APPLICATION NOTE

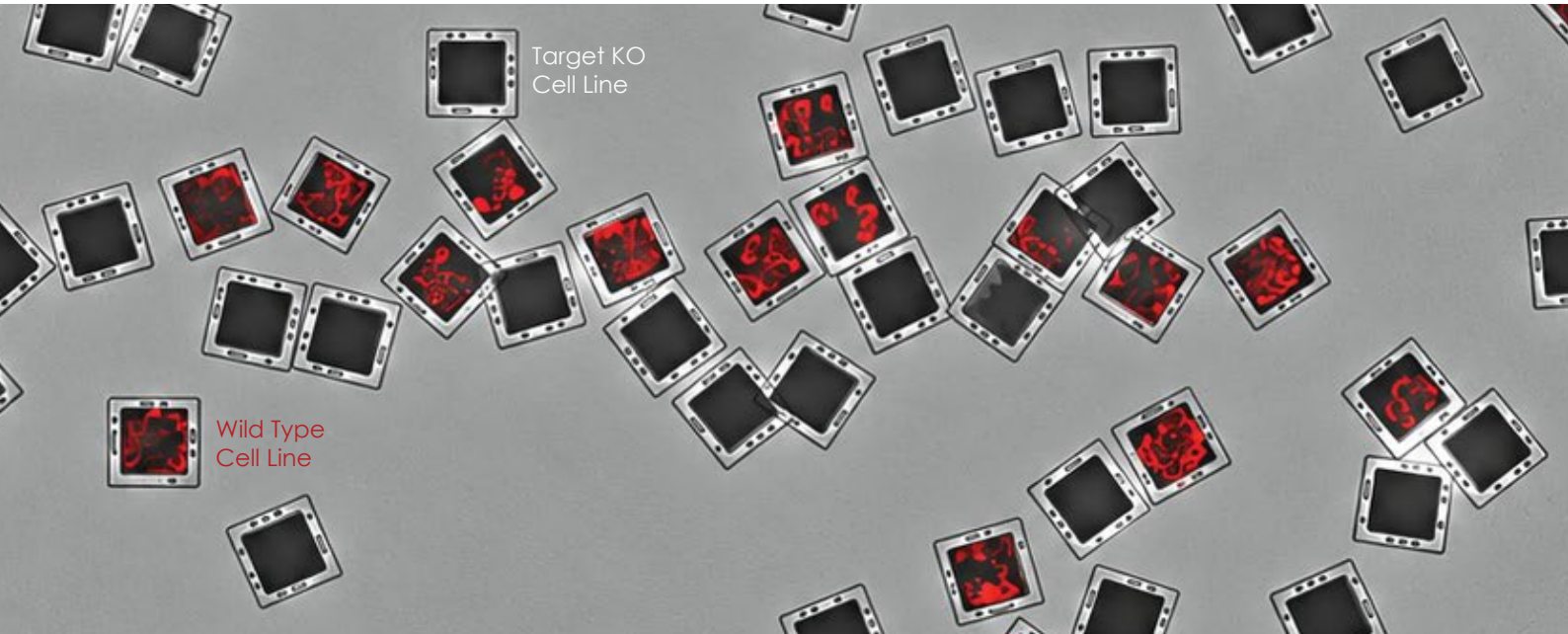
semarion

Scalable & High-Fidelity Antibody Screening

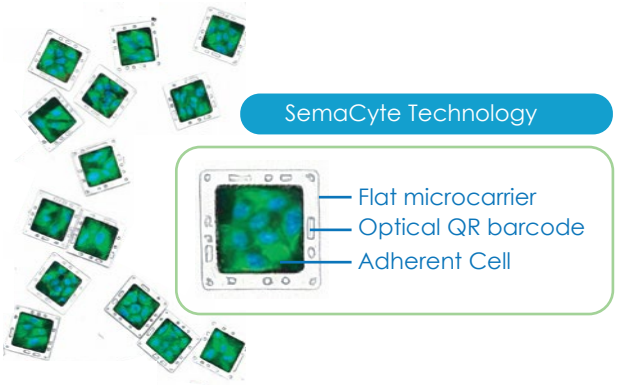
Imaging Multiplexed Cell Models for Antibody Specificity, Selectivity & Species Cross-Reactivity

From primary hit screening to antibody characterisation, SemaCyte® microcarriers enable multiplexed binding assays using adherent cells. Optically barcoded cell models, including target-expressing, species orthologue, and negative controls, can be pooled and analysed side by side in the same 384-well. Compatible with automated liquid handling and high-content imaging, SemaCytes support rapid, no-wash screening of antibody libraries and quantitative dose–response profiling of purified antibodies. Imaging enables deep model-speficic bidning analysis without affecting the native adhenert morpghology. The platform also supports downstream applications such as antibody internalisation and functional assays, including DNA damage response and T-cell engagement.

Data Richness: Specificity, selectivity, cross-reactivity within the same well
Scalability: Multiplex cell models to reduce plates, cells, antibodies and reagents
Automation: Broad tool compatribility for no-wash or multistep workflows



Barcode & Multiplex Adherent Cells for Data-Rich High Content Screening



SemaCyte® microcarriers are microscopic, optically barcoded wells that turn adherent cells into assay-ready reagents. Each cell model has a unique brightfield barcode, enabling multiple cell populations to be identified and analysed within the same well without additional fluorescent labels for cell identification.

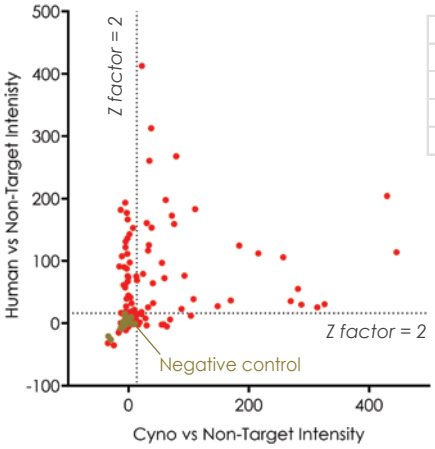
Compatible with automated 384-well workflows, SemaCytes can be cryopreserved, pooled and dispensed using standard liquid handlers. High-content imaging generates model-specific data for direct comparison of antibody binding across cell models.

Barcode. Bank. Pool. Screen.
From primary antibody hits to multiplexed binding profiles.

3-Plex Antibody Hit Screening & Species Cross-Reactivity

CHO-K1 cells transiently expressing human or cynomolgus target orthologues, alongside a non-targeting control, were multiplexed to screen B-cell supernatants. A homogeneous, no-wash assay combined supernatants and fluorescent secondary antibodies, followed by 4 h incubation and Opera Phenix imaging.

Model-normalised fluorescence enabled hit identification and species cross-reactivity assessment. Compared with iQue® screening, SemaCytes achieved 89% concordance, with a larger assay window, improved sensitivity and visual confirmation of binders.

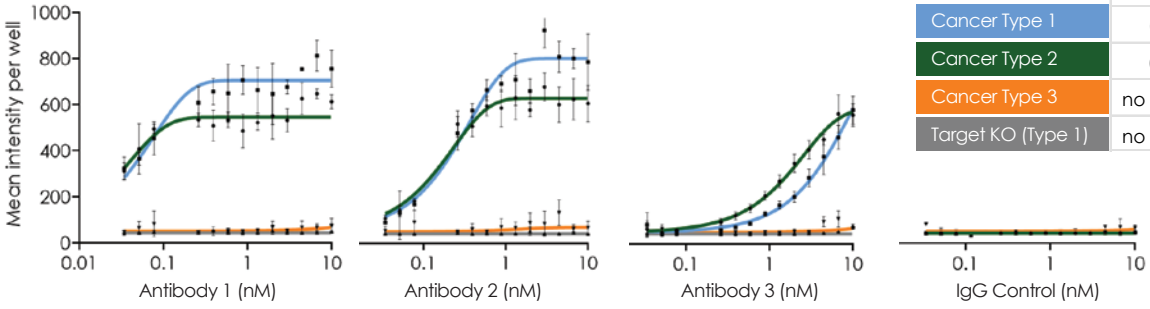


	Hits
Human + Cyno	35
Human only	8
Cyno only	37
None	43

RZ' = 0.87
Assay window = 150

4-Plex Antibody Characterisation: Binding Potency & Specificity

Four cancer-relevant and control cell models, including a target knockout, were multiplexed to profile purified antibodies across concentration ranges. Following 4 h antibody incubation, automated washing, fixation and secondary staining, Operetta imaging generated model-specific dose–response curves and apparent EC_{50} values. The assay enabled comparison of antibody binding potency and cell-model selectivity, while knockout and IgG controls supported assessment of on-target specificity and nonspecific binding.



	Ab 1	Ab 2	Ab 3
Cancer Type 1	0.03	0.16	1.79
Cancer Type 2	0.05	0.25	4.43
Cancer Type 3	no effect	no effect	no effect
Target KO (Type 1)	no effect	no effect	no effect

EC50 (nM)

RZ' = 0.74
Assay window = 21