

3D Tumor Spheroid Assay (mono-spheroids)

➤ Spheroids as in vitro tumor surrogates

Three-dimensional tumor cell culture has been shown to mimic the physiological cancer situation more closely than growth on a flat surface. Spheroid analysis has evolved as one of the major 3D methods of choice for compound analysis due to multiple advantages:

- Cells autonomously assemble based on endogenous adhesion and matrix proteins, not requiring artificial matrix addition.
- Spheroidal structure challenges compounds to penetrate typical cell conglomerate barriers.
- Compatible with high-throughput analysis

➤ Assay procedure

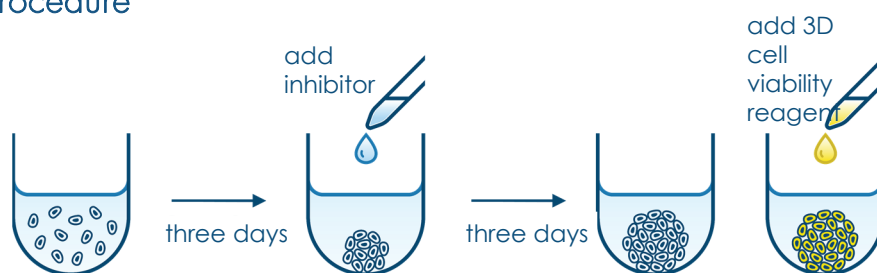


Figure 1: Assay procedure. Tumor cells are seeded in low attachment U-bottom 96-well plates in 20% Methocel in cell culture medium. Cells incubate for three days for spheroid formation. Subsequently, spheroids are treated with compounds for three days. After compound treatment, the cells are lysed by adding CellTiterGlo-3D™ reagent and luminescence is measured as a parameter for cell viability. IC₅₀ values are determined by testing 8 compound concentrations in duplicates.

This service is also available for co-culture spheroids containing tumor and stroma cells.

➤ Example U87MG mono-spheroids

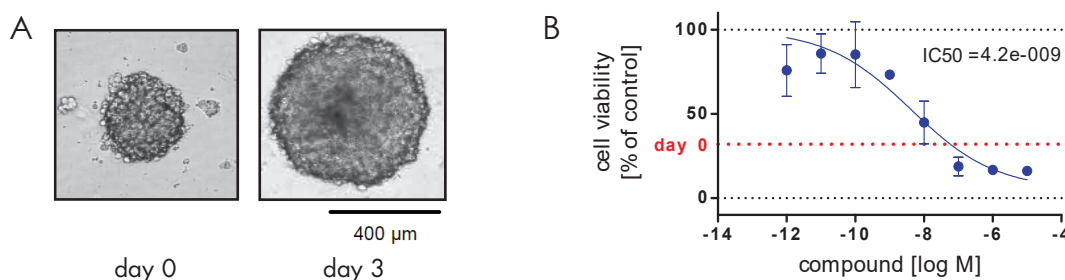


Figure 2: Study example. A. Comparison of size of U87MG spheroids at day 0 and 3. B. Dose-response of Trametinib treatment on U87MG mono-spheroids. Positive control (0% viability) is staurosporine treatment; negative control (100% viability) represents vehicle control. 'day 0' shows the number of viable cells present at the initiation of the experiment.