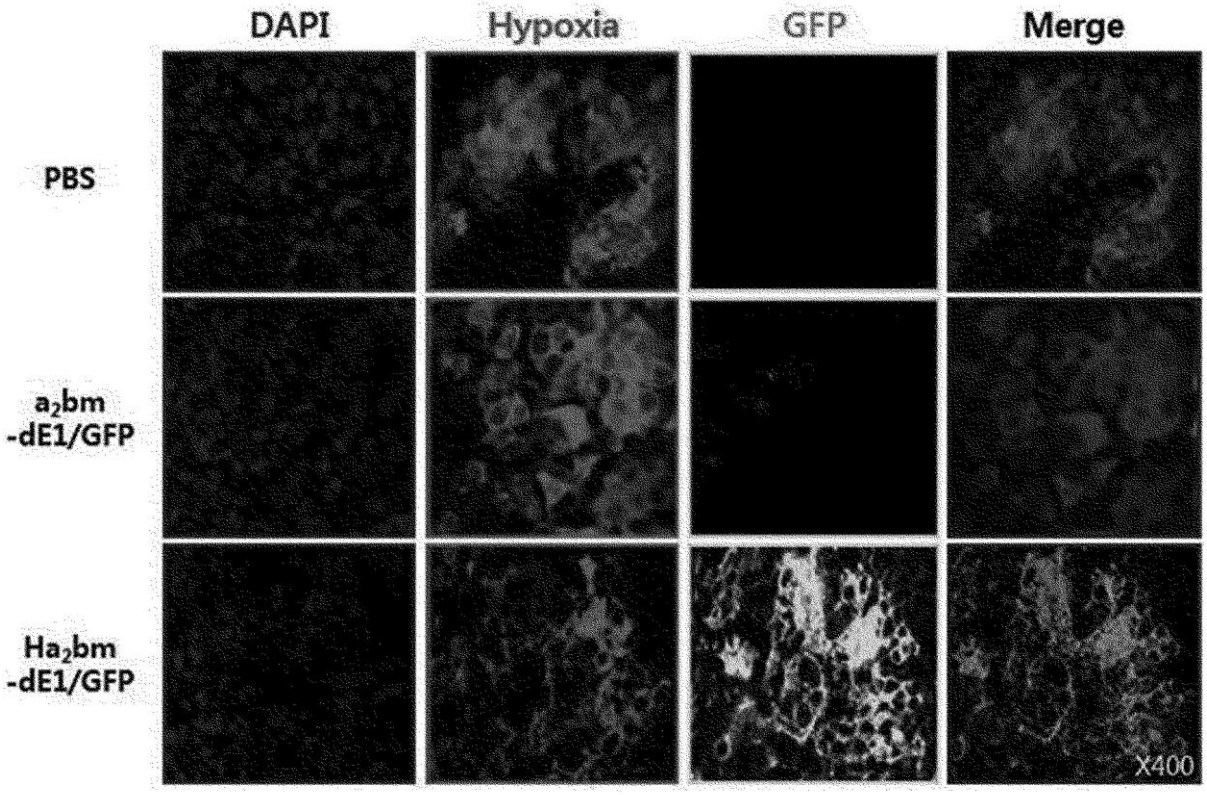


FIG. 8

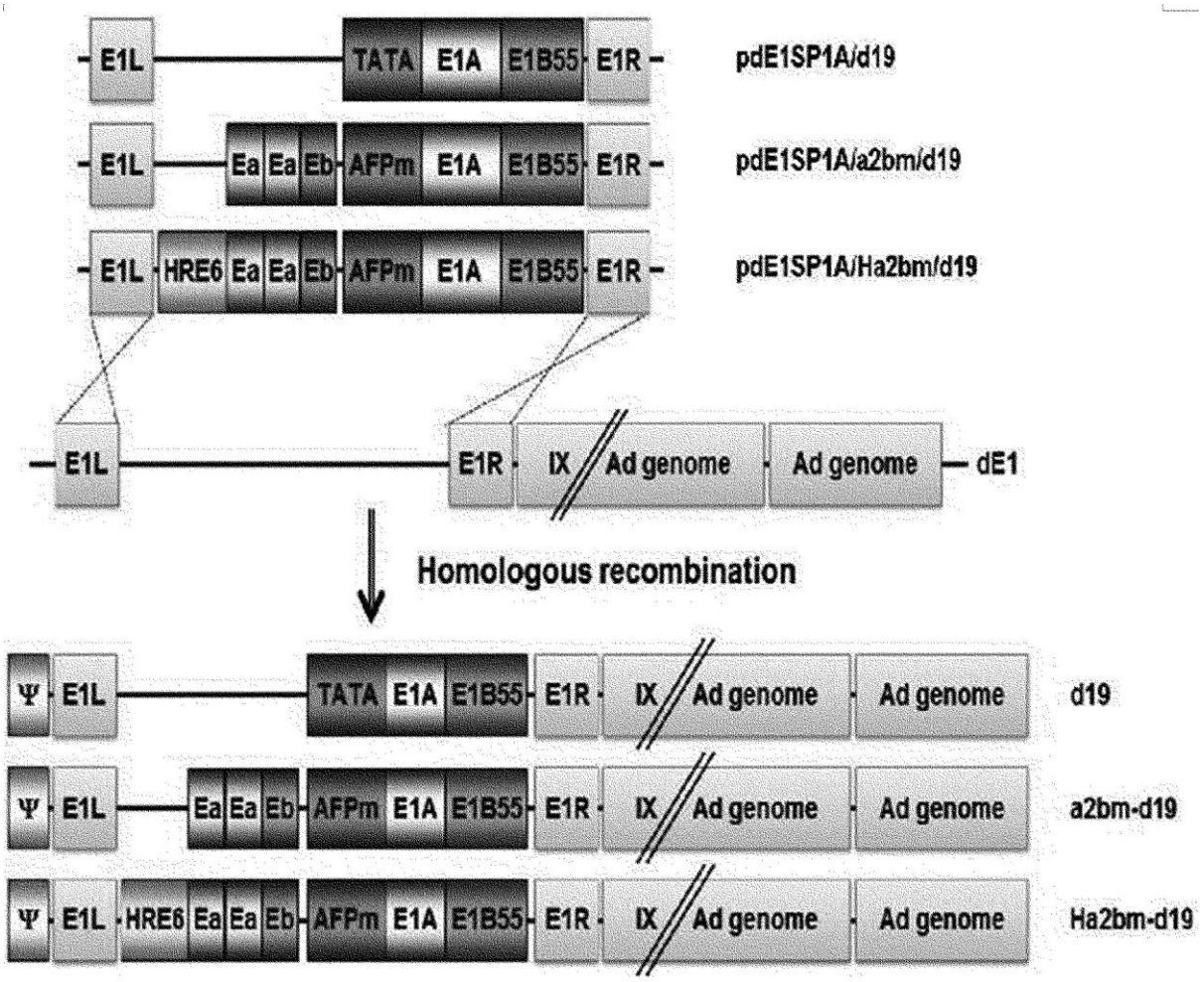

E1L
TATA
E1A
E1B55
E1R
pdE1SP1A/d19
Ea
Ea
Eb
AFPm
pdE1SP1A/a2bm/d19
HRE6
pdE1SP1A/Ha2bm/d19
IX
Ad genome
dE1
Homologous recombination
Ψ
d19
a2bm-d19
Ha2bm-d19

FIG. 9

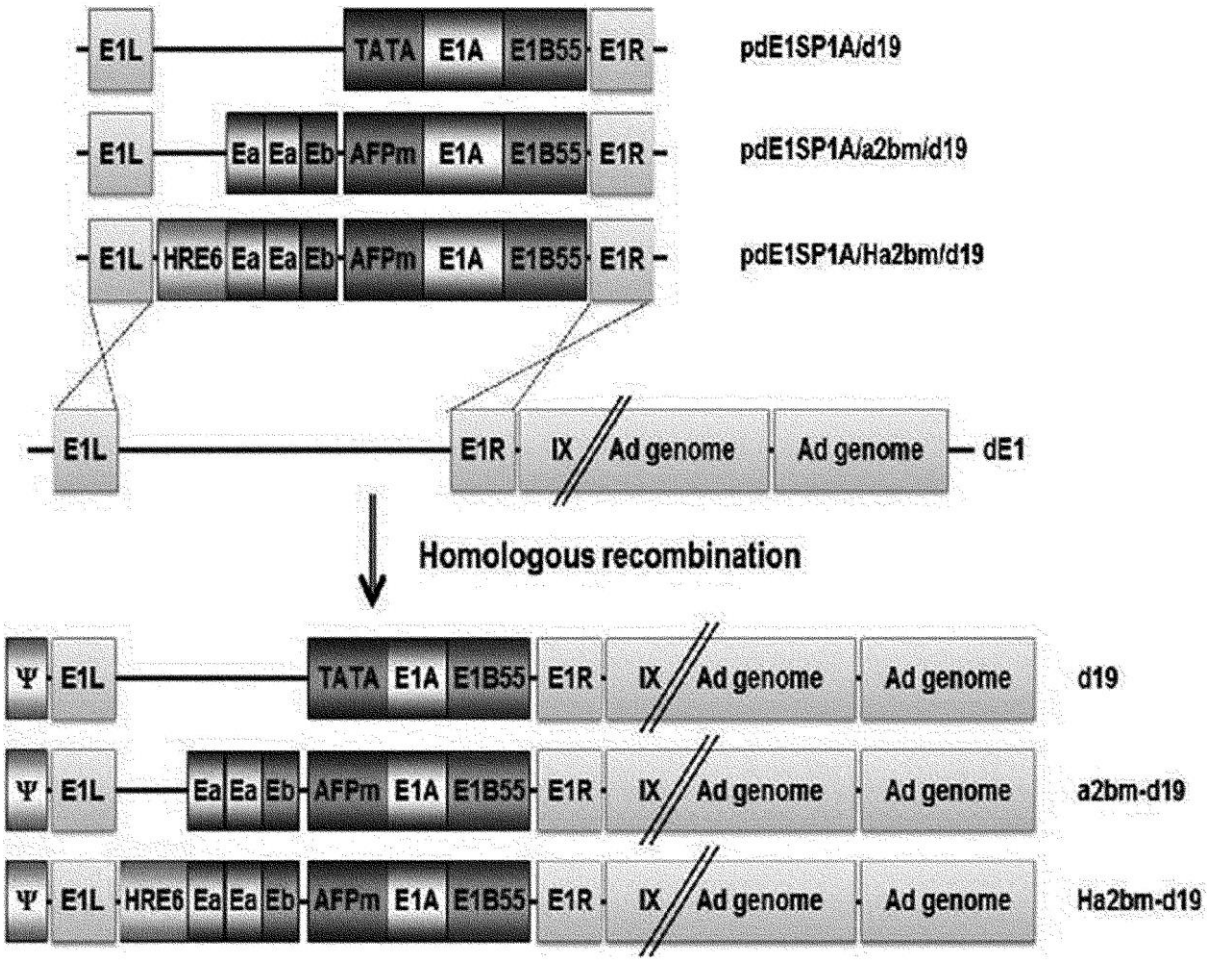

E1L
TATA
E1A
E1B55
E1R
pdE1SP1A/d19
Ea
Ea
Eb
AFPm
pdE1SP1A/a2bm/d19
HRE6
pdE1SP1A/Ha2bm/d19
IX
Ad genome
dE1
Homologous recombination
Ψ
d19
a2bm-d19
Ha2bm-d19

FIG. 10A

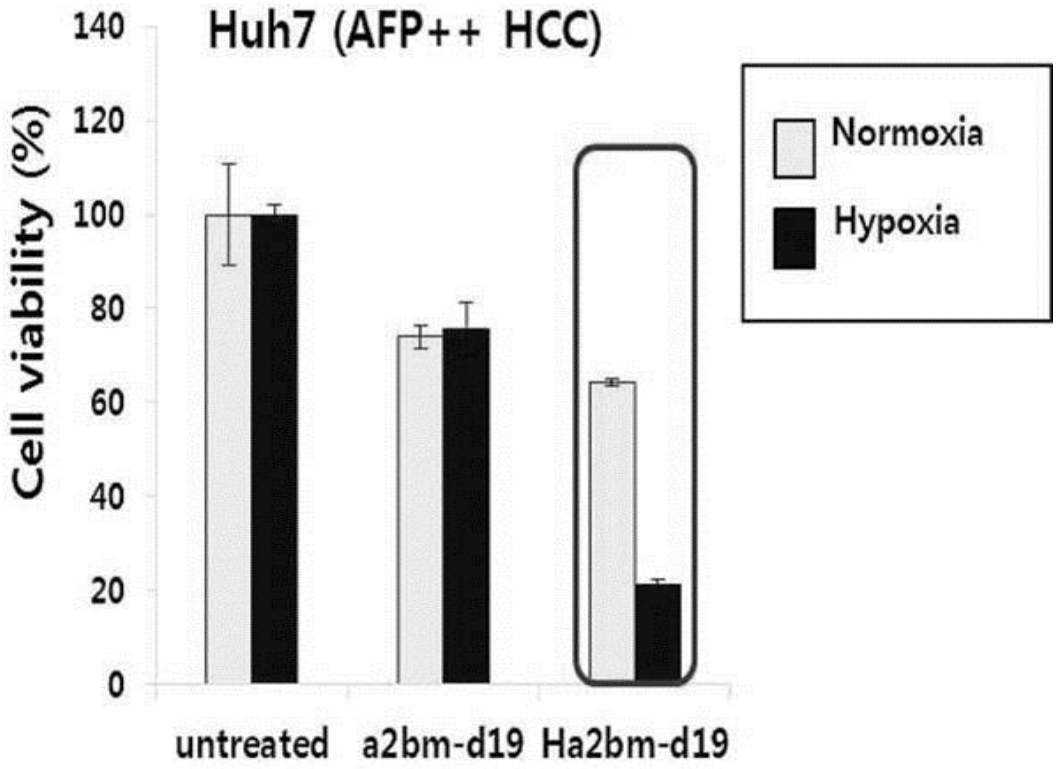



FIG. 10B

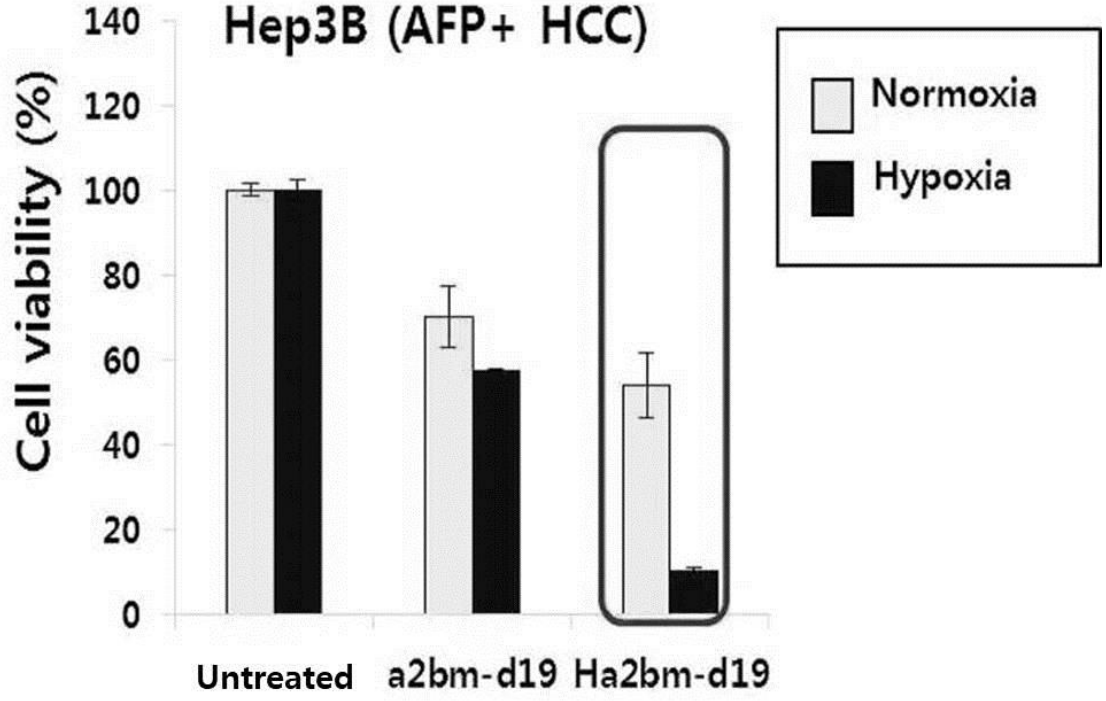


FIG. 10C

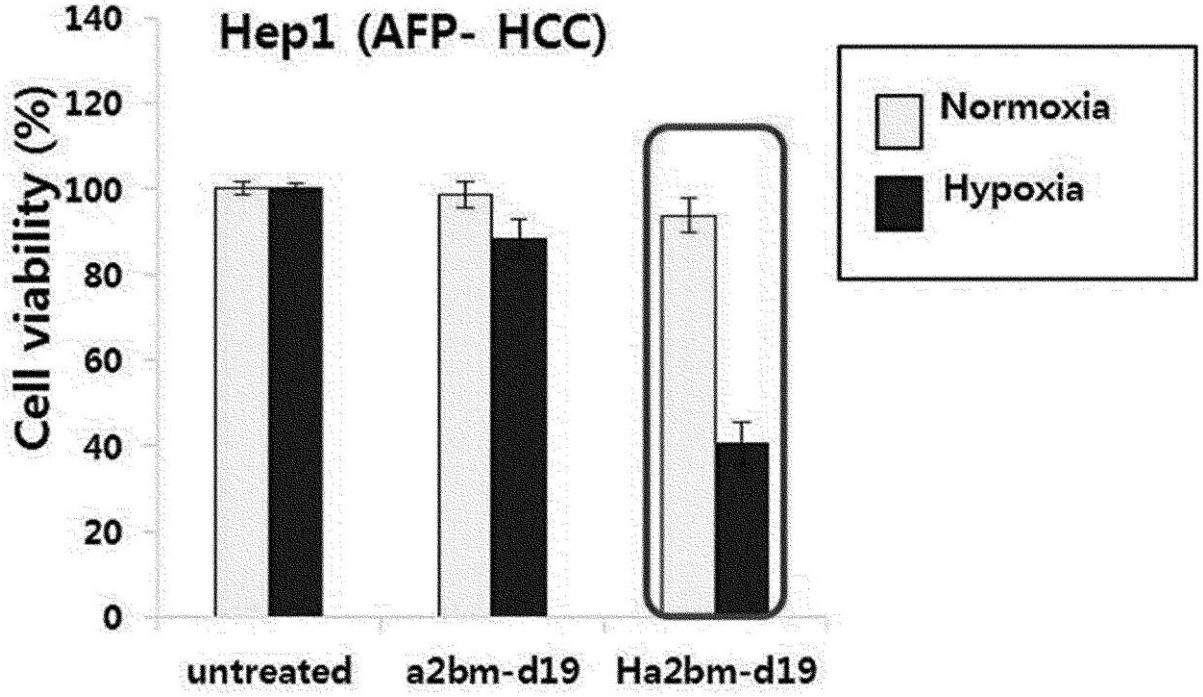



FIG. 10D

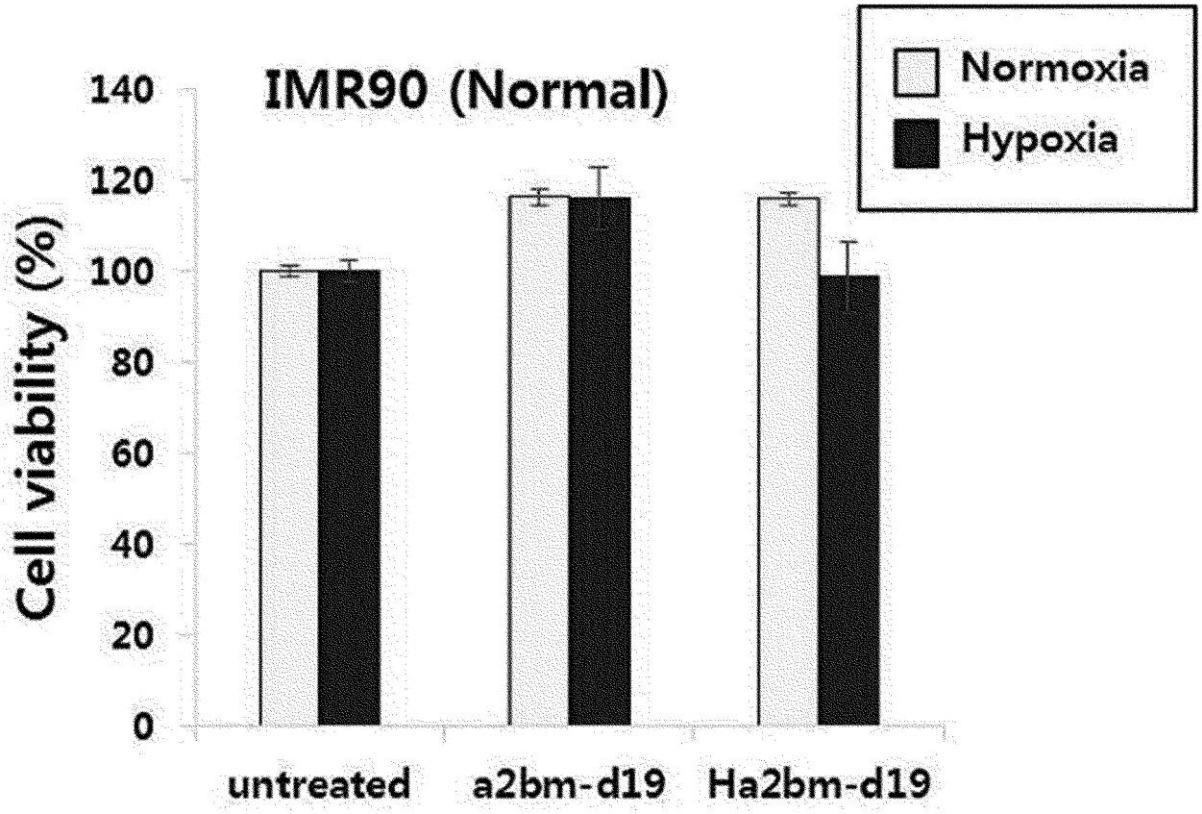


FIG. 10E

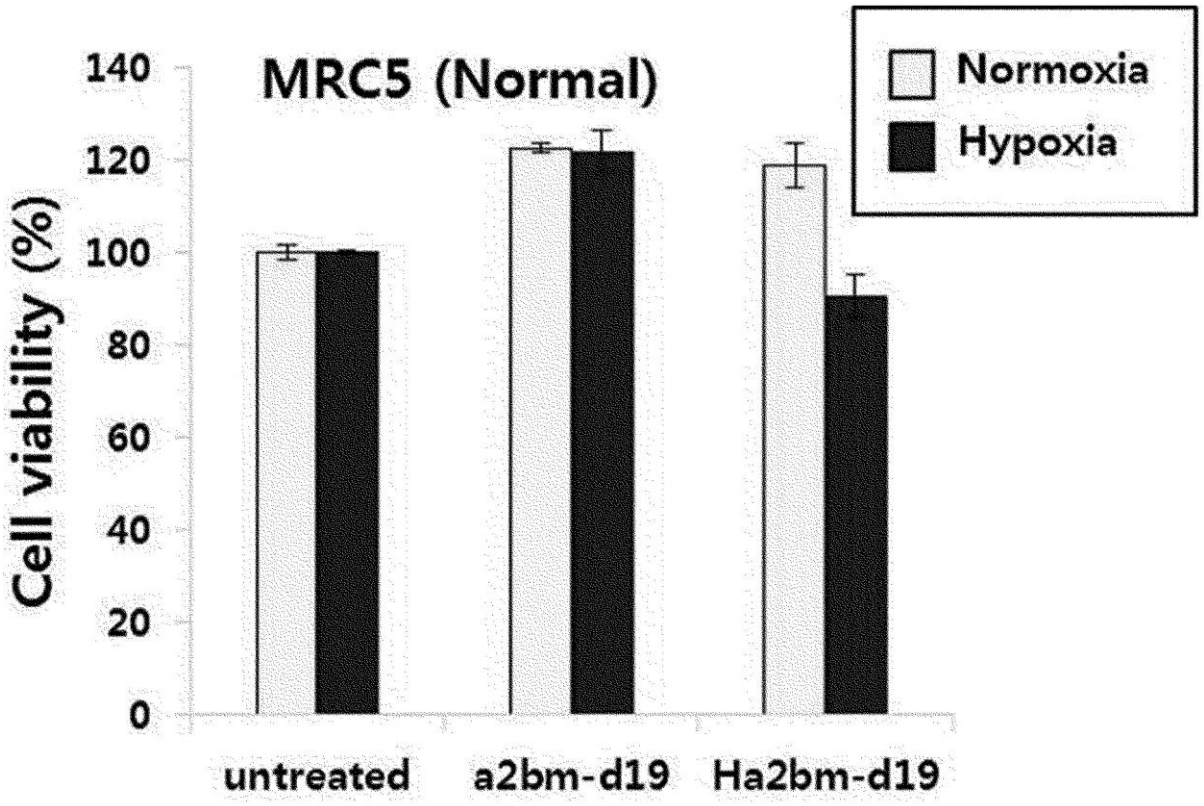

MRC5 (Normal)
Normoxia
Hypoxia
Cell viability (%)
140
120
100
80
60
40
20
0
untreated
a2bm-d19
Ha2bm-d19

FIG. 11

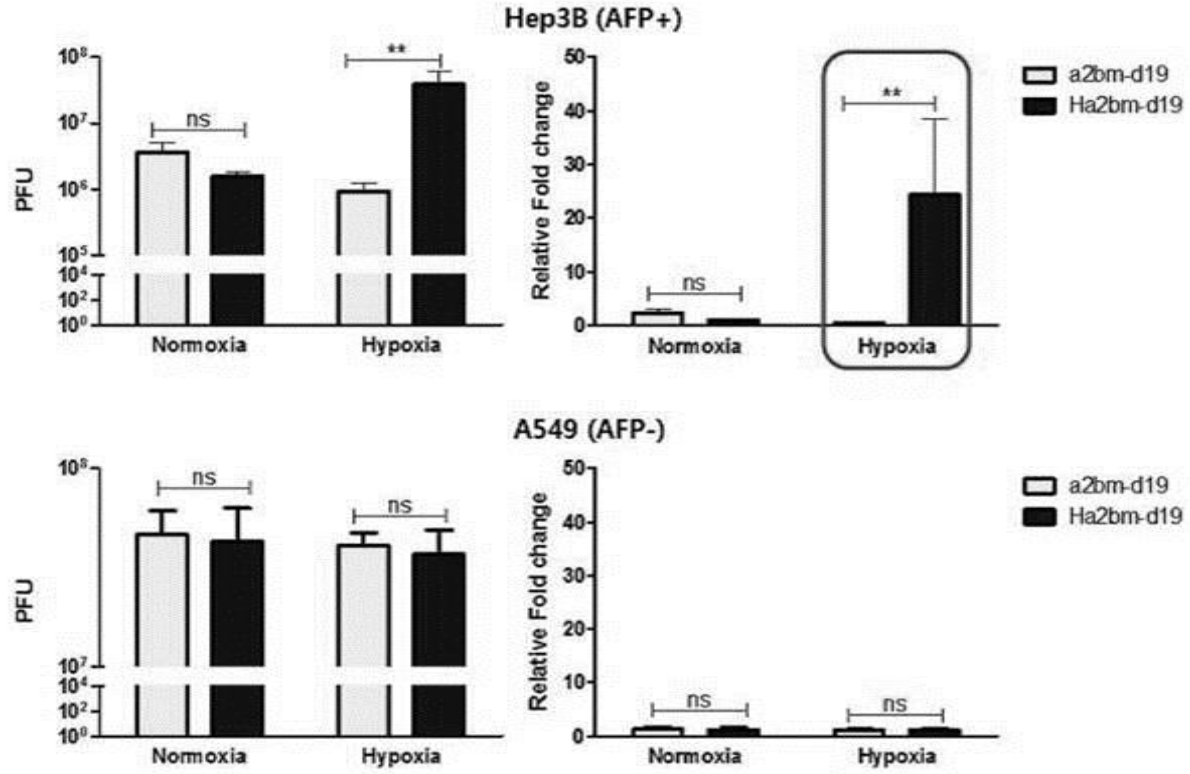

Hep3B (AFP+)
PFU
Normoxia
Hypoxia
ns
**
Relative Fold change
a2bm-d19
Ha2bm-d19
A549 (AFP-)

FIG. 12

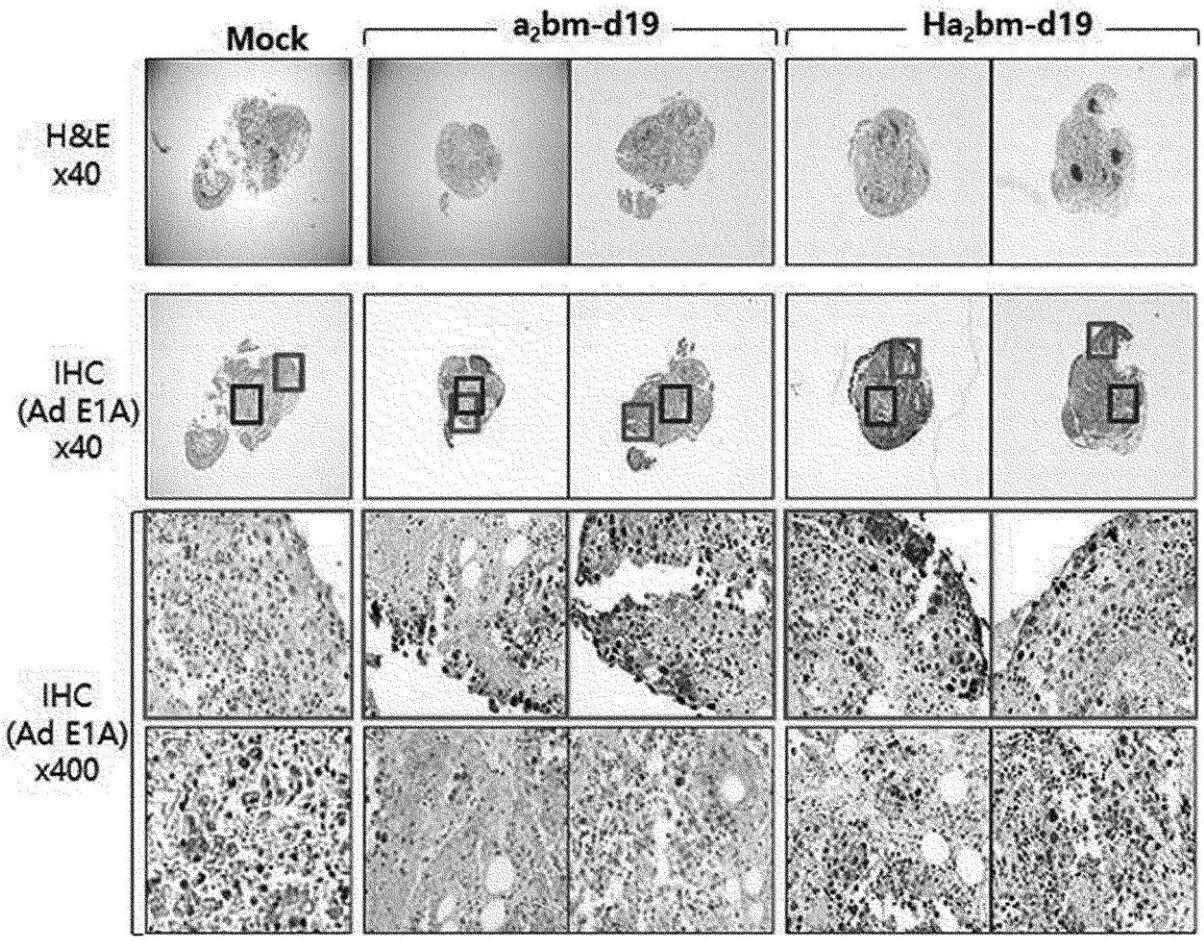


FIG. 13

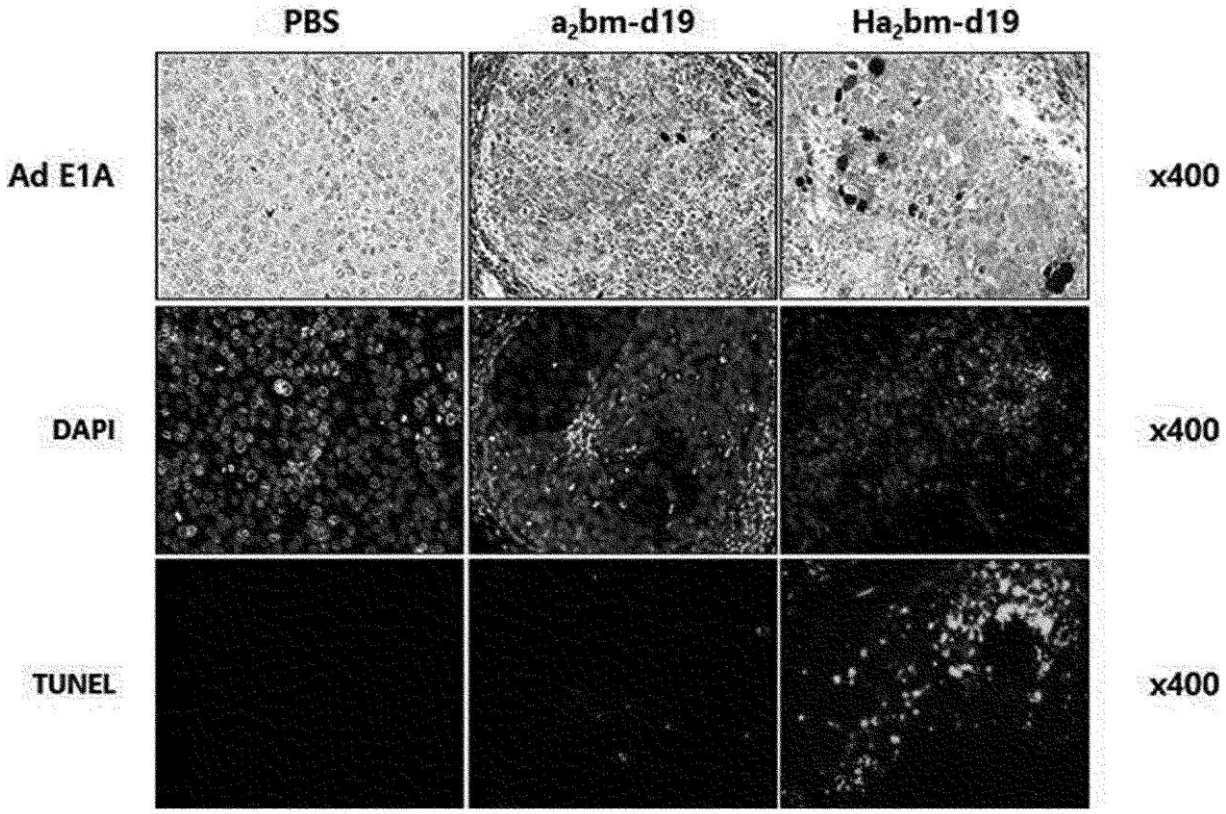


FIG. 14

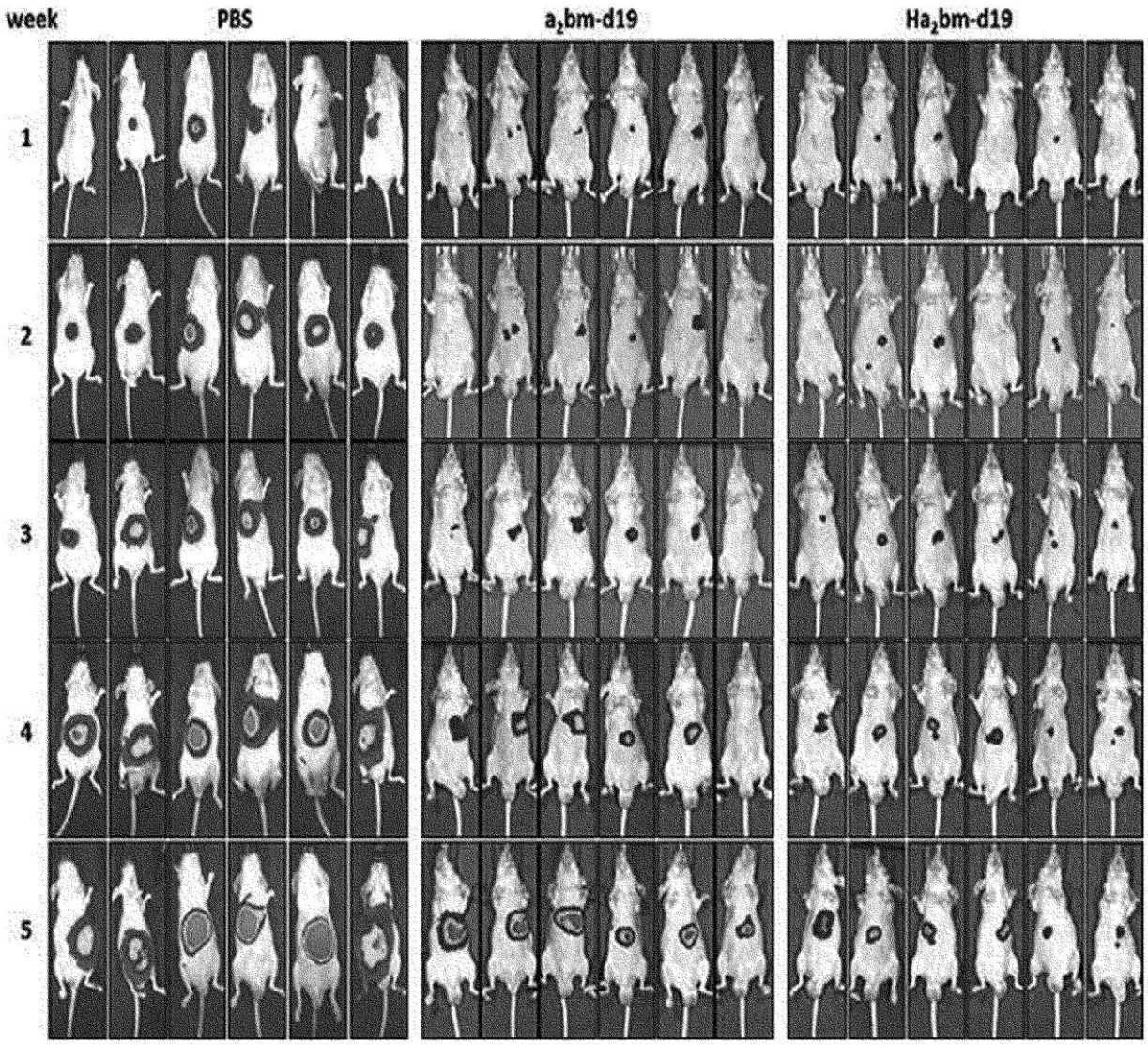


FIG. 15

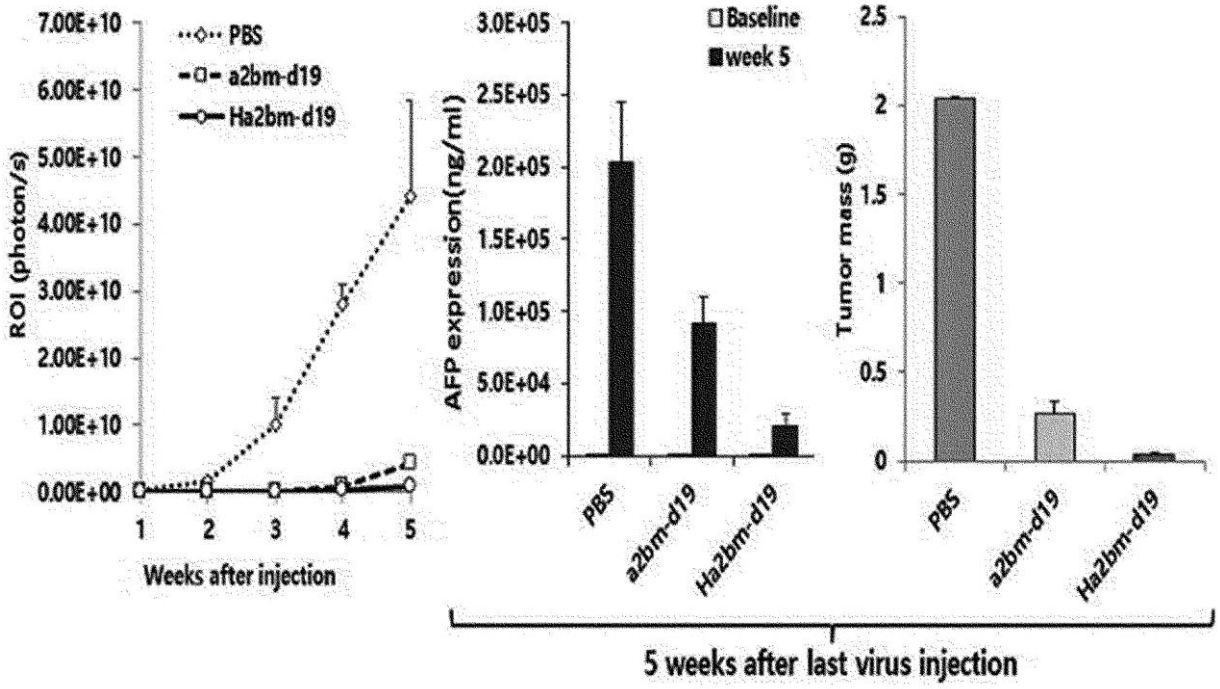



FIG. 16A

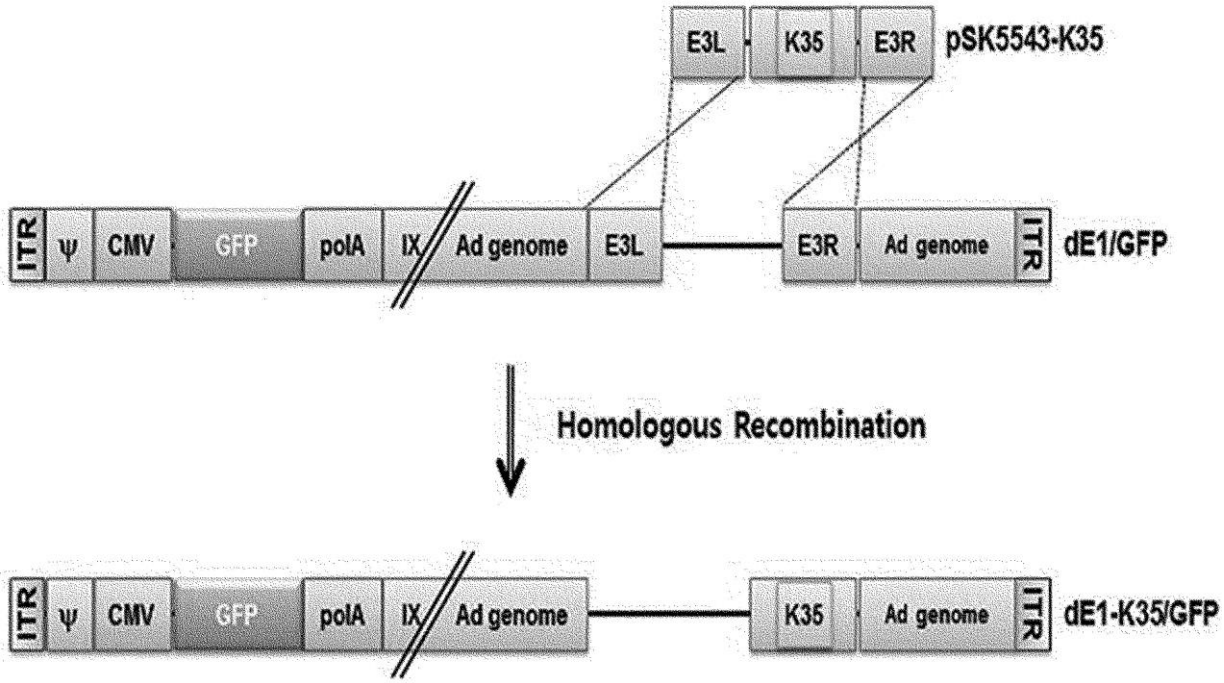


FIG. 16B

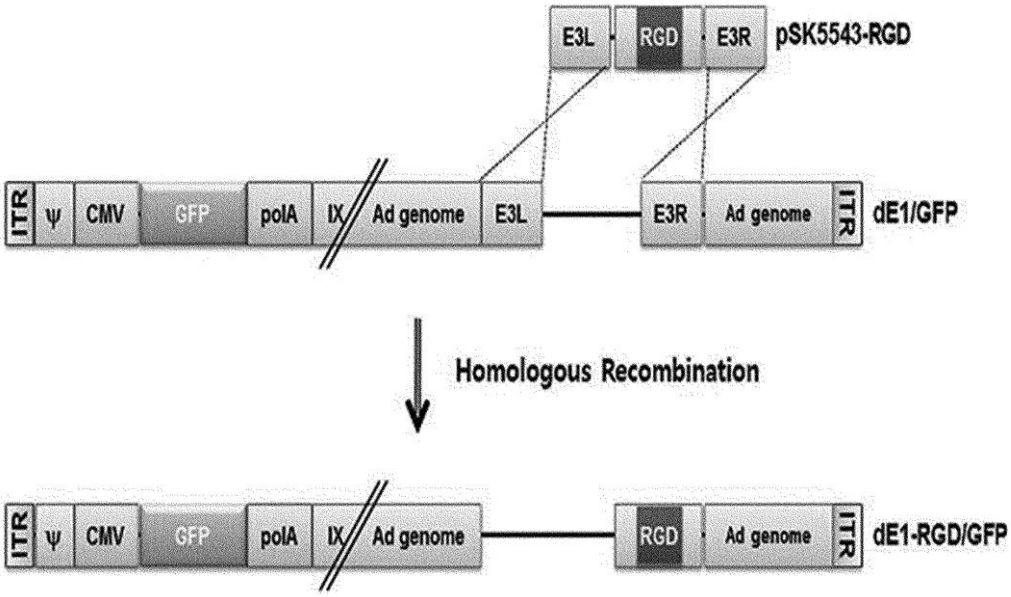



FIG. 16C

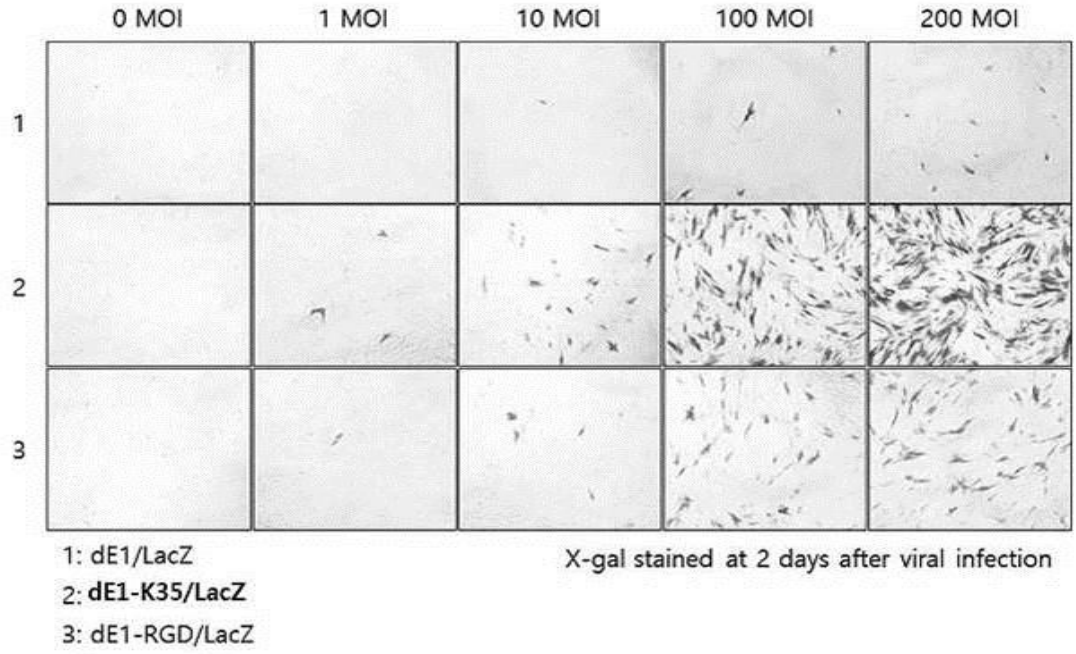


FIG. 17

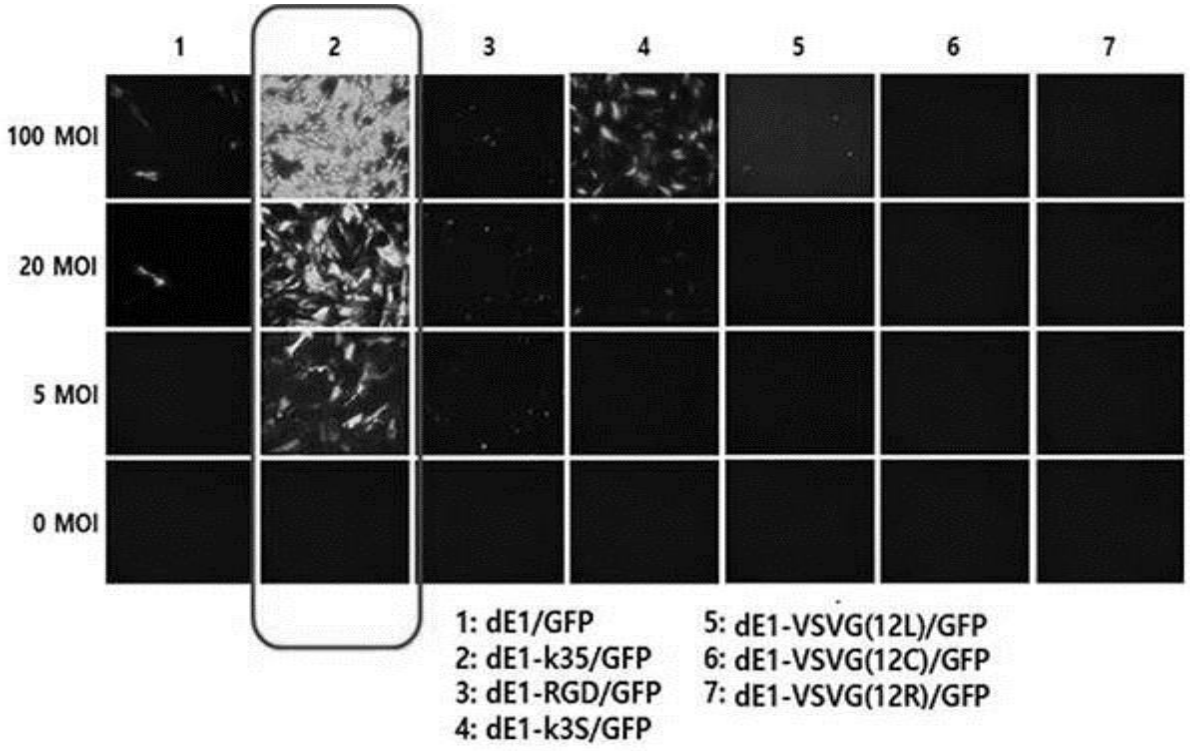

1
2
3
4
5
6
7
100 MOI
20 MOI
5 MOI
0 MOI
1: dE1/GFP
2: dE1-k35/GFP
3: dE1-RGD/GFP
4: dE1-k3S/GFP
5: dE1-VSVG(12L)/GFP
6: dE1-VSVG(12C)/GFP
7: dE1-VSVG(12R)/GFP

FIG. 18

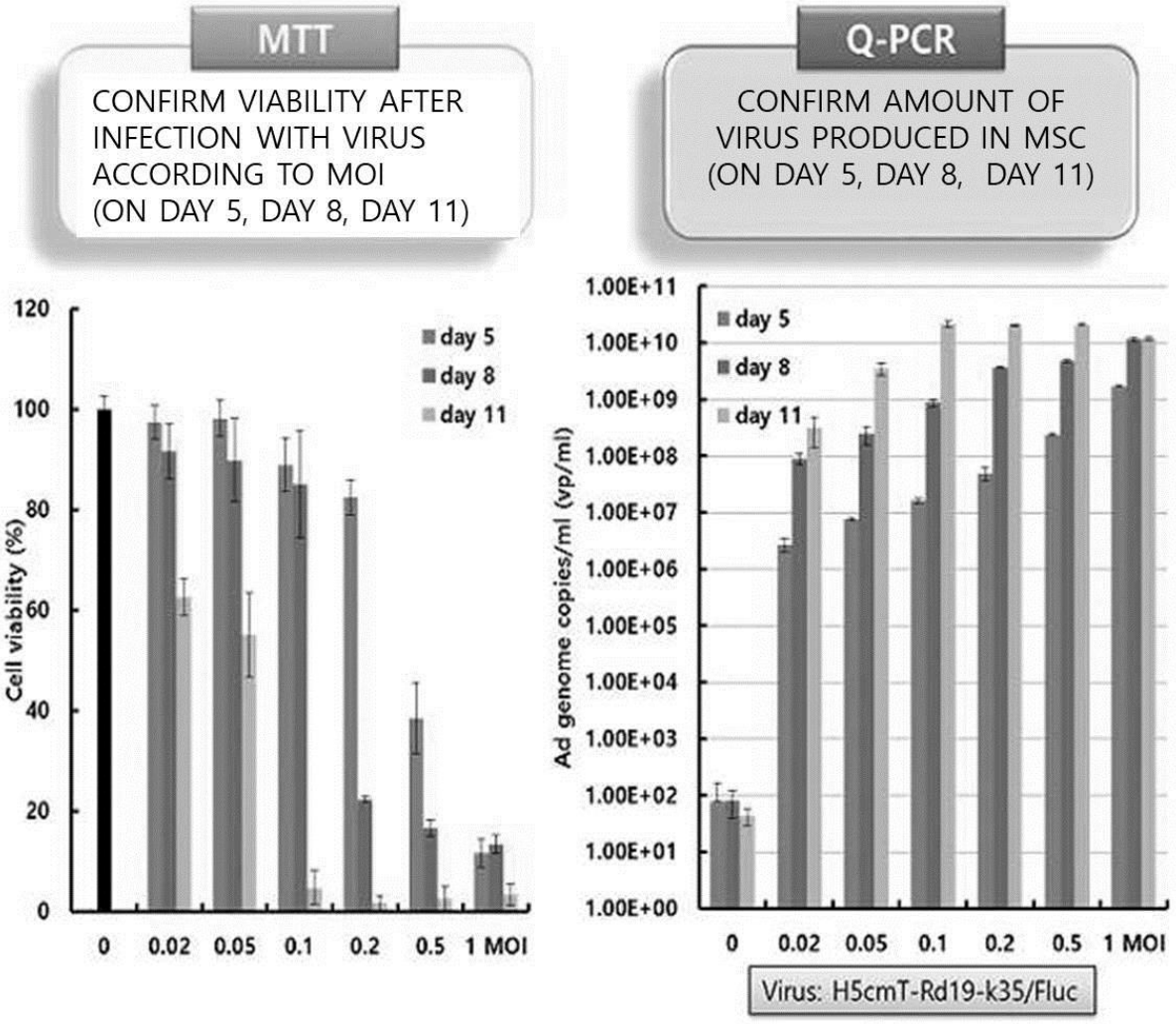

MTT
CONFIRM VIABILITY AFTER INFECTION WITH VIRUS ACCORDING TO MOI (ON DAY 5, DAY 8, DAY 11)
Cell viability (%)
120
100
80
60
40
20
0
day 5
day 8
day 11
0
0.02
0.05
0.1
0.2
0.5
1 MOI
Q-PCR
CONFIRM AMOUNT OF VIRUS PRODUCED IN MSC (ON DAY 5, DAY 8, DAY 11)
Ad genome copies/ml (vp/ml)
1.00E+11
1.00E+10
1.00E+09
1.00E+08
1.00E+07
1.00E+06
1.00E+05
1.00E+04
1.00E+03
1.00E+02
1.00E+01
1.00E+00
day 5
day 8
day 11
0
0.02
0.05
0.1
0.2
0.5
1 MOI
Virus: H5cmT-Rd19-k35/Fluc

FIG. 19

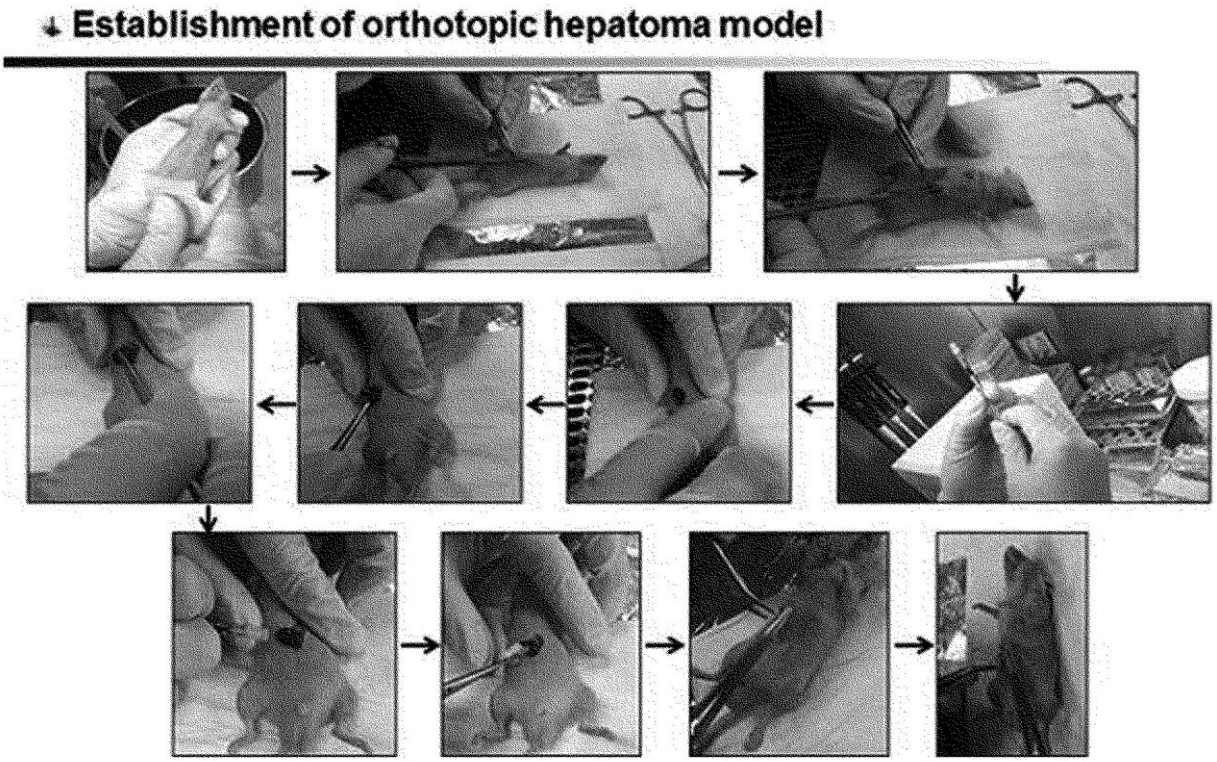


FIG. 20

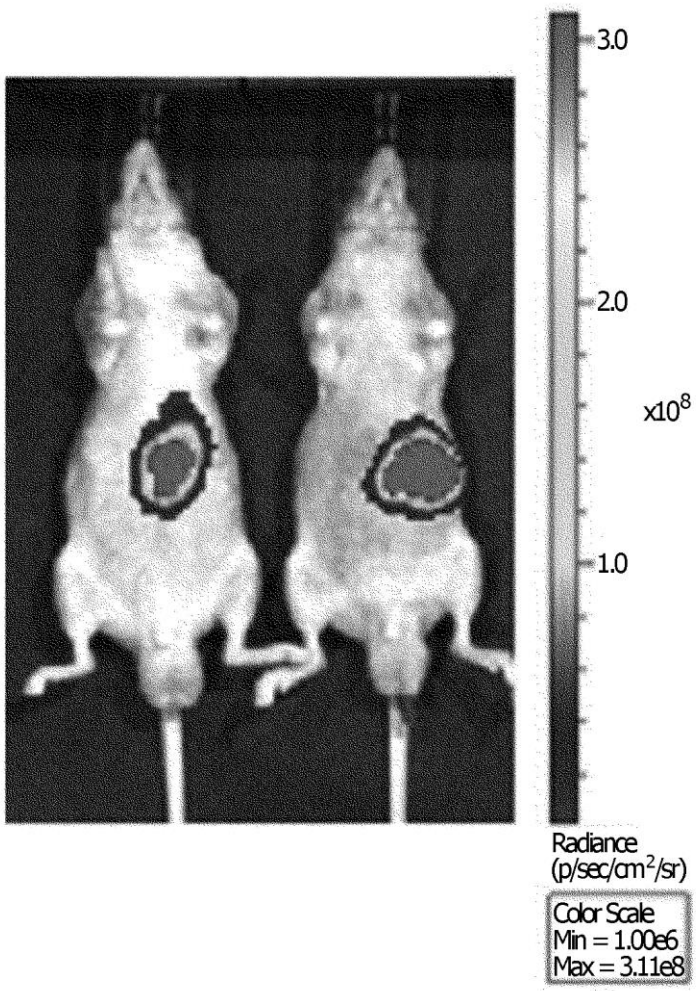
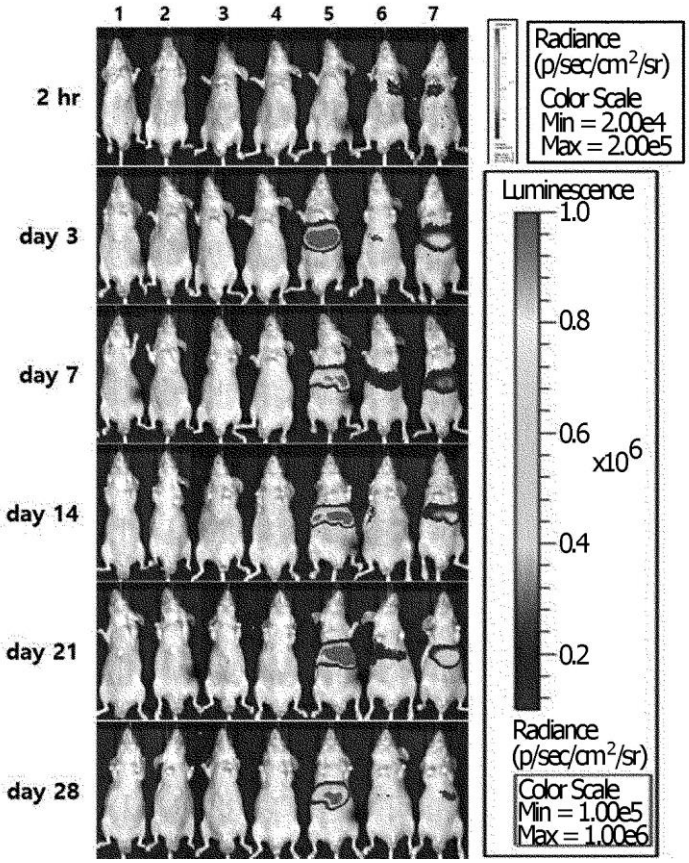

3.0
2.0
x10^8
1.0
Radiance
(p/sec/cm^2/sr)
Color Scale
Min = 1.00e6
Max = 3.11e8

FIG. 21




- **In vivo pilot study:**

**- Cell line: Hep3B**

**- MSC: 1X10$^6$ cells/마리**

**- Ad: H5cmT-Rd19-k35/Luc**

**1: PBS**
**2: PBS**
**3: Ad-MSC (0.2 MOI)**
**4: Ad-MSC (0.2 MOI)**
**5: Ad-MSC (0.5 MOI)**
**6: Ad-MSC (0.5 MOI)**
**7: Ad-MSC (1 MOI)**

FIG. 22

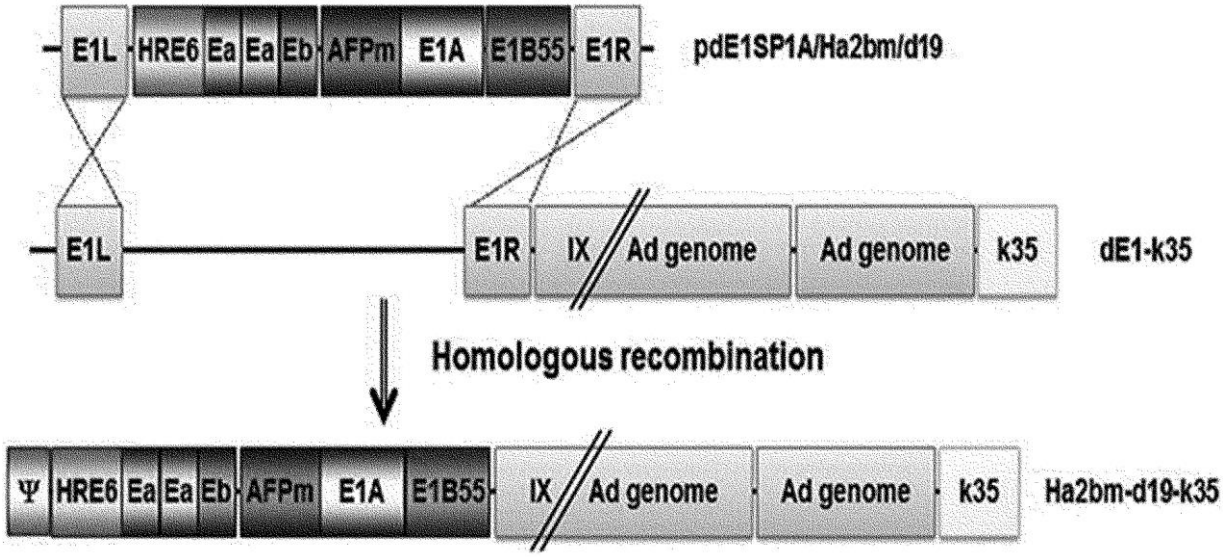



FIG. 23A

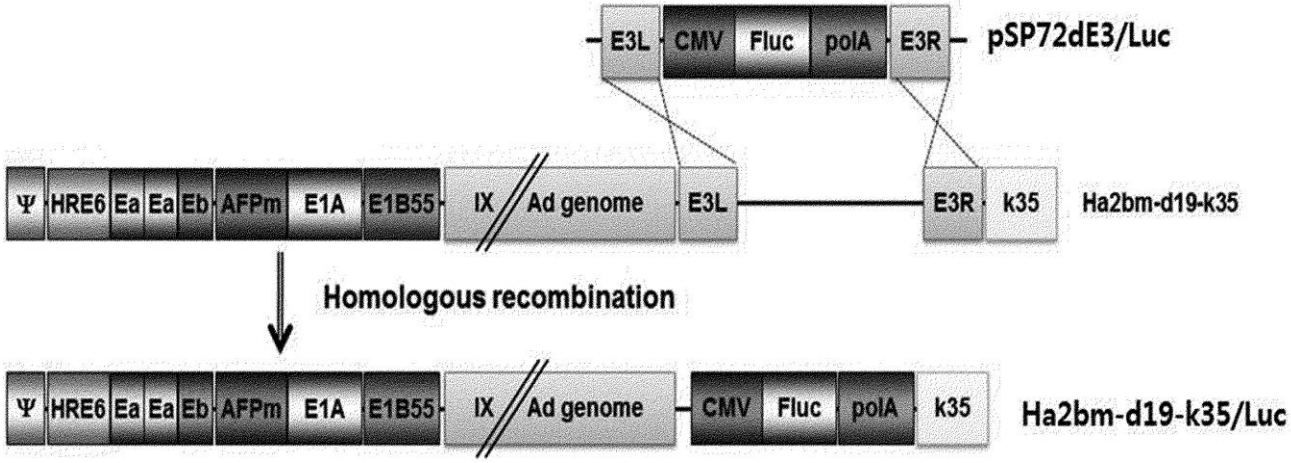



FIG. 23B

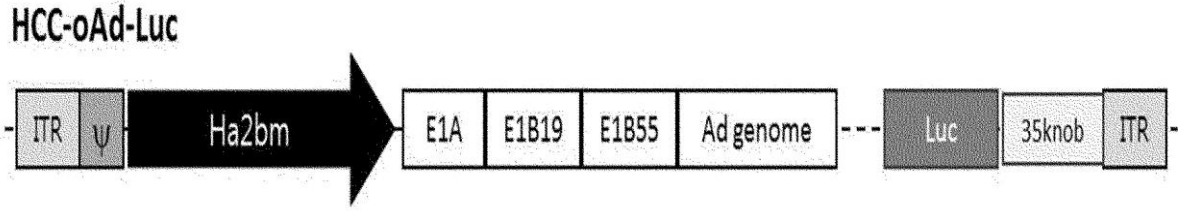


FIG. 24A

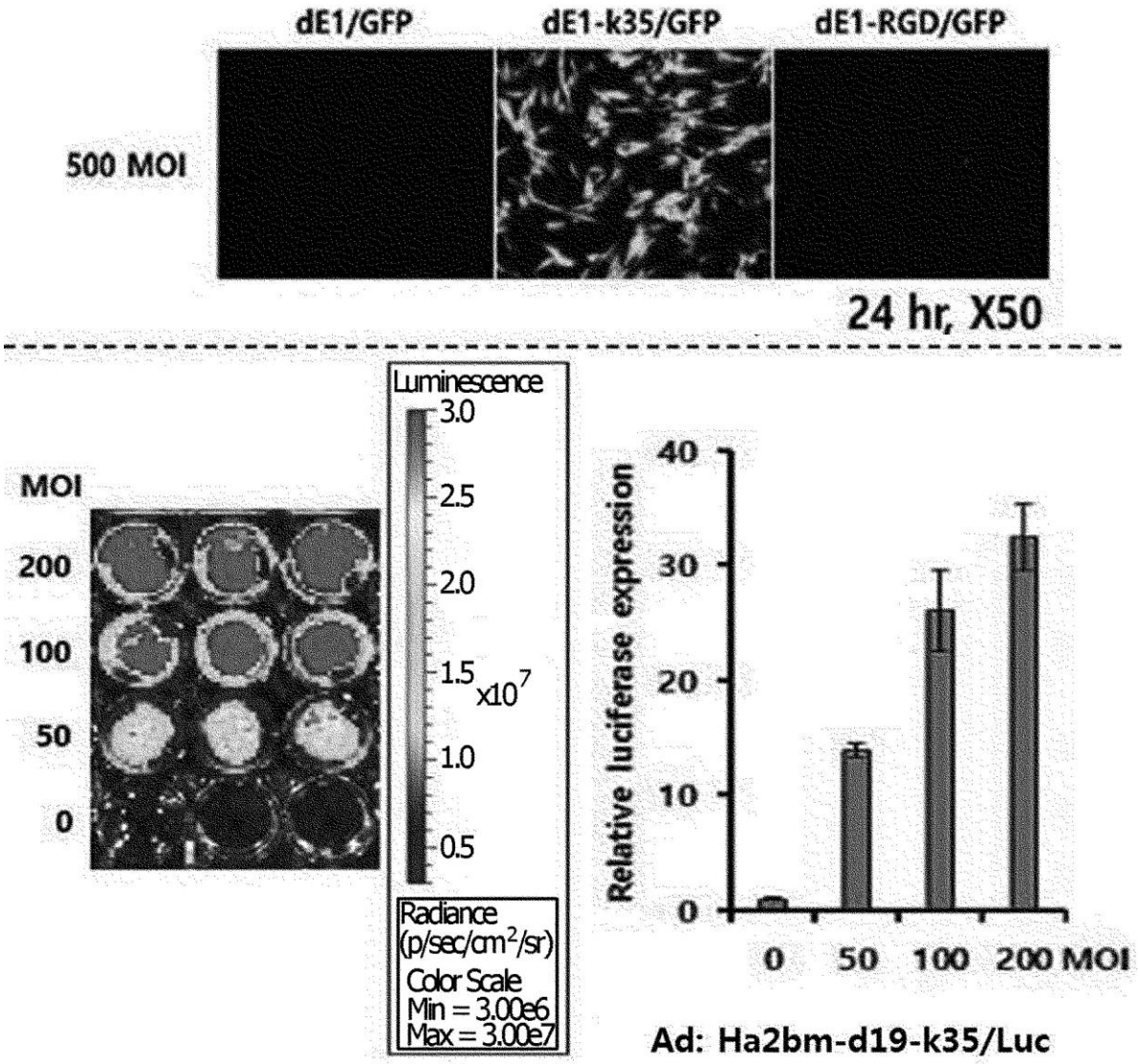

dE1/GFP
dE1-k35/GFP
dE1-RGD/GFP
500 MOI
24 hr, X50
MOI
200
100
50
0
Luminescence
3.0
2.5
2.0
1.5
x10^7
1.0
0.5
Radiance
(p/sec/cm²/sr)
Color Scale
Min = 3.00e6
Max = 3.00e7
Relative luciferase expression
40
30
20
10
0
0
50
100
200 MOI
Ad: Ha2bm-d19-k35/Luc

FIG. 24B

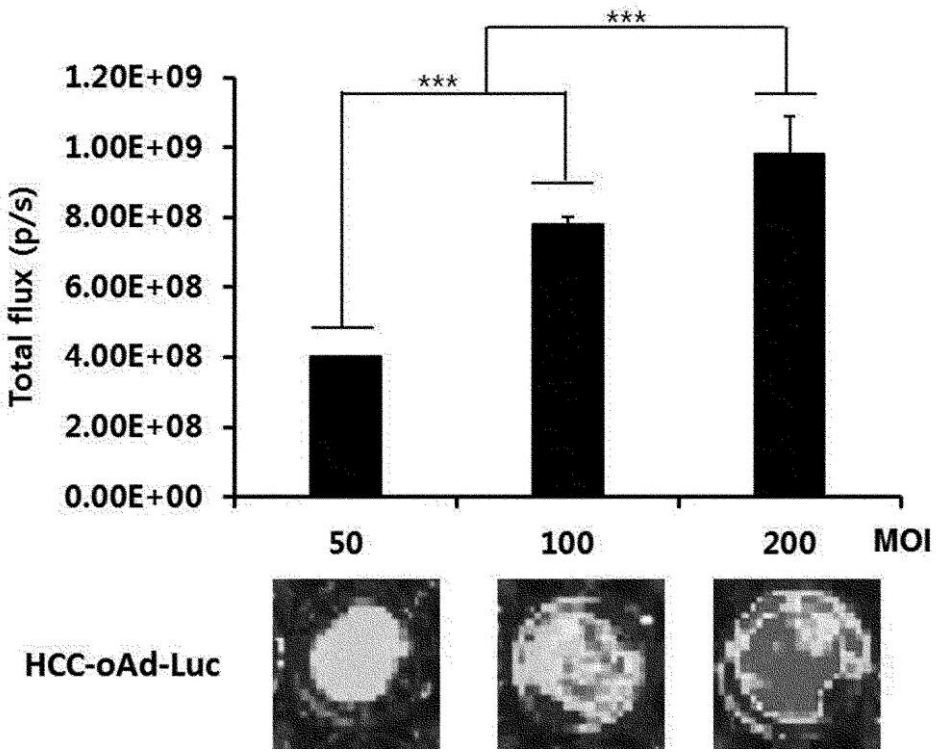



FIG. 25

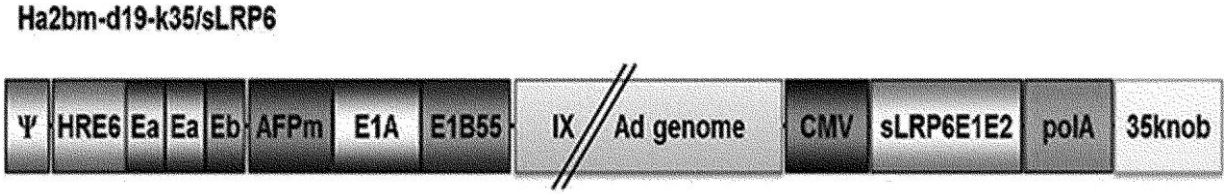


FIG. 26

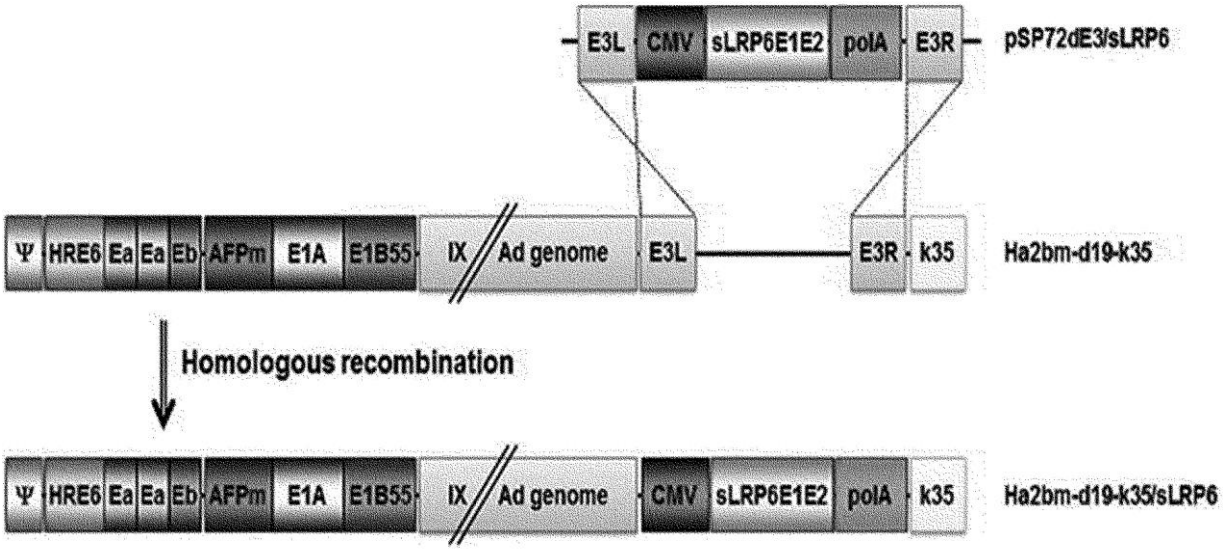

E3L
CMV
sLRP6E1E2
polA
E3R
pSP72dE3/sLRP6
Ψ
HRE6
Ea
Ea
Eb
AFPm
E1A
E1B55
IX
Ad genome
E3L
E3R
k35
Ha2bm-d19-k35
Homologous recombination
Ψ
HRE6
Ea
Ea
Eb
AFPm
E1A
E1B55
IX
Ad genome
CMV
sLRP6E1E2
polA
k35
Ha2bm-d19-k35/sLRP6

FIG. 27

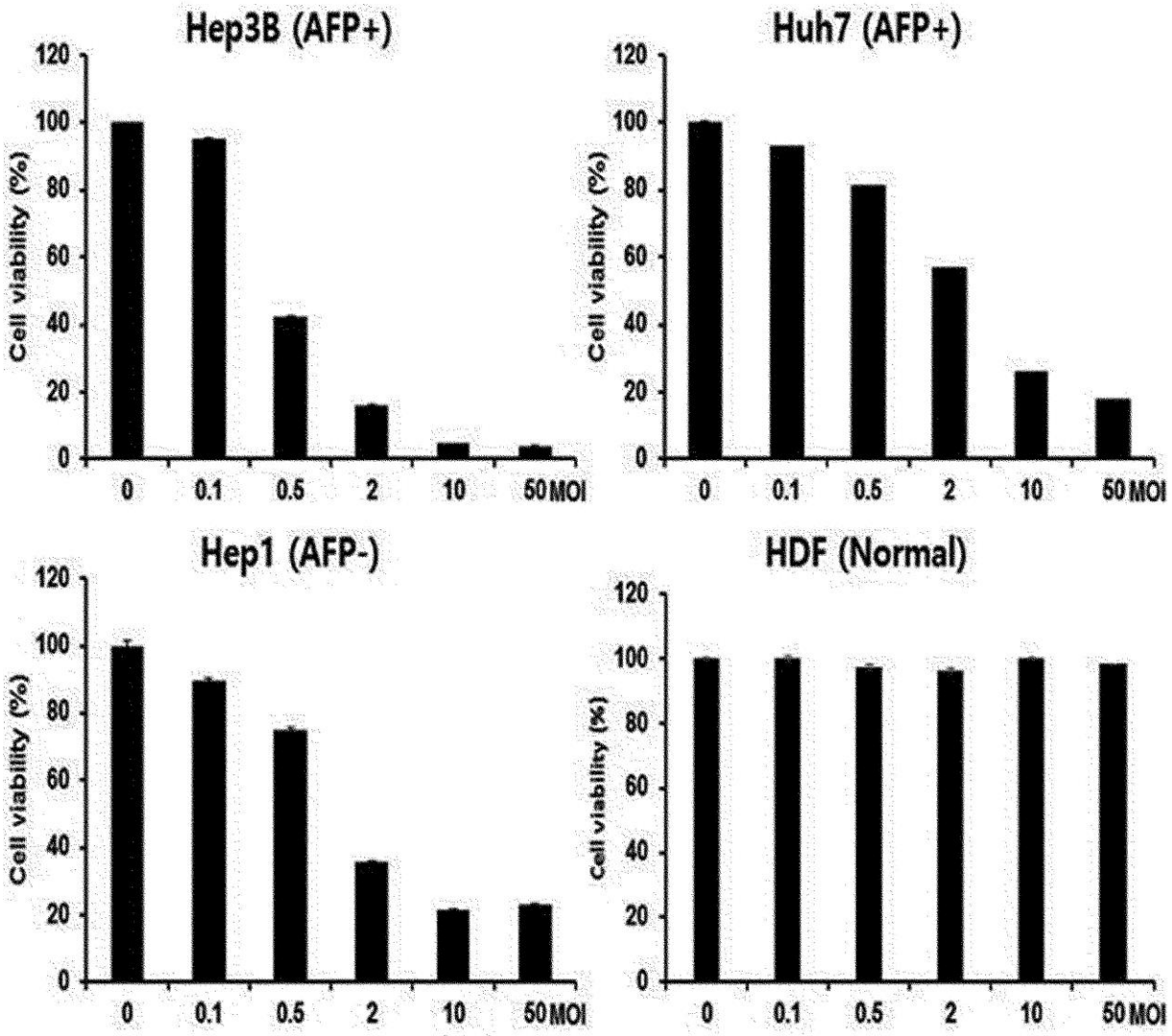

Hep3B (AFP+)
Huh7 (AFP+)
Hep1 (AFP-)
HDF (Normal)
Cell viability (%)
0
20
40
60
80
100
120
0
0.1
0.5
2
10
50 MOI

FIG. 28

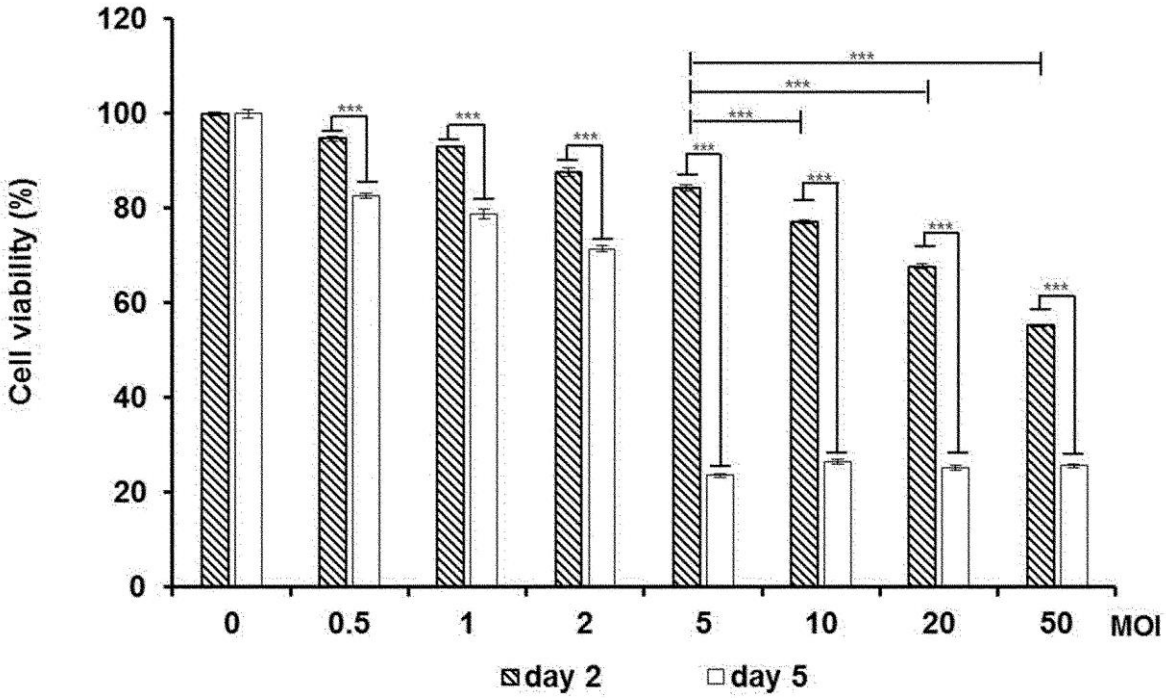

Cell viability (%)
120
100
80
60
40
20
0
***
0
0.5
1
2
5
10
20
50
MOI
day 2
day 5

FIG. 29

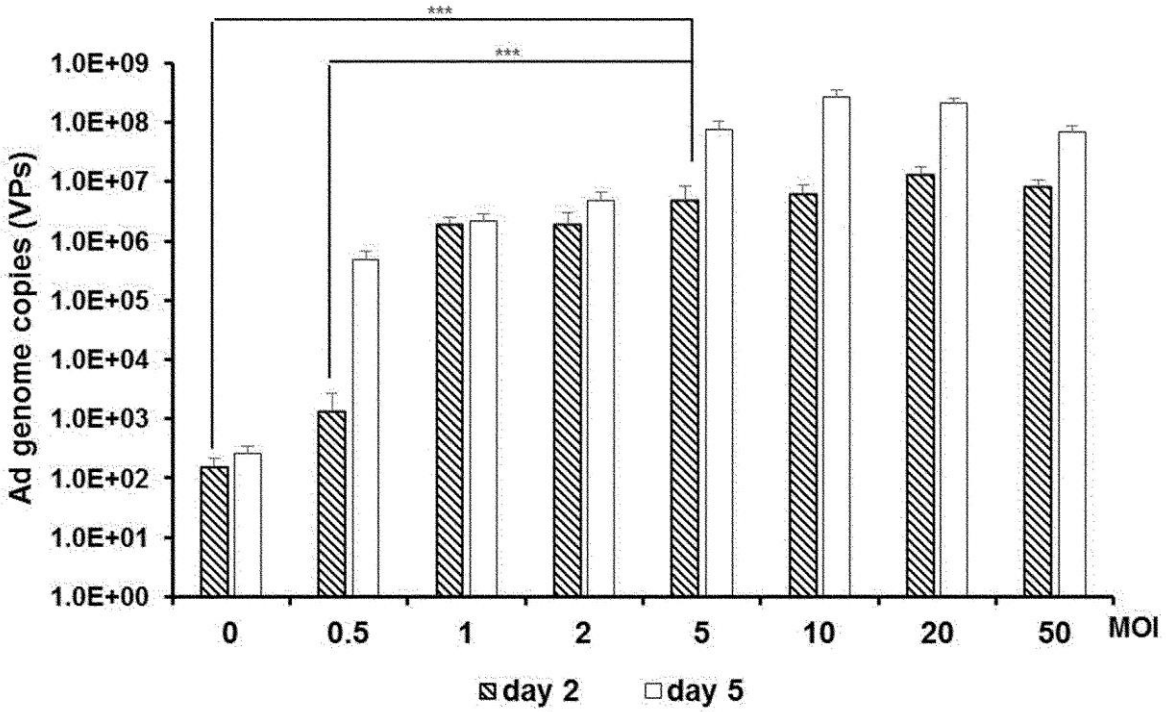

Ad genome copies (VPs)
1.0E+09
1.0E+08
1.0E+07
1.0E+06
1.0E+05
1.0E+04
1.0E+03
1.0E+02
1.0E+01
1.0E+00
***
0
0.5
1
2
5
10
20
50
MOI
day 2
day 5

FIG. 30A

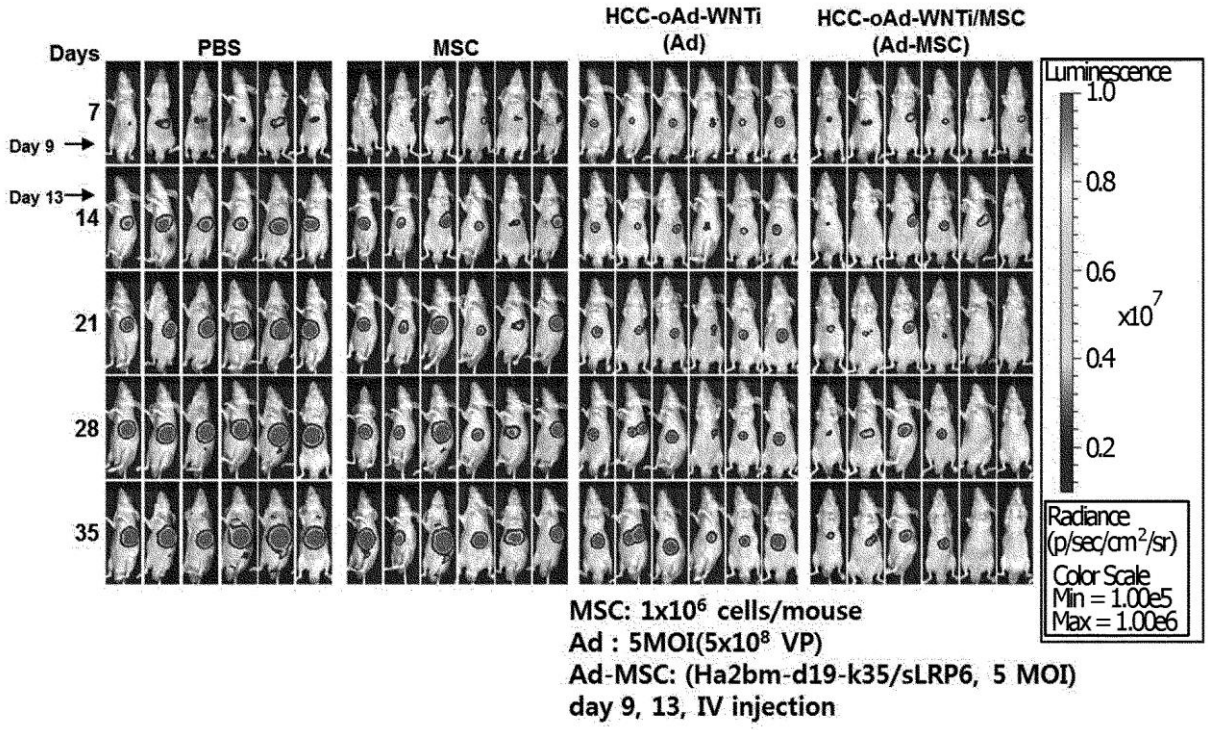



FIG. 30B

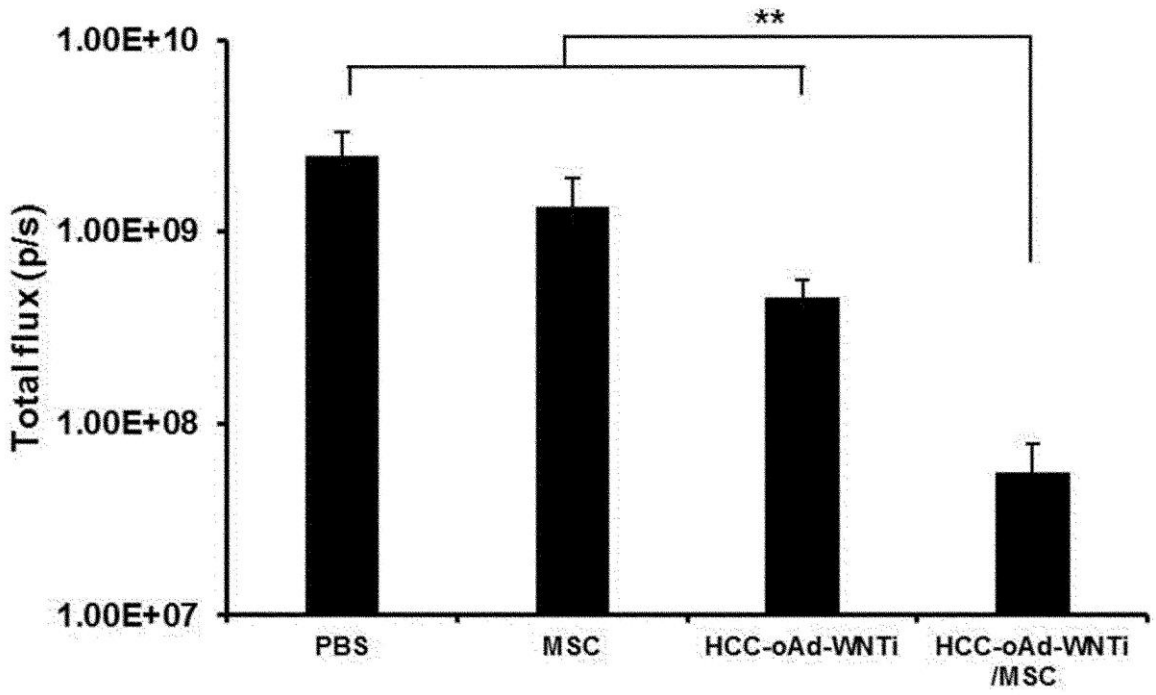


FIG. 31

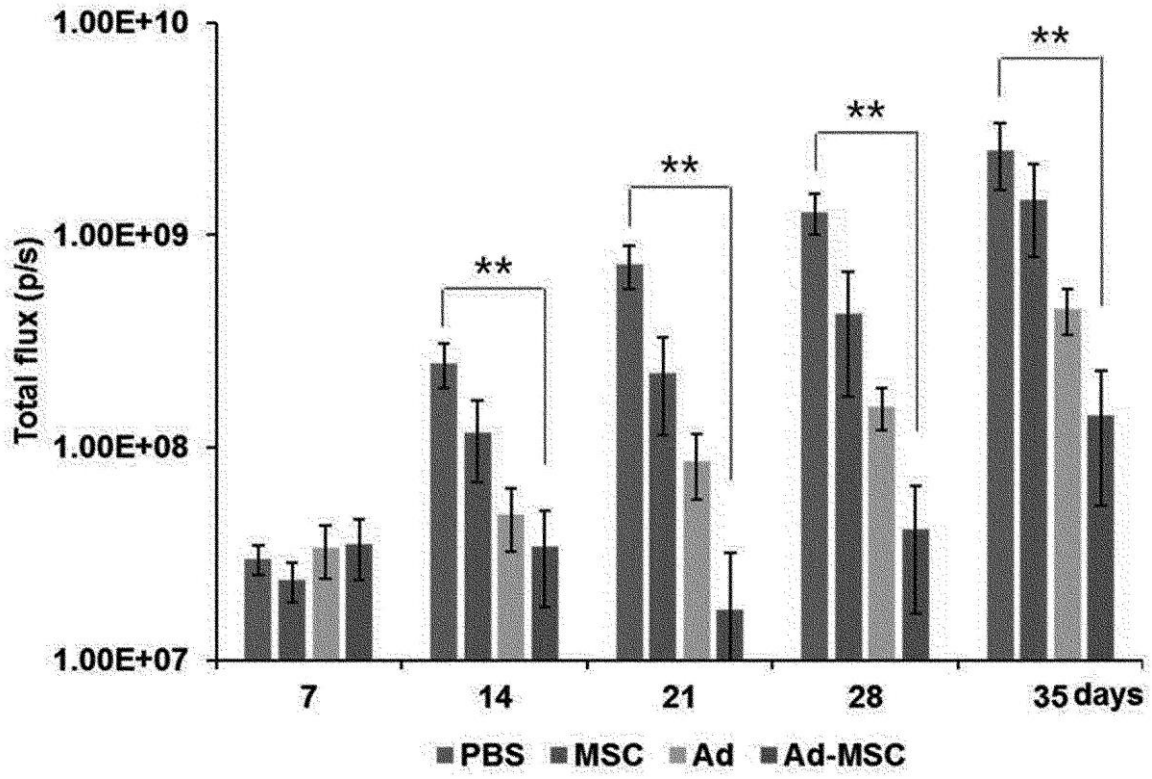

1.00E+10
1.00E+09
1.00E+08
1.00E+07
Total flux (p/s)
**
**
**
**
7
14
21
28
35 days
PBS
MSC
Ad
Ad-MSC

FIG. 32

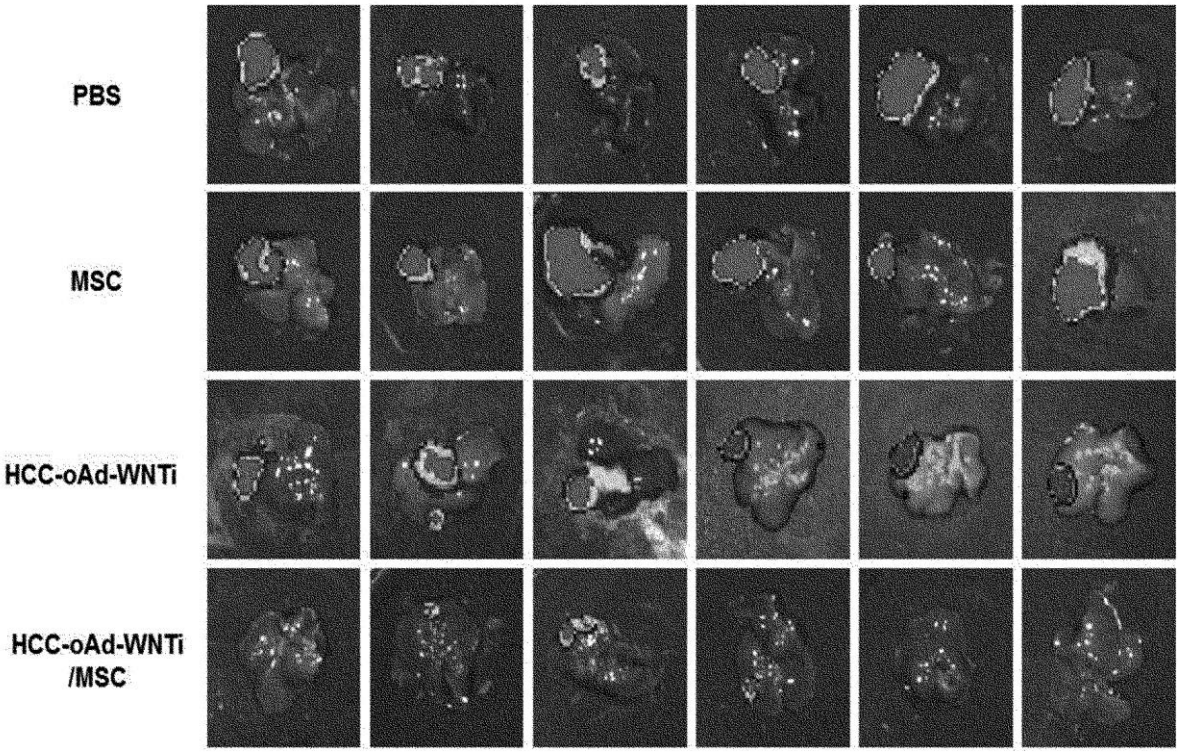

PBS
MSC
HCC-oAd-WNTi
HCC-oAd-WNTi /MSC
MSC: 1x10^6 cells/mouse
Ad : 5MOI(5x10^8 VP)
Ad-MSC: (Ha2bm-d19-k35/sLRP6, 5 MOI)
IV injection

FIG. 33A

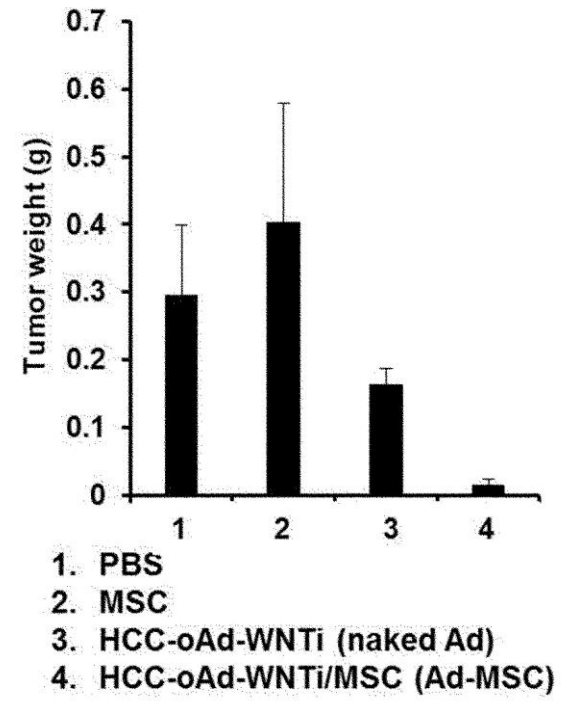



FIG. 33B

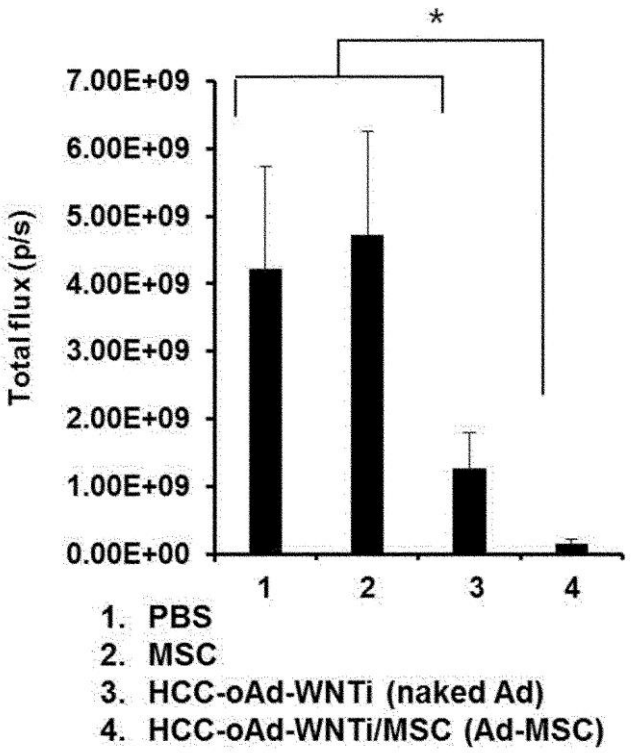


FIG. 34

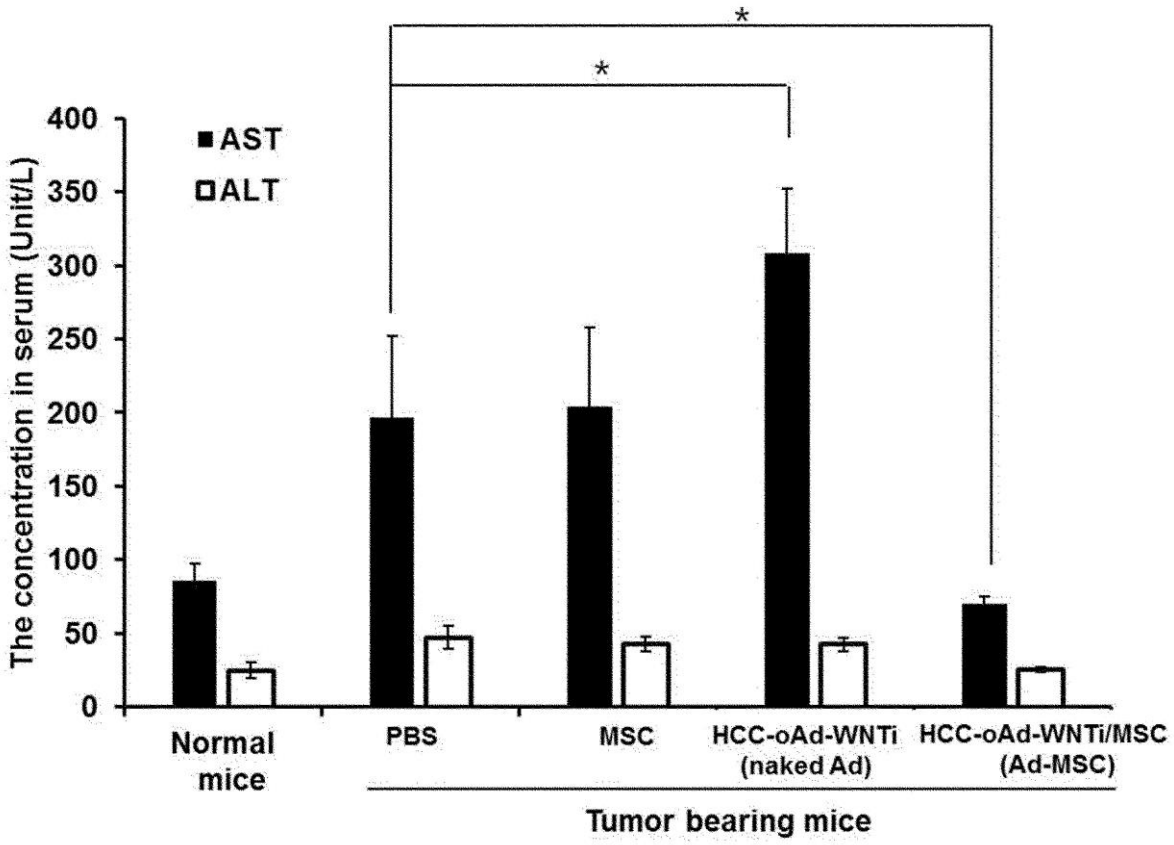
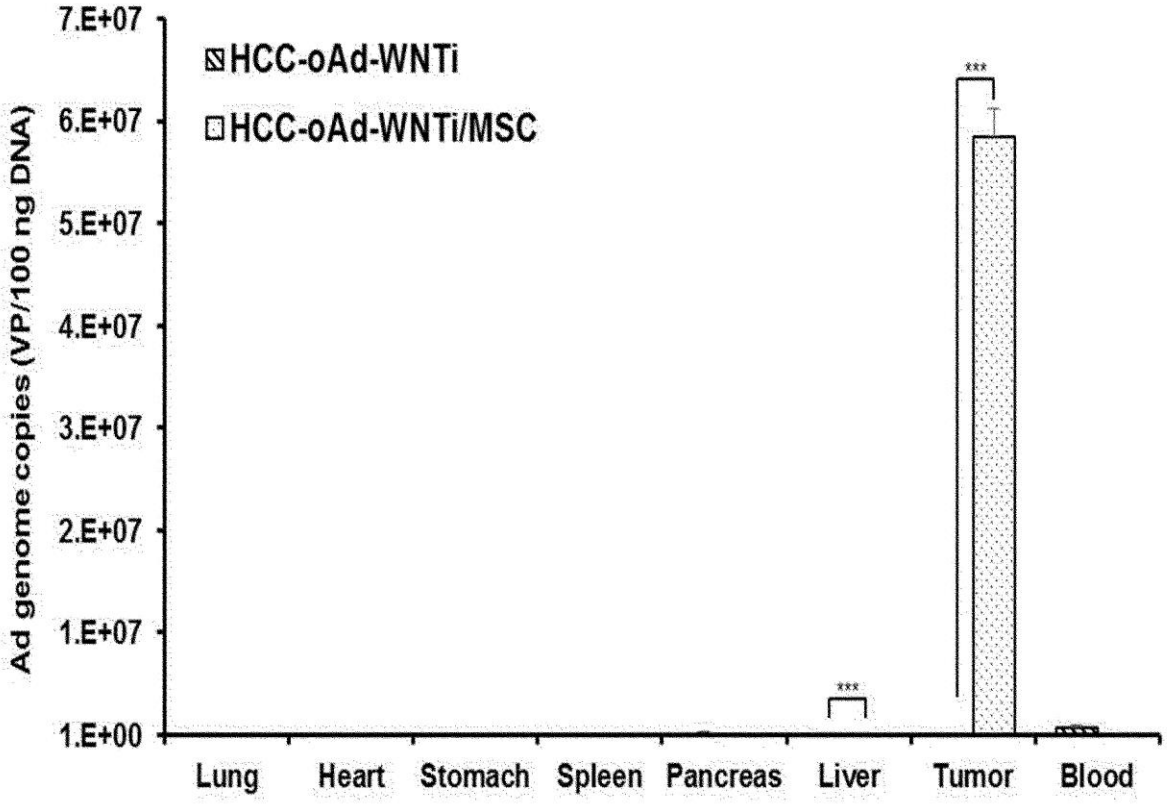

*
*
400
350
300
250
200
150
100
50
0
AST
ALT
The concentration in serum (Unit/L)
Normal mice
PBS
MSC
HCC-oAd-WNTi (naked Ad)
HCC-oAd-WNTi/MSC (Ad-MSC)
Tumor bearing mice

FIG. 35




| | Tumor | Liver | Tumor/Liver |
|---|---|---|---|
| HCC-oAd-WNTi | 1.37E+04 (1) | 9.24E+04 (1) | 0.147729 (1) |
| HCC-oAd-WNTi/MSC | 5.85E+07 ($4.3 \times 10^{3}$) | 6.11E+04 ($6.6 \times 10^{-1}$) | 957.40 ($6.48 \times 10^{3}$) |

FIG. 36

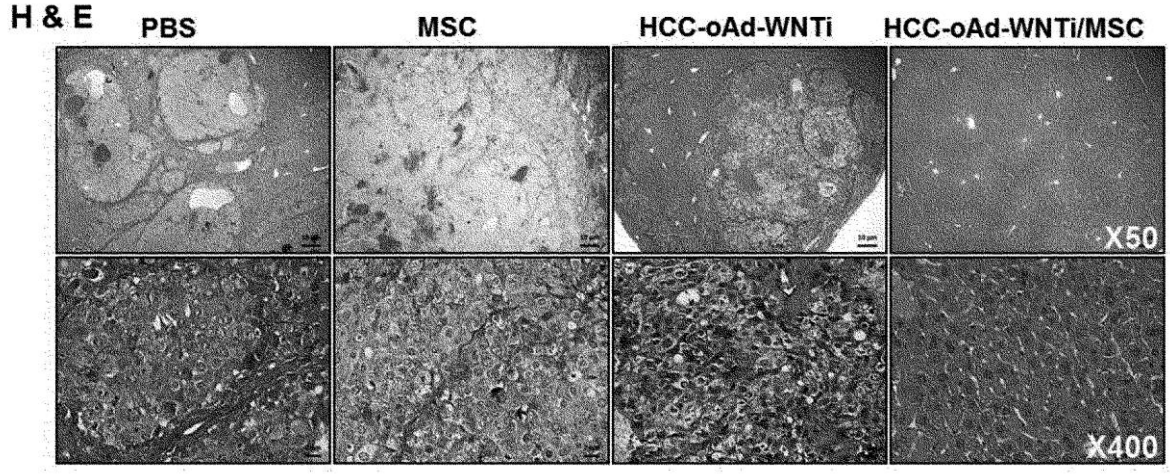


FIG. 37

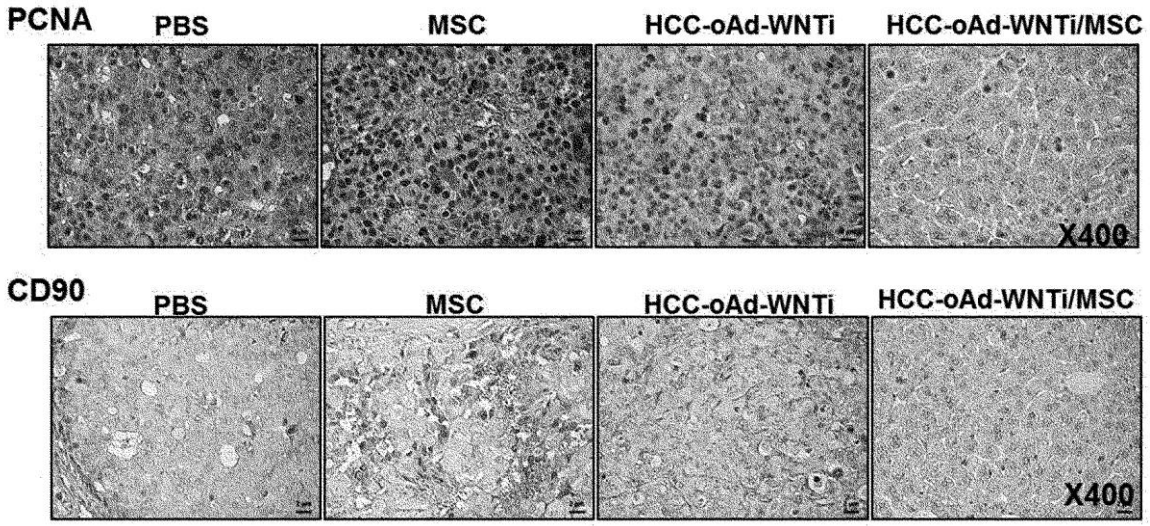



FIG. 38A

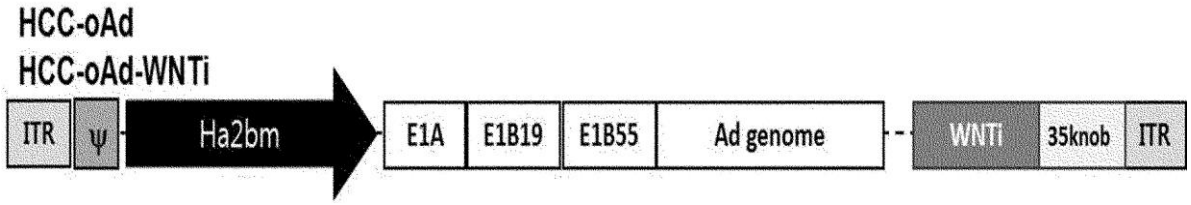



FIG. 38B

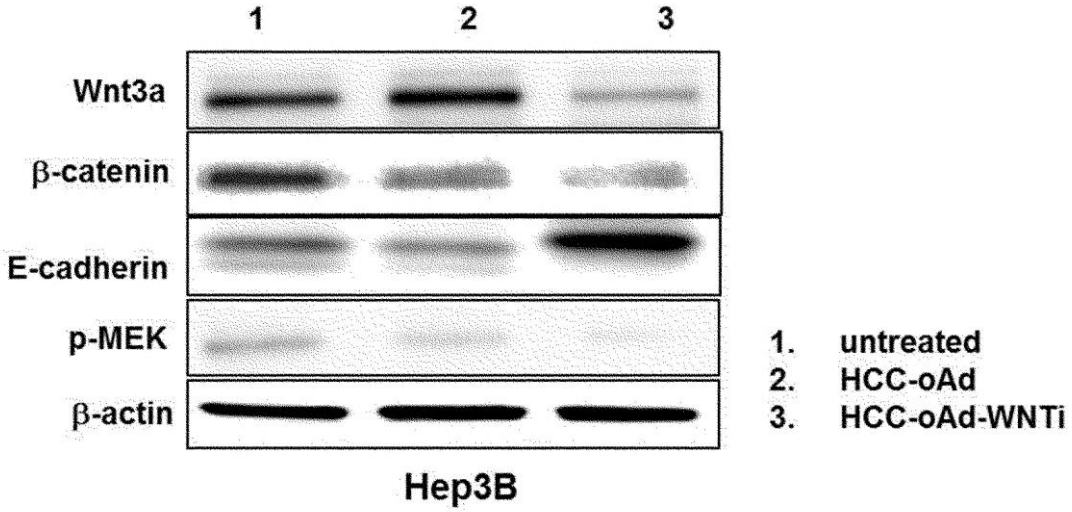


FIG. 38C

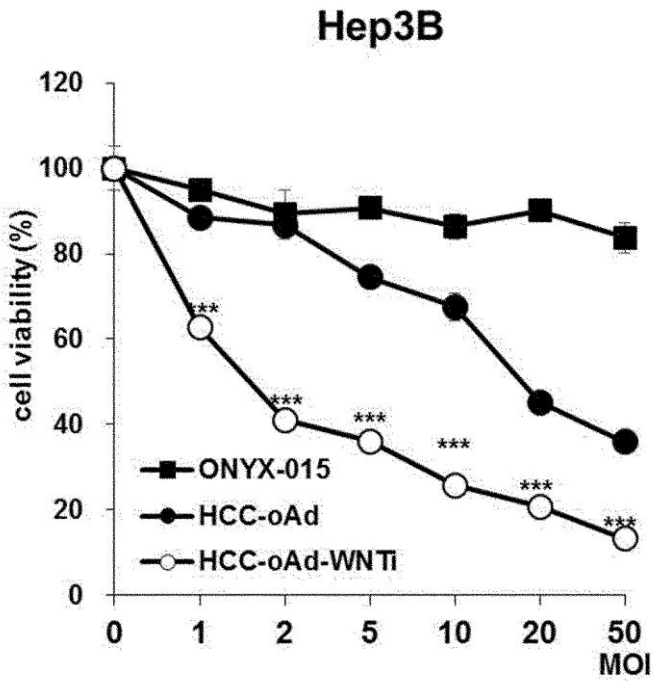



FIG. 38D

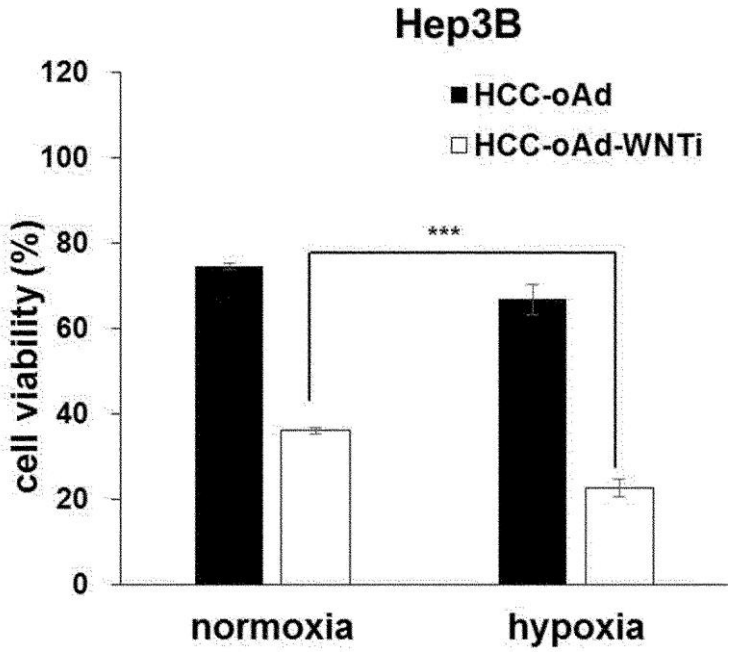


FIG. 39

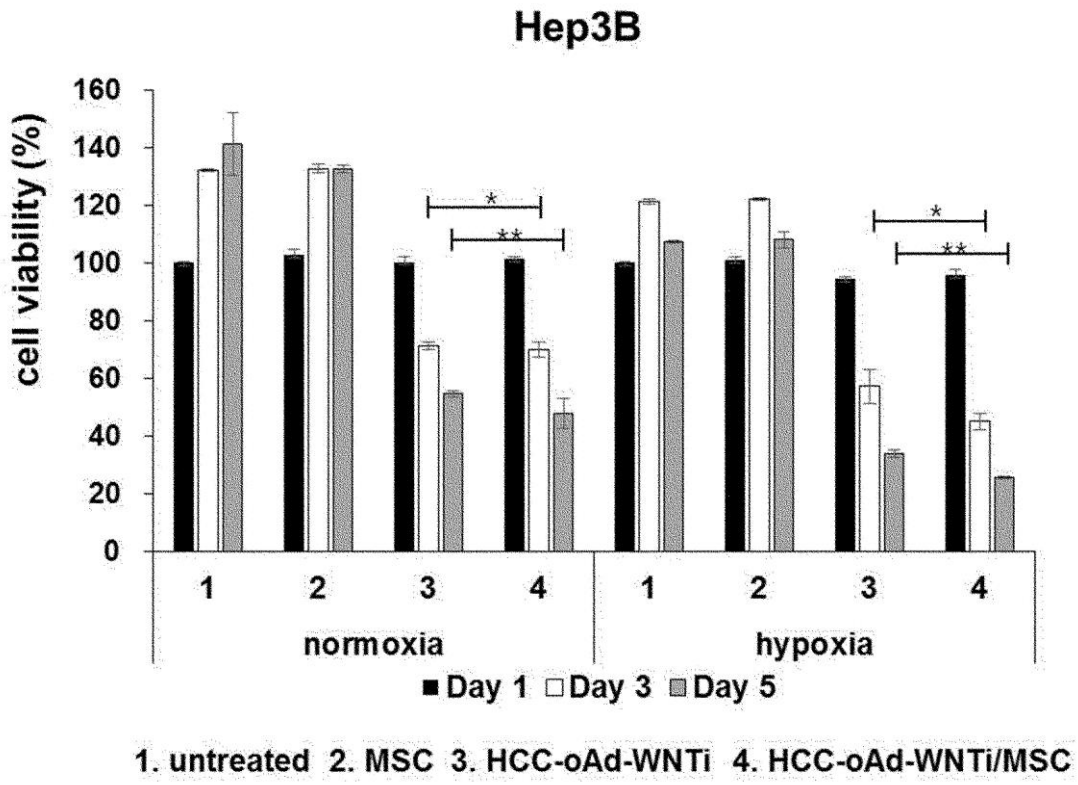
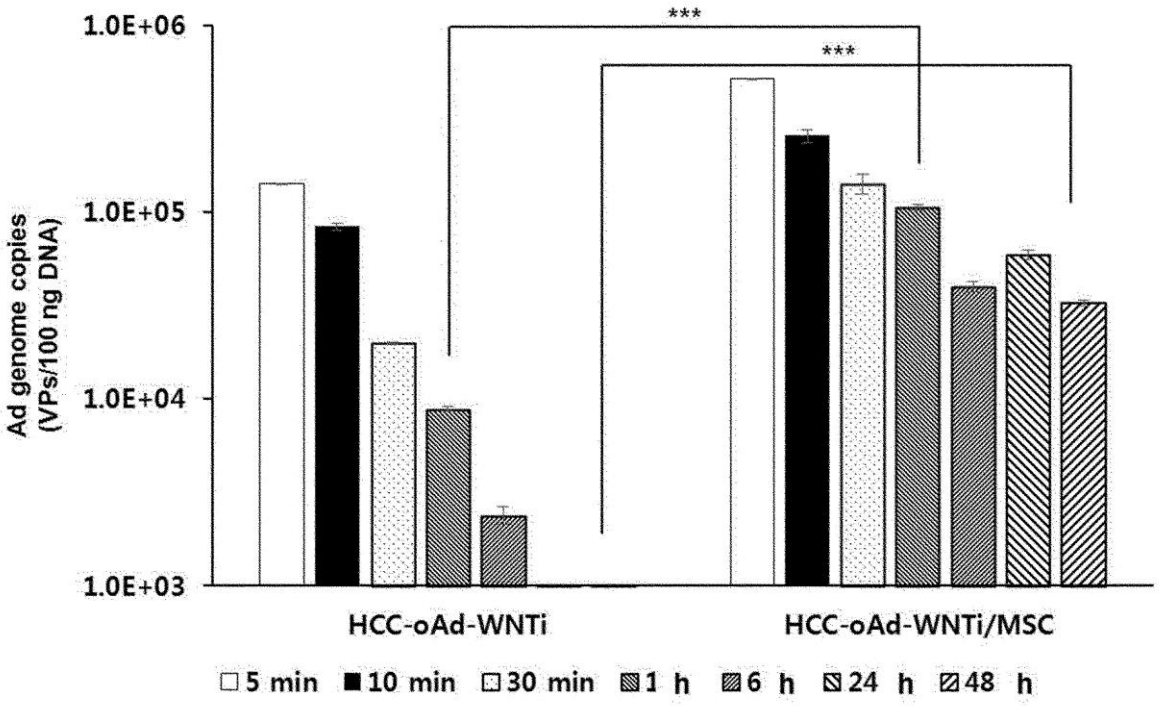

Hep3B
cell viability (%)
160
140
120
100
80
60
40
20
0
*
**
*
**
1
2
3
4
1
2
3
4
normoxia
hypoxia
Day 1
Day 3
Day 5
1. untreated 2. MSC 3. HCC-oAd-WNTi 4. HCC-oAd-WNTi/MSC

FIG. 40


***
***
1.0E+06
1.0E+05
1.0E+04
1.0E+03
Ad genome copies
(VPs/100 ng DNA)
HCC-oAd-WNTi
HCC-oAd-WNTi/MSC
5 min
10 min
30 min
1 h
6 h
24 h
48 h