iPSC Characterization Report

Information Summary

Organization: Neural Stem Cell Institute (NSCI)
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SCRC Code: NSCI5

Sample Name: GIH-56

Number of the Established iPSC Clones: 6

Characterization Performed:

1. Mycoplasma Test
2. Cell authentication Verification
3. Presence of the Pluripotency Marker
4. Spontaneous differentiation via embryoid body (EB) formation
5. Karyotyping

Qualified iPSC Clones: C1, C2, C4

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Mycoplasma Test

Method: Determination of the mycoplasmal enzyme activity using Lonza MycoAlert Plus Detection kit.

iPSC Clone	NSCI5-C1	NSCI5-C2	NSCI5-C4
Cell passage	P6	P6	P6
B/A Ratio	0.56	0.54	0.42
Mycoplasma	NEG	NEG	NEG
Pass or Fail	Pass	Pass	Pass

Cell Authentication Verification

Markers	NSCI5 (GIH-56)	NSCI5-C1	NSCI5-C2	NSCI5-C4
D3S1358	14, 18	14, 18	14, 18	14, 18
TH01	7, 9.3	7, 9.3	7, 9.3	7, 9.3
D21S11	31.2, 33.2	31.2, 33.2	31.2, 33.2	31.2, 33.2
D18S51	12, 14	12, 14	12, 14	12, 14
Penta E	10, 12	10, 12	10, 12	10, 12
D5S818	11, 13	11, 13	11, 13	11, 13
D13S317	11, 12	11, 12	11, 12	11, 12
D7S820	9, 10	9, 10	9, 10	9, 10
D16S539	11, 13	11, 13	11, 13	11, 13
CSF1PO	11, 13	11, 13	11, 13	11, 13
Penta D	9, 13	9, 13	9, 13	9, 13
vWA	14, 18	14, 18	14, 18	14, 18
D8S1179	8, 14	8, 14	8, 14	8, 14
TPOX	8, 12	8, 12	8, 12	8, 12
FGA	22.2, 25	22.2, 25	22.2, 25	22.2, 25
AMEL	X, Y	X, Y	X, Y	X, Y
Match with original Fibroblast		**Yes**	**Yes**	**Yes**

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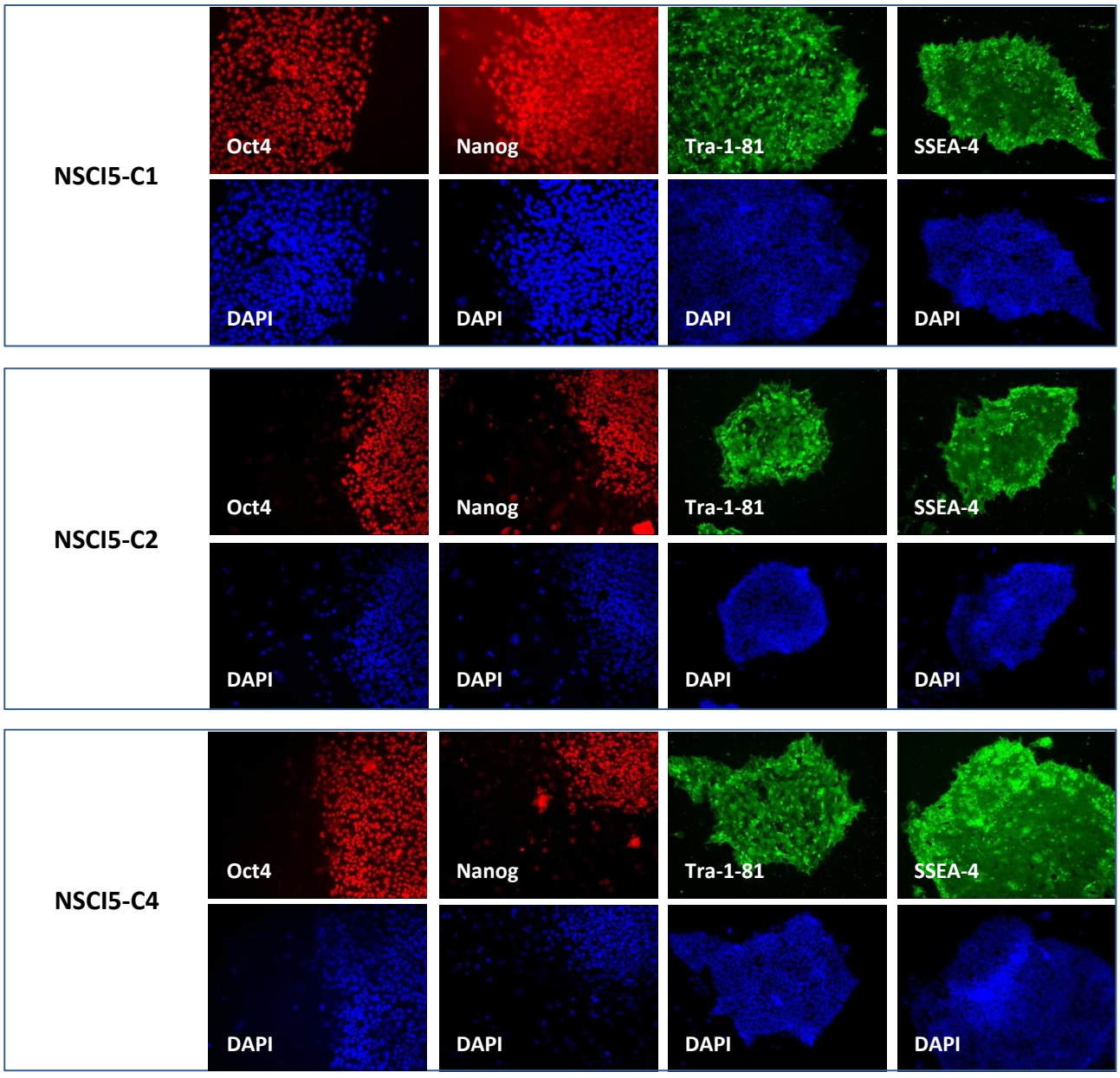
Expression of the Pluripotency Markers

Antibody staining for pluripotency markers: Oct4, Nanog, Tra-1-81, SSEA-4



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Differentiation Potential via EB Formation

Method: The iPSC cells were disassociated into small clumps to form EB in the low attachment dish. After a 10-day spontaneous differentiation, the EBs were harvested and RNAs were extracted . The expression levels of the selected 3-germ layer specific genes were analyzed by real-time PCR. Ct values are normalized for loading using a housekeeping gene.

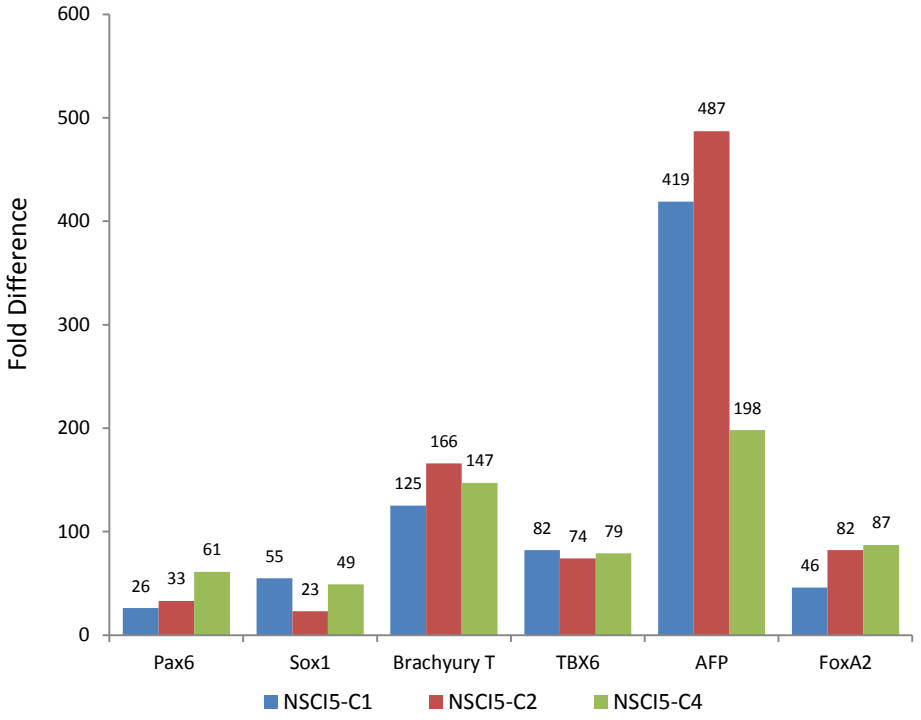

Figure: Lineage specific gene expression following EB differentiation. Fold difference is shown relative to undifferentiated iPS cell.

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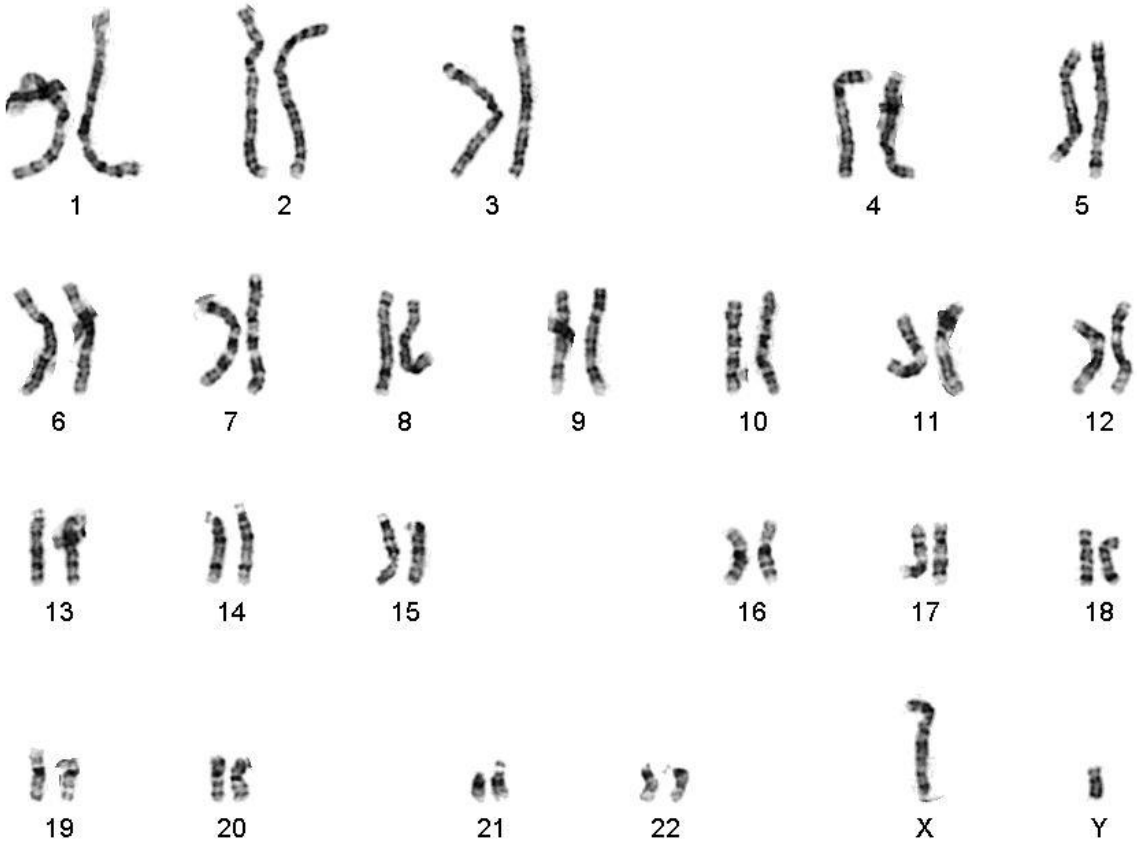
Karyotyping

Clone: NSCI5(GIH-56)-C1
Passage: P7

Tested: Number of metaphases counted: 20 Banding and level: GTG/550
Number of metaphases analyzed: 6 Number of karyotypes: 2

Result: 46,XY NORMAL MALE KARYOTYPE



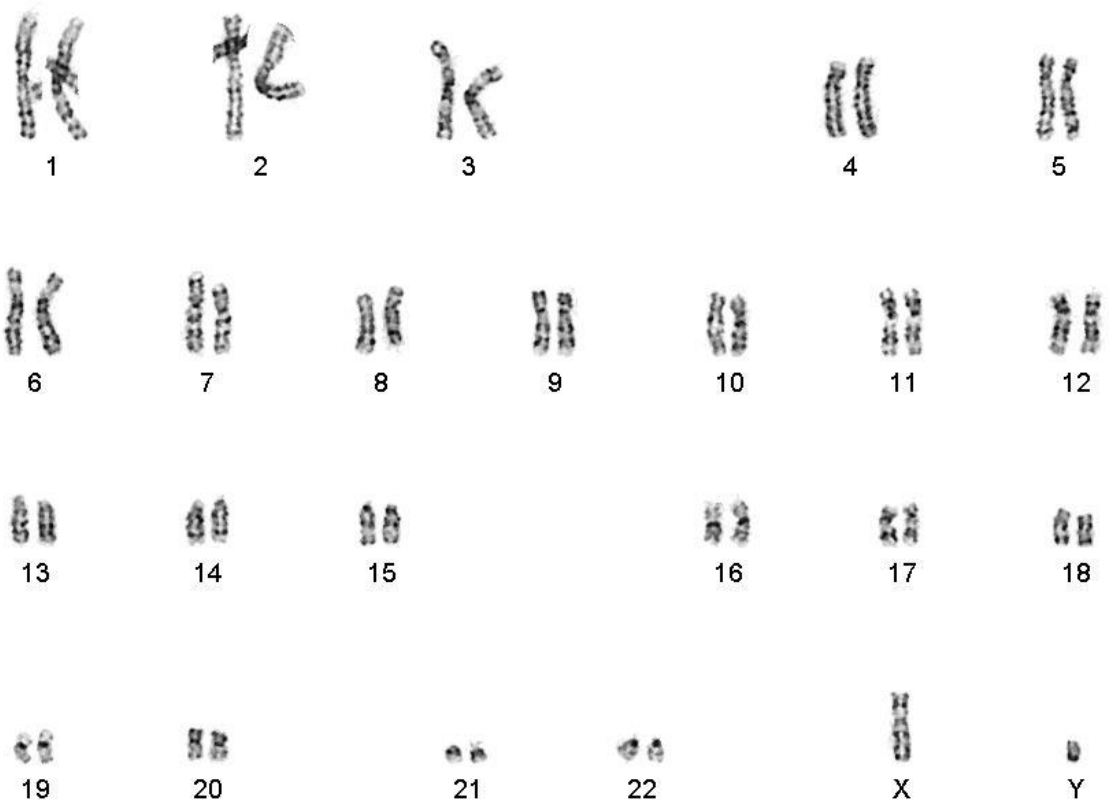
iPSC Characterization Report

Karyotyping

Clone: NSCI5(GIH-56)-C2
Passage: P7

Tested: Number of metaphases counted: 20 Banding and level: GTG/550
Number of metaphases analyzed: 6 Number of karyotypes: 2

Result: 46,XY NORMAL MALE KARYOTYPE



iPSC Characterization Report

Karyotyping

Clone: NSCI5(GIH-56)-C4
Passage: P8

Tested: Number of metaphases counted: 20 Banding and level: GTG/550
Number of metaphases analyzed: 6 Number of karyotypes: 2

Result: 46,XY NORMAL MALE KARYOTYPE