CORIELL INSTITUTE
FOR MEDICAL RESEARCH

GM 24683*C

Certificate of Analysis

Product description	Human Fibroblast reprogrammed with 4 factors (hOCT3/4 with shp53, hSOX2, hKLF4, hL-MYC) using episomal vectors	
Publication(s) describing iPSC establishment	None. Coriell in house generated iPSC line	
Parent cell line and cell type	GM01014	Fibroblast
Diagnosis	Cystic Fibrosis; CF	
Passage at freeze	24	
Media	DMEM/F12 + 20% KOSR + 5ng/ml FGF	
Feeder or Matrix Substrate	CF1 MEFs on 0.1% Gelatin	
Passage method	TrypLE Express	
Split ratio	1:6 every 5-7 days	

The following testing specifications have been met for the specified product lot:

Test Description	Test Method	Test Specification	Result
Post-Thaw Viable Cell Recovery	Colony Doubling	Colony formation and diameter doubling within 5 days	Pass
Sterility	Growth on agar	Negative	Pass
Mycoplasma	PCR	Negative	Pass
Karyotype	G-banding	46,XY	Pass
Identity Match	STR (THO-1, D22S417, D10S526, vWA31, D5S592, and FES/FPS)	Match parent fibroblast line	Pass
Surface Antigen Expression of Stem Cell Markers	Immunostaining	> 80% expression of SSEA-4	Pass
Pluripotency	Illumina Array and PluriTest Software (www.pluritest.org)	Pluripotency Score greater than 20 and a Novelty Score less than 1.62	Pluripotency: 34.90 Novelty: 1.489

Post-Thaw Viability

One vial of distribution lot was thawed. Cultures were observed daily. Colonies were photographed when they first appeared, then 3 days later (Colonies must double in diameter within 5 days).

Day 3	161 µm
Day 6	389 µm

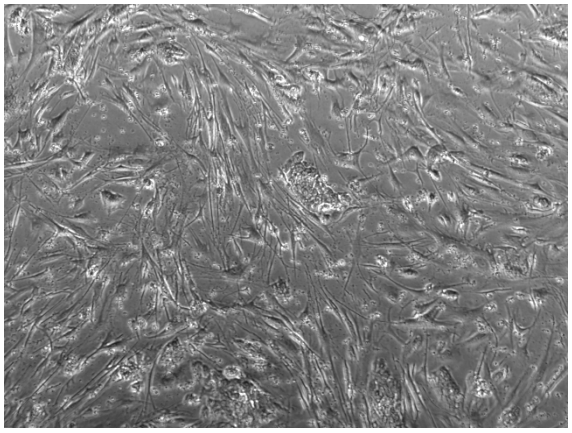

Figure 1A. Colony observed post thaw

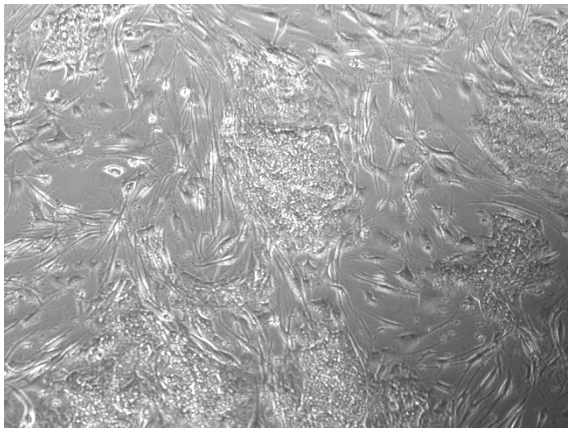

Figure 1B. Colony 3 days after first observation

Karyotype Analysis

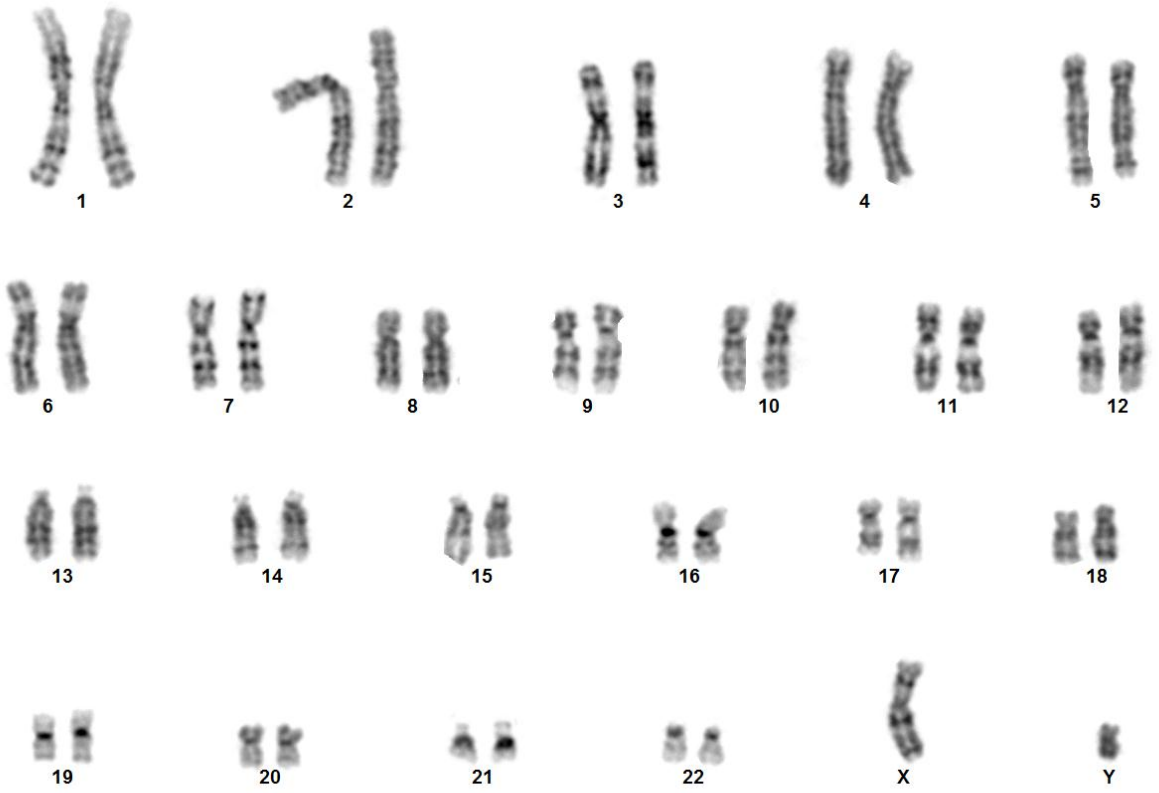

Figure 2: G-banded karyotype showing 46,X Y

Surface Antigen Expression of Stem Cell Markers

Undifferentiated cells are stained for the surface antigen, SSEA4. SSEA4 (stage specific embryonic antigen 4) is expressed on undifferentiated human pluripotent stem cells.

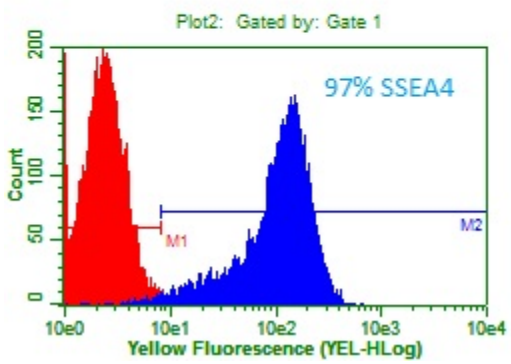

Figure 3: Representative histogram of SSEA-4 positive population. Histogram is an overlay of isotype control (red) and SSEA-4 positive population (blue).

Assessment of Pluripotency of a Cell Line

Cells are directed to differentiate to assess the pluripotency of the cell line. RNA is harvested and gene expression is analyzed by real-time PCR. Ct values are adjusted to the endogenous housekeeping gene , GAPDH. Relative gene expression is shown as fold difference in expression to that of the undifferentiated cells. Calculations were done using $2^{-\Delta\Delta CT}$ method.

Embryoid Body (EB) Formation Assay

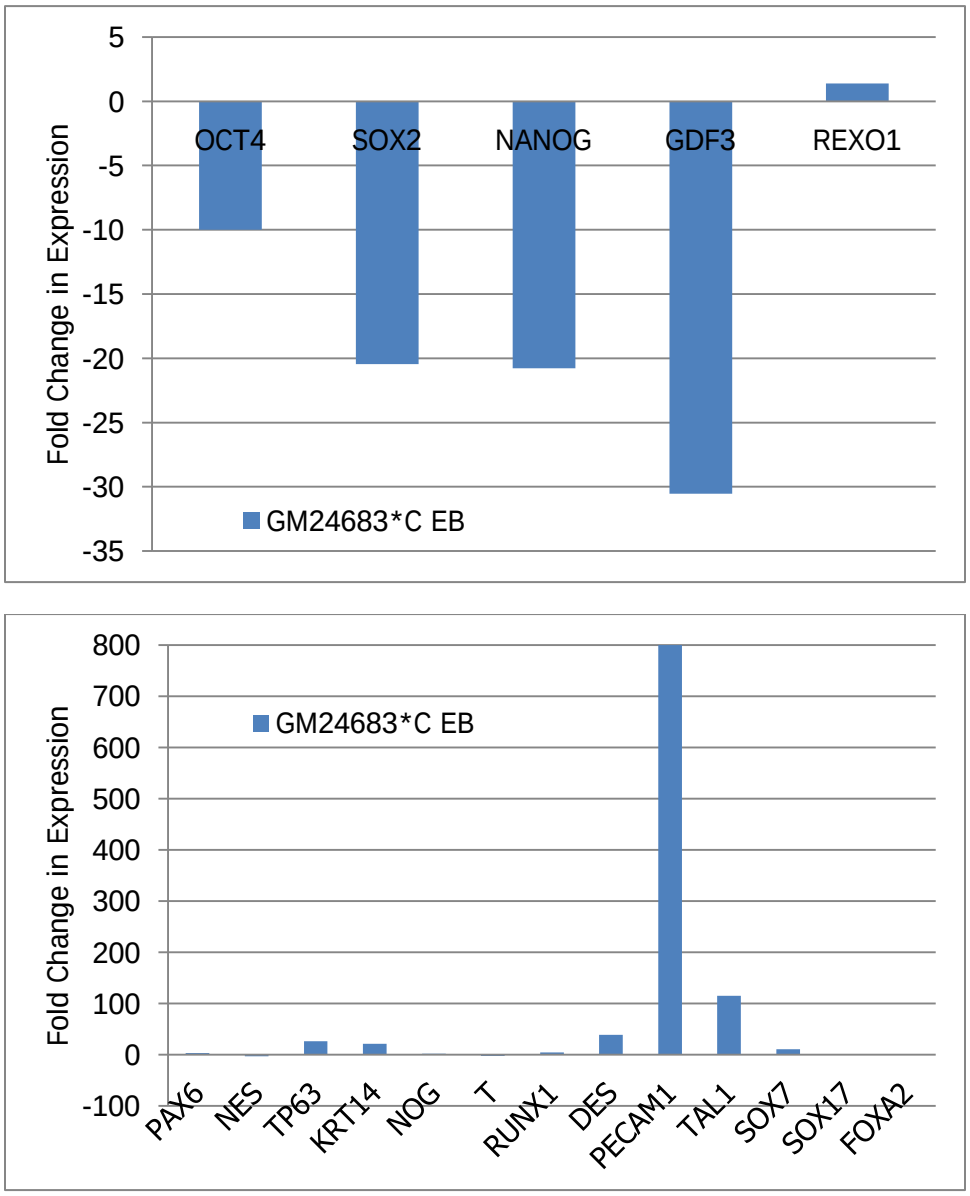

Figure 4. Gene expression following EB differentiation. Fold difference is shown relative to undifferentiated iPS cell line.

Pluripotency Markers

	OCT4	SOX2	NANOG	GDF3	REXO1
GM24683*C EB	-10	-20	-21	-31	1

Ectoderm

	PAX6	NES	TP63	KRT14	NOG
GM24683*C EB	3	-3	27	22	2

Mesoderm

	T	RUNX1	DES	PECAM1	TAL1
GM24683*C EB	-2	5	39	811	115

Endoderm

	SOX7	SOX17	FOXA2	AFP
GM24683*C	11	2	1	5086

Table 1. Fold difference values of gene expression of EB. Fold difference is relative to the undifferentiated cells. Ct values are normalized to that of GAPDH.

Stem Cell Biobank Technician
Date 10/28/14

☑ Pass
☐ Fail
☐ Other: _____

Shilpa Gandre-Babbe, PhD
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Date 10.30.2014