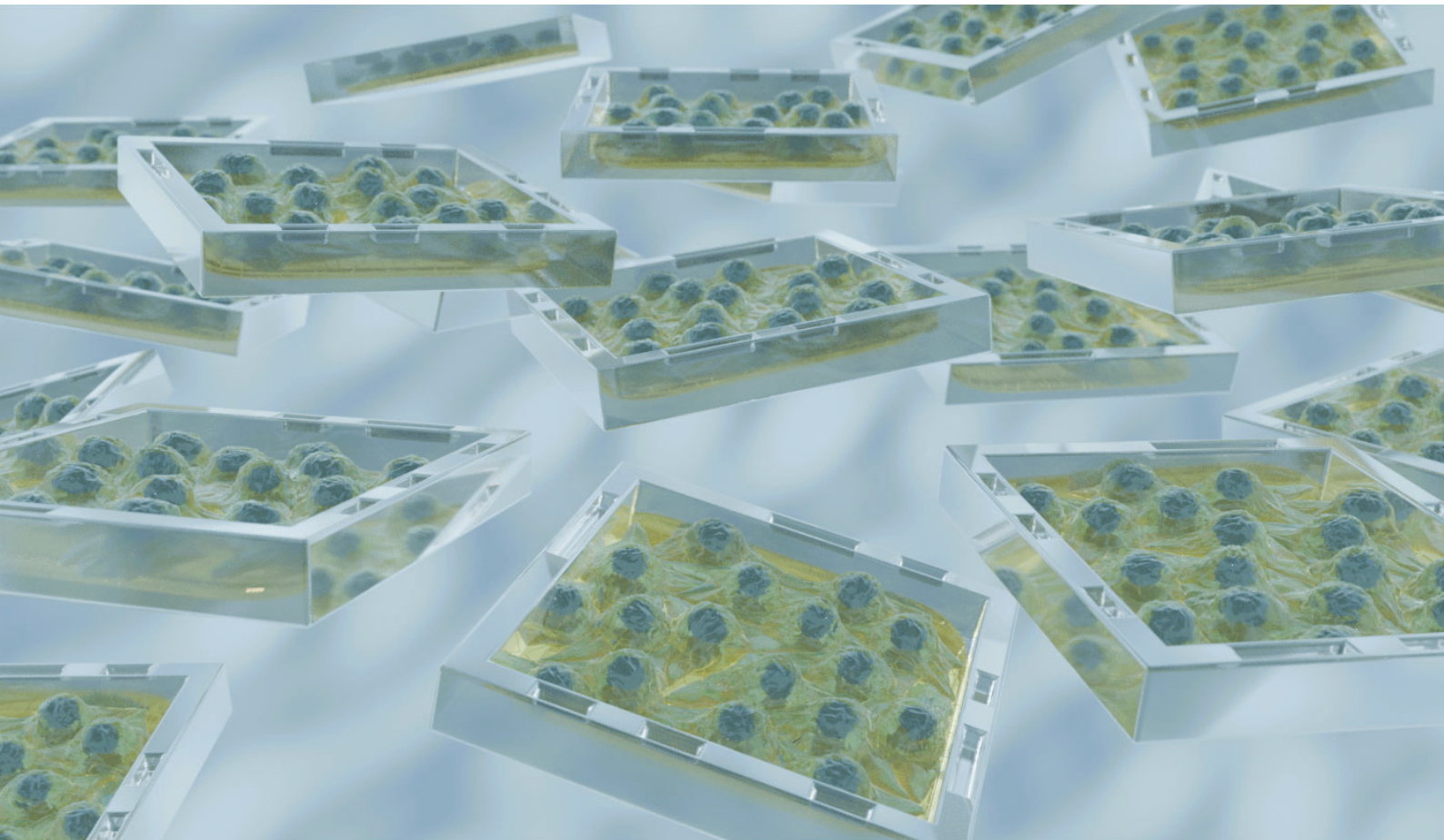




semarion

Flexibility for Adherent Cell Assays



SemaCyte[®] Microcarriers *for More Powerful Assaying Workflows*

Our SemaCyte[®] microcarrier platform leverages novel materials physics to move cells while retaining their adherent morphology. Our approach introduces flexibility, speed, and miniaturisation into existing drug discovery workflows. This unique approach to cell assays makes it possible to produce better data, faster.

The SemaCyte® Microcarrier Platform

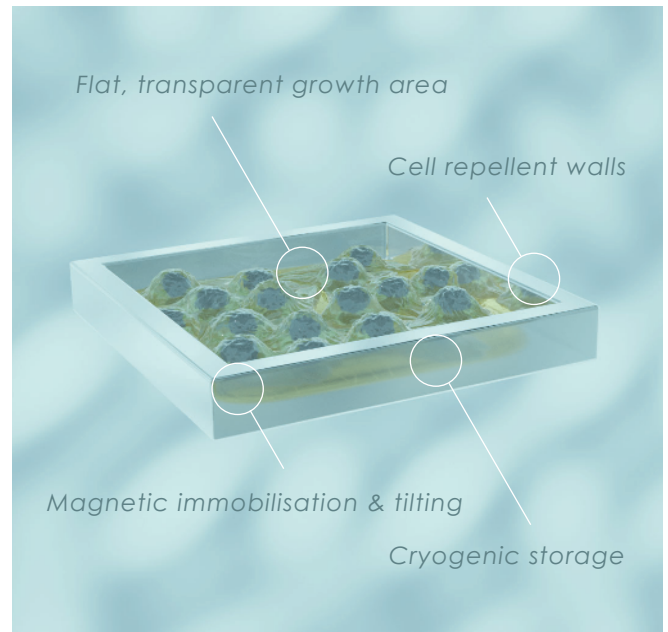
SemaCyte® assaying microcarriers function as ultra-miniaturised, mobile wells which carry small colonies of adherent cells.

These flat cell carriers can be moved with liquid handling tools and their orientation can be controlled magnetically.

The SemaCyte® products include Seeding Dishes and peripherals which integrate seamlessly with read-out equipment such as plate readers and microscopes.

Our Semalyse software can digitally isolate the microcarriers for downstream image analysis.

This unique approach enhances existing research workflows and enables novel assay methodologies.



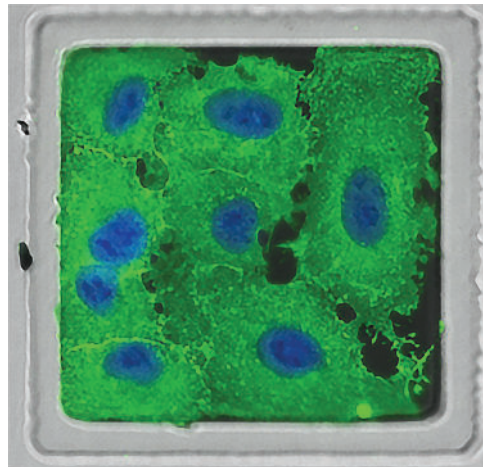
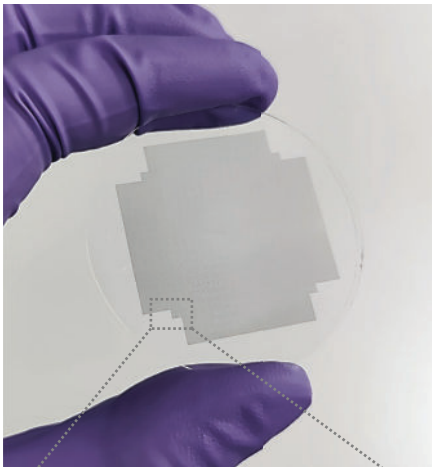
Move cells while retaining their adherent morphology



Measure Multiple Endpoints: 2-10x more data

Freeze Adherent Cells: 2-20x faster assays

Miniaturise Cell Assays: 5-100x less cells

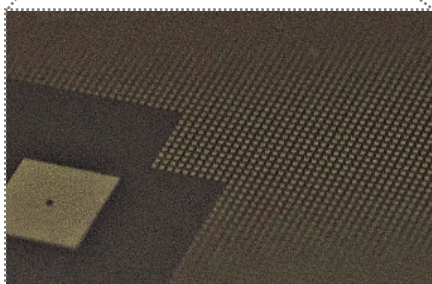


SemaCyte® microcarriers are made using microchip fabrication techniques, allowing for a high degree of control over their shape, properties, and features.

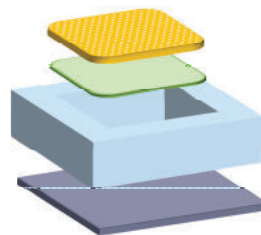
Our most common SemaCyte® microcarriers have a 100 x 100 µm² growth area and contain a synthetic fibronectin-mimetic extracellular matrix.

A standard 20 sq. cm Seeding Dish (SD-20) contains 50,000 immobilised SemaCyte® microcarriers.

Magnetic immobilisation and orientation is performed with our peripherals.



SemaCyte®
Microcarrier
Composition



Cell attachment surface

Magnetic actuator

Structural polymer

Smart release polymer



SemaCyte® Starter Kit

Seeding Dish 20, SD-20 (3x)
SemaPure15 (1x)
SemaPlate (1x)

Each SD-20 produces enough microcarriers for 2-10 multiwell plates worth of experiments.

The recommended density of SemaCytes per well ranges from 40-200 for 96-well plates and 10-50 for 384-well plates.

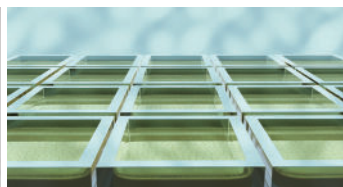
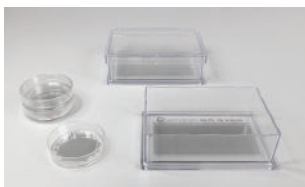
The SemaPure15 purifies SemaCytes after cell attachment. The SemaPlate orientates and immobilises SemaCytes inside microplates.

Cells on SemaCytes can be assayed in standard microwell formats for plate reader and microscopy endpoints.

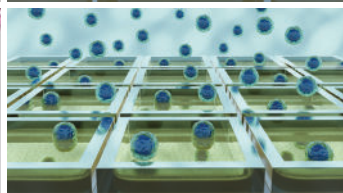
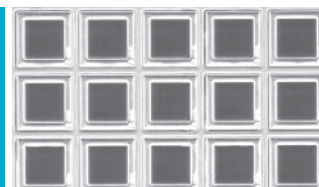


Integrate seamlessly with existing assaying workflows

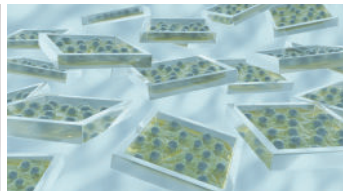
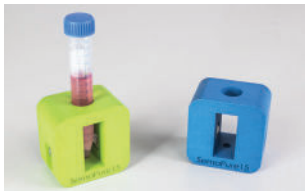
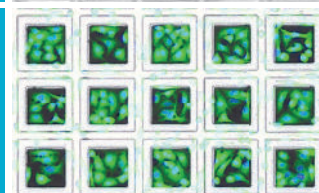
Preparation



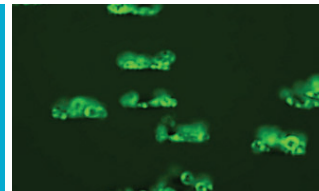
SemaCyte microcarriers are provided as immobilised arrays in Petri or ANSI/SLAS dishes.



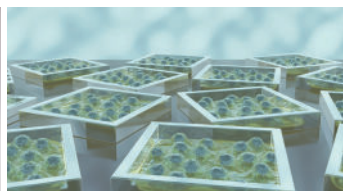
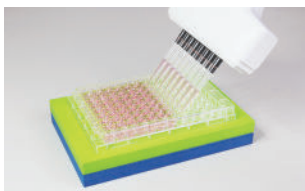
Cells are seeded and grown for a desired confluency and morphology.



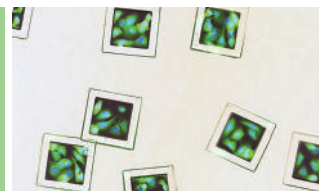
SemaCytes with cells are released into suspension, purified, and cryopreserved.

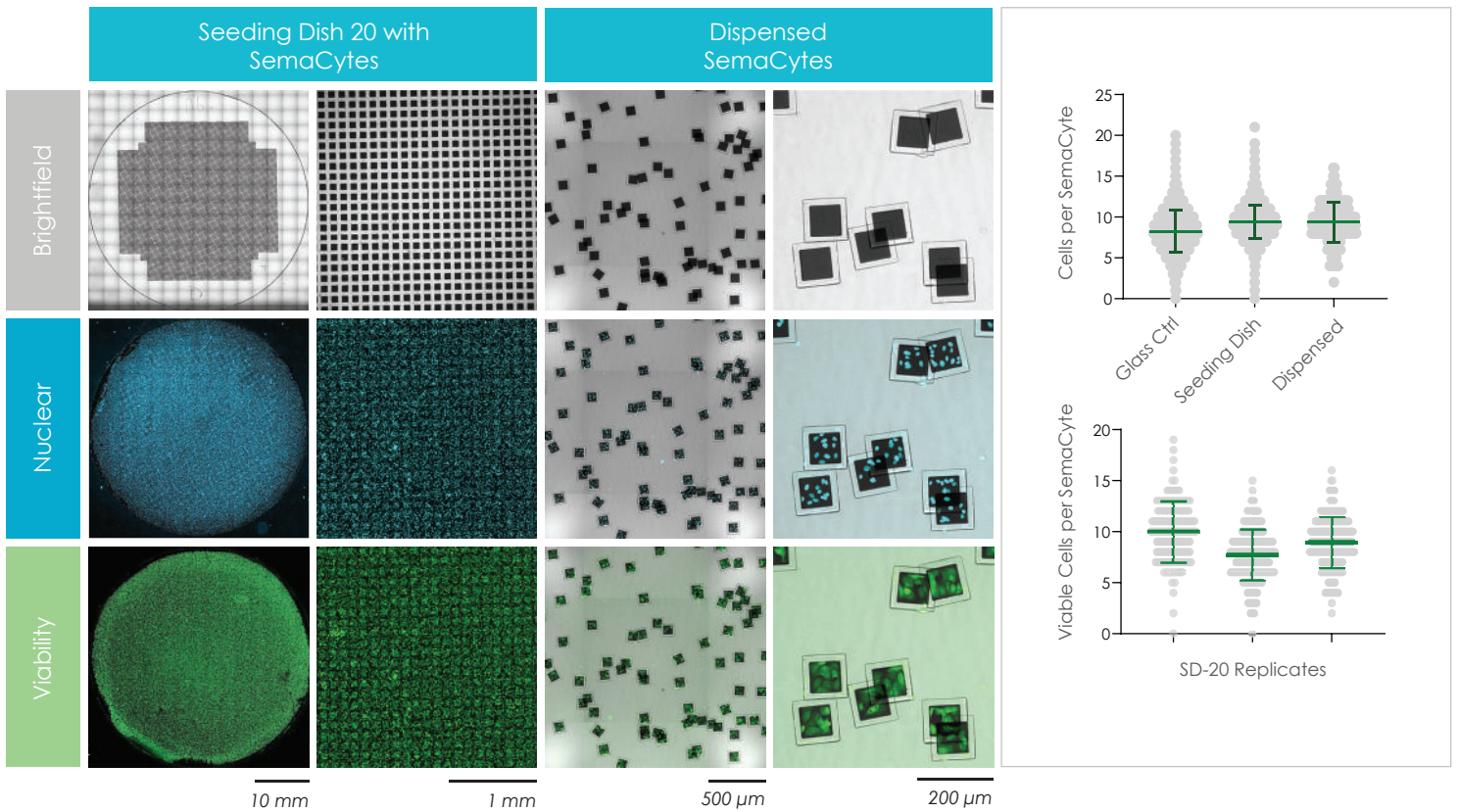


Assaying

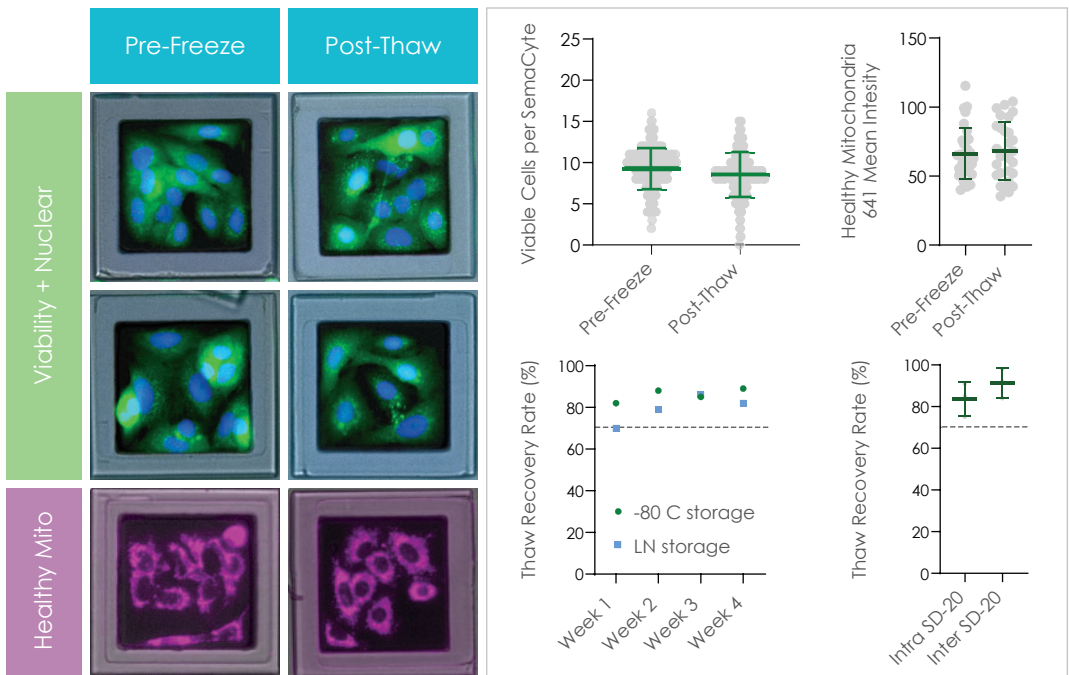


SemaCytes are dispensed at a desired density into microplates and assayed.





A549 pulmonary adenocarcinoma cells were seeded onto SD-20 Seeding Dishes at a density of 80,000 cells per cm². After 24 hours, SemaCytes were released into suspension, purified, and dispensed into 96-well plates at a density of 200 microcarriers per well. Cells were labelled with calcein AM and HCS NuclearMask blue and imaged with the ImageXpress® Pico Automated Cell Imaging System. ImageJ processing software was used to identify SemaCytes and determine the number of calcein AM positive cells. A glass control was used to compare cell densities.



SemaCytes with A549 cells were frozen as assay-ready cells in standard cryovials. After thawing, SemaCytes were magnetically purified and dispensed into microwell plates. Their viability (calcein AM) and mitochondrial health (PhenoVue 641) was assessed 1 hour after thawing.

SemaCyte® Seeding Dishes reproducibly generate carriers with healthy viable cells.

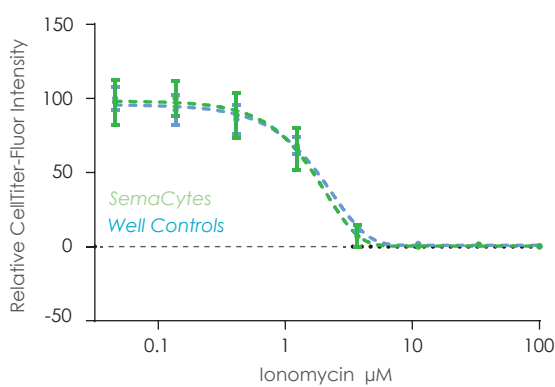
Adherent cells frozen on SemaCytes have consistently high recovery rates.

Cells on SemaCytes are healthy and assay-ready within 1 hour after thawing.

SemaCytes allow for dispensing of lower number of cells per well, while retaining high local confluency. These microcarriers enable instant assay-ready cells where confluency and phenotype are retained during freezing and thawing in cryovials.

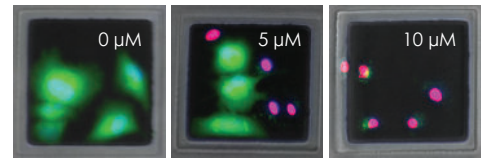
Below are examples of workflows with SemaCyte® Assay-Ready Cells compared to freshly plated cells.

Toxicity - 24 h Viability Assays



	Centre 1	Centre 2	Centre 3	
SemaCytes	2.21	3.15	1.67	IC50 (μM)
	0.89	0.57	0.51	Z-prime
Well Ctrl	2.26	3.15	1.82	IC50 (μM)
	0.76	0.83	0.76	Z-prime

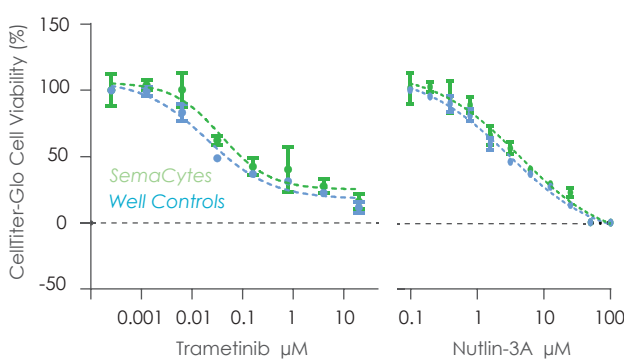
Assays performed by three research centres



Live (calcein AM) Dead (Ethidium)

Cell types: RCC-FG2 renal cells
Assay: CellTiter-Fluor™
Benefits: 2x faster data, 2x less cells

Drug Sensitivity - 72h Proliferation Assays

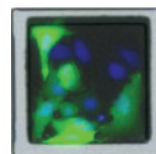
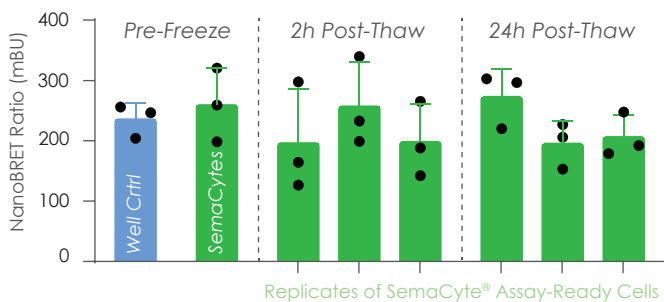


	Trametinib	Nutlin-3A	
SemaCytes	0.04	4.35	IC50 (μM)
	0.62	0.84	Z-prime
Well Ctrl	0.02	3.02	IC50 (μM)
	0.62	0.85	Z-prime

For timepoints of over 72h, the assay can be designed to let cells grow off SemaCytes onto the plate, akin to colony assays.

Cell types: A549 lung cancer cells
Assay: CellTiter-Glo™
Benefits: no 24h cell attachment

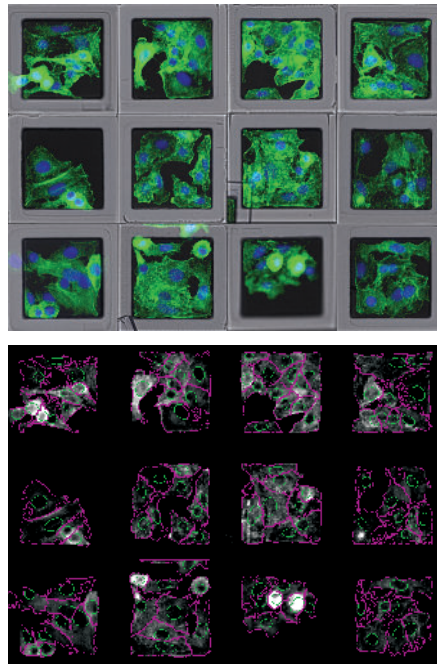
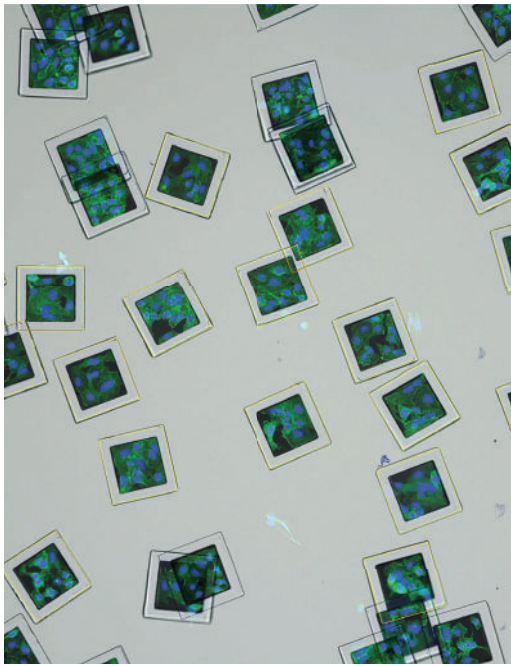
Protein-Protein Interaction - 3h Mechanistic Assays



56% GFP transfection efficiency

Cells were transfected using Lipofectamine 3000 while attached to SemaCytes. After 48h, microcarriers were aliquoted and batch frozen for on-demand deployment.

Cell types: A549 lung cancer cells
Assay: Live NanoBRET™
Benefits: 20x faster data, flexible



The Semalyse software can identify bona fide SemaCytes from microscopy images and digitally tile them together.

Tiled microcarrier images can be analysed using standard image analysis software such as cell profiler.

Below are examples of SemaCytes used for more flexible and miniaturised imaging workflows.

Cells can be fixed with PFA or methanol while attached to SemaCyte® microcarriers. Adherent cells can be moved before or after fixation for more flexibility and additional endpoints. With SemaCyte® assay-ready cells it is possible to assay and stain within 1h after thawing.

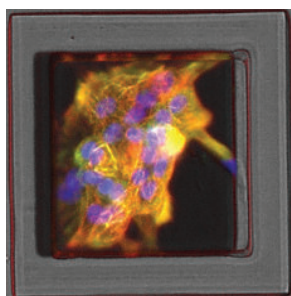
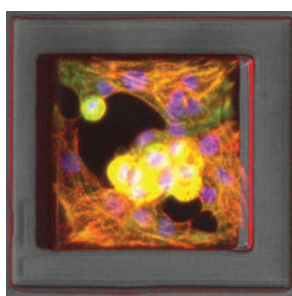
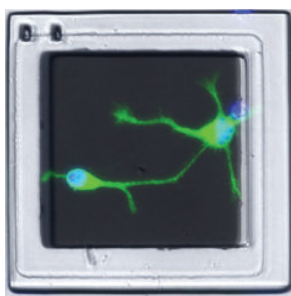
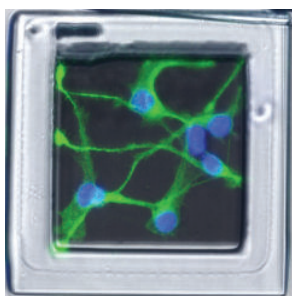
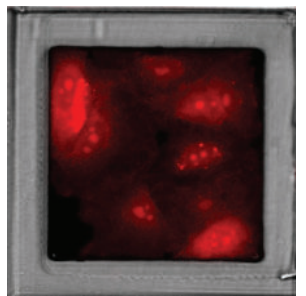
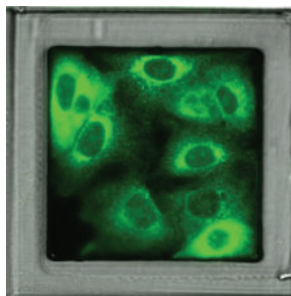
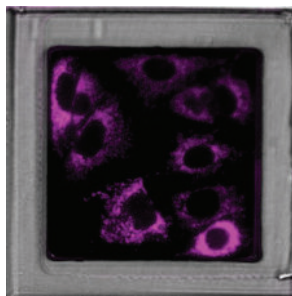
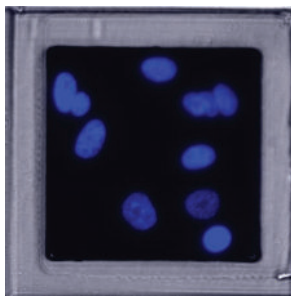
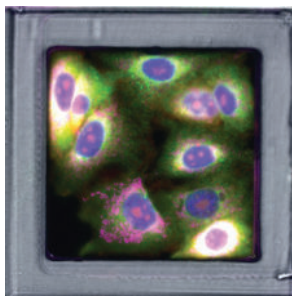
A549 Cell Painting

Hoescht

Mitochondrial Stain

Concavalin A

Phalloidin and WGA



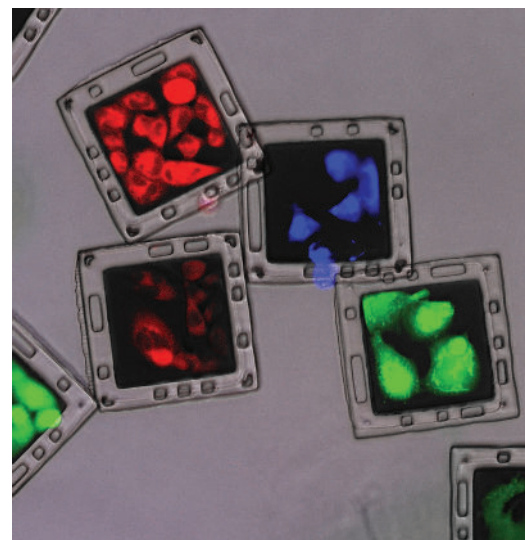
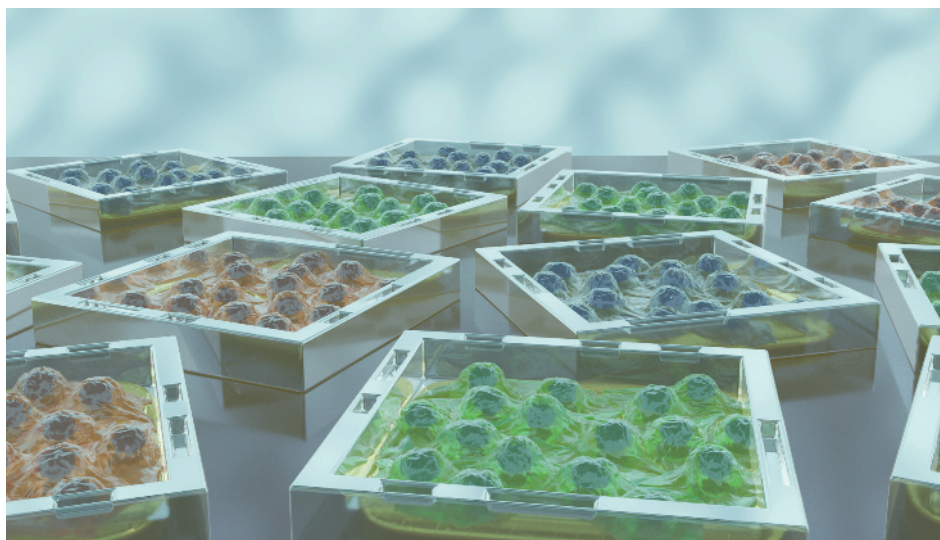
Novel methods to differentiate and mature iPS-derived cells on SemaCytes are being developed.

ioGlutamatergic Neurons from bit.bio
β3-Tubulin Nuclei

Ventricular Cardiomyocytes from Axol
α-Actinin β3 Tubulin Nuclei

PREVIEW: The SemaCyte® Multiplex Platform adds optical barcodes to the walls of the SemaCyte® microcarriers. This enables true high-order cell multiplexing for image-based assays. The barcodes are visible in brightfield and can be deconvoluted with the Semalyse software. Combined with the assay-ready features of the SemaCytes, this can drastically accelerate drug discovery campaigns.

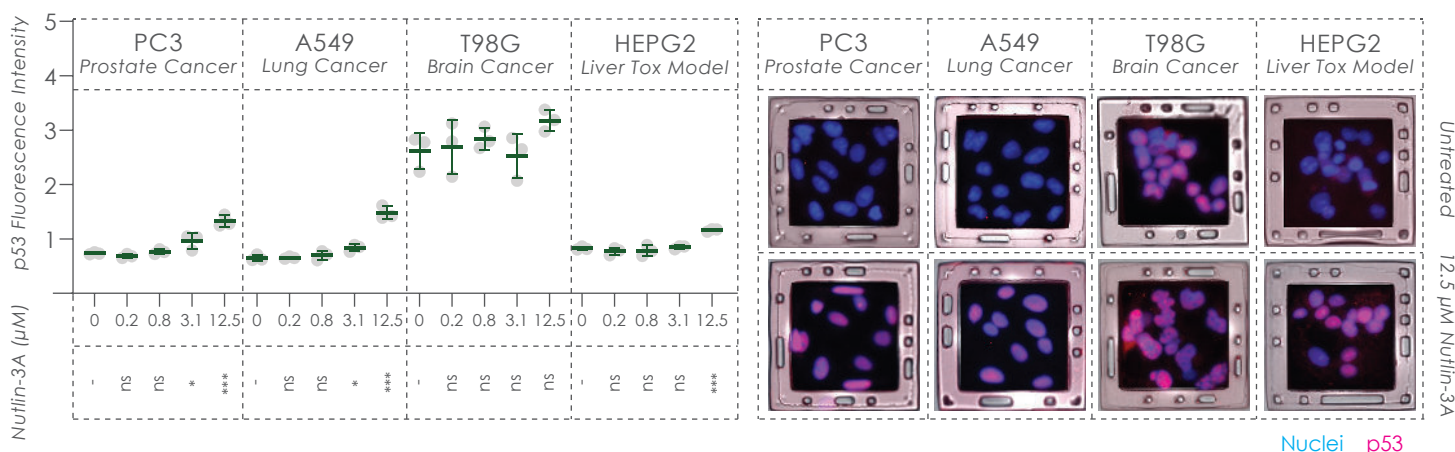
Currently under development



Optically Barcode & Pool Various Cell Types Together



Pooled Cell Panels: 2-10x less time & cost
Intrawell Controls: better data quality
Higher-Order Co-Cultures: 2-10 cells types



4-Plex Oncology Cell Panel Screen for p53 Stabilisation: Barcoded cell lines on SemaCytes are pooled and immediately drugged with Nutlin-3A. After 16h cells are fixed and stained p53 activity. When assay-ready, they accelerate timelines.

Benefits: 4x less reagents, 60x less cells, 2x faster



To learn more about our **Early Adopter Programme**, visit
www.semarion.com/early-adopter.

Let us enhance your cell assay workflows



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