Fig. 1

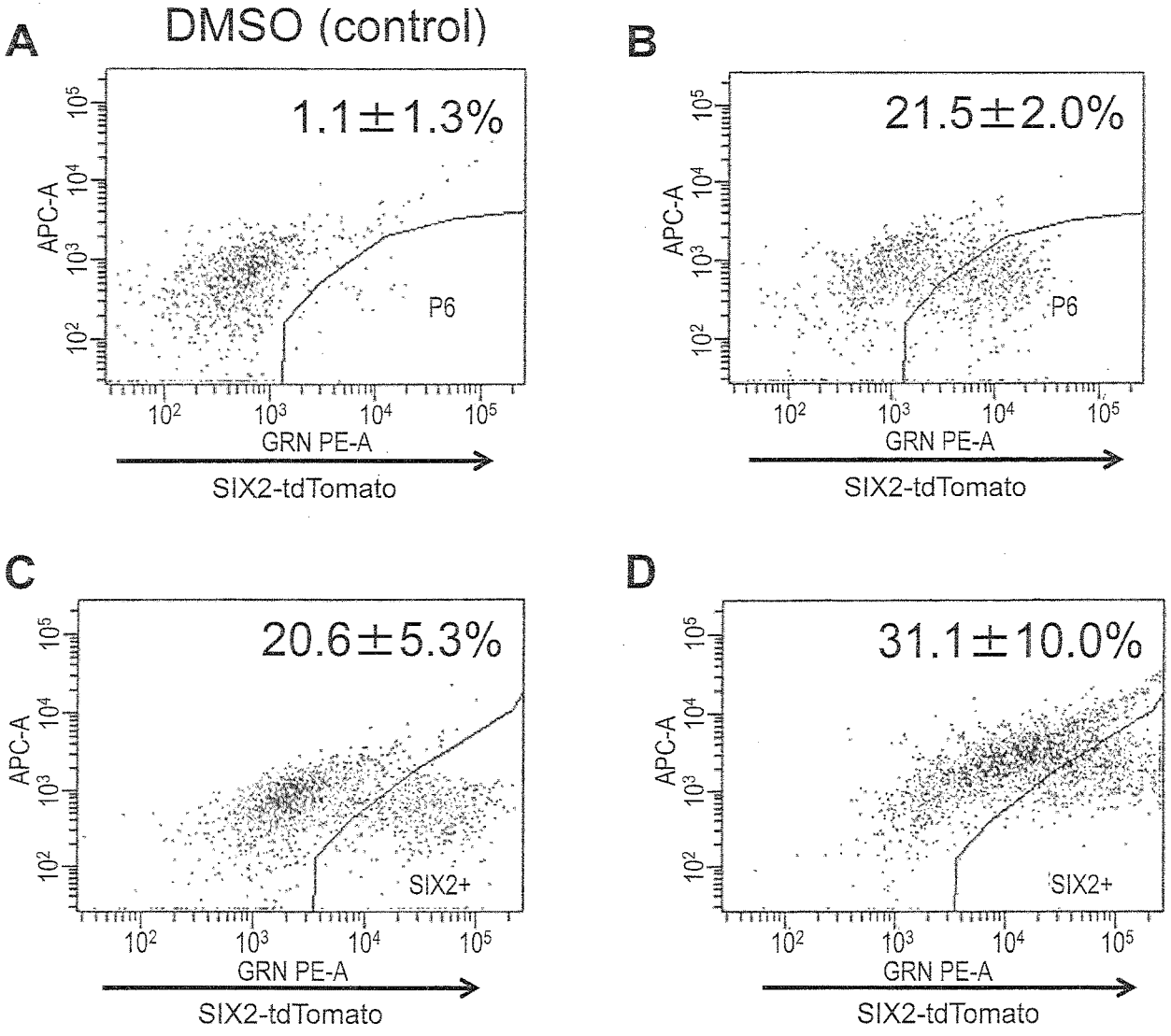

A
DMSO (control)
1.1±1.3%
P6
APC-A
GRN PE-A
SIX2-tdTomato
B
21.5±2.0%
P6
APC-A
GRN PE-A
SIX2-tdTomato
C
20.6±5.3%
SIX2+
APC-A
GRN PE-A
SIX2-tdTomato
D
31.1±10.0%
SIX2+
APC-A
GRN PE-A
SIX2-tdTomato

Fig. 2

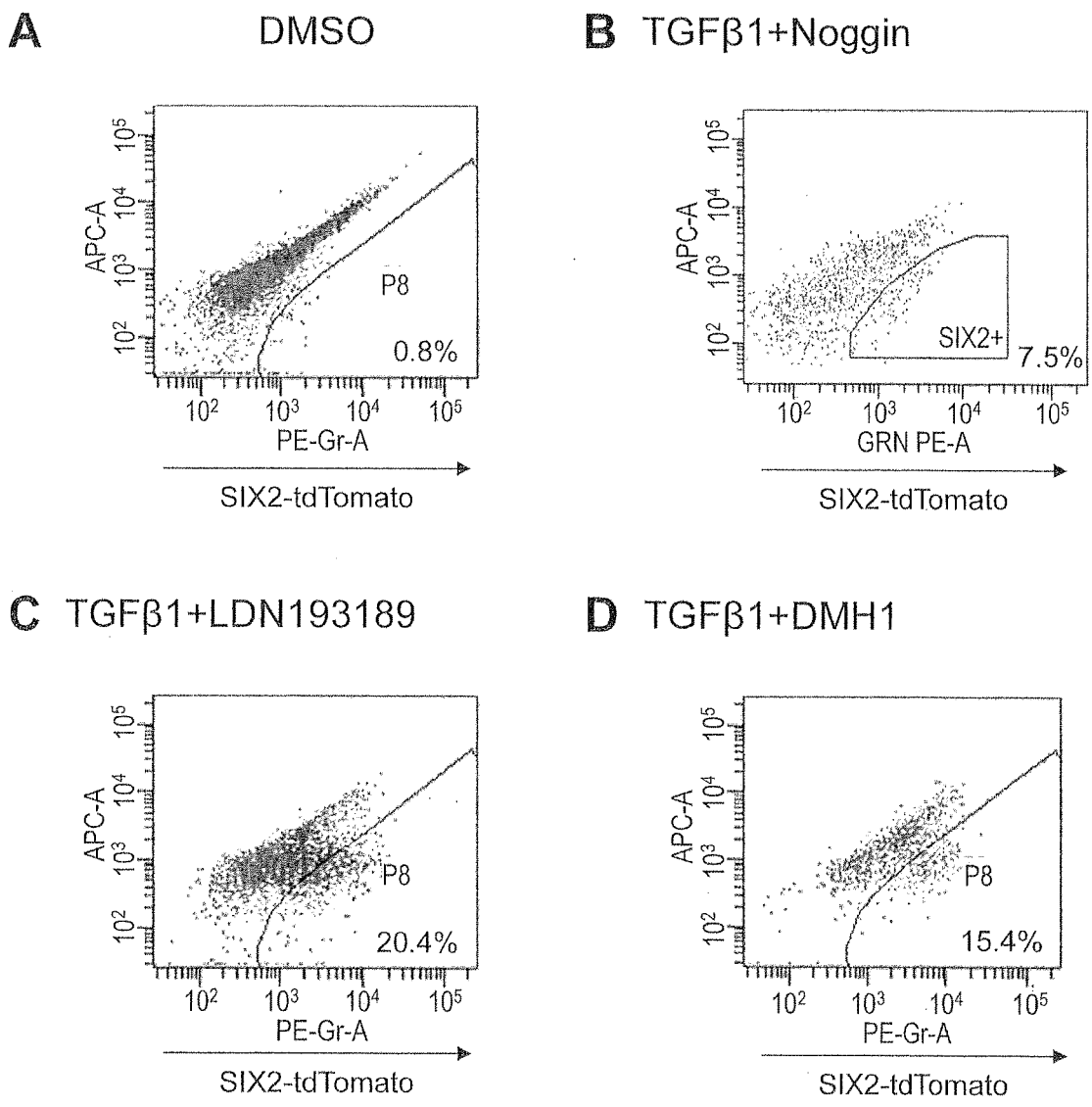
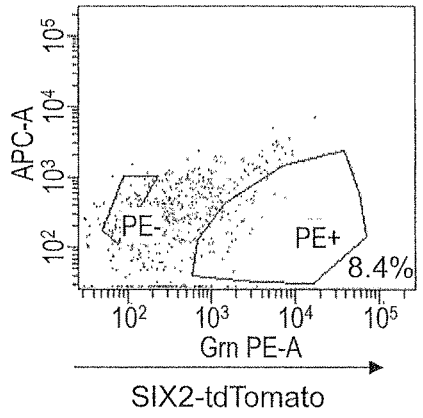
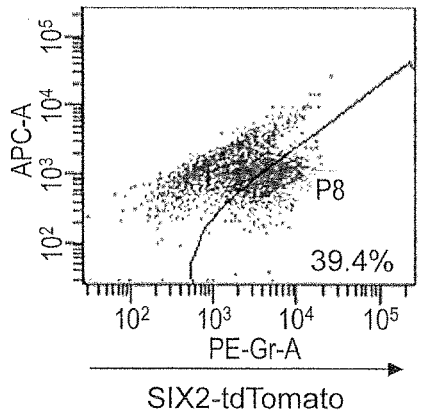

A
DMSO
APC-A
P8
0.8%
PE-Gr-A
SIX2-tdTomato
B
TGFβ1+Noggin
APC-A
SIX2+
7.5%
GRN PE-A
SIX2-tdTomato
C
TGFβ1+LDN193189
APC-A
P8
20.4%
PE-Gr-A
SIX2-tdTomato
D
TGFβ1+DMH1
APC-A
P8
15.4%
PE-Gr-A
SIX2-tdTomato

Fig. 2 (Continued)

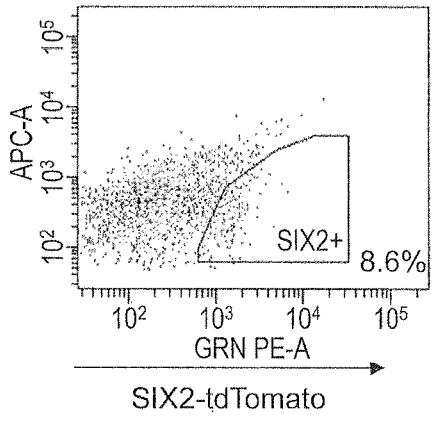
E IDE1+Dorsomorphin



F IDE2+Dorsomorphin



G TGFβ2+LDN193189



H TGFβ3+LDN193189

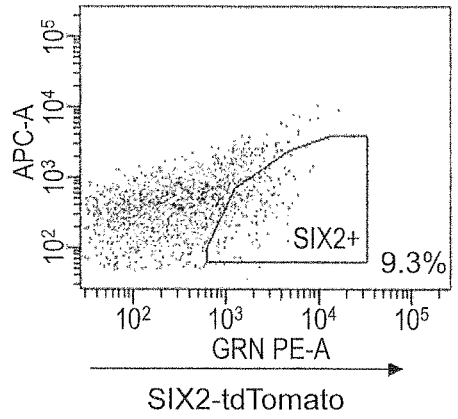
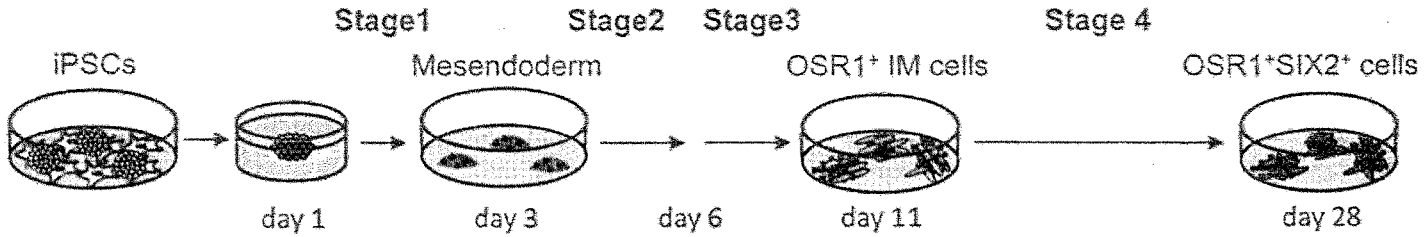


Fig. 3

A



Stage1	Stage2	Stage3	Stage4
Suspension culture	Adhesion culture		
DMEM/F12 Glutamax			
2% FBS, 0.5% P/SM	10% KSR, 50 μM 2ME, 1% NEAA, 0.5% P/SM		
activin A (100 ng/ml)	BMP7 (100 ng/ml)	TGFβ1 (5 ng/ml)	TGFβ1 (5 ng/ml)
CHIR99021 (1 μM)	CHIR99021 (1 μM)	TTNPB (1 μM)	DMH1 (0.5 μM)

B

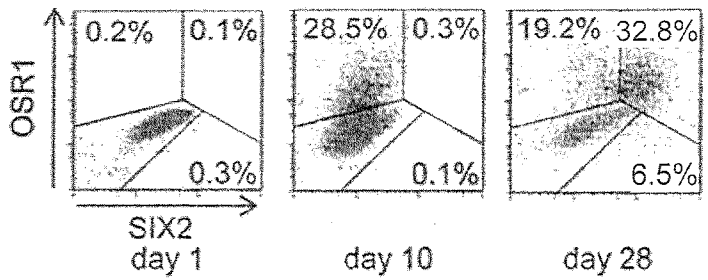


Fig. 3 (Continued)

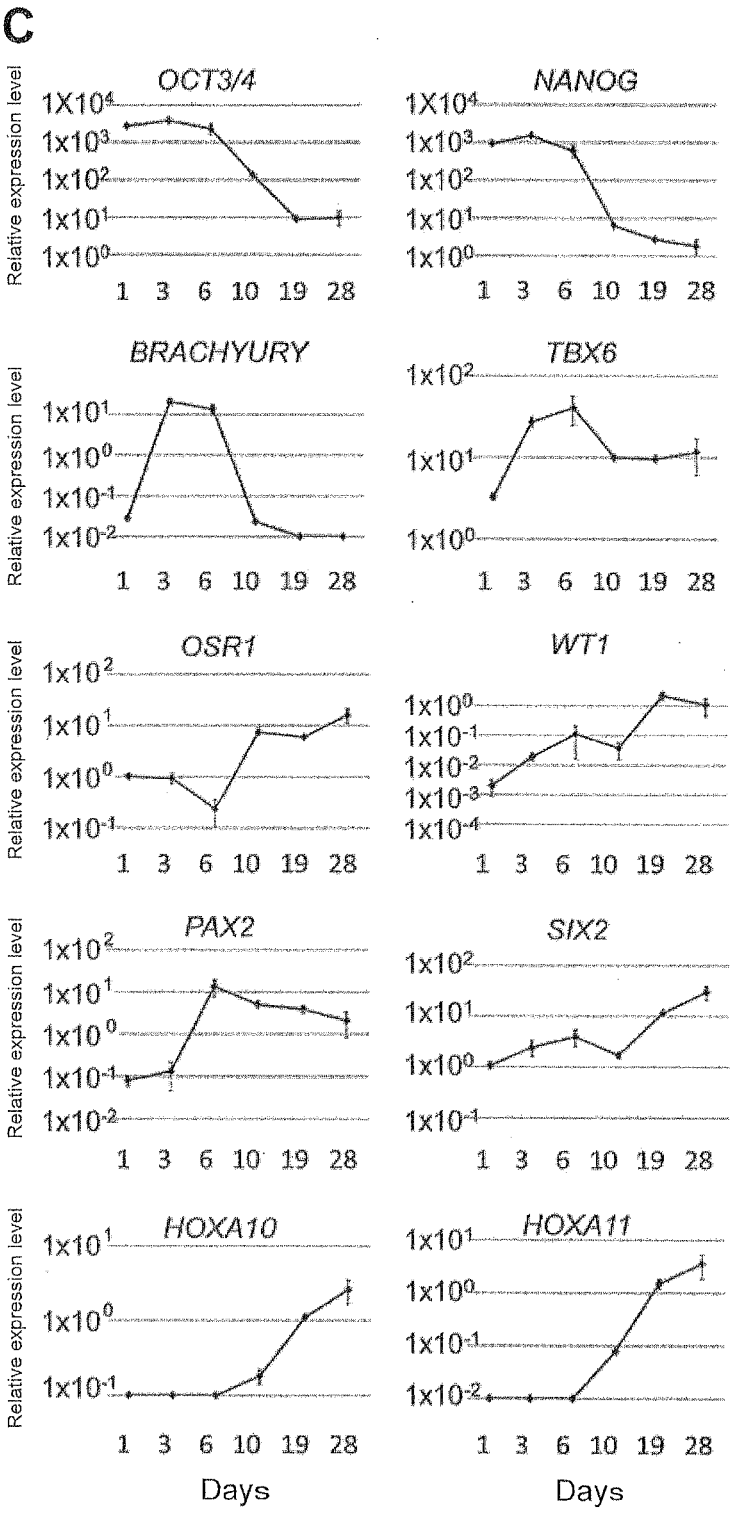


Fig. 3 (Continued)

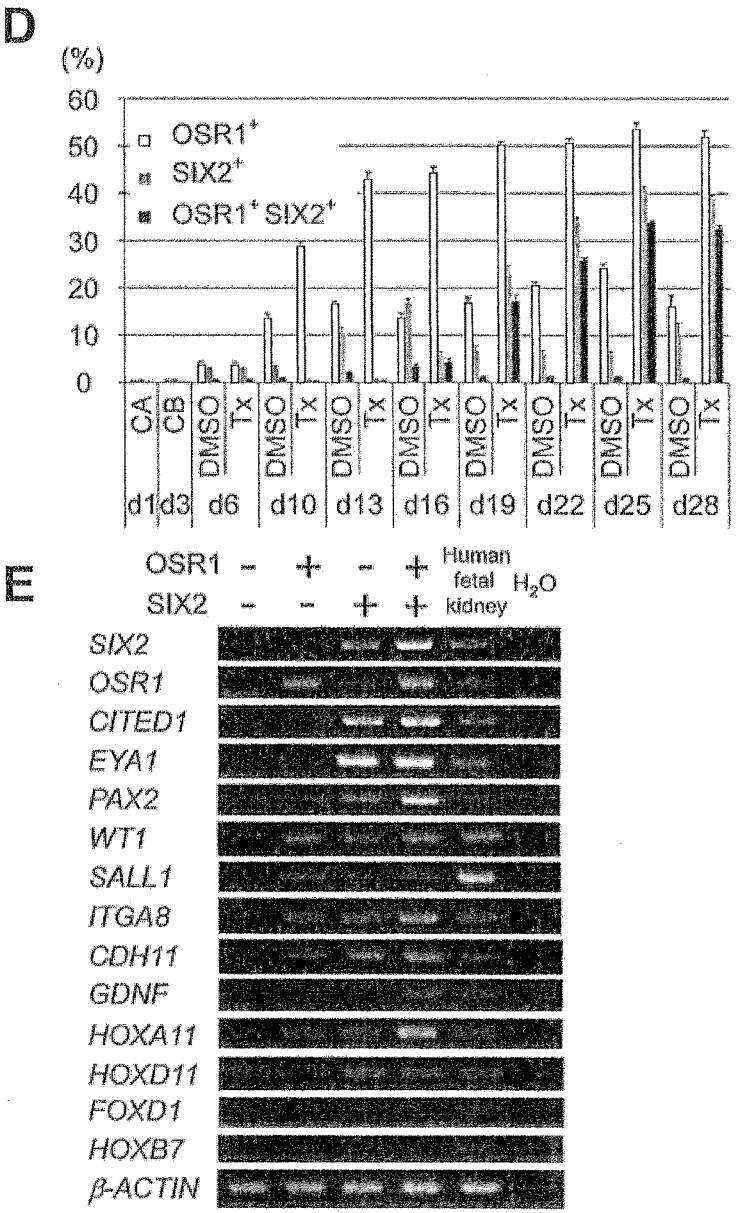


Fig. 4

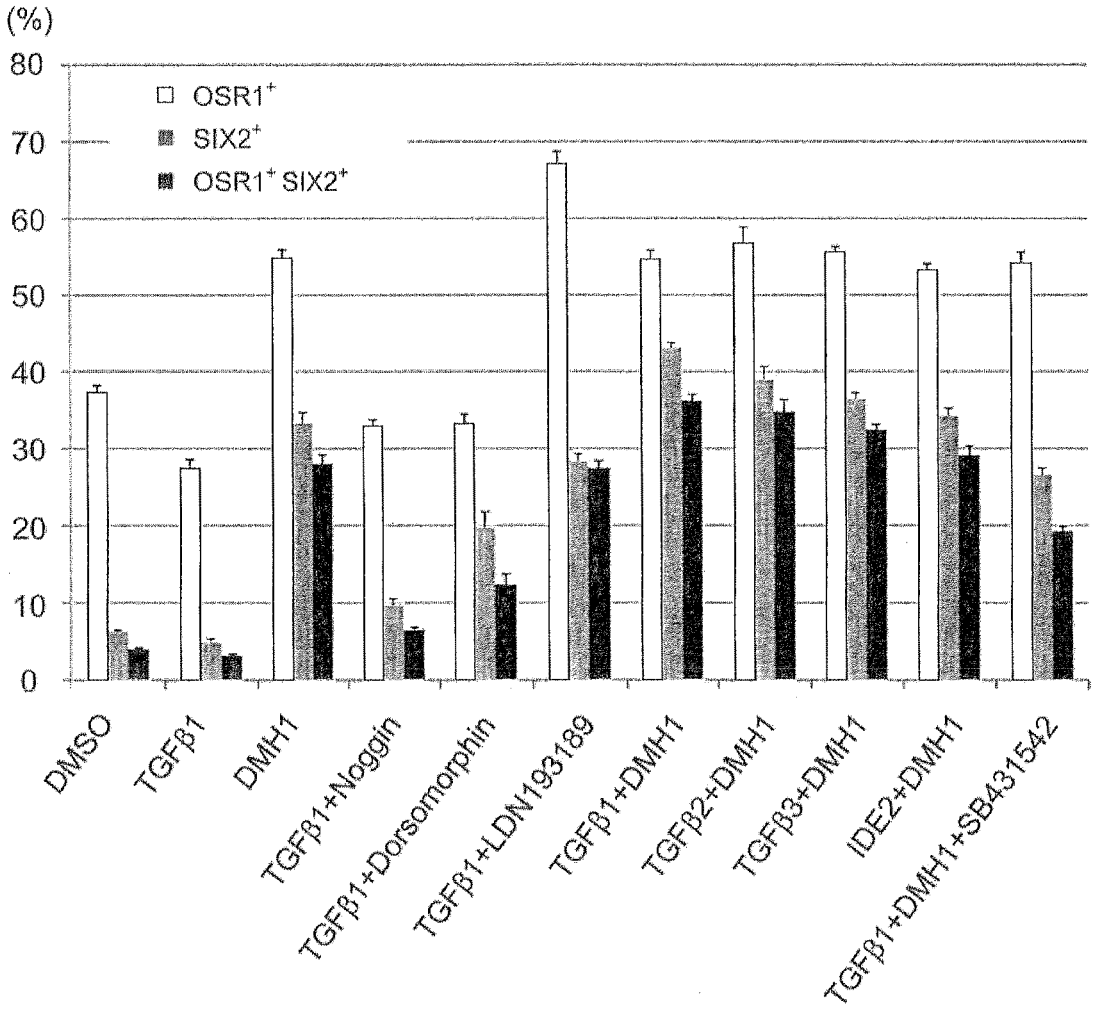

(%)
80
70
60
50
40
30
20
10
0
OSR1+
SIX2+
OSR1+ SIX2+
DMSO
TGFβ1
DMH1
TGFβ1+Noggin
TGFβ1+Dorsomorphin
TGFβ1+LDN193189
TGFβ1+DMH1
TGFβ2+DMH1
TGFβ3+DMH1
IDE2+DMH1
TGFβ1+DMH1+SB431542

Fig. 5

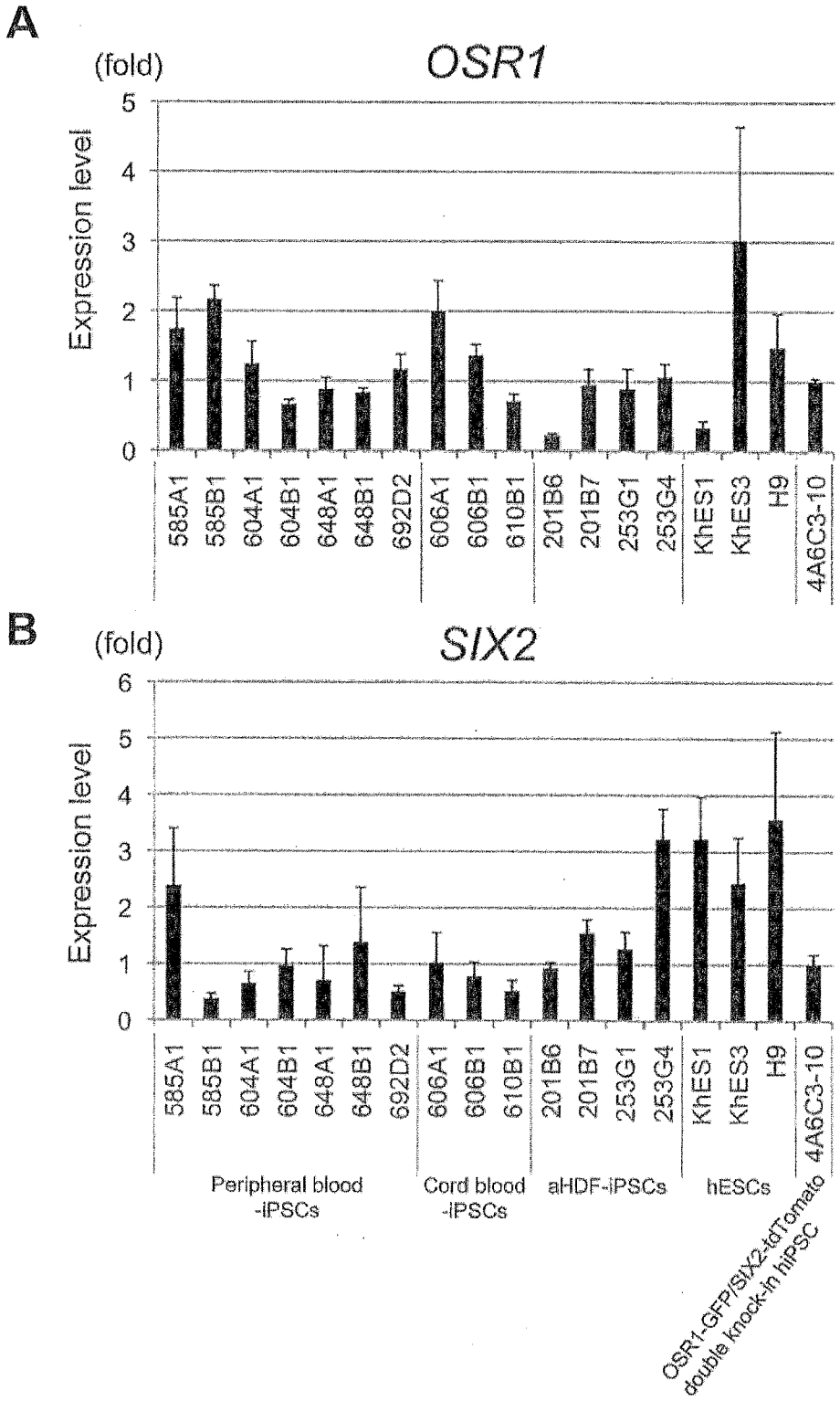

A
(fold)
OSR1
Expression level
5
4
3
2
1
0
585A1
585B1
604A1
604B1
648A1
648B1
692D2
606A1
606B1
610B1
201B6
201B7
253G1
253G4
KhES1
KhES3
H9
4A6C3-10
B
(fold)
SIX2
Expression level
6
5
4
3
2
1
0
585A1
585B1
604A1
604B1
648A1
648B1
692D2
606A1
606B1
610B1
201B6
201B7
253G1
253G4
KhES1
KhES3
H9
4A6C3-10
Peripheral blood -iPSCs
Cord blood -iPSCs
aHDF-iPSCs
hESCs
OSR1-GFP/SIX2-tdTomato double knock-in hiPSC

Fig. 6

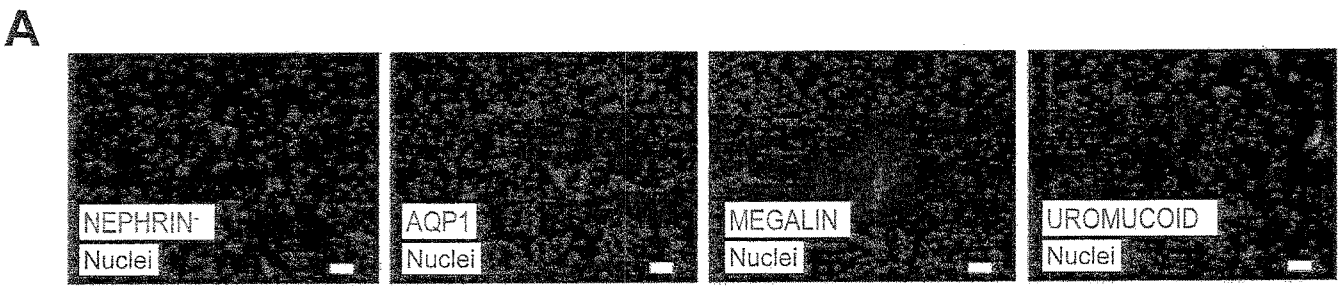

A
NEPHRIN-
Nuclei
AQP1
Nuclei
MEGALIN
Nuclei
UROMUCOID
Nuclei

Fig. 6 (Continued)

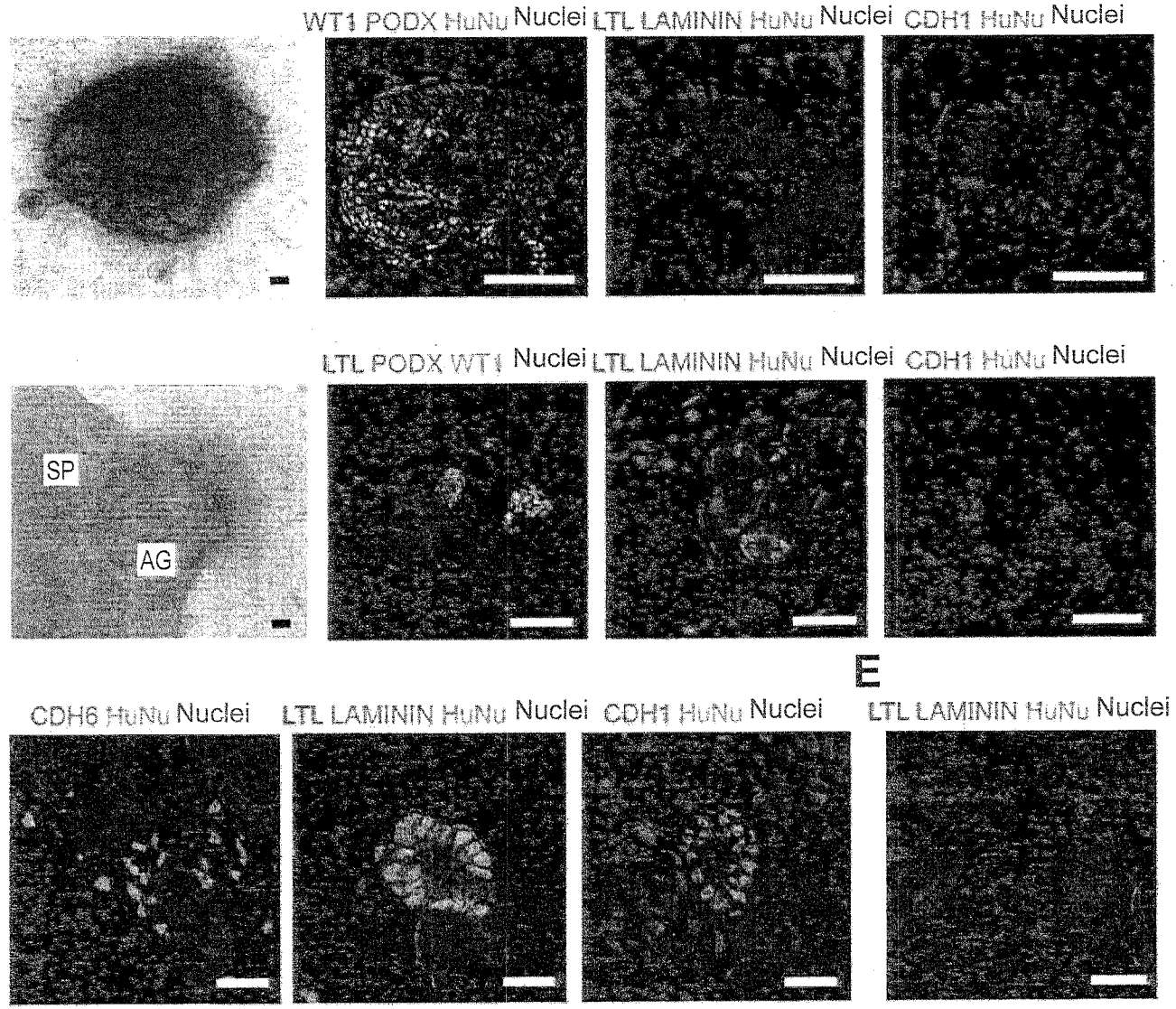


Fig. 6 (Continued)

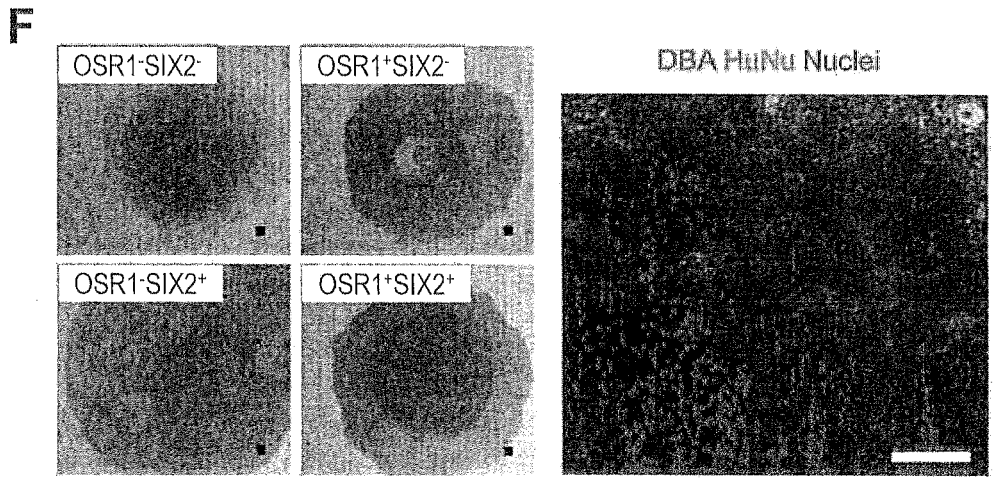


Fig. 7

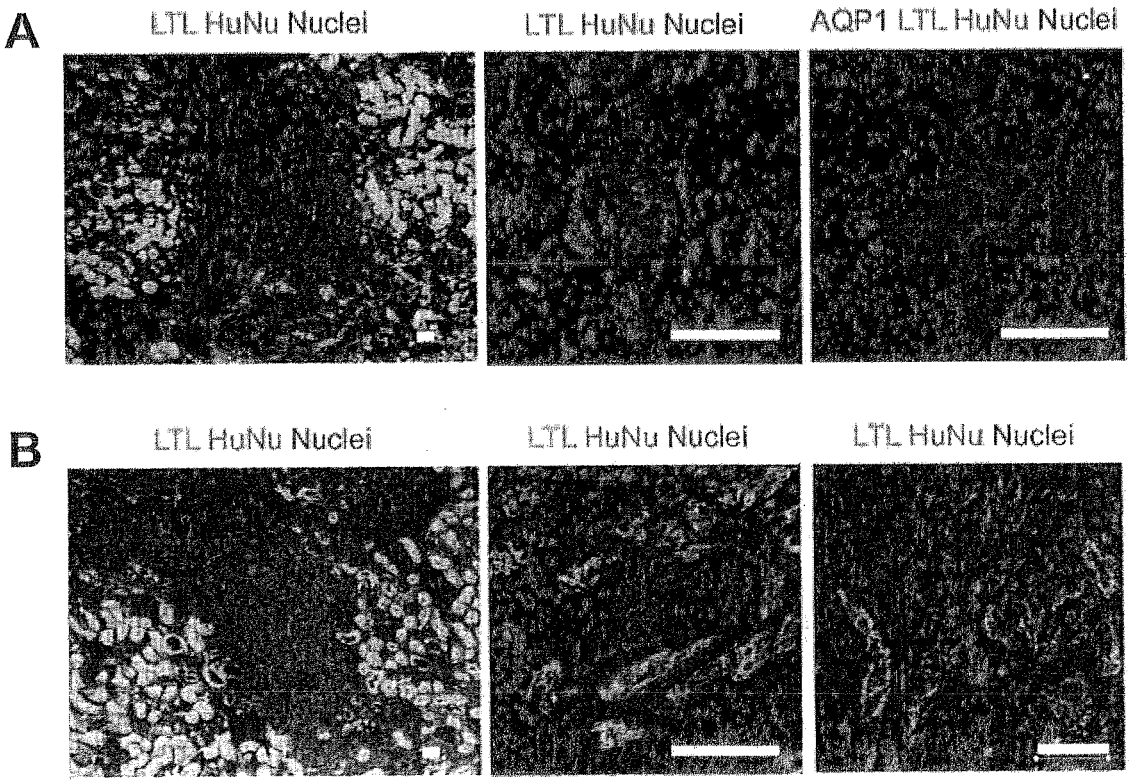

A
LTL HuNu Nuclei
LTL HuNu Nuclei
AQP1 LTL HuNu Nuclei
B
LTL HuNu Nuclei
LTL HuNu Nuclei
LTL HuNu Nuclei

Fig. 7 (Continued)

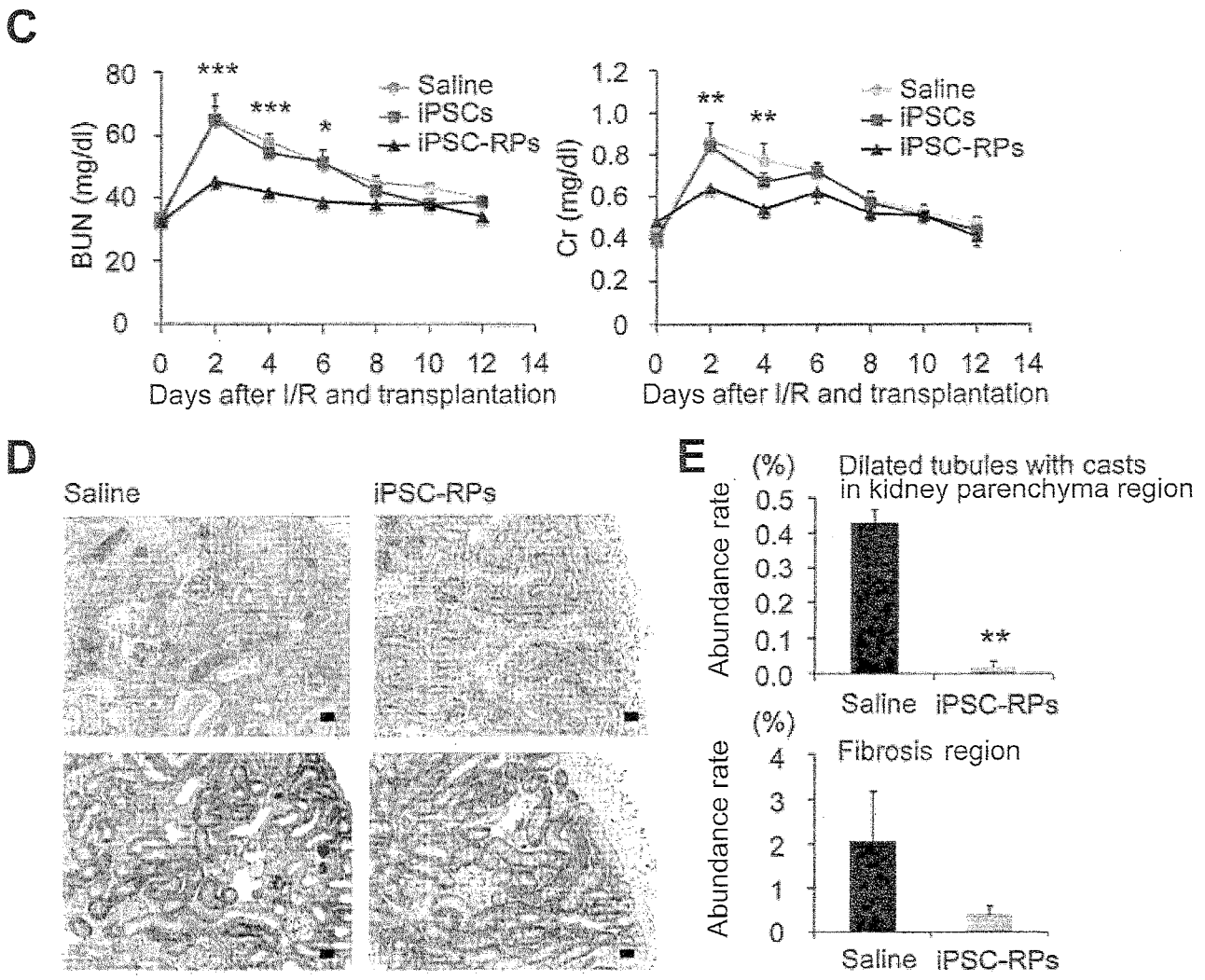


Fig. 1

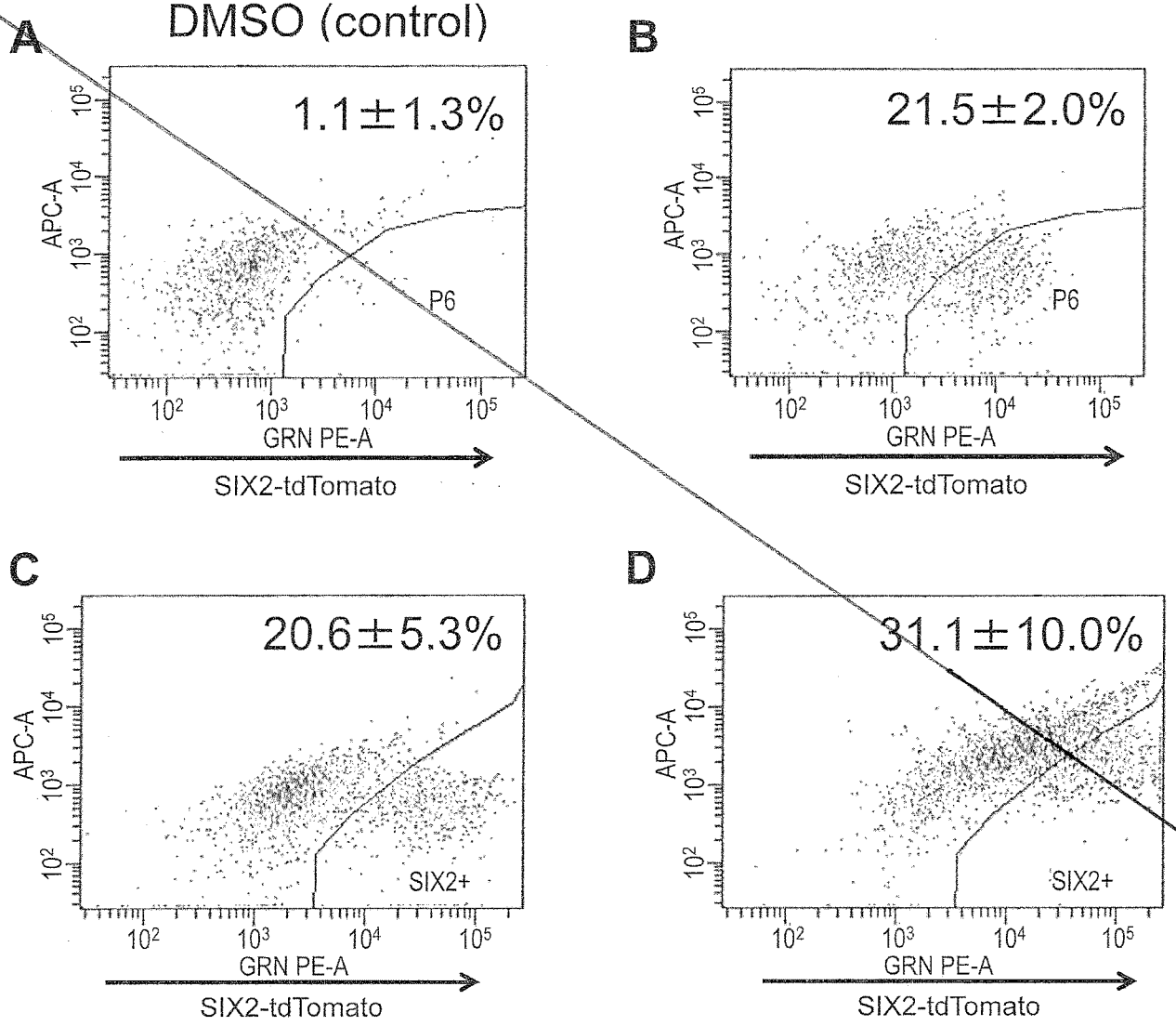

A
DMSO (control)
1.1±1.3%
P6
APC-A
GRN PE-A
SIX2-tdTomato
B
21.5±2.0%
P6
APC-A
GRN PE-A
SIX2-tdTomato
C
20.6±5.3%
SIX2+
APC-A
GRN PE-A
SIX2-tdTomato
D
31.1±10.0%
SIX2+
APC-A
GRN PE-A
SIX2-tdTomato

Fig. 2

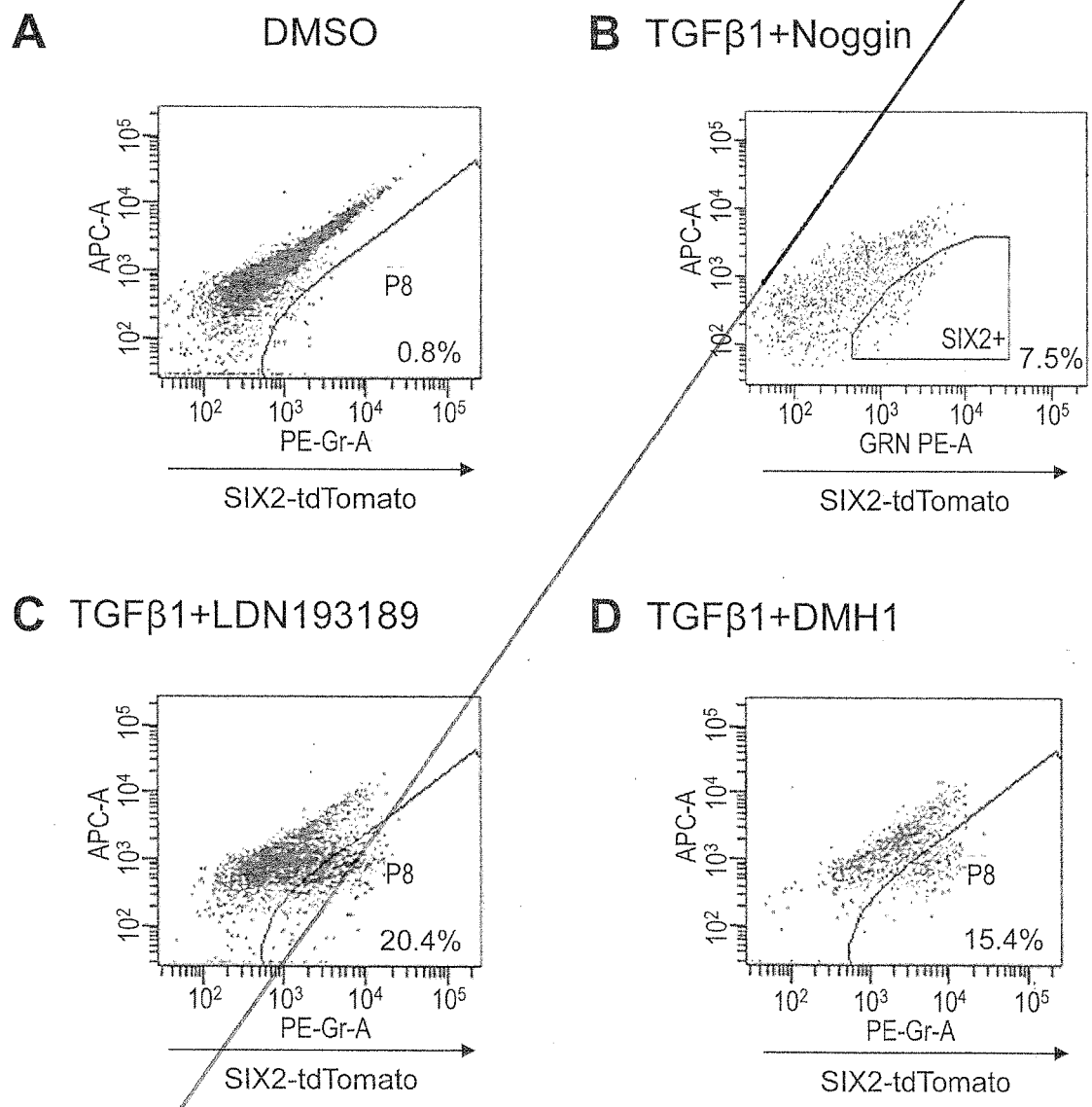
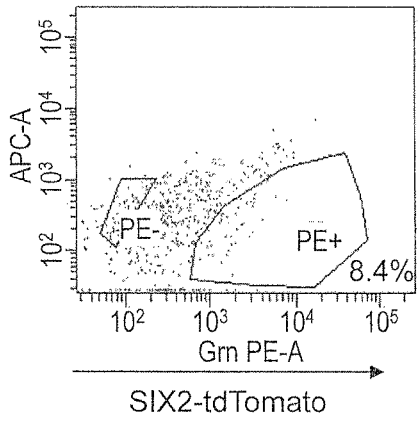
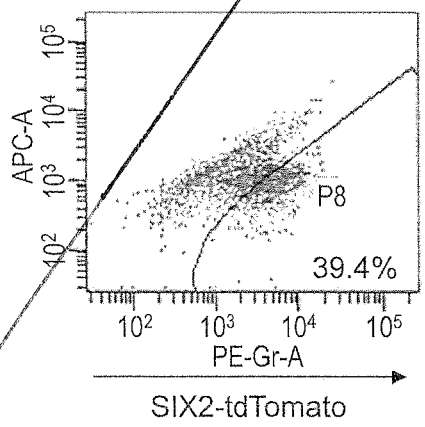

A
DMSO
APC-A
P8
0.8%
PE-Gr-A
SIX2-tdTomato
B TGFβ1+Noggin
APC-A
SIX2+
7.5%
GRN PE-A
SIX2-tdTomato
C TGFβ1+LDN193189
APC-A
P8
20.4%
PE-Gr-A
SIX2-tdTomato
D TGFβ1+DMH1
APC-A
P8
15.4%
PE-Gr-A
SIX2-tdTomato

Fig. 2 (Continued)

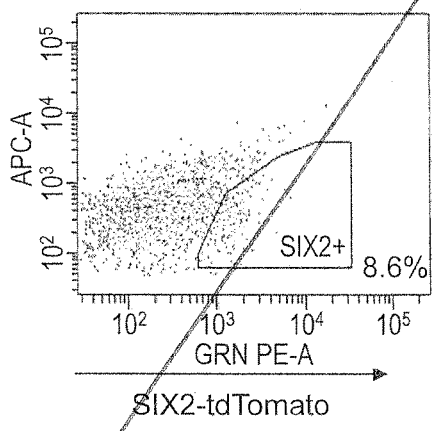
E IDE1+Dorsomorphin



F IDE2+Dorsomorphin



G TGFβ2+LDN193189



H TGFβ3+LDN193189

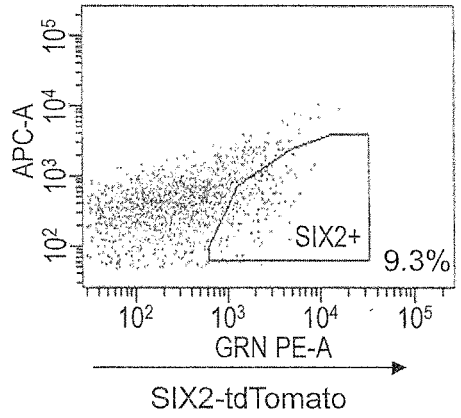
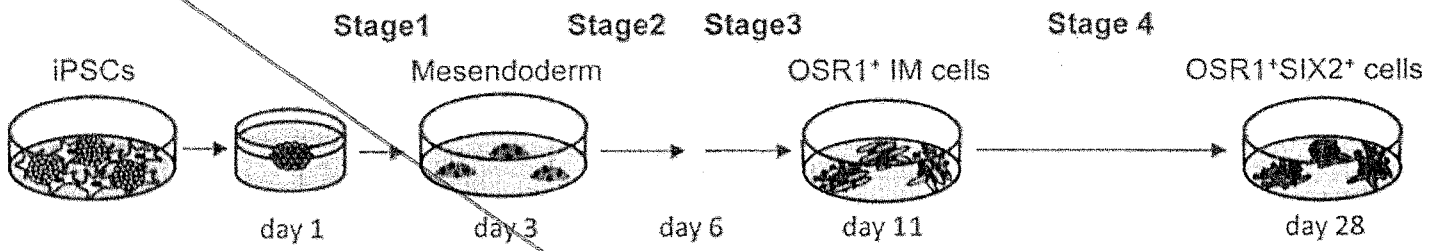


Fig. 3

A



Stage1	Stage2	Stage3	Stage4
Suspension culture	Adhesion culture		
DMEM/F12 Glutamax			
2% FBS, 0.5% P/SM	10% KSR, 50 μM 2ME, 1% NEAA, 0.5% P/SM		
activin A (100 ng/ml)	BMP7 (100 ng/ml)	TGFβ1 (5 ng/ml)	TGFβ1 (5 ng/ml)
CHIR99021 (1 μM)	CHIR99021 (1 μM)	TTNPB (1 μM)	DMH1 (0.5 μM)

B

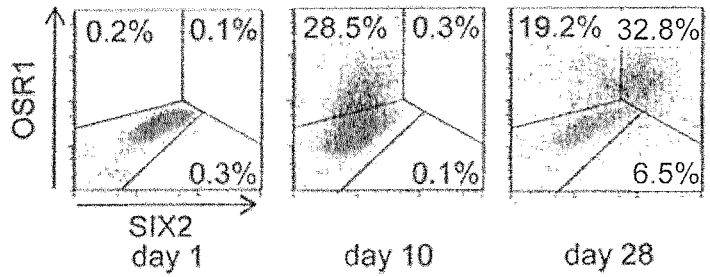


Fig. 3 (Continued)

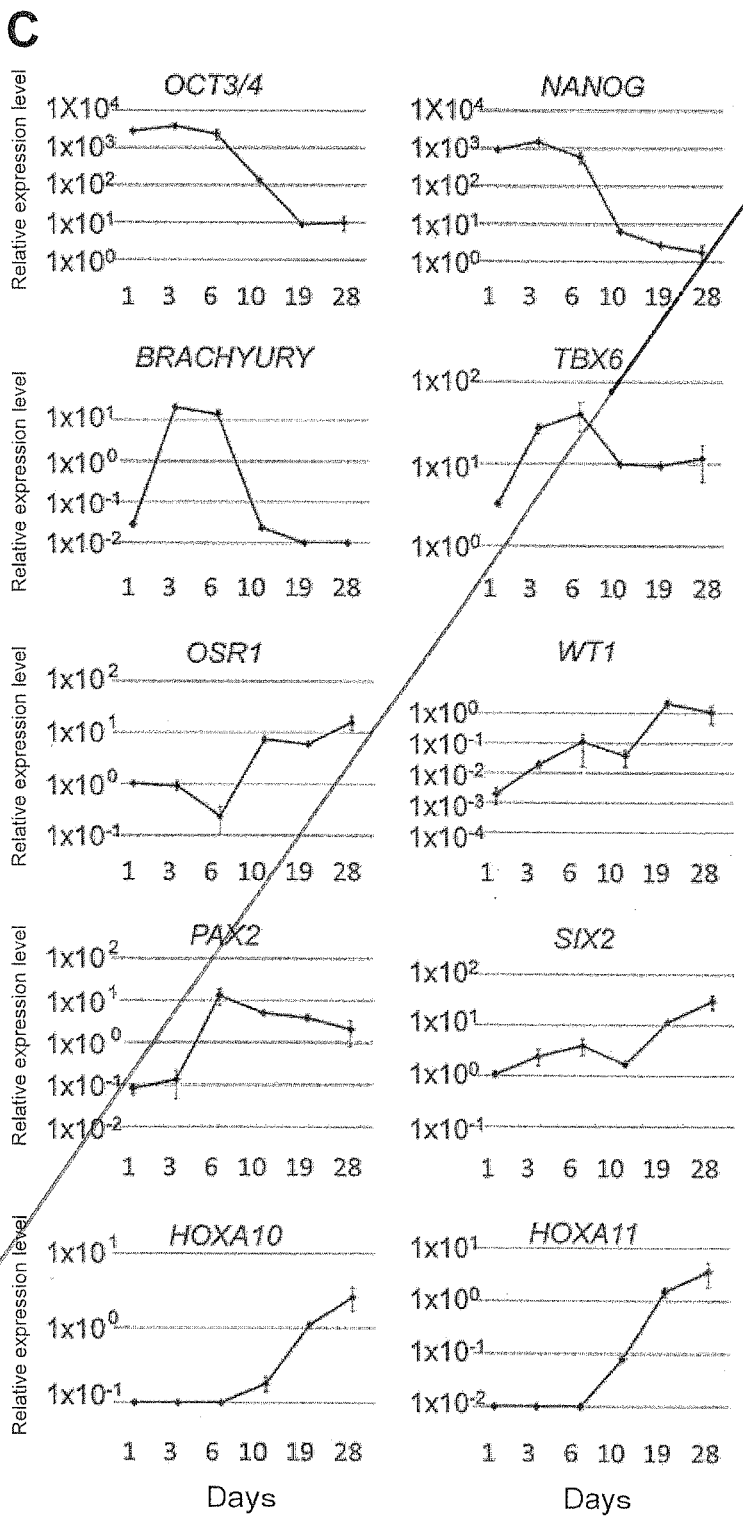


Fig. 3 (Continued)

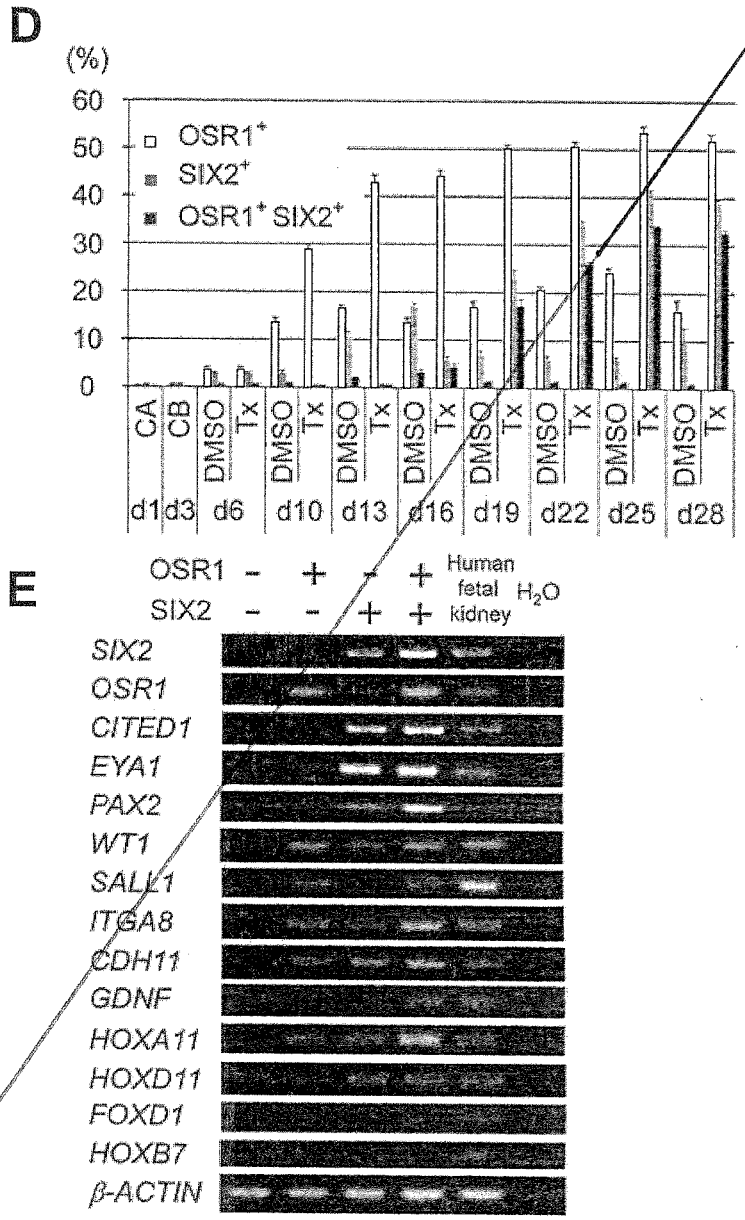


Fig. 4

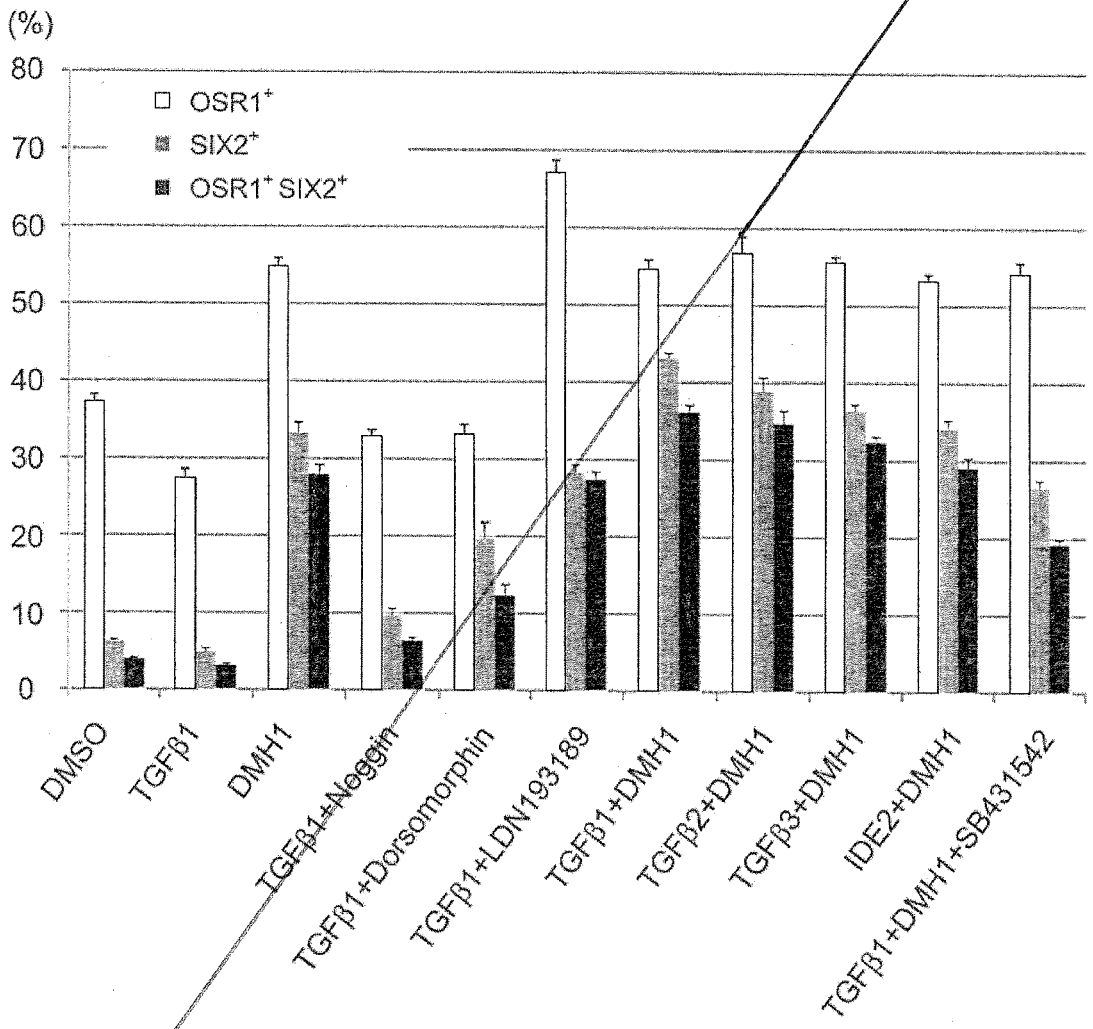

(%)
80
70
60
50
40
30
20
10
0
OSR1+
SIX2+
OSR1+SIX2+
DMSO
TGFβ1
DMH1
TGFβ1+Noggin
TGFβ1+Dorsomorphin
TGFβ1+LDN193189
TGFβ1+DMH1
TGFβ2+DMH1
TGFβ3+DMH1
IDE2+DMH1
TGFβ1+DMH1+SB431542

Fig. 5

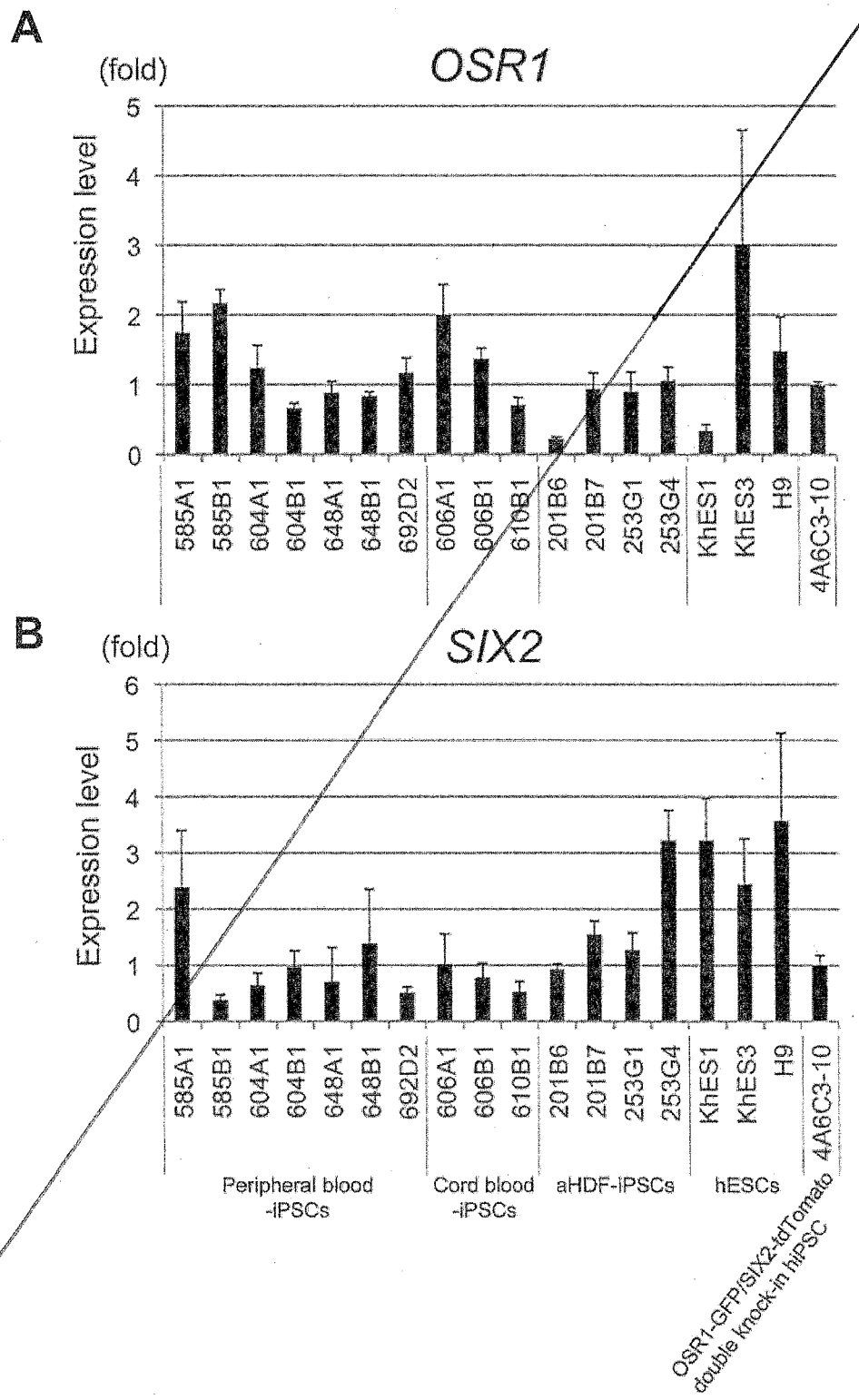

A
(fold)
OSR1
Expression level
5
4
3
2
1
0
585A1
585B1
604A1
604B1
648A1
648B1
692D2
606A1
606B1
610B1
201B6
201B7
253G1
253G4
KhES1
KhES3
H9
4A6C3-10
B
(fold)
SIX2
Expression level
6
5
4
3
2
1
0
585A1
585B1
604A1
604B1
648A1
648B1
692D2
606A1
606B1
610B1
201B6
201B7
253G1
253G4
KhES1
KhES3
H9
4A6C3-10
Peripheral blood -iPSCs
Cord blood -iPSCs
aHDF-iPSCs
hESCs
OSR1-GFP/SIX2-tdTomato double knock-in hiPSC

Fig. 6

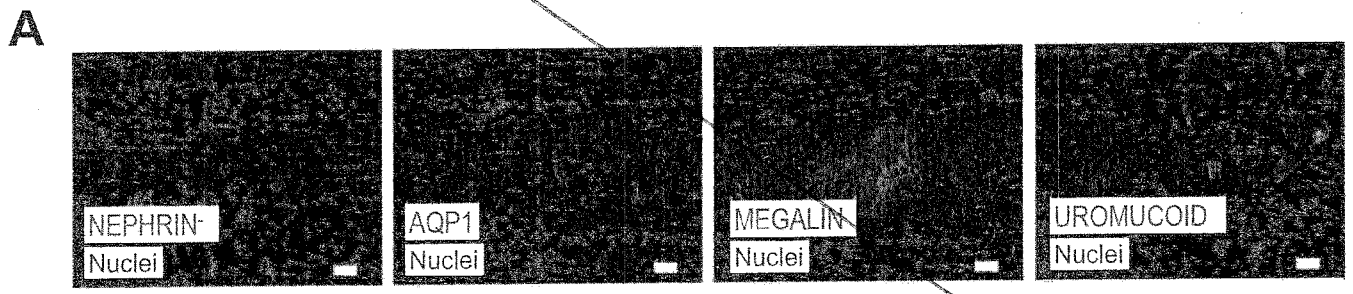

A
NEPHRIN
Nuclei
AQP1
Nuclei
MEGALIN
Nuclei
UROMUCOID
Nuclei

Fig. 6 (Continued)

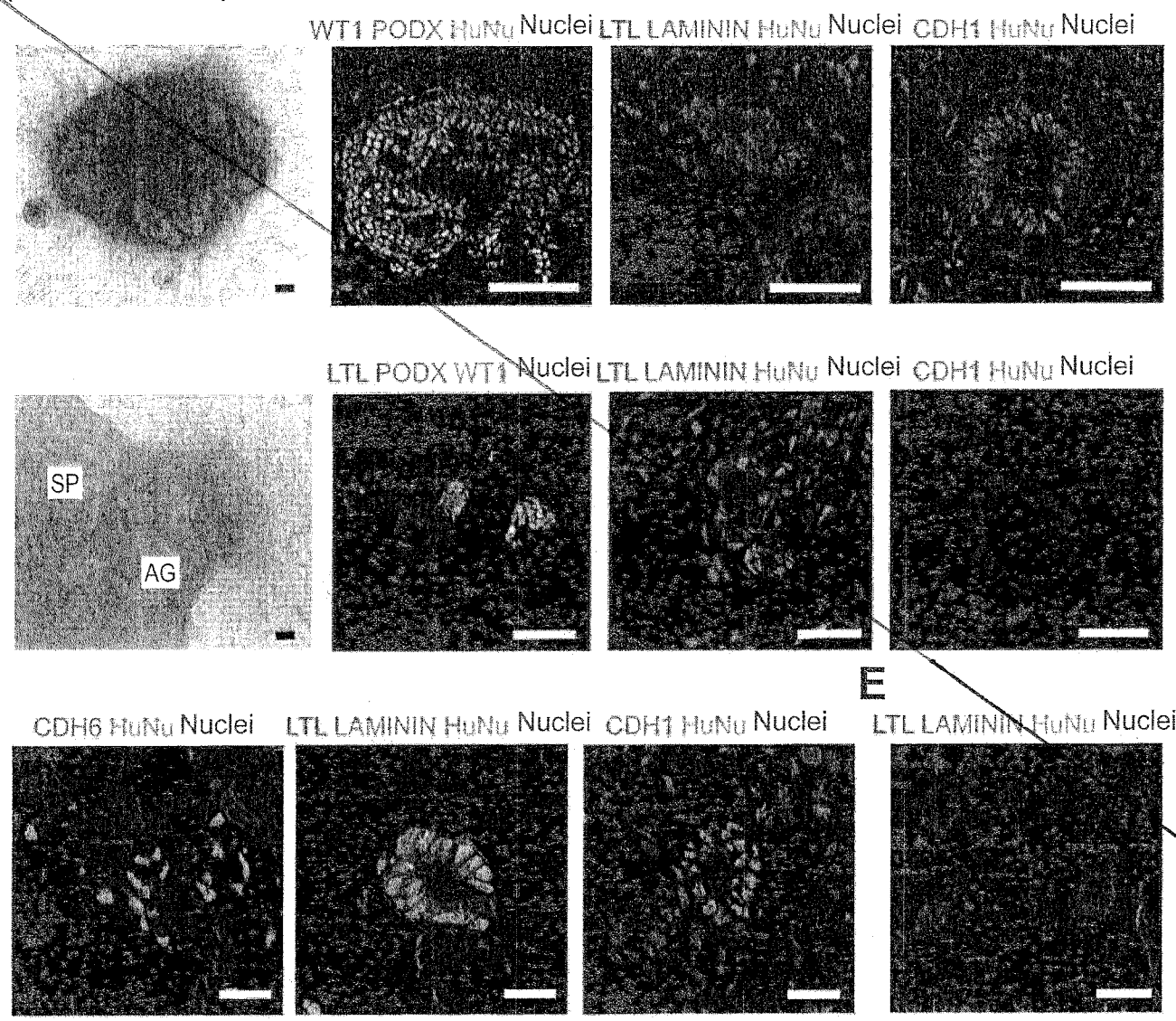


Fig. 6 (Continued)

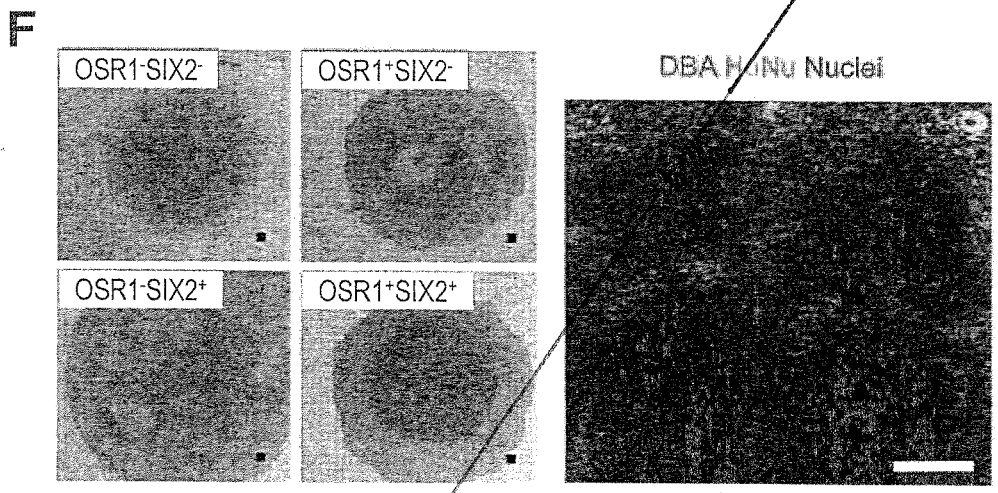


Fig. 7

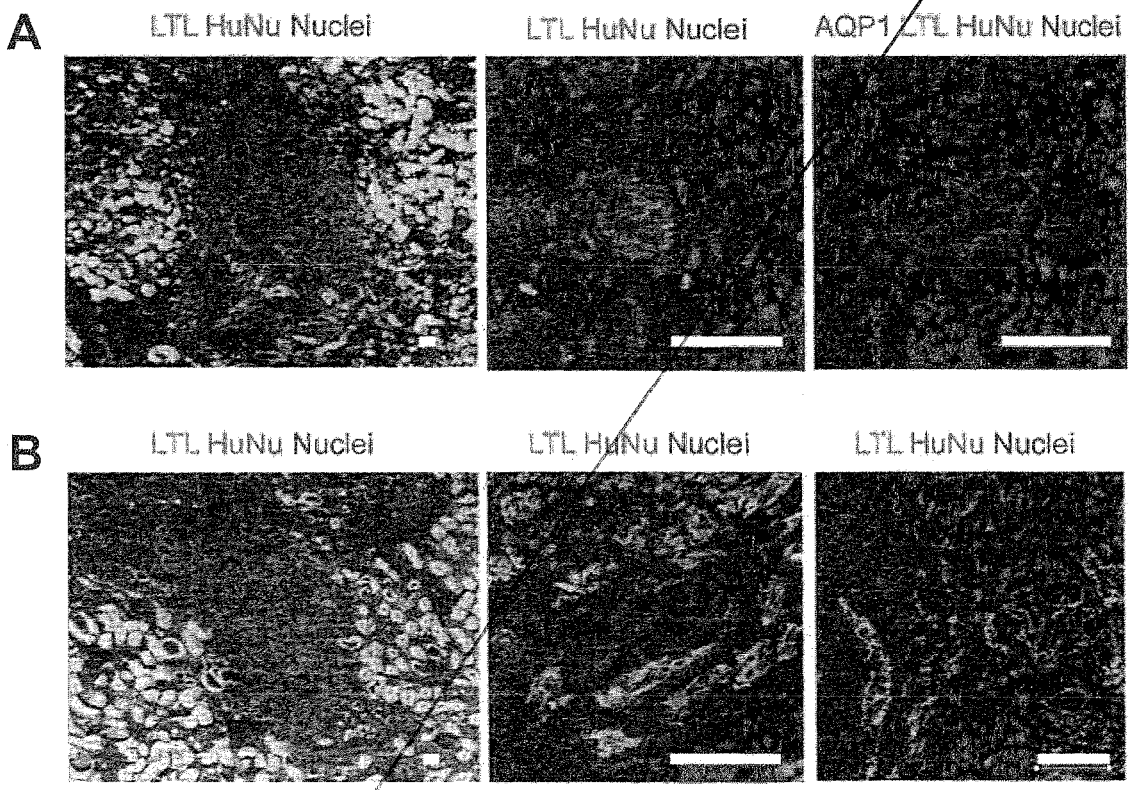

A
LTL HuNu Nuclei
LTL HuNu Nuclei
AQP1 LTL HuNu Nuclei
B
LTL HuNu Nuclei
LTL HuNu Nuclei
LTL HuNu Nuclei

Fig. 7 (Continued)

