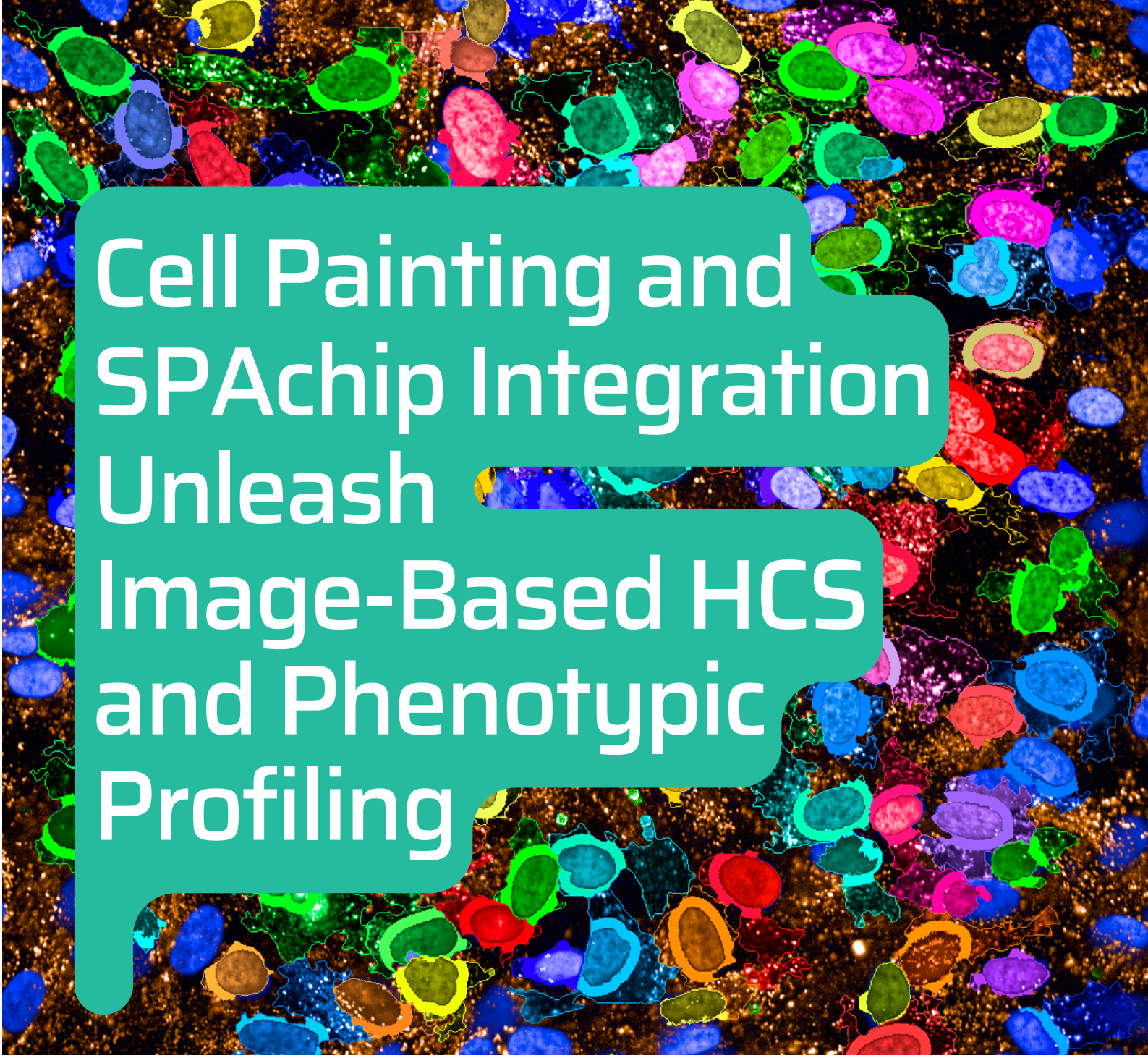


Cell Painting and SPAchip Integration Unleash Image-Based HCS and Phenotypic Profiling

Combine SPAchip® functional assays with Phenotypic Profiling

Image-based phenotypic profiling that can be applied in drug discovery for the biotech and pharma industry. SPAchip® functional readouts can offer valuable insights of cell homeostasis to be combined with phenotypic information from cell painting stains.

01

SPACHIP® FUNCTIONAL ASSESSMENT

Continuous monitoring of current SPAchip® assays can be evaluated to evaluate the effect of a chemical or genetic perturbation

02

MODE OF ACTION STUDIES

To study the mode of action of drugs or compounds by analyzing their phenotypic effects on cells

03

DRUG EFFICACY AND TOXICITY ASSESMENT

Drug efficacy and toxicity assessment: Assess the efficacy and toxicity of drugs in a high-throughput manner

04

PHENOTYPIC SCREENING AND HIT SELECTION

Cell Painting enables phenotypic screening of large compound libraries to identify hits with desired phenotypic effects

a4cell® nanodevices

OLS® OMNI Life Science

www.ols-bio.com

Purpose

Cell painting can be used to perform chemical or genetic perturbation studies of large libraries to select hits with changes in cellular morphology associated with specific phenotypes. This assay can aggregate data to obtain clusters with biological similarity that can reveal phenotypic fingerprints associated with the biological state of a cell.
As an image-based assay with subcellular resolution, Cell Painting focuses on specific organelle distribution important for cell homeostasis. Image analysis include signal intensity, texture, morphological features and correlation between these values that can uncover unsuspected off-target activities or help to infer mechanism of Action from reference databases. This methodology can be also implemented to empower toxicity testing with specific organelle targets.

SPAchip® Functional Assays and Cell Paint Selection

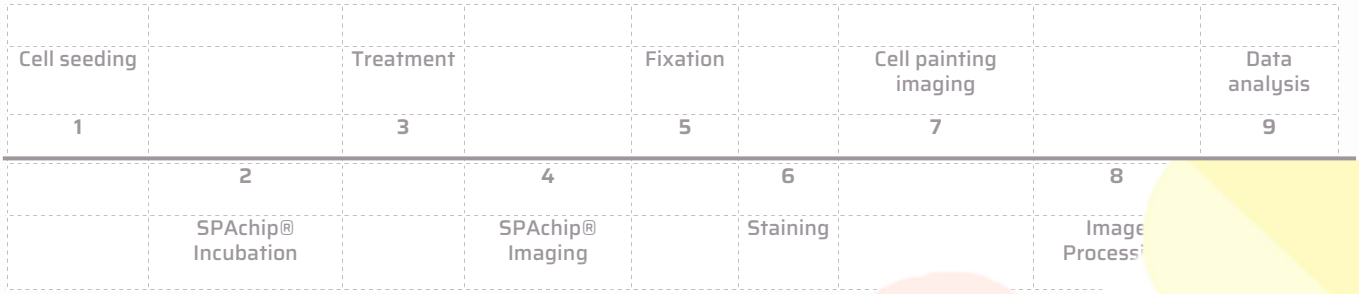
One important aspect to design your SPAchip® assay to strengthen Cell Painting data is spectral crosstalk, as some of the dyes stain with nearby excitation and can overlap with SPAchip® fluorescence channels. The features that can be analyzed with Cell Painting assays include:

- **SPAchip® Functional Assessment:** Continuous monitoring of current SPAchip® assays for real-time dynamic studies.
- **Nuclear/Mitotic Quantification:** Cell count and cell division state to evaluate cell proliferation and viability.
- **Cell Morphology & Organelle Distribution:** Spatial distribution and morphology of whole cell and subcellular structures.
- **Cytoskeletal Arrangement:** cell compartmentalization, cell-to-cell interactions and crosstalk.
- **Mitochondrial Health: number of** mitochondria, intensity and texture.

Consult SPAchip® protocol and A4cell staff for detailed information

Methodology

In this assay, eight cellular components are stained with six dyes and imaged in five channels on a fluorescence microscope. Image analysis software identifies individual cells and extracts a few thousand features from each, producing single-cell profiles. Cells are segmented to identify properties that results in features with associated values that can be quantified from the pixel information.
Cell Painting studies as described by the Broad Institute (Bray et al., 2016) and JUMP Consortium (Chandrasekaran et al., 2023) include different steps that can be combined with functional SPAchip® readouts with the following workflow:



Target Structure	Dye	SPAchip® Kit
Nuclei	Hoechst 33342	
Endoplasmic Reticulum	Concanavalin A – AF488	CytoCHECK SPAchip® pH GREEN single-Detection Kit CytoCHECK SPAchip® Calcium GREEN single-Detection Kit
Nucleoli/Cytosolic RNA	SYTO 14	
Golgi, Plasma Membrane	WGA – AF555	
Actin	Phalloidin AF568	CytoCHECK SPAchip® pH RED single-Detection
Mitochondria	MitoTracker Deep Red FM	CytoCHECK SPAchip® pH RED single-Detection

Cell painting assay done today!

Consult us to design your experimental protocol, perform cell testing, image your samples or analyse the cell painting data. Build you own case study and obtain a full report to improve your research.

References

Bray, M., Singh, S., Han, H., Davis, C. T., Borgeson, B., Hartland, C., Kost-alimova, M., Gustafsdottir, S. M., Gibson, C. C., & Carpenter, A. E. (2016). Cell Painting , a high-content image-based assay for morphological profiling using multiplexed fluorescent dyes. Nature Protocols, 11(9), 1757–1774. https://doi.org/10.1038/nprot.2016.105

Chandrasekaran, S. N., Ackerman, J., Alix, E., Ando, D. M., Arevalo, J., Boisseau, N., Borowa, A., Boyd, J. D., Brino, L., Byrne, P. J., Ch, C., Cimini, B. A., Clevert, D., Deflaux, N., & Doench, J. G. (2023). JUMP Cell Painting dataset: morphological impact of 136,000 chemical and genetic perturbations.