

Characterization of hPSC

Cell Line Name	SNUhES4		
Type of Cell Line	hESC		
Depositor (Institution)	Seoul National University		
Passage #	p28*		
Day of Cell Freezing	20120927*		
Analysis	Result	Passage #	Day of analysis
Cell viability (RCB phase)	Pass (49.4±11%)	p16	20130307
Cell viability (DCB phase-release test)	Pass (62.7±3.6%)	p28	20170825
Authentication (STR)	Pass	p18	20130328
Mycoplasma test (PCR)	Pass	p16	20130212
Mycoplasma test (Luminometer)	Pass	p16	20130212
Cell attachment & Colony morphology	Pass	p16	20130307
Microbial contamination test (Virus, Fungi, bacteria)	Pass	p18	20130425
Cell doubling time	Pass	p16	20130627
Karyotype (G-banding)	46,XY	p18	20130325
HLA genotype	HLA-A *02:01 *11:01G HLA-B *15:01(G62) *55:07 HLA-DRB1 *08:03 *13:02	p18	20130325
ABO genotype)	OO	p18	20130722
Stem Cell Marker Expression			
· AP	Pass (Positive)	p18	20130722
· ICC	Pass (Positive)	p18	20130723
· RT-PCR	Pass (Positive)	p19	20130527
· qRT-PCR	Pass (Positive)	p19	20130527
Differentiation Marker Expression			
· EB formation	Pass (Positive)	p19	20130822
· RT-PCR	Pass (Positive)	p19	20130916
· qRT-PCR	Pass (Positive)	p19	20130916

* Freezing media : 95% Serum with 5% DMSO

* The cryopreserved stock of this cell was provided by depositor (SNU).

Cell Culture Condition

· Feeder	STO (mouse embryonic fibroblast; ATCC CRC-1503)
· Media	hPS media (DMEM/F12 supplemented KSR and FGF2)
· Passage (Cell dissociation)	Manual detachment/Dispase II (Gibco, #17105-041)

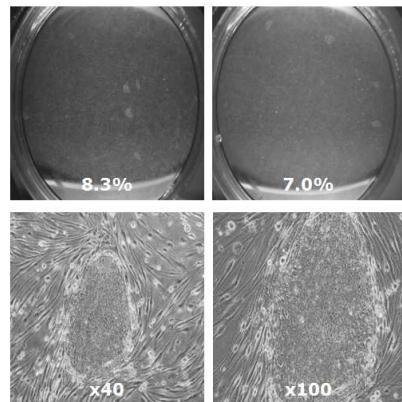
Mycoplasma contamination test

시료명	판정 (positive, negative or 재검사)					
SNUhES4 p16	negative					
	1st PCR		2nd PCR			
	1	2	3	1	2	3
	1. SNUhES4 p16	2. NC	3. PC			

시료명	A 값	B 값	B/A 비율†	판정 (positive, negative or 재검사)
SNUhES4 p16	3368	1445	0.43	negative
NC	3015	676	0.22	negative
PC	2255	97736	43.34	Positive

† < 0.9 Negative 마이코플라스마 미검출
≥ 1.2 Positive 마이코플라스마 오염
≥ 0.9~1.2 Borderline 제시험

Cell attachment & Morphology



세포부착률

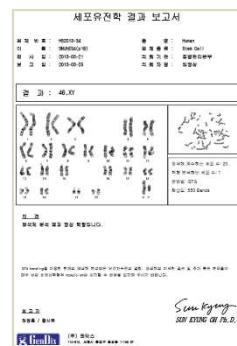
Colony d7/ Clump d0
(mean±SD)

7.7±0.9%

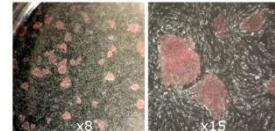
Microbial contamination test

검사종목	참고치	결과 SNUhES4
HBV DNA 정량	**	**
Copies/mL	<116	<116
IU/mL	<20	<20
Pg/mL	<0.0004	<0.0004
HCV RNA 정량	**	**
IU/mL	≤15	15이하
Copies/mL	≤40	40이하
HIV RNA 정량	<20	<20
HTLV I& II Ab (의주)	Negative	Negative
EBV PCR(정성)	Negative	Negative
CMV PCR(정성)	Negative	Negative
HPV DNA 정량(Type16,Type18)	**	**
고위험군HPV		Negative
HPV Viral load	<1	0.09
HSV Type I PCR(정성)	Negative	Negative
HSV Type II PCR(정성)	Negative	Negative
HSV PCR(정성)	Negative	Negative
HHV Type 6 PCR (정성)	Negative	Negative
Culture & ID	No growth	No growth
Fungus Culture	No growth	No growth

Karyotype

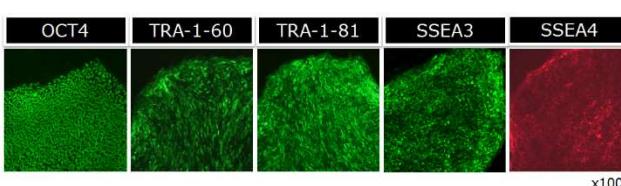


AP staining



Stem cell marker expression

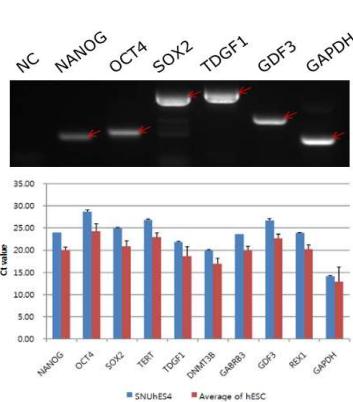
<ICC>



<qRT-PCR>

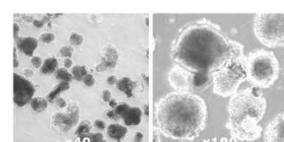
GENE	CTmean
NANOG	23.94
OCT4	28.71
SOX2	24.96
TERT	26.84
TDGF1	21.83
DNMT3B	19.92
GABRB3	23.60
GDF3	26.69
REX1	23.93
ACTB	18.88
RN18S1	14.08
18S	14.98
GAPDH	14.12

<RT-PCR>



Differentiation and marker expression

EB formation (day14)



<qRT-PCR>

GENE	CTmean
PAX6	32.78
SOX1	36.22
HNF3B	32.82
AFP	25.92
T	28.73
MYOG	35.37
ATCB	19.53
RN18S1	14.74
18S	16.07
GAPDH	19.79

<RT-PCR>

