

# Characterization of hPSC

<b>Cell Line Name</b>	<b>CHA-hES15</b>			
<b>Type of Cell Line</b>	<b>hESC</b>			
<b>Depositor (Institution)</b>	<b>CHA University</b>			
<b>Passage #</b>	<b>p25*</b>			
<b>Day of Cell Freezing</b>	<b>20160324</b>			
<b>Analysis</b>	<b>Result</b>	<b>Passage#</b>	<b>Day of analysis</b>	
Cell viability	Pass (88.0±4%)	p21	20161013	
Authentication (STR)	Pass	p26	20160531	
Mycoplasma test (PCR)	Pass	p22	20160302	
Mycoplasma test (Luminometer)	Pass	p22	20160302	
Cell attachment & colony morphology	Pass	p22	20160314	
Microbial contamination test (Virus, Fungi, bacteria)	Pass	p28	20141210	
Karyotype (G-banding)	46,XY	p29	20141010	
HLA genotype	HLA-A *31:01g *33:03	p29	20141010	
	HLA-B *44:03 *46:01			
	HLA-DRB1 *04:03 *13:02/39			
ABO genotype)	AO	p29	20141010	
<b>Stem Cell Marker Expression</b>				
· AP staining	Pass (Positive)	p23	20160329	
· ICC	Pass (Positive)	p23	20161006	
· qRT-PCR	Pass (Positive)	p23	20160727	
<b>Differentiation Marker Expression</b>				
· EB formation	Pass (EB14d)	p23	20160401	
· qRT-PCR	Pass (Positive)	p23	20160728	
* Freezing media : mFreSR (Stem Cell Technol. #05855)				

## Cell Culture Condition

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

### Mycoplasma contamination test

<PCR>

시험일자: 2016년 03월 02일

시료명	판정 (positive, negative or 재검사)
CHA-hES15 p22	negative

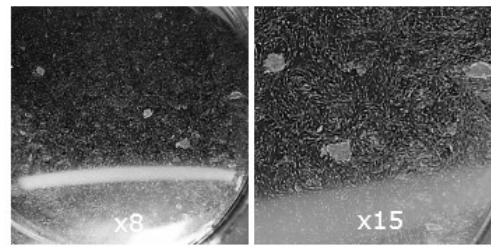
<루미노메터>

시험일자: 2016년 03월 02일

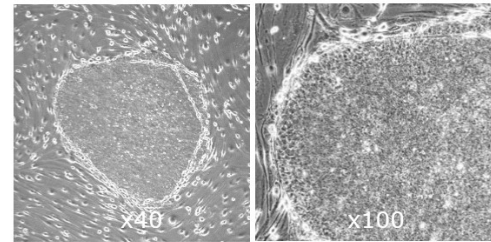
시료명	A 값	B 값	B/A 비율'	판정 (positive, negative or 재검사)
CHA-hES15 p22	3084	1264	0.41	Negative
NC	7881	783	0.10	Negative
PC	2950	163168	55.31	Positive

† < 0.9 Negative 마이코플라스마 미검출  
> 1.2 Positive 마이코플라스마 오염  
> 0.9~1.2 Borderline 재시험

### Cell attachment (Day 10)



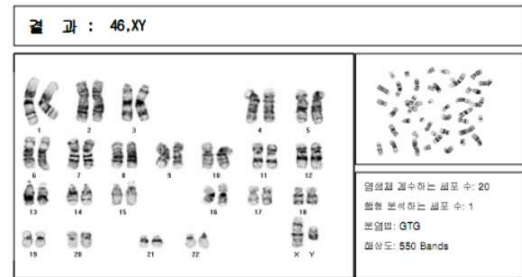
### Morphology



### Microbial contamination test

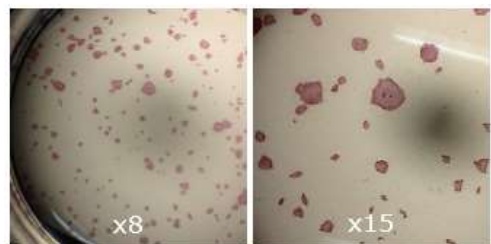
연번	검사종목	참고치	결과 CHA-hES15(p28)
1	HBV DNA 정량	**	**
	Copies/mL	<116	<116
	IU/mL	<20	<20
2	HCV RNA 정량	**	**
	IU/mL	≤15	15이하
	Copies/mL	≤40	40이하
3	HIV RNA 정량	<20	<20
4	HTLV I& II Ab (외주)	Negative	Negative
5	EBV PCR(정성)	Negative	Negative
6	CMV PCR(정성)	Negative	Negative
7	HPV DNA 정량(Type16, Type18)	**	**
	고위험군HPV HPV Viral load	<1	0.15
8	HSV Type I PCR(정성)	Negative	Negative
	HSV Type II PCR(정성)	Negative	Negative
	HSV PCR(정성)	Negative	Negative
9	HHV Type 6 PCR (정성)	Negative	Negative
10	Culture & ID	No growth	No growth
11	Fungus Culture	No growth	No growth

### Karyotype



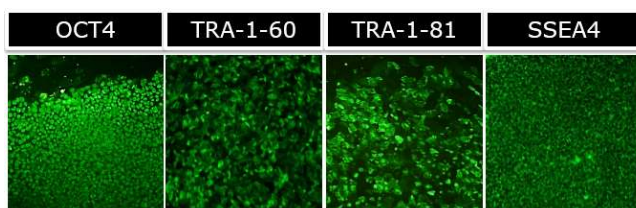
의견  
염색체 분석 결과 정상 현상입니다.

### AP staining



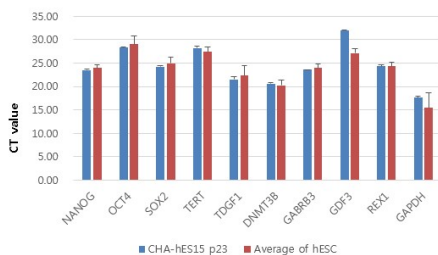
### Stem cell marker gene expression

<ICC>



<qRT-PCR>

GENE	CTmean
NANOG	23.48
OCT4	28.29
SOX2	24.27
TERT	28.18
TGDF1	21.55
DNMT3B	20.66
GABRB3	23.57
GDF3	32.02
REX1	24.37
ACTB	18.50
RN18S1	12.64
18S	12.46
GAPDH	17.60



### Differentiation marker gene expression

<EB formation>



<qRT-PCR>

GENE	CTmean
PAX6	23.28
SOX1	32.44
AFP	26.68
HNF3B	31.64
T	29.20
MYOG	35.50
ACTB	19.90
RN18S	12.59
18S	13.16
GAPDH	18.69