READY-2-GO
NEURITE OUTGROWTH & NETWORK DYNAMICS ASSAY SERVICES

BACKGROUND

Assessing the impacts of therapeutics targeting neurological disorders on neurite outgrowth or neuronal network integrity is essential to the success of any neurobiology-focused, drug-discovery/development campaign. Additionally, identifying potential neurotoxic effects of therapeutics targeting other non-neuronal disease classes is beneficial in flagging and addressing undesirable, off-target effects that could jeopardize clinical advancement and, ultimately, patient safety.

THE CHALLENGE

As iPSC-derived neuronal cells grow and proliferate *in vitro*, the neurites tend to overlap extensively, making accurate measurements of neurite length, branch points, *etc.* difficult or impossible. As a result, accurately measuring the impact of therapeutics upon neurite outgrowth requires extensive optimization of cell density to achieve minimal overlap in neurites that originate from adjacent cells, while maintaining a sufficient density to ensure cell health.

While these assays can be relatively straightforward to establish using immortalized cell lines such as PC-12 cells, those models may not be as physiologically relevant compared with human iPSC-derived neuron models.

Regarding detection, despite broad availability of robust detection antibodies for neuronal cell markers, a standard, antibody-based detection approach typically requires extensive wash steps that could mechanically disrupt the neuronal network.

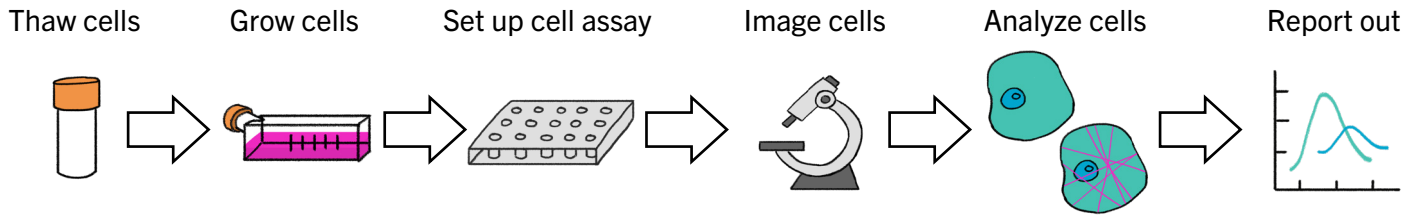
OUR SOLUTION

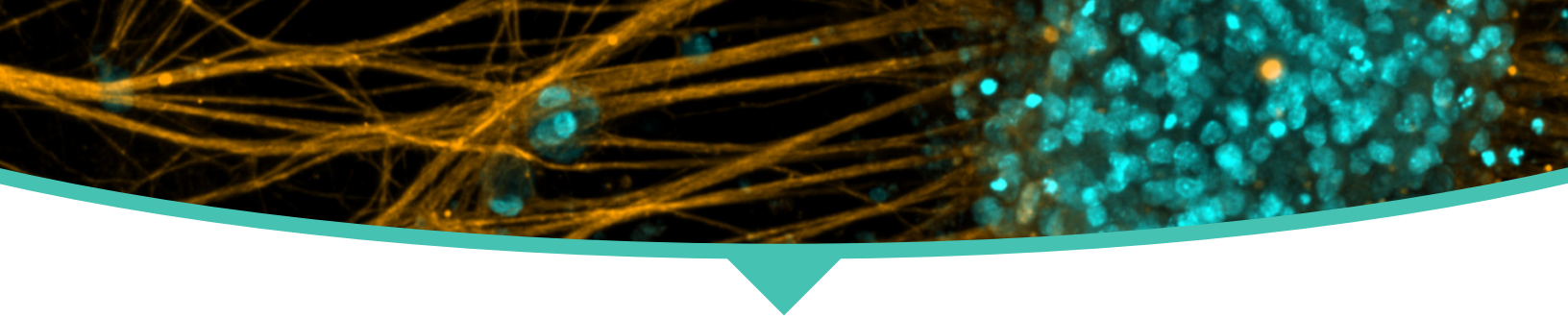
To address these challenges, PhenoVista has developed the Ready-2-Go (R2G) Neurite Outgrowth assay service and the R2G Neurite Network Dynamics Assay Service, enabling robust, accurate quantification of neurite outgrowth or network formation respectively, using human iPSC-derived neurons and a homogeneous assay workflow with dye-based detection. We have optimized cell-seeding densities to ensure accurate neurite measurements, with the flexibility to assess network formation.

KEY FEATURES & BENEFITS

- **Physiologically relevant cell model.** Human iPSC-derived neurons cultured in defined growth conditions.
- **Greater assurance of data quality than other models.** Homogeneous, dye-based detection limits artifactual neuronal network disruption and minimizes assay noise.

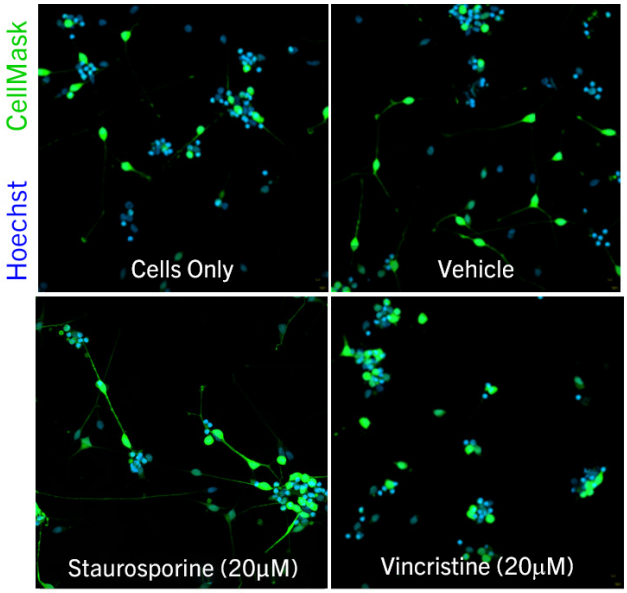
PROCESS OVERVIEW

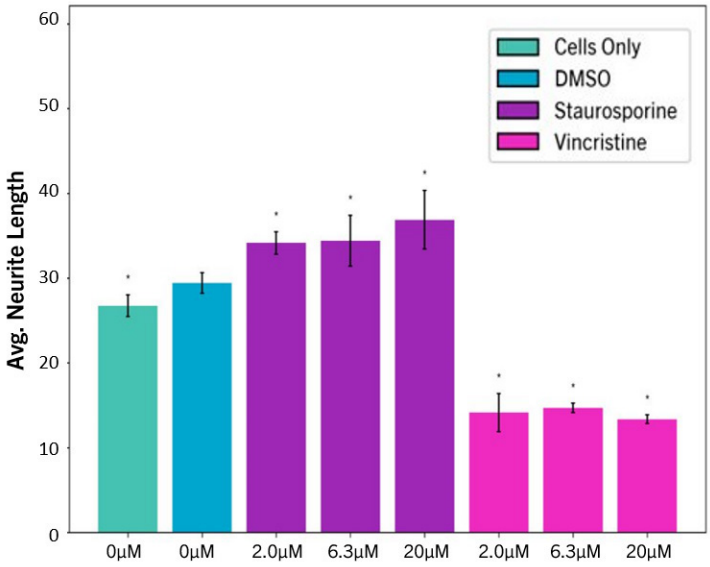




HOW IT WORKS

Cells are seeded into 384-well plates. For neurite-outgrowth assessment, cells are incubated in the presence of drug for 4 and 24 hours, when they are fixed and stained for imaging and analysis. For assessing neurite network, cells are treated with drugs on day 6 for 24 and 72 hours, and they are fixed and stained on days 7 and 9 for imaging and analysis. In the images below, human iPSC-derived glutaminergic neurons were treated with reference compounds, and neurites were measured at 24 hrs. Nuclei were stained with Hoechst (blue) and cell bodies and neurites with CellMask (green).





ASSAY SERVICE DETAILS

	Ready-2-Go Neurite Outgrowth & Network Dynamics^	Bespoke Assay Services
Cell Type	iCell Glutaneurons (Fujifilm Cellular Dynamics, Inc.)	If you would like to expand the service offering beyond R2G shown on left, please contact us at info@phenovista.com or reach out to your local sales representative.
Markers	Hoechst (nuclei), CellMask (cell body/projections)	
Dosing	6 doses of your test article	
Positive Control*	Staurosporine	
Negative Control*	Vincristine	
Time Points	Neurite Outgrowth^: 4, 24 hrs Network Dynamics^: 7, 9 days	
Assay Readouts	Total cell count, neurite length/area, # branch points	

* Vehicle and untreated controls also included.
^Neurite Outgrowth and Network Dynamics are distinct service offerings and can be purchased separately.

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