

## Assay Name: 3D multicellular tumor spheroid (MCTS) invasion screening

**Assay ID:** Celigo\_03\_0002

**Description:** Monitor the effect of a panel of drugs on the invasion of U87MG Glioblastoma MCTS into Basement Membrane Extract (BME) Matrigel

**Stains:** Label-Free

**Imaging channels:** Bright Field

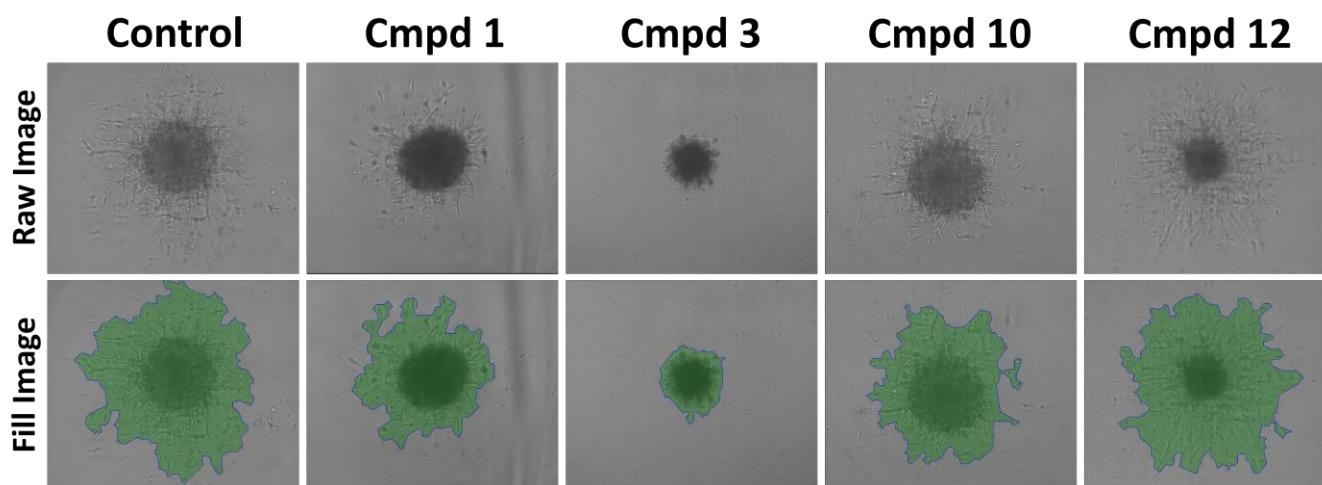
**Image analysis algorithm:** Celigo software Tumorsphere Migration

### Methods:

1. Seed 500 U87MG cells/well in ULA 384-well plates
2. On day 4, add different drug compounds at 2x or vehicle control in Basement Membrane Extract (BME) Matrigel
3. Monitor invasion by imaging and analyzing each 384-well plate at ~5 minutes/plate at 24, 48 and 72 hours on the Celigo imaging cytometer
4. Measure the invasion area on 24, 48, and 72 hours for each drug compound-treated MCTS
5. Compare the invasion area for each drug compound at each time point to characterize the tested compounds

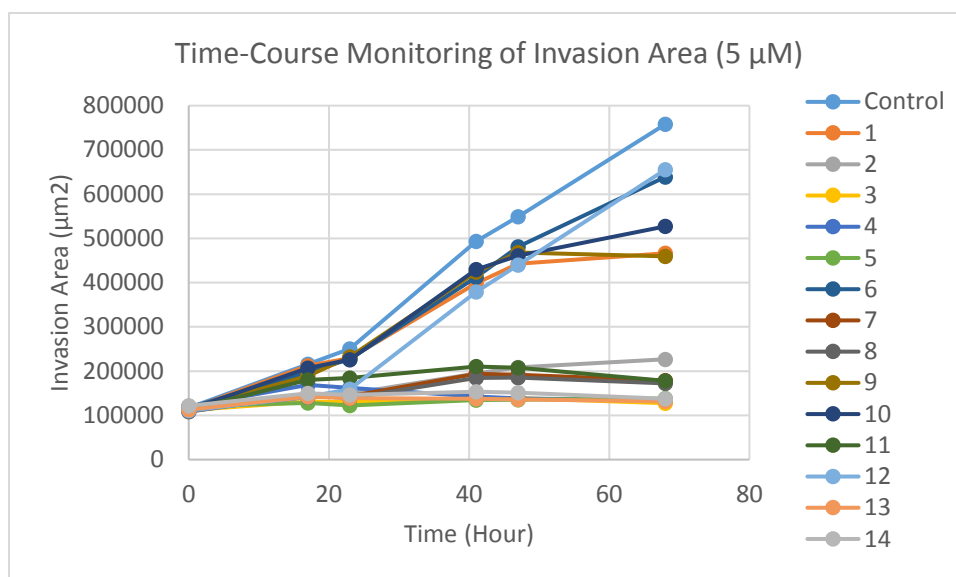
### Results:

Celigo captured Bright Field images showing invasion of the U87MG MCTS



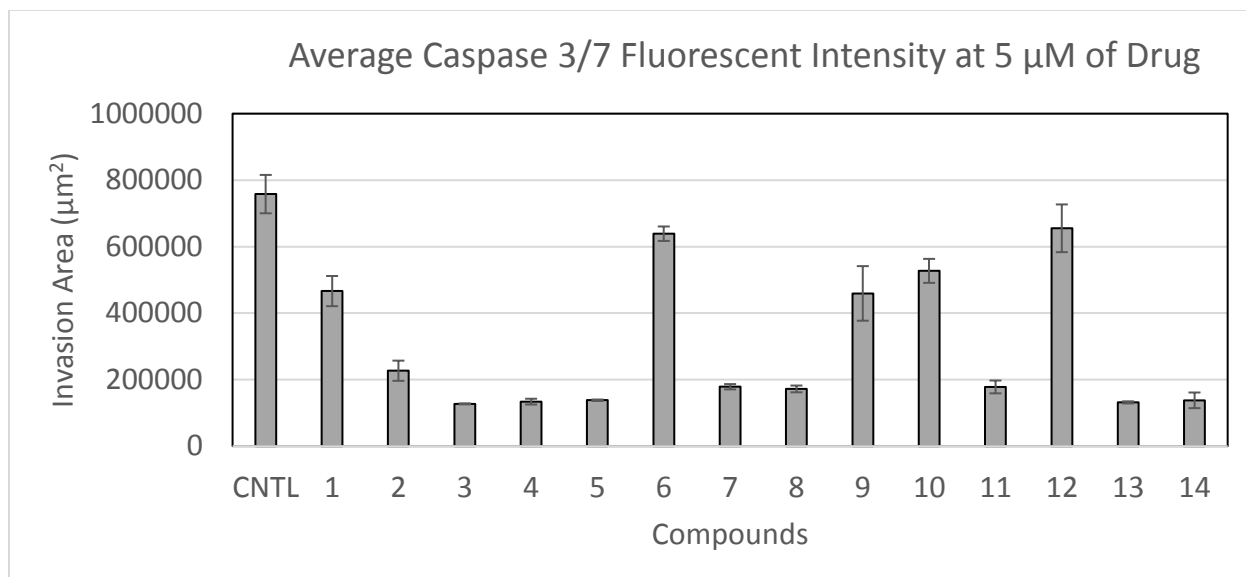
- The Celigo produces quantitative data and qualitative images, as displayed above
- The invasion area observed in the raw images can be outlined using a green fill option, allowing for simple visualization

## Kinetic measurement of 3D MCTS invasion inhibition



- The plot above was generated by exporting the data from Celigo into Excel and graphing the invasion area over time
- Kinetic monitoring allows the detection of the invasive inhibitory properties of compounds 10 and 12, which would not be detected in a 24 or 48 hour endpoint assay.

## Endpoint measurement of 3D MCTS invasion inhibition



- The plot above was an endpoint plot at Day 13. It shows the Control, and Compounds 1, 6, 9, 10, and 12 allowed normal invasion of the spheroids
- The rest of the drug compounds tested inhibited the invasion of MCTS into BME Matrigel