

Chordoma Foundation Cell Line Validation

U-CH17M

Cell Line Phenotype and Expression
Analysis Report

April 13, 2017

Cell Line Receiving

Format Received	Date Received	Condition	Quantity	Passage	Initial Cell Count	Initial Cell Viability
Frozen Vials	January 12, 2017	N/A	3	p (0),	7.8×10^5	79%

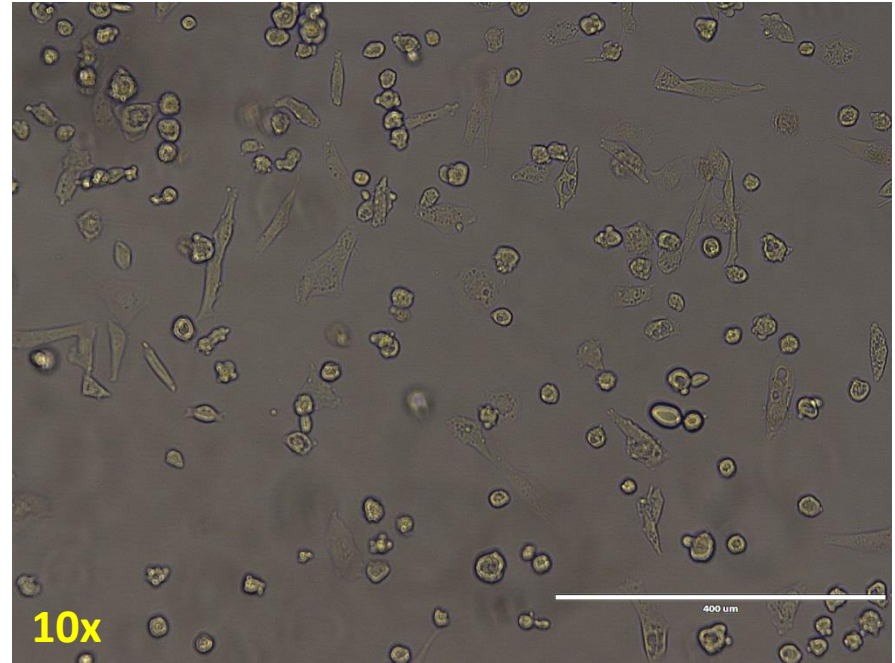
Growth Conditions

Media:

4:1 IMDM/RPMI + 10% FBS + l-Glutamine +
Pen/Strep

→ Passage when ~80-90% confluent (no more
than 1:2)

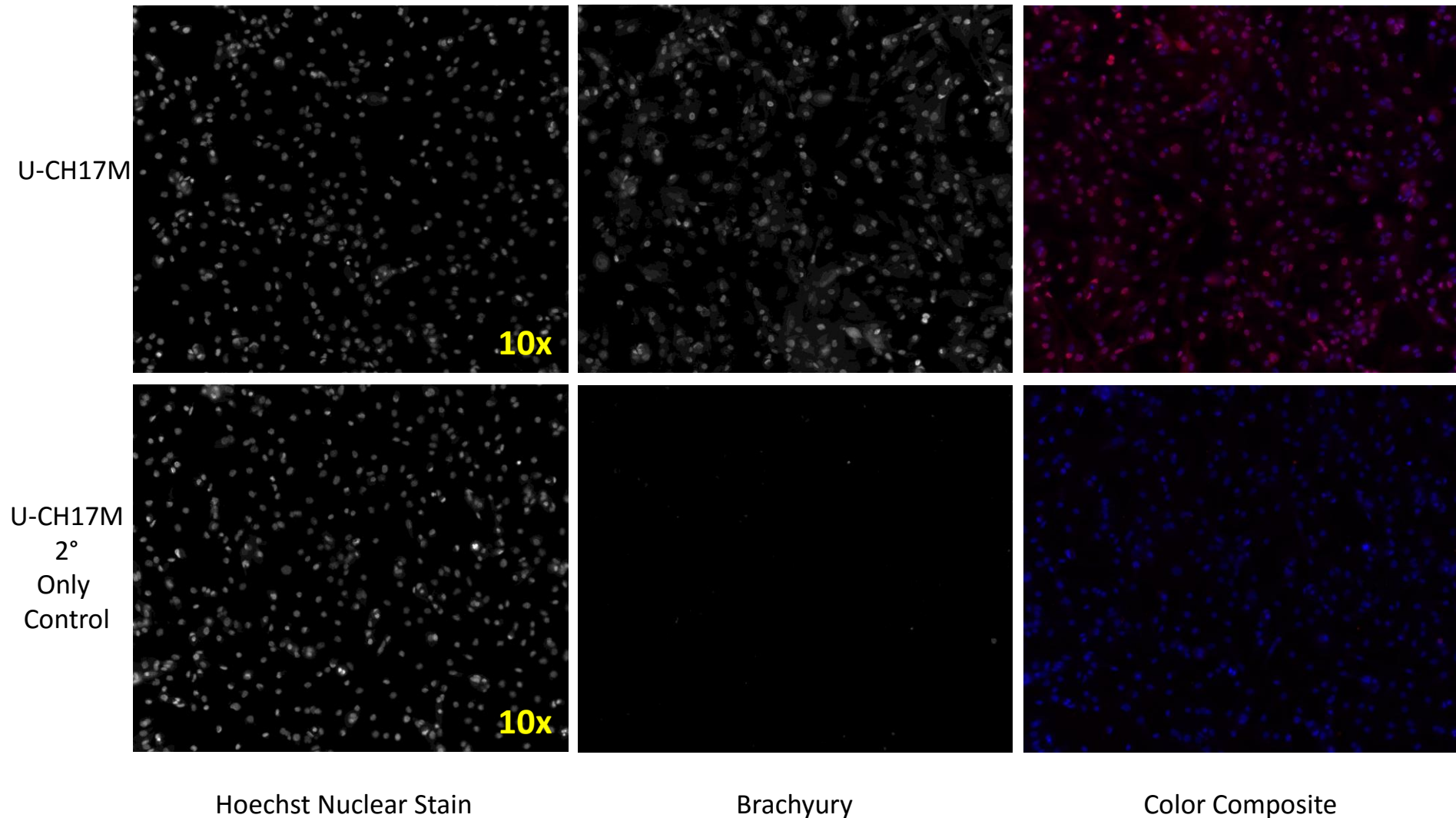
→ Change media every 5-7 days



U-CH17M 24hrs post thaw.

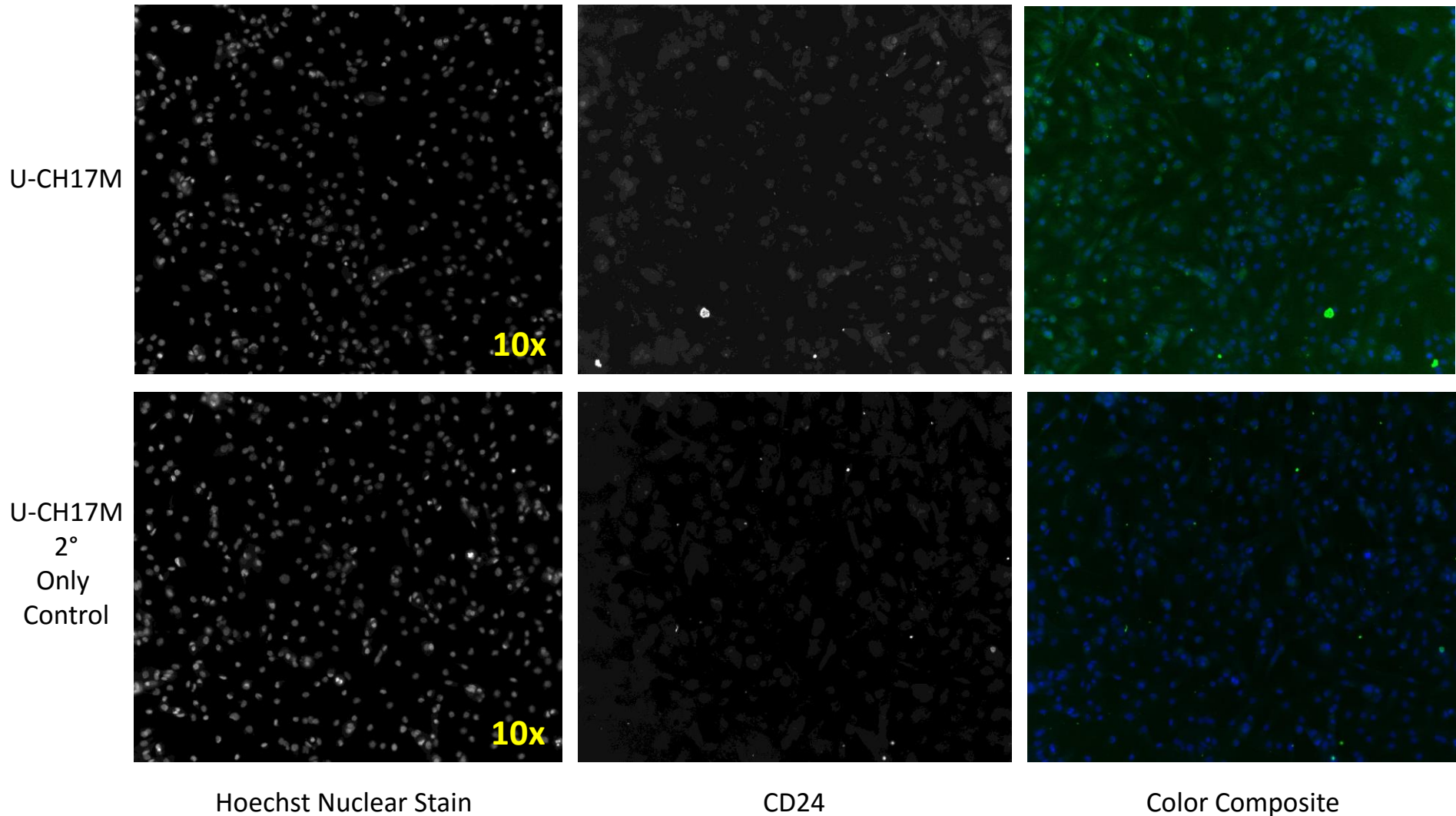
Cell Line Immunofluorescence Validation

U-CH17M p.(53) Anti-Brachyury versus Secondary-only Negative Control



Cell Line Immunofluorescence Validation

U-CH17M p.(53) Anti-CD24 versus Secondary-only Negative Control

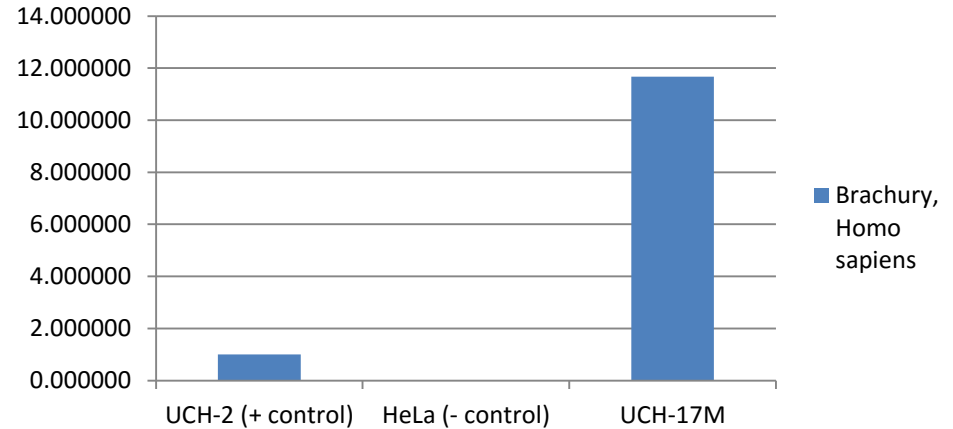


Cell Line PCR Validation

Relative quantification of Brachyury and CD24 gene in U-CH17M cell line

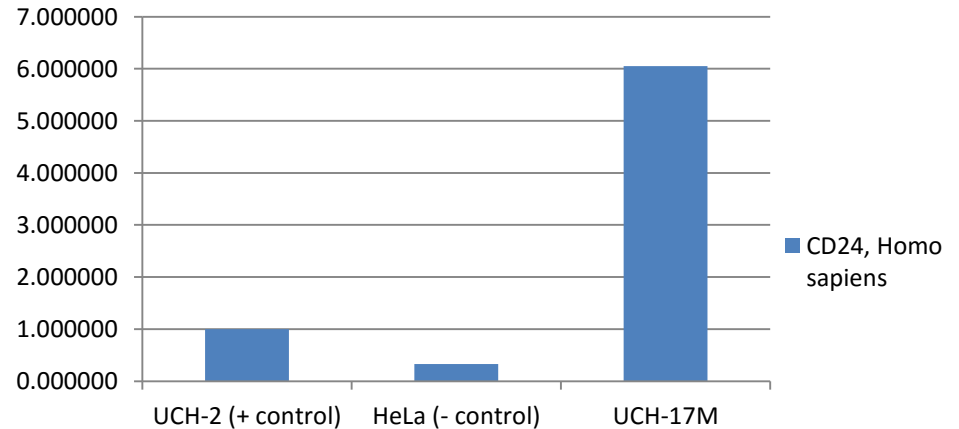
Sample	Brachury, Homo sapiens	Neg. Error	Pos. Error
UCH-2 (+ control)	1.000000	0.076062	0.082324
HeLa (- control)	0.000820	0.000052	0.000055
UCH-17M	11.675733	0.518295	0.542371

Brachury, Homo sapiens



Sample	CD24, Homo sapiens	Neg. Error	Pos. Error
UCH-2 (+ control)	1.000000	0.032074	0.033137
HeLa (- control)	0.326105	0.017748	0.018769
UCH-17M	6.052104	0.236485	0.246101

CD24, Homo sapiens



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

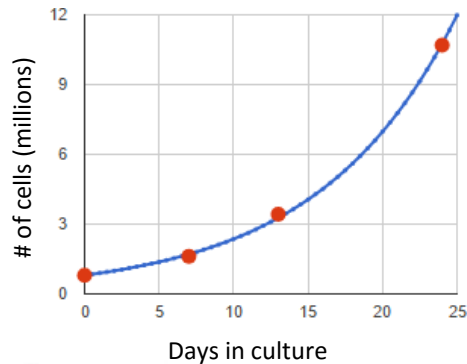
Results summary report of U-CH17M

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

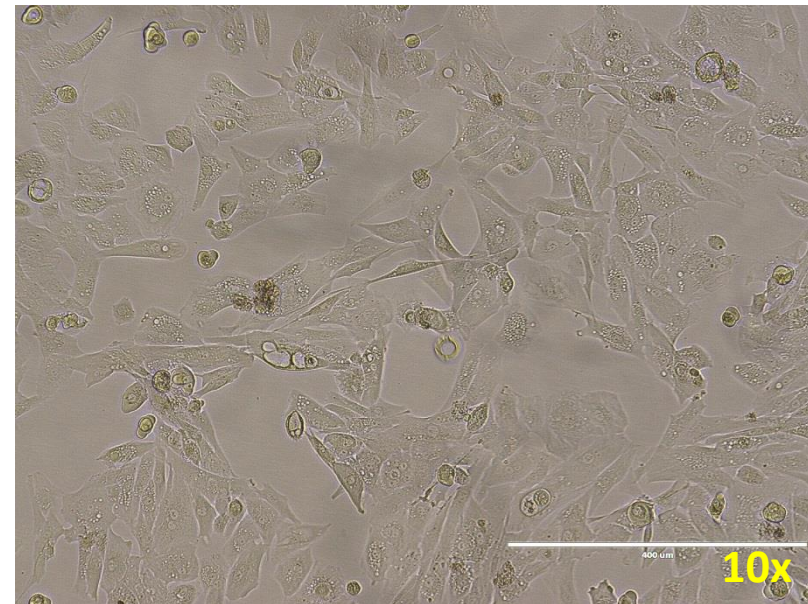
Cell lot generated

Stock Lot#	1027-002
Cells per vial	700k
Lot Viability	80%
Passages	5

Cell Line Growth: Cell doubling time= 6-8 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-CH17M, lot 1027-002.