UMASS MEDICAL SCHOOL MASSACHUSETTS HUMAN STEM CELL BANK

CERTIFICATE OF ANALYSIS

Cell Line: **HUES-8**

Bank Code: **MA11.019** Lot #: **001** Passage #: **23**

Cell Type: **hESC** Genetic Disorder: **N/A**

Depositor: **Douglas Melton** Depositing Institution: **Harvard University**

Initial Cell Line Testing	Passage #	Expected Result	Page #	Status
Mycoplasma	23	Negative	2	Pass
Genotype	30	Same as developer data (if available)	5	Pass
Karyotype	22	**46,XY**	7	Pass
Immunocytochemistry	27	Pluripotency markers positive	9	Pass
Flow Cytometry	26	SSEA4 > 80%; SSEA1 < 10%	10	Pass
qRT-PCR	25	Pluripotency markers positive	11	Pass
EB Formation	25	Germ layers detected by RT-PCR	12	As reported
Lot Testing (Post Thaw)				
Mycoplasma	24	Negative	13	Pass
Genotype	26	Same as initial result	14	Pass
Karyotype	28	46,XY	16	Pass
Flow Cytometry	31	SSEA4 > 80%; SSEA1 < 10%	18	Pass
qRT-PCR	29	Pluripotency markers positive	19	Pass
Post Thaw Recovery	24	Harvest Day: ≥ 15 Colonies; ≤ 30% Differentiation	20	Pass
Comments / Additional Information			21	

All results in this Certificate of Analysis have been reviewed according to the criteria listed above.

☑ **PASS** Lot is ready for distribution

☐ **FAIL** Lot eliminated

Date:

Reviewed by: Date:

Title: Quality Assurance and Compliance Officer

Certified by: Date:

Title: Senior Director, UHSCBR

MASSACHUSETTS HUMAN STEM CELL BANK
UMASS MEDICAL SCHOOL

Line Name:	HUES-8	Passage #:	23
Bank Code:	MA11.019	Before Cryopreservation:	X
Lot #:	000	Post Thaw/Passage #:	
Analysis by:	external	Date:	see below

Initial Mycoplasma Contamination Analysis

Test	Laboratory	Passage #	Test Date	Result
qPCR	GlobalStem	23	8/10/2011	Negative

[X] PASS [ ] FAIL [ ] Culture Eliminated

Reviewer Signature:

Initials: MJS

Date: 8/17/2011

Title: Lab Director

GlobalStem Laboratory Services Report

Mycoplasma test using quantitative PCR

WE TAKE OUR CELLS SERIOUSLY™

Case #:	20110809-1c	**Submission Date:**	08/09/2011
		No. of Samples:	**4**
Test Date:	08/10/2011	**Test Method:**	**Quantitative PCR**

Sample ID	Description	Other	PCR Suppression*	Mycoplasma
Negative Control	-	-	N/A	Negative
Positive Control	-	-	None	Positive
Hues 8.00 MA11.019	Frozen cells	Human	**None**	**Negative**

*Suppression of internal control amplification due to factors in sample. This is likely to reduce the sensitivity of mycoplasma detection. Sample resubmission may be necessary if suppression occurs.

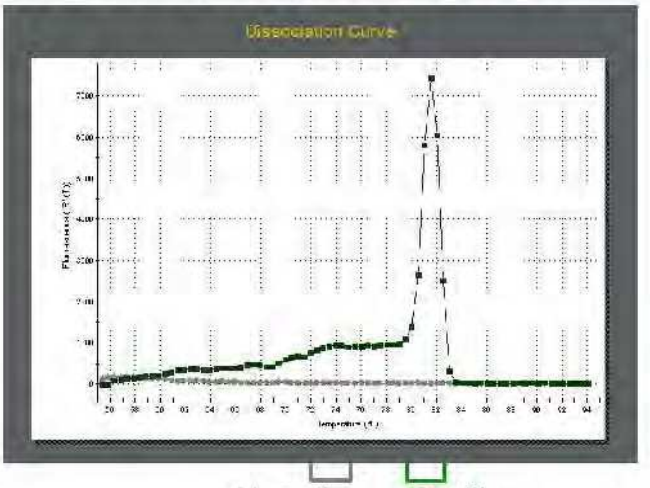
QC Scientist

08/11/2011
Date

GlobalStem Inc.
9430 Key West Avenue • Suite 130 • Rockville • Maryland • 20850 • USA
888-545-0238 • 301-545-0238 • Fax: 301-424-1989
www.globalstem.com • info@globalstem.com

Mycoplasma test report, 8-10-2011
qPCR

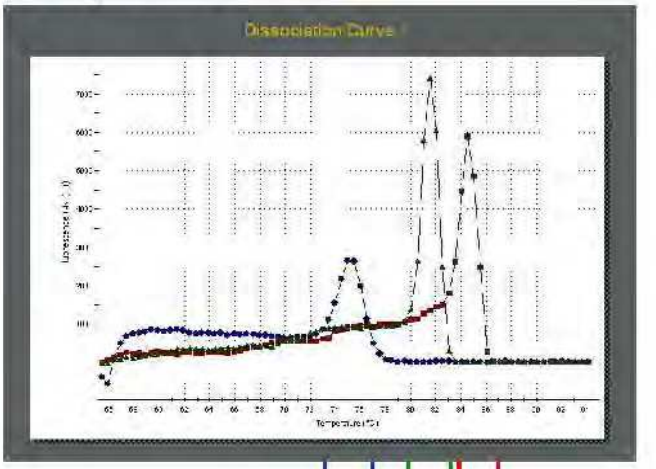
Positive and Negative Controls



Negative Control

Positive Control

Sample: MA11.019



Primer dimer

Positive control

Internal control

Well	Well Name	Assay	Ct (dR)	Tm Product 1 (-R'(T))	Result
F1	MA11.019	SYBR	No Ct	74.95	Negative
F3	MA11.019 + INT	SYBR	29.62	84.55	No Suppression
H1	Negative Control	SYBR	No Ct	57.38	negative
H3	Positive Control	SYBR	31.4	81.55	positive

Interpretation of results: Mycoplasma-negative samples show a predominant product with a Tm well below 80 C; this product is possibly a primer-dimer. The internal control band has a Tm of approximately 85°C, and amplification of the internal control means that the sample does not suppress the PCR reaction. Samples positive for mycoplasma, or the positive control amplicon, have a product with Tm of approximately 82°C.

Key:	Blue:	Customer sample
	Red:	Customer sample + Internal control
	Gray:	Negative control
	Green:	Positive control

MASSACHUSETTS HUMAN STEM CELL BANK
UMASS MEDICAL SCHOOL

Line Name:	HUES-8	Passage #:	30
Bank Code:	MA11.019	Before Cryopreservation:	X
Lot #:	000	Post Thaw/Passage #:	
Analysis by:	external	Date:	see below

Genotype (Fingerprint) Analysis

Laboratory	Report Date
Genetica DNA Laboratories	7/15/2011

X	Results match previously published genotype for this line
	Results match genotype from previous testing of this banked line
	There is no previous genotype available for this cell line

X	Results do not match any other genotypes in International Stem Cell Registry
	Results match genotype of a different cell line. Name (s) _____

	Results indicate that cell culture is not pure

X PASS FAIL Culture Eliminated

Reviewer Signature:

Initials: MJS

Date: 8/10/2011

Title: Lab Director

Cell Line Identity Profile

Genetica® DNA
Laboratories, Inc.
8740
Montgomery Rd
Cincinnati, OH
45236
USA
1-800-IDENTITY
1-800-433-6848
ph 513-985-9777
fax 513-985-9983
www.genetica.com

Submitted Sample MA11.019.000 (p30)

Date Received July 14, 2011
Test Number **93389-41**
3591
Report Date **July 15, 2011**

Genetic Systems Tested	Allele	
TPOX 2p23-2pter	8	11
D2S1338 2q35-37.1	17	
D3S1358 3p	15	19
FGA 4q28	20	25
D5S818 5q21-31	11	
CSF1PO 5q33.3-34	10	12
D7S820 7q	11	12
D8S1179 8	13	
THO1 11p15.5	7	9
vWA 12p12-pter	15	19
D13S317 13q22-31	12	14
D16S539 16q24-qter	9	12
D18S51 18q21.3	12	16
D19S433 19q12-13.1	13	15
D21S11 21q11.2-q21	28	29
Amelogenin X:p22.1-22.3 Y:p11.2	X	Y

Sample number 93389-41, labeled MA11.019.000 (p30) was submitted for identification. 15 autosomal short tandem repeat (STR) loci and the sex identity locus amelogenin were profiled using AmpFlSTR® Identifiler® PCR Amplification Kit. The electropherograms of analyzed data are attached.

Subscribed and sworn before me
on July 15, 2011

Martha E. Montenegro
Notary Public, State of Ohio
My Commission Expires: 10/15/2014

NOTARY PUBLIC THE GREAT SEAL OF THE STATE OF OHIO STATE OF OHIO

I, Elizabeth Panke, M.D., Ph.D., verify that the interpretation of results is correct as reported, and the above testing was conducted in accordance with the recommended guidelines for DNA testing set forth by AABB.

Elizabeth Panke, M.D., Ph.D.
Laboratory Director

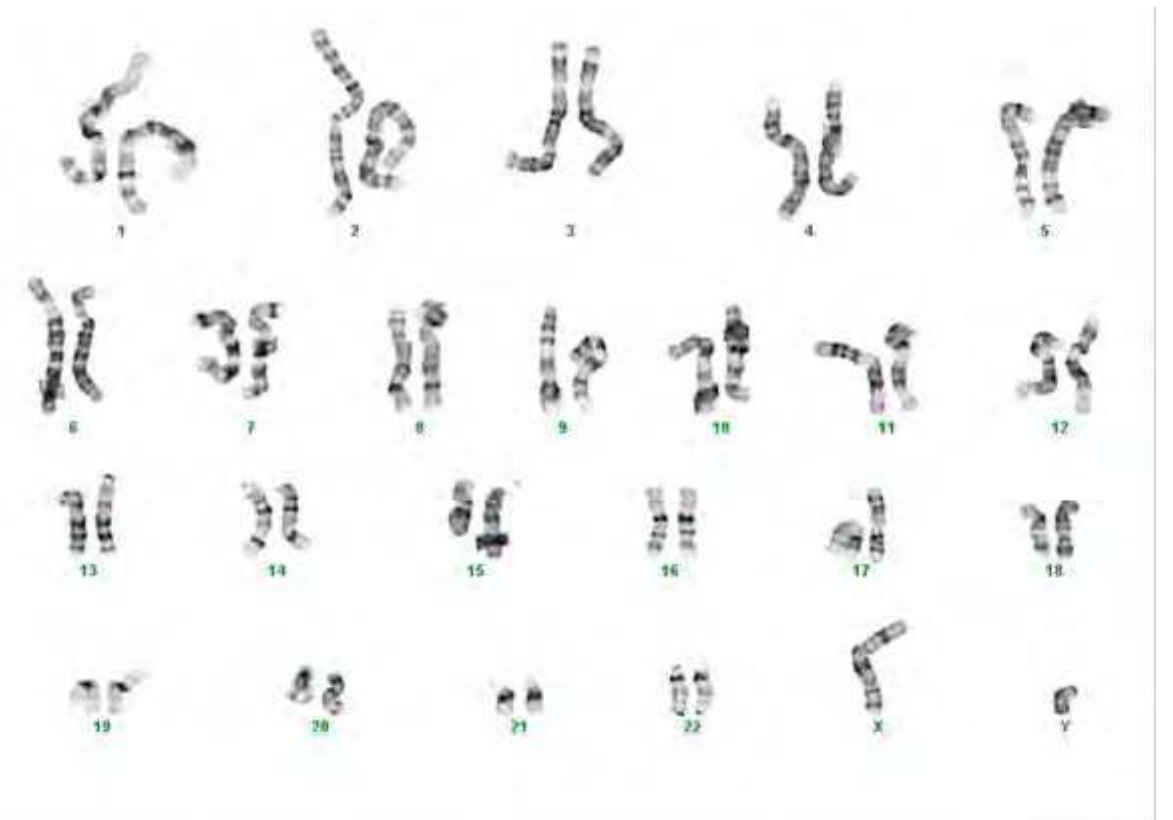
MASSACHUSETTS HUMAN STEM CELL BANK
UMASS MEDICAL SCHOOL

Line Name:	HUES-8	Passage #:	22
Bank Code:	MA11.019	Before Cryopreservation:	X
Lot #:	000	Post Thaw/Passage #:	
Analysis by:	external	Date:	see below

Karyotype Analysis

Laboratory	Report Date	Result
Cell Line Genetics	6/7/2011	46,XY

X	Karyotype is Normal
X	Non-clonal aberrations noted
	Karyotype is Abnormal



X	PASS		FAIL	Culture Eliminated

Reviewer Signature:

Initials: MJS

Date: 8/10/2011

Title: Lab Director

Cell Line GENETICS

Cell Line Characterization

Cell Line ID: HUES8/MA11.019.000 p22
Passage #: 22
Specimen Type: Human ESC Culture
Indication for Study: Routine Culture QC

Lab #: CLG-6763
PI: Meng-Jiao Shi
Contact Person: Chuanyou Zhang
Email: chuanyou.zhang@umassmed.edu
Address:
UMass Med School Stem Cell Bank
222 Maple Ave.
Shrewsbury, MA 01545

Test Code: 100
Account #: NA
PO #: NA

Date Received: 5/20/11
Date Reported: 6/7/11
Time in Culture: 3 days

Additional copies sent to:

Banding Technique: GTL **Band Resolution:** Good
Metaphases Counted: 20 Analyzed: 7 Karyotyped: 2

RESULTS: 46,XY[19] Apparently NORMAL Male Human Karyotype

Non-clonal Aberrations: 45,XY,-20 (one cell)

INTERPRETATION:

Cytogenetic analysis was performed on twenty G-banded metaphase cells from human cell line HUES8/MA11.019.000 p22. Nineteen cells demonstrated an apparently normal male karyotype, while one cell demonstrated a non-clonal chromosome aberration (listed above) which is most likely a technical artifact. No abnormal cells with trisomy 12 and/or 17 were detected.

Lorraine Faxon Meisner, Ph.D., FACMG

Julie A. Johnson, M.S., CLSp(CG)

MASSACHUSETTS HUMAN STEM CELL BANK
UMASS MEDICAL SCHOOL

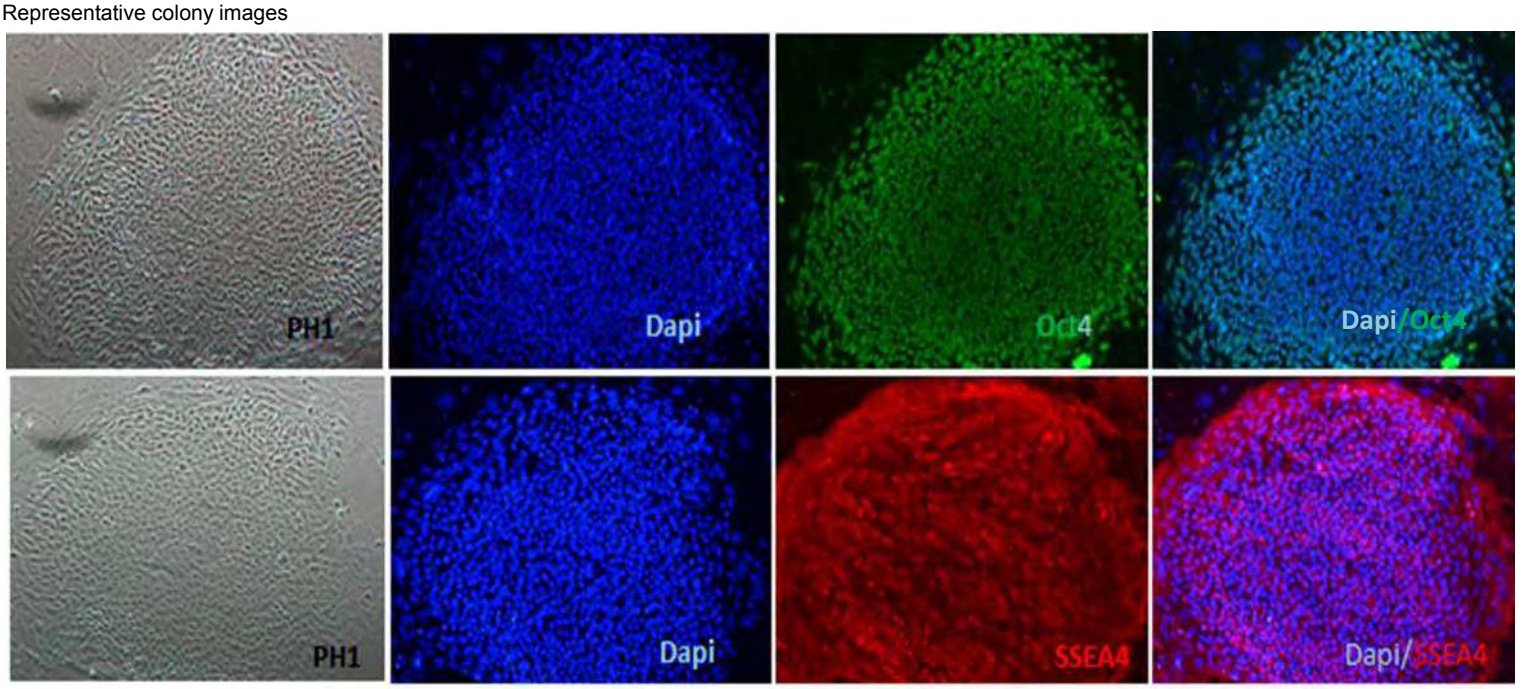
Line Name:	HUES-8	Passage #:	27
Bank Code:	MA11.019	Before Cryopreservation:	X
Lot #:	000	Post Thaw/Passage #:	
Analysis by:	CZ	Date:	6/22/2011

Immunocytochemical Analysis

Marker	Localization	Acceptance Criteria	# Positive Colonies*/ # Analyzed
Oct 3/4	Nuclear	16/20	20/20
SSEA4	Cell Surface	16/20	20/20

** Colonies are considered positive if the majority of cells stain with the marker by visual estimation.*

Representative colony images



[X] PASS [ ] FAIL [ ] Culture Eliminated

Reviewer Signature:

Initials: MJS

Date: 8/10/2011

Title: Lab Director

MASSACHUSETTS HUMAN STEM CELL BANK
UMASS MEDICAL SCHOOL

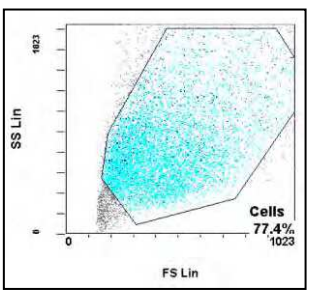
Line Name:	HUES-8	Passage #:	26
Bank Code:	MA11.019	Before Cryopreservation:	X
Lot #:	000	Post Thaw/Passage #:	
Analysis by:	MG	Date:	6/13/2011

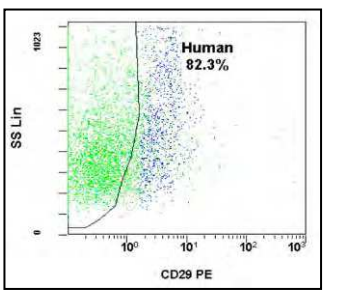
Flow Cytometry Analysis

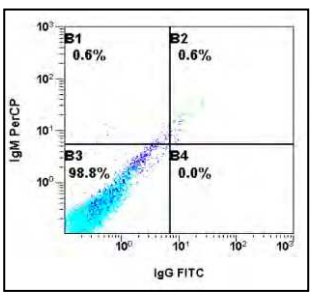
Marker	Target Population	% Positive	Acceptance Criteria (%)	Pass/Fail
SSEA4+	Pluripotent Cells	97.64%	> 80%	Pass
SSEA1+	Differentiating Cells	0.22%	< 10%	Pass

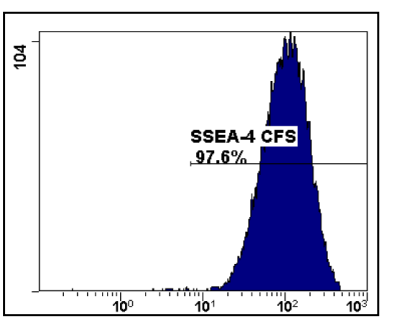
a. Sample picture

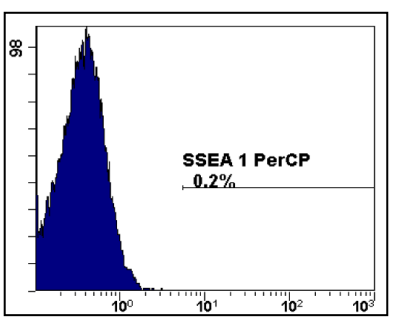
b. Flow cytometry data











[X] PASS [ ] FAIL [ ] Culture Eliminated

Reviewer Signature:

Initials: MJS

Date: 8/10/2011

Title: Lab Director

MASSACHUSETTS HUMAN STEM CELL BANK
UMASS MEDICAL SCHOOL

Line Name:	HUES-8	Passage #:	25
Bank Code:	MA11.019	Before Cryopreservation:	X
Lot #:	000	Post Thaw/Passage #:	
Analysis by:	LJ	Date:	6/8/2011

Analysis of Cells in Culture by RT Real Time-PCR

Note: Undifferentiated cells in culture should express markers of purlipotency, but not markers of differentiated cell types.

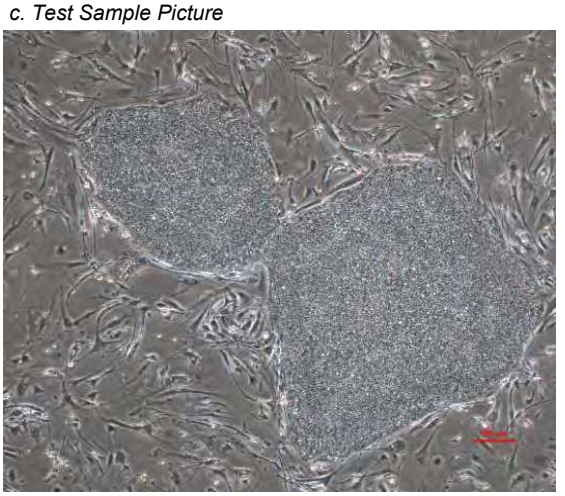
a. CT values from RT-real time PCR

	Pluripotency						Endoderm	Mesoderm	Ectoderm	Constitutive
	DNMT3B	*POU5F1*	*NANOG*	*ZFP42*	*TDGF1*	*TERT*	*AFP*	*GATA4*	*PAX6*	*ATCB*
H9 ES	23.42	22.265	24.805	25.695	23.085	30.475	39.475	31.695	34.745	18.95
MA11.019.000	23.965	23.635	26.52	27.605	24.54	31.31	33.86	33.135	33.925	20.37
H9 EB	29.14	30.81	32.33	30.75	31.435	32.63	20.61	27.81	29.085	20.23

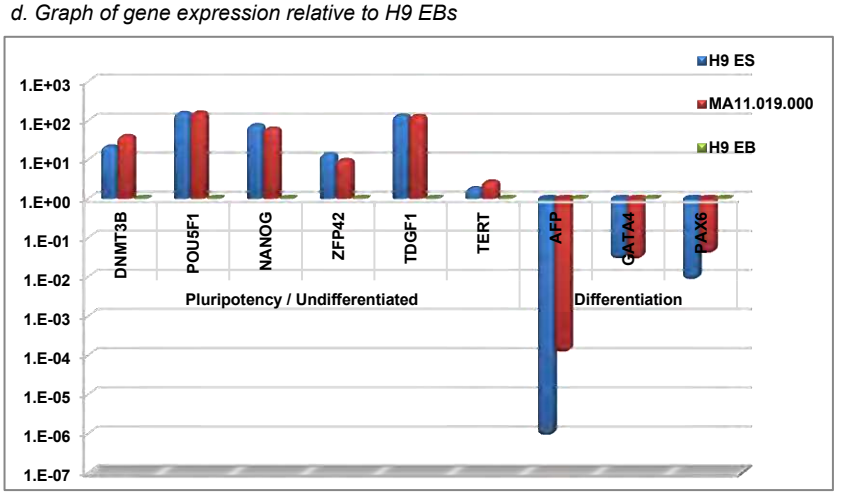
b. Fold-change relative to H9 EBs

	Pluripotency / Undifferentiated						Differentiation		
	DNMT3B	*POU5F1*	*NANOG*	*ZFP42*	*TDGF1*	*TERT*	*AFP*	*GATA4*	*PAX6*
H9 ES	21.71	153.81	75.85	13.69	134.36	1.83	8.62E-07	0.03	0.01
MA11.019.000	39.81	159.23	61.82	9.75	131.14	2.75	1.13E-04	0.03	0.04
H9 EB	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00

c. Test Sample Picture



d. Graph of gene expression relative to H9 EBs



X	PASS		FAIL			Culture Eliminated

Reviewer Signature:

Initials: MJS

Date: 8/10/2011

Title: Lab Director

MASSACHUSETTS HUMAN STEM CELL BANK
UMASS MEDICAL SCHOOL

Line Name:	HUES-8	Passage #:	25
Bank Code:	MA11.019	Before Cryopreservation:	X
Lot #:	000	Post Thaw/Passage #:	
Analysis by:	LJ	Date:	6/22/2011

Analysis of Embryoid Bodies by RT Real Time PCR

Note: Embryoid bodies developed from pluripotent cells should express markers of cell types representing all three germ layers, but not markers of pluripotent cells. Due to the limited number of markers assayed, lack of detectable expression of specific markers does not rule out germ layer formation.

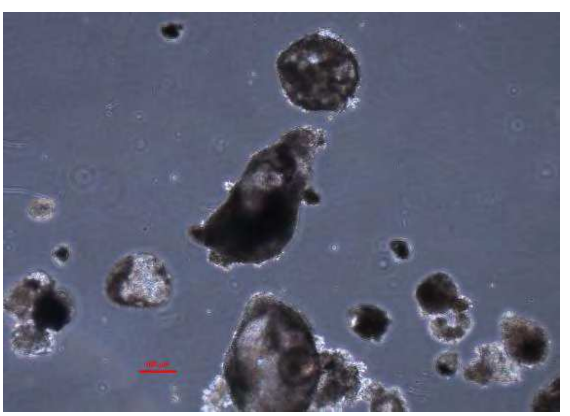
a. CT values from RT-real time PCR

	Pluripotency	Endoderm		Mesoderm				Ectoderm		Constitutive
	POU5F1	*AFP*	*GATA6*	*GATA4*	*RUNX1*	*PECAM*	*PTPRC*	*PAX6*	*NCAM1*	*ATCB*
H9 ES	22.27	39.48	31.92	31.70	32.98	37.07	No Ct	34.75	No Ct	18.95
MA11.019 EB	28.57	21.58	27.93	28.38	29.61	30.37	34.83	30.43	37.13	20.66
H9 EB	30.81	20.61	27.72	27.81	29.85	29.53	34.90	29.09	35.74	20.23

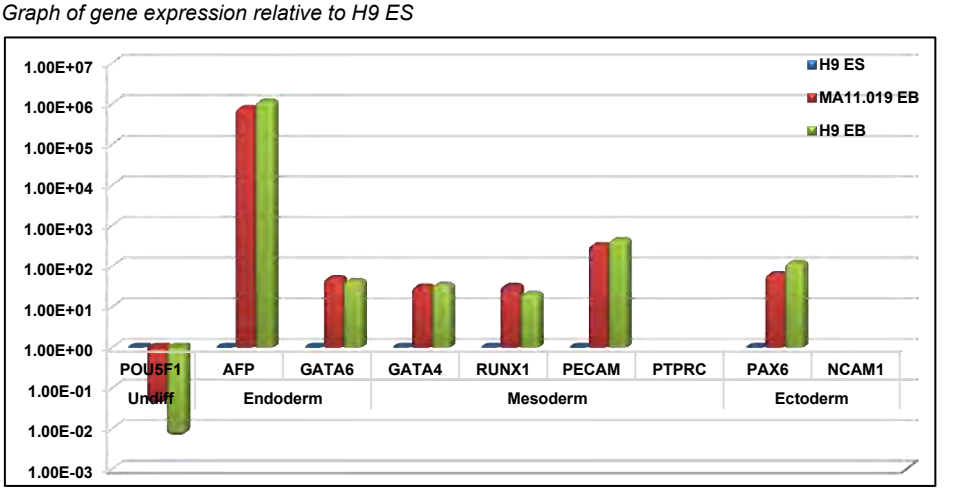
b. Fold-change relative to H9 ES

	Undiff	Endoderm		Mesoderm				Ectoderm	
	POU5F1	*AFP*	*GATA6*	*GATA4*	*RUNX1*	*PECAM*	*PTPRC*	*PAX6*	*NCAM1*
H9 ES	1.00	1.00	1.00	1.00	1.00	1.00		1.00	
MA11.019 EB	0.04	797430.91	51.80	32.45	33.71	338.97		65.12	
H9 EB	0.01	1159442.63	44.48	35.88	21.33	450.38		122.79	

c. Test Sample Picture EB day 14



d. Graph of gene expression relative to H9 ES



X	PASS		FAIL			Culture Eliminated

Reviewer Signature:

Initials: MJS

Date: 8/10/2011

Title: Lab Director

MASSACHUSETTS HUMAN STEM CELL BANK
UMASS MEDICAL SCHOOL

Line Name:	HUES-8	Passage #:	24
Bank Code:	MA11.019	Before Cryopreservation:	
Lot #:	001	Post Thaw/Passage #:	X / 1
Analysis by:	CZ	Date:	6/24/2011

MycoAlert Mycoplasma Data Log Sheet

Results Interpretation:

The ratio of Reading B to Reading A is used to determine whether a cell culture is contaminated by mycoplasma. Cells that are infected with mycoplasma will routinely produce ratios greater than 1. Results have been interpreted according to the table below:

B/A Ratio	Interpretation and Conclusions	
< 1	Negative	Sample uninfected
> 1.5	Positive	Sample infected
1 - 1.5	Borderline	To be retested with this method or another method

Results:

Sample ID	A Reading	B Reading	B/A Ratio	Conclusion
Positive Control	406	5131	12.64	Valid
Positive Control Sample 2	381	5876	15.42	Valid
Negative Control	521	142	0.273	Valid
Negative Control Sample 2	409	94	0.203	Valid
Sample 1	250	107	0.43	Negative
Sample 2	302	192	0.64	Negative

[X] PASS [ ] FAIL [ ] Culture Eliminated

Reviewer Signature:

Initials: MJS

Date: 8/10/2011

Title: Lab Director

MASSACHUSETTS HUMAN STEM CELL BANK
UMASS MEDICAL SCHOOL

Line Name:	HUES-8	Passage #:	26
Bank Code:	MA11.019	Before Cryopreservation:	
Lot #:	001	Post Thaw/Passage #:	X / 3
Analysis by:	external	Date:	see below

Genotype (Fingerprint) Analysis

Laboratory	Report Date
Genetica DNA Laboratories	7/15/2011

X	Results match previously published genotype for this line
X	Results match genotype from previous testing of this banked line
	There is no previous genotype available for this cell line
X	Results do not match any other genotypes in International Stem Cell Registry
	Results match genotype of a different cell line. Name (s) _____
	Results indicate that cell culture is not pure

X PASS FAIL Culture Eliminated

Reviewer Signature:

Initials: MJS

Date: 8/10/2011

Title: Lab Director

Cell Line Identity Profile

Genetica® DNA
Laboratories, Inc.
8 7 4 0
Montgomery Rd
Cincinnati, OH
4 5 2 3 6
U S A
1-800-IDENTITY
1-800-433-6848
ph 513-985-9777
fax 513-985-9983
www.genetica.com

Submitted Sample MA11.019.001 (p26)

Date Received July 14, 2011
Test Number **93391-41**
3591
Report Date **July 15, 2011**

Genetic Systems Tested	Allele	
TPOX 2p23-2pter	8	11
D2S1338 2q35-37.1	17	
D3S1358 3p	15	19
FGA 4q28	20	25
D5S818 5q21-31	11	
CSF1PO 5q33.3-34	10	12
D7S820 7q	11	12
D8S1179 8	13	
THO1 11p15.5	7	9
vWA 12p12-pter	15	19
D13S317 13q22-31	12	14
D16S539 16q24-qter	9	12
D18S51 18q21.3	12	16
D19S433 19q12-13.1	13	15
D21S11 21q11.2-q21	28	29
Amelogenin X:p22.1-22.3 Y:p11.2	X	Y

Sample number 93391-41, labeled MA11.019.001 (p26) was submitted for identification. 15 autosomal short tandem repeat (STR) loci and the sex identity locus amelogenin were profiled using AmpFlSTR® Identifiler® PCR Amplification Kit. The electropherograms of analyzed data are attached.

Subscribed and sworn before me
on July 15, 2011

Martha E. Montenegro
Notary Public, State of Ohio
My Commission Expires: 10/15/2014

NOTARY PUBLIC THE GREAT SEAL OF THE STATE OF OHIO STATE OF OHIO

I, Elizabeth Panke, M.D., Ph.D., verify that the interpretation of results is correct as reported, and the above testing was conducted in accordance with the recommended guidelines for DNA testing set forth by AABB.

Elizabeth Panke, M.D., Ph.D.
Laboratory Director

Genetica® DNA Laboratories, Inc. is accredited by The New York State Department of Health (NYSDOH), Forensic Quality Services-International (FQS-I, ISO/IEC 17025) and the College of American Pathologists (CAP) for DNA family relationship and identity testing for DNA family relationship and identity testing. Genetica® DNA Laboratories, Inc. headquarters are located in the United States of America.

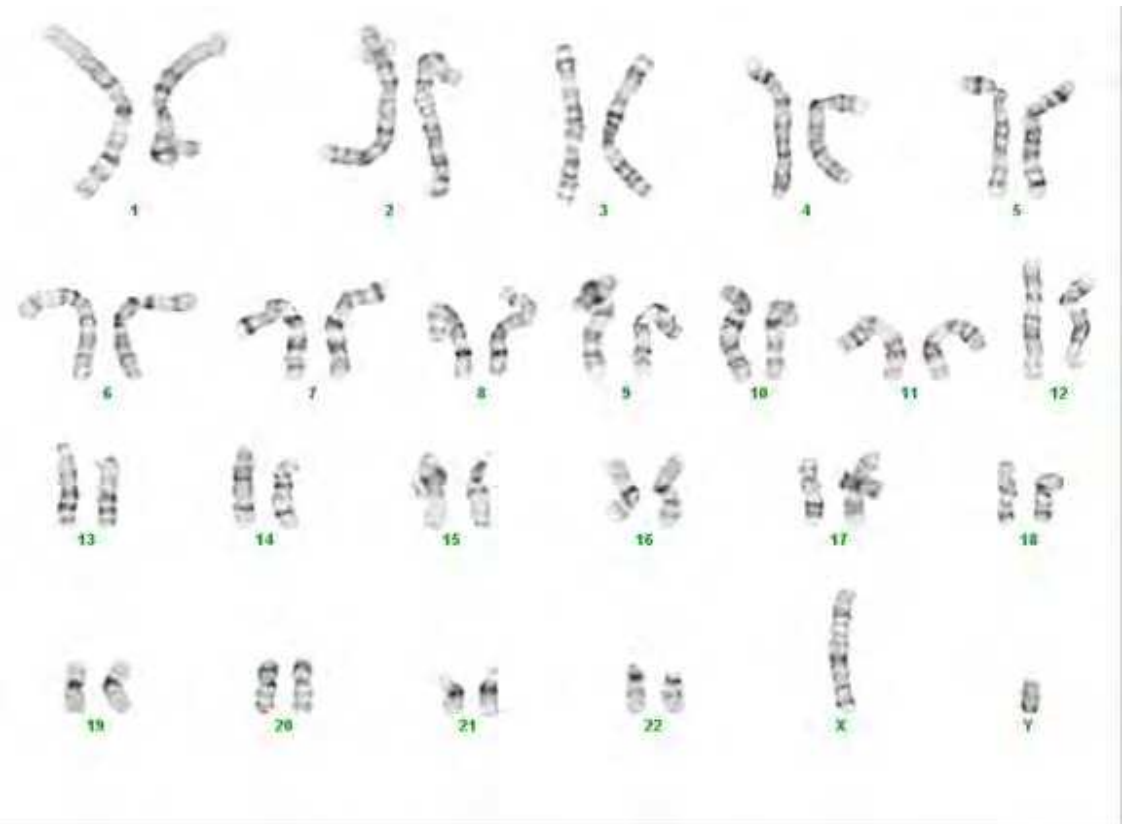
MASSACHUSETTS HUMAN STEM CELL BANK
UMASS MEDICAL SCHOOL

Line Name:	HUES-8	Passage #:	28
Bank Code:	MA11.019	Before Cryopreservation:	
Lot #:	001	Post Thaw/Passage #:	X / 5
Analysis by:	external	Date:	see below

Karyotype Analysis

Laboratory	Report Date	Result
Cell Line Genetics	8/2/2011	46,XY

X	Karyotype is Normal
X	Non-clonal aberrations noted
	Karyotype is Abnormal



X	PASS		FAIL		Culture Eliminated

Reviewer Signature:

Initials: MJS

Date: 8/10/2011

Title: Lab Director

Cell Line GENETICS

Cell Line Characterization

Cell Line ID: MA11.019.001 HUES-8 p28
Passage #: 28
Specimen Type: Human ESC Culture
Indication for Study: Routine Culture QC

Test Code: 100
Account #: NA
PO #: NA

Date Received: 7/22/11
Date Reported: 8/2/11
Time in Culture: 3 days

Lab #: CLG-7340
PI: Meng-jiao Shi
Contact Person: Lan Ji
Email: lan.ji@umassmed.edu
Address:
UMass Human Stem Cell Bank
222 Maple Ave.
Shrewsbury, MA 01545

Additional copies sent to:

Banding Technique: GTL **Band Resolution:** Good
Metaphases Counted: 20 Analyzed: 8 Karyotyped: 2

RESULTS: 46,XY[17] Apparently NORMAL Male Human Karyotype

Non-clonal Aberrations: 45,XY,-17
45,XY,-22
45,XY,-11

INTERPRETATION:

Cytogenetic analysis was performed on twenty G-banded metaphase cells from human cell line MA11.019.001 HUES-8 p28. Seventeen cells demonstrated an apparently normal male karyotype, while three cells demonstrated non-clonal chromosome aberrations (listed above) which are most likely technical artifacts. No abnormal cells with trisomy 12 and/or 17 were detected.

Lorraine Faxon Meisner, Ph.D., FACMG

Julie A. Johnson, M.S., CLSp(CG)

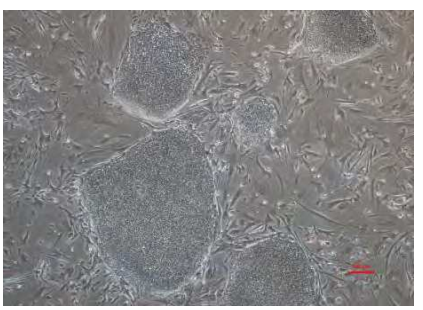
MASSACHUSETTS HUMAN STEM CELL BANK
UMASS MEDICAL SCHOOL

Line Name:	HUES-8	Passage #:	31
Bank Code:	MA11.019	Before Cryopreservation:	
Lot #:	001	Post Thaw/Passage #:	X / 8
Analysis by:	LJ	Date:	8/5/2011

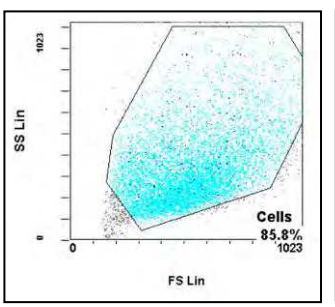
Flow Cytometry Analysis

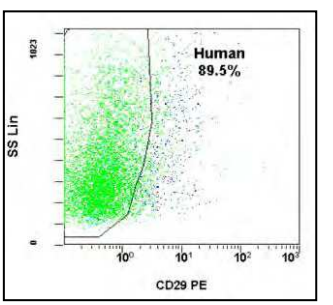
Marker	Target Population	% Positive	Acceptance Criteria (%)	Pass/Fail
SSEA4+	Pluripotent Cells	92.90%	> 80%	Pass
SSEA1+	Differentiating Cells	2.68%	< 10%	Pass

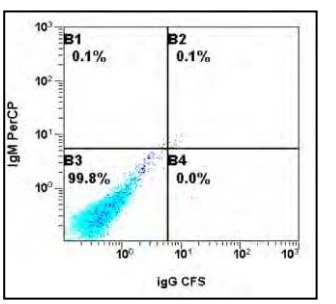
a. Sample picture

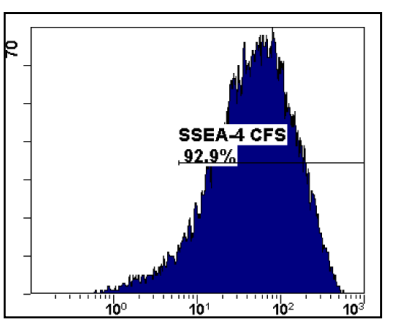


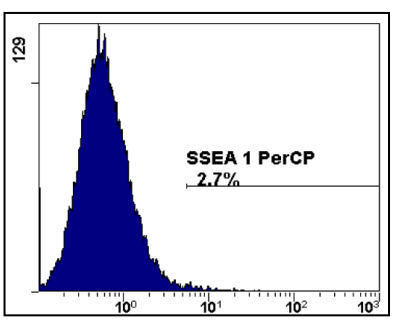
B. Flow cytometry data











[X] PASS
See Comments

[ ] FAIL

[ ] Culture Eliminated

Reviewer Signature:

Initials: MJS

Date: 8/10/2011

Title: Lab Director

MASSACHUSETTS HUMAN STEM CELL BANK
UMASS MEDICAL SCHOOL

Line Name:	HUES-8	Passage #:	29
Bank Code:	MA11.019	Before Cryopreservation:	
Lot #:	001	Post Thaw/Passage #:	X / 6
Analysis by:		Date:	7/27/2011

Analysis of Cells in Culture by RT Real Time-PCR

Note: Undifferentiated cells in culture should express markers of purlipotency, but not markers of differentiated cell types.

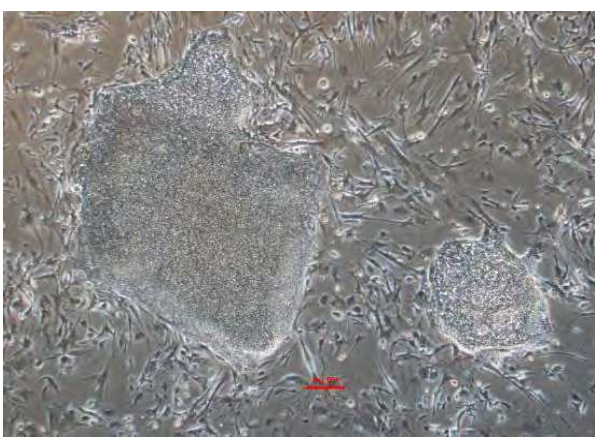
a. CT values from RT-real time PCR

	Pluripotency						Endoderm	Mesoderm	Ectoderm	Constitutive
	DNMT3B	*POU5F1*	*NANOG*	*ZFP42*	*TDGF1*	*TERT*	*AFP*	*GATA4*	*PAX6*	*ATCB*
H9 ES	23.67	22.715	25.215	26.335	23.755	30.65	38.855	32.355	34.745	19.295
MA11.019.001	24.53	23.765	26.52	28.03	24.885	32.4	35.05	31.17	34.14	20.56
H9 EB	29.255	30.7	32.62	31.025	31.965	32.895	20.74	28.38	29.165	20.41

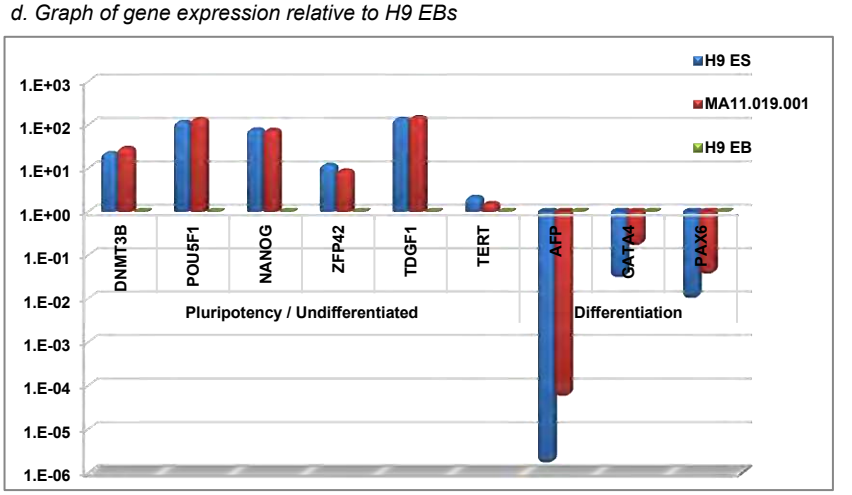
b. Fold-change relative to H9 EBs

	Pluripotency / Undifferentiated						Differentiation		
	DNMT3B	*POU5F1*	*NANOG*	*ZFP42*	*TDGF1*	*TERT*	*AFP*	*GATA4*	*PAX6*
H9 ES	22.16	116.97	78.25	11.92	136.71	2.19	1.63E-06	0.03	0.01
MA11.019.001	29.34	135.77	76.11	8.85	150.12	1.56	5.46E-05	0.16	0.04
H9 EB	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00

c. Test Sample Picture



d. Graph of gene expression relative to H9 EBs



X	PASS		FAIL		Culture Eliminated

Reviewer Signature:

Initials: MJS

Date: 8/10/2011

Title: Lab Director

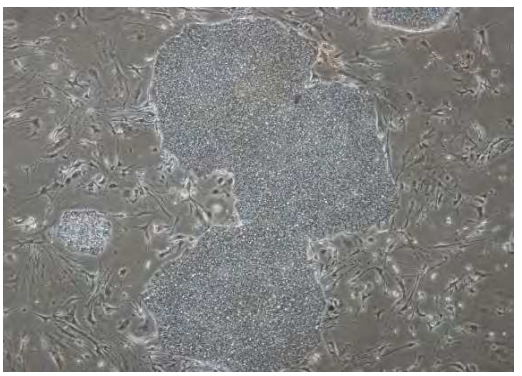
MASSACHUSETTS HUMAN STEM CELL BANK
UMASS MEDICAL SCHOOL

Line Name:	HUES-8	Passage #:	24
Bank Code:	MA11.019	Before Cryopreservation:	
Lot #:	001	Post Thaw/Passage #:	X / 1
Analysis by:	CZ	Date:	6/28/2011

Post Thaw Viability of Cryopreserved Cells

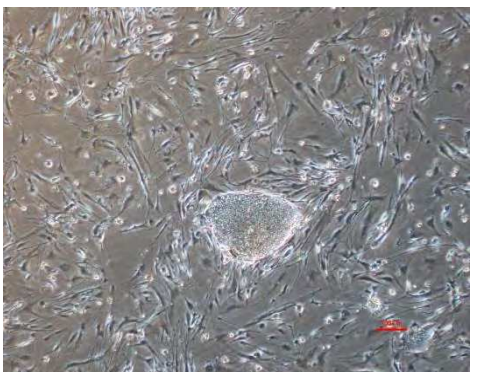
Harvest or Day Culture	Number of Colonies	% Estimated Differentiated Cells
Expected	***≥ 15***	***≤ 30%***
Actual	20	<5%
Pass/Fail	Pass	Pass

Prior to Cryopreservation



Post Thaw Day 2 Culture

Post Thaw Day 5 Culture



Comments:

X	PASS		FAIL		Culture Eliminated

Reviewer Signature:

Initials: MJS

Date: 8/10/2011

Title: Lab Director

Comments and Additional information

Flow cytometry was performed on Lot 001 of MA11.019 (HUES-8) at passage 29, 30 and 31. The lot passed acceptance criteria for the passage 30 and 31 testing, but failed testing for passage 29 due to the percentage of SSEA4 positive cells falling below the criteria of 80%. The percentages for each assay were:

Passage 29 – 76%
Passage 30 – 89%
Passage 31 – 93% (shown in this CofA, page 18)

While some instability in SSEA-4 expression can occur, possibly due to culture conditions, the SSEA-4 negative population seemed to be transient and the level of SSEA-1 was not above the acceptance criteria of 10% in any of the assays. In addition, significant changes in pluripotency marker expression by qRT-PCR were not observed (see page 19).

Reviewer Signature:

Initials: KPS
Date: 8/18/2011

Title: Lab Director