

# PRODUCT SPOTLIGHT

## *ThawReady™* by ATCC EFFORTLESS SPHEROIDS, CONSISTENT PERFORMANCE

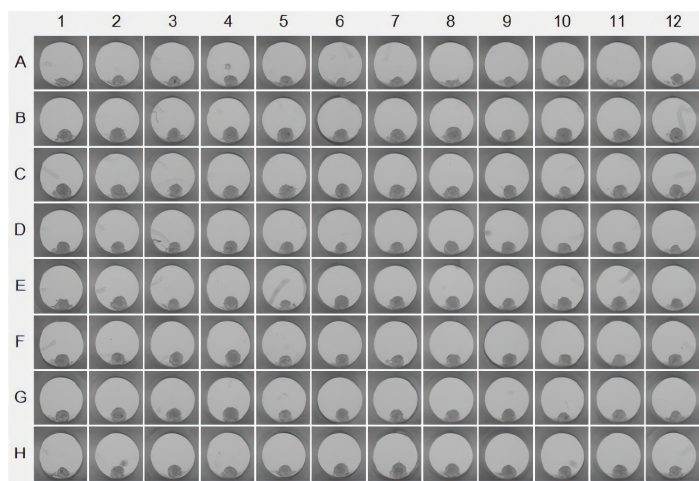
### THAWREADY™ HCT 116 SPHEROID KIT (ATCC® SCM-CCL-247-2PLT™)

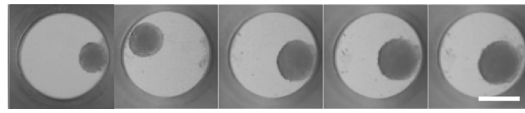
The ThawReady™ HCT 116 Spheroid Kit contains a 96-well plate that consists of 20  $\mu$ L of frozen HCT 116 cell solution in a cryoprotective agent within each well. The number of cells per well has been optimized to allow the formation of a single spheroid. This is achieved after 5 days of cell culture. The image on the right is a typical representation of HCT 116 spheroids on day 5 after thawing the plate using the Quick Start Guide protocol included in our ThawReady™ Spheroid Kit.

ThawReady™ HCT 116 plates can be employed to supplant conventional 2-dimensional cell cultures for all major assays.

#### REFERENCE DATA SET

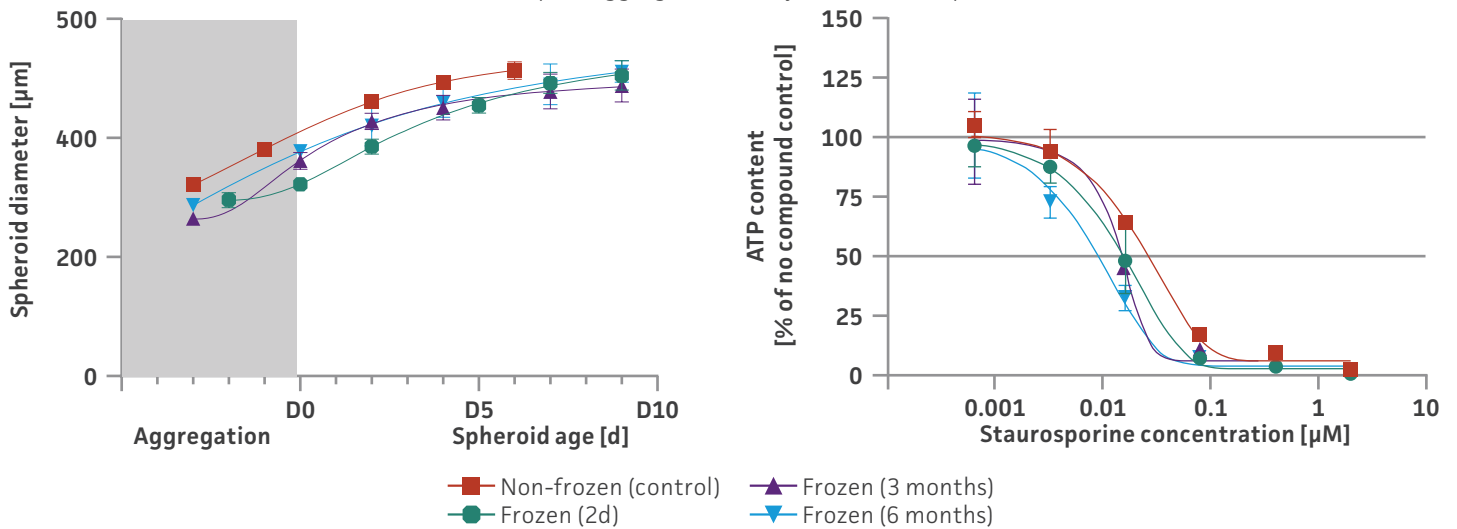
The HCT 116 cell line was evaluated for its growth rate, viability, and capacity to form 3-D spheroids. For the HCT 116 model, the recovery of cells and the formation and maturation of spheroids will take up to five days after the plate has been thawed. The completion of spheroid formation defines experimental day 0 as the point at which a typical treatment/experiment can be initiated. The growth in terms of proliferation and spheroid size was measured over a subsequent 9-day assay duration and benchmarked using the apoptotic agent staurosporine in a relevant concentration range.



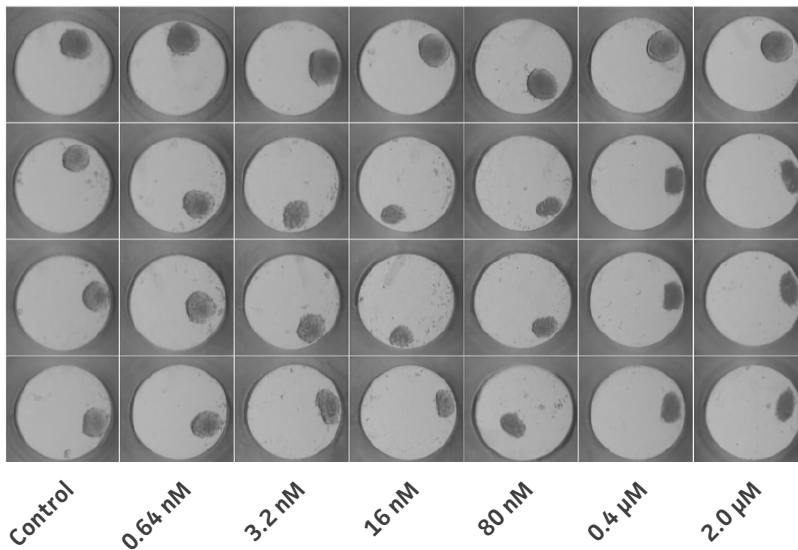


Day 0 → Day 9

Bright Field images of a typical HCT 116 spheroid; size over time from post aggregation to Day 9 (scale = 500  $\mu\text{m}$ )



The results from ThawReady™ plates with cell line HCT 116 from the same production run at different time points were compared to a non-frozen plate with cells from the same culture flask. The results demonstrate comparable growth and response to staurosporine.



### BENCHMARK DATA SET FOR DOSE RESPONSE

Brightfield images of technical replicates (N=4) of tumor spheroids on Day 9 for cell line HCT 116 showing consistent responses to increasing concentrations of staurosporine (scale = 500  $\mu\text{m}$ ). The ATP values are shown above.

## CELL LINE ORIGIN

HCT 116 cells were isolated from the colon of an adult male with colon cancer and have a mutation in codon 13 of the RAS proto-oncogene. This cell line can be used as a positive control for PCR assays of mutation in this codon. (ATCC® CCL-247™)

Cell Line		Model set up	
Product category	Human cells	Model type	Monotypic tumor spheroid
Organism	<i>Homo sapiens</i> , human	Assay-ready*	5 days in culture, 1 spheroid per well
Morphology	Epithelial-like	Morphology	Compact, spheroidal
Tissue	Large intestine; Colon	Size day 5 after thawing	~320 to 380 µm
Disease	Carcinoma; Colorectal	Size differences between spheroids	<15% in-plate <20% batch-to-batch
Applications	3-D cell culture Cancer research High-throughput screening Toxicology	Assay Application***	
		Assay window**	Up to 9 days
		Viability ATP Day 5	~15 to 25 pmol/MT
		Metabolism differences between spheroids	< 20% in plate < 20% batch-to-batch
		EC50 Staurosporine	0.01 µM
		Dose response variation	< 15%

\*Use recommended Aggregation Medium

\*\*Use Tumor Maintenance Medium

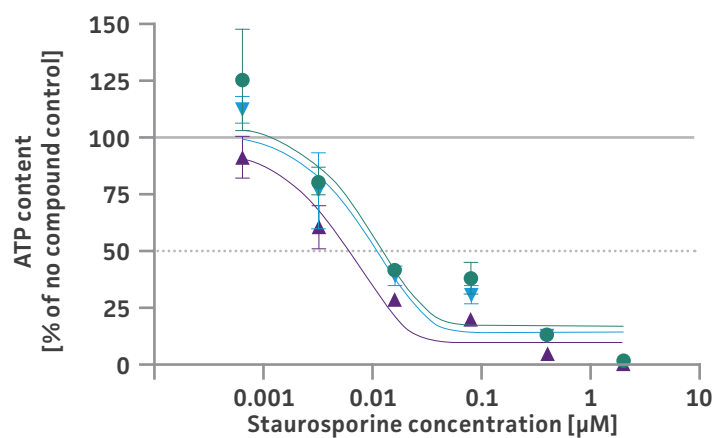
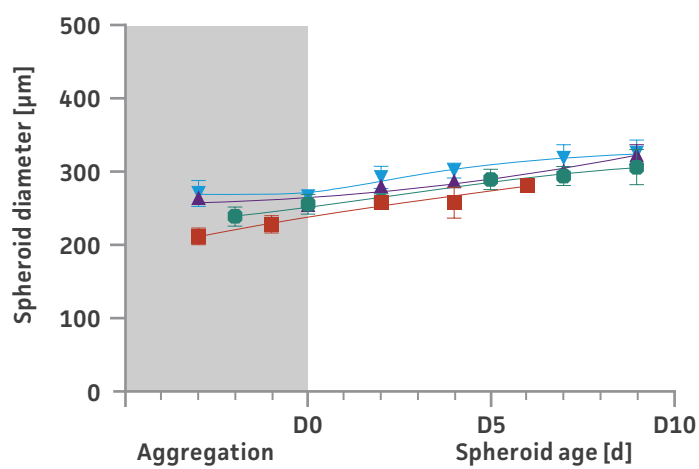
\*\*\*Results may vary on handling and assay conditions

**Table 1: Available ThawReady™ 3-D Spheroid Kits**

ATCC® No.	Designation	Components
SCM-HTB-133-2PLT™	ThawReady™ T-47D Spheroid Kit	2 plates of ThawReady™ T-47D cells 1 bottle ThawReady™ Spheroid Aggregation Media 1 bottle ThawReady™ Spheroid Maintenance Media 1 ThawReady™ Spheroid Assay Stand 1 ThawReady™ Spheroid Assay Balance Plate
SCM-CCL-185-2PLT™	ThawReady™ A549 Spheroid Kit	2 plates of ThawReady™ A549 cells 1 bottle ThawReady™ Spheroid Aggregation Media 1 bottle ThawReady™ Spheroid Maintenance Media 1 ThawReady™ Spheroid Assay Stand 1 ThawReady™ Spheroid Assay Balance Plate
SCM-CCL-247-2PLT™	ThawReady™ HCT 116 Spheroid Kit	2 plates of ThawReady™ HCT 116 cells 1 bottle ThawReady™ Spheroid Aggregation Media 1 bottle ThawReady™ Spheroid Maintenance Media 1 ThawReady™ Spheroid Assay Stand 1 ThawReady™ Spheroid Assay Balance Plate

**WWW.ATCC.ORG/THAWREADY-SPHEROIDKITS**





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