## 3D Tumor Spheroid Assay (co-culture 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.
- Combination of different cell types for co-spheroid studies possible.
- Compatible with high-throughput analysis.

## The Reaction Biology "3D Tumor Spheroid Assay" $\geq$

- Spheroid formation in low attachment u-bottom 96-well plates
- Viability assessment via luciferase-activity of Firefly or Renilla luciferase labeled cells.
- Available as mono- or co-spheroids.

( $\sqrt{}$  = good aggregates; 0= loose association)

- Simultaneous detection of two cell types in co-spheroids possible.
- Highly sensitive cell detection also allowing for low tumor/stroma ratios.

		Renilla Luciferase labelled stroma cells			
		none	HS5	HS27	NHDF
Firefly Luciferase labelled tumor cells	none		$\checkmark$	$\checkmark$	$\checkmark$
	A549	$\checkmark$	$\checkmark$	$\checkmark$	$\checkmark$
	DLD1	$\checkmark$	$\checkmark$	$\checkmark$	$\checkmark$
	НСТ116	$\checkmark$	$\checkmark$	$\checkmark$	$\checkmark$
	LN229	$\checkmark$	$\checkmark$	$\checkmark$	$\checkmark$
	MCF7	0	0	$\checkmark$	$\checkmark$
	U87MG	$\checkmark$	$\checkmark$	$\checkmark$	$\checkmark$

Figure 1: Co-Spheroid of 1000 Table 1: Cells currently available for mono- or co-spheroid studies.



**Co-Spheroid** 

U87MG tumor cells (Calcein stain) and 2000 HS27A stroma cells (RFP stain) one day after cell seeding.

## You ship your compounds – Reaction Biology performs the testing



- IC50 values are determined by testing 8 compound concentrations in deca- or semi-logarithmic steps (each concentration in duplicates).
- Quality assurance is provided by calculation of Z' factors for Low/High controls on each assay plate.

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