# H1 Oct4-EGFP

GFP expression linked directly to the Oct4 promoter. A GFP Neomycin construct was added to the H1 cell line via homologous recombination and targeted to the POUF1 gene. Cells will express GFP when Oct4 is expressed (e.g. undifferentiated state). Oct4/GFP positive cells are selectable using Neomycin. These cells tend to differentiate more frequently and are best grown in slightly sparser conditions. It is not unusual to have to manually remove differentiation at each passage in the absence of selection.

| | |
|---|---|
| **Alias** | WA01 |
| **Cell Type** | Modified Human ES |
| **Culture Platform / Protocol** | Feeder Independent - TeSR1 Medium |
| **Disease Model** | N/A |
| **Genetic Modification Keyword** | GFP (mediated) |
| **NIH Registry Approved** | Yes |
| **Karyotype** | 46,XY |
| **Blood Type** | O+ |
| **Publication** | Publication |
| **Provider** | University of Wisconsin (Thomson) |

| QTY | PRICE |
|---|---|
| 1 - 2 | $1,250.00/vial |
| 3 - 4 | $1,125.00/vial |
| 5+ | $1,062.50/vial |

Quantity 1

ADD TO CART

☐ Check if you received this cell line previously Why?

**Related Products**

H9 hOct4-pGZ
H9 hNanog-pGZ
WA01 (mTeSR™1/Matrigel™ Platform)

## Current Lot Information

| Lot Number | Lot Description | Passage Number | Banked By | Product Information and Testing |
|---|---|---|---|---|
| DL-02 | Modified ES Cell; Oct4 mediated GFP | 69(10) | WiCell | PDF |

## Historical Lot Information

| Lot Number | Lot Description | Banked By | Product Information and Testing |
|---|---|---|---|
| MCB-01 | Modified ES Cell; Oct4 mediated GFP | WiCell | PDF |

# Certificate of Analysis - Amended

| | | |
|---|---|---|
| Product Description | WA01 Oct4-eGFP Knock In | |
| Cell Line Provider | University of Wisconsin- Laboratory of Dr. James Thomson | |
| Lot Number | WA01(Oct4KI)-MCB-1 | |
| Date Vialed | 04-October-2008 | |
| Passage Number | p60 | |
| Culture Platform | Feeder Independent | |
| | Media[1]: TeSR | Matrix: Matrigel |

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

| Test Description | Test Provider | Test Method | Test Specification | Result |
|---|---|---|---|---|
| Post-Thaw Viable Cell Recovery | WiCell | SOP-CH-305 | ≥ 15 Undifferentiated Colonies, ≤ 30% Differentiation | Pass |
| Identity by STR | UW Molecular Diagnostics Laboratory | PowerPlex 1.2 System by Promega | Consistent with known profile | Pass |
| Sterility - Direct transfer method | Apptec | 30744 | Negative | Pass |
| Mycoplasma | Bionique | M250 | No contamination detected | Pass |
| Karyotype by G-banding | WiCell | SOP-CH-003 | Normal karyotype | Pass |

[1] These cells were cultured in the presence of G418 (Geneticin, Invitrogen catalog 11811) at a concentration of 50µg/ml. Not used for thawing, passaging, or freezing.

Appropriate biosafety precautions should be followed when working with these cells. The end user is responsible for ensuring that the cells are handled and stored in an appropriate manner. WiCell is not responsible for damages or injuries that may result from the use of these cells.

Amendment(s):

| Reason for Amendment | Date |
|---|---|
| CoA updated to include copyright information. | See signature |
| CoA updated for clarification of test specifications, added media foornote, and removed text regarding technical services | 05-October-2010 |
| CoA updated for format changes, clarification of test specifications, test method, addition of test provider, culture platform, and electronic signature, and reference to WiCell instead of the NSCB | 19-August-2010 |
| Original CoA | 22-April-2009 |

| Date of Lot Release | Quality Assurance Approval |
|---|---|
| 22-April-2009 | 12/30/2013<br>X AMC<br>AMC<br>Quality Assurance<br>Signed by: [redacted] |

UW Health
University of Wisconsin Hospital and Clinics

**Histocompatibility/Molecular Diagnostics Laboratory**

# Short Tandem Repeat Analysis*

**Sample Report: 6618-STR**
WA01(Oct4KI)-MCB-1

UW HLA#: 59824

Sample Date: 11/06/08
Received Date: 11/07/08

Requestor: WiCell Research Institute

Test Date: 11/07/08

File Name: 081107

Report Date: 11/16/08

Sample Name: (label on tube) **6618-STR**

**Description:** DNA Extracted by WiCell
251.81 ng/µL; 260/280 = 1.91

| Locus | Repeat # | STR Genotype |
|---|---|---|
| D16S539 | 5, 8-15 | 9,13 |
| D7S820 | 6-14 | 8,12 |
| D13S317 | 7-15 | 8,11 |
| D5S818 | 7-15 | 9,11 |
| CSF1PO | 6-15 | 12,13 |
| TPOX | 6-13 | 8,11 |
| Amelogenin | NA | X,Y |
| TH01 | 5-11 | 9.3,9.3 |
| vWA | 11, 13-21 | 15,17 |

**Comments: Based on the 6618-STR DNA submitted by WI Cell dated 11/06/08 and received on 11/07/08, this sample (UW HLA# 59824) matches exactly the STR profile of the human stem cell line H1 comprising 15 allelic polymorphisms across the 8 STR loci analyzed. No STR polymorphisms other than those corresponding to the human H1 stem cell line were detected and the concentration of DNA required to achieve an acceptable STR genotype (signal/ noise) was equivalent to that required for the standard procedure (~1 ng/amplification reaction) from human genomic DNA. This result suggest that the 6618-STR DNA sample submitted corresponds to the H1 stem cell line and was not contaminated with any other human stem cells or a significant amount of mouse feeder layer cells. Sensitivity limits for detection of STR polymorphisms unique to either this or other human stem cell lines is ~5%. These results were communicated via phone to the Cytogenetics laboratory of the WiCell Research Institute on Monday, October 27, 2008. A preliminary copy of this report was issued via electronic mail to the WI Cell Research Institute on Monday, November 17, 2008.**

* Testing to assess engraftment following bone marrow transplantation was accomplished by analysis of human genetic polymorphisms at STR loci. This methodology has not yet been approved by the FDA and is for investigational use only.

Test Facility:



WuXi AppTec



WiCell Research Institute

December 16, 2008
P.O. #:

# STERILITY TEST REPORT

**Sample Information:** hES Cells
3: WA01 (Oct4KI)-MCB-1

**Date Received:** November 25, 2008
**Date in Test:** December 01, 2008
**Date Completed:** December 15, 2008

**Test Information:** Test Codes: 30744, 30744A
Immersion, USP / 21 CFR 610.12
Procedure #: BS210WCR.201 (Modified: Alternate media used.)

| TEST PARAMETERS | PRODUCT | |
|---|---|---|
| **Approximate Volume Tested** | 0.5 mL | 0.5 mL |
| **Number Tested** | 2 | 2 |
| **Type of Media** | SCD | FTM-T-L-S |
| **Media Volume** | 400 mL | 400 mL |
| **Incubation Period** | 14 Days | 14 Days |
| **Incubation Temperature** | 20 °C to 25 °C | 30 °C to 35 °C |
| **RESULTS** | 2 NEGATIVE | 2 NEGATIVE |

**QA Reviewed:** Page 1 Signed

**Reviewed:** Page 1 Signed

*Testing conducted in accordance with current Good Manufacturing Practices.*

Mycoplasma Testing Services

Safe Cells

BIONIQUE TESTING LABORATORIES, INC

APPENDIX IV



Document#: DCF3013D
Edition#: 10
Effective Date: 07/15/2003
Title: **M-250 FINAL REPORT SHEET**

## M-250 FINAL REPORT

Direct Specimen Culture
Procedure 3008, 3011, 3013

**TO: Wicell QA**

BTL SAMPLE ID#: **55226** P.O.#: DATE REC'D: **11/04/2008**

TEST/CONTROL ARTICLE:

**6618 WA01 (OCT4 KI) MCB1**

LOT#: **NA**

DIRECT CULTURE SET-UP (DAY 0) DATE: **11/05/2008**

INDICATOR CELL LINE (VERO) SEE DNA FLUOROCHROME RECORD SHEET

| | | | | DATE |
|---|---|---|---|---|
| THIOGLYCOLLATE BROTH | DAY 7 | + | ⊖ | 11/12/2008 |
| | DAY 28 | + | ⊖ | 12/03/2008 |
| BROTH-FORTIFIED COMMERCIAL | | | | |
| 0.5 mL SAMPLE | DAY 7 | + | ⊖ | 11/12/2008 |
| 6.0 mL BROTH | DAY 28 | + | ⊖ | 12/03/2008 |
| BROTH-MODIFIED HAYFLICK | | | | |
| 0.5 mL SAMPLE | DAY 7 | + | ⊖ | 11/12/2008 |
| 6.0 mL BROTH | DAY 28 | + | ⊖ | 12/03/2008 |
| BROTH-HEART INFUSION | | | | |
| 0.5 mL SAMPLE | DAY 7 | + | ⊖ | 11/12/2008 |
| 6.0 mL BROTH | DAY 28 | + | ⊖ | 12/03/2008 |

(See Reverse)

Document#: DCF3013D
Edition#: 10
Effective Date: 07/15/2003
Title: M-250 FINAL REPORT SHEET

| SAMPLE ID#: **55226** | | AEROBIC | MICROAEROPHILIC | DATE |
|---|---|---|---|---|
| AGAR PLATES-FORTIFIED COMMERCIAL | DAY 7 | + ⊖ | + ⊖ | 11/12/2008 |
| | DAY 14 | + ⊖ | + ⊖ | 11/19/2008 |
| | DAY 21 | + ⊖ | + ⊖ | 11/26/2008 |
| AGAR PLATES-MODIFIED HAYFLICK | DAY 7 | + ⊖ | + ⊖ | 11/12/2008 |
| | DAY 14 | + ⊖ | + ⊖ | 11/19/2008 |
| | DAY 21 | + ⊖ | + ⊖ | 11/26/2008 |
| AGAR PLATES-HEART INFUSION | DAY 7 | + ⊖ | + ⊖ | 11/12/2008 |
| | DAY 14 | + ⊖ | + ⊖ | 11/19/2008 |
| | DAY 21 | + ⊖ | + ⊖ | 11/26/2008 |
| BROTH SUBCULTURES (DAY 7) | | DATE: 11/12/2008 | | |
| AGAR PLATES-FORTIFIED COMMERCIAL | DAY 7 | + ⊖ | + ⊖ | 11/19/2008 |
| | DAY 14 | + ⊖ | + ⊖ | 11/26/2008 |
| | DAY 21 | + ⊖ | + ⊖ | 12/03/2008 |
| AGAR PLATES-MODIFIED HAYFLICK | DAY 7 | + ⊖ | + ⊖ | 11/19/2008 |
| | DAY 14 | + ⊖ | + ⊖ | 11/26/2008 |
| | DAY 21 | + ⊖ | + ⊖ | 12/03/2008 |
| AGAR PLATES-HEART INFUSION | DAY 7 | + ⊖ | + ⊖ | 11/19/2008 |
| | DAY 14 | + ⊖ | + ⊖ | 11/26/2008 |
| | DAY 21 | + ⊖ | + ⊖ | 12/03/2008 |

RESULTS: No detectable mycoplasmal contamination

12/3/08
Date

Laboratory Director

M-250 Procedural Summary: The objective of this test is to ascertain whether or not detectable mycoplasmas are present in an in vitro cell culture sample, be it a primary culture, hybridoma, master seed stock or cell line. This procedure combines an indirect DNA staining approach to detect non-cultivable mycoplasmas with a direct culture methodology utilizing three different mycoplasmal media formulations. The indirect approach involves the inoculation of the sample into a mycoplasma-free VERO (ATCC) indicator cell line and performing a DNA fluorochrome assay after 72-120 hours of incubation. The direct culture aspect of the test utilizes three different mycoplasmal media including both broth and agar formulations. The sample is inoculated into each of the 3 broth formulations and also onto duplicate plates (0.1 mL/plate) for each of the 3 agar formulations. Subculture from broth to fresh agar plates is carried out after 7 days incubation. Agar plates are incubated aerobically and microaerophillically in order to detect any colony forming units morphologically indicative of mycoplasmal contamination. Issuance of the final report with signature of the Scientific Director/Study Director signifies that the required controls were performed concurrently with the test sample(s) as detailed in the referenced SOPs and that all test conditions have been found to meet the required acceptance criteria for a valid test, including the appropriate results for the positive and negative controls.

Mycoplasma Testing Services

MYCOPLASMA TESTING SERVICES

BIONIQUE TESTING LABORATORIES, INC

APPENDIX I

| | |
|---|---|
| Document #: | DCF3008A |
| Edition #: | 06 |
| Effective date: | 9/17/2003 |
| Title: | DNA FLUOROCHROME ASSAY RESULTS |

## DNA-FLUOROCHROME ASSAY RESULTS

Procedures 3008, 3009, 3011

Sample ID # **55226** **M-250** Date Rec'd: **11/04/2008** P.O. #

Indicator Cells Inoculated: Date/Initials: 11/6/08 / JA

Fixation: Date/Initials: 11/10/08 / KG

Staining: Date/Initials: 11/10/08 / KG

TEST/CONTROL ARTICLE:

**6618 WA01 (OCT4 KI) MCB1**

LOT# **NA**

**Wicell QA**
**WiCell Research Institute**

---

**DNA FLUOROCHROME ASSAY RESULTS:**

X **NEGATIVE:** A reaction with staining limited to the nuclear region, which indicates no mycoplasmal contamination.

____ **POSITIVE:** A significant amount of extranuclear staining which strongly suggests mycoplasmal contamination.

____ **INCONCLUSIVE:**

____ A significant amount of extranuclear staining consistent with low - level mycoplasmal contamination or nuclear degeneration.

____ A significant amount of extranuclear staining consistent with bacterial, fungal or other microbial contaminant or viral CPE. Morphology not consistent for mycoplasmal contamination.

COMMENTS: _____

Date: 11/10/08 Results Read by: KG Date of Review: 11/10/08 Reviewed by: SA

# WiCell Cytogenetics Report: 000782-103108
## WISC 6618

**Report Date:** November 11, 2008

## Case Details:

***Cell Line:*** *WA01(Oct4KI)-MCB-1 (6618)*

***Passage #:*** *62*

***Date Completed:*** *11/10/2008*

***Cell Line Gender:*** *Male*

***Investigator:*** *WiCell Stem Cell Bank*

***Specimen:*** *hESC on Matrigel*

***Date of Sample:*** *10/31/2008*

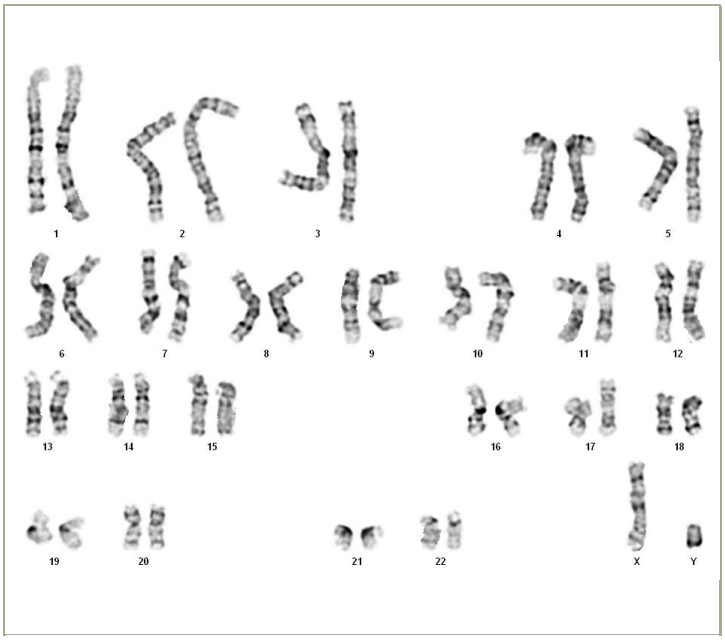
***Tests,Reason for:*** *Wisc Bank- FTDL*

***Results:*** *46,XY*

*Completed by ST, CLSp(CG), on 11/10/2008*

*Reviewed and interpreted by KDM, PhD, FACMG, on 11/10/2008*

***Interpretation:*** *No abnormalities were detected at the stated band level of resolution.*




***Cell:*** *S01-03*

***Slide:*** *B*

***Slide Type:*** *Karyotyping*

***Cell Results:*** *Karyotype: 46,XY*

***# of Cells Counted:*** *20*

***# of Cells Karyotyped:*** *3*

***# of Cells Analyzed:*** *7*

***Band Level:*** *450-550*

**Results Transmitted by Fax / Email / Post** **Date:**_____

**Sent By:**_____ **Sent To:**_____

# Product Information and Testing - Amended

## Product Information

| | |
|---|---|
| Product Name | H1 OCT4-EGFP |
| Lot Number | WA01(Oct4KI)-DL-02 |
| Parent Material | WA01(Oct4KI)-MCB-01 |
| Depositor | University of Wisconsin – Laboratory of Dr. James Thomson |
| Banked by | WiCell |
| Thaw Recommendation | Thaw 1 vial into 1 well of a 6 well plate. |
| Culture Platform | Feeder Independent |
| | Medium: mTeSR1 |
| | Matrix: Matrigel |
| Protocol | WiCell Feeder Independent Protocol modified to include feeding cells in the presence of G418 (Geneticin, Invitrogen catalog 11811) at a concentration of 50µg/ml. Not used for thawing, passaging, or freezing. |
| Passage Number | p69(10)<br><br>These cells were cultured for 68 passages prior to freeze, 10 of them in mTeSR1/Matrigel. WiCell adds +1 to the passage number at freeze so that the number on the vial best represents the overall passage number of the cells at thaw. |
| Date Vialed | 26-March-2009 |
| Vial Label | WA01(OCT4KI)-DL-2<br>P69(10) JT<br>26 MAR 2009<br>SOPCC038A |
| Biosafety and Use Information | Appropriate biosafety precautions should be followed when working with these cells. The end user is responsible for ensuring that the cells are handled and stored in an appropriate manner. WiCell is not responsible for damages or injuries that may result from the use of these cells.<br>Cells distributed by WiCell are intended for research purposes only and are not intended for use in humans. |

## Testing Performed by WiCell

| Test Description | Test Provider | Test Method | Test Specification | Result |
|---|---|---|---|---|
| Post-Thaw Viable Cell Recovery | WiCell | SOP-CH-305 | ≥ 15 Undifferentiated Colonies, ≤ 30% Differentiation | Pass |
| Identity by STR | UW Molecular Diagnostics Laboratory | PowerPlex 1.2 System by Promega | Consistent with known profile | Pass |
| Sterility - Direct transfer method | Apptec | 30744 | Negative | Pass |
| Mycoplasma | Bionique | M250 | No contamination detected | Pass |
| Karyotype by G-banding | WiCell | SOP-CH-003 | Normal karyotype | Pass |

Amendment(s):

| Reason for Amendment | Date |
|---|---|
| CoA updated to include copyright information. | See signature |
| CoA updated for format changes, including adding fields of thaw recommendation, vial label, protocol, and banked by, and incorporation of footnotes. | 28-Jun-2013 |
| CoA updated for clarification of test specifications and product description, added media footnote, and removed text regarding technical services | 05-Oct-2010 |
| CoA updated for format changes, clarification of test specifications, test method, addition of test provider, culture platform, and electronic signature, and reference to WiCell instead of the NSCB | 19-Aug-2010 |
| Original CoA | 21-Sept-2009 |

# *Product Information and Testing - Amended*

| Date of Lot Release | Quality Assurance Approval |
|---|---|
| 21-September-2009 | 12/30/2013<br>X AMC<br>AMC<br>Quality Assurance<br>Signed by: [redacted] |

UW Health

University of Wisconsin
Hospital and Clinics

Histocompatibility/Molecular Diagnostics Laboratory

# Short Tandem Repeat Analysis*

Sample Report: 8831-STR

UW HLA#: 61157

Sample Date: 06/18/09
Received Date: 06/18/09

Requestor: WiCell Research Institute

Test Date: 06/23/09

File Name: 090624

Report Date: 06/25/09

Sample Name: (label on tube) 8831-STR

Description: DNA Extracted by WiCell
271.19 ng/μL; 260/280 = 1.89

| Locus | Repeat # | STR Genotype |
|---|---|---|
| D16S539 | 5, 8-15 | 9,13 |
| D7S820 | 6-14 | 8,12 |
| D13S317 | 7-15 | 8,11 |
| D5S818 | 7-15 | 9,11 |
| CSF1PO | 6-15 | 12,13 |
| TPOX | 6-13 | 8,11 |
| Amelogenin | NA | X,Y |
| TH01 | 5-11 | 9.3,9.3 |
| vWA | 11, 13-21 | 15,17 |

**Comments: Based on the 8831-STR DNA submitted by WI Cell dated 06/18/09 and received on 06/18/09, this sample (UW HLA# 61157) matches exactly the STR profile of the human stem cell line H1 comprising 15 allelic polymorphisms across the 8 STR loci analyzed. No STR polymorphisms other than those corresponding to the human H1 stem cell line were detected and the concentration of DNA required to achieve an acceptable STR genotype (signal/ noise) was equivalent to that required for the standard procedure (~1 ng/amplification reaction) from human genomic DNA. This result suggest that the 8831-STR DNA sample submitted corresponds to the H1 stem cell line and was not contaminated with any other human stem cells or a significant amount of mouse feeder layer cells. Sensitivity limits for detection of STR polymorphisms unique to either this or other human stem cell lines is ~5%.**

Date

HLA/Molecular Diagnostics Laboratory

Date

HLA/Molecular Diagnostics Laboratory

* Testing to assess engraftment following bone marrow transplantation was accomplished by analysis of human genetic polymorphisms at STR loci. This methodology has not yet been approved by the FDA and is for investigational use only.

File: Final STR Report

WuXi AppTec



WiCell Research Institute

April 23, 2009
P.O. #:

# STERILITY TEST REPORT

**Sample Information:** hES Cells
8: WA01 (Oct 4 KI)-DL-2, 4976

**Date Received:** April 07, 2009
**Date in Test:** April 08, 2009
**Date Completed:** April 22, 2009

**Test Information:** Test Codes: 30744, 30744A
Immersion, USP / 21 CFR 610.12
Procedure #: BS210WCR.201

| TEST PARAMETERS | PRODUCT | |
|---|---|---|
| **Approximate Volume Tested** | 0.5 mL | 0.5 mL |
| **Number Tested** | 2 | 2 |
| **Type of Media** | SCD | FTM |
| **Media Volume** | 400 mL | 400 mL |
| **Incubation Period** | 14 Days | 14 Days |
| **Incubation Temperature** | 20 °C to 25 °C | 30 °C to 35 °C |
| **RESULTS** | 2 NEGATIVE | 2 NEGATIVE |

**Page 1 Signed**
**QA Reviewer** **Date**

**Page 1 Signed**
**Technical Reviewer** **Date**

*Testing conducted in accordance with current Good Manufacturing Practices.*

Mycoplasma Testing Services

BIONIQUE TESTING LABORATORIES, INC.

APPENDIX IV



Document#: DCF3013D
Edition#: 10
Effective Date: 07/15/2003
Title: **M-250 FINAL REPORT SHEET**

## M-250 FINAL REPORT

Direct Specimen Culture
Procedure 3008, 3011, 3013

**TO:**

BTL SAMPLE ID#: **57705** P.O.#: DATE REC'D: **06/11/2009**

TEST/CONTROL ARTICLE:

**WA01(Oct4K1)-DL-02-G #8831**

LOT#: **NA**

DIRECT CULTURE SET-UP (DAY 0) DATE: **06/11/2009**

INDICATOR CELL LINE (VERO) SEE DNA FLUOROCHROME RECORD SHEET

| | | | | DATE |
|---|---|---|---|---|
| THIOGLYCOLLATE BROTH | DAY 7 | + | (−) | 06/18/2009 |
| | DAY 28 | + | (−) | 07/09/2009 |
| BROTH-FORTIFIED COMMERCIAL | | | | |
| 0.5 mL SAMPLE | DAY 7 | + | (−) | 06/18/2009 |
| 6.0 mL BROTH | DAY 28 | + | (−) | 07/09/2009 |
| BROTH-MODIFIED HAYFLICK | | | | |
| 0.5 mL SAMPLE | DAY 7 | + | (−) | 06/18/2009 |
| 6.0 mL BROTH | DAY 28 | + | (−) | 07/09/2009 |
| BROTH-HEART INFUSION | | | | |
| 0.5 mL SAMPLE | DAY 7 | + | (−) | 06/18/2009 |
| 6.0 mL BROTH | DAY 28 | + | (−) | 07/09/2009 |

(See Reverse)

Document#: DCF3013D
Edition#: 10
Effective Date: 07/15/2003
Title: M-250 FINAL REPORT SHEET

| SAMPLE ID#: 57705 | | AEROBIC | MICROAEROPHILIC | DATE |
|---|---|---|---|---|
| AGAR PLATES-FORTIFIED COMMERCIAL | DAY 7 | + (−) | + (−) | 06/18/2009 |
| | DAY 14 | + (−) | + (−) | 06/25/2009 |
| | DAY 21 | + (−) | + (−) | 07/02/2009 |
| AGAR PLATES-MODIFIED HAYFLICK | DAY 7 | + (−) | + (−) | 06/18/2009 |
| | DAY 14 | + (−) | + (−) | 06/25/2009 |
| | DAY 21 | + (−) | + (−) | 07/02/2009 |
| AGAR PLATES-HEART INFUSION | DAY 7 | + (−) | + (−) | 06/18/2009 |
| | DAY 14 | + (−) | + (−) | 06/25/2009 |
| | DAY 21 | + (−) | + (−) | 07/02/2009 |
| **BROTH SUBCULTURES (DAY 7)** | | DATE: 06/18/2009 | | |
| AGAR PLATES-FORTIFIED COMMERCIAL | DAY 7 | + (−) | + (−) | 06/25/2009 |
| | DAY 14 | + (−) | + (−) | 07/02/2009 |
| | DAY 21 | + (−) | + (−) | 07/09/2009 |
| AGAR PLATES-MODIFIED HAYFLICK | DAY 7 | + (−) | + (−) | 06/25/2009 |
| | DAY 14 | + (−) | + (−) | 07/02/2009 |
| | DAY 21 | + (−) | + (−) | 07/09/2009 |
| AGAR PLATES-HEART INFUSION | DAY 7 | + (−) | + (−) | 06/25/2009 |
| | DAY 14 | + (−) | + (−) | 07/02/2009 |
| | DAY 21 | + (−) | + (−) | 07/09/2009 |

RESULTS: No detectable mycoplasmal contamination

7-9-09
Date

Laboratory Director

M-250 Procedural Summary: The objective of this test is to ascertain whether or not detectable mycoplasmas are present in an in vitro cell culture sample, be it a primary culture, hybridoma, master seed stock or cell line. This procedure combines an indirect DNA staining approach to detect non-cultivable mycoplasmas with a direct culture methodology utilizing three different mycoplasmal media formulations. The indirect approach involves the inoculation of the sample into a mycoplasma-free VERO (ATCC) indicator cell line and performing a DNA fluorochrome assay after 72-120 hours of incubation. The direct culture aspect of the test utilizes three different mycoplasmal media including both broth and agar formulations. The sample is inoculated into each of the 3 broth formulations and also onto duplicate plates (0.1 mL/plate) for each of the 3 agar formulations. Subculture from broth to fresh agar plates is carried out after 7 days incubation. Agar plates are incubated aerobically and microaerophillically in order to detect any colony forming units morphologically indicative of mycoplasmal contamination. Issuance of the final report with signature of the Laboratory Director signifies that the required controls were performed concurrently with the test sample(s) as detailed in the referenced SOPs and that all test conditions have been found to meet the required acceptance criteria for a valid test, including the appropriate results for the positive and negative controls.

APPENDIX I

| | |
|---|---|
| Document #: | DCF3008A |
| Edition #: | 06 |
| Effective date: | 9/17/2003 |
| Title: | DNA FLUOROCHROME ASSAY RESULTS |

## DNA-FLUOROCHROME ASSAY RESULTS

Procedures 3008, 3009, 3011

Sample ID # **57705** **M-250** Date Rec'd: **06/11/2009** P.O. #

Indicator Cells Inoculated: Date/Initials: 6/11/09 / JA

Fixation: Date/Initials: 6/15/09 / JA

Staining: Date/Initials: 6/15/09 / JA

TEST/CONTROL ARTICLE:

**WA01(Oct4K1)-DL-02-G #8831**

LOT# **NA**

---

**DNA FLUOROCHROME ASSAY RESULTS:**

X **NEGATIVE:** A reaction with staining limited to the nuclear region, which indicates no mycoplasmal contamination.

____ **POSITIVE:** A significant amount of extranuclear staining which strongly suggests mycoplasmal contamination.

____ **INCONCLUSIVE:**

____ A significant amount of extranuclear staining consistent with low - level mycoplasmal contamination or nuclear degeneration.

____ A significant amount of extranuclear staining consistent with bacterial, fungal or other microbial contaminant or viral CPE. Morphology not consistent for mycoplasmal contamination.

COMMENTS: _____

Date: 6/15/09 Results Read by: JA Date of Review: 6/15/09 Reviewed by: U

# WiCell Cytogenetics Report: 001123-052909

## NSCB 8831

**Report Date:** June 03, 2009

## Case Details:

***Cell Line:*** *WA01(Oct4KI)-DL-2 (8831)*

***Passage #:*** *73(14)*

***Date Completed:*** *6/3/2009*

***Cell Line Gender:*** *Male*

***Investigator:*** *National Stem Cell Bank*

***Specimen:*** *hESC on Matrigel*

***Date of Sample:*** *5/29/2009*

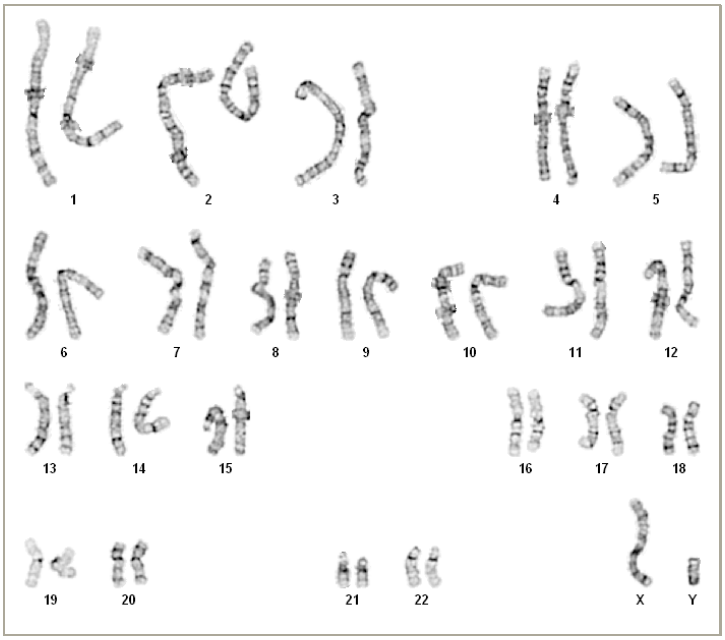
***Tests,Reason for:*** *FTDL Equivalent*

***Results:*** *46,XY*

*Completed by* *on 6/3/2009*

*Reviewed and interpreted by* *on 6/3/2009*

***Interpretation:*** *No abnormalities were detected at the stated band level of resolution.*




***Cell:*** *S01-01*

***Slide:*** *A*

***Slide Type:*** *Karyotyping*

***Cell Results:*** *Karyotype: 46,XY*

***# of Cells Counted:*** *20*

***# of Cells Karyotyped:*** *4*

***# of Cells Analyzed:*** *8*

***Band Level:*** *450-600*

**Results Transmitted by Fax / Email / Post** **Date:_____**

**Sent By:_____** **Sent To:_____**

**QC Review By: _____** **Results Recorded: _____**