

Chordoma Foundation Cell Line

U-CH1

Cell Line Phenotype and Expression
Analysis Report

June 22, 2015

Cell Line Receiving

Format Received	Date Received	Condition	Quantity	Passage	Initial Cell Count	Initial Cell Viability
Flasks (T25)	November 26, 2013	sparse	2	p. 37	N/A	N/A

Growth Conditions

Media:

4:1 IMDM/RPMI 1640 + 10% HI FBS

+ Pen/Strep

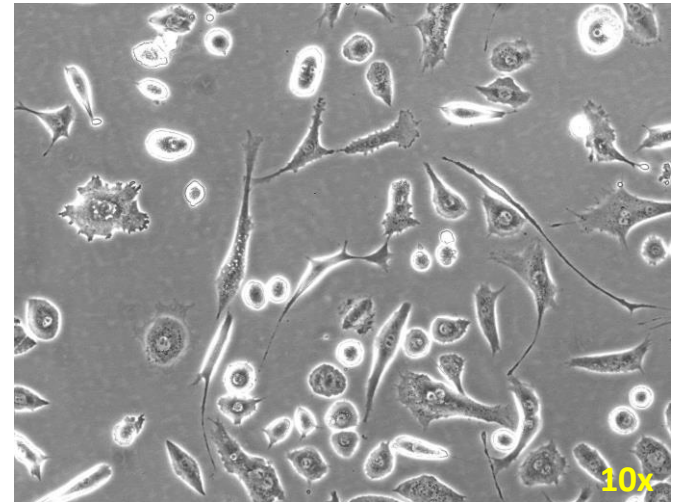
→ Use flasks coated with 0.1% gelatin

→ Passage when ~80-90% confluent (1:3, 1:5)

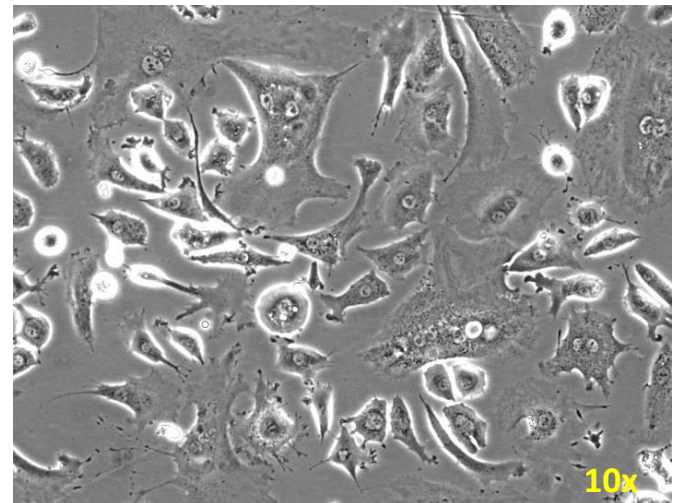
→ Change media every 3 days

Phase Contrast Image Review

Cells arrived live, in 2 T25 flasks. Were sparse looking when they arrived but recovered after 24 hours. They are visibly clear of contamination and grow well.



U-CH1 arrival day (11/26/2013)

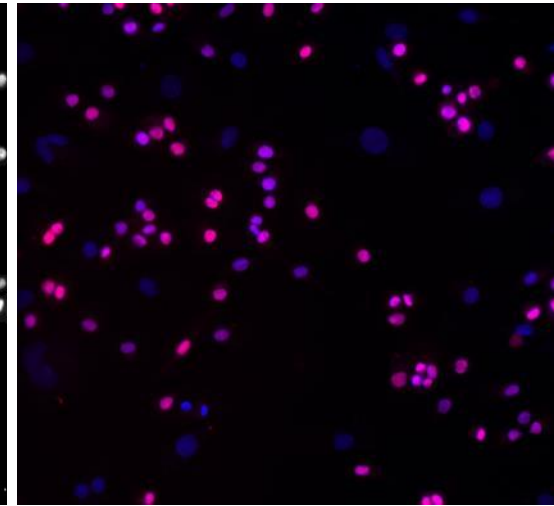
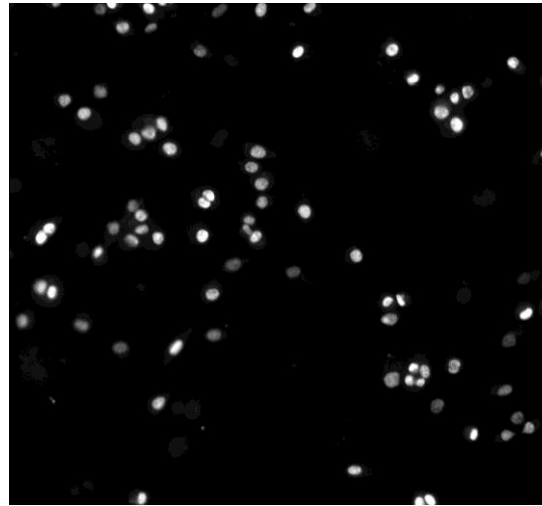
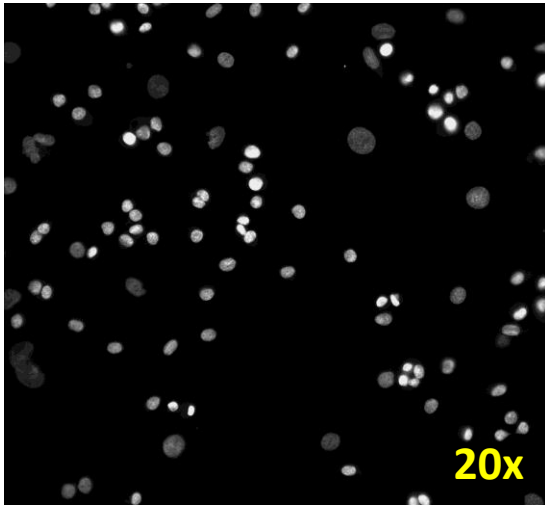


24 Hours After Receiving (11/27/2013)

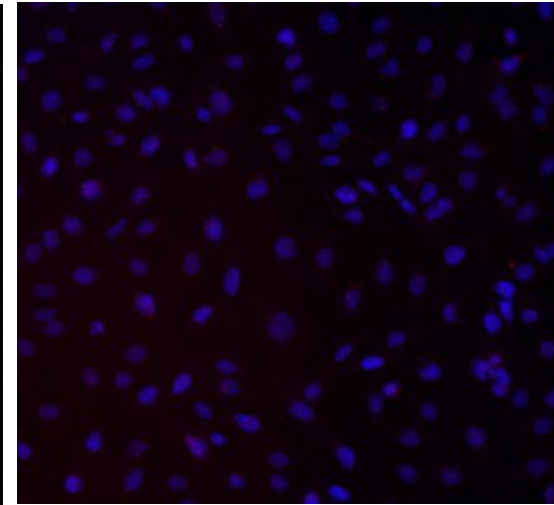
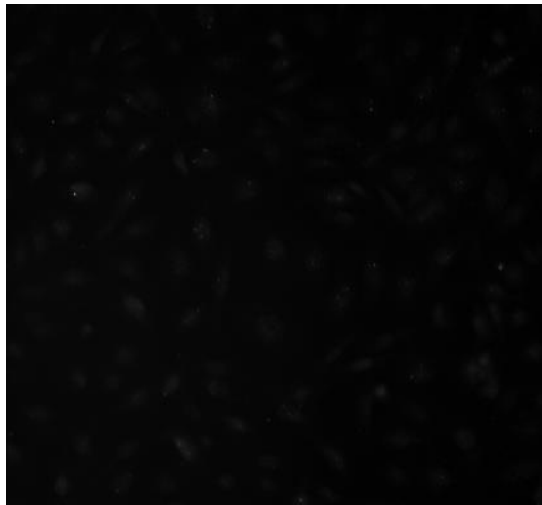
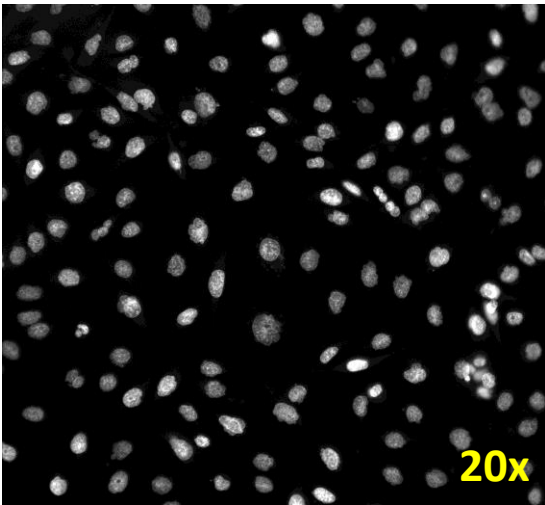
Cell Line Immunofluorescence Validation

U-CH1 (Supplied by Duke University) p.46 versus Non-Chordoma Negative Control

U-CH1
Duke



MDA-MB-231
Negative
Control



Hoechst Nuclear Stain

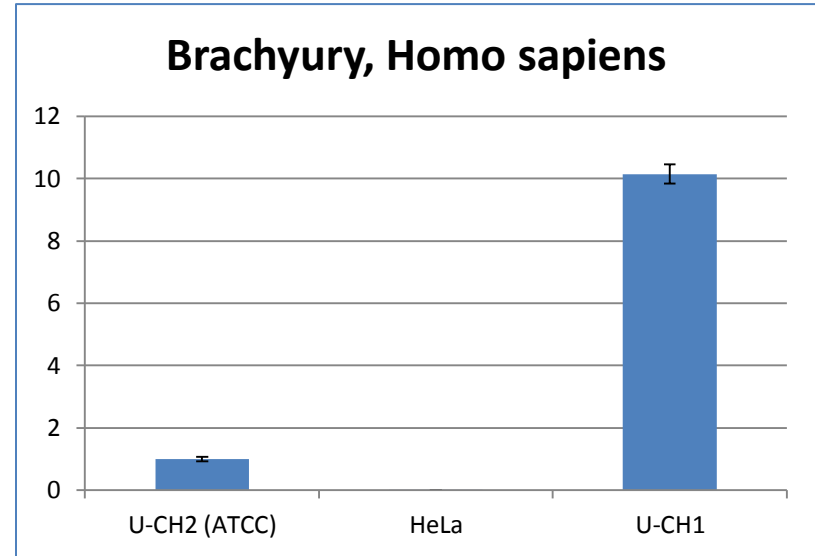
Anti-Brachyury Antibody

Color Composite

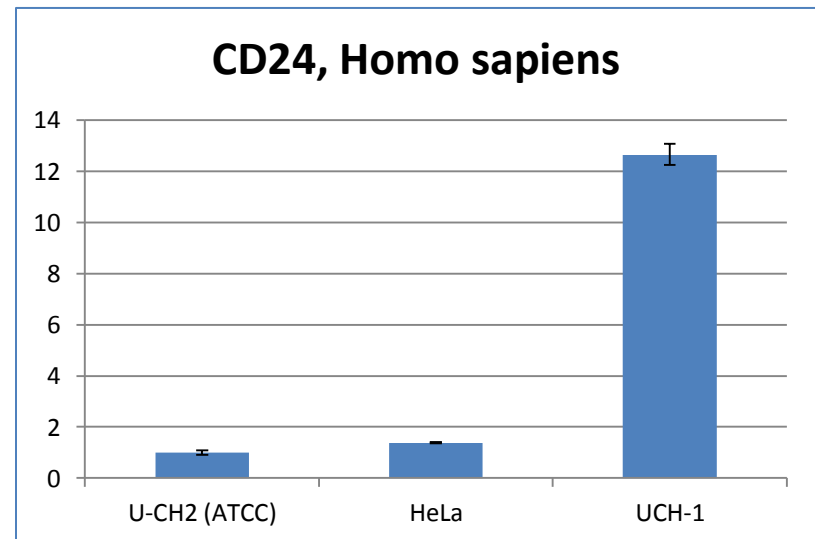
Cell Line PCR Validation

Relative quantification of Brachyury and CD24 gene in U-CH1 cell line

Sample	BRACHYURY, Homo sapiens	Neg. Error	Pos. Error
U-CH2 (ATCC)	1	0.071225	0.076687
HeLa	0.009183	0.00052	0.00055
U-CH1	10.143163	0.301634	0.310879



Sample	CD24, Homo sapiens	Neg. Error	Pos. Error
U-CH2 (ATCC)	1	0.078939	0.085705
HeLa	1.386653	0.03058	0.03127
UCH-1	12.643509	0.408367	0.421997



Tables and associated graphs depict relative quantification of N (top table and graph) and Z (bottom table and graph) gene expression/RNA in TEST cell samples. Gene expression across all assessed lines is set relative to the positive control sample, which is set at 1. The X-axis represents cell lines assessed and the Y-axis represents gene expression relative to positive control.

Cell Line Validation Results

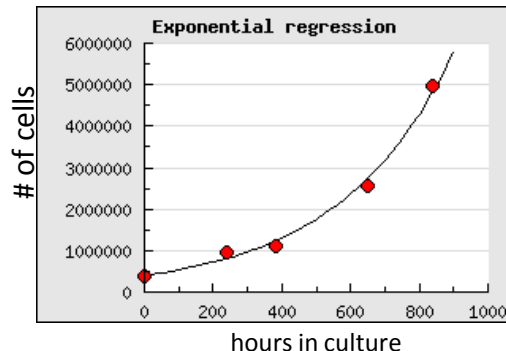
Results summary report of U-CH1

TEST	SPECIFICATION	RESULTS
Cell Growth	Immortalized	Doubling time = 9 days
STR Analysis	Human, unique	Pass
IF Validation	Signal in nucleus	Pass
PCR Validation	Expressing Brachyury and CD24	Pass

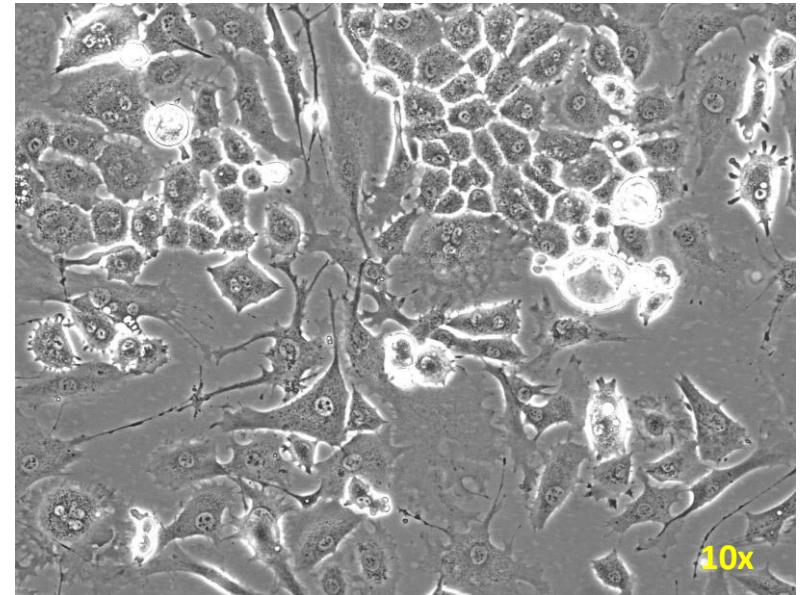
Cell lot generated

Stock Lot#	EB1013-056
Cells per vial	3.0×10^6
Lot Viability	98.3%
Passages	p.45

Cell Line Growth: Cell doubling time= 9 days



Cell growth rates were calculated from an actively growing culture for four passages. Growth rates will likely be slower when calculated from a fresh thaw.



**U-CH1 Vala cell lot EB1013-056
viability thaw**